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VIEWPOINTS

Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) Infection Challenge Experiments in Nonhuman Primates: An Ethical Perspective

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The coronavirus disease 2019 (COVID-19) pandemic has stimulated massive investment in biomedical research with the aims of understanding the disease and developing effective vaccine and therapeutic interventions. What role should animal research play in this scientific endeavor? Both the urgency to evaluate candidate interventions for human use and growing societal concern about ethical treatment of (nonhuman) animals put into question the justifiability of animal research as a precursor to clinical trials. Yet forgoing animal research in the rush to undertake human testing might expose human research participants to unacceptable risks. In this article, we apply a recently developed framework of principles for animal research ethics in exploring ethical questions raised by a severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) infection challenge experiment involving rhesus macaques, which evaluated the protective efficacy of the mRNA-1273 vaccine that was recently approved for emergency use. Our aim is to illuminate the ethical issues while introducing, and illustrating the use of, the framework.

Keywords. ethical framework; animal research; vaccine; infection challenge experiment; nonhuman primates.

The coronavirus disease 2019 (COVID-19) pandemic has stimulated massive investment in biomedical research, with rapid advancement of scientific knowledge aimed at understanding the disease and at developing effective vaccine and therapeutic interventions. What role should animal research play in this unprecedented scientific endeavor? Both the urgency to evaluate vaccines and treatments and the growing societal concern about ethical treatment of nonhuman animals (hereafter *animals*)—especially nonhuman primates [1]—provoke questions about the need for, and permissibility of, animal research as a precursor to human experimentation. Yet forgoing animal research in the rush to undertake human testing might expose human research participants to unacceptable risks.

In this article, we apply a recently developed framework of principles for animal research ethics in examining a severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) infection challenge experiment involving rhesus macaques. Our purpose is not to render a global judgment on this experiment—a task requiring more information about its details than we possess—but instead to identify and explore the most salient ethical issues provoked by this and other research on nonhuman primates for human benefit. Although those involved in the approval and conduct of such research tend to engage with some of these ethical issues, they rarely appreciate the full range of them. In addition to illuminating the ethical issues our discussion will introduce an ethical framework, illustrate how to use it, and demonstrate its practical value.

THE SIX PRINCIPLES FRAMEWORK FOR ANIMAL RESEARCH ETHICS

In evaluating the COVID-19 challenge study involving rhesus macaques, we employ the ethical framework for animal research that was recently presented in a book, *Principles of Animal Research Ethics* [2, 3], and has generated some interest in the scientific community [4]. Here we briefly describe what we call the 6 Principles Framework before deploying its principles in raising key ethical issues about the study.

The 6 principles framework was developed with the aim of earning as much consensus as possible between the biomedical-research and animal-protection communities, as well as the confidence of the broader public. It is limited to the extent that it is only a framework; it does not come with detailed instructions for implementation (one reason to provide an illustration of its use in this article). The framework's content lies somewhere between the poles of the American status quo of animal research regulation and practice [5], which ethicists tend to agree is not adequate as it stands [6], and the views of those who would prohibit all invasive, nontherapeutic animal research [7, 8]. Offering a more robust and defensible model than the canonical "3 Rs," [9] the 6 principles framework features 2 core values of animal research ethics and 6 principles that receive support from, and specify, these values.

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The 2 core values are *social benefit* and *animal welfare*. Social benefit, or benefit to society, is the ethically defensible end or purpose of animal research, the value that most credibly justifies the enterprise. More concretely, social benefit as understood in the framework consists of the value of biological and medical knowledge, especially (but not exclusively) in contributing to the understanding of human disease and developing safe and effective interventions for treatment or prevention. Animal welfare, or protecting the well-being of animal research subjects, sets ethical limits on the various possible means of pursuing the end of social benefit. Each of the 2 core values supports, and is specified by, 3 principles, which present more specific tests for the justifiability of particular animal studies.

