“Biomedical scientists’ recognition and alleviation of pain and distress in laboratory rats and mice”

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Overview

Scientific research on animals is a well-publicized ethical debate in our society. In part because of increasing attention to this issue, national legislation regarding laboratory animal welfare was created in 1966 with the Animal Welfare Act (AWA) and has been strengthened in the recent decades with its subsequent amendments, particularly the 1985 Improved Standards for Laboratory Animals. A separate law, the Health Research Extension Act, was also passed in 1985. However, research has shown that these regulations are not implemented uniformly throughout individual laboratories and institutions. Therefore, laboratory animals are not always adequately or equally protected among institutions. This variation is mainly due to the subjective nature of what constitutes proper laboratory animal use. For example, both of the above-mentioned laws require scientists' to recognize and minimize "pain and distress" in their research animals, but does not provide definitions of these states or directions for minimization. Therefore, because each research institution is left to its own interpretation of the regulations, the laws are difficult to enforce in a standardized manner. If the regulations are to work as intended on a broad scale, the intricacies of applying these welfare regulations must be understood by and represented from the viewpoint of those directly involved – the scientists. The following study focused on biomedical scientists' recognition of, experience with, and minimization of "pain and distress" in rats and mice. Participants were engaged in an in-depth interview. From an examination of their responses regarding this subjective area of laboratory animal welfare, the barriers that hinder standardized regulation were explored. Results from this study suggest that focusing on refinement of the existing regulations would improve the welfare of laboratory animals more than adding to the legislation. Documentation of procedures and results, dissemination of information, and providing objective measures for pain and distress are some of the solutions proposed to enhance regulatory compliance. In this way, strategies to ensure that laboratory animals are humanely treated are undertaken by the very group who is required to adhere to the animal welfare legislation and may thus be more viable.

Table of Contents

I. Introduction/Specific Aims

The recognition and alleviation of pain and distress in laboratory animals is one of the many government requirements in two Federal laws, passed in 1985, which address the welfare of research animals. These regulations exist to provide standardized oversight of the use of animals in scientific experiments, nationwide. Some states also have separate laws that govern the treatment of research animals, but the majority of the pertinent regulations are contained within the the Animal Welfare Act of 1966 and its subsequent amendments, and the Health Research Extension Act of 1985.

However, many components of these laws are difficult to enforce because they do not provide clear and uniform standards that are applicable to all research institutions.[10,11,39] The regulations address a range of concerns, including: housing and veterinary care standards, the establishment of an institutional board to oversee animal research (usually known as the Institutional Animal Care and Use Committee), a search for alternative methods, and the proper use of anesthetics, paralytics, multiple survival surgeries, and restraint devices. The requirements for these issues are straightforward and are therefore easier to standardize between different institutions. However, these kinds of specifics do not reflect the way the majority of the legislation is written. Most of the legislation states requirements in very general terms. For example, many of the mandates in the two Acts are based on "performance standards" rather than "engineering standards", meaning that a required outcome is stated within the text of the legislation, but there are no guidelines or standard methods given for how to achieve that outcome.[37] Therefore, investigators and their institutions are charged with achieving the specified outcomes, but are given flexibility for how to determine a methodology for compliance. This "performance" approach can be desirable because many variables such as, species, previous history of the animals, facilities, expertise of the people, and research goals often make prescriptive ("engineering") approaches impractical and unwarranted.[20,33]

Although allowing for flexibility in achieving required outcomes is beneficial for many reasons, it also makes some of the requirements not only difficult to comply with, but also difficult to enforce. As an example, both laws require the "psychological enrichment of primates", but give no further directions.[2,22] This means that an institution that provides a plastic ball to a primate living in a wire cage is just as compliant as an institution that houses primates in an outdoor/indoor enclosure that provides a variety of different activities and social contact with other primates. Both institutions can claim that they are

providing for the "psychological enrichment" of their primates because there are no further specifics with which they must comply. This is just one example of the wide range for standards of care that are legal according to the regulations, similar scenarios can be seen in the requirements for post-operative monitoring or exercise standards for dogs. Therefore, because of the general terms in which the legislation is written, quality care is difficult to regulate and standardize between institutions.

One of the most notable examples of this problem is the requirement to minimize laboratory animal "pain and distress."[24,30,38] The identification, control, and reporting of pain and distress is clearly required in both federal laws, but research institutions and individual scientists are not given specific directions for how to recognize, monitor and alleviate pain and distress.

Therefore how is compliance with the stated outcome of "minimization of pain and distress" to be judged if no clear definition for pain or distress is delineated? The abstract and subjective nature of defining "pain and distress" is only one of many reasons this particular part of the regulations poses problems. There are a variety of other factors that affect regulative compliance that should be further explored and documented, including: different requirements per institution or per laboratory for minimizing these states, specific characteristics of the animal model, the experience and background of the researcher, the research field and focus of the investigator, the investigator's personal feelings about the regulations and/or animal research.

In the following report, I explore these ambiguities with ten biomedical researchers who, on a daily basis, work with laboratory animals subjected to pain and distress. My original goal was to explore how pain and distress is recognized and alleviated in the laboratory by each participant. It was my intention to then expand the interview into a more generalized discussion of the existing animal welfare regulations and how they affect the participant. However, throughout the interviews and in my succeeding analyses, it became very obvious that this particular factor – how the participant views the regulations – is more closely tied to actual welfare of the animals than I had originally thought. The view of the regulations and why they exist took on a rather significant role in the participants' responses to the questions regarding their role in regulative compliance. Although the interview questions only concentrate on the regulations that pertain specifically to pain and distress, it became apparent that the controversy surrounding the use of animals in research affected how the participants felt about the regulations in general. That is, the participants through their own volition addressed the controversy surrounding animal experimentation

and readily expressed how they were affected by it. The controversy has been the impetus for most of the regulations the participants must abide by. Because of these responses, I have also included an analysis of this controversy in the United States and the origins of the regulations. Based on their responses, it is important to note that the participants' level of respect for these regulations may in turn influence their level of compliance.

For the goal of promoting recognition and alleviation of pain and distress in laboratory rodents, I believe it is also crucial to examine how this goal has been represented to the biomedical scientist. Therefore the concerns, questions, and examples contributed by the participants about pain and distress in laboratory rodents, as elucidated in the following report, offer an important and necessary viewpoint on how these regulations are transferred into actual practice in the laboratory. The findings of this report are based on the thoughts, questions, and concerns of those who are most affected by the regulations. At the end, a summary of recommendations is provided regarding possible ways to increase the welfare of laboratory rodents that experience pain and distress. These recommendations come directly from the following analysis of how the important aspects of this issue affect these participants and their work.

A. Specific Aims

The purpose of this study is to:

- document and analyze the participants' 1) recognition of laboratory animal pain and distress 2) experience of working with pain and distress in their laboratory animals and 3) perceptions of the government regulation of pain and distress.
- find common themes and problematic areas of the regulations from an assessment of the interviews
- provide a basis, rooted in actual experience, for the improving the oversight of laboratory animals' pain and distress

These aims are achieved through a qualitative analysis of interviews with biomedical researchers who work with rats and mice (see Appendix A). In the first section, the questions explore the participants' abstract relational definitions of the following terms: pain, distress, suffering, anxiety, aggression, and fear, as well as, each participant's detection of these states. In the second section, the participants are asked to discuss their experience with these states as shown in laboratory animals. They are asked for examples

from their own experience and are also asked to verbally evaluate and decide how they would handle hypothetical ethical scenarios and to discuss the most pertinent factors in their decisions. Lastly, participants are asked to share their knowledge and beliefs about the regulations that focus on pain and distress in the laboratory and how government involvement affects them as scientists.

B. Background

1. Animal experimentation and the controversy up to 1966

Documented accounts of animal experimentation go back thousands of years. The Greek physician, Claudius Galenus (AD 129-200) is thought to be the first to record the use of vivisection for scientific inquiry.[18] Societal respect for science grew during the period between 1500-1800 known as the Scientific Revolution.[5] Specifically, biological science progressed rapidly in the 18th and 19th centuries. This advancement was affected in part because better transportation during this period made traveling easier and therefore more extensive. As humans experienced more of the world, they were more educated about the similarities and differences in biological systems and this led to new theories about our own place among the living world. These ideas contributed to the growth of the anti-vivisection movement as well. Although the anti-vivisection movement was not new, it gained more popularity during the Victorian Age in England largely based on the same factors that supported the growth in biological inquiry.

For example, Charles Darwin based his theory of natural selection on his trip to the Galapagos Islands in 1835. He detailed what he had seen in his book *Origin of Species* and had a significant impact not only on biology, but also upon his culture. Based upon the analyses in this book he outlines the theory of human evolution linking us biologically with animals. Further scientific advancements during this period and after, such as Gregor Mendel's explanation of genes and Louis Pasteur's discovery of the unseen, but powerful, existence of bacteria and viruses not only gave credibility to the theory of evolution, but also gave more reason to utilize animals in scientific experiments based on biological kinship to humans. Specifically, for the fields of physiology, anatomy, and medicine animals proved to be very useful models, cheap and easily accessible, from which to gather a vast amount of information. Importantly, the use of animals for medical research was proving its effectiveness and value to the general populace with the advent of new vaccines for humans, such as polio, smallpox, and diptheria that were developed from the use of laboratory animals.[45]

Interestingly, the growing sophistication of the field of biology, specifically evolutionary biology, provided advanced arguments both for and against the use of animals in scientific exploration. Although animals were becoming imperative in the laboratory for investigating the function of the living body, socially their use was becoming more problematic. [1,18] As science advanced and the theory of evolution became more accepted, it also contributed more reason to question the previously-held belief of human-centered control of the environment and the animals within, including the use of animals in experimentation. For example, if the theory of evolution is true, then any "specialness" attributed to humans is negated through biological proof of our evolutionary connection. Furthermore, because there is scientific evidence that human arose from a similar source as other animals, it also diminishes the entitlement of humans to control whatever is not human. This idea is especially relevant to the background of the animal experimentation controversy because it offers a core explanation for why the human use of animals is so troubling to some. If humans and animals are linked biologically, doesn't that also mean that animals may share some of the same traits? Including the ability to feel pain and suffer? This idea was well articulated by Jeremy Bentham, an English philosopher in the early 1800s well before Darwin's famous journey, who asked the question, "It is not can they think, but can they *suffer*?[4] This statement provides a foundation for the simultaneously growing ethical debate about animal experimentation. Ironically, the growth in biological understanding escalated this school of thought.

Despite the growing regard for animal welfare, animal research was becoming the accepted norm in biomedical science, the number of animals used in laboratories increased significantly. In concert with this increase came an increase in groups that were dedicated to anti-vivisection. As early as 1824, the "Society instituted for the purpose of preventing cruelty to Animals" was established in England, later to become the Royal Society for the Prevention of Cruelty to Animals (RSPCA). In 1857, this Society started to focus on animal experimentation with the goal to abolish painful animal research.[13] The antivivisection movement in England captured the public's interest in a relatively short amount of time and by 1871, the British Association for the Advancement of Science developed guidelines for conducting physiological experiments.[24] And soon after, the Cruelty to Animals Act of 1876 was passed which was the first legislation to protect research animals.[37]

This ethical debate took hold in Europe well before it did in the United States where it wasn't until 1866 that an organization was even created that focused on animal welfare.

The American Society for the Prevention of Cruelty to Animals (ASPCA) founded by Henry Bergh was based upon the RSPCA in England that was formed 42 years earlier. Henry Bergh also introduced the first anti-vivisection legislation in New York in 1880, but it was defeated. But the popularity of the issue grew quickly and by 1910, 131 anti-vivisection societies existed in the United States, such as other states' SPCAs, like the Massachusetts Society for the Prevention of Cruelty to Animals (MSPCA), the American Anti-Vivisection society, an off-shoot of the Anti-Vivisection Society of England, the American Humane Association, and the New England Anti-Vivisection Society (NEAVS) to name a few of the most substantial groups. [3]

The scientific establishment contributed to the growing debate during its early stages by publicly extolling the necessity of animal research for medical advances. In 1896, the National Academy of Sciences issued a statement to United States Senate affirming the need for animals in laboratory research. This letter was in response to a federal anti-vivisection bill that had been introduced into Congress by Senator Jacob H. Gallinger which was later defeated.[6] In 1908, the American Medical Association (AMA) officially responded to the growing criticism about animal use in science by forming a Council for the Defense of Medical Research.[7] Its chairman, Walter B. Cannon, a physiologist at Harvard Medical School, regularly wrote articles to national newspapers and popular magazines articulating the need for animals in research.[7] Yet, he also understood the benefits of having a controlled program for laboratory animal use and in 1910 formed a committee at Harvard to devise a set of uniform laboratory rules for the care and use of research animals.[7] He also was one of the first to attempt to alleviate the growing controversy by collaborating with the opposition. In 1914, he made overtures to the AHA and the MSPCA, two of less radical anti-vivisection groups, to create and maintain an avenue of open communication between the welfare groups and the Council.[7]

The foundations for the animal research controversy had thus already been laid by the early 1900s, but were to grow even more acrimonious. Animal research boomed around the 1950s as public funding for science grew. Again, with the increase in animal use, there was an increase in its opposition. Membership in animal welfare groups not only increased toward the latter half of the twentieth century, but the groups started to diverge from each other into separate philosophies of animal *welfare* versus animal *rights*.[9]

Although the public was becoming more engaged in the controversy, the earliest attempts at providing guidelines and instructions for proper laboratory animal care on a national scale came from inside the scientific establishment. For example, in 1962, the

National Institutes of Health (NIH) with the National Academy of Sciences' Institute of Laboratory Animal Resources, published the *Guide for Laboratory Animal Facilities and Care.*[20] The *Guide* has been revised numerous times since the first edition in 1962 and is known today as the "Bible" in laboratory animal care. In addition, the American Association for Accreditation of Laboratory Animal Care (AAALAC) was organized in 1965 in order to provide a voluntary program for assessing laboratory animal facilities and animal care and use.[29] Still today, AAALAC encourages very high standards for the care and use of laboratory animals.

