Release of Two Nonindigenous Beetles, *Nanophyes marmoratus* and *N. brevis*, for Biological Control of Purple Loosestrife, *Lythrum salicaria*

Environmental Assessment

May 1994

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Finding of No Significant Impact

I. Introduction

The Animal and Plant Health Inspection Service (APHIS), U.S. Department of Agriculture (USDA), is considering issuing permits (e.g., Appendix 1) for importation into quarantine and subsequent release in several States of two nonindigenous insects (they are native to Europe and parts of Asia). The applicants wish to release these insects as biological control agents of purple loosestrife, an introduced weed. The permits would be issued by Biological Assessment and Taxonomic Support (BATS), Plant Protection and Quarantine (PPQ). Under the Federal Plant Pest Act (FPPA) as amended 1957 (7 U.S.C. 150aa *et seq.*) and the Plant Quarantine Act of 1908, as amended 1967 (7 U.S.C. 151 *et seq.*), USDA has broad authority to regulate the importation, interstate movement, and release into the environment of organisms that may directly or indirectly injure, cause disease, or damage plants. Biological control organisms are regulated by APHIS under the FPPA and involve a permitting process (7 CFR 330).

BATS prepared this environmental assessment (EA) prior to its permit decision in compliance with the National Environmental Policy Act (NEPA). The proposed authorization by APHIS for introduction and release of nonindigenous phytophagous insects into the environment comprises a Federal action that is not classified by APHIS for the purposes of the NEPA process (42 U.S.C. 4321 et seq). Regulations for implementing NEPA have been established by the Council on Environmental Quality (CEQ) (40 CFR 1500-1509), USDA (7 CFR 1b), and APHIS (44 FR 50381-50384 and 44 FR 51272-51274. Under the CEQ implementing regulations (40 CFR §1501.4(b) (1992), an EA must be prepared for such an action. An EA is a concise public document that must include brief discussions of the need for the proposed action, alternatives to the proposed action, and the environmental impacts of the proposed action and alternatives. Typically, EAs are developed to lead to one of two conclusions: either an environmental impact statement (EIS) is needed or a finding of no significant impact (FONSI) can be supported.

II. Proposed Action

A. Summary

Detailed information about the proposed experimental protocol is given in Appendix 2, Section 3.1., additional information is provided in Appendices 3, 4, and 5. In summary, the applicants propose to introduce and release into North America two species of beetles, the flower-feeding weevil *Nanophyes marmoratus* Goeze (Coleoptera: Curculionidae) and the seed-feeding weevil *N. brevis* Boh. for biological control of purple loosestrife. Several specific steps will be taken to ensure that beetles collected in Europe and subsequently released in North America are free from diseases and parasites (Appendix 5). Adult beetles of both species will be collected from purple loosestrife in Europe. Each individual will be examined under a dissecting microscope for species identity, external parasites (e.g., mites), and general condition; any individual appearing at all unusual will not be included. Before beetles intended for release are shipped to North America, samples will be dissected and examined for a variety of microorganisms, entomopathogenic nematodes, and any other external or internal parasites or parasitoids. Cleared beetles will then be shipped to the approved quarantine facility in the U.S. After ensuring that the beetle colonies

constituted pure cultures (i.e., free from any parasites, diseases, etc), several thousand beetles of one or both species would be transported to various States for release. An established network of specialists working on biological control of purple loosestrife would be used to propagate and establish *N. marmoratus* and *N. brevis*.

B. Biology of Purple Loosestrife

Details on the biology of purple loosestrife, *Lythrum salicaria* L. (Lythraceae), are provided in Appendices 2 and 3. Purple loosestrife is an herbaceous perennial of Eurasian origin that has become naturalized in North America. When mature, individual plants may exceed six feet in height and four feet in width. The weed first arrived during the 1800s, probably in ship ballast, but has also been introduced as an ornamental (Stuckey, 1980). Purple loosestrife has become a serious weed in marshes and alluvial wetlands (Thompson et al, 1987). The main problem areas are in the northeast and north-central portions of the United States and in parts of southern Canada. Its recent spread into the Midwest and West along major rivers and irrigation systems completes a cross-continent distribution and demonstrates that this weed is a problem of national importance (Thompson, et al. 1987).

C. Biologies of *Nanophyes marmoratus* and *N. brevis*

Nanophyes marmoratus Goeze is native throughout Europe and western Siberia. *N. brevis* Boh. is native throughout central and southern Europe, including the entire European Mediterranean and Asia Minor. Their geographic ranges and ecological requirements correspond to that of their host, purple loosestrife. The biologies of *N. marmoratus* and *N. brevis* are described more fully in Appendices 2 and 3. Feeding by the beetles damages leaves, flower buds, flower parts, and especially the ovaries of purple loosestrife; this feeding limits plant reproduction. Host specificity studies using beetles from northern Germany (*N. marmoratus*, Appendix 2, Section 5.1.7 and Table 2) and southern Germany (*N. brevis*, Appendix 2, Section 5.2.7 and Table 3) demonstrated that these beetles feed normally only on purple loosestrife. And in nature, purple loosestrife is the only known host for *N. marmoratus* and *N. brevis*.

III. Purpose and Need for Proposed Action

The purpose of the proposed Federal action (i.e., issuance of permits for release of *N. marmoratus* and *N. brevis*) is to provide applicants with the opportunity to establish natural enemies of purple loosestrife, an introduced weed. Although other biological control agents of purple loosestrife were introduced from Europe during 1992, (e.g., a root-mining weevil, *Hylobius transversovittatus*, and two leaf-feeding chrysomelid beetles, *Galerucella calmariensis* and *G. pusilla*), the applicants wish to complement the impact of these agents by introducing *N. marmoratus* and *N. brevis*, which are expected to reduce seed production in purple loosestrife and therefore reduce the plant's ability to compete with native plants (see also Appendix 2, Section 3).

Release is intended to address two primary needs: the need to facilitate use of self-perpetuating biological controls (should these beetles become established, they would provide continuous

contributions to purple loosestrife management without further human intervention) and the need to reduce dependence on the primary methods (e.g., herbicides) being used currently to manage purple loosestrife; these methods are expensive, inefficient, disruptive for the flora and fauna of treated areas, and temporary (Thompson *et al.*, 1987).

IV. Environmental Consequences of the Proposed Action

A. Summary

The proposed Federal action (i.e., issuance of permits for release of nonindigenous insects) has no environmental impacts *per se*. However, issuance of the permits result in environmental impacts caused by release of the insects. As such, the environmental impacts considered in this EA are those that would result from release of the insects. The petition to release *N. marmoratus* and *N. brevis* in North America (Appendix 2) addresses environmental impacts in several sections but especially in Sections 6-8. The weevils are not expected to have any direct effects on soil, water, air quality, wildlife, or human population. The potential environmental impact of primary concern to APHIS regarding issuance of permits for release of nonindigenous biological control agents is the impact that released agents may have on indigenous nontargets (e.g., feeding by the agent on native nontarget plants). This issue is discussed in the petition (Appendix 2, Section 5) and below. A secondary concern is the indirect effect that feeding by the agents may have on other species or systems.

B. Direct Effects

The host range of all known species of *Nanophyes* is restricted to plants within the Family Lythraceae. Host range studies were conducted on *N. marmoratus* collected from purple loosestrife in northern Germany Appendix 2, Section 5.1.7) and *N. brevis* collected from purple loosestrife in southern Germany (Appendix 2, Section 5.2.7). Results indicated that both species were highly specific to purple loosestrife. During external review of the petition (Appendix 2), the concern was raised that the host ranges of the beetles to be released (which are to be collected at various locations from Southern Europe to Finland) may differ from the host ranges of the beetles used in the host range studies (which were collected in Germany). This is an important consideration because it reflects directly on the potential of the released organisms to act as plant pests. The petitioners offered a response to this concern (Appendix 4, Section A.5., p. 4). With the assistance of its external reviewers, APHIS considered carefully the potential for *N. marmoratus* and *N. brevis* to act as plant pests, and collected additional information (see below) to assess more completely the plant pest risk posed by release of *N. marmoratus* and *N. brevis*.

It is reasonable to expect certain types of variation among populations of insects. For example, it is likely that the timing of seasonal events such as reproduction varies with latitude among *Nanophyes* populations (see Tauber *et al.* 1986). And species of herbivorous insects with wide host ranges typically demonstrate genetically—based host preferences (e.g., Via, 1984a; 1984b); specimens collected from a particular plant species would be expected to show a preference for that plant species. Thus, it has been suggested that specimens of *N. marmoratus* and *N. brevis*

collected from various source populations may show differences in host preference. The petitioners plan to collect beetles for release in North America from a variety of climatic regions in Europe because purple loosestrife occurs in a variety of climatic regions in North America. To test the hypothesis that the host ranges of beetles from the various areas are the same, the host ranges of beetles from each potential collection site would have to be tested individually. Currently, these data do not exist and uncertainty remains regarding the host ranges of beetles from all potential collection areas. For some reviewers of the release proposal, this concern was sufficient justification to recommend that authorization for North American releases be limited to beetles collected from sites in Germany (i.e., the source of material used for host range studies).

