



July 26, 2021

Via Electronic Submission to <https://rfi.grants.nih.gov/?s=6079cab8b801000072005032>

National Institutes of Health
Office of the Director

RE: Comments Submitted in Response to Notice Number NOT-OD-21-118, Request for Information

To Whom It May Concern:

The Council on Governmental Relations (COGR) is an association of 190 public and private U.S. research universities and affiliated academic medical centers and research institutes. COGR concerns itself with the impact of federal regulations, policies, and practices on the performance of research conducted at its member institutions. One area of significant interest and expertise among COGR member institutions is the appropriate conduct of basic and applied animal research to ensure proper protections for animal health, safety, and welfare, while reducing unnecessary burden on researchers and research institutions.

COGR appreciates the opportunity afforded by NIH to submit comments in response to [NOT-OD-21-118](#), “Request for Information on Flexibilities to Reduce Administrative Burden While Continuing to Apply the PHS Policy to Zebrafish Immediately After Hatching” (RFI). Although COGR member institutions fully support the flexibilities outlined in the RFI, we believe that NIH used an inappropriate standard to make its determination that the [Public Health Service \(PHS\) Policy on Humane Care and Use of Laboratory Animals](#) (“PHS Policy”) applies to zebrafish larvae immediately upon hatching. Thus, we respectfully request that NIH re-evaluate the application of PHS Policy to newly hatched zebrafish larvae for the reasons detailed below.

Background: Pursuant to the Section 2034(d) of the 21st Century Cures Act’s ([P.L. 114-255](#)) (“Cures Act”), NIH was directed to review regulations and policies governing the care and use of laboratory animals for ways to reduce administrative burden while ensuring the health and welfare of research animals. The RFI states that as part of these efforts, NIH considered a request from the research community in the report [Reducing Administrative Burden for Researchers: Animal Care and Use in Research](#) to “consider changing the applicability of the PHS Policy to zebrafish larvae from immediately after hatching . . . [until] larvae begin free feeding.” NIH, through its Office of Laboratory Animal Welfare (OLAW), rejected this request for delayed application of the PHS Policy. Instead, the RFI suggests the use of the following three existing flexibilities afforded to institutional animal care and use programs:

- a. Generate approximate number of animals to be used per PHS Policy IV.D.1.A;

- b. Employ the Institutional Animal Care and Use Committee's (IACUC) designated member review process for zebrafish protocols; and
- c. Include persons with zebrafish expertise as *ad hoc* consultants or subject matter experts to develop "tracking, animal care, and euthanasia methods" for zebrafish.

Re-Evaluation of PHS Policy Applicability:

The RFI is problematic in the following three important respects. First, the RFI does not detail or provide any reference to the scientific evidence that NIH reviewed in arriving at its conclusion that the PHS Policy should apply to just hatched larvae. Rather, it summarily concludes that the Policy should be immediately applied to such larvae "[b]ecause contrary evidence is not currently compelling." Second, the RFI evaluates this unnamed evidence in a vacuum because it does not describe what, if any, procedure is being performed upon the larvae. Third, as detailed below, the RFI applies an inappropriate evidentiary standard.

Specifically, the RFI states that "insufficient evidence currently exists to indicate *beyond a reasonable doubt*" (emphasis added) that procedures that may cause pain or distress in humans may cause pain or distress in zebrafish larvae at hatching. Yet, there is no legal or regulatory basis for this evidentiary standard. In fact, the "beyond a reasonable doubt" standard is the burden of proof applied in criminal proceedings. [See, *In re Winship*, [397 U.S. 358](#) (1970)]. In civil cases, a "preponderance of the evidence" is the standard most typically applied. [See, Cornell Law School, Legal Information Institute, "[Burden of Proof](#)" (2017)].

The RFI cites Principle IV of the [U.S. Government Principles for the Utilization and Care of Vertebrate Animals Used in Testing, Research & Training](#) (the "Principles") as support for application of the PHS Policy to zebrafish larvae at hatching. Principle IV states that "unless the contrary is established, investigators should consider that procedures that cause pain or distress in human beings may cause pain or distress in other animals." Nothing in the Principles, however, sets any standard for determining when scientific evidence establishes "the contrary." Additionally, the Health Extension Research Act of 1985 ([P.L. 99-158](#)), the PHS Policy, and the [Guide for the Care and Use of Laboratory Animals](#) do not suggest or propose the use of any specific evidentiary standard (e.g., "beyond a reasonable doubt," "clear and convincing," "preponderance of the evidence," etc.) in determining when procedures that cause pain or distress in humans may cause pain or distress in other animals.

The aforementioned statute, policies, and guidance do not support the use of any particular evidentiary standard for determining applicability of the PHS Policy, let alone suggest use of highest burden of proof in American jurisprudence, i.e., beyond a reasonable doubt. Nor do they support evaluating the question of potential pain and distress outside the context of a specific procedure. Accordingly, COGR respectfully requests that NIH re-evaluate the application of the PHS Policy to zebrafish larvae immediately at hatching. Such re-evaluation should include review of pertinent scientific evidence in the context of a specific procedure under the preponderance of evidence standard, which is more appropriate to the evaluation of scientific evidence in this context. Further, the Notice should clearly describe the scientific evidence that forms the basis for any decision.


Support of Flexibilities: Although, COGR believes that Notice NOT-OD-21-118's conclusion regarding the applicability of the PHS Policy should be reconsidered, COGR does fully support the use of the flexibilities that are detailed in the Notice. In particular, the flexibility concerning generation of the approximate number of animals to be used is necessary given the physical characteristics of zebrafish larvae. At the time of hatching (48 to 72 hours post-fertilization), zebrafish larvae are very difficult to accurately count because they are only between 3.1 and 3.5 millimeters long and are largely transparent with patches of camouflage pigmentation. [See, University of Oregon, Zebrafish Information Network (ZFIN) webpage, "[Zebrafish Developmental Staging Series](#)" (accessed July 6, 2021); C. Kimmel, *et. al.*, "[Stages of Embryonic Development of the Zebrafish](#)," *Developmental Dynamics*, 203: 253-310, Figure 39 (1995)]. At this early stage of development, individual counting of larvae would be unduly burdensome and would not materially change the guidance for proper housing and husbandry.

Conclusion

COGR and the institutions that it represents recognize the importance of animal research to basic and applied research and fully support and welcome changes that improve animal health, safety, and welfare. COGR institutions also recognize and applaud the Cures Act' mandate that federal agencies seek out and implement appropriate pathways for reducing administrative burden. We believe that the suggested re-evaluation process is a more appropriate path for determining if the PHS Policy applies to zebrafish larvae on hatching and may lead to a different conclusion. Finally, apart from the issue of re-evaluation, COGR supports all the flexibilities outlined in the RFI.

We hope that the information provided herein is useful to NIH. If you have any questions regarding these comments, please contact Kris West, Director of Research Ethics and Compliance, at kwest@cogr.edu.

Sincerely,



Wendy D. Streit
President