It was also during this period that the first national legislation was passed that outlined protection for research animals. The Animal Welfare Act of 1966 protected pets (dogs and cats) from being used as laboratory subjects.[12,17] The passage of this Act was influenced by an expose in two national magazines, *Life* and *Sports Illustrated.* The articles described the practice of unscrupulous dealers that stole family pets to then sell to research institutions for profit. The public outcry from these articles was tremendous and Congress received a vast amount of mail from concerned citizens demanding a bill to protect pets. Therefore, the first version of the Animal Welfare Act was not meant to control or inhibit the current practices of animal research within the laboratory in any way. The purpose and motivation of the first AWA is summarized well in the following quotation in 1966 from one of the Acts sponsors, Senator Warren Magnuson, of the state of Washington.

> "*I would like to emphasize that the issue before us today is not the merits or demerits of animal research. We are interested in curbing petnapping, catnapping, dognapping, and protecting animals destined for research laboratories, while they are in commerce. We are not considering curbing medical research. I have always considered myself a friend of the medical researcher... Yet, we do not think we can allow the needs of research, great as they may be, to promote either the theft of a child's pet or the growth of unscrupulous animal dealers*."[36]

2. Animal experimentation and controversy since 1966

As described, the animal protection legislation started quite minimally in this country. Not only did it cover only a selection of species used in research laboratories, but only addressed the proper *acquisition* of these laboratory animals with no mention of the care and use of the animal once it arrived at the research facility. It did not account for animals born within the facility, nor did it say anything about how the animal was to be treated *during* the experiment or after. This was to be corrected in a significant way twenty years later.

The limited nature of the 1966 Animal Welfare Act was apparent to the growing number of groups dedicated to anti-vivisection in the United States. According to one study, by the 1970s there were ten times as many anti-vivisection and animal rights groups as had been totaled in 1910.[3] There are several reasons for the increase in number of these groups during this time period. These include the increase in public funding, the role of the media, and changing attitudes regarding a mistrust of society's establishments and the federal government. [31] A thorough treatment of these issues is beyond the scope of this report. However, for the purposes of this report, it is important to analyze the circumstances leading up to, as well as the proponents of, the more substantial regulations that were passed in 1985.

Animal experimentation became an even more intense societal controversy during the 1970s and 1980s based on several occurrences that highlighted several animal welfare problems within highly respected research institutions. Just as important was how the problems were publicized in the media and the methodology used by anti-vivisection groups to uncover them.[28, 42] As detailed below, these instances helped create the emotional state of the debate.

In the late 1970s and 1980s, the idea of Animal Rights gained momentum and attracted many new members, and funding, to its cause.[27, 31] This was facilitated by several dramatic incidents that were heavily publicized and therefore gained mass attention. For example, in 1976, famed animal rights activist, Henry Spira, organized a successful protest outside the National History Museum that housed a laboratory that performed castration on cats for part of its research. Spira, by organizing numerous rallies outside the Museum was able to build public sympathy and was able to create a very useful forum for his arguments regarding the questionable cost-benefit analysis of the particular experiments.[42, 46] He skillfully attracted so much attention that the Museum and its funding agency, the NIH, had to react. As a result, a year after he had begun his public opposition to these experiments, NIH pulled the laboratory's funding. His appeal to the public to help him oppose these experiments worked in his interest very well. Spira used this approach again in his campaign against Revlon cosmetics. He bought a full-page ad in the New York Times that used a disturbing picture of a rabbit in pain from undergoing a test that measured the toxicity level of chemicals called the Draize eye test.[42, 46] Public outcry was enormous. Not only was he able to convince Revlon to stop these tests, but was also able to encourage them, along with other cosmetics companies, to contribute funding to create the Johns Hopkins Center for Alternatives to Animal Testing in 1981.[8] These successful efforts by

Henry Spira provided a new and exciting methodology for those involved in the Animal Rights movement. Using the media to gain sympathy and support from the public was catching on quickly among others within the movement since it was not only a successful strategy, but produced much faster results than supporting an often compromised piece of legislation that could take years to even be considered by Congress. The tactic used so successfully by Henry Spira was copied by many animal welfare/rights groups in the years to come.[28,36,37,42,44]

However, as the anti-vivisection movement grew in popularity, it came to be controlled by extreme Animal Rights groups that not only used media appeal to garner support for their cause, but eventually also used terrorist actions, such as laboratory break-ins, destruction of property, kidnapping laboratory animals, death threats, and even acts of outright violence toward scientists. In 1980, Alex Pacheco, co-founder of the militant animal rights group, The People for the Ethical Treatment of Animals (PETA), gained access into a primate laboratory by volunteering under the guise of a student volunteer. He videotaped the monkeys used in the laboratory's experiments and from this footage, animal cruelty charges were pressed against the Principal Investigator, Dr. Edward Taub. National attention was created for what came to be known as the Silver Springs Monkey Incident.[23, 35] This case was monumental because Dr. Taub was convicted by a Maryland court of 119 charges of both animal cruelty and inadequate veterinary care.[35] Maybe most damaging though was the revocation of his NIH grant because of the scandal. In a like manner, Dr. Thomas Generelli from the University of Pennsylvania lost his grant funding when videotapes of abuses in his laboratory also became publicized through an activists' infiltration into his laboratory.[14,15] Both of these instances ended in severe penalties for the researcher as well as mass public attention on the treatment of laboratory animals. Other researchers have been the recipients of similar raids, have been harassed at home, or threatened with death (Morrison, 2000) By 1989, it was reported that in the previous nine years, animal rights groups staged more than 29 raids on research facilities in the United States, stole over 2000 animals, and caused more than 7 million dollars in physical damage and loss of research documents.[49] And in 2002, the annual budget of People For the Ethical Treatment of Animals was estimated to be at $13 million. [31]

The growth of the animal rights movement and the specific tactics used brought media attention that was very effective at capturing the public's attention and sympathy. Largely in response to the escalating publicity of substandard animal care within research institutions and the growing concern of the voting public, two federal laws were passed. In

1985, an amendment to the Animal Welfare Act, the Improved Standards for Laboratory Animals and the Health Research Extension Act were both signed into law. These laws were far more comprehensive than the existing Animal Welfare Act and provided protection for animals used for scientific purposes covering a wide range of topics from the beginning to the end of each animal's use in the laboratory.[2, 22] It is important to note the time period of the 1985 legislation because it shows the influence that the public can have in the legislative arena.

II. Pertinent Regulations on Laboratory Animal Use

A. "Pain and Distress"

The following are the regulations, as written, that focus on pain and distress:

Animal Welfare Act – 1985 Standards for Laboratory Animals[2]

Sec. 2.31 Institutional Animal Care and Use Committee– "Procedures involving animals will *avoid or minimize discomfort, distress, and pain* to the animals. The principal investigator has considered alternatives to procedures that may cause more than *momentary or slight pain or distress* to the animals…procedures that may cause more than *momentary or slight pain or distress* to the animal will: be performed with appropriate sedatives, analgesics, or anesthetics, unless withholding of such agents is justified for scientific reasons…involve in their planning, the attending veterinarian. Animals that would otherwise experience severe or chronic *pain or distress* that cannot be relieved will be *painlessly* euthanized at the end of the procedure or, if appropriate, during the procedure." Section 2.36 Annual Report (7) – "State the common names and the numbers of animals upon which experiments, teaching, research, surgery, or tests were conducted involving accompanying *pain or distress* to the animals and for which appropriate anesthetic, analgesic, or tranquilizing drugs were used." (8) "State the common names and the numbers of animals upon which experiments, teaching, research, surgery, or tests were conducted involving accompanying pain or distress to the animals and for which appropriate anesthetic, analgesic, or tranquilizing drugs would have adversely affected the procedure, results, or interpretation…An explanation of the procedures producing *pain or distress* in these animals and the reasons such drugs were not used shall be attached…"

Health Research Extension Act of 1985– Public Health Service Policy on the Humane Care and Use of Laboratory Animals- Public Law 99-158[22]

Sec.495(c)- "The Director of NIH shall require each applicant for a grant…involving research on animals which is administered by the National Institutes of Health…to include in its application…(1) assurance satisfactory to the Director of NIH that scientists, animal technicians, and other personnel involved with animal care, treatment, and use by the applicant…and the concept, availability, and use of testing methods that limit the use of animals or *limit animal distress*;…"

"U.S. Government Principles for the Utilization and Care of Vertebrate Animals Used in Testing , Research, and Training"[26]

VI. "Proper use of animals, including the *avoidance and minimization of discomfort, distress, and pain* when consistent with sound scientific practices is imperative…investigators should consider that *procedures that cause pain or distress in human beings may cause pain or distress* in other animals."

VII. "Procedures with animals that may cause *more than momentary or slight pain or distress* should be performed with appropriate sedation, analgesia, or anesthesia…"

VIII. "Animals that would otherwise *suffer severe or chronic pain or distress* that cannot be relieved should be painlessly killed at the end of the procedure, or, if appropriate, during the procedure.

B. The Institutional Animal Care and Use Committee

Both Federal Acts also require a governing body, or committee, to oversee all aspects of the animal care and use program of research institutions. It is the role of these committees of every institution to make sure that institution is in compliance with the Federal regulations. Beyond several, general guidelines (see below), each Committee, referred to in the regulations as an Institutional Animal Care and Use Committee (IACUC), is able to devise its own Policies and procedural norms to serve its own institutional needs and goals. Although the two Federal laws differ slightly, the vast majority of the requirements are the same. The following are the major points included in both Acts that affect the IACUC:

i. Each IACUC member should be appointed by the Chief Executive Officer, President, or other Institutional Official.

ii. In the AWA, the Committee must include a veterinarian and an unaffiliated member and gives a minimum number, 3

iii. The PHS Policy makes the same requirements, but adds a non-scientific member to the necessary roster and its minimum requirement is higher at 5.
iv. The Committee must review the animal facilities and Approved procedural spaces at least semi-annually.
v. The Committee must review and approve, require modifications to, or withhold approval of each animal use protocol and review each protocol annually.
vi. The Committee must submit necessary reports to the United States Department of Agriculture (USDA) and the Office of Laboratory Animal Welfare (OLAW) at the National Institute of Health (NIH).
vii. Must comply with the standards of animal care as outlined in the *Guide for the Care and Use of Laboratory Animals*.
viii. Be authorized to stop or limit any research or activity that involves animals.
ix. Review concerns involving the care and use of animals at the institution(s).
x. Make recommendations to the Institutional Official regarding any aspect of the institution's animal program, facilities, or personnel training.[25]

II. Significance

A. How the Controversy Has Affected Biomedical Scientists' View of the Regulations

Attentiveness to the welfare of laboratory animals and ethical discussions about their use was increasing among scientists as well. One study charted the growing number of articles in the scientific and medical literature between 1966-1985 that openly discussed the ethics of animal experimentation and shifting opinions. The authors found a rise not only in the number of articles discussing the welfare of laboratory animals, but also a rise in the amount of research that focused on animal welfare.[37] This same study also compared the tone of these articles. The authors state that a difference is apparent between the articles written prior to the most intense animal rights activities (in 1978-1981) as they were more conciliatory in nature, than in the time period that followed, between 1982-1986 where a defensive tone was more prevalent. This study suggests that the extreme tactics of the animal rights groups had a detrimental affect on the scientific establishment and encouraged a defensive posture, as expressed in the journal articles. These activities, in hindsight, worked to stifle cooperative efforts between scientists and animal welfare workers for an increase in welfare standards within laboratories. In addition, because of the

media spotlight on the controversy, animal researchers often expressed a feeling that they are picked out unfairly for criticism by the public at large. [19]

Because the proponents of animal rights were the ones largely responsible for encouraging the public attention, they are also credited with being the impetus behind the passage of the laws during this period.[16,21] The majority of the animal research regulations were passed during a time period when animal rights proponents were publicized well, strong in number, and in financial resources. Since scientists who conduct their research on animals are the ones directly affected by the regulations this association between the regulations and animal rights (or welfare) proponents causes mistrust and bitterness toward the regulations. [10,16,21,36] This means that the overall goal of improved welfare regulations within research laboratories is often identified with animal rights groups only, creating a significant obstacle for the promotion of these ideals.

Often, the regulations are seen as a way of "giving in" to the activists. During a discussion of the Three Rs (reduction, refinement, and replacement)[43], a technique adopted by the research community regarding the use of animals, a biomedical scientist stated the following: "this response [establishment of the principles of the Three Rs] came across as a confession of guilt. Although scientists accept high standards for the use and care of research animals, they are not engaged in some kind of necessary evil. Appeasement is a losing game. To make concessions on a matter of principle is to concede the principle itself. Then defeat is only a matter of time, as opponents [animal right activists] demand complete consistency with their own principle."[19] This viewpoint was becoming increasingly popular among biomedical scientists. Unfortunately, their growing suspicions about welfare-related items within their institutions and on a broader scale was supported by the increasing number of protests and other activities, as well as, the rising membership of these groups even after significant 1985 regulations had been passed. "But even as the handling of research animals become more restrictive, the animal rights campaign became ever more demanding and violent."[19] Often, animal researchers were left feeling as though they had no voice in matters that were affecting them and no recourse to prevent their work from being jeopardized. "Scientists should realize that they are at the bottom of the policy food chain when it comes to deciding the future course of experimentation."[19]

Largely in reaction to the activities of the 1980s a severely polarized struggle now exists between the scientific community and the animal protection movement.[9,10,16,21,36] A study was done in 1995 that investigated the way members of each group viewed members of the opposition.[36] The author concluded that not only did both groups have a negative

view of the other, but the opposition was always viewed as having extreme characteristics and not having rational thoughts. The "other" group was always perceived as being almost incapable of logical thinking. This creates a very difficult basis on which to find common ground, although there may be plenty of room for agreement. This study also showed that in an analysis of the actual beliefs of the members of the opposing groups there was "agreement on the relative capacity of different animals to suffer" which would not be possible if the opposing groups differed in the extreme.

