

# FEDERAL AQUATIC NUISANCE SPECIES (ANS) RESEARCH RISK ANALYSIS PROTOCOL

## AQUATIC NUISANCE SPECIES TASK FORCE (ANSTF) November 30, 2010

### Introduction

This document is required by the Nonindigenous Aquatic Nuisance Prevention and Control Act of 1990 (“NANPCA, Public Law 101-646, 104 STAT. 4671, 16 U.S.C. 4701-4741), as amended by the National Invasive Species Act, 1996. Section 1202(f)(2) of NANPCA directs the Aquatic Nuisance Species Task Force (ANSTF) to establish a protocol “to ensure that research activities carried out under this subchapter do not result in the introduction of aquatic nuisance species to waters of the United States.”

Responsibility for actual use of this Protocol is specified in Section (f)(3): “The Task Force shall allocate funds authorized under this Act for competitive research grants to study all aspects of aquatic nuisance species, which shall be administered through the National Sea Grant College Program and the Cooperative Fishery and Wildlife Research Units. Grants shall be conditioned to ensure that any recipient of funds follows the protocol established under paragraph (2) of this subsection.”

Throughout this document, both the descriptors “nonindigenous” and/or “nuisance” are used when referring to aquatic species that are the target of this risk analysis protocol. Language used in NANPCA differentiates between a nonindigenous species and a nuisance species, with a “nonindigenous” label being solely based on the historic range of the species, while a “nuisance” designation is based on a species being both nonindigenous AND potentially harmful (“threatens the diversity or abundance of native species or the ecological stability of infested waters, or commercial, agricultural, aquacultural or recreational activities dependent on such waters” (NANPCA section 1003(1))). The ANSTF Research Committee adopted a precautionary approach by targeting this risk analysis to all aquatic nonindigenous species research, regardless of the “nuisance” designation. The intent of the procedures outlined herein is to minimize to the extent practicable the risk of release and spread of aquatic nonindigenous species into areas they do not yet inhabit, since any nonindigenous species may become a nuisance species. Not only is it often not possible to be sure that a species won’t become a nuisance (as defined) in the future, but the possession and/or release of nonindigenous species may be illegal under various federal, state or local laws which may or may not differentiate between nonindigenous and nuisance species.

## Background

This document (“the Protocol”) replaces the previously established “Protocol for Evaluating Research Proposals Concerning Aquatic Nonindigenous Species” (ANSTF, July 1994). It applies only to research involving aquatic nonindigenous species (ANS) and is designed to reduce the risk that research activities may cause introduction or spread of such aquatic species. Other potential means of introduction, such as bait movement, aquaria disposal, ballast water discharge, movement of recreational boats, movement of fishing gear, and horticultural sales, are not addressed here.

The original “Research Protocol,” adopted in draft form in 1992, was finalized and published by the ANSTF in July 1994. In 2008 the ANSTF requested the Research Committee (a Committee of the ANSTF) to evaluate and recommend revisions to the 1994 Protocol, as needed. According to the Society for Risk Analysis (SRA, <http://www.sra.org>), the elements or components of a “risk analysis” include risk assessment, risk characterization, risk communication, risk management, and policy relating to risk. This revised protocol incorporates three of those elements – it requires a risk assessment (Part I) and then, if needed, establishment and implementation of a risk management plan (Part II), with the combined results communicated to the funding agency as part of the proposal and funding process. Therefore this revised Protocol is renamed “Federal Aquatic Nuisance Species Research Risk Analysis Protocol.” It was adopted by the ANSTF [November 5, 2009].

This protocol supplements, but does not replace, other existing Federal guidelines established to control activities with specific major classes of organisms. This document does not eliminate or in any way affect requirements of the National Environmental Policy Act (NEPA; 42 U.S.C. 4321 et seq.).

