

Descriptive Guide to Observing Fish Lesions



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Andrew S. Kane, M.S., Ph.D.

Director, Aquatic Pathobiology Center
University of Maryland
Department of Epidemiology & Preventive Medicine
Division of Environmental Toxicology

VA-MD Regional College of Veterinary Medicine
8075 Greenmead Drive, College Park, MD 20742
Tel: 301-314-6808; email: akane@umaryland.edu
<http://aquaticpath.umd.edu>

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Additional copies can be obtained from the Lesion Guide website (<http://mybay.umd.edu/lesionguide.html>)

Introduction

Disease outbreaks and mass mortalities of fish generate notable public concern and often require investigation on the part of fish health diagnosticians and fishery managers. External lesions on fish are commonly observed in association with field morbidity or mortality events, and descriptions of these lesions can be helpful in diagnosing the cause of the etiologic agent(s), i.e., the cause of the event. However, the terminology (or lack thereof) used to describe external lesions is variable and may differ between agencies and individuals. This challenge can lead to reduced quality of observational data that is helpful to fish health managers and diagnosticians. With the support of the US Environmental Protection Agency, Office of Wetlands, Oceans and Watersheds, this guide to fish lesion observations was developed.

Purpose of this guide

This guide was developed to present an overview of observation techniques and provide terminology that can accurately describe external fish lesions. You do not need to be a fish pathologist to make observations that can be used to make a diagnosis. Using a consistent approach, and with a little practice, field technicians, fisheries biologists and environmental managers can make relevant observations, take high quality tissue samples, and make substantial contributions to field fish health studies. To make good observations they should not only make sense to you, but the descriptions should not be misunderstood by others reviewing your data. Further, it is helpful to be **systematic** (i.e., follow a prepared, itemized “game plan” for making observations) and **consistent** (treat all observations and samples in the study the same way) in your sampling strategy. This guide was intended to facilitate observations both in the field as well as in the laboratory.

This guide contains common terminology, sample descriptions and data sheets to assist with lesion observations. The data sheets have been provided for you to use “as is” or as a template to utilize data entry items or formats on your own data sheets. Good planning and use of appropriate data sheets plays an important role in determining the final outcome of your observations. Remember, you can never go back and get data that was not collected!!!

Sections to this guide:

- Protective Clothing
- Case Identity
- External Anatomy Review
- External Examination
- Descriptive & Systematic Observations
- Photographic Documentation
- Acknowledgements
- Selected References
- Vendors for supply materials
- Field and necropsy worksheets

Protective Clothing

Although making visual observations of lesions does not necessarily involve direct, physical contact with animals, careful inspection or disposal of individual fish carcasses often does involve some form of contact. Therefore, it is prudent to use protective clothing to avoid contact with potential disease agents. Gloves, lab coat, apron, goggles, and other protection should be worn as needed. A list of vendors that supply these items is included at the end of this guide.

Case Identity and History

When making collections or lesion observations each animal should have a unique ID number using a defined accession system. Useful information to include with your gross pathology observations includes: source of fish, water quality from source including temperature, source location, time and weather conditions during collection, collection method, general environmental observations, anomalous behavior (i.e., gulping at surface, flashing, morbidity, etc.), an estimate of the number of affected fish, relationship to previous reports, and nearby landmarks (i.e., marinas, sources of potential contamination, etc.).