Despite the absence of explicit host range studies on beetles from all intended collection sites, the potential for host range differences among specimens of *N. marmoratus* and *N. brevis* collected from southern Europe to Finland (including Germany) is considered to be acceptably low for two primary reasons. First, all host range studies have demonstrated, and all field collections suggest that these beetles have a strong preference for purple loosestrife over all other plants tested. And second, even if different populations of these beetles have different host preferences (and there are no data to support this hypothesis), the petitioners plan to collect *N. marmoratus* and *N. brevis* from wild purple loosestrife plants growing within the natural range of both purple loosestrife and the beetles. As such, APHIS has concluded that the probability is quite low that the host ranges of various populations of either *N. marmoratus* or *N. brevis* differ significantly from each other. Because normal feeding by *N. marmoratus* and *N. brevis* is expected to be restricted to purple loosestrife (see Section II.C. and Appendix 2, Sections 5.1.4, 5.1.7, 5.2.4 and 5.2.7), direct effects should be limited to populations of purple loosestrife. Indeed, these direct effects on populations of purple loosestrife are the intended, positive environmental effects.

Another valid concern raised during external review of the release petition concerns the fact that purple loosestrife has a showy and profuse floral display and has been used as an ornamental garden plant. In fact, nurseries in some States have developed horticultural stocks of purple loosestrife. If *N. marmoratus* and *N. brevis* individuals find these plants, they may feed on them and become a problem for purple loosestrife horticulturists. These individuals would probably consider *N. marmoratus* and *N. brevis* to be pests. However, as discussed in Appendix 4 (Section B), various States have recognized risks posed by ornamental plantings of purple loosestrife and have prohibited its sale and distribution. Nonetheless, although the direct impact of feeding on horticultural plantings of purple loosestrife may be important to a few individual horticulturists, these impacts are not expected to be significant even on a local scale, especially considering the positive environmental impacts associated with feeding by *N. marmoratus* and *N. brevis* on pest populations of purple loosestrife.

C. Indirect Effects

Indirect effects are those impacts associated with damage or mortality caused to individuals or populations of purple loosestrife. The most likely indirect effect is the natural or human-assisted recolonization by native plants on formerly loosestrife-infested lands. The reduction of *L. salicaria* is expected to increase land use by wildlife and open infested wetlands to a more productive environmental role. The reduction of chemical herbicides used to control the plant will have a positive effect on soil and water quality. Purple loosestrife is used by honey bees, but

is considered of minor importance as a nectar source. Since these agents will not eliminate purple loosestrife populations, *L. salicaria* can still provide a source of nectar.

Potential indirect effects caused by feeding of *N. marmoratus* and *N. brevis* on horticultural plantings of *Lythrum* spp. fall into three general categories: economic damage caused to professional horticulturists, nuisance to amateur horticulturists, and environmental damage caused by methods used to control *N. marmoratus* and *N. brevis* in horticultural plantings of purple loosestrife. However, due to the limited nature of purple loosestrife plantings on a national level (e.g., see Appendix 4, Section B), none of these indirect effects are expected to be significant.

D. Endangered and Threatened Species

Releases of *N. marmoratus* and *N. brevis* are not expected to have any direct impact on endangered or threatened species. There are no species in the Family Lythraceae listed by the Federal government as endangered or threatened. Although purple loosestrife has become established in various North American ecosystems, it is an aggressive nonindigenous pest with negative environmental impacts. Nor is release of *N. marmoratus* and *N. brevis* expected to have any negative indirect effect on endangered or threatened species. In fact, several State-listed endangered species are expected to benefit from reduction of *L. salicaria*, including a localized bulrush (*Scirpus longii*) in Massachusetts, dwarf spike rush (*Eleocharis parvula*) in New York, and the bog turtle in the northeastern U.S. (Malecki et al, 1993).

V. The No Action Alternative and Its Impacts

The Federal action being considered in this EA is issuance of permits for release of two nonindigenous beetles, *N. marmoratus* and *N. brevis*. The only alternative considered by BATS was not issuing the permits (i.e., No Action). Environmental impacts associated with the No Action alternative fall into two general categories. First, ecological damage caused by purple loosestrife would continue. Native vegetation would continue to be displaced and biotic diversity reduced. The replacement of cattails (*Typha* spp.) by purple loosestrife would continue to restrict the preferred habitat of muskrats and long-billed marshwren (Rawinski & Malecki, 1984). Second, in the absence of an effective overall biological control program, the environmental damage caused by repeated use of herbicides would continue. Both types of impacts are occurring currently. The proposed release of *N. marmoratus* and *N. brevis* is intended to reduce these negative environmental impacts. The extent to which releases of *N. marmoratus* and *N. brevis* will lessen these effects is unknown and will remain unknown until after the proposed releases are made and monitored.

VI. References

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Via, S. 1984a. The quantitative genetics of polyphagy in an insect herbivore. I. Genotype—environment interaction in larval performance on different host plant species. Evolution 38: 881-895.

Via, S. 1984b. The quantitative genetics of polyphagy in an insect herbivore. II. Genetic correlations in larval performance within and across host plants. Evolution 38: 896-905.

VII. Preparation, Consultation, and Review

This Environmental Assessment was prepared by the Animal and Plant Health Inspection Service (APHIS); Plant Protection and Quarantine (PPQ); Biological Assessment and Taxonomic Support:

Polly P. Lehtonen, B.S., Botanist, Preparer

Michael J. Firko, Ph.D., Entomologist, Preparer

Kenneth Lakin, Ph.D., Branch Chief

Matthew Royer, Ph.D., Chief Operations Officer

This Environmental Assessment was reviewed by other staff within APHIS:

Robert V. Flanders, Ph.D., National Biological Control Institute

The following technical specialists reviewed this Environmental Assessment:

Jack R. Coulson, B.S., USDA, Agricultural Research Service

A. C. Schmidt, M.S., Agriculture Canada

William Gimpel, Ph.D., Maryland Department of Agriculture

The following technical specialists reviewed the petition (Appendix 2).

Alfred F. Cofrancesco, Ph.D., U.S. Army Corps of Engineers

Jack R. Coulson, B.S., USDA, Agricultural Research Service

Peter Harris, Ph.D., Agriculture Canada

James Krysan, Ph.D., USDA, Agricultural Research Service

Dale Meyerdirk, Ph.D., Biological Control Operations, PPQ.

Matthew C. Perry, Ph.D., U.S. Department of the Interior

Janine Powell, Ph.D., USDA, Forest Service

James G. Saulman, Ph.D., U.S. Environmental Protection Agency

A. C. Schmidt, M.S., Agriculture Canada

David Sisneros, Ph.D., U.S. Bureau of Reclamation

Louis H. Waters, Ph.D., U.S. Department of the Interior

B. D. Wright, Ph.D., National Plant Board and Oregon Department of Agriculture.

VIII. Appendices

Appendix 1: Copy of permit application: importation from Europe of *Nanophyes marmoratus* and *N. brevis* into quarantine in the United States (Maryland), with subsequent release in Maryland, submitted by R. A. Malecki

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U.S. DEPARTMENT OF AGRICULTURE
ANIMAL AND PLANT HEALTH INSPECTION SERVICE
PLANT PROTECTION AND QUARANTINE
BIOLOGICAL ASSESSMENT AND TAXONOMIC SUPPORT
HYATTSVILLE, MARYLAND 20782

APPLICATION AND PERMIT TO MOVE LIVE PLANT PESTS AND NOXIOUS WEEDS

SECTION A - TO BE COMPLETED BY THE APPLICANT

1. NAME, TITLE, AND ADDRESS (Include Zip Code) 942640

Rich Malecki
NY Coop. Fish & Wildlife Research Unit
Fernow Hall - Cornell University
Ithaca, NY 14853

2. TELEPHONE NO. 607-255-2836 FAX

3. TYPE OF PEST TO BE MOVED ***Biological control agents
☒*** Arthropods ☐ Noxious Weeds ☐ Pathogens ☐ Other (Specify)

A. SCIENTIFIC NAMES OF PESTS TO BE MOVED	B. CLASSIFICATION (Orders, Families, Races, or Strains)	C. LIFE STAGES IF APPLICABLE	D. NUMBER OF SPECIMENS OR UNITS	E. SHIPPED FROM (Country or State)	F. ARE PESTS ESTABLISHED IN U.S.	G. MAJOR HOST(S) OF THE PEST
4. Nanophyes marmoratus	Col.:	Adults	50,000	Europe	No	Lythrum salicaria
5. Nanophyes brevis	Curculionidae	Adults	50,000	Europe	No	Lythrum salicaria
6.						