Although the terrorist activities still occur intermittently, they have subsided somewhat in the 1990s. Legislative attempts to further the rights of animals however, have not. Recently, a U.S. court recognized the legal standing of an individual to sue the federal government in order to force changes in the animal welfare regulations. And, the recent campaign to include rats, mice, and birds in the AWA has created a wave of protest from the scientific establishment. Researchers, largely because of the dramatics of the past, view this particular campaign very skeptically.

"But one need not be overly cynical to suspect that the real motive behind the push for inclusion is not a desire to protect the rodents' welfare so much as a wish to impede research by all means necessary."[31]

And on the international scene, there are far more restrictions in recent years. In England, Japan, and Australia, the use of primates for research has become illegal. In other countries, animals are gaining rights in small increments. For example, "Last year in Germany, the ruling Social Democratic and Green parties introduced legislation stating that animals have the right to be "respected as fellow creatures" and to be protected from "avoidable pain."[19] This differs dramatically from the present legal definition of animals as "property" in the United States, but these changes abroad heighten the sense of urgency among scientists within the United States that their research is being threatened.

The scientific community is currently in a period of "reaction" against laws and regulations that are perceived as imposed on them by people that scientist view as dangerous and unscrupulous. As we will see this can have unfortunate effects on how the animal welfare regulations are viewed and followed. Whether or not the federal regulations that surround the welfare of animals have an impact on their treatment inside laboratories depends greatly on the attitudes of the researchers themselves.[37] As stated by Dr. Adrian Morrison, a veterinarian whose research on the brain utilizes cats, "The government

bureaucrats may not like it, but it is our ethical sense that is the final arbiter of our animals welfare."[19]

No exploration into the effectiveness of the welfare regulations should be made without discussing the dramatic background of them in this country. As suggested, the effectiveness of the regulations depends in large part on how they are perceived by the working scientist. With this background in mind, how *are* the regulations viewed by biomedical scientists? Do the regulations have an effect on the everyday circumstances in laboratories? Is animal welfare a consideration when scientists are performing experimental procedures or writing protocols? What is their direct experience with their own lab animals welfare? Therefore, I propose to use the views of biomedical scientists to further the goal of increasing the standards of care these animals will experience. I believe that a thorough and candid exploration of laboratory animal welfare can only come from the researchers themselves. It is my belief that through this avenue a more practical understanding of research procedures and any obstacles to welfare compliance can be achieved. This will help create and encourage practical goals that can be more easily accomplished in each laboratory. Also, if the suggestions come from the researchers themselves, they may be more inclined to accept them.

B. Responsibility of the Investigator

The following are the regulatory duties that each investigator must be responsible for in order to comply with these Federal laws. The *U.S. Government Principles for the Utilization and Care of Vertebrate Animals Used in Testing, Research, and Training*[26] lists these responsibilities. In summary, the *Principles* expect investigators to:

- Design and perform procedures on the basis of relevance to human or animal health, advancement of knowledge, or the good of society;
- Use the appropriate species, quality, and number of animals to obtain valid results;
- Avoid or minimize discomfort, distress, and pain in concert with sound science;
- Use appropriate sedation, analgesia, or anesthesia;
- Establish experimental end points;
- Provide appropriate animal husbandry directed and performed by qualified persons;
- Conduct experiments on living animals only under the close supervision of qualified and experienced persons.

C. Focus on Pain and Distress

The recognition and alleviation of laboratory rodent "pain and distress" in an appropriate area in which to investigate ways to increase the welfare of research animals because it is starting to gain more attention not only within laboratory animal medicine, but also in the legislative arena. [30,32,34,40,41] Therefore, this enables the participants to be questioned about welfare by using explicit and familiar examples and gives them a format with which to discuss regulative control more broadly. An analysis of the monitoring and alleviation of these states is useful for the overall goal of understanding how welfare is actualized in different laboratories.

Within both the Animal Welfare Act and the Health Research Extension Act are requirements for the "minimization of pain and distress" in laboratory animals. Yet, this remains a difficult area of the regulations to comply with and enforce, for several reasons. First and foremost, there is no widely agreed upon objective standard for deciding what constitutes "pain and distress". Only one area of the legislation gives a somewhat detailed explanation of "pain and distress". In the Animal Welfare Act, Sec. 1.1, Definitions of Terms, "…*painful procedure,* as applied to any animal, means any procedure that would reasonably be expected to cause more than slight or momentary pain or distress in a human being to which that procedure was applied, that is, pain in excess of that caused by injections or other minor procedures." This definition, in itself, can be understood differently. For example, what distress means to one person, may not be distressful to another. Or what exactly is meant by "momentary"? The definition could easily be interpreted in different ways. The description attempts to give some guidance by stating examples such as injections; but then becomes too general again with the use of "other minor procedures." And if pain and distress is not even characterized in the same way by all humans, how can the alleviation, minimization, and reporting of it be consistent throughout all institutions and all laboratories?

Secondly, scientific exploration into "pain" and the processes that produce and signal a pain response in animals is a relatively new area. Even scientists who specialize in pain research cannot define it in absolute terms. Therefore, how exactly is this requirement handled by research institutions and by scientists individually? And if pain is hard to test using objective measures, then one is forced to rely on the visual appearance of an animal. However, this leads to a host of other unreliable variables, such as, differences among individual animals in pain tolerance, prey behavior (hiding pain and sickness to avoid death by predators), sensitization, increased tolerance, etc.

In addition, if there is not enough knowledge about pain, there is even more of a deficiency about "distress." This suggests that a cognitive component is involved with animal pain and suffering, but animal cognitive research is even less developed than pain research. Along with the initial dilemma about how to properly define distress are the following additional questions. Do some procedures become more or less distressful after a certain length of time? How does the intensity of the procedure affect the level of distress? How does the social/behavior component of an animal's life affect its ability to feel distress? How do the living conditions affect the animal's ability to feel distressed? The questions are endless.

Therefore, it is useful to explore how scientists perceive these aspects of pain and distress since they are responsible for its "minimization". How can these issues be handled well by research institutions that, by law, need to prove that adequate standards of animal welfare take place within the laboratory environments of their facilities? What are the constraints that exist in understanding and monitoring laboratory animal pain and distress? We need an investigation of the current problems surrounding this issue, so that we know where to focus our efforts to promote animal welfare in research.

And, most importantly, what are the suggestions and concerns, *from the biomedical scientists,* about this issue and how it is regulated. It is imperative that the suggestions and solutions come largely from the biomedical scientist. If they are involved they will be able to create a system that works for them instead of being imposed upon them.

II. Research Design

A. Participant Selection

I interviewed ten biomedical scientists. The research from those selected centered around mammalian neural activity. The participants represented three different ranges of age and experience. Each participant is delineated by his/her title below. To make the analysis and discussion of the interviews easier, each participant is assigned a name for the purposes of this report.

Participant 1 – Adam – Research assistant

Participant 2 – Ben – Graduate student

Participant 3 – Corinne – Associate professor

Participant 4 – David – Graduate student

Participant 5– Elijah – Research assistant

Participant 6 – Frederic - Professor

Participant 7 – George - Professor

Participant 8 – Hank – Assistant professor

Participant 9 – Isabella – Assistant professor

Participant 10 – Jacqueline – Graduate student

The following criteria were used to select the pool from which all ten participants came:

- Principal Investigators who work at a large research institution that includes a medical school, graduate school, and a hospital.
- Principal Investigators who work in biomedical research
- From a listing of PIs within the medical and graduate schools, only those who work with rats and mice were chosen.

A different methodology was employed for contacting the first five participants (Adam, Ben, Corinne, David, and Elijah) than for the last five (Frederic, George, Hank, Isabella, and Jacqueline). They were solicited by an outside friend of mine who worked within the institution. One of the participants in this first group of five (Ben) then asked the other four to participate. The results from this first group served as a pilot study before solicitation of the second group of five participants. This second group of five was solicited after the study was approved by the Institutional Review Board (IRB) of the medical and graduate schools of the same institution. Therefore, different requirements existed for the last group of five than for the first group of five. The following description only applies to this last group of five participants (Frederic, George, Hank, Isabella, and Jacqueline).

The investigators of this study sent out a letter to thirteen biomedical researchers who fit the criteria of the study, including:

- 4 from the Department of Neuroscience
- 4 from the Department of Anatomy and Cellular Biology
- 2 from the Department of Psychiatry
- 1 from the Department of Biochemistry
- 2 from a specialized center that focused on sensory research

The letter of invitation was sent only to the Principal Investigator of each laboratory. In the letter, the study was very generally explained (see Appendix B) and the addressee was asked to respond to the student investigator if interested in participating. Both the student investigator's email and phone number were given. Because of IRB restrictions, the student investigator was not allowed to initiate any further contact with those who received letters of invitation. If the student investigator received an email or phone call from a potential participant, only then was the interaction allowed to go forward. If the participant then

initiated further contact, the logistics of meeting were discussed and confidentiality was again assured. Four out of twenty who received a letter of invitation responded. The fifth participant (Jacqueline) was solicited by one of the other four respondents (Isabella), as is explained below.

B. Interview Methodology

Before the start of the interview, the participants were asked to sign a Confidentiality Agreement, all of which I kept. After this, the participant was informed of the background of the student investigator, a very general explanation (so as not to affect the results) of the study, and the completely voluntary nature of the interview and that he/she may discontinue the interview at any time. After each interview, I asked the participant if there was anyone else in his/her laboratory who might also participate in the study. This produced one additional participant (Jacqueline).

Data collection

The interview consists of fifteen questions that all required elaboration (see Appendix A). The questions used were pre-tested and modified through the analysis of the pilot study (Appendix D). Therefore, the interview questions differ between the first set of five (pilot interviews - Adam, Ben, Corinne, David, and Elijah) and the second set of five (final – Frederic, George, Hank, Isabella, and Jacqueline). Two of the questions from the pilot interviews focus on pain and distress and three focus on regulations (Appendix D). The second set of interview questions was more focused on working with pain and distress and gave scenarios for the participants to work through. Therefore, throughout the analysis of this report, only the responses of five of the first set of interview questions (pilot study) are included because they are similar, if not the exact same, to the questions asked of the last five participants.

The following descriptions of methodology apply to all ten interviews. During the interview, the investigator prompted the participant only if a significant portion of the question was left unanswered. The investigator did not signal agreement, concern, or closure to any of the questions. Therefore, the length of the interviews were different for each participant depending on his/her level of involvement and degree of elaboration of the questions.

I transcribed the interviews exactly as they occurred. Each participant was given a number for identification to ensure confidentiality throughout the process of transcription

and this number is replaced by a name in the following analysis. After transcription, I combined each participant's answers according to the question, so that all the answers were directly compared. From this comparison, similarities, discrepancies, and any overall trends can be highlighted.

Confidentiality

The interviews were tape-recorded and the participants were informed of this within the letter of invitation and again in person just prior to the interview. Each participant was allocated his/her own tape for the study and each tape was labeled with the participant's identification number only. The participant did not identify him/herself or the details of his/her specific research on tape. To ensure complete confidentiality of each participant's answers, any biographical details and institutional affiliations that could possibly link an individual to his/her statements were omitted or altered. No one other than me knew the actual identities of participants. Furthermore, all records linking individual respondents and the interview transcripts were destroyed at the conclusion of this project. There are no copies made of the tapes.

IV. Findings/Analyses

A. Knowledge base about pain and distress in laboratory rats and mice

RELATIONAL DEFINITIONS

All ten participants were asked to differentiate between the following: pain, distress, suffering, anxiety, fear, and aggression. Everyone had significant difficulty defining these states and differentiating them. "I wouldn't necessarily distinguish [between the terms]...I don't know that I can" (Frederic) or "anxiety I can't define and I don't think that people can really define it although they try to" (Hank). "I don't think I would know the difference [between some of the terms] if I saw it (Isabella). The pain and suffering postures, or the ones that appear to be those, I think are the same. See, pain, suffering, and distress do seem to kind of fall together, only in that their appearance is kind of similar..." (Jacqueline). The most articulate comparison between pain and suffering came from Isabella, "I guess I think there's a slight difference between pain and suffering because pain can end and when I think of suffering I think of it being a longer term condition."

There were generalities that could be seen among the participants' definitions. For example, suffering was recognized by all ten participants to be the term that represented the greatest intensity. Furthermore, a cognitive component of "pain and distress" was

introduced by eight (except for Frederic and Hank) of the ten participants. Also, all but two (Frederic and Hank) of the participants mentioned psychological suffering in some manner. Six of the ten participants specifically mentioned "psychological suffering/distress". Adam, Ben, David Elijah, George, and Jacqueline all mentioned the anticipation of pain to be one of the causal factors of distress and suffering (more explicit examples are included in Section 2 below). None of the subjects qualified any of their answers based on phylogenetic characteristics meaning that no one classified any of the above states as only being felt by certain species.