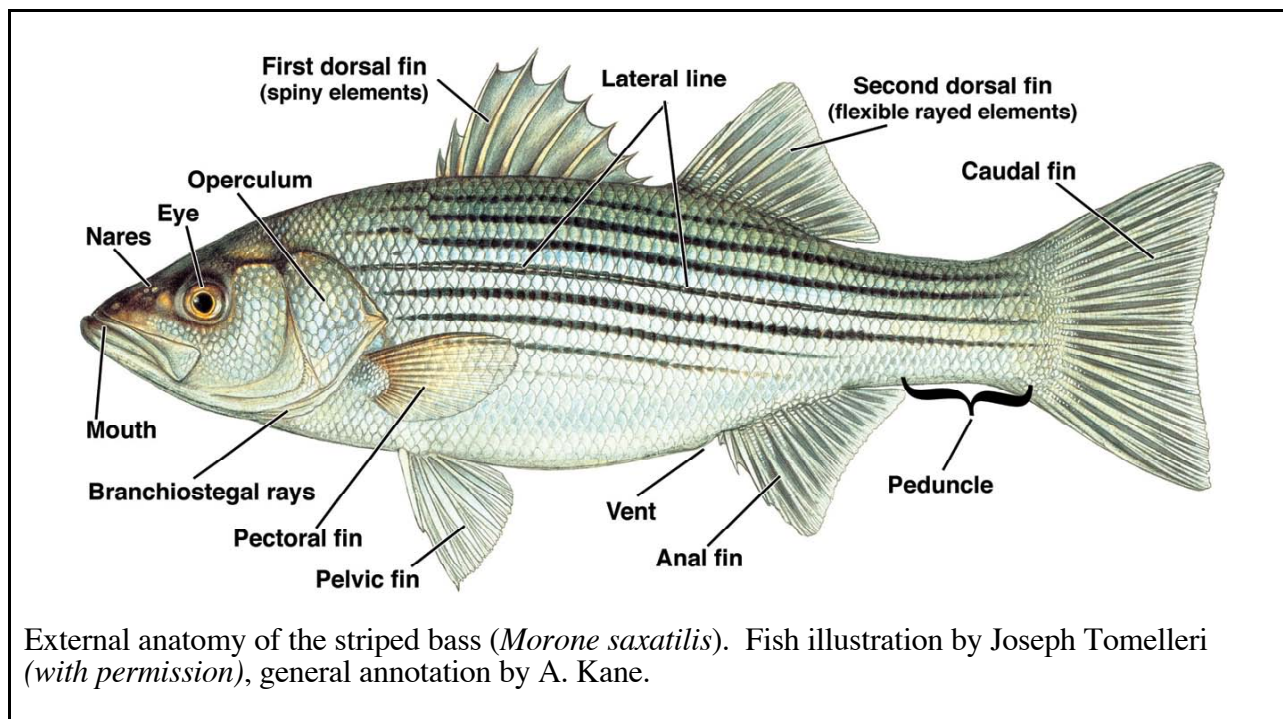
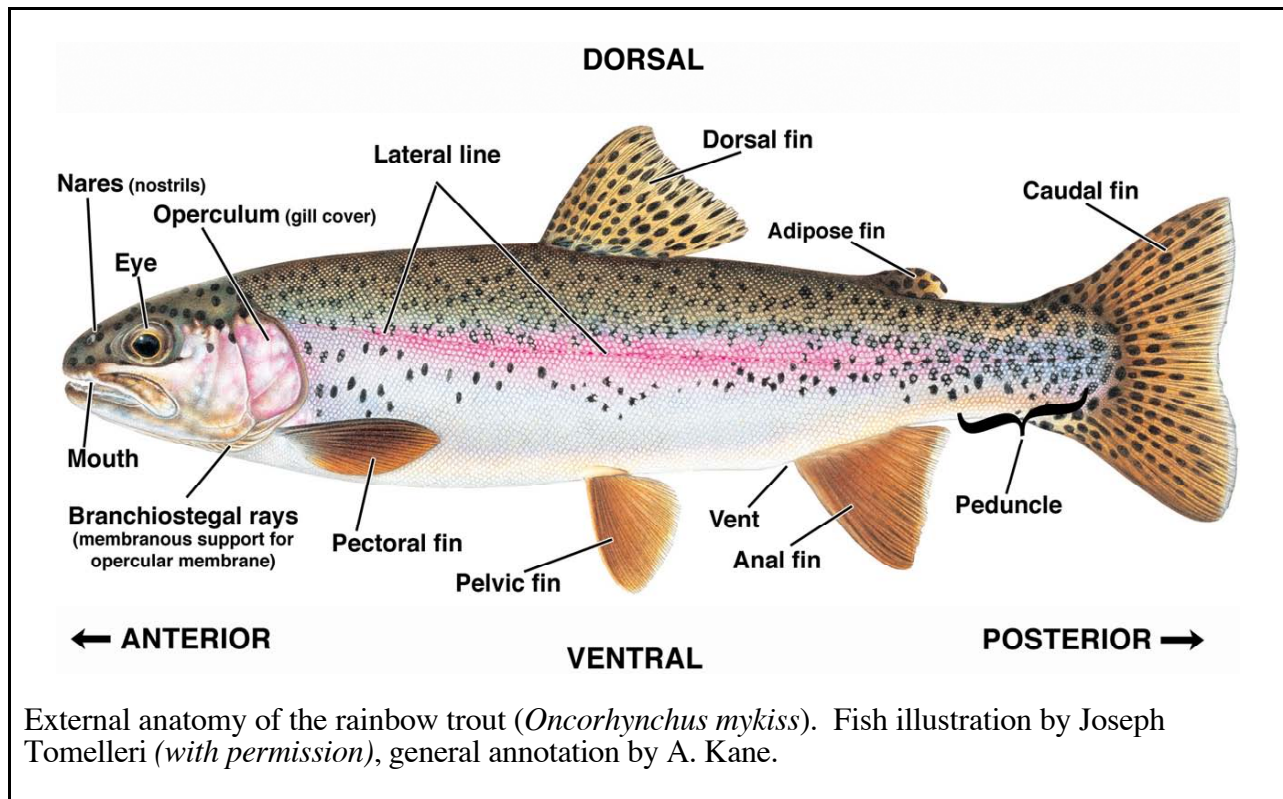
Review of External Anatomy

