May 4, 2023

Arati Prabhakar, Ph.D. Director Office of Science and Technology Policy The White House 1600 Pennsylvania Ave. NW Washington, DC 20500

Dear Dr. Prabhakar:

I hope this correspondence finds you well. I am writing on behalf of People for the Ethical Treatment of Animals U.S.—PETA entities have more than 9 million members and supporters globally—to respectfully request that your office address chronic and serious animal welfare violations, which very likely compromise the integrity of research, in the National Institutes of Health's (NIH) intramural laboratories.

As part of the White House Office of Science and Technology Policy's (OSTP) important mission, your office seeks to "strengthen[] and advance[e] American science and technology." NIH positions itself as the "steward of medical and behavioral research for the Nation," and toward that end, the agency receives substantial appropriations courtesy of U.S. taxpayers—more than \$45 billion in FY 2022—and operates massive intramural laboratories on its campus in Bethesda, Maryland and elsewhere. However, records obtained by PETA through Freedom of Information Act (FOIA) requests document shocking violations of basic federal animal welfare guidelines in NIH's own intramural laboratories.

PETA has analyzed NIH violations reports from October 2017 to January 2023,¹ tabulating an astounding 169 violations over this period. A clear pattern of repeated, systemic, and egregious violations of basic animal welfare standards emerges from the analysis. Animals suffer miserable deaths in NIH's laboratories because they're deprived of food, water, or air. Animals are used in painful, invasive procedures, but are denied post-procedural pain relief. Experimenters routinely deviate from their own protocols approved by the agency's animal care and use committees (ACUCs)—jeopardizing both the studies and the welfare of the animals used.

In response, NIH's feeble assurances of strengthened training, updated SOPs, enhanced ACUC oversight, discussions with staff, and experimenters' apologies forwarded in NIH's self-reports to its own Office of Laboratory Animal Welfare (OLAW) have not decreased the number of violations and have been entirely insufficient to address the serious compliance issues in NIH's laboratories. NIH's failure to comply with minimum animal welfare laws and guidelines as well as its failure to correct ongoing patterns of sloppy techniques call into question its PEOPLE FOR THE ETHICAL TREATMENT OF ANIMALS

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¹Exhibit 1: Table of Violations in NIH's Intramural Laboratories Self-Reported to OLAW, October 11, 2017 – January 4, 2023.

commitment to good research, likely diminishing its contribution to American science and technology.

A Monkey Who Died of Thirst, a Mouse Who Was Allowed to Drown, and an Escaped TB-Infected Marmoset Who Was Gassed to Death

NIH documents reveal 33 serious animal welfare violations just in the eight month period from May 13, 2022, to January 4, 2023. Among these: A monkey died after being left without water for the better part of a long weekend; a mouse drowned while being used in a water maze test simply because the experimenter didn't want to disturb the video tracking system used to record the test by pulling the mouse from the water; and a marmoset held in a BSL-3 laboratory and infected with Mycobacterium tuberculosis—transmissible to humans—escaped from his cage and climbed in the HVAC system. He was gassed to death with carbon dioxide after experimenters failed to capture him

Inadequate Oversight and Limited Accountability and Transparency

While universities and commercial enterprises that use animals covered under the Animal Welfare Act are required to be inspected at least once a year by the U.S. Department of Agriculture, federal laboratories—including those operated by NIH—are exempt from such inspections. A system of self-reporting and self-regulation is intended to ensure compliance with federal animal welfare standards. However, this appears to have become an administrative exercise in which NIH sends a written report of an animal welfare violation to OLAW, outlining any planned remedies aimed at ensuring the violation will not be repeated; in turn, OLAW may request clarification or further information—but eventually closes the case. There are no citations, no enforcement actions, no true accountability. Moreover, the violations that are self-reported to OLAW are not published on NIH's website or otherwise made public, and it falls on concerned citizens or non-profit organizations like PETA to employ FOIA requests to uncover transgressions.

NIH has thus far chosen to turn a blind eye to the systemic problems in its laboratories, so we turn to you to intercede for the sake of animal welfare, sound science, public trust, and the U.S.' position as a global leader in biomedical research. We respectfully request that your office use its authority to ensure that investigators and staff adhere to laws and guidelines and that all scientists and staff who refuse to do so be terminated.

I would be happy to meet with your office to answer any questions. Thank you for your time and consideration.

Sincerely,

Alke Chede

Alka Chandna, Ph.D. Vice President Laboratory Investigations Cases <u>AlkaC@peta.org</u> | 757-803-6447

Exhibit 1

Table of Violations in the Intramural Laboratories of the National Institutes of Health (NIH) Self-Reported by NIH Institutes to NIH's Office of Laboratory Animal Welfare (OLAW) and Obtained by PETA Via Freedom of Information Act Requests October 11, 2017 – January 4, 2023

The original federal case reports can be found at the following link (please copy the link and paste it in your browser): <u>https://www.dropbox.com/sh/g2sf23d5cgx5jk0/AABABIVMvUWkg60ULr95YgqAa?dl=0</u>

Number	Case No.	Date of Initial Self-Report to OLAW	Details
1	12A	Oct. 11, 2017	After mice were injected with an anticancer agent suspended in DMSO, 45 became immediately sick post-injection and an unspecified number required euthanasia. An investigation revealed that the anticancer agent had been delivered in 100% DMSO, resulting in toxicity. The anticancer agent and DMSO were not listed on the Animal Study Protocol (ASP).
2	12B	Oct. 27, 2017	Approximately 205 mice received the wrong experimental diet. This occurred when the research food ran out and another, different diet was fed by a caretaker who assumed it was the same formulation. All the mice were euthanized because the experimental aims had been disrupted.
3	12C	Feb. 22, 2018	Lights in nonhuman primate animal rooms had been on for 24 hours for almost five months. The facility manager had not been monitoring the building automated systems reports which reflected the problem. On Dec 7, 2017, a DVR veterinary technician informed facility management staff that while working late the previous night, he noticed the lights in animal rooms in the animal facility corridor were still on. It was determined that the lights in that area had been on 24 hours a day, 7 days a week, since July 18, 2017.
4	12D	Feb. 22, 2018	A veterinarian failed to provide timely care to a 6.5-year-old female Aotus monkey (owl monkey who had been used for