The 3 principles of social benefit are *no alternative method*, *expected net benefit*, and *sufficient value to justify harm*. No alternative method requires of any prospective animal study that there be no ethically permissible alternative to the use of animals that could be expected to generate the knowledge sought in the study. It is therefore similar to replacement in the "3 Rs" [9]. As the science of alternative methods advances [10, 11], this principle might become increasingly difficult to satisfy.

Expected net benefit requires of any animal study that meets the first principle—thereby offering the prospect of a unique benefit to human society—that the ex ante prospect of this unique benefit outweigh its anticipated costs to human beings. The prospect of benefit is a function of both (1) the importance or value of the knowledge sought in the study and (2) the likelihood of actually obtaining this knowledge. (Although the second factor might be difficult to estimate, it is essential to any honest, prospective cost-benefit assessment.) Expected net benefit requires that the prospect of benefit outweigh anticipated costs to human beings such as financial costs, lost time or opportunity costs, and even any risks that human subjects might face in clinical trials as a result of toxicity false negatives in animal studies. In view of low translation rates from successful animal studies to clinical use in human beings [12–14], there is reason to believe that at present this principle is frequently not satisfied. Expected net benefit is the first element of a 2-part cost/benefit assessment, the part that focuses on human interests.

If the first 2 principles are satisfied, then sufficient value to justify harm comes into play. This third principle considers "costs" to animals and requires that the prospect of unique net benefit to society be sufficiently important to justify anticipated harms to the study's animal subjects. Together, the 3 principles of social benefit require that an animal study offer the ex ante prospect of a unique benefit that outweighs costs to human beings and is sufficiently important to justify anticipated harms to animal subjects.

An animal study that meets the principles of social benefit is a promising candidate for ethical justification. But now the particular means of pursuing the research question come into focus: Do the details of study design, housing, handling, and the like adequately protect the animal subjects' welfare? Three principles of animal welfare—*no unnecessary harm*, *basic needs*, and *upper limits to harm*—together call for an affirmative answer to this question.

No unnecessary harm requires that the harm imposed on animal research subjects be minimized to what is required in pursuing scientific objectives (in a study that satisfies the principles of social benefit). Accordingly, the animals should not be harmed through negligence, and any harms associated with procedures, handling, living conditions, or the like should not exceed what is strictly necessary for the study. Basic needs requires that animal subjects' basic needs for food, water, rest, exercise, companionship, and whatever else is generally needed for a decent life for the sort of animal in question be satisfied except when strictly required for the pursuit of socially beneficial scientific objectives. The 6 principles framework leaves open the difficult question of whether (painlessly) killing an animal that could be expected, if permitted to live, to have a decent quality of life—that is, whether "premature death"—counts as a harm to the animal or, equivalently, as a failure to meet a basic need. We examine this question as it pertains to monkeys when we apply the framework to the study under consideration.

In view of the fact that the first 2 principles of animal welfare permit harms, or particular failures to meet basic needs, when legitimate scientific objectives call for them, a final principle sets a limit to permissible harm. Upper limits to harm requires that animal subjects not be caused to endure severe suffering for a lengthy period of time. Like the European Union requirement that prohibits (with few exceptions) long-lasting, severe pain, suffering, or distress [15], the present principle does not attempt to operationalize what counts as "severe suffering" or how long a "lengthy" period of time is. However, together with the other 2 principles of animal welfare, this final principle advances the reasonable goal of providing decent lives for animal subjects by prohibiting procedures, housing conditions, or deprivations that are likely to cause them to endure prolonged agony or misery.

APPLICATION TO THE RHESUS MONKEY INFECTION CHALLENGE STUDY

With this overview of the 6 principles framework, we turn to the COVID-19 vaccine challenge study involving rhesus macaques. Following the sequencing of the SARS-CoV-2 genome by Chinese investigators in January 2020, the Vaccine Research Center of the US National Institute of Allergy and Infectious Diseases (NIAID) and biotechnology company Moderna rapidly developed the novel mRNA-1273 candidate vaccine. Moderna began human safety testing in March 2020 at the same time that NIAID investigators launched an infection challenge study to evaluate the vaccine's efficacy in rhesus macaques [16]. The accelerated process of vaccine evaluation in response to

the urgency of the pandemic departed from the typical trajectory in which animal studies precede testing in human subjects. Phase III human efficacy trials of mRNA-1273 commenced in late July 2020. On 30 November, Moderna announced that the vaccine had produced efficacy of 94% in preventing COVID-19. In December, the US Food and Drug Administration issued Emergency Use Authorization for the vaccine.