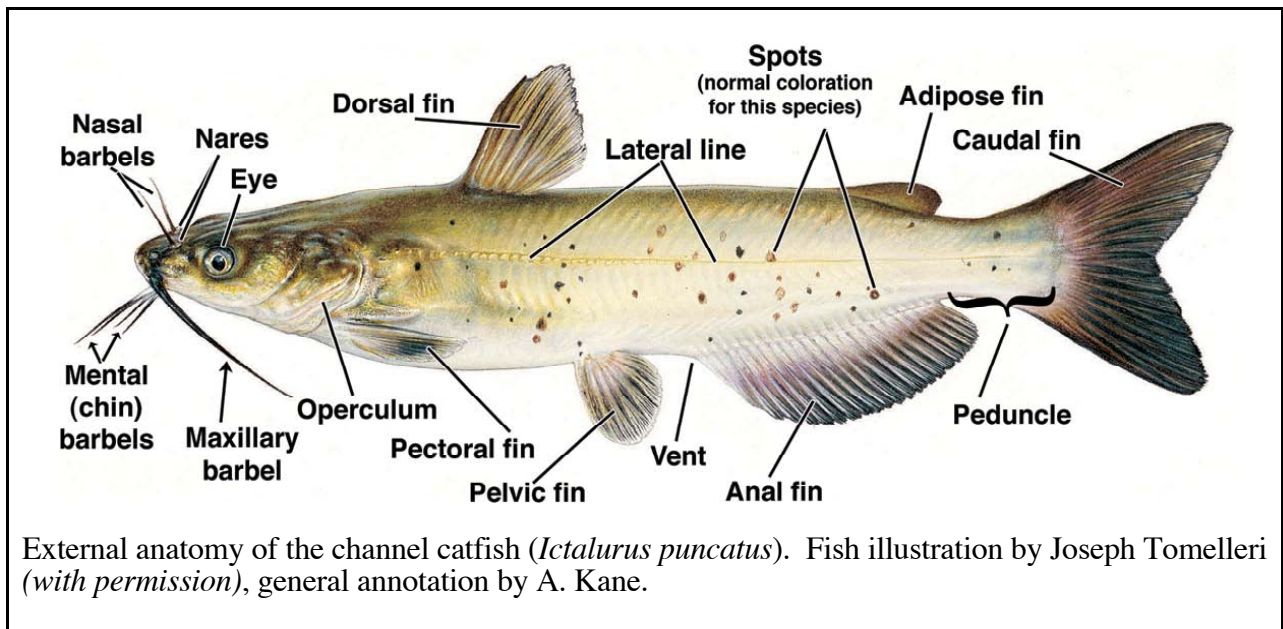
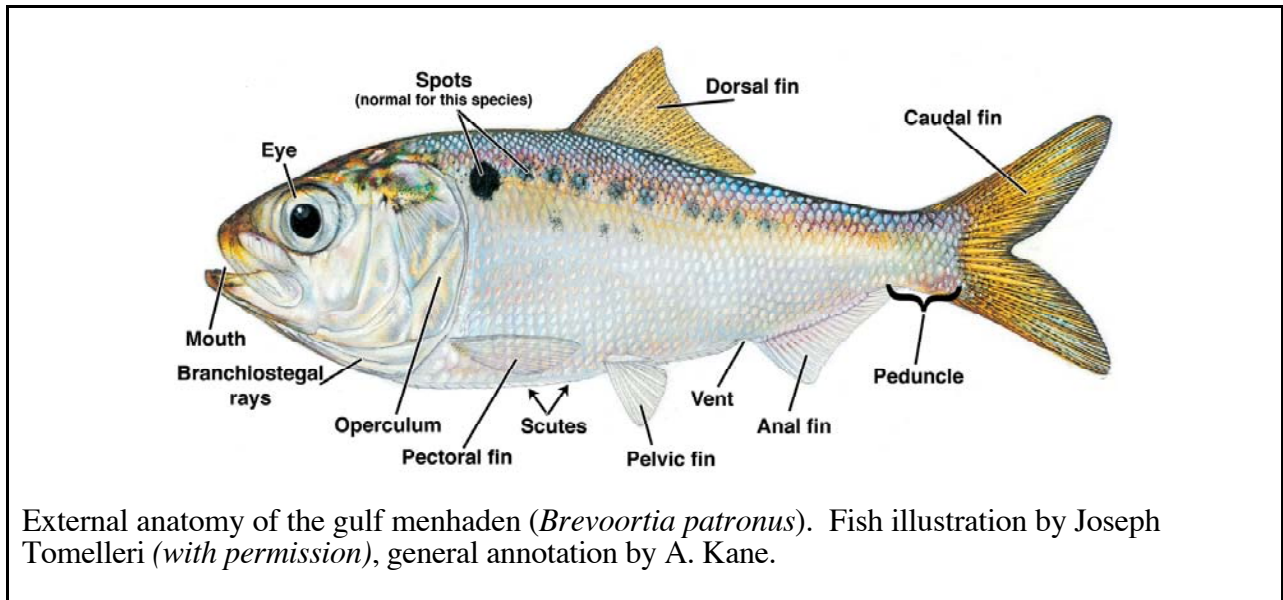
This section will allow you to clearly specify the anatomical location of any gross pathology observed. Examples of four fishes with varying anatomical features are annotated below for reference.

These illustrations were contributed, with permission, by Joseph Tomelleri (Cimmeron Trading Co. <http://www.americanfishes.com>). These images may not be reproduced, used or distributed except as part of this intact outreach guide to fish lesions.

The first fish image shown, rainbow trout, is annotated with dorsal-ventral and anterior-posterior orientation:

External Anatomy





External Examination (checklist of parameters that may be relevant for comment):

Identify the species, length and weight.

Is the animal alive, weak, moribund or dead? How many animals are affected?

Are there any areas of abnormal (or different) coloration? Does the fish appear generally pale or have areas of dark coloration?

What color are the gills? (healthy gills are usually a bright cherry red)

Is there excessive mucus on the skin or gills?

Is the body shape normal? Abdominal distention, skeletal deformities, bulging eyes?

Are observations of abnormalities on one side (left or right) or bilateral?