5	12E	March 5, 2018	 breeding) with significant (20%) weight loss. Although a veterinary technician had notified the veterinarian, the animal was not promptly examined and there was no record of observation or a treatment plan. The monkey was noted to have a low hematocrit (i.e., she was severely anemic) and subsequently died. A necropsy of the monkey indicated that she had a severe abdominal hemorrhage, thoracic effusion, and cardiomyopathy. A dog sustained skin burns from an electric blanket. The blanket
			had been used because of the cold temperature in the procedure room and inability of other sources to maintain adequate heat.
6	12F	March 29, 2018	 Experimenters deviated from an approved septic shock protocol involving dogs ("Effects of Adjunctive Therapy with Early Transfusion of Granulocytes in a Canine Model of S. <i>aureus</i> Pneumonia."): A small dose of an experimental compound was given to a dog, an unapproved additional transfusion unit of blood was given, silk suture was used which caused infection, dogs were dosed with an experimental agent before the acclimation period had ended, and an experimental agent was used on an unapproved dog. The study was suspended.
7	12G	June 11, 2018	Five mice died due to lack of food and water. When the cage was set up, no food or water was provided and the problem was not noted during the daily health checks. The problem was not reported to the veterinarian for one week.
8	12H	June 11, 2018	One mouse died and two had exceeded the endpoint parameters in a study involving administration of an experimental compound in drinking water. In total, 38 mice dosed with the experimental agent had not been removed from dosing when their body weight dropped by more than 15%, animals were returned to dosing before regaining weight, and animals were kept on study beyond the approved endpoints in the protocol. This laboratory has had prior instances of noncompliance.

9	12I	July 11, 2018	Two mice were kept alive one day longer than described in the protocol. The mice on this study were to be euthanized as soon as symptoms developed in one animal.
10	12J	July 12, 2018	A malfunction of a metabolic caging system resulted in the deaths of 12 mice due to elevated temperature and decreased oxygen levels. Three surviving mice were immediately removed.
11	12K	Aug. 14, 2018	Three individuals who had performed survival surgery procedures on mice used unacceptable surgical techniques, failed to adhere to the ACUC-approved protocol, and failed to use appropriate analgesics post-surgery.
12	12L	Sept. 4, 2018	On July 16, 2018, a principal investigator (PI) practiced injecting Cortland's salt solution into 15 zebrafish although this procedure had not been approved. Four of the 15 fish died immediately. The procedure was repeated in 18 fish on August 6, 2018, although the procedure still had not been approved. Over the next three days, 11 fish were euthanized and 7 were found dead.
13	12M	Oct. 2, 2018	On Aug 25, 2018, a group of four transgenic mice was discovered with footpad swelling, blackened hard digits, and/or skin lesions. One mouse was immediately euthanized and the others were placed under veterinary care until Sept 4, 2018, when they were euthanized. The mice had been subjected to formalin injection (although this procedure had only been approved for wild-type mice – and in that protocol, the mice were to be euthanized 90 minutes after the injection of formalin). Also, the transgenic mice had previously received surgery, which should not have been followed by formalin injection.
14	12N	Oct. 2, 2018	Between the evening of July 30, '18 and the morning of July 31, 2018, two rhesus macaques escaped their cages due to unsecured cage floors; and a third escaped due to an unsecured cage divider allowing the animal access to an unlocked cage. The three macaques fought and sustained injuries to the hands, arms, faces, and tongues. They were captured on the morning of July 31 st and treated. Two additional caged monkeys were also treated for

			injuries apparently obtained from interactions with the escaped animals.
15	120	Oct. 9, 2018	Imaging studies were performed over two days in two live mice without prior review and approval by the ACUC. Specifically, the mice were anesthetized, imaged, and recovered—without approval. The mice were also held in the Mouse Imaging Facility without approval.
16	12P	Oct. 15, 2018	Mice on a bone fracture study were not given post-operative analgesia (buprenorphine), as required by the protocol. The mice had received a short acting dose of analgesia (buprenorphine) prior to the procedure. The report stated: "Withholding analgesia from mice that have undergone major surgical procedure is a serious violation of approved ASP procedures, PHS policy, and the Guide, potentially subjecting the animals to unnecessary pain and distress." The notes specify: "Mice [who] undergo the bone fracture surgery should be provided with SR buprenorphine for as long as the mice require analgesia The duration of treatment can be based on known human requirements with similar injuries; this may mean that one week of analgesia treatment may be required. Mice, as prey animals, tend to mask pain/distress, which would make it difficult to clearly see the magnitude of pain they are experiencing."
17	12Q	Oct. 29, 2018	Experimenters failed to use aseptic technique and failed to provide appropriate post-surgical analgesia to mice used in a blood vessel cannulation surgery.
18	12R	Dec. 11, 2018	Blood had been collected from five rats although this procedure had not been approved. One of the animals was subsequently euthanized due to bruising at the collection site.
19	125	Jan. 4, 2019	Mice who were used in a study were of a different strain than those listed on the protocol.
20	12T	Jan. 17, 2019	Three mice died and one was moribund (s/he was euthanized) due to lack of food in the cage. An investigator had set up the cage, but

21	12V	April 3, 2019	 failed to provide food. The problem was not identified by husbandry staff during the daily health checks, which was compounded by the room being on a reverse light cycle, which made observations more difficult. Also, some lights were not working at all. During the evening of Jan. 28, 2019, "a faulty HVAC valve
			feeding an animal housing room at the NIH Animal Center caused the temperature in the room to reach 100 degrees Fahrenheit." The alarm did not notify of the excessive temperature. At 7:30 am on Jan 29, 13 dead mice were found in four cages.
22	12W	April 8, 2019	Several mice had infected surgical incisions due to the apparent lack of proper surgical preparation (i.e., shaving the surgical site). Six mice were euthanized due to infection. The employee who carried out the surgeries had not been approved to carry out surgeries on animals in the animal study protocol.
23	12X	May 2, 2019	 On April 5, 2019, it was determined that mice on a protocol had not been given the protocol-required post-surgical analgesia. There was a misunderstanding by the PI that the non-NCI facility housing the mice would administer the analgesia. "This oversight probably affected 105 – 140 mice overall." The NCI ACUC Chair wrote: "[A]lthough it's not clear how long this has been going on, this has probably affected 105-140 mice since the last protocol renewal." The nature of the surgery is not specified.
24	12Y	May 1, 2019	An experimenter was observed conducted surgery on a mouse without using proper aseptic technique. The surgery was not described in the approved protocol. Two additional mice who had been used in the unapproved surgery had not been given analgesics. The mice were euthanized.
25	12Z	June 11, 2019	Three mice died due to lack of food. "The animal caretaker had failed to provide food when the cage was changed and the problem was not identified during the daily health checks by husbandry staff or when the cage was handled by an investigator." The mice had been without food for 48 hours.