Although everyone was initially uncomfortable with quantifying and describing "pain and distress", their knowledge base seemed extensive. This was shown from the length of their answers and the number of examples that respondents gave in an attempt to describe these states. As they started verbally working through the differences and characteristics of pain and distress, they came up with more ideas and, incidentally engaged the interviewer in abstract discussions of these states, using their own research animals and their experience with them as examples. They also seemed quite qualified in how to determine causation of these states, as shown below.

The participants were more comfortable when a term could be defined in an objective manner. Pain was the term most often linked to a measurable state. "Pain is something that can be measured objectively by looking at the chance to avoid pain or measuring the electrical potentials" (George). "I recognize distress when an animal huddles and is pilo-erect [hair stands on end, often seen when an animal fails to groom normally] and is not engaging in the usual activities of a rat or mouse" (Frederic). "...well, if there is long-term distress you would see it in the condition of the animal, if it doesn't eat or drink, or if it has diarrhea" (George). "I generally look for withdrawal of the animal from other animals" (Hank).

DETECTION

Since the recognition and subsequent alleviation of pain and distress comes from the investigators themselves, it is also imperative that each of these participants explains how they detect pain and/or distress. For the majority of the life of experimental animals, the investigators, research staff, and animal care personnel are the only ones who observe them at all. Therefore, do those who are responsible for the "minimization and alleviation of pain and distress," have the ability to determine these states?

Eight of then ten respondents believed they were rather good at the detection of pain and distress. "I think that in my lab we are hyper-sensitive to causing pain and

distress" (Isabelle), "Well, it's my job to teach these kinds of things to others in my lab. To observe the mice and know what is normal and to watch for what is abnormal" (Hank) "I'm probably very sensitive to causing animals pain and distress. I have a background in animal behavior, so I'm very used to observing behavior" (Isabella). Frederic thought that knowledge of pain and distress was straightforward enough that as long as people were taught what to look for then recognition of it was very similar among those that had previously worked with research animals. Most of the respondents also mentioned that knowledge of these states is imperative to good science. "With almost any research...you want to be sure you don't introduce undue stress or extraneous strain on the animal because that messes up your research" (Frederic). George stated that he was "pretty comfortable" in how to detect these states "because a lot of my research analyzes these kinds of things." Likewise, Isabella stated that "we need to be very cautious of disturbing the animals in any way that would affect their behavior" and Hank, "we work with genetically manipulated mice, so we have to know 'what a mouse does' and be cognizant of the fact that we've changed it and could give it a predisposition to [different behaviors]. We have to know the [biological] background, so this is the kind of thing that I have to teach people (research assistants, veterinary staff, and animal care technicians) that are taking care of the colony".

Interestingly, Corinne was very hesitant to describe her comfort level with detection of these states. She instead viewed this kind of answer as projecting her own understanding onto the animals and she explained that "people *think* [they are seeing suffering in the animal]..." and left the door open to the possibility that the way humans understand or feel these states is unknown in animals. However, Corinne was the only one to mention the influence that each animal's individual nature may have on each of the states and how they are experienced by that animal. Therefore, I do not believe that she did not think that animals could experience pain and distress, I think rather she was unwilling to hypothesize about these unknowns.

I also asked the participants to explain how well others could detect these states. Benjamin, David, George, Hank, Isabella, and Jacqueline thought that there was a definite division among scientists of different ages. "Younger investigators are more clued into [behavioral indications of distress] because that's what they've been doing...so I think you get the associate or assistant professor that knows more about it than the full professor" (Hank). However, Hank also stated "It seems to me that students really don't' have any experience, coming in as a graduate student, for instance. You get people coming in here with a background in chemistry or something like that and suddenly they work here and

they have no idea that there's even more than one kind of mouse." And two participants believed there was division, but that it was more along the lines of research field. "I find that there is a very big difference between people whose research depends upon the animals being in a stable state and unstressed and appreciate that and recognize signs when the animals are distressed and those who just want to grind up tissue and the animals are just another material in their lab. I think there is a very big difference" (Isabella). "I think that people that don't do surgery probably have more difficulty recognizing pain, distress, suffering, or people that don't see their animals on a regular basis" (Jacqueline). Elijah was visibly troubled by the way some investigators treated their animals "Yeah, I think with some post-docs' animal practice, especially foreigners...[gave example's name]. I hated watching him deal with the animals, just because he was so numb to using them, so I would bet if you were to talk to older scientists it would be very similar to that. They're just numb to it. They learned science when it was not regulated."

TRAINING

The participants were also asked to explain any formal training they had received regarding laboratory animal welfare or the detection of adverse states. There was a general feeling that they had not been assisted in a formal way with the more intricate details of pain and distress in lab animals. "I mean [we] went through animal training, but none of this [animal pain and distress] has ever been pointed out" (Elijah). Although Isabelle stated that she had watched instructional videotapes on injections and handling as a graduate student, most of the training that the participants had been through came from individual laboratories where they had worked. "Didactic training? Well, I didn't really have anything, only hands-on" (Hank). Frederic explained that just recently he had seen more forums advertised within his facility that presented detailed explanations of animal procedures, dealing with distress, and proper care and use. And Hank mentioned the availability of guidelines and policies within his institution "although [they] are bureaucratic, [they are] also kind of helpful."

But all participants agreed that the vast majority of procedural training or learning about the care and use of laboratory animals came from individual laboratories they had previously worked in. "When I first started, regulations were much less evolved and so most of my training I received early on was at an apprenticeship sort of level where someone would teach me how to do things side by side" (Frederic). "Most of my training had been oral learning from the P.I. in the lab or a senior investigator who was able to

describe what to look for and then that was reinforced by a senior technician who had more years of experience (Jacqueline). All of the participants elaborated on how much they had learned simply from their own observations and experience. “You really get to know [colony behavior] just by going into the mouse room everyday” (Hank).

B. Working with pain and distress

CAUSATION

After the participants had been encouraged to think in abstract terms about “pain and distress” in laboratory rats and mice, they were asked to characterize it using examples. Each participant was asked to name and describe what kinds of procedures or experiments could cause pain, distress, anxiety, fear, aggression, and suffering.

The participants organized the procedures they had experience with on a less invasive to more invasive scale. For example, pain was associated with procedures that were relatively quick, without long-lasting effect. Some of these named procedures include injections, tail flick tests, tail cautery, ear punches, and tail snip biopsies. “I also do things like tail-snip biopsies or ear-tagging for genotyping which I would consider to be moderate, short-lived pain. Tails have to be cauterized with a cautery pen, so I would think that those things are painful, but I wouldn’t consider it extreme. That’s the kind of thing that the mouse deals with relatively well and that kind of pain, I would assume, the mouse is pretty much back to baseline after a few hours” (Hank). “I much prefer the tail-flick method...the way I produce pain is by shining a light through a lens on the mouse’s tail and when the mouse feels the pain it flicks its tail and that turns off the light automatically, so that mouse controls its own response” (George).

The participants were more unsure and inconsistent about what kinds of procedures would cause suffering. “Invasive and consecutive surgeries would cause greater pain [than what was previously mentioned]” (Hank). “Surgical procedures I would certainly consider painful, like ovariectomies” (Jacqueline). These statements were compared against the two participants’ previous answers to the kinds of procedures that caused “pain”, but still neither person mentioned suffering, just *greater pain*.

The participants gave more examples for situations that would cause distress or anxiety. “Animals are distressed when they are being closely confined and when they are being acutely, even an injection, will get them to vocalize and respond” (Frederic). “Ear punching is painful, distressing, and causes anxiety. Injecting animals, I would say is painful, they don’t understand why you are coming at them with a needle, but that is the

type of thing, I am comfortable doing without anesthesia or without any type of pain killer, at least subcutaneous injections" (Jacqueline). "Injections, I would call momentary distress, I don't call that pain. Before, when we would do ear-tagging and not sterilize the tags, the animals would get infections which then I would start to put in the category of more moderate distress, I wouldn't call it pain, but it's definitely distressing because you are constantly wiping at it" (George). Throughout his answers, George was more comfortable with naming reactions as distressful rather than painful.

The participants did not limit the states of distress and anxiety of being caused by physical procedures only. "Well, distress is something that we can inflict on animals by certain things we do like restraint, that's very stressful. Drilling through the wall [building construction], loud sounding vibrations, those things cause great distress to the animals, or some kind of conditioned response, or even just that some animals exist better in a cage than others [with less anxiety]" (Isabella). "...on the simplest level, as far as anxiety goes, changing cages can cause anxiety, changing housing situations as far as what animals are together is very anxiety-causing [disrupting pre-existing social structures within the conspecifics]" (Jacqueline). Other explicit examples included: "caused by the separation of mother and babies" (Elijah), "immobilization, for example, putting an animal in something for a long time, that is not pain per se, but it's severe distress to the animal" (Isabella), "some animals just have a nervous disposition..." (Jacqueline), "of course an animal can be distressed if a water bottle opens up in their cage and they don't have anywhere dry to lay, but that wouldn't be painful necessarily" (Jacqueline), "poor housing conditions or bad handling situations" (Jacqueline). David also recognized that inadequate housing and handling situations could cause distress.

ALLEVIATION

Most of the participants were quite active in taking steps to alleviate pain or distress experienced by their research subjects. Frederic, Hank, Isabella, and Jacqueline routinely use analgesics after performing surgical procedures; such as abdominal, thoracic, and central nervous system surgeries. All five participants gave post-operative care beyond what was required by their institution. Some examples include, using a heating pad to keep the recovering animals warm and to facilitate healing, multiple injections of analgesia, and establishing laboratory practices/protocols to ensure their research animals are well cared for. This latter point is important as these practices are not specifically required by their institution, nor by any federal regulation. For example, both Hank and Isabella explained

that they have protocols for secondary and tertiary administrations of anesthetic to ensure the animal is well below a conscious plane during the entire length of surgery. They also both explained that this was a common practice in their laboratory based on having previous experience with animals waking up during surgery. These situations had made them uncomfortable enough to actively do something to prevent its recurrence. Isabella also explained about a separate experiment done by her laboratory that specifically focused on the post-operative pain felt by their research animals. This was not a study done specifically for her research purposes, but rather to learn more about their research animals, so that the laboratory could provide better care. Isabella explained that they had experimented with giving an analgesic before inducing an anesthetic state because prior research had shown that the animals have better recovery time and less post-operative sequellae when this is done. But, her laboratory had problems using the analgesia in conjunction with the particular anesthetic drug that they used since both drugs depressed respiration. Therefore, in a series of experiments with careful record-keeping, they came up with safe doses of each drug in combination, so that they could use both and provide the best care for their animals.

This last example is going above and beyond what is the norm and what is required to alleviate laboratory animal pain. In this case, Isabella felt that the problem of post-operative pain in her animals was an important enough issue to put in significant time and effort to devise a way to alleviate it. She could have easily rationalized that a better analgesic drug/protocol was known, but would simply not work with her particular research. Rather, she made it work.

Among some of the other participants however, there was a fair amount of rationalizing their responsibility in causing pain and distress. It was articulated the most directly by Hank "...it was hard to tell...if the animal was actually suffering, it was distressed, I'm sure. But I don't know what we could do for that. My impression is that there [are] actually limited resources for use to take care of these things. And when you're working with mice, it's just worse. What works in one strain doesn't work in another. It's so difficult. That's why I think it's hard to do this with mice, other than gross observation, it's hard to tell." He also described previous situations he was in that required some rationalization because it was bothersome to him. "I used to be involved in a research project that required removing the cerebellum of a cat and to do that required extensive surgery and would always result in a cat spinning around on its axis for a number of days, so because of that the animal had to be restrained and given fluids. That's probably the

most drastic thing I've been involved in and you just have to kind of forget about all the rules, or definitions of what is pain and what is distress because this is all from what you did to the animal."

All the participants were engaged in some level of pain/distress alleviation, though animal welfare was not the driving impetus in all cases. Everyone, even those that were more concerned with animal welfare, agreed that the animal's state of well-being should be a consideration for the experimental results as well as for welfare concerns. Bad animal care means bad data and therefore a bad experiment. "It seems to be that the animals that are having real problems 24 hours after surgery probably won't be a good subject to use because probably the surgery was botched somehow" (Hank). "If an animal is in too much pain or appears to be suffering they have to be sacrificed because in our study you really can't have one animal going through a different scenario than another and compare them to each other and sacrifice data" (Jacqueline).

ETHICAL DILEMMAS - HYPOTHETICAL

In an effort to analyze the experience of working with pain and distress from different angles, the participants were asked to verbally work through two "real world" scenarios.

Scenario #1: If you were involved in a brain surgery that held the rat's head in a stereotaxic apparatus and near the end of the procedure, the rat starts to struggle, do you give more anesthesia knowing that more drug could very possibly cause overdose to the animal and loss of data and that the procedure only has another five minutes at most until done?

All participants made the decision to give more anesthetic even though the animal (data) could be lost. This may have been too easy a question for the participants to answer in theory. Three of the five participants answered the question quite readily without hesitating to consider what the interviewer thought posed a credible dilemma. "I would definitely give more anesthesia. There is no reason to make an animal suffer like that for five minutes or even one minute. That would just be cruel.\" (George). "I would give more anesthesia because, first of all, I couldn't stand it..." (Isabella). The other two participants stated other considerations, such as "Well, how much is it struggling? Is the animal actually coming awake or is it reflexive?" (Hank) "...I guess it depends on timing. I mean what are you doing to them at that point? Stimulating nerves or just something physical? If an animal twitches when you are doing their wound clipping and they are all stitched up and you are doing something that is just pinching their skin together, I would feel comfortable

letting them wake up. But I don't feel comfortable having an animal feel anything internal..." (Jacqueline).

Scenario #2: The participants were asked if it was better animal welfare practice to use the same group of animals for multiple surgeries, thereby increasing the pain and suffering that each animal endures, or is it better to minimize the pain and suffering that each animal goes through, but therefore use more animals in total?

