

CHAPTER 15

Procedure for Revisions to the National Wild Fish Health Survey Laboratory Procedures Manual

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I. Introduction

The National Wild Fish Health Survey Laboratory Procedures Manual is intended to provide optimum detection methods and standardized protocols for all aspects of the Survey. This document is intended to be adaptive to follow both the changing body of scientific knowledge and to address emerging pathogens. The following chapter outlines the mechanism for revising to the NWFHS Manual.

II. Format of the Manual

A. TITLE AND CITATION

National Wild Fish Health Survey Procedures Manual – Second Edition (June 2004).

True, K., (ed.) 2004 National Wild Fish Health Survey Laboratory Procedures Manual, Second Edition. U.S. Fish and Wildlife Service.

B. CHAPTERS

The Manual consists of 15 chapters and necessary appendices. The Quality Assurance Program (Chapter 15 in the 2001 First Edition of the Manual) still pertains to the NWFHS, but is now contained in a separate volume of the USFWS Handbook:

Chapter 1 Overview of the National Wild Fish Health Survey

Appendix 1.A Glossary of Terms

Appendix 1.B APHIS Memorandum regarding OIE Notifiable Diseases

Chapter 2 Sample Collection and Submission

Appendix 2.A Shipping Addresses and Contacts for the Fish Health Centers

Appendix 2.B NWFHS Submission Form

Chapter 3 Sample Receipt and Laboratory Tracking

Appendix 3.A Case History Record

Appendix 3.B Chain of Custody Form

Chapter 4 Standard Necropsy Procedures for Finfish

Chapter 5 Bacteriology

Appendix 5.A Media Formulations

Appendix 5.B Reagents

Appendix 5.C Profiles Obtained with API-20E for Known Fish Pathogens

Appendix 5.D Flowchart for Targeted Gram-Negative Fish Pathogens

Appendix 5.E Flowchart for Targeted Gram-Negative PRI

Appendix 5.F Some Characteristics of Long Gram-Negative Bacteria

Chapter 6 - ELISA for Detection of Renibacterium salmoninarum Antigen in Fish Tissue

- Appendix 6.A Quality Control Program for ELISA
- Appendix 6.B Standardization of Reagents
- Appendix 6.C Collection and Processing of Tissue Samples for ELISA
- Appendix 6.D Reagents, Supplies and Equipment Lists

Chapter 7 Corroboration Testing of Renibacterium salmoninarum by PCR

- Appendix 7.A Worksheet for DNA Sample Data
- Appendix 7.B Worksheet for Initial Amplification of Rs DNA by PCR
- Appendix 7.C Worksheet for Nested (Second Round) Rs PCR
- Appendix 7.D Photodocumentation and Report of Results

Chapter 8 Parasitology

- Appendix 8.A Reagents and Solutions
- Appendix 8.B Common Parasites of Fish

Chapter 9 Corroboration Testing of Parasites by PCR

- Appendix 9.A Equipment, Supplies and Reagents
- Appendix 9.B Mc PCR Worksheet
- Appendix 9.C Analysis of Extracted DNA using an UV Spectrophotometer

Chapter 10 Tissue Culture of Fish Cell Lines

Chapter 11 Virology

- Appendix 11.A Glossary of Terms used in Tissue Culture and Virology
- Appendix 11.B Media Used in Tissue Culture and Virology

Chapter 12 Corroborative Testing of Viral Isolates

- Appendix 12.A. PCR Worksheets: Corroborative Testing of Viruses
- Appendix 12.B. Photodocumentation of Agarose Gel Electrophoresis of PCR Products
- Appendix 12.C. General Procedures for PCR Protocols
- Appendix 12.D. Analysis of Extracted DNA using Gene Quant UV Spectrophotometer
- Appendix 12.E. Quality Assurance/Quality Control for PCR
- Appendix 12.F. Reagents