26	13A	Sept. 12, 2019	On March 29, 2019, an animal health technician noticed a cage that had been marked for euthanasia the previous day (March 28 th). The cage contained nine mice, two of whom were dead and one with severe fight wounds. The remaining seven mice were immediately euthanized by the technician. Upon investigation, it was determined that the mice in the cage had come from six different cages; SOP permits such combining for <i>immediate</i> euthanasia (which obviously didn't happen) and SOP also only allows for a maximum of five animals per cage.
27	13B	June 19, 2019	A bone marrow biopsy procedure was carried out on a nonhuman primate without first clipping the fur and cleaning the skin. The procedure was not performed aseptically. The experimenter also failed to provide post-procedure care to the primate (although details are not specified).
28	13C	Sept. 15, 2019	On June 9, 2019, an experimenter performed vasectomies on 11 mice. The following morning, two of the mice were found with "suture failure and dehiscence with protrusion of the viscera." They were euthanized. A third mouse was later found with suture failure. He was re-sutured and "recovered well." An additional mouse was found dead on June 11, 2019. It was also determined that the experimenter failed to implement post-surgical monitoring procedures and failed to provide post-surgical analgesia as specified in the protocol.
29	13D	Sept. 12, 2019	On Friday, July 25, 2019, 12 mice were fasted overnight for a study to be conducted the next day. The mice were fed a food pellet the next day but the experimenter forgot to then provide <i>ad</i> <i>libitum</i> chow and to remove the "Food Restriction" cards. He also forgot to make notations in the log book. On Monday morning, two dead and four dehydrated, listless mice were found. One of the dehydrated mice was euthanized.
30	13E	Sept. 12, 2019	On Feb. 21, 2019, it was determined that there were multiple noncompliance issues in connection with surgeries conducted on mice. In particular, it was not clear that the experimenters were

			properly trained (as they did not carry out the procedures in the presence of the Attending Vet as required in this particular study) and wound closure procedures did not adhere to what was specified in the protocol. Several mouse were found dead in their cages.
31	13F	Aug. 5, 2019	On May 28, 2019, an animal caretaker reported three dead mice in a cage. The cage had been set up by an experimenter on May 23 rd with water, but no food. The lack of food was not noticed on any of the intervening days.
32	13G	Dec. 22, 2020	On April 23, 2019, a rhesus macaque escaped into a room from his/her cage and bit an animal technician on the hands and thighs while loose; this injury was handled by the NIH Occupational Medical Service (OMS) office. On May 1, 2019, another rhesus macaque escaped while being transferred (via pole and collar) from a transport cage to his/her home cage and bit an experimenter on the thighs (the employee was taken by ambulance to the hospital, but was released that night). On May 2, 2019, a marmoset bit an animal technician on the finger while being handled.
33	13H	Nov. 5, 2019	Two mice died and one was in poor condition due to lack of food. A fourth mouse died because s/he had a malocclusion and could not eat.
34	13I	Jan. 28, 2020	Five zebrafish died at a satellite facility when the temperature dropped 10 degrees. There was a malfunction in the heating system.
35	13J	Oct. 15, 2019	Six mice died and two were euthanized due to severe dehydration. Staff failed to replace the bottles which contained small amounts of water.
36	13K	Oct. 24, 2019	A mouse was subjected to a survival surgery under an expired protocol. Because the procedure had been performed under an inactive animal study proposal, the mouse was euthanized.
37	13L	May 1, 2020	A PI was found performing surgical procedures that had not been approved by the ACUC.

38	13M	Oct. 21, 2019	An experimenter failed to monitor mice after they were injected with 5 fluorouracil as required by the protocol. Eight mice were found dead or moribund in their cages. The failure to monitor the mice may have prevented the mice from being euthanized at the designated humane endpoint.
39	13N	Dec. 6, 2019	An experimenter administered tamoxifen in mice (number not specified), but did not prepare the tamoxifen in the manner approved. Six mice were found dead and another 20 mice required euthanasia.
40	130	Dec. 6, 2019	One mouse was killed and another required euthanasia when, during a routine cage changing, a technician dropped a cage of mice after being startled when a rack was hit by the door when another individual entered the room.
41	13P	Dec. 6, 2019	Eight knockout mice were found dead in their cages. They were to be provided water treated with nitisinone (NTBC), but were not.
42	13Q	Jan. 21, 2020	After 50 African thicket rats were administered Azithromycin by gavage, 18 were found dead or euthanized due to adverse clinical signs. Although Azithromycin was approved to be administered, the dosage administered exceeded that listed on the animal study protocol. The remaining rats were also euthanized.
43	13R	Jan. 28, 2020	Three dead and two moribund recently weaned mice were found in two cages on an animal housing rack. The cages had water, but no food. The moribund mice were euthanized.
44	13S	Sept. 23, 2020	An NEI laboratory staff member injected a drug into seven mice. However, the drug was not approved for use on the study and the laboratory staff member was not listed in the animal study protocol. All seven mice became ill and one was euthanized for failure to respond to veterinary treatment. Also, because the drug was not listed in the animal study protocol, the husbandry staff were unaware of the safety issues and required handling precautions related to the drug.
45	13T	Oct. 17, 2019	There was a spike in zebrafish mortality in the NICHD Aquatics Facility between July 7, 2019 to July 11, 2019. The report from