The incorporation of a Hazard Analysis and Critical Control Point (HACCP) approach for prevention planning and developing Containment Plans specific to particular research activities is encouraged. Information about the use of HACCP is available at <http://www.seagrant.umn.edu/ais/haccp>. A web site detailing the application of HACCP to natural resource pathways, plus a link to download a HACCP wizard that helps create HACCP plans, can be found at <http://www.haccp-nrm.org>.

## **Federal ANS Research Risk Analysis Protocol**

The Federal ANS Research Risk Analysis Protocol consists of a risk assessment (risk characterization and communication, Part I, below) to be completed by the Principal Investigator to evaluate proposed research for its potential to result in the introduction or spread of aquatic nonindigenous species to or within the waters of the United States.

If indicated by the risk assessment (Part I), the Principal Investigator must develop risk management plans by developing and documenting Containment Plans (Part II). Containment Plans specify and describe the Standard Operating Procedures that will be used throughout the research project to prevent escape or unintentional transfer of aquatic nonindigenous organisms by the research activities conducted under the project. Due to the number of federally funded programs and facilities and the differing characteristics and distributions of potential research organisms and types of research, it is impractical to specify a generic Containment Plan that would be suitable for every situation.

### **Responsibilities**

#### **1. The Principal Investigator (PI)**

The Principal Investigator is responsible, along with his/her institution, for determining that the proposed research complies with all applicable local, state, and national laws and regulations.

Under the Protocol, the Principal Investigator is responsible for:

- Conducting and documenting the research risk analysis outlined in this document.
- Including the completed risk assessment (Part I) as part of the research proposal.
- If Part I indicates the need for Containment Plans, a statement must be included with Part I that the appropriate Containment Plans will be prepared and implemented by the Principal Investigator prior to initiation of the research. See Appendix IV.

Containment Plans should document (1) the control and containment procedures that will be used during research and throughout the time that the species is present and viable—this will usually be accomplished by attaching appropriate Containment Plans; (2) a training plan to assure that all staff associated with the research are aware of the Containment Plan and the Standard Operating Procedures for conducting the research; and (3) a plan showing how, upon completion of the study, the research organisms will be humanely euthanized and disposed of properly.

## 2. The Research Institution

An authorized administrative representative of the Research Institution other than the PI, and from the chain of authority above the PI (such as a Department Chair, Section Chief, Director, etc.), must provide a signed statement as part of the proposal cover pages acknowledging that:

1. The Research Institution has reviewed and approved the proposed research and the Federal ANS Research Risk Analysis Protocol documentation completed by the Principal Investigator.
2. Based on the outcome of the risk assessment (Part I of the Protocol), creation and implementation of appropriate Containment Plans to prevent the introduction of aquatic nonindigenous species to the waters of the United States will be implemented by the Principal Investigator prior to initiation of the research.
3. The Research Institution and the PI are responsible for complying with all applicable local, state, and national laws and regulations related to possession of nonindigenous species. The researcher and/or research institution is responsible for contacting the appropriate state and federal agencies to obtain permits, as required, for transporting and possessing the species of interest.
4. The Principal Investigator and his/her Research Institution are responsible for ensuring that students and staff involved with this research comply with all provisions of the appropriate Containment Plans and legal requirements associated with this research.

## 3. The Funding Agency

NANPCA (1990) section 1202(f)(3) requires that competitive research grants authorized and funded under the Act be conditioned on use of the Protocol to ensure that any recipient of funds follows the protocol. It is the responsibility of funding agencies to determine the applicability of this requirement to any research they fund.

## PART I

### Risk Assessment

Sufficient information and detail must be provided to enable the funding agency program manager and/or proposal reviewers to evaluate the accuracy and completeness of the risk assessment and the need for Containment Plans.

Answer each of the following questions in writing. Provide enough detail so that a reviewer can evaluate and understand the basis for your answers. Use additional pages as needed.

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(Questions 1-4 relate to the risk of introduction.)