This question was recognized as a familiar ethical dilemma by all the participants, indicating they had all previously pondered it. Some of the participants articulated other factors that may influence the decision. For example, the particular research goal may require multiple surgeries, as in determining the consequence of a brain lesion on a particular behavior of an animal subject or performing ovariectomies on a rodent to later implant embryos (an increasingly common procedure used to create transgenic animals). When encouraged to make a decision, Frederic, George, and Hank favored more animals thereby undergoing fewer procedures and Isabella and Jacqueline favored using fewer animals and maximizing the use of a smaller number of animals. Some of the explanations for the participants' decisions follow: "It's better to use more rats in total. Because for one thing, I don't think the results would be as reliable with multiple surgeries, aside from the pain factor" (George). "That kind of question, I probably wouldn't think about what is better for the rat, but rather, what is better for the experiment. It depends on the surgery that was done beforehand and it depends on how you want to use them" (Hank). "I guess our tendency is to use fewer rats if we can, but that is very dependent on whether the data will be influenced. But that's very good question, I don't know what the right answer is" (Isabella). "I tend to go by fewer animals, multiple procedures on a single animal than using more and causing more animals to die" (Jacqueline).

ETHICAL DILEMMAS – PERSONAL EXPERIENCE

When specifically asked about the ethics of working with laboratory animal pain and distress, the participants readily shared instances when they had encountered particularly troubling situations. They all brought up these examples without prompting by the interviewer and freely elaborated and responded to follow-up questions. No one was hesitant to talk about their personal feelings about animal research and all recognized the ethical difficultly that sometimes is attached to their work. All the participants gave an example of a time when they were particularly troubled by doing animal research:

(Frederic) "I guess the best example is when I was doing surgery on neonatal rat pups and it required that the usual anesthesia for newborn pups is hypothermia and that is very effective at eliminating any kind of movement or any problems with the animals. However, this particular experiment that I had to do to required that there be anesthesia other than hypothermia and it was difficult to get the animals to a proper surgical plane and not have them die and in the end some of the animals probably were not adequately anesthetized, but the only way to get that data was to carry on with the surgery". This response was particularly interesting to me because this dilemma described by Frederic is very similar to Scenario #1. They both present a situation where the investigator must choose between the welfare of the animal or the experimental results. Frederic chose the animal's welfare in they hypothetical scenario, but here has explained that the experiment was continued even while sacrificing the animal's well-being. Therefore, he has given two conflicting answers on the same issue.

(George) "Well, I remember...we were treating mice for leukemia with some new drugs and they wiped out the leukemia completely and the rest of the body seemed fine until weeks later their eyes just started to push out because there was a tumor growing in their brain. So, in a case like that, what do you do? Do you study it further or just sacrifice the animal? Or I've done a lot of work where the mice end up getting really sick from cocaine, but I had to establish what the cocaine was doing and using different doses and such."

Hank's story about a troubling experiment for him (removal of a cat's cerebellum)) is explained previously.

(*Isabella*) "I think the thing that probably marred me for life was in graduate school when we had labs where we had to decapitate animals and some people were really good at it and some people weren't and we had some really horrible things happening in that lab where people would try to decapitate and only get part of the head and that was just a horrible, horrible thing to see and I think it influenced my life because I would never allow someone who was not experienced to handle an animal in any way. It made me realize how important it is to train people."

(Jacqueline) "Well, one of the things that I've struggled with is, how ethical is it to do surgery when you, the surgeon, are not at your best level of performance. Like if you're tired and that's something that I try to think about at each time and have contemplated."

C. Regulations on laboratory animal pain and distress

Once the participants had shared their personal experiences with research involving animal pain and distress, the control of it was explored. The rest of the questions in the interview asked how the participants felt about the regulations that surround research animals, in theory and in practice.

In general terms regarding governmental control over research institutions, the participants explained their thoughts.

> *(Frederic)* "I don't think [the regulations] are necessarily an effective way of achieving the end. The regulations are such that they can be gotten around if someone was particularly motivated to do so. I think the better way is to insist on some level of training and sensitization to the issues, but the regulations, quite honestly, can be onerous in terms of paperwork and other things that are required."

> *(George)* "[The regulations are a] necessary evil. Necessary because you can't just leave it up to individual people to make up their minds. Working with animals is a kind of privilege that you don't want to abuse. ...but it's gotten to the point where some things are almost ridiculous. Where just to look at a mouse cage you have to put things on your feet and put on a disposable lab coat and gloves, but somebody said its necessary, so I can't fault our own people. You get used to it."

> *(Hank)* "...I don't like filling out my IACUC form, there's just no specific standard way of answering the questions, so when I fill out the forms and try to justify the different procedures that I want to do I have to think of what the most common occurrence is and sometimes its easy, like procedures that I've been doing for 10 or 20 years, but generally you have to fill them out at a time when you are applying for new grants and are doing things that you haven't done before and you just have to kind of wing it. Honestly, it takes me a whole day to fill out my IACUC form and I know the IACUC forms are mandated by the universities, not by the governmental system, at least that's my impression, but they need them to show that animal welfare is something that is actually controlled at the university. ...the IACUC form has to be filled out and there is a lot of cutting and pasting going on and before I became involved in these things, there were things in the IACUC form that had absolutely nothing to do with the project [that was proposed] and the stupid thing was that it really proved a point, it was like as long as you put down something, anything, and you are willing to sign off on it, then it doesn't matter and that kind of thing pisses me off because then why am I wasting my time trying to do it right? I think what you find is that more and more, administration is shoving off what I think is their duty, onto the researcher. I mean they ask how many animals we are going to use in the next three years and if you are going to be honest about it, it takes a long time to figure it out. I always try to err as an overestimate, but those numbers are ultimately filtering back to the Feds and then other people get ahold of them and say things like, 'there have been 3 billion mice killed this year alone in the U.S. by medical research and we have to stop this.' And all these ideas are just based on things that are theoretical, pie-in-the-sky, 'I think I might use this,' you know none of these numbers are real. It kind of forces us to invent things, because there's no other option, you know?"

> *(Isabella)* "I have a problem with governmental oversight when it comes from people that don't work with animals and have impressions that are not necessarily true and so I guess I would like to live in a world where investigators themselves were their own monitor, where they were educated and it wouldn't take legislation to monitor them. But I think the legislation has definitely improved animal facilities

> and just the physical environment the animals are in, but sometimes the legislation goes too far. You need a little bit to make sure that the animals are well cared for, that they live in conditions that provide for them, but I think then you go to the point where you are legislating certain things that cannot be done." (Jacqueline) "I guess the government role in my mind would be to make sure that all the institutions are on the same page that they are all being equally critical about the same things, that they are not being manipulated by someone who has or brings in all the money. I guess that is where it is helpful to have government intervention."

When asked about their knowledge of the Federal regulations for animal research, the participants brought up most of the general requirements, such as; the IACUC, approval of a protocol, protocol renewals, investigative staff training, and an alternative search for the procedures proposed. However, the participants had just as much information that was incorrect. (Hank) "My experience is that the Committee doesn't know a whole lot about science. My impression is that they just believe what we say and although I look [for alternatives], I'm sure other people do not. It's a little bit of a grey area when we want to do something new, a pilot study, whether we should put in a new IACUC form. I don't know if other people do it when they want to do a pilot experiment, I doubt it." (Isabella) "Well, that [the regulations] seem to be a changing target. We try to design our experiments to comply with the laws that we are aware of." (Jacqueline) "I know our protocols have to be approved by an IACUC and reviewed, I think bi-annually. I know there is also some set of standards that are national that people try to follow. But I don't really know anything about the IACUC."

Once their overall knowledge about the regulations was explored, the participants were asked about their specific knowledge of the regulations that surround "pain and distress". The Animal Welfare Act requires that animals be placed within a certain pain category on an animal protocol (Appendix C). All the participants knew the general categorizations about the pain categories that are listed on a protocol form, but were confused about which letter indicated which category. They all knew that the pain categories delineated between pain with and without alleviation (the use of anesthetics) and some also thought there may be different categories for minimal or intense pain.

The participants were then given the correct definitions of each category and asked to classify one of the following four scenarios within a category.

a. <u>induction of cancer in mice throughout a period of three months</u>

(Frederic) "I'd say that's probably **A**"

(George) "I guess it's unavoidable pain because you have to study the growth of the cancer and you can't keep the animal anesthetized all the time, so Category **E**"

(Hank) “If you are injecting cancer cells, I would consider that momentary pain from the injection and then depending on the cancer, I would think that could be maximum pain.” **Undecided**

(Isabella) “Well, three months is a long time for a rat. I don’t know, that’s a really grey area, because you are doing something to an animal at the time is not as bad, but eventually it can become unalleviated pain for the animal, so how does the IACUC look at that? That’s a very hard question. I think it would really depend on the type of cell you are injecting.” **Undecided.**

(Jacqueline) “Well, my first question would be, ‘What would the induction entail?’ If its injection then it would fall under minimal pain, I think, under Category C. However, with that in mind, that doesn’t cover the animal once it finally gets the cancer. We know from human studies that cancer is painful, so while the induction is painless, the actual progression of the cancer could be painful. So, my next question would be, ‘Would it get to a painful stage?’ Or how long is the study’s progression before the animal is removed from it, or sac’d. …so if it were going to a late cancer stage then it would have to fall under the highest category of unalleviated suffering.” **Undecided.**

b. <u>genetic engineering by creating a mouse unable to use its forelimbs</u>

(Frederic) “**A**”

(George) “It would require considerable justification. It can’t indulge in normal behavior.” **E**

(Hank) “I would see distress, but not any pain. I would consider this to be just behavioral then. In the end, if the mouse can’t use its forelimbs, then no analgesia will help.” **C**

(Isabella) “Well, I don’t know if its pain and distress if they were never able to use their forelimbs, they wouldn’t know about it. I don’t know about pain, but definitely stress! “I think I would have to put it in **E**”

(Jacqueline) “It really bothers me that so many transgenic studies don’t seem to think about what the animal is going to feel like when it has all this stuff done to it that is wrong. I would say this falls under the highest category of pain and distress - **E**. And can’t be relieved. While it may not be painful on a daily basis it is going to be emotionally distressing. Should an animal inherently know that it should be able to use its forelimbs? I should think so. This should be pretty tightly regulated.”

c. <u>rats subjected to skin based toxicity tests throughout a 24 hour period</u>

(Frederic) "**D**"

(George) "Well, again, it would require real justification. It can be painful if its reminiscent of the Draize test. It can't just be done lightly. It's unrelieved pain – **E**"

(Hank) "Well, I would think it straddles between momentary pain and just observation (**C or B)**. Then again, depending on what's going on, perhaps a need for analgesia to be administered."

(Isabella) "So, you are talking about injecting animals with compounds that are caustic. That would be **E**."

(Jacqueline) "So, here are the things I would think about…when I've accidentally gotten chemicals on my skin, it can be pretty painful. So what do we know about the substance and its affect on humans? Has the study been done on the top of the skin? I'm uncomfortable when the substance is a cosmetic or something that is not going to be injected into humans anyway. If its going to be injected into humans down the road, then obviously it has to be tested in animals. However, if it's a substance that people are addicted to and that why it has to be studied, then its kind of superfluous. So, if [the animals] are going to die from it and go through pain, then it would fall under a high classification of pain and distress." **Undecided.**

d. <u>isolating infant rats for the extent of their lives (one year), five days after birth</u>

(Frederic) "Umm, I think that's **A**."

(George) "That's more hypothetical suffering because no one really knows enough about it to know how much or whether they really are suffering, you would assume it would be very cruel to human infants, but I don't know…its deprivation without relief. You'd have to have a good justification, you can't just do it out of curiosity." **Undecided.**

(Isabella) "That's pretty cruel. Are their needs being met, like eating and warmth, etc.? I would probably put that in alleviated pain and distress. **D**"

(Jacqueline) "That would be lonely, so it definitely falls under emotionally distressing, whether its physically distressing, probably not, but who knows? My impression is that there aren't any regulations about emotional distress. The guidelines here would probably qualify it as minimal pain, but whether that's right is hard to say because it is definitely going to be emotionally distressing at that age. Once they were weaned maybe it would be

different. I would be concerned and uncomfortable doing the research. I mean these are animals that are used to living in groups and they are social." **Undecided.**

The participants were then asked how *they* think experimental procedures should be monitored. Their suggestions ranged widely and were quite creative.

(Frederic) "The way it works right now, is that folks prepare a protocol and submit it, and then they find that there is some problem with it and they have to go and resubmit it and it can be a multi-step progression to finally get to something that is considered acceptable. I think that's not a particularly efficient way to do it and I think that you could probably get away with a little better efficiency if the IACUC methodology was to actually help the investigator assemble something that is appropriate by having on file what they expect, for example. You would pick modules that you need to help and assemble your protocol and then it wouldn't be a two-day process. And I've never gotten a straight answer as to whether or not that is permissible under the regulatory scheme or if its something that folks have just not attempted to do. I think that the husbandry requirements and that sort of thing are all very reasonable and important and should be in place. I don't' have any question about that. One aspect of the process that I find particularly onerous and I think not very sensible in terms of the regulatory scheme is this requirement to specify numbers. I think it's rare in my work and I think probably in other investigators' work to be able to specify with any degree of certainty really how many animals it would take to complete the experiment. And so if you can't do a good job then why do a job at all? I spend a lot of time trying to figure it all out and then it's irrelevant and they know it's irrelevant and then you are asked to justify why the numbers are inaccurate and you say 'well, the experiment didn't go necessarily the way it was supposed to.' I mean it is an experiment. So that I find silly and it should be eliminated.