7. WHAT HOST MATERIALS WILL ACCOMPANY WHICH PESTS (Indicate by line number)
leaves

8. DESTINATION: Maryland
9. PORT OF ARRIVAL: Washington International Airport
10. APPROXIMATE DATE OF ARRIVAL OR INTERSTATE MOVEMENT: June/July 1994
11. NO. OF SHIPMENTS: 3
12. SUPPLIER: R.A. Malecki
13. METHOD OF SHIPMENT: ☒ Air Mail ☐ Air Freight ☐ Baggage ☐ Auto

14. INTENDED USE (Be specific, attach outline of intended research)
Biological control of Lythrum salicaria (purple loosestrife)

15. METHODS TO BE USED TO PREVENT PLANT PEST ESCAPE
sealed escape- proof containers

16. METHOD OF FINAL DISPOSITION
field release

17. Applicant must be a resident of the U.S.A. I/We agree to comply with the safeguards printed on the reverse of this form, and understand that a permit may be subject to other conditions specified in Sections B and C.
SIGNATURE OF APPLICANT (Must be person named in Item 1): R. A. Malecki
18. DATE: 4/12/94

SECTION B - TO BE COMPLETED BY STATE OFFICIAL

19. RECOMMENDATION: ☒ Approve ☐ Disapprove ☐ Accept USDA Decision
20. CONDITIONS RECOMMENDED: Voucher specimens to be deposited with Maryland Department of Agriculture
21. SIGNATURE: C. Ditmars
22. TITLE: Entomologist
STATE: MD
23. DATE: 4 May 1994

SECTION C - TO BE COMPLETED BY FEDERAL OFFICIAL

PERMIT

24. PERMIT NO. 942640

(Permit not valid unless signed by an authorized official of the Animal and Plant Health Inspection Service)

Under authority of the Federal Plant Pest Act of May 23, 1957 or the Federal Noxious Weed Act of 1974, permission is hereby granted to the applicant named above to move the pests described, except as deleted, subject to the conditions stated on, or attached to this application. (See standard conditions on reverse side).

See Supplemental Permit Conditions

on Reverse

CC: MD NER Coulson

This permit does not authorize the introduction, importation, interstate movement, or release into the environment of any genetically engineered organisms or products

25. SIGNATURE OF PLANT PROTECTION AND QUARANTINE OFFICIAL	26. DATE	27. LABELS ISSUED	28. VALID UNTIL	29. PEST CATEGORY
M. J. Fuchs	6/1/94	5 PPQ 599	Revoked	B2

PQ FORM 526 (OCT 88) Previous edition may be used.

Appendix 1

STANDARD SAFEGUARDS OF PERMIT

1. All pests must be shipped in sturdy, escape-proof containers.

2. Upon receipt of pests, all packing material and shipping containers shall be sterilized or destroyed immediately after removing pests.

3. Pests shall be kept only within the laboratory or designated area at the permittee's address.

4. No living pests kept under this permit shall be removed from confined area except by prior approval from State and Federal regulatory officials.

5. Without prior notice and during reasonable hours, authorized PPQ and State regulatory officials shall be allowed to inspect the conditions under which the pests are kept.

6. All pests kept under this permit shall be destroyed at the completion of the intended use, and not later than the expiration date, unless an extension is granted by this issuing office.

7. All necessary precautions must be taken to prevent escape of pests. In the event of an escape, notify this office.

Public reporting burden for this collection of information is estimated to average 0.17 hours per response, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing and reviewing the collection of information. Send comments regarding this burden estimate or any other aspect of this collection of information, including suggestions for reducing this burden, to Department of Agriculture, Clearance Officer, OIRM, Room 404-W, Washington, D.C. 20250; and to the Office of Information and Regulatory Affairs, Office of Management and Budget, Washington, D.C. 20503

SUPPLEMENTAL PERMIT CONDITIONS (Section C, PPQ Form 526)

Permit Number: 942640
Permittee: Malecki, Rich
Organism: *Nanophyes brevis* Boh., *N. marmoratus* Goeze (Coleoptera: Curculionidae).
State: MD

1. Note conditions specified by Maryland officials.

2. This permit authorizes release into the environment as per the Environmental Assessment dated May 1994 and the Finding of No Significant Impact dated June 1, 1994.

3. The applicant will provide to the Organism Permitting and Risk Analysis (OPRA) branch of Biological Assessment and Taxonomic Support (BATS, PPQ, APHIS, USDA) a list of counties where the permitted organism was actually released into the environment.

Project Title: Biological Control of Purple Loosestrife (*Lythrum salicaria* L.)

Proposed Action: Introduction and field release of *Nanophyes marmoratus* and *N. brevis*, two additional control agents of purple loosestrife.

Date: June 1993

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1 Abstract

Purple loosestrife (*Lythrum salicaria*) is a herbaceous perennial of European origin that has become a serious weed in marshes and alluvial wetlands over much of the temperate parts of the United States and Canada. Dense monospecific stands of loosestrife replace the more diverse native North American flora resulting in the degradation of these wetland habitats. There are presently no satisfactory physical, mechanical, or chemical means of controlling this plant. Biological control offers the most promising method of resolving this problem.

A root-mining weevil (*Hylobius transversovittatus*) and two leaf-feeding chrysomelids (*Galerucella calmariensis* and *G. pusilla*) from Europe were released in North America in 1992. Two additional flower- and seedfeeding species (*Nanophyes marmoratus* and *N. brevis*) are proposed for introduction in 1994. Both species were studied and screened between 1990 and 1992. They have a wide geographic and ecological range, and live monospecific on *L. salicaria* within its native European range. Their introduction is expected to further reduce seed output and lessen the competitive ability of purple loosestrife.

We are confident that both species can be established throughout the range of purple loosestrife in North America. We are also confident that these species will have no detrimental impact on native, agricultural, or ornamental plant species.

2 Introduction

Purple loosestrife (*Lythrum salicaria* L.) (Lythraceae) is a herbaceous perennial of European origin that has become naturalized in North America. It arrived during the early 1800s in ship ballast and was also introduced as a herb and ornamental (Stuckey 1980). The plant has since become a serious weed in marshes and alluvial wetlands (Thompson et al. 1987). The main problem areas occur in the northeast and north-central portions of the United States and in parts of southern Canada. Recently it's spread into the arid Midwest and West along major rivers and irrigation systems is causing concern. Where thoroughly established, *L. salicaria* tends to form dense

monospecific stands that displace the more diversified native flora (Malecki and Rawinski 1985).

Profuse seed production and a high rate of germination makes purple loosestrife an aggressive invasive species. Once established, mature plants develop large rootstocks that function as storage organs. These rootstocks provide resources for regrowth after the aboveground shoots are cut, burned or killed by application of foliar chemical herbicides. There is currently no satisfactory means of controlling purple loosestrife (see Thompson et al. 1987). Herbicides used most often against purple loosestrife are glyphosate (Rodeo), 2,4-D, and, on an experimental basis, triclopyr (Garlon 3A). These are non-specific herbicides and are disruptive to sensitive aquatic or semi-aquatic habitats, expensive and do not provide control over a prolonged period of time (Skinner et al. 1993).

The target weed is not attacked to any noticeable degree by native North American phytophagous insects or plant pathogens (Hight 1990). This gives it a competitive advantage over native wetland plants. The aim of biological control is to counterbalance the present competitive advantage of this plant by the establishment of several effective host specific herbivores.

Following the identification and host range screening of three biological control agents of purple loosestrife, i.e. the weevil *Hylobius transversovittatus* (Goeze) and the two chrysomelids *Galerucella calmariensis* (L.) and *G. pusilla* (Duft.), a petition was drafted for their introduction and release into North America (see Malecki et al. 1991). The three agents were released in summer 1992 in seven states (Virginia, Maryland, Pennsylvania, New York, Minnesota, Washington, Oregon). To date, observations in Washington, Oregon, Minnesota, Pennsylvania and New York have demonstrated that they survived the winter and started to mate and oviposit.

Two additional weevil species *Nanophyes marmoratus* (Goeze) and *N. brevis* (Boh.) were selected and investigated during the period 1990-1992 (Blossey and Schroeder 1992). Both of these species have a considerable impact on seed production of *L. salicaria* and are expected to complement the impact of the leaf-feeding and root-boring beetle species first released in 1992. *N. marmoratus* occurs throughout the entire range of purple loosestrife, while the

distribution of *N. brevis* seems restricted to areas south of the 50th parallel. Because of the ecological similarity between loosestrife infested areas in North America and large parts of the plant's native range in Europe (Blossey et al. 1993), it is expected that both species will have little difficulty becoming established.