			NICHD states: "We currently house approximately 161,000 animals in the facility and typical mortality per system is 30 fish per day but [over a span of] 4 days [7/7/19 - 7/11/19], we saw an average of 115 deaths." It is believed that a combination of environmental stresses (construction issues and floor resurfacing) contributed to the deaths.
46	13U	July 6, 2020	Two post Baccalaureate fellows working in a laboratory at NIMH were instructed to euthanize mice, but were not trained or supervised on appropriate methods of euthanasia. Euthanasia was performed by a method not described in the protocol. The fellows were permanently removed from the laboratory.
47	13V	April 13, 2020	A mouse was found dead with his/her forelimb stuck in the side of the cage. The cage was broken (allowing for the mouse to become trapped), but was still used.
48	13W	April 13, 2020	An adult mouse was found dead in the home cage. It was determined that a husbandry staff member failed to euthanize the mouse within the time period directed by the clinical veterinarian and the animal study proposal.
49	13X	March 2, 2020	Five mice were found dead after not receiving adequate food in their cage.
50	13Y	March 12, 2020	Four mice were found dead in their home cage. The mice had been gassed with isoflurane, but no secondary, physical method was used to confirm death as required.
51	13Z	April 13, 2020	Six mice died after they were injected with a compound (retrorsine) that was not included in the approved animal study proposal.
52	14A	June 30, 2020	Three recently weaned mice were found dead in a cage. There was food, but the water bottle was empty and the bedding was completely caked and wet. The water bottle was leaking, most probably because the rubber stopper was not tightly sealed in the bottle. It was determined that the wet cage had been missed on more than one health check by two different animal care technicians.

53	14B	May 4, 2020	Two mice went missing, but husbandry and research staff members failed to report the missing mice. The mice were later
			found in animal traps.
54	14C	June 30, 2020	Four retired breeder female mice were found dead in a cage. There
			was food, but the water bottle was empty. There was no sign of a leaking water bottle.
55	14D	June 29, 2020	On May 4, 2020, a guinea pig pup who was born on May 1, 2020 was found dead outside the cage in a drop water reservoir container beneath the automatic watering system. It seems that the
			pup was able to fit through a space between a port hole and the lixit valve, whereupon s/he fell into the reservoir container.
56	14E	June 29, 2020	On two consecutive days, the "caretaker" noticed the cages had water but no food, but failed to take action. This person was reassigned to a different facility.
57	14G	Sept. 23, 2020	Four of 10 mice died as they were not able to access food. Food pellets were clumped together and did not drop down in the feeder. It was determined that husbandry staff had failed to follow the SOP of breaking up the food before putting it in the feeder. The responsible individual was transferred to a cage wash position in another facility.
58	14H	Aug. 19, 2020	Two male rhesus were allowed pair access when returned to their home quad cage after TB testing. They were supposed to be individually housed, but were not. Fighting occurred during which one monkey sustained several serious injuries to his left arm, left leg, and back. The monkey "required extensive surgical and medical treatment." The report states: "This injury resulted in prolonged treatment, stress, and immune responses which will impact the long-term HIV study."
59	14I	Aug. 19, 2020	An experimenter administered a drug to mice that was not included in the relevant animal study protocol.
60	14J	Aug. 27, 2020	Between February 6, 2020 and July 16, 2020, 142 mice held in NCI animal facilities were given expired sustained-release

			buprenorphine. Three expired vials had been used past the
<u>(1</u>	1.417	9 4 0 2020	expiration date.
61	14K	Sept. 9, 2020	A moribund mouse was found in a cage with water but no food. The mouse was euthanized.
62	14M	Sept. 30, 2020	A mouse who was subjected to embryo transfer surgery was missing a staple necessary to close the incision properly. The card on this mouse's cage listed the wrong principal investigator.
63	14N	Jan. 12, 2021	A marmoset died after becoming entangled in an enrichment device chain.
64	140	Feb. 25, 2021	Two cages were identified with no food (each cage housed 4 recently weaned mice). Six of the mice were dead. The other two mice recovered.
65	14P	Oct. 19, 2020	A cage was found with an empty water bottle and an older mouse was observed to be in poor condition ("hunched, lethargic, with an ocular concern"). The mouse was euthanized.
66	14Q	Jan. 29, 2021	A rhesus macaque was subjected to two survival surgeries under an animal study protocol for which only a single surgery had been approved. The ASP described only one surgery involving an injection into the inner ear of only one ear. However, the PI performed the same surgery on the opposite ear one week later.
67	14R	Oct. 19, 2020	A male rhesus (NHP ZA28; 18 years of age) suffered a burn (2 x 3 inches, 1/2 inch deep) to the abdomen caused by the use of a gel pack heating device during a procedure. The gel pack was supposed to be wrapped prior to being placed on the animal's abdomen. Instead, the gel pack was placed directly on the abdomenand two days later, a lesion developed on the abdomen. The macaque was not on an active protocol, but had been anesthetized for a tooth extraction.
68	14S	Jan. 21, 2021	Four mice were found dead in a cage where no food was present.
69	14T	Feb. 3, 2021	A 6-year-old female marmoset was being prepped for an MRI, but died while under anesthesia. The connection to the oxygen source was not intact (i.e., the tubing had become disconnected).

70	14U	Feb. 11, 2021	While using electrocautery during surgery on a mouse, small
			flames came up from "the surgery site" (i.e., the mouse). The
			procedure was stopped and the mouse was euthanized. It was
			determined that alcohol fumes from the surgical prep built up
			under the drapes and was ignited when using the electrocautery.
71	14V	Feb. 25, 2021	From Dec. 20, 2020 through Jan. 7, 2021, 15 rhesus macaques
			received expired buprenorphine as part of a multimodal analgesia
			regimen.
72	14W	Feb. 25, 2021	A quad holding four monkeys was found to be without adequate
			water (it appeared that the monkey may have not had access to
			water for three days). The water line from the wall to the quad was
			not completely connected. The line was reconnected and three of
			the monkeys immediately began drinking. All four animals
			received fluids subcutaneously and one of the four required
			additional treatments, but fully recovered.
73	14X	March 1, 2021	A mouse was found dead, entrapped between the side of the food
			hopper and the steel bars containing the food. The carcass was
			partially hanging down into the cage from the hopper.
74	14Y	March 3, 2021	Hamsters were placed in transmission caging [two cages
			connected through a 3" port on the sides of the cages, which are
			blocked using a mesh screen to prevent the hamsters from crossing
			into the other box]. One animal inserted his/her head into an
			opening in the cage, but could not extricate him/herself from the
			opening, suffocating. A second hamster had likely attempted to
			insert his/her head into the opening and also died.
75	14Z	March 30, 2021	Experimenters failed to follow veterinary directives and failed to
			comply with institutional policies. Buprenorphine was prescribed
			for a mouse on Jan. 29th, but was not started until Feb. 2nd (i.e.,
			five days after directed). Repeated doses prescribed were missed.
76	15A	May 3, 2021	Experimenters failed to provide water to animals. A water bottle
			containing medicated water to a mouse cage had become clogged
			with sediment, blocking the mice from getting water. Six mice
			were found dead from dehydration. Animal care staff failed to

