DEPARTMENT OF HEALTH & HUMAN SERVICES

PUBLIC HEALTH SERVICE
NATIONAL INSTITUTES OF HEALTH

FOR US POSTAL SERVICE DELIVERY:
Office of Laboratory Animal Welfare
6700B Rockledge Drive, Suite 2500, MSC 6910
Bethesda, Maryland 20892-6910
Home Page: http://grants.nih.gov/grants/olaw/olaw.htm

FOR EXPRESS MAIL:
Office of Laboratory Animal Welfare
6700B Rockledge Drive, Suite 2500
Bethesda, Maryland 20817
Telephone: (301) 496-7163
Facsimile: (301) 402-7065

July 29, 2022

Re: Animal Welfare Assurance
A3194-01 [OLAW Case 1P]

Matthew Tremblay, Ph.D.
Chief Operating Officer
Scripps Research Institute
10550 North Torrey Pines Road
Mail Code: BCC-506
La Jolla, CA 92037

Dear Dr. Tremblay,

The Office of Laboratory Animal Welfare (OLAW) acknowledges receipt of your July 12, 2022 letter providing additional requested information regarding an adverse event within the animal care and use program at The Scripps Research Institute. According to the additional information provided, OLAW understands that the veterinary intervention consisted of transferring the 9 live frogs into clean tanks by 9:15 am and performing a veterinary assessment.

Although the frogs began to develop slightly more muscle tone, the veterinarians cautioned the investigator that stress is known to induce gram negative septicemia in frogs. At 2:50 pm, one frog was euthanized per the lab manager's request; at 3:50 pm, four frogs were found dead; and at 3:57 pm, the remaining 4 frogs were euthanized per veterinary instruction.

OLAW appreciates your cooperation as Institutional Official, and your program's transparency. Based on the original and additional information provided, OLAW is satisfied that appropriate actions were taken by The Scripps Research Institute to investigate this incident and prevent recurrence. We find no cause for further action by this office. Thank you.

Sincerely,

Brent C. Morse -S

Digitally signed by Brent C. Morse -S
Date: 2022.07.29 10:04:21 -04'00'

Brent C. Morse, DVM
Director
Division of Compliance Oversight
Office of Laboratory Animal Welfare

cc: IACUC contact

McCoy, Devora (NIH/OD) [E]

From: McCoy, Devora (NIH/OD) [E]
Sent: Friday, July 29, 2022 10:42 AM
To: mtremblay@scripps.edu
Cc: OLAW Division of Compliance Oversight (NIH/OD); juanct@scripps.edu; (b) (6) (b) (6)
Subject: OLAW Case A3194-1P
Attachments: A3194-1P.pdf

Good morning Dr. Tremblay,

Attached please find Dr. Morse's final response to OLAW Case A3194-1P. If you have any questions, feel free to contact us by phone or by e-mail.

Best,
Devora

Devora McCoy, BS, MBA
Program Analyst
Division of Compliance Oversight
Office of Laboratory Animal Welfare
National Institutes of Health
301-435-2390

McCoy, Devora (NIH/OD) [E]

From: McCoy, Devora (NIH/OD) [E]
Sent: Monday, July 18, 2022 11:42 AM
To: mtremblay@scripps.edu
Cc: juanct@scripps.edu; OLAW Division of Compliance Oversight (NIH/OD)
Subject: OLAW Case A3194-1P
Attachments: A3194-1P.pdf

Good morning Dr. Tremblay,

Attached please find Dr. Morse's final response to OLAW Case A3194-1P. If you have any questions, feel free to contact us by phone or by e-mail.

Best,
Devora

Devora McCoy, BS, MBA
Program Analyst
Division of Compliance Oversight
Office of Laboratory Animal Welfare
National Institutes of Health
301-435-2390

A3194-1P

Scripps Research

IACUC Office

10550 North Torrey Pines Road
La Jolla, California 92037
Mail Drop: TPC 3
iacuc@scripps.edu

CONFIDENTIAL

July 12, 2022

Brent Morse, D.V.M.
Director, Division of Compliance Oversight
Office of Laboratory Animal Welfare
National Institutes of Health
6700B Rockledge Drive
Suite 2500, MSC 6910
Bethesda, MD 20817
olawdco@mail.nih.gov

RE: Animal Welfare Assurance D16-00122 (A3194-01) – OLAW Case #A3194-1P

Dear Dr. Morse:

In response to your letter dated July 01, 2022, please find the additional clarifying information.

