Protocol Detail Report

Printed By: Eggert, Lori S.
8/23/2016 10:48:04 AM

Report Comments

Protocol Information

Version #

Reference Number	8826		
Protocol Number			
Protocol Type:	Original		
Principal Investigator:	Eggert, Lori S.	**Approval Date:**	
Submittal Date:	8/23/2016	**Effective Date:**	
Author:	Eggert, Lori S.	**Renewal Date:**	
Status:	Submitted	**Next Review Date:**	
Inactive Date:		**Expiration Date:**	

Basic Information **1**

*** Save Often and TOPAZ Problem Tracker** **1.1**

Make sure you click on the "black disk" icon above to save your work as you answer questions. If you receive an error, the program will only be able to restore your work to the last SAVED VERSION.

If you encounter a problem, please send a report to the Topaz Problem Tracker at https://research.missouri.edu/acqa/tracker Someone will respond to your inquiry as soon as possible.

Include Attachments **1.2**

Include ALL relevant attachments to this protocol here. To add the attachment click on the "paperclip" icon in the top right of this question header.

Animal Care and Use Committee Policies **1.3**

For more information about the standard policies of the MU Animal Care and Use Committee please visit the ACQA website.

Protocol Number **1.4**

Protocol Number (filled in by ACUC)

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Reference Number 1.5

Reference Number (automatically assigned)
8826

Principal Investigator 1.6

Eggert, Lori S.	eggertl@missouri.edu	(573) 884-3685 ext

Created By 1.7

Eggert, Lori S.	eggertl@missouri.edu	(573) 884-3685 ext

Department 1.8

Click on the drop down list to select the department.
Biological Sciences

Title 1.9

Protocol Title
Validating the taxonomic and distributional status of Neosho Smallmouth Bass in Missouri, Arkansas, and Oklahoma

Triennial Re-Write 1.10

Is this protocol a re-write of an ACUC protocol that was previously approved at the University of Missouri?
- ☐ Yes
- ☑ No

1.10.1

Yes

1.10.2

No

Species Section 2

Species 2.1

Click on the green plus sign to select the species.

2.1.1

Fish #1

Strain/Stock/Breed 2.1.1.1

List the Strain/Stock/Breed you will be using in this protocol.
Neosho smallmouth bass (Micropterus dolomieu velox) n=160; Other smallmouth bass (M. dolomieu) n=30; Spotted bass (M. punctulatus) n=30; Total = 220

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Authorized Amounts 2.1.1.2

Authorized Amounts Per Pain/Distress Category
(ALWAYS click the green cross to select from a predefined list of options) Please note, the total number of animals requested is the amount of animals you will need for a 3 year period. This number should include all experimental animals plus animals used for colony maintenance (breeders and offspring produced that are not used for experiments). These numbers should match the amounts in the Justify Animal Numbers section. If this is a triennial re-write these amounts should also include any animals on the previous protocol that will be transferred to the new protocol.
As of December 1, 2010, any animals that are not covered by the Animal Welfare Act (laboratory mice and rats, farm animals used in agriculture research, birds, amphibians, and reptiles) do not need USDA Pain/Distress Category classification in the TOPAZ protocol submission form unless they are used in Category E. Category E is pain produced but due to the nature of the research, pain relieving drugs cannot be used.

If you are creating a protocol using non-USDA species, you may choose “Undefined (non-covered species only)”. Please continue to complete the question regarding whether or not the proposed use is USDA Category E (unrelieved pain/distress) regardless of the species of animals used.

Pain Category	Authorized	Requested	On Order	Received	Available
Undefined (non-covered	220	0	0	0	220
Totals		0	0	0	220

USDA Category E 2.1.1.3

Have any animals been included in Category E (above)?

- ☐ Yes
- ☑ No

2.1.1.3.1

Yes

2.1.1.3.2

No

Age/Weight of Animals to be Used 2.1.1.4

Age/Weight:

All fishes sampled will be adults - no fish less than 200 mm in length will be sampled for this study.

Proposal Overview 3

Purpose 3.1

Purpose of the study:

The taxonomy of this fish has been questioned for many years - we will use genetic and genomic data to compare it to other smallmouth bass.

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Value 3.2

Please provide the information necessary to allow the ACUC to evaluate the objectives of the study against potential animal welfare concerns.

This will allow the Missouri Department of Conservation to better manage this natural resource in the field.