1. Will the research involve ONLY the use of preserved samples of water, sediment, and/or biota?

YES  NO

YES: Score = 0; Proceed directly to Question 3

NO: Score = +1

Score, Q1: \_\_\_\_\_

*Proceed to Next Question.*

\* \* \* \* \*

2. Are any nonindigenous disease-causing parasites, pathogens, or other disease-causing agents known to be carried by the species to be used in this research, not already in the ecosystem(s) where the research will be conducted, and OIE (*World Organisation for Animal Health*, [http://www.oie.int/eng/en\\_index.htm](http://www.oie.int/eng/en_index.htm)) reportable or known to be harmful to the health of native and/or stocked species?

YES  NO

YES: Score = +1; prevention/containment procedures are required.

NO: Score = 0

Score, Q2: \_\_\_\_\_

*Proceed to Next Question.*

\* \* \* \* \*

3. Will this research involve transportation of unpreserved water, sediment, and/or biological samples or specimens in any life stage between or through

water bodies or ecosystems not interconnected with the source ecosystem of the samples?

YES  NO

YES: Score = +1; *preventive/protective shipping and transportation procedures may be required.*

NO: Score = 0

Score, Q3: \_\_\_\_\_

*Proceed to Next Question.*

\* \* \* \* \*

4. Will this research involve use of field sampling equipment that is, has been, or will be used in different natural water bodies and/or sediments located in unconnected ecosystems?

YES  NO

YES: Score = +1; *appropriate field equipment decontamination procedures are required; development of a HACCP plan for field gear should be considered.*

NO: Score = 0

Score, Q4: \_\_\_\_\_

Sum of Scores Q1-4: \_\_\_\_\_

**If the Sum of Scores for Questions 1-4 is “0”, STOP HERE - you do not need to take further action. There is low risk that the research activities would result in the introduction or spread of aquatic nonindigenous species, or expose the ecosystem to associated diseases, parasites, or pathogens.**

**If the Sum of Scores for Questions 1-4 is >0, proceed to the next question.**

\* \* \* \* \*

(Questions 5-7 relate to the risk of establishment.)

5. Are there reasons to conclude that the nonindigenous species used in this research **cannot survive and/or reproduce** in any of the ecosystems, watersheds, or drainage networks through which or where live or unpreserved samples will be transported, used, or stored for this research?

YES  NO

YES: Score = 0

*If Yes, please attach a narrative that provides the basis for this answer.*

NO: Score = +1

Score, Q5: \_\_\_\_\_

*Proceed to next Question.*

\* \* \* \* \*

6. Are there reasons to conclude that the nonindigenous species used in this research would NOT become aquatic nuisance species, as defined by NANPCA (1990, as amended), if it/they escaped or were released? (*Note: this does not refer to survival, but rather the likelihood that the species could or will become an aquatic nuisance as defined by NANPCA Section 1003).*)

YES  NO

*Yes: Score = -1; There is low risk that the research activities under this project will result in the establishment or spread of an aquatic nuisance species.*

*If Yes, please attach a narrative that provides the basis for this answer.*

NO: Score = +1

Score, Q6: \_\_\_\_\_

*Proceed to next Question.*

\* \* \* \* \*

7. What was your answer to Question 2?

YES  NO

*YES: Score = +1; prevention/containment procedures are required, even if the host species is believed or known not likely to become established or become a nuisance if released, unless the samples will be preserved at the site of collection in a manner that is known to also kill or deactivate viruses and other pathogens.*

NO: Score = 0

Score, Q7: \_\_\_\_\_

\* \* \* \* \*

(Question 8 establishes if there are existing regulations that require use of specific containment procedures.)

8. Are collection, possession, and/or transportation of any of the species to be used in this research regulated by any local, state or federal laws?

YES  NO

YES: Score = +6

*If "Yes" please attach a citation and brief description/summary of the applicable regulations.*

NO: Score = 0

Score, Q8: \_\_\_\_\_

Sum of Scores Q5-8: \_\_\_\_\_

**If the Sum of Scores, Questions 5-8 is “≤0”,** NO Containment Plan is needed and no further action is required. However, to avoid the spread of nonindigenous species by incidental means during the conduct of this research, care should be taken to decontaminate all field equipment by appropriate means before reusing it in another ecosystem. Development of a HACCP plan for field gear is recommended.