One of the other requirements I think is silly is sterile surgery for rodents. Rodents, just invariably, are able to survive surgery under clean conditions without any requirement for sterility or asepsis, so I think that treating a rat like a human on an operating table is not appropriate and its not particularly beneficial to anybody."

(Hank) "Right now, I [am involved in everything]. Either I do with a student [to teach] or with a more senior person to make sure that they knew what they were doing…and that our understanding [was] the same as far as what pain is and if they understood how to deal with particular problems of a mouse colony. At my previous institution, the animal tech taking care of the room would email the P.I. with any problems in a daily report. So, we'd know who is getting watched and what is going on. They don't

do that here. I would be nice if something like that were in place. In the end, the most important thing is training the people and getting the P.I. to understand about training and where it has to be bolstered to align with the standards of each lab. I mean I think I know what is going on with diagnosing pain or other disorders, but maybe there are other things I should be looking for. So maybe there should be some type of advisory that goes out to people relating to this. Something like, 'are you checking for this', or 'do you feel comfortable with that', if you are, then you throw the thing in the garbage, if you are not, then you can contact so-and-so."

(Isabella) "I think the facility can help the investigators a lot by basically teaching the investigators how to do things [procedures] in a better way. I think the facilities would be a really good resource for people trying to do things that they haven't done before or trying to find better ways to do them."

(Jacqueline) "I think we are on the right track by having a review board that oversees the protocols. One place that I was, they wanted to observe you doing the procedures which I thought was not too bad of an idea because someone can write something in the protocol, but how do you know that they are actually going to do it that way or know enough to do it that way? I think at least the first time that you submit a protocol for something that you've never done before that it should be observed. One thing I wish existed was surgery classes. Since some protocols [procedures] are the same wherever you go, like ovariectomies, there should be recommended ways to do the procedure, some type of guidelines, like type of anesthesia, amount, whatever..if those types of things were available, I'm sure most people would be happy to comply."

The last question asked of the participants was to discuss the pros and cons of the following policies and requirements within the laboratory environment.

1. A more thorough classification system of pain and distress utilizing factors such as behavioral and social needs of the animal and the time and intensity of the procedure. Would this classification system be useful? Why or why not?

(Isabella) "I think no matter what you do there is no way to really make lines around some of these issues, no matter how well you describe them. I think they would still be very hard to define within a specific category. The only thing that would help is to make the investigator more aware of what it is that he/she is doing. But I don't think in the long run it would be helpful defining where something belongs in a category."

(Jacqueline) "It sounds like nightmare! Certainly there would be advantages to a behavioral component. I mean I guess the time and intensity would be the most easy to

quantify, but how would you classify social and behavioral variances? Anything that gets more info about the protocol is better, you know anything that gets the investigator thinking about those issues is good, but I think its gets very difficult to regulate. But for information gathering, it would be helpful."

2. encouraging more observation and subsequent documentation of pain and distress to assure adequate reporting

(Frederic) "A pro would be that there is some level of new information that the investigator could get from that, but again I think it would be a regulatory burden."

(George) "That would be good, except that these observations would either be very subjective or just performed as a checklist. I don't know how much they would mean to other scientists."

(Hank) "I don't mind that and in some degree, I actually do that, so I guess it depends on the level of bureaucracy that comes with it. I don't like the idea of bureaucracy, I think that it has to be for our own use. Let's say it somehow got out that all my animals were abused or whatever, I would think that my notes wouldn't reflect that anyhow. If I'm a secret mouse torturer, I'm not going to write it in my notebook."

(Jacqueline) "Again, documentation is only as good as someone making sure that its done."

3. requiring the IACUC to validate the categorization of experiments through periodic/random IACUC review of activities and outcomes

(Frederic) "The pros is that you would use this as kind of an oversight function. The con is that I don't think it is necessary."

(George) "I think that would be great because they would have some actual experience instead of sitting around a table and discussing these things at nauseating length."

(Hank) "The hesitation that I have about the whole thing is who would be looking at the data? ..at some point you have to trust the person to do things right or you don't give them grant money."

(Jacqueline) "The pro would be that you would definitely catch people who are lying about their protocol. The con would be that it would diminish the sense of community trust."

As a wrap-up, the participants were encouraged to summarize their main concerns and suggest different methods or changes to the present regulations or institutional practices concerning laboratory animal pain and distress.

> *(Frederic)* "I think the crucial thing is to have the dissemination of information and I think that then becomes less of a regulatory scheme and more of a willingness on the part of the staff of the IACUC."

> *(George)* "Well, I don't think there is enough information about the long-term effects of transient pain and distress. That is something at least if people had in mind they would be more aware of."

> *(Hank)* "To me, regulation means that somehow you've solved the problem and all you can do is trust that people are doing what you are trying to regulate. I think that, in the end, regulations don't stop anything. I think there should be guidelines, definitely. Because people need to know what is best, people need to have some sort of framework in which to think about things when they are first starting out. I mean how can you regulate pain and distress? How many times do people have these situations, like the rat in the stereotaxic apparatus that wakes up, and then they just go ahead and finish the experiment and it doesn't get written anywhere, so that is something that is against regulations, but the whole thing is gone so quickly. And how is anyone going to find out? Regulations are only good for things you can actually test."

> *(Jacqueline)* "Well, we should make protocols more specific about why the research is being done, how long the animal is going to be used in the conditions, how much distress do you think the animal is going through, for how long, is it going to be emotionally distressed, is it stressed through other ways? The first thing would be to get more info out there. But it bothers me when the regulations are made by people who have no idea."

IV. Conclusion

Although this study cannot be extrapolated beyond the participants, it is useful in that in brings to light some important points and concerns regarding how pain and distress, and the control of it, is viewed in laboratories. These points will become increasingly important since the focus on this particular area of the regulations promises to intensify as the regulations become more inclusive and specified and research in the fields of neurobiology and animal cognition become more sophisticated.

Although the sample size is small, some trends are suggested from the responses of the participants. There are differences in how comfortable the scientist is with his/her research being regulated. The participants that were the most senior (Frederic, George) spoke often of "before the regulations" meaning before the late 80s when they were affected by the 1985 laws. Based on the frequency with which this time period was

discussed, suggests that there was a memorable change in how their animal research was done, or at least the amount of paperwork they were responsible for. But the other Principal Investigators (Corinne, Hank, Isabella) that had begun their schooling around the time of these regulations had no such comparison to make and instead focused on more of the specifics of the regulations that affected them. In other words, it is suggested that the act of being regulated was taken for granted by these younger investigators and they were able then to focus on the specifics of *how* they were regulated and ways in which the entire oversight process could be improved. And lastly, the youngest investigators (Adam, Ben, David, Elijah, and Jacqueline) were the most affected by how their animals responded to the procedures and had the least to say regarding the regulations. It should be added however, that the youngest investigators also have the least direct experience with the realities of the regulations such as the paperwork and policies involved, so this also should be taken into account.

I also found suggested differences in the participants' field of study. For example, Isabella and Jacqueline were the most in tune with ways in which their animals' emotional/mental states may be affected by research. Both the these researchers had a background in animal behavior and their present research was in neuroendocrinology, therefore they needed to be quite aware of how the animal's stress level was affected by outside influences in order to produce quality research. Likewise, Hank was the participant that spoke the most about the social environment of the animals. His research depends on his control of breeding colonies and therefore he has direct experience with the potential health care concerns and distressors that are caused by social interactions. None of the participants used the animals for tissue harvest only, all ten were involved in survival surgery of their animals, so they all needed to be concerned with how the animal was affected by the procedures that they performed. However, it would be interesting to analyze the responses of investigators that use animals only for tissue harvest. Are they less aware of the animals' ability to experience pain and distress since they do not need to be cognizant of these states for the success of their research? This possibility was even suggested by one of the participants – Isabella.

Experience is not standardized among scientists, so therefore compliance and adequate animal care will not be either. Differences among researchers and their specific fields of expertise should be taken into account by the institution. Some investigators may need more help in providing proper animal care. This may simply require more education and procedure training, or it may require verification of pain and distress categories stated

within their protocol, or assistance in proper post-operative monitoring, etc. These are ways in which the institution can become more involved and provide help to investigators to maintain compliance. In this way, the institution provides solutions rather than providing only obstacles to the investigator. In a similar manner, the expertise of the investigators can be utilized to assist the institution with maintaining compliance. Interestingly, it was shown that although the investigators did not believe that they knew much about the issues of animal care and welfare, in fact, they were an exceptional source of this information. Not only are some of them very highly qualified in their particular knowledge of surgical or non-surgical procedures and procedural effects, but also carry with them a wealth of information regarding indications of animal pain and distress. Much of the indications of animal pain and/or distress that the participants utilize every day in the laboratory would contribute to animal welfare in substantial ways if used in to help create the legislation that research is subjected to. This is a strong argument for involving the scientists when developing the guidelines and regulations that are involved in proper animal care in the laboratory.

The aim of this study was to highlight important concerns from the view of the biomedical scientist. Through their own words, they have provided provocative analyses and have openly discussed their own beliefs regarding the subject matter. More specifically, the participants articulated how the regulations affect them as scientists and explained how they perceive the issue of laboratory animal welfare and its proponents. This finding is extremely important because, as shown, the individual scientist is the person most responsible for the care of his/her own research animals and therefore he/she must understand the importance of the regulations and proper care. It only hurts the animal if the ones responsible for their care feel threatened by the regulations because in the end this affects the level of compliance. If closer attention was paid to the scientists when developing these regulations, better procedural oversight and more uniform compliance could be achieved. This report also suggests that another significant obstacle to achieving compliance has been created by the very groups that push for increased humanity for animals. Because the opposition has been very aggressive and combative instead of collaborative they have in many ways sabotaged their own cause. Ironically, the desire to "liberate" animals makes improving their care more difficult.

It is the hope of the author that in the future these issues will be discussed in more detail and that those people who are most affected by research animal pain and distress will be at the center of any active solution.

V. Recommendations for Improving Animal Care Standards:

1. *How to write a protocol*

As was elucidated by the participants, they often are confused about what is expected of them in terms of the welfare regulations. Each investigator is required to write an animal protocol that details the experimental procedure that the animals will undergo. The participants view the protocol writing process as a useless regulative busywork. Institutions can assist their investigators by making this process easier. This was suggested by Frederic, "and I think that you could probably get away with a little better efficiency if the IACUC methodology was to actually help the investigator assemble something that is appropriate by having on file what they expect, for example. You would pick modules that you need to help and assemble your protocol and then it wouldn't be a two-day process." There is no standard requirement for how to design a protocol as long as specific information is given, like a lay description, the alternative search, pain and distress categorizations, and number of animals. Therefore, institutions have the flexibility to create their own protocols. Investigators would need to do less paperwork if the protocols were more directed toward their individual research. For example, a brief questionnaire can be used for a basic overview that includes the specifics that are listed are included in all protocols. Beyond that, the investigator then could choose between different modules based on the individual procedures and care that is needed for his/her own experiments. These modules could be separated into ones for surgery, breeding, behavioral testing, genetic engineering, ascites production, monoclonal/polyclonal antibody production, etc. They were explain what kinds of details about each procedure the Committee will need to examine and, when appropriate, give specific examples or templates for the investigator to use.

This would diminish the paperwork that the investigators are responsible for and because the institution (through the IACUC) is taking on a role in the regulative paperwork requirements, the administration is showing support for the investigators by attempting to take on some of the paperwork.

The investigator can also be assisted during this process if the IACUC is clear about exactly what is expected in the protocol. For example, often investigators do not justify their requested number of animals, list enough detail about the surgery, or provide enough information about how the procedure affects the health of the animals. Yet often this occurs simply because the investigators do not know to put more details into

the protocol or what exactly the IACUC will be analyzing. If the IACUC provide the investigator with an instructional pamphlet that explains these key points, lists common problems of protocols, and includes examples of approved protocol sections, then the guesswork would be taken out of the process and the time put into writing the protocol would diminish.

2. *Provide lists of procedures per pain and distress categorization*

Often, the investigator does not fully understand the USDA categorization for pain and distress. To confound this problem, institutions sometime define them differently from other institutions. Therefore, the categorization of some procedures may be somewhat subjective. This is indicated from the wide ranges given by the participants for USDA categorization when questioned about the hypothetical research protocols. Not only does this suggest that not everyone is equally educated about animal well-being, but also does not take into account the same considerations before, during and after the procedure. The USDA categorization is an attempt by the government to provide standardization between experimental procedures, but if the same procedure can be categorized under either A or E by five experienced researchers, this shows that this kind of disparity is probably common between research institutions. Therefore, it may be useful to list procedures that would be classified under each category. This would not be possible in all cases, but may be possible for some standard procedures. To help in this effort, IACUCs should provide their investigators with a list of which procedures are to be included in which category. Even if the investigator includes something in their protocol that is not on the general list, it would at least give them a starting point that helps them to understand that particular IACUC's classification system.

3. *Provide Standard Operating Procedures (SOPs) for drugs and procedures*

As mentioned previously, often the investigator does not include as much detail as is required in their protocol. If the IACUC were to provide standard operating procedures, such as common surgeries like ovariectomies, thoracotomies, or implantations, then the investigator could simply follow this format and be directed for what to include in their own protocol. Likewise, SOPs could be provided for use of anesthetic or analgesic drugs, monitoring during surgery, post-procedural care, and methods of euthanasia. This would also serve the overall purpose of increased and *standardized* animal care because since the SOPs are provided by the IACUC and/or veterinary staff, they would explain the optimal

methodology and could therefore take the place of out-of-date methods used in laboratories. This is another way that guesswork is taken out of the animal care part of the research. The veterinary staff and IACUC are taking on more responsibility in the planning of the research, but at the same time helping the investigator by providing useful information that ordinarily would not be the focus of the investigator.

