



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

February 7, 2022

Re: Animal Welfare Assurance  
A3865-01 [OLAW Case 1C]

Professor Joseph Incandella  
Vice Chancellor for Research  
University of California-Santa Barbara  
Office of Research  
(b) (4) Cheadle Hall  
Santa Barbara, CA 93106-2050

Dear Dr. Incandella,

The Office of Laboratory Animal Welfare (OLAW) acknowledges receipt of your January 20, 2022 letter reporting an adverse event within the animal care and use program at the University of California – Santa Barbara. According to the information provided, OLAW understands that, on July 9, 2021, 53 small juvenile Pacific tree frogs (i.e., frogs that have recently lost their tails) were found dead in their cages. The most likely cause of death was desiccation due to a recent modification to the cages. The mortalities returned to normal, background levels after the cage configuration was changed back. There have been no incidences of desiccation since these changes were made. The activity was PHS-funded.

The IACUC has asked the PI to modify their housing SOP to state that daily mortalities are expected for frogs in a metamorphic stage and to establish guidance for when lab members providing husbandry should contact the AV regarding higher than normal rates of mortality.

OLAW believes that the corrective and preventative measures put in place by the University of California – Santa Barbara are consistent with the provisions of the PHS Policy on Humane Care and Use of Laboratory Animals. We appreciate being informed of this incident and find no cause for further action by this office.

Sincerely,

Brent C. Morse -S

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

Digitally signed by Brent C.  
Morse -S  
Date: 2022.02.07 08:31:24  
-05'00'

cc: IACUC Contact

## UC SANTA BARBARA

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Institutional Animal Care and Use Committee

Santa Barbara CA 93106-5062

Tel: (805) 893-5855

Fax: (805) 893-2005

Email: [iacuc@lifesci.ucsb.edu](mailto:iacuc@lifesci.ucsb.edu)

<https://www.research.ucsb.edu/animal-subjects/about>

January 20, 2022

Animal Welfare Assurance A3865-01 (D16-00497)

Dr. Axel V. Wolff, Director  
Division of Compliance Oversight  
Office of Laboratory Animal Welfare  
National Institutes of Health  
6700B Rockledge Drive, Suite 2500  
Bethesda, MD 20892-6910

Dear Dr. Wolff,

In accordance with our Assurance and PHS Policy IV.F.3, I am writing to report an incident of protocol non-compliance involving the care and use of animals at the University of California, Santa Barbara. Below is the full investigation report into IACUC protocol #8-21-966, "Alternative hosts for the amphibian chytrid fungus". The IACUC is convinced that appropriate remediations were enacted to prevent recurrence of the incident described below.

### Summary of Investigation:

On July 9, 2021, an undergraduate caretaker reported finding 53 small juvenile Pacific tree frogs (i.e., frogs that have recently lost their tails) dead in their cages. This was immediately reported to the PI and graduate student, who notified the Attending Veterinarian (AV) via email on July 12, 2021. The graduate student's email identified the most likely cause of death as desiccation due to a recent modification to their cages and noted that mortality rates returned to normal levels (~5/day) after undoing the change to the cages. When the AV returned from vacation on July 16, 2021, the notification from the graduate student was forwarded to the IACUC Office. After receiving this notification, the IACUC Chair appointed a sub-Committee, comprised of himself and the AV, to investigate the matter further.

On July 22, 2021, the sub-Committee met with the PI and graduate student. The mortalities occurred in five separate cages containing juvenile frogs of various sizes and life stages. The graduate student explained that the frog cages are misted with water from a spray bottle, covered with a clear plastic wrap, and the bottom of the cage is lined with a wet paper towel to ensure that the container stays humid. Frogs are also given a water dish and a hide (i.e., a small plastic flower pot cut in half to seek shelter under). However, on July 4, 2021, the lab switched from using plastic wrap on the cages to a thin, sheer (organza) fabric. The rationale for this switch was that the fabric would work better for containing the fruit flies that are fed to the frogs and would allow for better air flow. The graduate student said that after making the switch, the lab did notice

that there were some areas of the cage/paper towel that were drying out more quickly than usual. The lab used a spray bottle to mist the cage with water whenever this was noticed. The graduate student acknowledged that the weather had become warmer and dryer in the days leading up to the incident. After finding the desiccated small juvenile frogs, the lab determined that the fabric allowed for too much airflow into the cage and resumed using clear plastic wrap to cover the top of the cage. Additionally, the lab has now propped up one side of the cage. This creates a gradual slope on the bottom of the cage and causes water to pool only on one side. There have been no incidences of desiccation since these changes were made.