	150	M 2 2021	follow the SOP requiring periodic shaking of the medicated water bottles to avoid sediment build-up. In addition, staff had failed to administer the topical ointment to the mice, as required in the protocol.
77	15B	May 3, 2021	 Experimenters failed to comply with humane endpoints. On March 17, 2021, a mouse was reported for hunched posture and was evaluated as emaciated and lethargic. The veterinarian instructed the PI to euthanize the mouse by 2 pm the following day. However, the mouse was not euthanized. The veterinarian found the mouse on March 22nd with a large abdominal mass and emaciation—and euthanized the mouse at that time.
78	15C	May 3, 2021	In two separate incidents, on March 11, 2021 and April 12, 2021, vivarium staff identified a used cage with several neonates that had been sent to cage wash to be cleaned. The neonates were discovered before the cages were run through the cage washer, and they were euthanized.
79	15D	May 3, 2021	A mouse escaped from a cage. However, the investigator did not inform the Animal Program Director or husbandry personnel. The mouse was found dead in a live trap, and a necropsy determined the cause of death was likely starvation.
80	15E	May 14, 2021	Experimenters failed to end a study at the ACUC-approved study end date. The expiration date was recorded incorrectly in the Internet Animal Study Proposal as being a month later than the protocol was actually scheduled to expire. Mice were subjected to procedures in which the ends of their tails were cut off ("tail snip") and tumors on their bodies were measured with calipers. The issue was noticed two weeks after the study was supposed to end.
81	15F	April 27, 2021	A chemical (clenbuterol) was administered to mice, but this compound was not in the ACUC-approved protocol. Five mice died after administration of this compound, and it was suspected that the reason the mice died was because of advanced age and obesity. Younger mice had previously tolerated the compound.

82	15H	June 7, 2021	Experimenters failed to handle animals appropriately. There were two cases of accidental injury of Aotus monkeys associated with their removal from housing units. In the first case, the monkey was noted to have a fracture to P1 of digit 4 of the right hand. The monkey's finger was amputated down to the metacarpal. In the second case, a veterinary technician was removing a monkey from the cage when the monkey's left leg became caught in part of the cage. The technician didn't notice but proceeded to pull the monkey out of the cage. This resulted in a mid-shaft fracture of the tibia and fibula. The monkey underwent surgery to address the fracture.
83	151	Sept 7, 2021	A mouse died due to lack of food. A note on the cage mistakenly stated "No Food on Floor" rather than "Food on Floor".
84	15J	Aug. 3, 2021	The thermostat in an animal holding room read 90 degrees F at 5:59 AM. Subsequently, seven mice in the room were found dead. A bad valve in the holding room was believed to be the cause of the temperature spike.
85	15K	Aug. 3, 2021	An experimenter found a single-housed eight-week-old female mouse dead, and reported that there was no water in the water bottle, and dry bedding. It was assumed that the animal had died of dehydration.
86	15L	Aug. 3, 2021	Two male rhesus macaques gained access to each other's cages, due to a padlock not securing a dividing panel. The monkeys fought, with both animals sustaining injuries.
87	15M	Sept 27, 2021	Three rhesus macaques failed to receive their minimum, protocol- required daily water intake of 20 ml/kg. This occurred 2-3 times per week prior to the PI discovering the issue. It was determined that the protocol was not followed regarding the gradual decrease, and then gradual increase, of the water rations. Protocol deviations were likely caused by both miscommunication among laboratory/animal care staff and faulty record keeping.

88	15N	Aug. 17, 2021	A small litter of mouse pups in a nestlet was discovered inside a clear trash bag, located in a hallway receptacle containing discarded bedding material from cage changes. The pups were euthanized.
89	150	Aug. 17, 2021	An adult mouse died after being run through the high-temperature, mechanical cage washer. The carcass of the mouse was discovered inside an "enrichment" tube on the clean side of the cage washer. The mouse was hidden in the tube and was not observed before being run through the cage washer.
90	15P	Aug. 17, 2021	An experimenter used "toe clipping" (i.e., toe amputation) as a means to identify eight adult mice. This was not included in the animal study protocol.
91	15Q	Aug 31, 2021	Three mice were found dead in a cage and two additional hunched and scruffy mice were found in a cage. As they were in poor condition, they were killed. Food for the mice was clumped in the top of the feed bin and the mice couldn't reach it; they starved to death.
92	15R	Sept 7, 2021	On Aug. 18, 2021, while using electrocautery during surgery on a mouse, small flames were observed from the surgery site (i.e., the mouse's body). The procedure was stopped and the mouse was killed.
93	158	Sept 14, 2021	Water flow was turned off on a row of tanks housing zebrafish. As a result, water temperature decreased and water quality declined, likely causing the death of seven fish.
94	15T	Sept 20, 2021	Veterinary orders for analgesia were not followed on a weekend. As a result, a mouse did not receive a second dose of Buprenorphine Sustained-Release and the ibuprofen medicated water was removed and replaced with plain water. The mouse exhibited signs of pain and was killed two days later.
95	15U	Oct 21, 2021	One hundred and three cavefish were found dead in tanks at NICHD. Based on the physical appearance of the affected animals and on the accumulation of air bubbles in the tanks, gas bubble