Lay Term Description of Experimental Design 3.3

To put something in layman's terms is to describe a complex or technical issue using words and terms that the average individual (someone without professional training in the subject area) can understand. This section should be written so that someone with a 10th grade science education can easily understand the project.

We will collect Neosho smallmouth bass ((Micropterus dolomieu velox) in their natural range where habitat is most suitable, and where hybrids with other species of bass are unlikely to occur. We will also collect representatives of other forms of smallmouth bass and spotted bass (M. punttulatus). Fish will be cught by hook and line and placed in a cooler with water pulled from the stream when not being handled. A picture of each fish will be taken by placing it in a pan containing wter from the stream, and measurements will be taken using calipers. A clip will be taken from the upper caudal fin (no more than 1/3 of the fin) using scissors. The total handling time is approximately 2 minutes. Fish will be moved using dip nets and will be released at the site of capture.
The collected tissue will be used for genetic and genomic analyse to address our research questions.

Scientific Description of Experimental Design 3.4

In language scientific colleagues outside of your discipline would understand, provide a step-by-step, general description of the animal experiments you will perform. Include an overview of all procedures and manipulations, and explain why they must be performed. For complicated experimental designs, a flow chart, diagram, or table is strongly recommended to help the IACUC understand what is proposed. DO NOT describe details of the surgical procedures, or non-surgical manipulations here. Such details are requested later in the form.

Same as Lay Term Description. s

Justify 4

Attaching Files/Documents 4.1

Attach all relevant files and images under Section 1.

Justify Use of Animals in your Research 4.2

Justify the use of animals for your experimental goals. DO NOT describe details of the experimental design or justify animal numbers here.

Because it involves genetic studies of the species in its natural range, this project could not be done without collecting samples in the field. There is no known scientific archive of samples that could be used.

Justify Animal Species 4.3

Justify the choice of species for your study.

We are using the species that are the target of the study - we could not do this with surrogate species.

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Justify Animal Numbers 4.4

Attachments: Animal Numbers Justification The Logical Determination of "N" in Animal Experimentation Non-Statistical Approach for Calculating the Optimum Number of Animals Needed in Research Statistics and the Issue of Animal Numbers in Research JUSTIFY ANIMAL NUMBERS EXAMPLE

Justify numbers of animals to be used (attach timeline or flow chart and power analysis, if possible, to describe study groups). This section should include a description of animals used for colony maintenance (breeders and all offspring produced) as well as a description of experimental animal numbers. Total numbers should match the requested numbers in the Authorized Amounts section.

We will collect appoximately 100 samples in Missouri, 40-60 in Arkansas and Oklahoma. We will also collect 20-30 samples of other small mouth bass and 20-30 spotted bass. The maximum number of fishes collected will be 220, which represents the minimum number of samples needed for statistical significance in our study.

Project Information 5

Attaching Files/Documents 5.1

Attach all relevant files and images under Section 1.

Protocol Associates 5.2

You must add Protocol Associates HERE - (CLICK on the green cross to select from a list of staff. The PI needs to be included in this section as well.) If you need staff members added to the list e-mail acuc@missouri.edu. All personnel identified as "key associate" or "Co-PI" will receive correspondence regarding the protocol, including reviewer's comments.

Anderson, Michelle

Responsibilities	Michelle, an MDC employee, will be responsible for fish sample collection and DNA analysis and will consult with Lori Eggert on data analysis, data interpretation and manuscript preparation.
Comments	

☐ **Co-Investigator** ☑ **Key Associate** ☐ **Authorized to Order Animals**

Eggert, Lori S.

Responsibilities	Lori Eggert will supervise the project, including experimental design, DNA analyses, data analyses, data interpretation and manuscript publication.
Comments	

☑ **Co-Investigator** ☐ **Key Associate** ☐ **Authorized to Order Animals**

Training and Qualifications 5.3

Provide a description of the training and qualifications for each individual listed above under Protocol Associates. Provide adequate detail to allow the ACUC to determine if the individual has adequate training and experience with the species and procedures to perform their role proficiently. If they do not have prior training or experience, how will this be obtained?

Michelle Anderson and Lori Eggert have been trained in fish handling and fin clip collection by Jeff Koppelman, MDC Resource Scientist, who has 20 years of experience in fish handling and fish resource management. Lori has worked with animal DNA analyses since 1992.