The PI also explained that in nature this is the most vulnerable stage of development for these frogs and there is a very high mortality rate. The immune system of these animals only offers limited protection until about 6 weeks after a juvenile frog's front legs have developed, at which point the immune system becomes more robust. Additionally, the PI explained that it is possible for a cage to be too wet for this stage of juvenile frog since it is no longer fully aquatic after developing from a tadpole. A small juvenile frog can get stuck in the surface tension of the water in the cage and drown. It is not uncommon for 5-10 of the animals at this stage to be found dead in their cages each day. It is likely that the daily mortality rate for these animals would be even higher in nature. Cages of juvenile frogs are regularly consolidated as the animal numbers in each cage decrease due to these mortalities. The sub-Committee has asked the PI to modify their housing SOP to state that daily mortalities are expected for frogs in a metamorphic stage and to establish guidance for when lab members providing husbandry should contact the AV regarding higher than normal rates of mortality.

On September 17, 2021, the IACUC Chair presented this investigation report to the IACUC at a convened meeting. The Committee agreed that the desiccation of these frogs coincided with the changing of the material used to cover the cages from plastic wrap. Additionally, it was noted that this building does not have a humidifier component in its HVAC system, so the humidity must be controlled by the lab at the primary enclosure level. It was discussed whether a humidifier, designed for use in amphibian or reptile terrariums, could be used to more accurately regulate the humidity in each cage. While this is a possibility to be considered if a similar incident occurs in the future, the IACUC believed that this was probably not necessary if the lab would continue to regularly use a spray bottle to mist the cage with water and cover the cage with plastic wrap. This project is funded by NIH grant 5R01GM135935. The Committee voted that this incident is reportable to both OLAW and AAALAC.

On November 12, 2021, the graduate student clarified which protocol the animals were being used under. Additionally, s/he submitted the housing SOP with the modifications requested by the sub-Committee, as well as the IACUC protocol with revisions to the number of animals they anticipate using for the remainder of the protocol. On December 2, 2021, the IACUC approved the modifications to the protocol and housing SOP. If you have any additional questions or comments, please feel free to contact us. Thank you for your time.

Sincerely,

DocuSigned by:

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Prof. Joseph Incandela

Institutional Official

Vice Chancellor for Research

(b) (4) Cheadle Hall

University of California, Santa Barbara

Santa Barbara, CA 93106-2050

incandela@research.ucsb.edu

Cc: Dr. Stuart Feinstein, IACUC Chair  
Dr. Manuel Garcia, Campus Veterinarian  
IACUC Office

**Wolff, Axel (NIH/OD) [E]**

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**From:** OLAW Division of Compliance Oversight (NIH/OD)  
**Sent:** Friday, January 28, 2022 7:39 AM  
**To:** IACUC Office  
**Cc:** OLAW Division of Compliance Oversight (NIH/OD)  
**Subject:** RE: [EXTERNAL] Incident Report for A3865-01

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

Axel Wolff, M.S., D.V.M.  
Deputy Director, OLAW

-----Original Message-----

**From:** IACUC Office <iacuc@lifesci.ucsb.edu>  
**Sent:** Thursday, January 27, 2022 6:24 PM  
**To:** OLAW Division of Compliance Oversight (NIH/OD) <olawdco@od.nih.gov>  
**Cc:** Joseph Incandela <incandela@research.ucsb.edu>; Manny Garcia <manuel.garcia@ucsb.edu>; Stu Feinstein <feinstei@lifesci.ucsb.edu>  
**Subject:** [EXTERNAL] Incident Report for A3865-01

CAUTION: This email originated from outside of the organization. Do not click links or open attachments unless you recognize the sender and are confident the content is safe.

To Whom It May Concern:

Attached is a letter that details an incident of protocol noncompliance at the University of California, Santa Barbara (A3865-01).

The UCSB IACUC has investigated and reviewed the incident. This report includes the summary of the incident, as well as any Committee corrective actions.

Please let me know if there are any questions.

Sincerely,

(b) (6)

\*\*\*Due to COVID, Research Integrity Staff are working remotely but remain fully operational\*\*\* For up-to-date information about COVID and research as it pertains to human and animal research, please visit our website: <https://www.research.ucsb.edu/office-research-mission-continuity-plan>

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## (A3865-01) - University of California - Santa Barbara

**IO Name** Incandela, Joseph

**IO Title** Vice Chancellor for Research

**IO Credentials** Ph.D.

**IO Email** incandela@research.ucsb.edu

**IO Address** (b) (4) Cheadle Hall  
Santa Barbara, CALIFORNIA 931062050  
UNITED STATES

**IACUC Title**

**IACUC Email** iacuc@lifesci.ucsb.edu

**POC Info**

(b) (6)