4. *Provide objective measures of pain and distress*

This can be done on a national and institutional level. Nationally, guidelines from the American Veterinary Medical Association could be provided in general ways. For example, just like with the list of procedures per USDA pain and distress category, the IACUC can expand of this list by also providing information on the kind of pain that results, how to detect it, and how to alleviate it. These guidelines would explain the differences regarding major and minor surgeries (disruption of body cavity, physiology, or orthopedic structure) from other kinds of surgery, such as subcutaneous implants or venipuncture cut-downs. Information regarding characteristics of that particular procedure or characteristics of the species would also be useful. For example, surgeries of the eye and periorbital structures are painful to most animals, as are involving the ears, nose, and teeth. Orthopedic procedures are generally painful because of trauma to large muscle masses. Amputation, especially high on the limb, and thoracotomy induce severe pain. Surgeries involving the cervical vertebrae are usually more painful than are procedures involving the thoracic or lumbar vertebrae. Perirectal procedures are generally painful and animals can be seen rubbing and scooting on their perineum in response to pain. Abdominal pain may be difficult to detect or the animal may appear to hunch its back and tuck in its abdomen. [32,48]

Investigative staff may benefit from explanations of the necessary observations to make on the recovering animals. For example, on awakening from anesthesia, animals may shake their heads excessively, rub or paw at the painful area, and vocalize. Changes in eating, drinking, excrement, socializing, or grooming (pilo-erection) are also important to note. Investigators who work with rodents should also know the important characteristics of their animal that would affect their behavior, such as the "prey behavior" of hiding their pain. These animals may not vocalize very readily, but there are other specifics to be aware of that can signify pain.

These are all ways to involve the investigators in the humane care of their research animals, by involving them in the process and educating them about how the animals are affected from their specific procedures.

5. *Require documentation of procedures that may cause pain and distress*

It was hard for everyone to differentiate between pain and distress. The participants were more confident discussing examples of how the states would be caused. This can be put to use within the requirements of the animal protocol. This would be an ideal format in which to include further documentation and requirements about pain and distress potential for each procedure. For example, an institution can require that post-procedural care of experimental animals include objective measures of pain indication; such as post-operative records that note biological and behavioral characteristics. It would also be helpful to require statements in the animal protocol that address these objective measures. For example, the investigator could be required to explain what can be expected from observing the animal after (or during as with tumor growth) a procedure and/or what the investigative staff will look for during post-procedural monitoring; such as, inactivity, pilo-erection, change in feeding, drinking, or excreting, change in social behavior, hunched posture, etc. To this end, it would also be beneficial if the investigator were required to directly explain the kind or level of pain and distress that is expected from the proposed procedure. This can be done in the previous suggestion about detailing the post-procedural care or it can be done through a more thorough explanation of USDA animal categorization (see Question #11). In this way, the investigators are able to use their prior experience and are given the opportunity to contribute what they know about pain and distress with their particular experimental procedures. This information is often not recorded or shared among scientists as it is not usually part of the direct research results that will be published. However, it can be of significant value not only for the requirement of reporting and minimizing pain and distress, but also for further refinement of the experimental procedures. Also, these explanations within the animal protocol provide a record for what the investigative staff will be watching for in their animals and gives a clear expected endpoint for the animal. The IACUC can also use this information to monitor the animal's potential pain and distress and require earlier endpoints or proper alleviation through the use of analgesics, if not originally used.

5. *Provide institutional polices or guidelines that address the psychological well-being of research animals*

A cognitive component of animals was recognized by all of the participants. Within the Federal regulations are requirements for addressing the "psychological well-being of

primates and dogs", but the results of this study indicate that most investigators believe there is a psychological component to most research animals, not just dogs and primates. Therefore, institutional policies that account and provide for the psychological health of the research animals may be readily accepted. Further policies or guidelines could be implemented to provide for these needs in research animals. There is an increasing amount of research that focus on these issues in research animals. Some of the many solutions that have been explored include: toys and videotapes for primates, dogs, pigs, sheep, and goats. Environmental enrichment activities for rodents, including balls, nestlets, nuts and bolts, tunnels, wheels, etc. Policies could exist not only for the enrichment of individual animals, but also for encouraging proper socialization among animals, such as limits for seclusion, visual and aural awareness of each other, proper weaning lengths, proper cage populations, etc.

Also, most of the participants talked of situations that would cause distress among their research animals that were not related to a certain experimental procedure. This is additional information that can be used to create guidelines that address the psychological health of the animals. For example, there should be directions on proper handling, living conditions, and awareness about the effects of changes in the animals' environment. The institution could provide policies to educate the investigative staff about these issues, which would also serve to improve the facility conditions.

Although all of the above-mentioned examples would be good starting points for how to address these kinds of concerns, it is important that the administration of the institution, most notably the IACUC, also proceed cautiously with new policies that affect the animals. Even if new policies are implemented to increase the standards of care for the research animals, it is also important to protect the particular research of each investigator and providing for the psychological well-being of animals also introduces more variables into the research which could adversely affect the results. By proceeding cautiously, the IACUC is validating the importance of the research and showing the animal welfare does not have to come at the cost of sacrificing good scientific practice.

6. *Provide more educational opportunities regarding research animal health and well-being.*

Generally, the participants seemed to believe that training is important because they had concerns about the abilities of other investigators to recognize pain and distress in their animals. They also made points about the difference between investigators either on a field-

specific, age, or cultural level. Throughout the interview everyone had additional questions about pain and distress, the recognition of it, and other abstract parts of the issue, indicating that they would be interested in additional or more focused training on these issues. Frederic and Hank even directly suggested that informational pamphlets be made available to investigative staff on a regular basis. And Isabella and Jacqueline both talked of the idea of hands-on instruction for particular procedures from the veterinary staff. I think that it is significant that my respondents were the ones to suggest these ideas. It suggests that there is a real need and that investigators would be open to participating. It should be the responsibility of the institution to provide adequate training of procedures, surgeries, handling, behavior, breeding, etc. Every institution is mandated to provide training for their investigative staff, but again this is all that is stated, so that "training" could mean a variety of things. But, this report suggests that quite extensive training is needed and would be accepted readily. Along with hands-on instruction, they should also be provided with resources of topical interests from journal articles or online resources. The laboratory animal medicine division should maintain a library that is accessible to the investigative staff. Also, topical seminar, training workshops, protocol instruction sessions are also important to provide on a regular basis.

7. *Involve investigators in the training on a broad scale. Use their line of research, or past experience and expertise with specific procedures*

If the investigative staff is included in the goal of high standards of animal care, they will be more willing to participate on an individual scale. Their own research and knowledge and experience of certain procedures should be utilized. For example, if a particular lab is very good at retro-orbital blood collection, can a representative give an instructional session open to other labs on their methodology? This helps everyone and also builds up the collegial atmosphere, with the inclusion of the IACUC and animal care division.

8. *More research on pain and distress needed and more dissemination of existing information on a national scale.*

An increased amount of scientific data on these issues can only provide cleaner guidelines on the recognition and subsequent alleviation of them. George, in fact, directly stated that more data was needed. Although the laboratory animal division can be involved with research that focuses on laboratory animal care, there is not much that can be done on

an institutional level. However, the IACUC or other divisions within the institution can help to disseminate the information. As with the suggestion in #6, the investigative staff can be kept aware and educated about the latest refinements and issue with laboratory animal care and welfare.

9. *The messenger is important, utilize an already respected entity to encourage increased welfare standards*

The older participants noted the improvement in the animal facilities since the implementation of the Federal regulations. For example, the regulations have provided guidelines to all research institutions regarding housing conditions, husbandry standards, and adequate veterinary care. This, in turn, creates more standardized conditions in animal facilities across the country and therefore better and cleaner animal use data. And this was recognized by some of the participants. The animal facility personnel of the laboratory animal medicine unit of each institution may be useful in providing direction to the investigative staff on increased standards of care within each laboratory. The discussion of training from the participants also supports this theory, as they readily suggested an increase in training.

It was also directly stated or implied by the participants that the identity behind regulations, policies, and guidelines is important to them. This fact further supports the idea of the *institution itself,* by way of the laboratory animal medicine personnel, the veterinarian, the IACUC, the veterinary technicians, and animal caretakers should be given the responsibility to ensure adequate animal well-being. These participants have indicated that this message may be better received from these messengers, rather than outside groups with increased governmental regulations. However, it is important to analyze each research institution individually since the animal care staff may not, in all situations, be the best group to encourage increased standards of welfare among the research scientists. Depending on the characteristics of the particular culture of each institution, different groups may be better than others to exemplify high standards of care and/or monitor each laboratory for regulatory compliance.

10. *Validate investigators' concerns and knowledge of their own research.*

Respect and trust is a difficult feeling to maintain between the IACUC and investigators if there is a constant requirement for documentation and follow-up verification of the protocol, but an atmosphere of collaboration can be supported if their

interests and concerns about the way they are regulated by the institution is given a platform and taken seriously. For example, before the IACUC approves a new policy, they could have a comment period for the investigative staff to read it and relay their thoughts or concerns. They may contribute many important considerations that the IACUC should take into account. Therefore, IACUC policies and procedures for regulating compliance is done in concert with the investigators. This may help the overall state of compliance because the investigators were part of the solution rather than something new and foreign being imposed upon them.

VI. References

[1]Aldhous, P, Cochlan, A. & Copley, J. (1999). Let the people speak. New Scientist 22, 26-31.

[2]Animal Welfare Act of 1966. (Public Law 89-544), as amended in 1970 (Public Law (91-79), in 1976 (Public Law 94-279), in 1985 (Public Law 99-198).

[3]Benison, S. (1970). In defense of medical research. *Harv Med Alumni Bull 44,* 3:16

[4]Bentham, J. (1962). *The Works of Jeremy Bentham, Volume 1,* pp 142-143, ed. J. Bowring. New York: Russell and Russell.

[5]Brinton, C. (1957). The Scientific Revolution, 1500-1800 – The Formation of the Modern Scientific Attitude. *American Historical Review 62 (4)*: 889-890.

[6]Carson, Gerald (2002). Leaders of the movement. In Kelly Wand (ed.) The animal rights movement (American social movements).Greenhaven Press.

[7]Catlett, Stephen J. (1984). Walter B. Cannon Papers (1905-1928) in *A Guide to Manuscript Sources in the History of Medicine in the American Philosophical Library*. Philadelphia, PA

[8]Center for Alternatives to Animal Testing, The Johns Hopkins University website: http://caat.jhsph.edu/

[9]Dawkins, MS (1980). *Animal suffering: The science of animal welfare.* New York: Methuen, Inc.

[10]Dodds, W.J. & Orlans, F. B.(eds.) (1982). Scientific Perspectives on Animal Welfare. New York: Academic Press

[11]Dresser, R. (1989). Developing standards in animal research review. Journal of the American Veterinary Medical Association, 194(9):1184-1191

[12]Erbe, NA. (1966). *Anti-cruelty laws and scientific use of animals in the United States.* London: Research Defense Society, pp 1-12.

[13]Federation of American Scientists (1977). Animal rights. *Pub Interest Rep 30,* 8: 1-8, as quoted in Eskridge NK (1978).

[14]Finsen, Lawrence and Susan. (2002). The ALF and the "Unnecessary Fuss" video. In Kelly Wand (ed.) The animal rights movement (American social movements). Greenhaven Press.

[15]Fox, JL. (1984). Lab break-in stirs animal welfare debate. *Science 224:* 1319-1320.

[16]Fox, MW. (1984). Reconciling humane concerns and laboratory animal research. *N Eng J Med 310,* 5:325-326.

[17]Free, Ann Cottrell (September 12, 1996). "Creating the Climate: A Journalist's Role and Observations on Events Leading to Passage of the Laboratory Animal Welfare Act." Animal Welfare Act: Historical Perspectives and Future Directions Symposium Proceedings, Riverdale, Maryland. WARDS, 1998.

[18]French, RD. (1975). Animal experimentation and humanitarian sentiment before 1870. In: *Antivivisection and medical science in Victorian society.* Princeton: Princeton University Press, pp 16-17.

[19]Goodwin, Frederick & Morrison, Adrian. (2000). Science and self-doubt. Why animal researchers must remember that human beings are special. *Cerebrum: The Dana Forum on brain science.*

[20]*Guide for laboratory animal facilities and care.* Department of Health, Education, and Welfare, Publication 1024 (1963).

[21]Hart, L. (Ed.) (1998). Moving toward a less-troubled middle ground. In Responsible conduct with animals in research (pp. 3-17). New York: Oxford University Press.

[22]PHS (Public Health Service). 1996. Public Health Service Policy on Humane Care and Use of Laboratory Animals. Washington. D.C.: U.S. Department of Health and Human Services, 28 pp. [PL 99-158. Health Research Extension Act. 1985]

[23]Holden, C. (1981). Scientist convicted for monkey neglect. *Science 214*: 1218-1220.

[24]Holden, C. (2000). Researchers pained by effort to define distress precisely. *Science 290:* 1474-1475.

[25]Institutional Animal Care and Use Committee Guidebook. ARENA/NIH/OLAW. 2002. NIH. Pub. (IACUC duties, special considerations, federal regulations, references, and resources.

[26]IRAC (Interagency Research Animal Committee). 1985. U.S. Government Principles for Utilization and Care of Vertebrate Animals Used in Testing, Research, and Training. Federal Register, May 20.1985. Washington. D.C.: Office of Science and Technology Policy.

[27]Jacobs, FS. (1984). A perspective on animal rights and domestic animals. *J Am Vet Med Assoc 184,* 11:1344-1345.