**If the Sum of Scores, Questions 5-8 is >0, but less than +5 and the proposed research is selected for Federal funding,** the PI is responsible for developing and documenting appropriate Containment Plans prior to initiation of research.

**If the Sum of Scores, Questions 5-8 is +5 or greater and the proposed research is selected for Federal funding,** it is the responsibility of the PI and his/her research institution to assure that the research meets all legal requirements for permits and for implementation of any containment procedures specified in regulations.

- If there are applicable containment procedures that are already specified by local, state or federal regulatory agencies, they should be identified by reference in the proposal.
- The existence of legally mandated or specified containment requirements does not preclude the need for the PI and his/her research institution to develop, document, and implement additional Containment Plans that are identified as necessary by this Risk Assessment.



## PART II

### Containment Plan(s)

If the outcome of the risk assessment (Part I) indicates the need for Containment Plans, the Principal Investigator is responsible for developing and implementing a plan to prevent nonindigenous species from escaping or being accidentally released, and for decontaminating associated equipment. The specific procedures will depend on the species involved, their life stages and sizes, the characteristics of the research location(s) with regard to the species' critical environmental factors, and the potential for the species to survive and reproduce in that/those locale(s). If any of the species is or is known to carry nonindigenous disease-causing parasites, pathogens, or other disease-causing agents, extra precautions may be necessary.

The Containment Plan should use a combination of physical, biological, environmental, and/or chemical barriers to contain or confine all life stages of the organism possibly present during the research. **The development and inclusion of the HACCP approach tailored to natural resource pathways is recommended** (see <http://www.haccp-nrm.org/>).

Appendix I provides sources of information related to containment.

Considerations when developing a Containment Plan for research:

- Know and follow all federal, state, local, and institutional regulations pertaining to the species you intend to obtain, especially the need for specific permits for collection or possession of those species; obtain required permits prior to proposing the research, if possible. (See Appendix II for a partial list of laws and regulations. However, the researcher is responsible for ascertaining all applicable local, state, and federal regulations that apply to his/her research).
- Understand the biology and behavior of the organisms relative to potential escape or unintentional release. Are the organisms prone to escape from captivity? Are there highly resistant or physiologically tolerant life stages (e.g., eggs resistant to desiccation)? Are there life stages with high dispersal potential?
- Understand the distribution and physiological tolerances of the organisms. What is the previous invasion/introduction history? Can they survive within the research area(s) if escape or release occurs? Would escape or release likely result in sustainable new populations?
- Learn and maintain good management practices, such as: Clean and disinfect systems (and if appropriate, personnel) between activities; do not leave water or organisms in systems after work is complete (unless

maintaining as research stock); isolate systems (e.g., have separate nets and cleaning equipment for each system).

- Establish a written standard operating procedure (SOP) for proper handling, housing, husbandry, and disposal of specimens. These may be simple or complex as dictated by the organism, the types of activities involving the organism, the housing facility, and applicable regulations. For example, maintenance of nonindigenous species in outdoor facilities will require more containment safeguards than the use of an indoor laboratory facility. Protocols should incorporate redundant safeguards to contain organisms if one level of containment is breached. Practicality is also an important characteristic of effective protocols.
- Unnecessarily stringent and ridged SOPs may make research impossible to conduct and thus ignored or bypassed by research staff. The written SOPs should be rigorous, but allow flexibility and application of judgment where appropriate.
- Train colleagues, staff, and students in proper handling, housing, husbandry, and disposal of specimens. Do not allow unsupervised access to facilities holding live or viable specimens by untrained personnel or the public. Do not allow untrained personnel to perform procedures where escape would be possible.
- Take precautions when moving field gear (e.g., boats, trailers, nets, waders, scuba, snorkeling and similar personal gear, etc.) between locations if transport of nonindigenous species is possible. Field personnel should also be checked if the situation dictates. For example, someone snorkeling in a body of water infested with a species like *Cercopagis pengoi*, the fishhook waterflea, could easily carry a significant number of those organisms caught up in their hair and clothing. Even though adults of that species would quickly die out of water, they could be carrying eggs that can remain viable after such exposure. Procedures such as visual inspection, washing, removal of plant material or sediments, drying, and disinfection can reduce the probability of moving organisms between field sites.