Are there any sores, areas of reddening, lesions or frayed fins? (focus of this guide)

Parasite survey: skin scrape, gill biopsy, fecal sample results.

See attached Necropsy Worksheet that provides space for these observations.

Making observations:

Good lesion observations are descriptive and systematic. Record detailed observations of any lesion or area that looks abnormal. In any samples taken for histopathology, include pieces of the abnormal tissue with a bit of surrounding, apparently normal tissue. To provide maximal imagery of your observations, include comments on:

size (provide measurement or estimate dimensions);

number (how many)

location (where on body)

presentation (focal, multifocal, diffuse)

shape (round, oval, thread-like, ribbon or tube-like, etc.)

texture (smooth, granular, nodular, cheesy)

color and opacity (transparent, translucent, opaque)

firmness (hard, soft)

3-D description (depressed, flat, raised)

severity (minimal, mild, moderate, marked, severe)

extent (% body coverage; Note that this is a difficult estimate to make accurately and can easily be over-estimated. Consider that if one entire side of a fish is affected, that would equate to 50% coverage)

Use the above comment categories and “food descriptions” to convey the lesion imagery, particularly when the anomaly which you are describing is unknown to you. Examples of material or food imagery which can be used to describe lesions include: cauliflower, cottage cheese, cream cheese, pea soup, cream and coffee, marbles, sandpaper, etc. Imagery can also be used for size comparisons if no ruler is available, e.g., size of a pin head, pencil eraser, nickel, dime, quarter, orange, basketball, etc. Please refer to the simple lesion classification system below for additional information.

Further, we suggest that you draw a picture or use data sheets that contain a fish line drawing and draw in the abnormality indicating location. Complement the drawing with descriptive words. A sample data sheet is provided that you can use to record lesion observation data.

A SIMPLE LESION CLASSIFICATION SYSTEM:

1. LESION OBSERVATION (describe primary lesion first; make additional observations for other lesions):

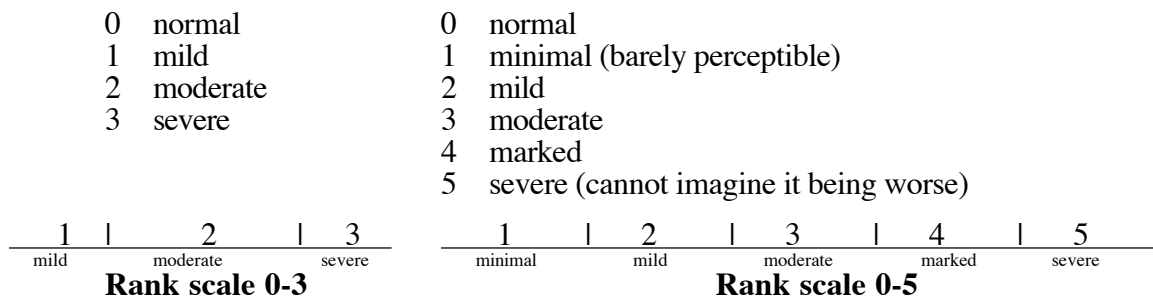
- a) **Loss of Scales (LOS):** there is no reddening or other changes; lesion does not obviously penetrate beneath the surface of the skin and scales.
- b) **Reddening:** red or reddish color (diffuse or focal) associated with the observation; this is not necessarily a penetrating lesion
- c) **Ulcer:** there is a penetration of the lesion through the skin and scales. Underlying dermis (deep skin beneath the scales), muscle or viscera may be visible.
- d) **Other:** describe

2. LESION LOCATION (e.g., head, vent, left pectoral fin, lateral line, etc.)

3. LESION PRESENTATION

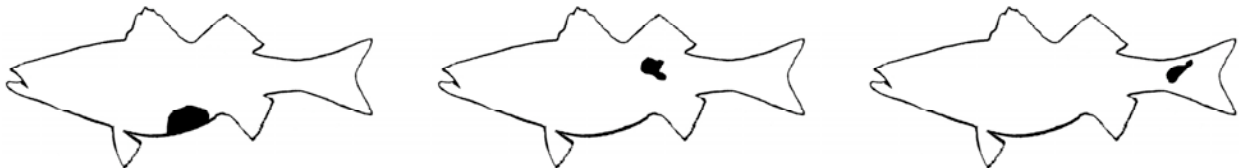
- a) **focal:** single affected area, typically the lesion margins are well-circumscribed
- b) **multifocal:** more than one affected area
- c) **diffuse:** affected area(s) widely distributed (like chicken pox); margins of the lesion(s) may not be well circumscribed

4. LESION SEVERITY: May be ranked on a scale of 1-3 or 1-5; 0=no lesion. If using the 0-3 scale, the mid-range severity ("moderate," with a rank of 2) covers a broader range of severity than mild or severe in the --5 scale. See diagram below. The 0-5 scale affords greater range of observation.



Cartoon of Fish Lesion Presentation:

Focal



Multifocal



Diffuse



Additional Examination Techniques: (included for conceptualization, but not a focus of this guide)

Systematic necropsy: Although necropsy techniques are outside the scope of this guide, we can mention that non-professionals can learn to sample all organ systems for preservation and subsequent histological examination. Several references are provided at the end of this guide that include necropsy techniques. All organs should be sampled for necropsy, regardless of the presence of grossly observable lesions. Organs should include gills; heart with ventricle, atrium and bulbus arteriosus; skin; muscle; bone/cartilage; brain; eye; lateral line; esophagus; stomach; intestine; anterior kidney; posterior kidney; swim bladder; liver; spleen; mesentery and gonad.