[28]Jasper, James M. (2002). Postwar organizations and reforms. In Kelly Wand (ed.) The animal rights movement (American social movements).Greenhaven Press.

[29]McPherson, CW & Mattingly, SF. (eds.)(1999). 50 years of laboratory animal science (1950-2000). American Association of Laboratory Animal Science (AALAS). Memphis, TN:AALAS Foundation Press.

[30]Moberg, GP (ed.) Animal stress. "Regulations and guidelines for animal care. Problems and future concerns." *American Physiological Society.* P. 281-296.

[31]Morrison, Adrian. (2002). Understanding the effect of animal-rights activism on biomedical research. *Actas de Fisiologia, 8:* pp 9-22.

[32]National Research Council, (1992). Recognition and alleviation of pain and distress in laboratory animals. Institute of Laboratory Animal Resources. Washington, D.C.: National Academy of Sciences Press.

[33]National Research Council. (1996). *The guide for the care and use of laboratory animals*. Institute of Laboratory Animal Resources. Washington, D.C.: National Academy of Sciences Press.

[34]New directives for Animal Welfare Act. (1975). *J Am Vet Med Assoc 167, 4*:260.

[35]Pacheco, Alex (2002). The Silver Springs Monkeys Case. In Kelly Wand (ed.) The animal rights movement (American social movements).Greenhaven Press.

[36]Paul, Elizabeth S. (1995). Us and Them: Scientists' and animal rights campaigners' views of the animal experimentation debate. *Society and Animals, Vol. 3, No. 1.*

[37]Phillips, Mary T. & Sechzer, Jeri A. (1989). Animal research and ethical conflict. An analysis of the scientific literature: 1966-1986. New York: Springer-Verlag.

[38]Phillips, M. T. (1993). Savages, drunks, and lab animals: The researcher's perception of pain. Society and Animals 1:61-81.

[39]Plous, S. & Herzog, H. (2001). Reliability of protocol reviews for animal research. Science, 293(5530):608-9.

[40]Rowan, A. (1984). *Of mice, models, and men: A critical evaluation of animal research.* Albany, NY: State University of New York Press.

[41]Rowan A. (1998). The search for animal-being. In Lynette Hart, (Ed.) Responsible Conduct with Animals in Research. (pp.119-131). New York: Oxford University Press.

[42]Rowan, Andrew N. & Loew, Franklin, M. (1995). The animal research controversy: Protest, process, and public policy. An analysis of strategic issues. Center for Animals and Public Policy, Tufts University School of Veterinary Medicine.

[43]Russell, W.M.S. & Burch, R.L. (1959). The principles of humane experimental technique. Methuen:London

[44]Samuels, WM. (1984). Public affairs. Reporters and researchers both to blame in telling the real story. *Physiology 27,* 6:403-404.

[45]Sechzer, JA. (ed.) (1983). *The role of animals in biomedical research.* Annals of the New York Academy of Sciences, vol. 406. New York: The New York Academy of Sciences, pp 5-12.

[46]Singer, Peter. (2002). Henry Spira and the Draize Campaign. In Kelly Wand (ed.) The animal rights movement (American social movements).Greenhaven Press.

[47]United States Department of Agriculture website, Animal Care History, at http://www.aphis.usda.gov/ac/awahistory.html

[48]Use of Laboratory Animals in Biomedical and Behavioral Research. National Research Council and Institute of Medicine, Committee on the Use of Laboratory Animals in Biomedical and Behavioral Research. 1988. Washington, D.C.: National Academy Press.

[49] World Medical Association Statement on Animal Use in Biomedical Research. Adopted by the 41st World Medical Assembly. Hong Kong, September 1989.

VII. Appendix A

Knowledge base about pain and distress issues

1. Do you distinguish between the terms: Pain, suffering, distress, anxiety, aggression, fear, as seen in laboratory animals?
2. How comfortable are you at detecting these states?
3. How would you rate the ability of your colleagues to make these distinctions?
4. Describe any training you have had about working with laboratory animals?

Working with pain and distress

5. Describe what kinds of procedures would cause each of the conditions listed above.
6. Describe any steps you, or your laboratory, take to minimize pain and distress if observed in an animal.
7. What would you do if involved in the following scenarios:
    a. During a brain surgery, the rat regains consciousness and starts struggling. Knowing that 1) giving more anesthesia could very probably cause death (loss of data) 2) finishing the procedure would only take another five minutes approximately 3) the rat is in a stereotaxic device so finishing the procedure despite the struggling is possible – do you give more anesthesia or do you quickly finish the experiment?
    b. Is it better to use the same group of rats for multiple surgeries, thereby increasing the pain and suffering that each rat individually endures and then euthanize or is it better to minimize the pain and suffering that each rat goes through, but therefore use more rats in total?

8. Please give an example of a time when you encountered a difficult ethical decision or a time when you were particularly uncomfortable.

Regulations on laboratory animal pain and distress

9. How do you feel about governmental oversight of laboratory animal use?

10. Describe what you know about the regulations that protect laboratory animals? Include also, the process that each laboratory must go through to ensure compliance with these regulations.
11. Do you know about the five pain categories in an IACUC form?
12. How would you classify the following according to an appropriate pain category:
    a. induction of cancer in mice throughout a period of three months
    b. genetic engineering by creating a mouse unable to use its forelimbs
    c. rats subjected to skin based toxicity tests throughout a 24 hr. period
    d. isolating baby rats for the extent of their lives (1 year) five days after birth
13. How should experimental procedures be monitored and carried out? Considerations: skill level of researcher, number of animals involved, importance of procedure, P&D classification of procedure, etc.
14. Discuss the possible pros and cons of the following:
    a. encouraging more observation and subsequent documentation of pain and distress to assure adequate reporting.
    b. requiring the IACUC to validate the categorization of experiments through periodic/random IACUC review of activities and outcomes
    c. more intensive training and restrictions on who conducts the procedures

15. What are your suggestions concerning the regulation of pain and distress of laboratory animals? What do you think are the most pertinent factors to keep in mind?

VII. Appendix B July , 2002

Dear

We are writing to ask you to participate in a research study entitled "The Recognition of Pain and Distress in Laboratory Rats and Mice by Biomedical Researchers". This study is part of a final project conducted by a graduate student in Animals and Public Policy at Tufts University School of Veterinary Medicine.

The impetus for this project has come from research that shows that the government regulations on the use of laboratory animals are often difficult to follow in the laboratory for a variety of reasons. In particular, the legislation is most often introduced and organized by proponents of animal welfare that come from outside the research community. Therefore, too often the regulations fail to incorporate the needs and goals of the researchers themselves. In general, the regulations, as written, are difficult to enact because the details of laboratory animal use are not well represented from the viewpoint of the scientist.

A particular area of difficulty is the monitoring of "pain and distress" in laboratory animals. Specifically, the Animal Welfare Act and the Public Health Service Policy outline requirements for the alleviation of "pain and distress" in laboratory animals. Yet, the definition of these states is very subjective and thus creates a wide range of variability among scientists and institutions as to how these states are recognized and alleviated for different species. The aim of this study is to document the researchers' first-hand experience of recognizing and working with these conditions in rats and mice. As a result, the most pertinent factors that surround these issues can be identified by representatives of this community.

As a participant, you will be asked fifteen questions in an interview format. Interviews will take approximately one hour. Interview questions are general and do not require specific information about the laboratory's research or personnel. The identity of the participants and the details of the laboratory's research will not be disclosed. To ensure complete confidentiality of each respondent's answers, all biographical details and institutional affiliations that could possibly link an individual to his/her statements will be omitted or altered. No one other than the student investigator will know the actual identities of participants. Furthermore, all records linking individual respondents and the interview transcripts will be destroyed at the conclusion of this project.

Thank you for considering participation in this study. It is our hope that you will take this opportunity to contribute your views and concerns as a scientist. In the continuing debate about how to properly regulate the use of laboratory animals, it is very important to include the perceptions of those who are directly affected by governmental regulation.

In the near future, you will be contacted by phone by the student investigator of this study, Valerie Parkison. If you have any further questions, please feel free to call or email.

Phone number: *617.596.4970* Email: *vparki01@tufts.edu*

Thank you for your time,

Valerie Parkison, BS, MS candidate
Center for Animals and Public Policy, Tufts University School of Veterinary Medicine

Allen Rutberg, PhD, advisor
Center for Animals and Public Policy, Tufts University School of Veterinary Medicine

Jan Dizard, PhD, advisor
Department of Anthropology and Sociology, Amherst College

IX. Appendix C

*** Pain and Distress Categories (USDA, 2002)[47]**

Category	Description
(A)	Behavioral observation *
1 (B)	Little or no discomfort or stress.
2 (C)	Minor stress or pain of short duration.
3 (D)	Moderate to severe distress.
4 (E)	Severe pain near, at or above the pain tolerance threshold. **Requires annual institutional USDA report.**

* this category was added by the author

Appendix D

Interview Questions

1. Please describe your field of study and your lab's specific focus.
2. *Please describe the use of animals in your laboratory, invasive and otherwise.*
3. Please describe your responsibility, as a scientist, to the welfare of the animals under your control.
4. When you read a journal publication, 1) do you understand the experimental situation of the animals involved and 2) in which category of distress you would classify it?
5. How do you compare and rate the terms: Pain, suffering, distress, anxiety, aggression, fear
6. How would you rate your ability (and separately, those of your lab coworkers) to detect the previously listed states in the animal before, during, or after the experiment?
7. Please give examples of what you would classify as an "alternative" in the broad realm of animal testing?
8. What alternatives to the direct use of animals, such as procedures, methodology, or equipment are you aware of in your specific field and in others?
9. What do you recognize as the pros and cons of alternative use in your field of study?
10. How do you feel about government regulations on research that push the use of alternatives? For example: rats, mice, and birds controversy/ ICCVAM regulations/ cloning controversy.
11. How representative are your beliefs/views of others in your field? In biomedical research as a whole?
12. Do you believe any regulations that derive from the Animal Welfare Act should be updated, removed, or otherwise modified?
13. What circumstances, if any, do you believe would lead to/promote greater use and acceptance for animal alternatives by people in your field?
14. What are your beliefs (or concerns, interests, questions) about the current state of laboratory animal use?
15. What are your beliefs (or concerns, interest, questions) about the future state of laboratory animal use?

***** *italicized question was not allowed per laboratory manager*

Appendix E

Center for Animals and Public Policy
Tufts University School of Veterinary Medicine

"The Recognition of Pain and Distress of Rats and Mice by Biomedical Scientists"

Allen T. Rutberg, PhD
Valerie Parkison
Jan E. Dizard, PhD

PURPOSE OF STUDY

The aim of this study is to document biomedical researchers' first-hand experience of recognizing and working with pain and distress in laboratory rats and mice. Government regulations require researchers to recognize, assess and report pain and distress in their laboratory animals, however these states are subjective and therefore difficult to define. This study can help to outline similarities and differences in how the states of pain and distress are recognized by those who determine their existence.

PROCEDURES TO BE FOLLOWED

You were contacted first by a letter of invitation. The student investigator, Valerie Parkison then called you at your listed office number to answer any further questions and to invite you to participate in this research study. The general logistics of the interview may also have been discussed. If you decide to participate, the interview session will include only yourself and the student investigator. The interview will be audiotaped and will take approximately one hour.

CONFIDENTIALITY

The audiotapes will be labeled with a unique identification number assigned to you. The actual identities will only be known by the student investigator. Research details that could provide identifying information such as current project, the focus of the laboratory, and specific procedures will not be included on the tapes. The tapes and transcripts will be locked in an office at the Center for Animals and Public Policy at the Tufts University School of Veterinary Medicine and only the student investigator, Valerie Parkison and the Principal Investigator, Allen Rutberg will have access to the key. At the end of the study, all the tapes and transcripts will be destroyed.

RISKS

Material to be discussed is sensitive and could be potentially disturbing to you. You can discontinue the interview at any time and/or not answer any of the questions.

BENEFITS

There are no direct benefits to the participants.

ALTERNATIVES

The alternative to participate is not to participate in the study.

PRINCIPAL INVESTIGATOR'S PHONE NUMBER

If you have any questions about this study, call:

Allen T. Rutberg, PhD	508.887.4769
Valerie Parkison* **(contact person)**	**617.596.4970**
Jan E. Dizard, PhD	413.542.2742

COST

There will be no cost to you for participating in this study.

STIPEND

No stipend will be offered to you for participation.

PARTICIPANT'S STATEMENT

I have read this consent form and have discussed with Dr. Rutberg or his representative, Valerie Parkison, the procedures described above. I have been given the opportunity to ask questions, which have been answered to my satisfaction. I understand that any questions that I might have will be answered verbally or, if I prefer, with a written statement.

I understand that I will be informed of any new findings developed during the course of this research study.

I understand that my participation is voluntary. I understand that I may refuse to participate in this study. I also understand that if, for any reason, I wish to discontinue participation in this study at any time, I will be free to do so, and this will have no effect on my future care or treatment by my physicians or this hospital.

I understand that no funds to provide financial compensation for research-related injury or illness are available.

If I have any questions concerning my rights as a research subject in this study, I may contact the Tufts-New England Medical Center Human Investigation Review Committee at (617) 636-7512.

I have been fully informed of the above-described study with its risks and benefits, and I hereby consent to the procedures set forth above. I have received a signed copy of this consent form.

I understand that as a participant in this study my identity and my medical records and data relating to this research study will be kept confidential, except as required by law.

_____ _____

Date Participant's Signature

I have fully explained to _____ the nature and purpose of the above-described study and the risks that are involved in its performance. I have answered all questions to the best of my ability.

_____ _____

Date Principal Investigator or Representative's Signature

_____ _____

Date Witness's Signature