3 Purpose and Need

The distribution and establishment of new *L. salicaria* populations occurs almost entirely by the spread of seeds. The three root- and leaf-feeding beetles released in 1992 are expected to considerably reduce plant vigor and survival. This will also affect flowering and seed output. However, even at high insect population levels a certain proportion of plants will still produce seeds. These seeds will build up the existing seed bank, and provide a continuous source for new recruitment. Release and establishment of the two *Nanophyes* species is expected to minimize this additional source of seed.

Both, *N. marmoratus* and *N. brevis*, occupy a well defined niche on the flower spike of *L. salicaria*. Adults and larvae of *N. marmoratus* feed on the closed flower buds. Following attack, these buds are generally aborted. In Europe, under high rates of larval parasitism, *N. marmoratus* maintained an average attack rate of about 30%. The closely related species *N. brevis* attacks seeds in flowers which escape attack by *N. marmoratus*. *N. brevis* occurs in similar or even higher densities in Europe. Since competition between the two species is minimal, their effect on seed output is often additive.

For purple loosestrife, we expect that the establishment of the three already released control agents will reduce the density of the plant to about 10% of its above ground biomass over 90% of the area actually infested. This optimistic estimate is based on observations within the native European range of *L. salicaria* where it normally occurs in small scattered stands. Large dense populations in Europe are rare exceptions and do not persist. The release and establishment of the two flower-feeding species is expected to reduce markedly the seed-production of *L. salicaria* plants escaping from attack by the root- and leaf-feeding species.

Successful biological control of *L. salicaria* in North American wetlands will increase the diversity of these habitats and drastically reduce or eliminate the need for chemical and mechanical control measures. An analysis by Thompson et al. (1987) of the benefits and costs of biological control of purple loosestrife produced a ratio of 1:27, i.e. an estimated 27 units of benefits for each unit of cost.

3.1 Description of program

A national program for the biological control of purple loosestrife is presently being implemented by the U.S. Fish and Wildlife Service (Malecki et al. 1993). Much of the coordination and funding comes from this agency. However, extensive participation and input is received from scientists associated with the USDA Agricultural Research Service, the U.S. Army Corps of Engineers, Agriculture Canada, the International Institute of Biological Control, and a number of state organizations and academic institutions. These scientists constitute a "Working Group for the Biological Control of *Lythrum salicaria*" which serves to assist in the coordination of activities, the production of comparable results, and the reduction in costly duplication of research. All research, monitoring and management of the three species introduced in 1992 and the two species addressed in this petition are subject to review and approval by this group.

Procedures for the introduction and release of *Nanophyes marmoratus* and *N. brevis* are as follows:

In spring 1994 hibernated adults of both species will be field collected on sites from southern Europe to Finland. The collection of adults is favored because they are not attacked by parasitoids. Their subsequent release during the oviposition period should facilitate establishment in North America. Likewise, insects collected at climatically different sites should increase the likelihood of establishment throughout the current range of purple loosestrife in North America. The collections will be carried out in cooperation with the International Institute of Biological Control, Delémont, Switzerland and coordinated by R. Malecki, U.S. Fish and Wildlife Service.

Insects will be shipped to a quarantine facility in Annapolis, MD where beetles and shipping material will be checked for diseases and other contaminants. From quarantine, both species will be released into field

cages for brief periods to prevent their immediate spread from the release site. Established links to participants and sites in 7 states (Pennsylvania, Maryland, New York, Minnesota, Oregon, Washington, Montana) where release of *H. transversovittatus*, *G. calmariensis* and *G. pusilla* has occurred will be used to propagate and establish *N. marmoratus* and *N. brevis*.

4 Description of Target Organism

4.1 Taxonomy

Family: Lythraceae
Subfamily: Lythraceae
(7 genera -*Ammannia*, *Rotala*, *Peplis*, *Lythrum*, *Cuphea*, *Diplusodon*, *Lafoensa*).
Genus: *Lythrum* (30 species)
Species: *L. salicaria* L.
Synonyms: *L. palustre* Salisb., *L. spicatum* S.F. Gray, *L. spiciforme* Dulac)

The taxonomy of *L. salicaria* is discussed in detail by Thompson et al. (1987). The main facts are quoted below.

Lythrum is the type genus of the family Lythraceae with some 500 species in 22 genera worldwide. Representatives include herbs, shrubs and trees that reach their greatest abundance in the American tropics where two of the largest genera (*Cuphea* and *Diplusodon*) are endemic (Gleason 1963). Several of the genera include species that are cultivated as ornamentals, but none are food or fiber producers. About 35 species of *Lythrum* occur throughout the world, of which 12 species are found in the continental United States (Shinners 1953). Three of these species, *L. hyssopifolia* L., *L. virgatum* L. and *L. salicaria*, are introductions from Europe. The status of *L. virgatum* is unclear, it is most likely a special form of *L. salicaria* (Anderson and Ascher 1993, Skinner et al. 1993).

Hultén (1971) referred to varying chromosome numbers of 30, 50 and 60 for *L. salicaria* occurring throughout the world. In a recent paper summarizing information concerning purple loosestrife, Mal et al. (1992) state that the normal chromosome number of purple loosestrife

is 60 and that other numbers are suspect. Thompson et al. (1987) examined specimens of *L. salicaria* from nearly all sectors of its continental range in North America and found that, morphologically, they were fairly uniform. Although they recognize the variability of *L. salicaria*, they believe that the North American target populations belong to a single well defined species. This assumption is supported by host acceptance tests performed in Europe with purple loosestrife obtained from Washington, Minnesota, Maryland and Washington; all plants supported the normal development of the proposed insect control agents

4.2 Distribution

L. salicaria has European and Asian distribution centers. The European segment is largest and extends from Great Britain across Europe to the central USSR. Its northern limit is near the 65th parallel (Tutin et al. 1968). *L. salicaria* is common throughout central and southern Europe and along the coastal fringe of the Mediterranean basin. In Asia, Hultén (1971) indicated that the main islands of Japan are the core of the species' native range, with outlying populations extending from the Amur River south across the lowlands of Manchuria and China to southeast Asia and India.

As an invading species, *L. salicaria* is found in eastern Africa (Ethiopia), Australia, Tasmania, New Zealand, the U.S.A. and southern Canada. The distributional history of *L. salicaria* in North America is described in detail by Stuckey (1980) and Thompson et al. (1987).

4.3 Life history

The biology and life history of *L. salicaria* is discussed in detail by Thompson et al. (1987). Therefore only a short summary is given.

Depending on site conditions, mature plants of *L. salicaria* are between 0.5 and 3 m high, with a maximum of 30-50 shoots emerging from a common rootstock. In Europe, in the presence of several specialized herbivores, plants are generally below 2 m high and produce a maximum of 5-10 shoots.

Seed production varies with plant size and age. Large plants can produce up to 1,000 capsules per stem with 90-120 seeds per capsule.

Seeds remain viable for many years and germination can occur on a variety of substrates with a wide range of pH.

Successfu! seedling establishment only occurs on moist soil, mainly in late spring or early summer when temperatures are high (Shamsi & Whitehead 1974). *L. salicaria* is found in a great variety of sites, from rock crevasses to gravel, sand, clay and organic soils, both in Europe and North America. Moisture is the most important factor for growth and reproduction, but well established plants can persist at dry exposed sites for many years.

4.4 Related ornamental and native plants

There are no related food crop or fiber plants, but *L. salicaria* itself has been and, to a certain extent, is still being used as an ornamental garden plant in Canada and the United States. This is because of its profuse and attractive floral display and the ease with which it is grown. Several large nurseries have developed their own horticultural stocks, including strains claimed to be infertile. However, studies by Anderson and Ascher (1993) found that cultivars are highly fertile when used as male or female parents with wild purple loosestrife, native species, or other cultivars. These cultivars grown in gardens are a major factor for the continued spread of purple loosestrife. As stated by Thompson et al. (1987), the economic value of *L. salicaria* as an ornamental is very small and is far outweighed by the damage caused to wetlands.

Twelve species of *Lythrum* are recognized in the continental United States, three of which are naturalized species of European origin (*L. hyssopifolia, L. virgatum* and *L. salicaria*). Only three of the nine native species, *L. alatum, L. lanceolatum* and *L. lineare* L., occur in the central and northeastern United States and southern Canada (Fernald 1950). None of the North American species in the genus *Lythrum* are listed as endangered or threatened species under the US Fish and Wildlife Endangered Species Act.