Chapter 13 Histology for Finfish

Chapter 14 Non-Lethal Methodology for Detection of Fish Pathogens

Chapter 15 Protocol for Revision to the National Wild Fish Health Survey Laboratory Procedures Manual

- Appendix 15.A. Second Edition (2004) Manual Revision Committee Members
- Appendix 15.B. Associate Editors – Previous Contributors to the First Edition (2001)

C. LAYOUT

The layout of the document will be in outline form as follows:

1. Roman numerals for main chapter headings: I, II, III, IV, V.
2. Sub-heading formatting will follow : A,B,C....1,2,3,.....i, ii, iii.
3. Each chapter, or a specific section of a chapter, will have its own bibliography.
4. Each chapter will have its own appendices with chapter number and capitol letters.
 - a. Appendix one of Chapter 3 would be titled and referenced as (3.A)
 - b. Appendix two of Chapter 3 would be titled and referenced as (3.B)

III. Stringency

1. The appropriateness of methodologies shall be determined by the Center Directors based on several factors:
 - The sensitivity of the assay
 - The specificity of the assay
 - Availability of reagents
 - Availability of technology and/or required personnel training
 - Labor requirements
 - The cost of the assay.
2. The incorporation of additional pathogens into the Manual shall be contingent on the availability of appropriate screening and corroborative tests.

IV. Revision Process

1. The Manual will be reviewed on an annual basis by all USFWS Fish Health Centers.
2. Recommends for revision will be submitted at the annual Fish Health Biologist Meeting.
3. When revisions include proposal of the inclusion of a new pathogen, the Center submitting the proposal will provide the appropriate screening and corroborative test methods, and references for the protocols.
4. Center Directors will adopt revisions by a consensus vote.
5. One member from each Fish Health Center will be assigned to the Manual Revision Committee (Manual Committee).
6. The Committee will make appropriate revisions to the Manual and submit the final draft to all Fish Health Centers for review.
7. The revised Manual will be adopted at the subsequent Fish Health Biologist Meeting.
8. A mastercopy of the Manual will be maintained on the National Wild Fish Health Survey website. In instances where serious typographical or technical errors occur, the web version of the Manual will be updated to reflect immediate changes that cannot wait until the next revision cycle.
9. The National Aquatic Animal Health Coordinator for the USFWS will be responsible for coordinating revisions to the NWFHS Database when new pathogens or procedures are adopted.

V. Committee Members

- A. The Committee shall consist of nine people, the NWFHS representatives from each Fish Health Center. Additional members may be included by a consensus vote of the Committee, if additional expertise is needed.
- B. The Committee will have one chairperson, to be nominated and elected by simple majority by the Committee members each year.

VI. Committee Authority

- A. The Committee will have the authority to make minor revisions defined as edits that update, optimize, or support current methodologies without changing the screening or corroborative methods. These revisions can be incorporated by consensus vote of the members of the revision committee. If a consensus decision cannot be made, the chair will take the issue to the Center Directors for a final decision (by consensus).

- B. Major revisions are changes that involve the addition of new pathogens, new methods, or significant deviations from the current protocol. These revisions will require a consensus vote by Center Directors. Proposals for new pathogens, or revisions of current methods will be discussed at the Annual Fish Health Biologists meeting. Center Directors will decide by consensus whether to accept, reject, or amend the recommendation(s) for revision.

- C. The Committee Chair will maintain a record of all decisions made during the annual meeting and direct the Committee to incorporate these revisions in the annual revision process.

VII. Manual Revisions

- A. If the changes are substantive (more than typos or clarifications), then an additional page(s) will be inserted into the Manual, stating the change and the date of the change.

- B. The Chair will keep the master copy that indicates the changes, the date, and the rationale for change.

- C. The Chair will update the web version of the Manual that is maintained on the NWFHS website. This master copy will be the definitive manual.