1) Veterinary intervention consisted of:
   a. transfer of the 9 live frogs into clean tanks containing water of adequate quality for supporting frogs and isolating them from the unknown toxic agent;
   b. veterinary assessment noting that the frogs were lethargic and their limbs were somewhat flaccid; no additional clinical signs were present;
   c. upon transfer to the clean tanks, the frogs began to develop slightly more muscle tone however the veterinarians cautioned the investigator that stress is known to induce gram negative septicemia in frogs, which was likely to develop within 24-72 hours and be manifested as a cutaneous hemorrhage along with skin sloughing.

2) After transfer into clean tanks by 9:15 am the following timeline was recorded:
   a. the remaining 9 frogs, although responsive, did not improve significantly;
   b. at 2:50 pm, one frog was euthanized per the lab manager's request;
   c. at 3:50 pm, four frogs were found dead; and
   d. at 3:57 pm, the remaining 4 frogs were euthanized per veterinary instruction.

We expect that this additional information addresses outstanding concerns. If further information is needed, please don't hesitate to contact us at iacuc@scripps.edu.

Sincerely,

(b) (6)

Matt Tremblay, Ph.D.
Chief Operating Officer
Institutional Official

(b) (6)

Juan Carlos de la Torre, Ph.D.
Chair, Institutional Animal Care and Use Committee
Professor, Dept. of Immunology and Microbiology

McCoy, Devora (NIH/OD) [E]

From: IACUC Department <iacuc@scripps.edu>
Sent: Tuesday, July 12, 2022 2:53 PM
To: McCoy, Devora (NIH/OD) [E]
Cc: OLAW Division of Compliance Oversight (NIH/OD); Matt Tremblay; Juan Carlos de la Torre; (b) (6)
Subject: [EXTERNAL] RE: OLAW Case A3194-1P
Attachments: OLAW Ltr - DNC - frogs addtl info 07-12-22.pdf

Devora,

The attached letter provides the additional clarifications requested. We expect that this addresses any outstanding concerns, however let us know if additional information is required.

Thanks and have a good afternoon

(b) (6)

From: McCoy, Devora (NIH/OD) [E] <devora.mccoy@nih.gov>
Sent: Tuesday, July 5, 2022 5:38 AM
To: Matt Tremblay <mtremblay@scripps.edu>
Cc: OLAW Division of Compliance Oversight (NIH/OD) <olawdco@od.nih.gov>; Juan Carlos de la Torre <juanct@scripps.edu>; (b) (6)
Subject: OLAW Case A3194-1P

Good morning Dr. Tremblay,

Attached please find Dr. Morse's response to OLAW Case A3194-1P. Please note that additional information is being requested by July 29, 2022.
If you have any questions, feel free to contact us by phone or by e-mail.

Best,
Devora

Devora McCoy, BS, MBA
Program Analyst
Division of Compliance Oversight
Office of Laboratory Animal Welfare
National Institutes of Health
301-435-2390

DEPARTMENT OF HEALTH & HUMAN SERVICES

PUBLIC HEALTH SERVICE
NATIONAL INSTITUTES OF HEALTH

FOR US POSTAL SERVICE DELIVERY:
Office of Laboratory Animal Welfare
6700B Rockledge Drive, Suite 2500, MSC 6910
Bethesda, Maryland 20892-6910
Home Page: http://grants.nih.gov/grants/olaw/olaw.htm

FOR EXPRESS MAIL:
Office of Laboratory Animal Welfare
6700B Rockledge Drive, Suite 2500
Bethesda, Maryland 20817
Telephone: (301) 496-7163
Facsimile: (301) 402-7065

July 1, 2022

Re: Animal Welfare Assurance
A3194-01 [OLAW Case 1P]

Matthew Tremblay, Ph.D.
Chief Operating Officer
Scripps Research Institute
10550 North Torrey Pines Road
Mail Code: BCC-506
La Jolla, CA 92037

Dear Dr. Tremblay,

The Office of Laboratory Animal Welfare (OLAW) acknowledges receipt of your June 28, 2022 letter reporting an instance of noncompliance with the PHS Policy on Humane Care and Use of Laboratory Animals at The Scripps Research Institute. Your letter supplements the information contained in the prompt preliminary telephone report to OLAW on March 4, 2022. According to the information provided, OLAW understands that on the morning of March 03, 2022, 21 out of 30 adult Xenopus were found dead. Irrespective of veterinary intervention, the remaining nine animals were flaccid and barely moving. Four of these animals were found dead later that day and the remaining five had not improved and were euthanized per veterinary recommendation. The animals associated with this incident were supported in part by NIH grants, with no unallowable costs charged to the awards.