Tissue sampling and preservation techniques for histopathology:

- Tissue samples taken for pathology should be no more than 5 mm (one-half inch) thick to insure proper fixation.
- Use a 1:10 tissue to fixative ratio to insure proper fixation.
- Use neutral buffered formalin to avoid certain fixation artifacts (recipe below)

Recipe for 10% Neutral Buffered formalin (to make 1 liter):

Formaldehyde (37%)	100 mL
Distilled water	900 mL
NaH ₂ PO ₄ (sodium phosphate, monobasic)	4.0 g
NaHPO ₄ (sodium phosphate, dibasic)	6.5 g

Safe and proper sample shipment and cleanup:

Sample shipment: Whirlpak bags; double or triple bags (particularly if tissues have spines or bony protrusions), paper insulation between bag layers; protective packing materials (wrap bags in newspaper and surround with crumpled newspaper to protect in shipping); study box. Include copies of all pertinent data sheets and accession numbers for samples. If using FedEx or other special service, be sure to include recipients proper address and telephone number and verify with recipient that the package will be arriving at a specified time and location.

Cleanup: Common sense applies. Double bag all carcasses and bloody materials and gloves. Have a dedicated container for “sharps,” i.e., syringes, broken glass and microscope slides and cover slips. Large lidded coffee cans, labeled as “sharps” works well. Good housekeeping speaks for itself: do not leave anything behind and clean up any blood/mucus from working surfaces.

Photographic documentation of lesion observations

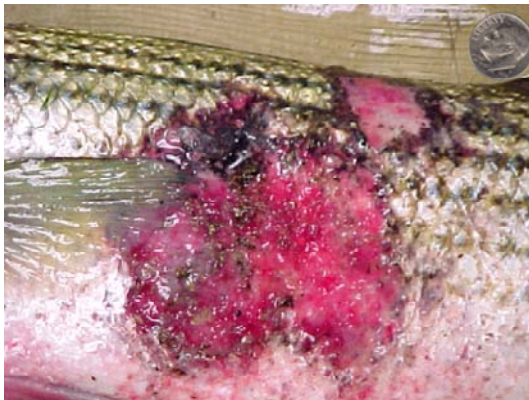
If possible take a photograph (note photograph number of the lesion/specimen). Include a ruler (or common object, e.g. a coin) in the picture to indicate relative size and a tag with the specimen number or ID. Photographs provide the pathologist with accuracy and the greatest possible information).

- a) practice in advance - take test pictures with exposure bracketing;
- b) entire specimen (including accession number and ruler for scale);
- c) close-up of suspect areas (i.e., lesions or abnormalities).

Thanks to “modern times,” digital photography has become user-friendly and affordable. Use of digital photography and a computer with internet accessibility, allows fish culturists and other persons to take digital pictures of whole fish, lesions and microscopic images, and forward them to colleagues or fish health experts for evaluation. Low-end, but highly utilitarian digital cameras now cost between \$150 and \$350. If you go this way, be sure to have “macro ability,” i.e., the ability to photograph small objects like lesions at 1:1 (where a dime could fill the image frame). If you are already in this “digital age,” you can share your observations with others easily over the internet! This concept can be a valuable tool to send observation data to a fish health manager or pathologist.



This goldfish (*Carassius auratus*) has a focal, well-circumscribed, deep oval ulcer (severity ranking of 5, “severe”), 25 x 18 mm, located on the left mid-flank, above the pelvic fin. The ulcer penetrates through the muscle and reveals rib bones and some of the viscera. The margins of this ulcer do not appear reddened, but the posterior margin has a cream-colored section extending approximately 3-4 mm. The distal edges of the anal and caudal fins are frayed (severity ranking of 3, “moderate”).