Besides the native *Lythrum* species, there are four other genera in the family Lythraceae which are an important component of the marsh and wetland biomes in North America, i.e. the genera *Ammania, Cuphea, Decodon* and *Rotala*.

Appendix 2

Scientific classification:
see TAG petition p5

General life history:
see TAG petition p6 life history

Pest status:
noxious weed in 13 states
see BioScience paper (Biological control of purple loosestrife, November 1993, p 681: "Current Problem")

4. Biology of organisms to be released:

Common name:
no common names

Scientific classification:
see TAG petition p8 (*N. marmoratus*) and p11 (*N. brevis*)

Taxonomic specialists and voucher specimens:
see TAG petition p8 (*N. marmoratus*) and p11 (*N. brevis*)

Natural geographic range:
throughout Europe and Siberia (*N. marmoratus*)
south and central Europe; in Germany as far north as the river Main (*N. brevis*)

Location where organisms were originally collected:
Adult weevils will be collected on sites from southern France to Finland. Source populations will be kept separately and released to study the potential effect of climatic preadaptations on their establishment and control success

General life history:
see TAG petition p8 (*N. marmoratus*) and p11 (*N. brevis*)

Dispersal capabilities:

N. marmoratus has good dispersal capabilities. The species colonizes new sites of purple loosestrife in their first or second growing season and was found on almost all sites studied in Europe by scientists of the International Institute of Biological Control.

N. brevis was present and abundant on most purple loosestrife sites studied within its range in southern Europe. This indicates a good dispersal capability.

Host range - field:

The only field host for *N. marmoratus* and *N. brevis* is purple loosestrife

Host range - laboratory:

The only laboratory host for both species was purple loosestrife (see TAG petition p10, Host range screening and Table 2 (*N. marmoratus*) and p13, Host range screening and Table 3 (*N. brevis*)

Short list of references on organisms:

Alonso-Zarazaga, M. A. (1989). Revision of the supraspecific taxa in the Palaearctic Apionidae Schoenherr, 1823. 1. Introduction and subfamily Nanophyinae Seidlitz, 1891. Fragm. Entomol., Roma, 21 (2), 205-262.

Dieckmann, L. (1963). Die mitteleuropäischen Arten der Gattung *Nanophyes* Schönh. nebst einer neuen Art aus Bulgarien. Reichenbachia Dresden 1, 169-194.

List of known parasites:

see TAG petition p10, Mortality factors (*N. marmoratus*) and p13, Mortality factors (*N. brevis*)

The organisms to be released are phytophagous insects and do not act as hyperparasites.

5. Expected attainable range in North America

Throughout the distribution range of purple loosestrife. We have some reservation about the potential for *N. brevis* to colonize the northern part of this range.

The only other plant in the family Lythraceae, *Lagerstroemia indica* L. (crape-myrtle) is an ornamental shrub or tree introduced from Asia.

5 Description of the Organisms for Release

5.1 *Nanophyes marmoratus*

5.1.1 Taxonomy

Order:	Coleoptera
Family:	Curculionidae
Subfamily:	Nanophyinae
Tribe:	Nanophyini
Genus:	Nanophyes Schoenherr 1838
Species:	N. marmoratus (Goeze 1777)
Synonyms:	Rhynchaenus lythri auctt., Curculio lythri Fabricius

Identification of the species was confirmed by the late Dr. L. Dieckmann, Department of Insect Taxonomy, Institute for Plant Protection, Eberswalde, Germany. Voucher specimens will be deposited in the collection of the International Institute of Entomology, MNH London, and the collection of the USDA Systematic Entomology Laboratory, Beltsville, MD, USA.

The subfamily Nanophyinae presently includes 18 genera and some 276 species, all associated with Lythraceae and distributed in all regions, but poorly represented in the New World (4 species). In the Palearctic region it is represented by 4 genera, 1 subgenus and 87 species (Alonso-Zarazaga 1989). According to Dieckmann (1963), the genus *Nanophyes* in Europe consists of 13 species.

5.1.2 Life history

After hibernation, adults in northwestern Germany appear on *Lythrum salicaria* during the second half of May. The beetles start feeding on the youngest leaves in the shoot tips. However, this is of little significance to the plant except where beetles occur in large numbers.

As soon as flower buds develop, adult beetles move to the upper parts of the flower spikes where they copulate and feed on the receptacle and the ovaries within closed flower buds. The attacked buds are later aborted.

Oviposition starts at the end of June and extends into September. The relatively large eggs are deposited into the tip of young flower buds before the petals are fully developed. Generally, only a single egg is deposited per bud through a hole chewed by the female. Attacked buds remain closed and do not flower. Observations indicate that females lay 1-2 eggs per day, i.e. some 60-100 eggs in total. The main oviposition period is during July and August.

The larvae first consume the stamens, and in most cases the petals, and then eat the ovary. Only a single larva can complete development per flower bud. Mature larvae form a pupation chamber from their frass at the bottom of the bud. The new generation beetles appear mainly in August and feed on the remaining green leaves of purple loosestrife before finding overwintering sites. Some remain on the plants into October. The complete development from egg to adult takes about one month. There is one generation per year. The species feeds and develops exclusively on *L. salicaria*.

5.1.3 Distribution

Throughout Europe and western Siberia (Dieckmann 1963). With few exceptions, *N. marmoratus* was abundant and found at all sites visited in northern and central Europe. The species tolerates a wide range of environmental conditions and has an excellent host finding capacity. Adults were present on isolated sites with single plants.

5.1.4 Impact on host plant

The total impact of *N. marmoratus* was difficult to determine. Flower buds abort due to adult and larval feeding. To collect data on flower bud abortion and its causes, flower spikes were beaten over a net and the flower buds that dropped were collected for inspection. These collections were made in August at a locality near Darmstadt, German Rhine Valley, where both species of *Nanophyes* occurred.

An evaluation of the flower buds in six subsamples, each with 100 buds, revealed that an average of 23±4.9 (SD) were suitable for oviposition by *N. marmoratus*, while 77±4.9 (SD) buds were too small. In 10 additional subsamples each with 60 flower buds suitable for oviposition by *N. marmoratus* about one fifth of the buds contained various developmental stages of *N. marmoratus*. (Table 1) The frequency of stages was as follows: eggs 2.9%, first instar larvae 5.9%, intermediate larval instars 26.5%, mature larvae 35.3% and pupae

Appendix 2

29.4%. These results indicate that the majority of the buds dropped as soon as larval feeding had severely damaged the vascular bundles.

Observations indicate that a large proportion of small buds drop prematurely because of adult feeding by *N. marmoratus* at the top of the flower spikes. In this way the total impact of *N. marmoratus* on seed production of *L. salicaria* is considerably increased.

Average larval attack rates in buds not aborted was approximately 30%. Based on field observations and dissections of flower buds the total reduction of seed output by *N. marmoratus* is estimated at about 50%. We speculate that the total impact of *N. marmoratus* on seed production of *L. salicaria* in North America could be relatively higher since inflorescences on North American plants are less dense and the buds are more accessible for oviposition.

5.1.5 Mortality factors

In Europe, nine species of ecto-parasitoids of prepupae and pupae were found. All of them belong to the Chalcidoidea (i.e to the genera *Aprostecetus*, *Pteromalus*, *Trichomalus*, *Eutetrastichus* and *Habrocytus*). Only *Trichomalus pexatus* Wlk. (Hym., Torymidae) and the polyphagous hyperparasitoid *Eupelmus urozonus* Dalm. have been identified at species level. Further species identification is pending.

Rates of parasitism vary considerably with time, from plant to plant and site to site, but regularly attain high levels of 60-90%.

5.1.6. Field host plants

Throughout its known European distribution, *N. marmoratus* feeds and develops exclusively on *L. salicaria*.

5.1.7. Host range screening

The host range of all known species of *Nanophyes* is restricted to plant species in the family Lythraceae. To provide evidence for this restriction we included 28 plant species of another 15 plant families in our screening program (Table 2). Larval transfer tests were not carried out because normal larval development occurs in undamaged and still developing flower buds. Ovipositing females are able to oviposit without causing immediate bud drop. However, experimental larval or egg transfers failed due to the damage caused to plant tissue and subsequent bud abortion.

The beetles used for the screening tests were field collected in northern Germany (near Hamburg and Kiel) in May. Oogenesis only occurs after adults have fed on flower buds of *L. salicaria*. Therefore adult feeding tests were carried out in glass tubes as no-choice tests containing cut leaves as well as flower- and leaf-buds. Multiple choice oviposition tests were carried out in gauze cages (3x2x2m) in the garden with potted test plants.