			disease was diagnosed. The bio-filter air-bubbler was located
<u> </u>			abnormally close to the pump intakes.
96	15V	Oct 29, 2021	A NINDS investigator failed to label cages with appropriate safety signage regarding mice who had been treated with the chemical hazard tamoxifen.
97	15W	Nov 1, 2021	Mice were used in an experiment in which they were fed a high fat diet. The ACUC-approved protocol included an end point of 40 weeks, but mice remained in the facility receiving the diet at 48 weeks after diet initiation.
98	15X	Nov 1, 2021	Multiple mice were injected with tumor cells. The tumor cells used were not approved; they were more aggressive than had been expected and the lab did not monitor mice appropriately after injection, resulting in mice progressing beyond the expected endpoints. The mice suffered "adverse health effects."
99	15Y	Nov 1, 2021	Fifteen mice underwent surgical procedures, but they were not given systemic analgesics as described in the protocol. This likely resulted in untreated post-operative pain.
100	15Z	Nov 5, 2021	A cage was found that contained four mice, two animals were found dead, and two others were reported to be in very poor health (cold and dehydrated), necessitating euthanasia.
101	16A	Nov 15, 2021	A group of mice was sent to necropsy after developing unexpected clinical signs and one animal died unexpectedly. The mice were found to have been injected with a tumor cell line that was not approved on the approved ACUC protocol and treated with an agent that was also not approved.
102	16B	Dec 9, 2021	Three cages of mice had been found without water bottles. One mouse was dead. The cages had been set up on Friday, but staff neglected to set up water bottles; the matter was discovered on Monday.
103	16C	Dec 9, 2021	A rabbit was given ketamine prior to euthanasia and tissue harvest. The tech was distracted and left the room; when they returned, the rabbit was dead. According to the report: "The failure of a

104	16D	Dec 9, 2021	research technician to appropriately monitor an anesthetized rabbit may have contributed to [his/her] death before the initiation of the ACUC-approved experimental procedure."
104	10D	Dec 9, 2021	A laboratory member administered a drug to mice for a procedure that was not listed in the approved protocol. Ten mice "were affected"—"most likely because of the injection given to them." It seems "the reagents used in the injection were not freshly prepared and they were consequently formulated inappropriately."
105	16E	Jan 12, 2022	Twenty-two mice experienced weight loss that exceeded the humane endpoint approved in the protocol. Lab staff held mice 24 hours past the approved endpoint—resulting in one moribund animal and two deaths.
106	16F	Jan 12, 2022	Rats who were used in an experimental surgery were observed with bloating. It was determined that laboratory personnel had used a drug called Equithesin (a mixture of pentobarbital, chloral hydrate, and magnesium sulfate; this is used as a general anesthetic in horses) to anesthetize the rats, rather than pentobarbital, which is what had been approved. Laboratory personnel lied when asked what anesthesia had been used—and they later admitted they were not aware of the differences between the two drugs. Of note, according to this article: <u>https://link.springer.com/article/10.1007/BF02246672</u> : "In 1977 it was reported that chloral hydrate could cause adynamic ileus in rats, leading to morbidity and death. The symptoms were a swollen abdomen, lethargy and anorexia."
107	16G	March 23, 2022	Three mice were administered lipopolysaccharide (LPS) at a dose that was about ten times the dose listed in the protocol. Two of the mice were ill and another was moribund. All three mice were euthanized.
108	16H	April 1, 2022	Five recently weaned mice died as a result of food deprivation. The cage may have been incorrectly assembled by care staff when the animals were weaned and the missing food from the hopper was not identified during the subsequent twice daily checks.

109	16I	April 1, 2022	Six mice were intravenously injected with a suspension of tumor cells in Matrigel, and one animal died. However, the protocol had specified subcutaneous injection.
110	16J	April 1, 2022	An animal (species not indicated, but it appears that the animal was either a mouse or a rat) on a protocol was at the humane endpoint, but was not euthanized until a day later—meaning that the animal suffered protracted pain and distress.
111	16K	April 1, 2022	A small 10-day old guinea pig pup was injured by a vertical metal grate in the back of the guinea pig cage. A gap allowed entrapment and fracture of one of the animal's hind legs. Due to the severity of the injury, the animal was euthanized.
112	16L	April 4, 2022	A cage of chickens was found with two dead chickens in it, and the remaining five chickens were in a lethargic state. Two of these five chickens were euthanized. The food hopper was in a configuration that precluded access to the food by the chickens.
113	16M	April 27, 2022	Fifteen mice died after experimenters failed to properly restrain the mice for an experimental procedure in which the mice were exposed to radiation.
114	16N	April 27, 2022	Eight mice who had undergone intracranial injection had not received the required analgesic and had been kept past the experimental endpoint. The mice were supposed to be euthanized within 48 hours after surgery, but were still alive 1-3 weeks following the surgery. Also the surgeon was not approved to perform procedures on the protocol and had known that the procedure was not approved. The surgeon admitted to performing 38 such surgeries from November 2021 to March 1, 2022; and confirmed that he did not use a post-operative analgesic for any of the animals receiving surgery from sometime in January 2022 until March 2022. The violations occurred for many weeks and remained unchecked because of poor communication, faulty

			record keeping, and apparent efforts by the surgeon to hide any evidence of his activities.
115	160	April 29, 2022	An experimenter performed abdominal surgery on two adult mice; however, the abdominal site had not been approved in the surgical plan described in the ACUC-approved animal study protocol.
116	16P	May 11, 2022	Mice in three cages were subjected to tail biopsies; however, this procedure had not been included in the approved protocol. Moreover, the tail biopsies were not performed according to institutional guidelines, in that the mice were older than 21 days of age when biopsied, more than 2 mm of tail was collected for the biopsy, inadequate analgesia was used (only topical lidocaine was used prior to the tail biopsies), and inadequate hemostasis/monitoring occurred. The mice likely experienced unrelieved pain.
117	16Q	May 11, 2022	Mice in 11 cages were subjected to a major survival surgery (orthotopic pancreatic cell transplant). However, the experimenters did not provide the approved analgesics. Also, the staff failed to record any post-surgical observations. The mice likely experienced unrelieved post-surgical pain.
118	16R	May 11, 2022	Mice in three cages were noted to have hunched postures, unsteady gait, generalized dermatitis, and rough hair coats. The mice were beyond the endpoints specified in the protocol. Also, three of the mice had been reinjected with a cell line, a procedure that was not approved by the ACUC.
119	168	May 13, 2022	A moribund mouse was improperly euthanized by an animal husbandry technician who failed to adequately perform a second method of killing following CO2 gassing. It appears that the technician attempted to break the mouse's neck (the skin on the back of the mouse's neck was found to be lacerated), but failed— and the mouse regained consciousness. The mouse was observed moving while an employee was preparing to perform necropsy.