For containment of diseases, parasites, small species, or the early life stages of larger species, the procedures outlined in the National Institutes of Health Guidelines for Research Involving Recombinant DNA Molecules, published May 7, 1986 (51 FR 16958, p. 16959), or guidelines developed by the U.S. Department of Health and Human Services (see references) are the most comprehensive.

For containment or confinement of larger forms, the guidelines developed for whole plants or animals by the Office of Agricultural Biotechnology, USDA, are

the most appropriate, especially if the research is to be conducted outside the laboratory (see Appendix I).

The Principal Investigator and the Research Institution are responsible for ensuring that research activities do not violate laws or regulations and do not result in spread of nonindigenous species.

### **Reporting**

Escape or release of a nonindigenous aquatic species must be reported in compliance with applicable local, state, and federal laws, as well as to the federal agency funding the research. Violation of any conditions attached to funding by a federal agency may have consequences that will be determined and administered by the funding agency.

## **APPENDICES**

**Warning:** The information presented in Appendices I and II was last updated in April 2009 and is believed to be accurate as of that date, but is subject to change. In addition, there may be other sources of information not included here. The reader is advised to check for additional and/or more recent content and guidelines, as needed.

## **APPENDIX I**

### **Existing Guidelines and Protocols**

#### **Guidelines for Recombinant DNA Molecular Research:**

The following is a list of guidelines and protocols used to confine or contain nonindigenous species or organisms involved in recombinant DNA research. These can also be applied to nonindigenous aquatic species proposals. Consulting one or more of these will help investigators to identify physical, biological, chemical, and/or environmental preventative measures that may be used to confine or contain the nonindigenous aquatic species during research, transportation, and storage.

Federal Register 51, No. 8, pg. 16958;  
Federal Register 51, No. 123, pg. 23367  
Federal Register 52, No. 154, pg. 29800  
Federal Register 56, No. 22, pg. 4134  
Federal Register 51, No. 88, pg. 16959

For the most updated information visit <http://oba.od.nih.gov/oba/index.html>

#### **Guidelines for Microorganisms**

National Institutes of Health (NIH). 1968. Guidelines for Research Involving Recombinant DNA Molecules. Published in Federal Register May 7, 1986 (51 FR 16958-16961) with additional major actions August 24, 1987 (52 FR 31838); July 29, 1988 (53 FR 28819); October 26, 1988 (53 FR 43410); March 13, 1989 (54 FR 10508); March 1, 1990 (55 FR 7438); and August 11, 1987 (52 FR 29800) with appendix P for plants and Q for animals; and May 28, 2002 (NOT-OD-02-052). For the most updated information visit <http://oba.od.nih.gov/oba/index.html>

#### **Guidelines for Whole Plants and Animals**

ICES Code of Practice on the Introductions and Transfers of Marine Organisms 2004. <http://www.ices.dk/reports/general/2004/ICESCOP2004.pdf>

U.S. Department of Agriculture (USDA). 1984. Coordinated Framework for Regulation of Biotechnology. Federal Register December 31, 1984 (49 FR 50856) and June 26, 1986 (51 FR 23302).

USDA. 1986. Advance Notice of Proposed USDA Guidelines for Biotechnology Research. Federal Register June 26, 1986 (51 FR 23367-23393) and February 1, 1991 (56 FR 4134-4149).