The NIAID infection challenge study, conducted in conjunction with the contract research organization Bioqual, received approval from Animal Care and Use Committees at the US National Institutes of Health Vaccine Research Center and at Bioqual, where the research took place [17]. The study involved 24 rhesus macaques, aged 3 to 6 years, who underwent the following procedures. Vaccine or placebo was injected twice into their hind legs at the beginning of the study and at 4 weeks. At week 8, all of the study subjects received a challenge of SARS-CoV-2 by intratracheal and intranasal administration. Following the infection challenge, the macaques received blood draws on days 0, 7, and 14; nasal swabs on days 1, 2, 4, and 7; and bronchalveolar lavage on days 2, 4, and 7. In order to conduct lung pathology studies, all 24 macaques were killed at 1 of 2 time points during 15 days after the infection challenge.

According to the study report, "The predefined primary endpoints of the study were the difference in the viral load in BAL [bronchoalveolar lavage] fluid between the vaccine groups and the control groups [17]." These primary endpoints were examined to assess prevention of SARS-C0V-2 disease and transmission following infection challenge. The purpose of obtaining lung specimens from the euthanized monkeys was to investigate signs of protection against severe disease. The study report abstract concluded, "Vaccination of nonhuman primates with mRNA-1273 induced robust SARS-CoV-2 neutralizing activity, rapid protection in the upper and lower airways, and no pathologic changes in the lung [17]."

As with human research, the ethical evaluation of animal research is prospective. Did the prospect of generating valuable knowledge with the monkey infection challenge study meet the 3 principles of social benefit? The phase 1 clinical trials did not await completion (or even commencement) of the animal study—a point that might seem to suggest that the study was unnecessary and therefore violated no alternative method (where the alternative would be conducting clinical trials without any study involving nonhuman primates). Yet it seems reasonable to assume that promising results from such an animal study were critical for proceeding with later-phase clinical trials. That is, had the monkey infection challenge study yielded no significant efficacy signal or indicated excessive toxicity, the trial sponsor and investigators would have had compelling reason not to invest major resources and put human volunteers at risk in a large phase 3 clinical trial. In this way, the monkey infection challenge study arguably offered a unique benefit in the form of vital scientific information.

Moreover, the study offered a reasonable *ex ante* prospect of advancing an enormous eventual benefit to humanity in the form of an effective vaccine—a prospect that, we now know, was realized. The enormous value of an effective vaccine to be deployed in response to the COVID-19 pandemic presumably dwarfs the costs to humanity of conducting the monkey infection challenge study, even when we factor in the earlier uncertainty that the study and later trials would yield positive results. (In this connection it is noteworthy that rhesus macaques, due to their genetic similarity to humans, are especially good models for human physiology and susceptibility to infectious diseases [18].) Thus, expected net benefit was apparently satisfied.

Was the ex ante prospect of unique net benefit sufficiently valuable to justify the harms incurred by the monkeys involved in the study, as required by sufficient value to justify harm? To address this question requires considering the harms incurred by the test subjects.

As mentioned earlier, each of 24 rhesus macaques was exposed to the following: 2 injections of either vaccine or placebo in their hind legs; a challenge of SARS-CoV-2 administered intratracheally and intranasally; 3 blood draws; 4 nasal swabs; 3 administrations of bronchalveolar lavage; and termination of their lives, presumably by humane methods. In addition, we may assume that at least some—possibly all—of the deliberately infected monkeys who received a placebo, rather than the experimental vaccine, became ill (because rhesus macaques can acquire the illness) [19]; here we note that illness in the animal subjects was not discussed in the study report or supplementary materials [17]. The overall burden incurred by monkey subjects, especially those who became sick from the infection, is far from trivial. Yet again, the prospect of benefit associated with a promising vaccine for SARS-COV-2 was enormous, presumably enough to satisfy sufficient value to justify harm.