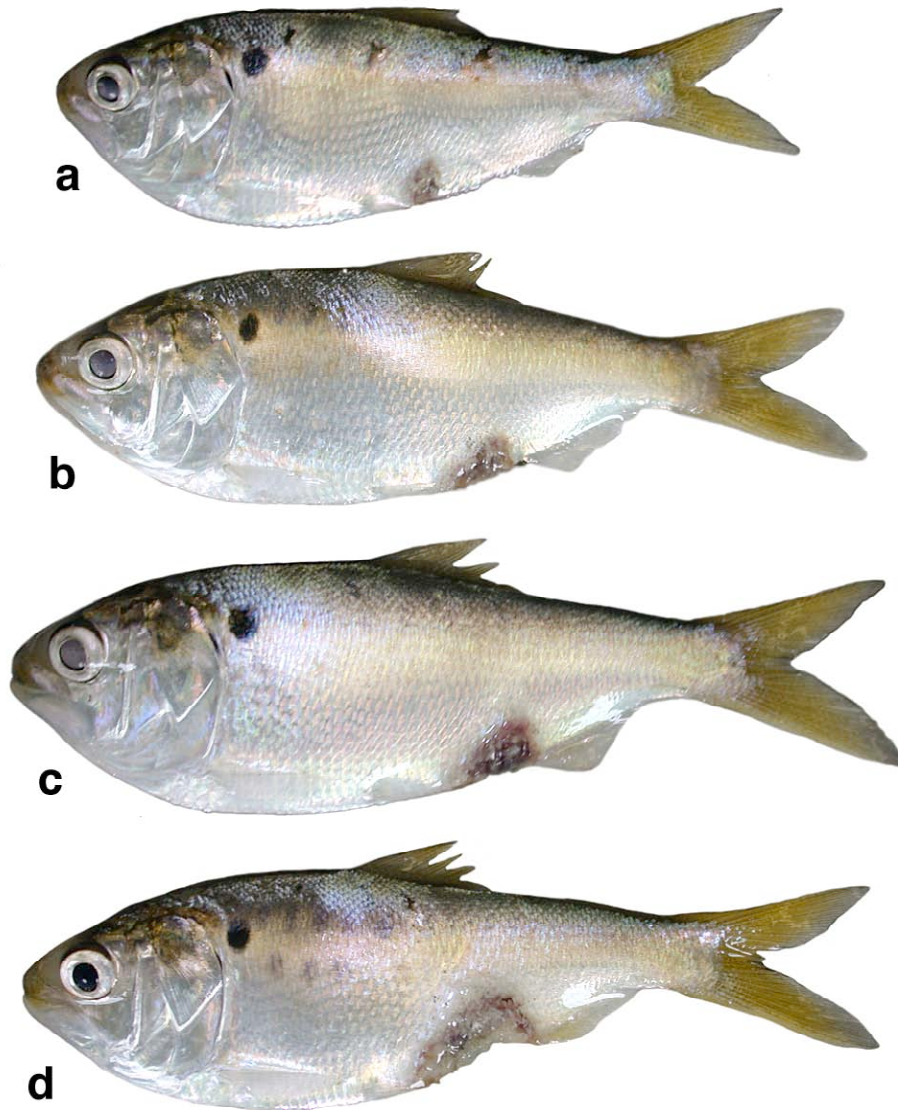
The image of the striped bass (*Morone saxatilis*) on the left has a large (7 x 6 cm), focal, diffuse irregularly-shaped, red lesion (ulcer) that penetrates the skin and scales and reveals underlying dermis (severity ranking 4, “marked”). The lesion is located on the left side of the specimen, extending posteriorly from the distal edge of the pectoral fin; a portion of the lesion extends to the dorsal surface of the fish, anterior to the first dorsal fin. Portions of the lesion, as well as the lesion margins, have superficial black coloration (possibly pigmentation). The lesion margins are irregular. The surface of the lesion has a sandpaper-like appearance. Ventral to the lesion, there is a large, diffuse area of reddening, with no loss of scales, that extends from the branchiostegal membrane to the vent. The fish was in otherwise good body condition. In contrast, the image of the striped bass on the right shows a multifocal, extensively distributed lesion.



This young-of-the-year Atlantic menhaden (*Brevoortia tyrannus*) measured 58 mm total length and presented with spinal deformity. The deformity consisted of curvature in the dorsal-ventral plane, and extended from the middle of the dorsal fin to the caudal fin. Severity ranking 3, "moderate." The animal was alive and otherwise non-remarkable when observed.



This specimen is an Atlantic menhaden (*Brevoortia tyrannus*) that presented with a raised, irregularly-shaped, off-white, cauliflower-like growth (5 x 8 mm) on the right side, extending from the peduncle into the fork of the caudal fin. The mass has well-defined edges and had reddened margins. The smaller photograph on the right shows the same image and indicates the appropriate size tissue sample that would be taken for preservation and subsequent histologic analysis. This sample contains both the abnormal tissue as well as adjacent normal tissue.



This composite image of four young-of-the-year Atlantic menhaden (*Brevoortia tyrannus*) depicts ulcerative lesions with varying ranks of severity. The ulcer seen on fish “a” has a severity ranking of 2, “mild.” Fish “b,” “c” and “d” have severity rankings of 3 (moderate), 4 (marked) and 5 (severe), respectively. Of course these rankings are relative and subject to the experience of the observer. Nevertheless, if personnel follow the basic tenets of severity rankings as illustrated in this guide, there should be a fair degree of consistency between observers, plus or minus one severity ranking. Note that the ulcer on fish “a” is located halfway between the pectoral fin and the anal fin, whereas ulcers on the other three specimens are perianal. These perianal lesions have burgundy margins with central red to grayish areas. Also, careful observation of the image of fish “a” shows a small (3 x 3 mm), diffuse area of reddening on the left side, just below the distal insertion of the dorsal fin.

Selected References:

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- ASTM (American Society for Testing and Materials) 1996. Standard Guide for Fish and Wildlife Incident Monitoring and Reporting. ASTM E1849-96.
- Ferguson, H.W. *Systemic pathology of fish*. Iowa State University Press. Ames, Iowa, 1989.
- Gratzek, J.B., Matthews, J.R. *Aquariology. The science of fish health management*. Tetra Press. Morris Plains, NJ, 1992.
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- Noga, E. *Fish disease diagnosis and treatment*. Iowa State University Press. Ames, Iowa, 2000.
- Reimschuessel, R., Bennett, R.O., Lipsky, M.M. A Classification system for histological lesions. *J. Aquat. Anim. Health.*; 4:135-143, 1992.
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- Ribelin, W.E., Migaki, G. eds., *The pathology of fishes*. The University of Wisconsin Press. 1975.
- Roberts, R.J. ed., *Fish Pathology*. 2nd. Ed. Bailliere Tindall. London, England, 1989.
- Stoskopf, M. *Fish Medicine*. W.B. Saunders Company, Philadelphia, P.A., 1993.