USDA. 1986. Introduction of Organisms and Products Altered or Produced Through Genetic Engineering Which are Plant Pests or for Which There is Reason to Believe are Plant Pests. Federal Register June 26, 1986 (51 FR

23352-23366) and June 16, 1987 (52 FR 22892-22915) and Code of Federal Regulations January 1, 2008 (7 CFR 340.0).

Coulson, J. R. & R. S. Soper. 1989. Protocols for the introduction of biological agents in the United States, pp. 1-35. *In*: R. P. Kahn (ed.), Plant Protection & Quarantine, Vol. 3, Special Topics. CRC Press, Inc., Boca Raton, FL. 215 pages.

USDA, Office of Agricultural Biotechnology. 1988. USDA Guidelines for Research Outside the Laboratory Involving Biotechnology, also Federal Register June 26, 1986 (51 FR 23367-23313) and February 1, 1991 (56 FR 4134-4149). <http://www.aphis.usda.gov/brs/pdf/abrac%201991.pdf>

### **International Guidelines and Protocols:**

Daszak P, Cunningham AA, Hyatt AD. Draft guidelines for international translocation of amphibians with respect to infectious diseases. Attachment 6. In: Speare R and Steering Committee of Getting the Jump on Amphibian Disease. Developing management strategies to control amphibian diseases: Decreasing the risks due to communicable diseases. School of Public Health and Tropical Medicine, James Cook University: Townsville. 2001: 150-156.

European Inland Fisheries Advisory Commission. 1988. Code of Practice and Manual of Procedures for Consideration of Introductions and Transfers of Marine and Freshwater Organisms. FAO. EIFAC. Occasional paper No. 23. 52 pages.

The FAO/NACA Asia Regional Technical Guidelines on Health Management for the Responsible Movement of Live Aquatic Animals: lessons learned from their development and implementation. R. P. Subasinghe & M. G. Bondad Reantaso, 55-63.

International Council for the Exploration of the Sea. 1982. Proposed Guidelines for Implementing the ICES Code of Practice Concerning Introduction and Transfer of Marine Species. 23 pages.

The World Organisation of Animal Health (OIE). Aquatic Animal Health Code 2008. [http://www.oie.int/eng/normes/fcode/en\\_sommaire.htm](http://www.oie.int/eng/normes/fcode/en_sommaire.htm)

### **Disease-Related Guidelines and Protocols:**

Anonymous. 1989. Operating Procedures for the Alma Quarantine Facility. Prepared for the Alma Research Station, Guelph, Ontario, Canada. 16 pages.

Scarfe, A. D., C-S Lee, and P.J. O'Bryan. 2006. Aquaculture Biosecurity: Prevention, Control and Eradication of Aquatic Animal Diseases. Blackwell Publishing Professional. Ames, Iowa.

Horner, R. W., and R. L. Eschenroder. 1993. Protocols to Minimize the Risk of Introducing Salmonid Disease Agents with Importation of Salmonid Fishes. Great Lakes Fish Disease Control Committee Spec. Pub, 27-37.

U.S. Department of Health and Human Services. 2007. Biosafety in Microbiological and Biomedical Laboratories. 5th Edition. U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control, Atlanta, Georgia 30333, and National Institutes of Health, Bethesda, Maryland 20892. 11 pages

American Fisheries Society - Fish Health Section Blue Book. 2007. Suggested Procedures for the Detection and Identification of Certain Finfish and Shellfish Pathogens. <http://www.afsbooks.org/x70314cxm1.html>

An additional 17 references on laboratory disease and pathogen control methods can be found listed in the Federal Register, May 7, 1986 (51 FR 16965).

#### **Other Guidelines and Protocols:**

Klingman, D. L., and J. R. Coulson. 1983. Guidelines for Introducing Foreign Organisms into the United States for Biological Control of Weeds. Bulletin of Entomological Society of America. Fall 1983:55-61.

Guidelines for the Importation, Interstate Movement, and Field Release of Foreign Arthropod-Parasitic Nematodes into the United States for Biological Control of Arthropod Pests of Plants, Man, and Domestic Animals, and Vectors of Plant, Human, and Animal Pathogens, and for the Interstate Movement and Export of Foreign and Native Arthropod-Parasitic Nematodes for Research on Biological Control of Such Pests.