Adult feeding tests showed that normal feeding on flower buds was restricted to *L. salicaria* (Table 2). A lesser amount of feeding was observed on *L. hyssopifolia* and nibbling occurred on another four species in the genus *Lythrum*, and on *Rhexia mariana*, *Epilobium angustifolium*, and *Rumex crispus*. All other plants offered were refused. Additional field cage observations revealed that newly emerged beetles feed on *L. hyssopifolia* toward the end of the field season after *L. salicaria* plants have dried up.

Oviposition and complete larval development was restricted to *L. salicaria* (Table 2).

5.2 *Nanophyes brevis*

5.2.1 Taxonomy

Order:	Coleoptera
Family:	Curculionidae
Subfamily:	Nanophyinae
Tribe:	Nanophyini
Genus:	Nanophyes Schoenherr 1838
Species:	N. brevis Boh. 1845
Synonyms:	none

Identification of the species was confirmed by the late Dr. L. Dieckmann, Department of Insect Taxonomy, Institute for Plant Protection, Eberswalde, Germany. Voucher specimens will be deposited in the collection of the International Institute of Entomology, MNH London, and the collection of the USDA Systematic Entomology Laboratory, Beltsville, MD, USA.

5.2.2 Life history

After hibernation, the adults appear together with those of *N. marmoratus* on young shoots of *L. salicaria* and feed on the youngest

leaves. The two species separate as soon as the flower spikes develop. *N. marmoratus* occupies the upper part of the flower spike with unopened flower buds, while *N. brevis* concentrates in the lower part with opened flowers. Here *N. brevis* feeds exclusively on the receptacles. Oviposition is restricted to fertilized flowers and the eggs are laid directly into the ovary. The larvae feed on the developing seeds within the inflated ovaries. Normally, only a single larva is found per ovary. Complete larval development takes about seven weeks. Pupation occurs within the ovary. Fully developed beetles leave the ovaries through a hole which they gnaw. The main emergence period is August and September, but beetles can still be found on their host plant in late October.

In Europe, *N. brevis* occurs as far north as the river Main in central Germany. In areas where both *Nanophyes* species occur, direct larval competition is completely avoided. *N. marmoratus* oviposits into closed flower buds and *N. brevis* into fertilized flowers. Larvae of both species never occur together in the same bud because they complete their development at the site of oviposition.

5.2.3 Distribution

Throughout central and southern Europe including the entire European Mediterranean and Asia Minor. Also recorded in Egypt (Dieckmann 1963).

5.2.4 Impact on host plant

N. brevis supplements the effect of *N. marmoratus* by destroying seeds in flowers which escaped *N. marmoratus* attack in the bud stage. Unfortunately, not all seeds of attacked ovaries are consumed. Dissections indicate an average seed reduction of 50%. Attack by *N. brevis* has been checked at six localities in Europe. It was found that attack rates vary considerably between individual plants. Mean attack rates were around 30%. At southern localities *N. brevis* seems to be the dominant seed-eating species of *L. salicaria*.

As shown in Table 1, 42.6% of the buds dropping from beaten flower spikes showed feeding damage by adult *N. brevis*. The receptacle had been partially or completely consumed. Thus, adult feeding on the receptacle of opened flower buds (i.e. those which escaped attack by *N. marmoratus*) considerably add to total seed destruction. It should be mentioned that in contrast to buds attacked by *N. marmoratus*, flower

buds containing developmental stages of *N. brevis* remain attached to the flower spike. It is expected that *N. brevis* could attain higher rates of attack in North America in the absence of its parasitoids.

5.2.5 Mortality factors

N. brevis is attacked by the same parasitoids as *N. marmoratus* and observations reported under 5.1.5 can be extended to *N. brevis*. Dissections from different sites revealed over 80% parasitism.

5.2.6 Field host plants

N. brevis feeds and develops exclusively on *L. salicaria* throughout its known European distribution.

5.2.7 Host range screening

Adult feeding and oviposition tests with *N. brevis* were carried out under exactly the same conditions as described for *N. marmoratus*. The same restrictions to experimental larval transfer tests as discussed for *N. marmoratus* apply. The beetles used for the screening tests were field collected in the southern German Rhine Valley (near Offenburg and Darmstadt) after hibernation in spring.

Adult feeding tests showed that normal feeding was restricted to *L. salicaria* (Table 3). Moderate feeding occurred on *L. hyssopifolia* and some nibbling was found on *L. alatum*, *Decodon verticillatus* and *Ludwigia alternifolia*. All other species were refused.

Oviposition and complete larval development was restricted to *L. salicaria* (Table 3).

6 Environmental Consequences of Release

6.1 General

There are at present no satisfactory means of effective long-term control of *L. salicaria*. The measures presently in use to lessen competition from *L. salicaria* in selected areas are expensive, disruptive for the flora and fauna of the treated areas, and their effect is not persistent. Therefore, all state and federal agencies involved in the program (USDA, USDI, the Army Corps of Engineers, etc.) set a high priority on the biological control of *L. salicaria*.

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All *Lythrum* species have attractive flowers and several species, including the target species, are grown as ornamentals. A considerable reduction in the abundance of purple loosestrife flowers in marshes and wetlands is a price that must be paid for biological control. The expected end result is restoration of the natural floral diversity within the areas presently dominated by the invasive *L. salicaria*.

6.2 Physical environment

6.2.1 Air

N. marmoratus and *N. brevis* are phytophagous insects that will have no impact on air quality.

6.2.2 Water

N. marmoratus and *N. brevis* by themselves will have no impact on water quality.

6.2.3 Land

There are no satisfactory chemical or mechanical methods to control purple loosestrife in marshes and alluvial wetlands where it occurs. Consequently *L. salicaria* is displacing the native vegetation in these habitats. The reduction of purple loosestrife by biological control will have a positive effect on both the native flora and fauna of infested wetlands. It will also provide a control option that is consistent with the conservation of a valuable and ecologically important biome.

6.3 Human health

N. marmoratus and *N. brevis* will have no impact on human health.

7 Ecological Relationship

7.1 Wildlife

Except for its nectar, purple loosestrife is largely unused by native wildlife or insects. On the contrary, *L. salicaria* is an aggressive invasive weed which is forming dense uniform stands and replacing a diversified native flora, including important waterfowl food plants. The loss in quality of these habitats as waterfowl production sites and as refugia for a variety of associated wildlife species is well documen-

ted (Rawinski & Malecki 1984). Thus, biocontrol will be beneficial by opening the habitat to plants used by wildlife.

7.2 Endangered and threatened species

There are no endangered or threatened *Lythrum* species listed in the US Fish and Wildlife Endangered Species Act. No plants in the genus *Lythrum* are at risk from the introduction of *N. marmoratus* or *N. brevis* into the United States or Canada.

7.3 Non-target invertebrates

The proposed biocontrol agents are monospecific phytophagous insects restricted to the target weed. *L. salicaria* is not a host of any specialized North American insect species. The reduction of purple loosestrife density will be beneficial to native organisms due to the increase of native plant species.

7.4 Other biocontrol agents

The first three biocontrol agents of purple loosestrife, i.e. the root-feeding weevil *Hylobius transversovittatus* and the two leaf-feeding chrysomelids *Galerucella calmariensis* and *G. pusilla* were field released in the summer of 1992 in seven states in the U.S. It is expected that once firmly established these three species will drastically reduce the population density of *L. salicaria*, but will not be able to suppress seed production completely. Since new recruitment of *L. salicaria* depends primarily on seeds and since seeds remain viable in the seed bank for more than ten years, a reduction of seed output is of importance (Welling and Becker 1990).

Based on field observations in Europe, it is expected that the combined action of both agents proposed could reduce seed production of *L. salicaria* by 50-70%. Since the high rates of parasitism observed for both species in Europe are not likely to occur within the proposed release area, the total percentage of seed destruction will be higher.

7.5 Pollinators

Purple loosestrife is used by honey bees. However, an analysis by Thompson et al. (1987) of the ten leading honey-producing states in

1976 shows that *L. salicaria* is of minor importance as a nectar source. Since biological control of *L. salicaria* will only reduce but not completely eliminate purple loosestrife from the invaded areas, *L. salicaria* can still provide a source of pollen and nectar.

8 Cumulative Impacts

The establishment of *N. marmoratus* and *N. brevis* is expected to further increase the destruction of flower buds and seeds by primarily attacking those which have escaped adult and larval feeding of the two *Galerucella* species. Their effect is enhanced because each species occupies a well defined niche on the flower spike of *L. salicaria*. Therefore, their effect on seed reduction is additive. No adverse effects are seen from their introduction except for the further reduction in the abundance of the attractive purple loosestrife flowers. This is a consequence of any purple loosestrife control and not peculiar to these control agents.