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Training Requirements 5.4

Attachments: Occupational Health and Safety Program

Note: The ACUC required Basic Training can be found at: https://www.research.missouri.edu/acqa/. This training must be updated every three years in order to receive protocol approval.

Note: It is the Principal Investigator's responsibility to ensure that all persons listed in Protocol Associates above participate in the MU Occupational Health and Safety Program. See Section 7:020 MU Business Policy and Procedures Manual for details. For enrollment procedures visit the OHSP website.

Funding Source 5.5

What is the funding source for this project? (Note: If funded internally or by a non-peer-reviewing agency, a peer review of scientific merit may be required.)
Missouri Department of Conservation grant

Wild Animals 5.6

Are WILD ANIMALS to be used or studied?

- [x] Yes
- [ ] No

5.6.1

Yes

Permits 5.6.1.1

Scientific Collection Permit # and issuing agency:
Michelle Anderson has a collecting permit in Missouri (MDC #17002), and a collectors permit in Arkansas (Arkansas Fish and Game #060720163). The permit application is pending in Oklahoma, but may not be needed if we are able to obtain samples from our collaborators.

5.6.2

No

Client-Owned Animals 5.7

Are CLIENT-OWNED animals to be used or studied?

- [ ] Yes
- [x] No

5.7.1

Yes

5.7.2

No

Animal Husbandry 6

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Facility 6.1

In which animal facility will animals be housed? (Give specific building(s) and room number(s) if known):
Animals will not be taken from their natural habitat - no housing is required.

Housing Outside of Facility 6.2

Will animals be housed anywhere other than a designated animal housing facility for more than 12 hours (e.g., a laboratory)?

- [ ] Yes
- [x] No

6.2.1

Yes

6.2.2

No

Use Areas 6.3

List all places (buildings and room numbers) where non-surgical procedures to animals will be performed, and the type of procedure(s) to be performed in each area:
no surgical procedures will be performed

Surgery - Facility 6.4

List all surgery facilities that will be used.
n/a

Transportation Between Animal Housing/Use Facilities 6.5

Will animals be transported with a private vehicle between animal housing/use facilities?

- [ ] Yes
- [x] No

6.5.1

Yes

6.5.2

No

Exceptions to the Guide or Ag Guide 6.6

Does this protocol contain any exceptions to either the Guide or the Ag Guide? This includes nonstandard husbandry, single housing of social species, prolonged cage change interval, altering sanitation schedule, food/fluid restriction, multiple major survival surgery, and prolonged restraint.

- [ ] Yes
- [x] No

6.6.1

Yes

6.6.2

No

Nonstandard Husbandry **6.7**

Explain any nonstandard husbandry requirements (feeding, watering, housing, environment).
NOTE: Singly housing social species is considered non-standard husbandry. Wire-bottomed caging is considered non-standard housing for rodents.

Length of Departure **6.8**

List the length of time the animal will undergo nonstandard husbandry.

Food and/or Fluid Restriction **6.9**

Will FOOD or FLUIDS be restricted?

- [ ] Yes
- [x] No
- [ ] Overnight only

6.9.1

Yes

6.9.2

No

6.9.3

Overnight only

Description of Non-Surgical Procedures **7**

Attaching Files/Documents **7.1**

Attach all relevant files and images under Section 1.

Sample Collection **7.2**

Will samples, such as blood or tissues, be collected from live animals? (Include sampling for genotyping.)

- [x] Yes
- [ ] No

7.2.1

Yes

Sample Type **7.2.1.1**

Type of sample(s):
Fin clip - no more than 1/3 of the upper caudal fin will be taken from each fish.

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Sample Volume 7.2.1.2

Volume of sample(s):
< 1 gram

Sampling Frequency and Duration 7.2.1.3

Frequency of collection and for how long:
Once

Sampling Method 7.2.1.4

Method of collection:
The fin clip will be taken with scissors that are cleaned with ethanol between uses

7.2.2

No

Induced or Spontaneous Neoplasia 7.3

Will induced or spontaneous neoplasia occur in live animals?

- ☐ Yes
- ☑ No

7.3.1

Yes

7.3.2

No

Animal Restraint 7.4

Will there be animal restraint lasting longer than 60 minutes?