Guidelines for the Importation, Interstate Movement, and Field Release of Foreign Microbial Pathogens (Fungi, Bacteria, Rickettsia Viruses, Protozoa) into the United States for Biological Control of Arthropod Pests of Plants, Man, and Domestic Animals, and Vectors of Plant, Human, and Animal Pathogens, and for the Export of Foreign and Native Arthropod Pathogens for Research.

Guidelines for the Importation, Interstate Movement, and Field Release of Foreign Arthropods and Nematodes into the United States for Biological Control of Weeds, and for the Interstate Movement and Export of Foreign and Native Arthropod and Nematode Natural Enemies of Weeds.

Guidelines for the Importation, Interstate Movement, and Field Release in the United States of Foreign Microbial Pathogens for Biological Control of Weeds, and for the Interstate Movement and Export of Foreign and Native Pathogens of Weeds for Research.

Guidelines for the Importation, Interstate Movement, and Field Release of Foreign Beneficial Organisms (Microbial Pathogens and Antagonists) into the United States for Biological Control of Plant Nematodes and Plant Pathogens, and for the Export of Such Organisms (Foreign and Native) for Research.

Southeastern Cooperative Wildlife Disease Study. 1985. Model for State Regulations Pertaining to Captive Wild and Exotic Animals. University of

Georgia, Athens, Georgia. 48-page manuscript. Prepared in response to Resolution #9. U.S. Animal Health Association, Milwaukee, Wisconsin 10/27-11/1/85.

Reid, D.F., J. Bidwell, J. Carlton, E. Marsden, and S. Nichols. 1993. Zebra-Mussel-Specific Containment Protocols. Aquatic Nuisance Species Task Force, Approved Species-Species Protocol. 72 pages

Jennings, G. P., and J. A. McCann. 1991. Research Protocol for Handling Nonindigenous Aquatic Species. National Fisheries Research Center, U.S. Fish and Wildlife Service, Gainesville, Florida. 43 pages.

#### **Brown Tree Snake Protocol:**

Pacific Basin Development Council. 1991. Recommended Protocol for Transport of Live Brown Tree Snakes (*Boiga irregularis*). Prepared for Plant Quarantine Branch, State of Hawaii Department of Agriculture and Biological Survey, and the U.S. Fish and Wildlife Service.

#### **Guidelines for Animal Care and Welfare:**

Guidelines for Use of Live Amphibians and Reptiles in Field Research and Laboratory Research. 2004. Second Edition, Revised by the Herpetological Animal Care and Use Committee (HACC) of the American Society of Ichthyologists and Herpetologists.

Office of Science and Technology Policy. 1985. Interagency Research Animal Committee's Report: U.S. Government Principles for the Utilization and Care of Vertebrate Animals Used in Testing, Research, and Training. Federal Register, Vol. 50, No. 97, May 20.

Guidelines for the Use of Fishes in Field Research. 1988. American Society of Ichthyologists and Herpetologists (ASIH), American Fisheries Society (AFS), and American Institute of Fisheries Research Biologists (AIFRB). Fisheries, Vol. 13, No.2, 16-23.

#### **Guideline for Quarantine Procedures**

Fisher, T. W. & L. A. Andrés. 1999. Quarantine: concepts, facilities, procedures. *In*: Principles and Application of Biological Control. Academic Press, San Diego, CA. 1046 p.



## **APPENDIX II**

### **Other Relevant Legislation and Executive Orders**

Applicable State Laws, Regulations, Permit and Notification Requirements - Must be determined on an individual basis by Principal Investigators and Research Institutions.