9 Mitigative Measures

No measures to mitigate the effects of the biocontrol agents should be necessary or desirable. The population densities of specialized biocontrol agents are regulated by the abundance of their host plant. Thus, the populations of the control agents will decrease as their host plant populations decrease.

10 Conclusions

The European weevils *N. marmoratus* and *N. brevis* have a wide geographic range, and live monospecific on the target host plant. Therefore, native plants and ornamental species are not at risk and the two species proposed are expected to have an important negative impact on seed production of *L. salicaria*.

11 References

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Appendix 2

Table 1. The causes of *L. salicaria* bud dropping (%) at Groß Umstadt, Germany, August 15, 1991. Numbers are means±SD of 10 samples containing 60 buds each.

Larval attack of *N. marmoratus*	Adult feeding of *N. marmoratus*	Adult feeding of *N. brevis* in the receptacle	Unknown/ unattacked
21.5± 4.1	30±6	42.6±9.7	8.7±7.1

Appendix 2

Table 2 Host specificity screening of *Nanophyes marmoratus*

		Adult feeding No. of holes/replicate	Oviposition cage
	Exposure time	(2 pairs/3 days)	(several weeks)
A. Taxonomically Associated Plants			
1.	Lythrum salicaria L.	43/44/49/35/57	numerous
2.	L. lineare L.	2/6/1/2/0	-----
3.	L. alatum Pursh.	14/3/0/0/0	-----
4.	L. californicum Torr. & Gray	0/1/0/0/0	-----
5.	L. hyssopifolia L.	21/12/0/14/15	-----
6.	Decodon verticillatus (L.) Ell	-----	-----
7.	Rotala ramosior (L.) Koehne	-----	-----
8.	Ammania coccinea Rottb	-----	-----
9.	A. robusta Heer & Regel	-----	-----
10.	Cuphea viscosissima Jacq	-----	-----
11.	C. wrightii Gray	1/0/0/0/0	-----
12.	C. laminuligera Koehne	-----	-----
13.	C. lanceolata Alton	-----	-----
14.	C. lutea Rose	-----	-----
15.	Lagerstroemia indica L.	-----	
Punicaceae			
16.	Punica granatum L	-----	
Melastomataceae			
17.	Rhexia mariana L	0/1/0/0/0	
Onagraceae			
18.	Ludwigia alternifolia L	-----	-----
19.	Epilobium angustifolium L	1/1/0/4/3	-----
20.	Oenothera biennis L	-----	-----
21.	Gaura parviflora	-----	-----
22.	G. biennis	-----	-----
23.	Circaea quadrisulcata (L.) Hara	-----	---
B. Associated Wetland Plants of Wildlife Importance			
Typaceae			
24.	Typha latifolia L	-----	-----
Sparganiaceae			
25.	Sparganium eurycarpum Engelm.	-----	
Alismataceae			
26.	Sagittaria latifolia Willd	-----	
Poaceae			
27.	Zizania aquatica L	-----	

Appendix 2

Cyperaceae			
28.	Scirpus americanus Pers.	-----	-----
29.	S. acutus	-----	-----
30.	Carex comosa Bostt.	-----	-----
Salicaceae			
31.	Salix interior Rowlee	-----	
Polygonaceae			
32.	Rumex crispus L.	0/1/0/0/0	-----
33.	Polygonum coccineum Muhl.	-----	
Chenopodiaceae			
34.	Chenopodium.album L.	-----	-----
Ranunculaceae			
35.	Ranunculus sceleratus L.	---	-----
36.	R. bulbosus L.	-----	-----
C. Important Agricultural Plants			
Poaceae			
37.	Triticum aestivum L.	-----	-----
38.	Oryza sativa L.	-----	-----
39.	Zea mays L	-----	-----
Chenopodiaceae			
40.	Beta vulgaris L.	-----	
Fabaceae			
41.	Glycine max L.	-----	-----
Malvaceae			
42.	Gossypium hirsutum L.	-----	
Asteraceae			
43.	Helianthus annuus L.	-----	-----

- no feeding/oviposition

* each bar represents one replicate

Appendix 2

Table 3 Host specificity screening of *Nanophyes brevis*

		Adult feeding	Oviposition cage
	Exposure time	(2 pairs/3 days)	(several weeks)
A.	Taxonomically Associated Plants		
Lythraceae			
1.	Lythrum salicaria L.	++/++/++/++/++	numerous
2.	L. lineare L.	-----	-----
3.	L. alatum Pursh.	(+)/(+)/---	-----
4.	L. californicum Torr. & Gray	-----	-----
5.	L. hyssopifolia L.	+/+/+/+/+	-----
6.	Decodon verticillatus (L.) Ell	(+)/(+)/(+)/(+)/(+)	
7.	Rotala ramosior (L.) Koehne	-----	-----
8.	Ammania coccinea Rottb	-----	-----
9.	A. robusta Heer & Regel	-----	-----
10.	Cuphea viscosissima Jacq	-----	-----
11.	Lagerstroemia indica L.	-----	
Punicaceae			
12.	Punica granatum L	-----	
Onagraceae			
13.	Ludwigia alternifolia L	(+)/(+)/(+)--	-----
14.	Epilobium angustifolium L	-----	-----
15.	Oenothera biennis L	-----	-----
16.	Gaura parviflora	-----	-----
17.	G. biennis	-----	-----
18.	Circaea quadrisulcata (L.) Hara	-----	---
B.	**Associated Wetland Plants of Wildlife Importance**		
Typaceae			
19.	Typha latifolia L	-----	-----
Sparganiaceae			
20.	Sparganium eurycarpum Engelm.	-----	
Alismataceae			
21.	Sagittaria latifolia Willd	-----	
Poaceae			
22.	Zizania aquatica L	-----	
Cyperaceae			
23.	Scirpus acutus	-----	-----
24.	S. americanus Pers.	-----	-----
25.	Carex comosa Bostt.	-----	-----

Appendix 2

Salicaceae
26. Salix interior Rowlee -----

Polygonaceae
27. Rumex crispus L. ----- -----
28. Polygonum coccinеum Muhl. -----

Chenopodiaceae
29. Chenopodium.album L. ----- -----

Ranunculaceae
30. Ranunculus sceleratus L. ----- -----
31. R. bulbosus L. -----

C. Important Agricultural Plants

Poaceae
32. Triticum aestivum L. ----- -----
33. Oryza sativa L. ----- -----
34. Zea mays L. ----- -----

Chenopodiaceae
35. Beta vulgaris L. -----

Fabaceae
36. Glycine max L. ----- -----

Malvaceae
37. Gossypium hirsutum L. -----

Asteraceae
38. Helianthus annuus L. ----- -----

++ normal feeding
+ slight to moderate feeding
(+) nibbling
- no feeding/oviposition

* each bar represents one replicate

Appendix 2

INFORMATION REQUESTED FROM APPLICANTS IN SUPPORT OF PERMIT APPLICATION (USDA-APHIS-PPQ-BATS)

1. Brief description of the proposed action:

Introduction and field release into the U.S. of two European insect species, *Nanophyes marmoratus* and *N. brevis*, as additional control agents of the exotic wetland perennial purple loosestrife (*Lythrum salicaria*).

2. Detailed description of the proposed action

Purpose of the release:
see TAG petition p3

Need for the release:
see TAG petition p1 Introduction

Specific location of rearing facility and release:
see TAG petition p4 last three paragraphs

Number to be released:
depending on availability 500-1000/site/species

Timing:
June/July 1994

Methods to be used:
Insects will be shipped to cooperators and immediately released into sleeve or walk-in cages. Cages will be removed after 1-2 weeks to allow adults to spread.

3. Biology of target(host) organism(s)

Common name:
purple loosestrife

6. Expected environmental impacts

Human impacts:
none

Others (direct and indirect):
see TAG petition p13-16

Mitigative measures:
see TAG petition p16

Appendix 3

Supplementary information to Petition Number 93-05, release of *Nanophyes marmoratus* and *N. brevis* on *Lythrum salicaria*.

provided by

Richard A Malecki and Bernd Blossey

A. Comments by A. C. Schmidt

General: Media coverage and public interest

This petition is written for a review process of the TAG, not for the media. The public will not see this paper and the scientific community will be presented with all information provided to reviewers last year and this year. Screening results and environmental risk assessments for *Galerucella calmariensis*, *G. pusilla* and *H. transversovittatus* will appear in the next issue of "Weed Science". A paper covering the screening results of the flower feeding insects on purple loosestrife has been submitted to "Biological Control".

The risk assessment, especially for *Lythrum alatum* and *Decodon verticillatus* was covered in the petition to introduce the first three insects. It is available to the TAG members in case additional information is desired. We felt it need not be repeated here. (See our cover letter).