Animal records noted that all frogs were observed to be in good health as late as 4:00 pm the day before, March 02, 2022. Water quality parameters noted daily were within normal range for pH, temperature, and conductivity, except for a brief increase in chlorine/chloramine on April 30, 2022. The equipment was functioning properly. Water samples were sent to a laboratory for further analysis and the results were consistent with measurements made 'in-house'. It was determined that this acute mortality incident resulted from exposure to normal components of the municipal supply water. There were no husbandry practice errors or equipment malfunction. Varying levels of chloramines are thought to be the most likely underlying cause. Current Scripps water filtration procedures have been in place since 2009 without major incidents.

Potential corrective and preventive measures: It is understood that adult frogs will no longer be required on this project. However, the following recommendations were provided should Scripps decide to re-establish a Xenopus colony:

a) Implement a reverse osmosis supply water purification system in addition to the existing four-stage filtration system.
b) In the absence of a reverse osmosis water purification system, upgrade the current supply water prefilters to include catalytic carbon-activated filters.
c) Implement use of the manufacturer's Monitoring System and Sensors that send alarms via emails and texts if parameters are out of range.
d) Avoid use of copper pipes for supply water to prevent copper leaching into the water.

e) If compatible with recirculating water systems, consider use of commercially available detoxifiers, particularly in emergency situations, to bind and neutralize ammonia, chlorine/chloramine, and nitrite.

OLAW appreciates the consideration of this matter by The Scripps Research Institute, which was consistent with the philosophy of institutional self-regulation. Based on the information provided, OLAW is satisfied that appropriate actions have been taken to investigate this incident and prevent recurrence. OLAW concurs that the incident warranted reporting. **OLAW is requesting additional clarifying information.** Please provide a timeline from when the nine remaining animals were found "flaccid and barely moving" to when "Four of these animals were found dead later that day". Also, please provide a brief description of the "veterinary intervention" provided. We appreciate being informed of this matter and ask that you provide the requested additional information **by July 29, 2022.** Please refer to OLAW case #A3194-1P in your reply. Thank you.

Sincerely,

Brent C. Morse -S

Digitally signed by Brent C. Morse -S
Date: 2022.07.01 12:53:27 -04'00'

Brent C. Morse, DVM
Director
Division of Compliance Oversight
Office of Laboratory Animal Welfare

cc: IACUC contact

A3194-1P

Scripps Research

IACUC Office

10550 North Torrey Pines Road
La Jolla, California 92037
Mail Drop: TPC 3
iacuc@scripps.edu

CONFIDENTIAL

June 28, 2022

Brent Morse, D.V.M.
Director, Division of Compliance Oversight
Office of Laboratory Animal Welfare
National Institutes of Health
6700B Rockledge Drive
Suite 2500, MSC 6910
Bethesda, MD 20817
olawdco@mail.nih.gov

RE: Animal Welfare Assurance D16-00122 (A3194-01)

Dear Dr. Morse:

We are writing to finalize the oral report to OLAW of March 04, 2022 with regards to unanticipated death of 21 Xenopus maintained within an Aquaneering Aquatic Housing Rack system. The incident was reported to the IACUC at their March 16, 2022 meeting and they were kept apprised of the investigation. A final report was provided to the IACUC at their June 15, 2022 convened meeting. An overview of the event and information reviewed by the IACUC is provided below.

1) On the morning of March 03, 2022, 21 out of 30 adult Xenopus, maintained for breeding to obtain required experimental tadpoles, were found dead. Irrespective of veterinary intervention, the remaining 9 animals were flaccid and barely moving. Four of these animals were found dead later that day and the remaining 5 had not improved and were euthanized per veterinary recommendation.

2) Animal records noted that all frogs were observed to be in good health as late as 4:00 pm the day before, March 02, 2022. Water quality parameters noted daily were within normal range for pH, temperature, and conductivity, except for a brief increase in chlorine/chloramine on April 30, 2022. Equipment function is checked daily by husbandry staff and monthly by Aquaneering. Following the incident, Aquaneering was consulted, and a determination made that the equipment was functioning properly.