120	16T	May 13, 2022	A surgical pilot study was performed on six mice without approval. In particular, a "surgical decidualization" was performed on six mice. This was carried out in the Reproductive & Developmental Biology Laboratory (RDBL) Pregnancy and Female Reproduction group. "The mice were killed at the end of the pilot study.
121	16U	May 13, 2022	An unspecified number of neonatal rats died from lack of oxygen after a veterinary technician used a 4-chamber anesthesia machine as a staging area from which to microchip the pups. The technician did not realize that oxygen was not flowing in one of the chambers of the machine; and the rat pups held there died.
122	16V	May 13, 2022	A Yucatan mini-pig became injured from being held in an enclosure with perforated, raised flooring. The pig was observed to be limping on his left hind foot and radiographs revealed a fracture of his left third distal phalanx. It was suspected that the pig got his left hind third toe stuck in the webbed flooring and fractured the toes as he was trying to get his foot freed.
123	16W	May 13, 2022	Two mice held in two different cages were found dead; the laboratory failed to provide food to the animals. These were "breeder cages" housing two animals each; it was presumed that the partners were cannibalized. Employees had failed to check these cages over a nine-day period.
124	16X	June 9, 2022	Mice were given expired ketamine (an anesthetic) on ten occasions over a six-month period.
125	16Y	June 9, 2022	A mother mouse and her litter of seven pups were found dead in a cage. It was determined that there was very little water in the water bottle—and the caretaker had failed to provide water over the weekend. The mice had sunken eyes with a general look of dehydration.
126	16Z	June 9, 2022	A marmoset infected with Mycobacterium tuberculosis escaped from an isolate cage during a procedure and crawled up the containment caging exhaust tubing and into the HVAC system located in the building interstitial space. When attempts to lure the

107	17.4	Lung 0, 2022	marmoset out failed, the laboratory resorted to gassing the marmoset with CO2. This incident took place in Building 33's ABSL-3 suite.
127	17A	June 9, 2022	Twenty-three dead and three moribund mice were found in 11 cages in the same area of a mouse rack. The dead animals had been cannibalized. It was determined that husbandry personnel failed to adequately monitor and report the health status of the mice in the 11 cages.
128	17B	June 10, 2022	Animal technicians failed to appropriately install valves into Hydropac pouches. As a result, mice held in the cages began showing clinical signs of disease consistent with potential dehydration. All of the animals were killed.
129	17C	July 5, 2022	Four mice were given a food that was laced with an antibiotic, even though this was not in the approved protocol. The mice were killed.
130	17D	July 5, 2022	Twenty-seven mice underwent an approved surgical procedure; however, the surgical technique used differed from the approved description. The lab exceeded the number of mice approved to be used. Also, the experimenters used inappropriate surgical technique, inadequate analgesia, inappropriate aseptic technique, and lack of appropriate hemostasis.
131	17E	Aug 10, 2022	Three mice were injected with an experimental compound at a higher dose than that listed in the approved protocol.
132	17F	Aug 10, 2022	A dead mouse was found floating in a water maze. The experimenter had failed to see the struggling mouse and remove him/her from the water. According to the report: "The researcher failed to see this struggling mouse because [s/he] had swum to the wall of water tank where the researcher did not have good vision of the mouse. The researcher also said they were afraid of moving as this might disturb the testing and the video tracking system. The researcher failed to fish the mouse out of the water as the net on hand was too short and they did not drain the tub as they needed

			more supplies to make the water murky for following experiments."
133	17G	Aug 10, 2022	A lab carried out a pilot experiment on 25 mice, which had not been approved by the ACUC. Three of the mice died, but the NICHD veterinarian was never contacted during this time.
			The experiment involved inflicting combined stressors on mice— including 24-hours food and water deprivation, behavioral testing, and the reintroduction of high sucrose water. The three mice died after sucrose water reintroduction.
134	17H	Aug 10, 2022	Thirty young zebrafish ("zebrafish fry" – these particular individuals were 13 days old) held in a dish were discovered to be dead in an incubator. They apparently had not been fed, nor did they receive water changes. One staff member had not been trained to check incubators, while the other just failed to check the incubators.
135	17I	Aug 10, 2022	Over the holiday weekend of June 18-20, three monkeys lacked water for some portion of the weekend. One monkey died. It was discovered that a portion of the waterline going to the specific quad unit in which the four monkeys were held was not properly assembled. The weekend caretaker admitted that she had not performed the required lixit check the day before, but had falsely reported that she had. That worker is no longer at NIH.
136	17J	Aug 10, 2022	Three weanling mice held in a cage were found to have no food; they did have access to water. One mouse was found dead, and the two others were moribund and were immediately killed. According to the investigation conducted by NICHD's ACUC, several people failed to notice the lack of food.
137	17K	Aug 16, 2022	A cage confining a litter of seven two-week-old mice was found to be flooded. All seven mice had died of hypothermia. It was determined that the animal caretaker had not conducted the mandated health checks properly and therefore did not notice the flooding in time to save the mice.

138	17L	Aug 16, 2022	Twenty-four mice were administered an unapproved drug via oral gavage, daily for 13 days. Six of the 24 mice were deemed to be in poor enough condition that the investigator was instructed to kill them. The investigator's access to the lab was suspended for 6 weeks.
139	17M	Sept 16, 2022	An individual began using monkeys in a protocol, even though their name had not been added to the protocol ("Determinants and long-term consequences of opioid and cocaine self-administration in squirrel monkeys").
140	17N	Sept 16, 2022	Four mice were noted by animal care staff as being lethargic, and were killed. It was determined that four hours earlier, the mice were administered a drug designed to cause muscle injury. The concentration of the drug had been approved by the ACUC, but the drug was administered at a greater volume than approved.
141	170	Sept 16, 2022	A caretaker mistakenly opened a divider panel latch between two cages and removed the divider panel—giving the two rhesus macaques in the cages access to one another. The macaques fought, resulting in injuries to both animals. One monkey suffered multiple small lacerations, requiring sutures, antibiotics and analgesics and recovered well. The other monkey sustained severe injuries that required surgical repair and follow-up sedations and treatment by the veterinarian due to infections and complications.
142	17P	Sept 16, 2022	Mice were subjected to retro-orbital blood collection as part of a training session for veterinary technicians. The mice were under general anesthesia for the procedure, but were allowed to recover—even though the protocol specified that the mice should be killed at the end of the procedure.
143	17Q	Aug 10, 2022	A female African Green monkey was observed to have a laceration to the base of her tail. X-rays confirmed that she suffered with a tail fracture. The cause of the injury could not be determined. Staff were reminded to be more vigilant while working with monkeys with long tails.