1. European host plants:

Contrary to Dr. Schmidt's suspicion concerning the lack of information on wetland plants and their associated phytophagous insects, the European insect fauna is very well known. Generally, the literature, especially for weevils, gives lists and dates of plant species from

Appendix 4

5. Introduction into North America

In response to this comment about differences in host specificity of different populations of the same insect species, we again reviewed the literature (BIOSIS) and the Proceedings of the last four International Symposia on the Biological Control of Weeds (Brisbane, Vancouver, Rome, Christchurch). We were unable to find any evidence that host specificity differs between populations occurring in different regions. On the contrary, some researchers recommend collecting from as many different populations as possible to increase genetic variability.

Attention is called to a paper by Harris (1991) who states that "without investigation, it is not certain that collections from other sites have the same host range as those approved for release". The cited example of *Rhinocyllus conicus*, does not prove his point. *R. conicus* was collected from a different host plant than the originally screened adults and host race formation is a common phenomenon in insects. Thus, it should be no surprise that the weevil attacked thistles in the genus *Cirsium* from which the species was collected. Host range tests were performed using beetles collected from *Silybum* sp. (Harris 1991).

Since we will collect *Nanophyes* spp. from the only host plant of both species, *L. salicaria*, there should be no difference in host specificity in any of the populations.

B. Comments by David Sisneros, Bureau of Reclamation

Risk to ornamental varieties of purple loosestrife

It is very likely that both *Nanophyes* species will attack ornamental varieties of purple loosestrife. Ornamental plantings are documented as a major source for the spread of purple loosestrife. The plant is on the noxious weed list of at least 13 states that prohibit its sale or distribution. Most likely, other states will follow this legislation.

C. Comments by Jack Coulson

Keys for species identification

Two papers (Alonso-Zarazaga 1989, Dieckmann 1963) have been published giving species identification and drawings of the main discriminating characters. We do not see the need to repeat this information in this petition.

Literature cited (other than in petition)

Harris, P. (1991). Screening classical weed biocontrol projects and agents. In *Biological Control Quarantine: Needs and Procedures.* (eds. Coulson, J. R. ; Soper, R. S. , and Williams D. W.) Pro-ceedings of a workshop sponsored by USDA-ARS. U. S. Department of Agriculture, Agriculture Research Service, ARS-99, pp 61-68.

NEW YORK COOPERATIVE FISH & WILDLIFE RESEARCH UNIT
FACSIMILE TRANSMISSION SHEET

DATE: May 26, 1994
TO: Mike Firko, USDA, APHIS
FAX:
FROM: Dr. Rich Malecki/ Dr. Bernd Blossey
FAX: 607 255 1895 **PHONE:**607 255 5314 **E-MAIL:**bb22@cornell.edu
REMARKS:

Pathogen-Parasitoid check of *Nanophyes marmoratus* and *N. brevis*, biological control agents to be introduced for control of purple loosestrife (*Lythrum salicaria* L.).

To insure the introduction of disease free weevils into the United States we implement the following procedures:

From each sampling area in Europe, subsamples will be examined for internal and external diseases, parasites and parasitoids. Samples of adult weevils will be shipped to specialists who have agreed to inspect the organisms before they are released from quarantine.

Dr Jean Adams
USDA-ARS
Insect Biocontrol Lab, Bldg. 011A
BARC-West
10300 Baltimore Ave.
Beltville, MD 20705-2350

has agreed to examine the insects for pathogenic viruses, bacteria and microsporidian diseases.

Appendix 5

Dr. Ralf-Udo Ehlers
Institut of Phytopathology
Universität Kiel
Kiel
Germany

will examine the insects for entomopathogenic nematodes.

Dr. Bernd Blossey
Department of Natural Resources
Fernow Hall, Cornell University
Ithaca, New York 14853

will examine individuals for the presence of external parasites or other diseases before they are shipped to the United States. Any questionable specimens will be immediately destroyed.

Dr. Blossey will also examine individuals from each sampling area for internal parasitoids. At least 20 individuals per collection area will be dissected under a dissecting microscope.

Appendix 5

Rec'd 8/2/93

MONTANA STATE UNIVERSITY
1893•CENTENNIAL•1993

Entomology Research Laboratory

Montana State University
Bozeman, Montana 59717

Telephone 406-994-3860
FAX 406-994-6029

July 26, 1993

Deborah Knott
USDA-APHIS-PPQ
Plant Pest Permit Section
RM. 625, Federal Bldg.
6505 Belcrest Road
Hyattsville, MD 20782

Dear Ms Knott:

Dr. Chuck Quimby with the USDA-ARS, Rangeland Weeds Laboratory has asked me to review the petition: "Introduction and field release of Nanophyes marmoratus and N. brevis, two additional control agents of purple loosestrife" submitted by Malecki et al. After reveiwing the petition, I feel that these two species are sufficiently host specific to warrant their introduction and release into North America. Although lesser amounts of feeding were observed on other test plants, it did not appear to be significant and, in my opinion, not of great concern. The introduction of these two species should compliment the potential damage caused by the three insect species that have been cleared for field release, and thus be of benefit.

Should you have any questions please contact me.

Sincerely,

Jeffrey L. Littlefield
Quarantine Officer/
Research Entomologist

cc. J. Coulson

93-05
Purple
Loosestrife

TAGIBCW #86 (93-05)

Nanophyes marmoratus (Goeze) and N. brevis Boheman (Col.: Curculionidae) — From Europe — For purple loosestrife (Lythrum salicaria L.)

Petition for importation of both species into quarantine in Maryland (Annapolis) for field release in the United States (initially Pennsylvania, Maryland, New York, Minnesota, Oregon, Washington, and Oregon) for biological control of purple loosestrife. Received in BCDC 6/16/93.

Copies sent by BCDC to J. Pakaluk, SEL (curculionid taxonomist) and P. C. Quimby (biocontrol specialist) for comment, 7/1/93.

Comments, as TAG member, sent to TAG Executive Secretary (Acting), 7//9/93.

Comments (no objections) from taxonomist received in BCDC 7/15/93; copy forwarded to TAG Exec. Secr. 7/19/83.

Comments by biocontrol specialist (J. L. Littlefield, MTSU) sent to Acting TAG Exec. Secr. 7/26/93; copy received in BCDC 8/2/93 (no concerns expressed).

Comments from Canadian Review Group sent directly to TAG Chairman, 9/16/93; copy received in BCDC 9/23/93.

Copies of all reviewer's responses (total of 10 from TAG with 9/16/93 concurring comments from Canada) and letter dated 11/24/93 from TAG Chair recommending release of both species, sent to M. H. Royer, Chief Operations Officer, BATS, PPQ, received in BCDC 12/3/93.

Environmental Assessment, with FONSI signed 6/1/94, received in BCDC 6/7/94.

State (New York, Oregon, Pennsylvania, Washington, Maryland) approvals for release of both species obtained 12/7/93-5/2/94; PPQ permits for release in those states issued 6/1-2/94; copies of permits received in BCDC 6/9/94.

Copy of Canadian comments on draft EA and of request for permits for release in Canada received in BCDC 6/20/94.

Request for TAG comments on discovery of nematodes in quarantine populations of beetles received from Europe received in BCDC from Ken Lakin, Chief, OPRA, BATS, PPQ, 7/6/94. Copy of request faxxed to Canada by BCDC 7/8/94. Response to Lakin as TAG member sent 7/8/94.

Comments from Canada received 7/13/94; filed, not circulated.

Stephen D. Hight
Department of Entomology
University of Maryland
1300 Symons Hall
College Park, Maryland 20742
301-504-3944 (PHONE)
301-314-9290 (FAX)

DATE: 28 June 1994

TO: Mike Fitzo
USDA-APHIS-PPQ-BATS

FAX NUMBER: 301-436-8700

NO. OF PAGES:

MESSAGE: I am sending you this FAX to update you on the shipment of Nanophyes marmoratus and N. brevis that were received last Saturday (25 June) from Europe. This shipment of 8,800 adults is being maintained at the MDA, Annapolis Quarantine Facility. As I mentioned on the phone, we have a problem with nematodes. The following information should be considered preliminary. The Europeans are continuing to work on identifying the nematode and the extent of infestation, and I am trying to line up some expertise in the states. I'll forward any new information to you once it is received.

Identification: Superfamily - Tylenchida
Species Group - Entaphelenchidae (Poinar, 1975)

Biology: group not pathogenic; egg and juvenile nematodes found, no adults.

Infestation: 100 N. marmoratus dissected and no confirmed nematodes found; 100 N. brevis dissected and all females had nematodes, no nematodes in males, female N. brevis had developing eggs, nematodes found in specimens from all collection sites (southern France and southern Germany).

cc: P. Tipping
B. Blossey & R. Malecki