- ☐ Yes
- ☑ No

7.4.1

Yes

7.4.2

No

Additional Explanation of Non-Surgical Procedures 7.5

Describe all animal treatments or manipulations including, injections, behavior studies, imaging, non-routine blood collections, exercise protocols, etc.

n/a

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(OFFICE USE ONLY - SKIP THIS QUESTION, DO NOT ANSWER) 7.6

This information is now listed in the "Test Substances" Section of the form.

Substance	Amount/Dosage	Route/Volume of Admin.

Test Substances 8

Test Substances Table 8.1

List the test substances you will give the animals here (including vehicles given to controls):

Substance	Amount/Dose/Volume	Route	Hazard (yes or no)	Pharmaceutical Grade (yes or no)
None	none	none	no	

Non-Pharmaceutical Grade Substances 8.2

For those substances that are marked "no" as pharmaceutical grade, list a justification in the space below. Also, include instructions for how they will be mixed to maintain sterility and adjust pH.

none

Hazardous Agent 8.3

If you marked "yes" under Hazard, please complete the "Hazardous Materials" Section that follows.

Hazardous Materials 9

Hazardous Materials 9.1

Will you use any of the below hazards when working with live animals?
Biological Hazards (ex. pathogens, biological toxins, recombinant DNA, cell/cell lines, blood/blood products, tissues and fluid, etc.)
Chemical Hazards (ex. non-biological hazmat, carcinogens, teratogens, mutagens, nanoparticles, etc.)
Radiation Hazards (ex. radiolabeled chemicals, irradiators, lasers, etc.)

- ☐ Yes
- ☑ No

9.1.1

Yes

9.1.2

No

Description of Anesthetic Procedures 10

Anesthetics, Preanesthetics & Tranquilizers 10.1

Will any anesthetics, preanesthetics, or tranquilizers be used?

- ☐ Yes
- ☑ No

10.1.1

Yes

10.1.2

No

Description of Surgical Procedures **11**

Attaching Files/Documents **11.1**

Attach all relevant files and images under Section 1.

Surgical Procedures **11.2**

Will there be any surgical procedures?

- ☐ Yes
- ☑ No

11.2.1

Yes

11.2.2

No

Potential Pain or Physical Stress **12**

AWA Regulations **12.1**

Note: Animal Welfare Act regulations define a painful procedure as "any procedure that would reasonably be expected to cause more than slight or momentary pain ... in a human being to which that procedure was applied, that is, pain in excess of that caused by injections or other minor procedures." Procedures reasonably expected to cause pain in the absence of anesthetics or pain relieving drugs should be considered to have the potential to cause pain even with the use of such drugs.

Potential Side-Effects and Adverse Health Effects **12.2**

Describe any potential side-effects or anticipated adverse health effects of all procedures listed in the preceding sections: animal husbandry, description of non-surgical procedures, anesthetic procedures, and surgical procedures.

We do not anticipate any adverse health effects - we will not collect from fish < 250 mm in size, and will clean the scissors with ethanol between uses to minimize the chance that pathogens woulc be transferred between fishes. If a fish swallows a hook, we will use a needle-nose tool to remove it. If we cannot reach the hook with the tool, we will cut the line and allow the hook to stay in the fish. Studies have shown that a much larger proportion of fish that deeply swallow a hook survive if the hook is left in the fish than if it is removed (http://fishandboat.com/images/pages/qa/fishing/catch_rel.htmv).

Assurance of Limited Discomfort and Pain **12.3**

Describe how it is assured that discomfort and pain are limited to that which is unavoidable for the conduct of this experimentation.

We will handle the fish for the minimum amount of time possible and will not release fish < 250 mm in size without collecting fin clips. Studies have shown that fin-clipping is a low risk procedure that does not debilitate the fish.
Carmichael, G.J. et al. (1986) Transactions of the American Fisheries Society 115:455-459.
Tranquilli, J.A. and William, .F. (1982) North American Journal of Fisheries Management 2:184-187.

Disposition of Animals **13**

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Animal Disposition 13.1

Check all that apply

- [ ] Adoption (See MU adoption policy)
- [ ] Market
- [ ] Euthanasia
- [ ] Transfer to different project, PI, or another institution
- [x] Other

13.1.1

Adoption (See MU adoption policy)

13.1.2

Market

13.1.3

Euthanasia

13.1.4

Transfer to different project, PI, or another institution

13.1.5

Other

Other, Animal Disposition 13.1.5.1

Explain other animal disposition below.