Lacey Act of 1900 - 16 U.S.C. 3371-3378 and 18 U.S.C. 42 Item 2,58 amended with the 2008 Farm Bill

[http://www.aphis.usda.gov/plant\\_health/lacey\\_act/index.shtml](http://www.aphis.usda.gov/plant_health/lacey_act/index.shtml)

Endangered Species Conservation Act of 1973— 16 U.S.C. 1531-1543 plus Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES) — 16 U.S.C. 1531-1543.

Executive Order 11987 dated March 1977 - Exotic Organisms

Plant Quarantine Act of 1912 (7 U.S.C. 151 et seq.)

Terminal Inspection Act of 1915

Federal Plant Pest Act of 1957 (7 U.S.C. 150aa et seq.)

Federal Noxious Weed Act of 1974 (Public Law 93-629-Jan. 3, 1975) (7 U.S.C. 2801 et seq. + 21 U.S.C. 111 et seq.)

National Environmental Policy Act of 1969 (NEPA)

Occupational Safety and Health Act of 1970 - Federal Register April 12, 1984 (50 FR 14468) (29 U.S.C. et seq.)

Animal Welfare Act. 7 U.S.C. 2131-2155; 80 STAT. 350, 84 STAT. 1560, 90 STAT. 417, 99 STAT. 1645.

The Plant Protection Act of 2000 – replaced the Plant Quarantine Act, the Federal Pest Act, and the Federal Noxious Weed Act and seven other statutes.

## **APPENDIX III**

### **Definitions**

**Aquatic Nuisance Species (NANPCA, 1990, as amended):** an aquatic nonindigenous species that threatens the diversity or abundance of native species or the ecological stability of infested waters, or commercial, agricultural, aquacultural or recreational activities dependent on such waters.

**Established Population:** when used in reference to a species, means the species is reproducing and self-sustaining in an open ecosystem, i.e. in waters where the organisms are able to migrate or be transported to other waters.

**Nonindigenous Species:** any species or other viable biological material that enters an ecosystem beyond its historic range, including any such organisms transferred from one country to another. Nonindigenous species include both exotics and transplants. [Note: Historic range is interpreted to mean the territory occupied by a species at the time of European colonization of North America.]

**Pathogen:** as defined in USDA guidelines, a virus or microorganism (including its viruses and plasmids, if any) that has the ability to cause disease in another living organism.

**Surrounding Waters:** any free flowing or standing waters in the immediate vicinity of the research facility that are connected with public waters either directly or indirectly.

**Survive:** when used in reference to biological species, means the species is able to live in an ecosystem during its normal life span, but not necessarily that it is able to reproduce itself.

**Unintentional Introduction:** an introduction of nonindigenous species that occurs as a result of activities other than the purposeful or intentional introduction of the species, usually involving the release, often unknowingly, of nonindigenous organisms without any specific intent.

**Waters of the United States:** the navigable waters and the territorial sea of the United States. Since aquatic species can move or be transported by currents into navigable waters, all internal waters of the United States, including its territories and possessions, are included. The Territorial Sea of the United States is that established by Presidential Proclamation Number 5928 of December 27, 1988.

## **APPENDIX IV**

### **Suggested Content for Containment Plans**

Identification of Principal Investigator and Research Institution

Description of research

Description and location(s) of research facility(ies) and sampling sites

Source of specimens if not from sampling sites (e.g., provided by another researcher or research institution)

Nonindigenous species to be collected or used in the research

Summary of

- Biology, including Diseases and Parasites
- Life History
- Ecology
- Environmental Factors
- Prior Invasion History
- Present Distribution and
- Status of the Species in the Study Area(s).

Permits required (if any)

Containment procedures specified by regulations, if any

HACCP analysis

Containment procedures to be used for physical, biological, chemical, and environmental containment, in addition to any required by regulation

- Shipping and transportation precautions

Training and qualifications of personnel

Security at facilities where live specimens will be maintained

- Plan for extreme events (hurricanes, floods, etc.)
- Plan for securing facility and limiting access

Emergency Plans in case of escape or release

Procedures for terminating research

- Fate of Surviving Specimens – Close-Out Procedures

Administrative controls, roles, responsibilities

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