VIII. Revision Time Line

- A. March 1 – April 1 - Changes are proposed and adopted at the annual Fish Health Biologist meeting. Adopted revisions are submitted to the Committee Chair for inclusion in the next Manual revision.
- B. September 1 - November 1 – Revision Committee is formed, a new Chair is elected, and Chair distributes proposed changes to the committee members.
- C. November 1- December 1 – The Committee meets via teleconference to determine whether to accept or reject proposed changes.
 - 1. If the Committee rejects the proposal, due to lack of appropriate screening and corroborative test, this decision is communicated to the Center Directors, who may provide further direction to the Committee.
- D. December 1- January 1 – Committee makes proposed revision to the Manual and any additional minor revisions adopted by the Committee.
- E. January 1- February 1 – Committee distributes the revised Manual to Center Directors and Fish Health Center staff for review.
- F. February 1- March 1 – Centers submit comments to the Committee.
- G. April – The Chair or his/her designee(s) distributes final draft to Fish Health Centers and posts final draft on web page.
- H. April – A summary of the revisions and future recommendations are presented to the group at the annual Fish Health Biologist meeting.

Appendix 15.A – Second Edition (2004) Manual Revision Committee

Patricia Barbash
Lamar Fish Health Unit
Non-Lethal Methodology

John Coll
Lamar Fish Health Unit
Quality Assurance*

Norm P. Heil
Warm Springs Fish Health Center
Sample Collection and Submission

Laura Kessel
Idaho Fish Health Center
Rs-PCR

Ken Lujan
Lower Columbia Fish Health Center
Corroborative Testing of Parasites

Sonia Mumford
Olympia Fish Health Center
Histology
Manual Revision

Terrance Ott
La Crosse Fish Health Center
Tissue Culture Section

Kenneth Peters
Bozeman Fish Health Center
Virology
Corroborative Testing of Viruses

Kimberly True, Editor
California-Nevada Fish Health Center
Manual Revision Committee Chair
ELISA, Parasitology,
Manual Revision Oversight

Linda Vannest
Bozeman Fish Health Center
Corroborative Testing of Parasites
Corroborative Testing of Viruses

Jason Woodland
Pinetop Fish Health Center
Bacteriology, Corroborative Testing of Viruses

Appendix 15.B Associate Editors

Fish health biologists from the nine regional Fish Health Centers developed and contributed individual chapters for the initial version of the Manual (Version 1.0 – June 2001). The following individuals made significant contributions:

Patricia Barbash
Lamar Fish Health Unit
Chp 7 - Rs PCR
Chp 14 – Non-lethal Methodologies

Ray Brunson
Olympia Fish Health Center
Chp 11- Virology Section

Norm P. Heil
Warm Springs Fish Health Center
Chp 2 - Sample Collection and Submission

Becky Lasee
La Crosse Fish Health Center
Chp 8 Parasitology – Section 1
Chp 3 – Sample Receipt
Chp 15 – QA/QC*

Jerry Landye (formerly with)
Pinetop Fish Health Center
Chp 8 Parasitology – Section 3

Beth McCasland (formerly with)
California-Nevada Fish Health Cntr
Chp 5 - Bacteriology

Kenneth Peters
Bozeman Fish Health Center
Chp 8 - Cs PCR

Terrance Ott
La Crosse Fish Health Center
Chp 10 - Tissue Culture Section

Kimberly True, Editor
California-Nevada Fish Health Center
Chp 5 – Bacteriology
Chp 6 - ELISA
Chp 8 - Parasitology – Section 2

*Quality Assurance/ Quality Control (QA/QC) is now a separate volume of the USFWS Handbook

Other Contributors to the First Edition:

William E. Knapp and Mary Ellen Mueller – Chapter 1
USFWS - Division of Hatcheries
Arlington, Virginia

Theodore R. Meyers – Chapter 4, 12, & 13
Sally Short – Chapter 13
Alaska Fish & Game
C.F. Division
Juneau & Anchorage

James R. Winton and William N. Batts – Chapter 12
Western Fisheries Research Center
Seattle, Washington