144	17R	Sept 27, 2022	Mice held in multiple cages were observed to be receiving tamoxifen. However, the protocol failed to flag tamoxifen as a chemical hazard. "The facility practice is for the investigator to place a standard hazardous chemical sticker on the cage card. It was subsequently determined that the wrong protocol number was placed on the cage card, thus the facility staff were not aware of the hazardous chemical present in the cages."
145	17S	Sept 21, 2022	Four mice were observed to be in poor health by the vet staff. Two of the mice improved with supportive care, but two mice died. It was subsequently determined that the mice had been treated with tamoxifen, but this chemical was not included in the protocol. Also, the cage was not labeled to indicate the chemical hazard.
146	17T	Sept 29, 2022	 Four mice held in a cage were found to have no food in the hopper. Two mice had died, and two mice recovered with supportive care. In a second incident, three mice held in a cage died after employees failed to give food to these animals. "It was determined that in both cases, a new cage was inadvertently set up without food late in the week, and in each case the oversight went undetected until the following Monday."
147	17U	Dec 1, 2022	Experimenters drew blood from mice, even though this procedure had not been approved by the ACUC.
148	17V	Dec 7, 2022	After rats were used in experimental surgeries, experimenters failed to fill out post-operative cage cards—failing to list the date of the surgery or the treatments given. This particular laboratory had failed to fill out post-operative cage cards on at least three previous occasions over a period of a few months.
149	17W	Dec 29, 2022	Four mice died because they were given the wrong water following a surgery to remove their adrenal glands. The mice were supposed to receive saline water, but received deionized water instead. There was a misunderstanding as to which individuals were responsible for providing this water.

150	17X	Dec 29, 2022	Six mice were used in an unapproved pilot study. They experienced weight loss greater than what had been approved in the original animal study protocol (20% weight loss), thus exceeding the approved humane endpoint.
151	17Y	Jan 4, 2023	A subadult male monkey, unintentionally paired with another male macaque, was reported to have significant fight wounds in multiple locations. The sounds required sutures, pain medications, and antibiotics.
152	NCI-Frederick- 2Z	March 1, 2018	A mouse with an infected uterine prolapse had been inappropriately treated with ointment and kept alive rather than being immediately euthanized per institutional policy. According to the report: "The prolapse was necrotic, malodorous, infected, and completely prolapsed (2-3 cm)."
153	NCI-Frederick- 3A	July 3, 2018	Experimenters failed to follow euthanasia procedures. A baby mouse was found alive in a refrigerator intended for dead animals. The protocol required that the mouse be decapitated (as a secondary method of killing) after carbon dioxide gassing.
154	NCI-Frederick- 3B	Aug. 2, 2018	Experimenters failed to remove animals from a "biohazardous bag" in a timely manner. The animals were thought to be biohazardous waste and were disposed of.
155	NCI-Frederick- 3C	Sept. 14, 2018	Experimenters failed to provide food to two mice being used as "sentinels."
156	NCI-Frederick- 3D	Dec. 4, 2018	Experimenters failed to provide post-surgical analgesia to mice who suffered with open wounds.
157	NCI-Frederick- 3E	March 22, 2019	A post-doctoral student was stuck by a needle while performing injections on an animal assigned to a protocol; the student was not listed on the protocol.
158	NCI-Frederick- 3F	July 25, 2019	Several unattended mouse cages were found in the carbon dioxide gas chamber, but the gas flow was shut off. One mouse was still alive, but under sedation.
159	NCI-Frederick- 3G	Dec. 9, 2019	In one animal room, three mice in a cage were found dead of apparent dehydration; another cage was discovered with one mouse found dead and two additional mice in the cage had to be

			euthanized due to apparent dehydration. In the same room as above, five mice were found with severe malocclusion and were euthanized. The previous day, an additional mouse had been euthanized due to malocclusion.
160	NCI-Frederick- 3H	Oct. 13, 2020	Two animals were found dead in their cage. Two additional animals were moribund and had to be euthanized. Two other animals survived, but were dehydrated; since they could not be used experimentally, they were also killed. The lixits on the water bottles had been turned up, preventing the animals from accessing water, and this was not noticed for 3 days.
161	NCI-Frederick- 3I	Dec. 23, 2020	Two cages on a rack were found without food and one of the cages contained one dead mouse and two moribund mice who were subsequently euthanized.
162	NCI-Frederick- 3J	Feb. 25, 2021	A mouse was found dead after his or her tail became caught in the lid of the cage—and the mouse could not access food or water.
163	NCI-Frederick- 3K	April 13, 2021	After 20 mice were injected with insulin for an insulin tolerance assay, the animals quickly succumbed to toxicity from the injection. Five animals died and the remaining were euthanized due to their poor condition. A review of the incident determined that the mice had received an overdose of insulin due to a calculation error during formulation.
164	NCI-Frederick- 3L	Oct 26, 2021	Two mice were found dead in their cage. The mice had been placed on the side of the rack that had deprivation caps rather than on the side where automatic watering was available.
165	NCI-Frederick- 3M	Dec. 20, 2021	Eight mice were placed on a biocontainment rack on Friday, Dec. 3, 2021. On Monday, Dec.6, 2021, seven of the eight mice were found dead, and the remaining mouse was euthanized due to his/her poor condition. It was determined that ventilation to the biocontainment rack had not been turned on, resulting in poor ventilation.

166	NCI-Frederick- 3N	April 1, 2019	A cage holding mice flooded, resulting in the death of 12 animals. The flooding occurred because the cage was improperly seated, causing a water valve leak.
167	NCI-Frederick- 3O	Oct. 24, 2022	Mice were given an expired analgesia (buprenorphine) prior to being used in a training session. The mice were euthanized at the end of the session. In a separate protocol, mice on a pancreatic tumor injection study were administered an expired analgesia (buprenorphine).
168	NCI-Frederick- 3P	Nov. 28, 2022	Three cages holding mice flooded, resulting in the deaths of 13 mice.
169	NCI-Frederick- 3Q	Nov. 28, 2022	Five mice underwent mammary fat pad implantation surgery. However, the individual who performed the procedure was not yet fully certified to perform the procedure.