Fishes will be released after sample collection at the site of capture.s

Literature Search 14

Attaching Files/Documents 14.1

Attach relevant files to Section 1 (you will need to click on the “paperclip” icon to do so).

Painful Procedures 14.2

Any procedure that may potentially cause more than momentary or slight pain or distress requires a literature search for animal alternatives.

Are you performing any procedures that may potentially cause more than momentary or slight pain or distress?

- [x] Yes
- [ ] No

14.2.1

Yes

USDA Covered Species 14.2.1.1

Does this protocol utilize animals covered by the Animal Welfare Act or assigned to Category E? (AWA covered species include all warm blooded animals except birds, rats of the genus Rattus, and mice of the genus Mus, bred for use in research, horses not used for research purposes, and other farm animals.)

- [ ] Yes, includes USDA covered species or Category E
- [x] No

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14.2.1.1.1

Yes, includes USDA covered species or Category E

14.2.1.1.2

No

Refinements 14.2.1.1.2.1

The Guide requires that the 3R's (replacement, reduction, and refinement) be considered. Replacement and reduction have already been addressed. Refinement refers to modifications of husbandry or experimental procedures to enhance animal well-being (e.g., enrichment) and
minimize or eliminate pain and distress (e.g., alternative methods, analgesia, etc.).

Please describe any refinements used or considered for this protocol.
These studies have shown that fin clipping is a low-risk procedure that doees not debilitate the fish:
Carmichael, G.J. et al. (1986) Transactions of the American Fisheries Society 115:455-459.
Tranquilli, J.A. and William, .F. (1982) North American Journal of Fisheries Management 2:184-187.

14.2.2

No

Exceptions to the Standards and Regulations 15

Exceptions to the Standards and Regulations 15.1

Exceptions to the standards and regulations promulgated under the Animal Welfare Act, for the care and use of warm-blooded animals must be specified and explained by the principal investigator in the space below.

N/A

Pre-submission Veterinary Review 16

Pre-submission Veterinary Review 16.1

Complete these steps:
If this is a new protocol or triennial rewrite a pre-submission veterinary review is required.
Please click the “Submit” icon (blue and yellow arrows) above to submit the protocol for pre-submission veterinary review.
Confirm that you want to submit by clicking on “set status”
The e-signature box will appear and the current logged in user must enter his/her username and password, click on “save”
By hitting “send” on the “animal protocol submitted for approval” e-mail that appears, notification of submission will be sent via e-mail to the ACUC Coordinator. The ACUC Coordinator will notify the veterinary reviewer that you require a pre-submission review. If you do not hear back from the pre-submission reviewer within 3 days please contact the veterinary reviewer directly.

☑ Check this box after completing steps 1-5.

16.1.1

Check this box after completing steps 1-5.

Investigator Assurances 17

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ABSL-2 Assurance **17.1**

I will provide training to the husbandry/veterinary staff at least 48 hours prior to exposing animals to a biohazard regarding (but not limited to): the health hazards and symptoms of the biohazard(s) being used; husbandry related research specific SOP's (e.g. handling live exposed animals and contaminated cages); and animal/carcass disposition.

- ☐ Yes, I will meet the requirements of this statement.
- ☐ No, I will not meet the requirements of this statement.
- ☐ Not Applicable

17.1.1

Yes, I will meet the requirements of this statement.

17.1.2

No, I will not meet the requirements of this statement.

17.1.3

Not Applicable

E-signature Assurance **17.2**

1. The information provided herein is accurate to the best of my knowledge.

2. Procedures involving vertebrate animals will be performed only by trained or experienced personnel, or under the direct supervision of trained or experienced persons.

3. Any change in the care and use of vertebrate animals involved in this protocol, will be promptly forwarded to the MU ACUC for review; such changes will not be implemented until the committee's approval is obtained.

4. The number of animals proposed is the minimum necessary to conduct valid experimentation.

5. I ensure that I am not unnecessarily duplicating previous experiments.

6. I have considered alternative methods to using animals.
7. I understand that animal housing must be coordinated with the facility veterinarian and/or facility manager and that approval of this protocol does not guarantee space to house animals.

- ☐ Yes, I have met the requirements of the 7 items listed above.
- ☐ No, I have not met the requirements of the 7 items listed above.

17.2.1

Yes, I have met the requirements of the 7 items listed above.

17.2.2

No, I have not met the requirements of the 7 items listed above.