Selected websites:

- Aquatic Pathobiology Center, University of Maryland (<http://aquaticpath.umd.edu>)
- Fish and Human Health in the Chesapeake Bay (<http://mybay.umd.edu>)
- NRAC publication providing an introduction to freshwater aquaculture water quality (<http://mybay.umd.edu/nrac170.pdf>)
- NRAC publication on general principles of fish health management (<http://mybay.umd.edu/fhmgmtnrac111.pdf>)
- Virginia Marine Institute of Marine Science (VIMS) *Pfiesteria* research relating to fish health (<http://www.vims.edu/env/projects/pfiesteria>)
- Fish Health Section of the American Fisheries Society (<http://www.fisheries.org/fhs>)
- USGS press release on Chesapeake Bay fish health issues (<http://biology.usgs.gov/pr/newsrelease/1998/9-23d.html>)
- Bay Journal article (by Karl Blankenship) (4/02) providing historical reference to *Pfiesteria*-related fish health issues (<http://www.bayjournal.com/article.cfm?article=236>)
- Bay Journal article (by Karl Blankenship) (6/05) discussing mycobacteriosis in Chesapeake Bay striped bass (<http://www.bayjournal.com/article.cfm?article=2551>)
- Fathead minnow histology atlas (<http://aquaticpath.umd.edu/fhm>)
- Maryland Department of Natural Resources, Fisheries Service (<http://www.dnr.state.md.us/fisheries>)

Ordering Information for Fish Necropsy Supplies

You can request a catalog from these and other companies so that when you need to make budgeting decisions and/or order supplies, the information is at your fingertips. Mention of suppliers in this guide does not constitute endorsement.

Argent Chemical Laboratories - MS222 and other chemicals/drugs

Telephone- 425 885-3777/800 426 6258
Fax- 425 885-2112
Internet- <http://www.argent-labs.com>
Address- 8702 152 Ave. NE Redmond, WA 98052

Aquanetics Systems Inc. - Aquaculture hardware. Great catalog

Telephone- 619-291-8444
Address- 5252 Lovelock Street, San Diego, CA 92110
Fax- 619-291-8335

Biomedical Research Instruments, Inc. - medical and dissection instruments

Telephone- 301-881-7911/800-327-9498
Address- 12264 Wilkins Avenue, Rockville, MD 20852
Fax- 301-881-8762
Internet- www.biosupplynet.com

Fisher Scientific - General scientific supplies

Telephone- 1-800-766-7009
Address- 600 Business Center Dr., Pittsburgh PA
Internet- www.fishersci.com
Fax- 1-800-926-1166

IDE Interstate Inc. - medical equipment and instruments, pharmaceuticals and proprietaries

Telephone- 1-800-666-8100
Address- 1500 New Horizons Blvd., Amityville, N.Y. 11701
Fax- 1-800-IDE-FAX1
Internet- <http://ideinterstate.com>

Thomas Scientific - General scientific supplies

Telephone- 609 467-2000/800 345-2100
Fax- 609 467-3087
Internet- www.thomasci.com
Address- 99 High Hill Rd. at I-295, P.O. Box 99, Swedesboro, NJ 08085-0099

VWR Scientific Products - General scientific supplies

Telephone- 1-800-932-5000
Address- 405 Heron Drive, P.O. Box 626 Bridgeport, NJ 08014
Fax- 410 418- 8398
Internet- www.vwrsp.com

Acknowledgements

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John Heisler

FIELD FISH LESION WORKSHEET

Date: _____ Collection Time (begin): _____ (end): _____
 Collection Location: _____
 Collection Gear: _____ Biologist: _____

LESION TYPE: Loss Of Scales (LOS); **Reddening**=area of reddening; **Ulcer**=penetrating the skin; **Other** (describe).
LESION LOCATION: **Head**; **Fin** (one or more fins); **Vent**; **Body** (other than head or fins); **Other** (describe; use back of sheet and identify observations by fish #).
LESION PRESENTATION: **Focal** (affected area with defined edges); **multifocal** (more than one focal lesion on the body); **diffuse** (edges of affected area not clearly defined, or affected areas are adjoining/coalesced).
SEVERITY CODES: **0** =normal; **1**=mild; **2**=moderate; and **3**=severe.
T=mark if lesion(s) is due to obvious capture/net trauma.
✓=check if sample(s) taken for supporting studies (or mark for histopathology (H), bacteriology (B), virology (V), etc.)