3) Water samples were sent to Weck Laboratories for further analysis and the results were consistent with measurements made 'in-house'. The San Diego municipal supply water consistently meet EPA standards for potable water and the Aquaneering four-stage water filtration system, with biofilter, is designed to provide water quality conditions suitable for the maintenance of Xenopus

4) Based on the results of exhaustive testing, it was determined that this acute mortality incident resulted from exposure to normal components of the municipal supply water. There were no husbandry practice errors or equipment malfunction. Varying levels of chloramines are thought to be the most likely underlying cause. Current Scripps water filtration procedures have been in place since 2009 without major incidents, attributed to supply water, however Aquaneering representatives indicated that problems with municipal supply water do occur locally and nationally from time to time.

5) During the time period of analysis of this incident, the principal investigator obtained required experimental tadpoles from a commercial source and determined that adult frogs would no longer require maintenance at the institute. However, the final report provided the following recommendations should Scripps decide to re-establish a Xenopus colony.
    a. Implement a reverse osmosis supply water purification system in addition to the four-stage Aquaneering filtration system.
    b. In the absence of a reverse osmosis water purification system, upgrade the current supply water prefilters to include catalytic carbon-activated filters.
    c. Implement use of Aquaneering Monitoring System and Sensors that send alarms via emails and texts if parameters are out of range.
    d. Avoid use of copper pipes for supply water to prevent copper leaching into the water.
    e. If compatible with recirculating water systems, consider use of commercially available detoxifiers, such as AmQuel, particularly in emergency situations, to bind and neutralize ammonia, chlorine/chloramine, and nitrite.

6) The Xenopus affected by this incident were maintained as a source of experimental tadpoles supported in part by NIH grants R01EY031597 and R21NS114975 with no unallowable costs charged to this award.

The Scripps Research Institute remains committed to ensuring the proper care and treatment of animals used in research and teaching at the institution. If you require any further information, please contact us at iacuc@scripps.edu.

Sincerely,

(b) (6)

Matt Tremblay, Ph.D.
Chief Operating Officer
Institutional Official

(b) (6)

Juan Carlos de la Torre, Ph.D.
Chair, Institutional Animal Care and Use Committee
Professor, Dept. of Immunology and Microbiology

Wolff, Axel (NIH/OD) [E]

From: OLAW Division of Compliance Oversight (NIH/OD)
Sent: Wednesday, June 29, 2022 7:09 AM
To: (b) (6)
Cc: OLAW Division of Compliance Oversight (NIH/OD)
Subject: RE: Report for D16-00122 (A3194-01)

Thank you for this report, (b) (6) We will send a response soon.
Axel Wolff

From: (b) (6)
Sent: Tuesday, June 28, 2022 3:13 PM
To: OLAW Division of Compliance Oversight (NIH/OD) <olawdco@od.nih.gov>
Cc: Matt Tremblay <mtremblay@scripps.edu>; Juan Carlos de la Torre <juanct@scripps.edu>; (b) (6)
(b) (6)
Subject: [EXTERNAL] Report for D16-00122 (A3194-01)

Dear Dr. Morse,

Please see the attached final report for Animal Welfare Assurance D16-00122 (A3194-01) as a follow up to the oral report of March 03, 2022.

If you have any questions or concerns, please write or call.

Sincerely, (b) (6)

(b) (6)

A3194-1P

Initial Report of Noncompliance

By: BCM

Date: **3/4/2022**

Time: Voicemail

Person Reporting: (b) (6)
Telephone #: (b) (6)
Fax #:
Email:

Name of Institution: __Scripps Research Inst.
Assurance number: ___A3194_____

Did incident involve PHS funded activity? __?____
Funding component: _____
Was funding component contacted (if necessary): _____

21 frogs died overnight. Remaining 9 frogs not doing well. Aquarium system just serviced by manufacturer.

Species involved: Xenopus laevis
Personnel involved: ??
Dates and times: ?
Animal deaths: yes

Projected plan and schedule for correction/prevention (if known): _____

Manufacturer techs returning.

Projected submission to OLAW of final report from Institutional Official:

< 60 days

OFFICE USE ONLY
Case #_____