FISH #	SPECIES	TL (mm)	LESION TYPE			T	LESION LOCATION				LESION PRESENTATION			SEVERITY				✓
			LOS	Reddening	Ulcer		Head	Fin	Vent	Body	Focal	Multifocal	Diffuse	0	1	2	3	
			LOS	Reddening	Ulcer		Head	Fin	Vent	Body	Focal	Multifocal	Diffuse	0	1	2	3	
			LOS	Reddening	Ulcer		Head	Fin	Vent	Body	Focal	Multifocal	Diffuse	0	1	2	3	
			LOS	Reddening	Ulcer		Head	Fin	Vent	Body	Focal	Multifocal	Diffuse	0	1	2	3	
			LOS	Reddening	Ulcer		Head	Fin	Vent	Body	Focal	Multifocal	Diffuse	0	1	2	3	
			LOS	Reddening	Ulcer		Head	Fin	Vent	Body	Focal	Multifocal	Diffuse	0	1	2	3	
			LOS	Reddening	Ulcer		Head	Fin	Vent	Body	Focal	Multifocal	Diffuse	0	1	2	3	
			LOS	Reddening	Ulcer		Head	Fin	Vent	Body	Focal	Multifocal	Diffuse	0	1	2	3	
			LOS	Reddening	Ulcer		Head	Fin	Vent	Body	Focal	Multifocal	Diffuse	0	1	2	3	
			LOS	Reddening	Ulcer		Head	Fin	Vent	Body	Focal	Multifocal	Diffuse	0	1	2	3	
			LOS	Reddening	Ulcer		Head	Fin	Vent	Body	Focal	Multifocal	Diffuse	0	1	2	3	
			LOS	Reddening	Ulcer		Head	Fin	Vent	Body	Focal	Multifocal	Diffuse	0	1	2	3	
			LOS	Reddening	Ulcer		Head	Fin	Vent	Body	Focal	Multifocal	Diffuse	0	1	2	3	
			LOS	Reddening	Ulcer		Head	Fin	Vent	Body	Focal	Multifocal	Diffuse	0	1	2	3	
			LOS	Reddening	Ulcer		Head	Fin	Vent	Body	Focal	Multifocal	Diffuse	0	1	2	3	
			LOS	Reddening	Ulcer		Head	Fin	Vent	Body	Focal	Multifocal	Diffuse	0	1	2	3	
			LOS	Reddening	Ulcer		Head	Fin	Vent	Body	Focal	Multifocal	Diffuse	0	1	2	3	
			LOS	Reddening	Ulcer		Head	Fin	Vent	Body	Focal	Multifocal	Diffuse	0	1	2	3	
			LOS	Reddening	Ulcer		Head	Fin	Vent	Body	Focal	Multifocal	Diffuse	0	1	2	3	

Fish Pathology Report

Lab use only:

Accession number:

Date received:

Cut in by:

DATE SUBMITTED:

SOURCE (NAME AND ADDRESS):

TELEPHONE:

()

COMMON NAME:

SCIENTIFIC NAME:

SOURCE CASE #:

SOURCE TANK I.D.:

AGE:

SEX: _ M _ F

ANIMAL USE:

Exhibit Quarantine
 Research Breeding
 Holding Other

SPECIMEN SUBMITTED:

Live animal Preserved tissue
 Dead animal Gross only
Fixative:

MANNER OF DEATH:

Natural
 Euthan. agent:

PRESERVED TISSUES SUBMITTED (CHECK):

all tissues present

skin brain mouth spleen thyroid
 muscle eye(s) esophagus pancreas gonad
 bone nares stomach head kidney uterus
 cartilage lateral line intestine caudal kidney epigonal
 fin(s) swim bladder liver interrenal
 other:
 other:

HISTORY AND CLINICAL SUMMARY:

- 1) Problems first noticed?
- 2) Symptoms? Eating? Behavior?
- 3) Treatments?
- 4) Water quality?
- 5) Biopsy results?
- 6) Supporting studies (please attach results):

Parasitology Microbiology Hematology Viro-serology Other:

Notes:

Necropsy Work Sheet

Prosector: _____

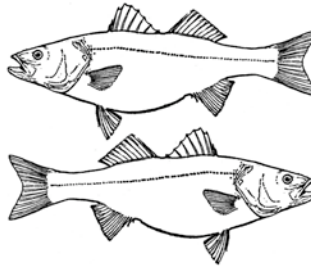
Slides (#): _____

General examination:

(physical condition, fins, orifices, skin, operculum)

Weight (g): _____

Length (mm): _____



Wet mounts:

Skin scrape: _____

Body cavity:

(peritoneum, mesenteries, etc.)

Scrape/smear: _____

Respiratory system:

(gills pseudobranch)

Gill biopsy: _____

Central nervous system and Sensory organs:

(meninges, brain, spinal chord, eyes, nares, lateral line)

Impression smear: _____

Circulatory system:

(pericardial sac, heart, valves, great vessels)

Impression smear: _____

Hematocrit (%): _____

Total plasma solids mg/dl: _____

Gastrointestinal system:

(mouth, esophagus, stomach, small & large intestine, rectum)

Gut scrape: _____

Liver and gall bladder:

Impression smear: _____

Spleen & Pancreas:

Impression smear: _____

Urinary system:

(anterior & posterior kidney, ureters)

Impression smear: _____

Reproductive system:

(external genitalia, gonads, uterus, accessory organs)

Impression smear: _____

Swimbladder:

Int/ext. scrape: _____

Musculo-skeletal system:

(bones, muscle, cartilage; Bouin's fixative for hard structures?)

Scrape/smear: _____

Endocrine system:

(thyroid, interrenal, pituitary)

Impression smear: _____