Turning now to the principles of animal welfare, we begin with no unnecessary harm. At first glance, it might appear that all anticipated harms associated with the monkey infection challenge study were necessary in the course of a well-designed study that looked for a signal of vaccine efficacy. We suggest, however, that the harm involved in killing the monkeys might have been unnecessary and therefore unjustified.

The book presenting the 6 principles framework flags but remains agnostic on the question of whether or not premature death counts as a harm—or, equivalently, whether avoidance of premature death is a basic need [2]. (Because of this equivalence, we may here consider the 2 principles, no unnecessary harm and basic needs, with the same set of reflections.) We cannot address the distinct, important question of whether such basic needs as sufficient space, exercise, and companionship were met, due to the absence of such details about the monkey subjects' housing and activities in the study report and supplementary materials.

The present authors maintain that creatures with the degree of cognitive and social complexity that characterizes rhesus monkeys [20–23] incur a harm when they die prematurely: a loss of the sorts of experiences and relationships that they enjoy and care about. Rhesus monkeys, in captivity, can live up to 30–35 years [24], whereas the monkeys in this experiment were killed between the ages of 3 and 6 years. Although for some sentient animals premature death may involve no, or very little, harm to them, that judgment seems implausible in the case of such animals as canines, pachyderms, cetaceans, apes, and monkeys [25]. Moreover, killing the monkey subjects does not appear to have been required for the principal aim of looking for a signal of vaccine efficacy in terms of the primary study endpoints, described above, regarding viral load as measured by bronchoalveolar lavage. Therefore, it is doubtful whether this element of the study was consistent with the principle of no unnecessary harm. However, killing the subjects enabled examination of lung pathology and thereby evaluation of whether infected monkeys showed signs indicative of severe disease. We submit that a substantial burden of proof—in the form of explicit, detailed justification—should be required to permit the killing of monkeys in this sort of experiment. This expectation was not met in the published research report or supplemental materials describing this particular infection challenge experiment.

The final principle to consider is upper limits to harm. In discussing sufficient value to justify harm, we noted that the overall harm to individual test subjects was far from trivial. We would describe the overall harm as at least moderate. Was it compatible with upper limits to harm? That is, were the monkeys ever caused to undergo severe suffering for an extended period of time without relief from anesthesia, analgesics, sedatives, or the like? As far as we know, even the most stressful procedure, bronchalveolar lavage, is unlikely to cause so much experiential harm. On the other hand, monkeys who became ill as a result of infection presumably experienced some significant degree of discomfort. Did the sickness cause them, or some of them, to suffer severely for lengthy periods of time? In the absence of data about illness in the monkeys, it is impossible to answer this question.

It is important to note that the 6 principles framework explicitly acknowledges that this final principle may be subject to occasional exceptions [2]. Although such exceptions are to be carefully justified and documented, it is plausible that well-designed animal studies that seek an effective intervention (vaccine or treatment) in the context of a raging, highly lethal pandemic are precisely the sorts of studies that might merit exceptions to upper limits to harm. On the other hand, if pain relief could have been provided to monkey subjects who became ill as a result of infection without interfering with the study's principal aims, then all 3 principles of animal welfare—upper limits to harm as well as no unnecessary harm and (because freedom from experiential harm is a basic need) basic needs—would call for such pain relief.

CONCLUSION

In this article, we have explicated a recently developed ethical framework for animal research and applied it to an infection challenge study in rhesus monkeys aimed at evaluating the protective efficacy of a novel mRNA vaccine for the pandemic virus SARS-CoV-2. Our principal aim has been to identify and preliminarily examine key ethical issues regarding this study. Rarely is the full range of these issues squarely confronted, yet they are pertinent to trial sponsors, animal research ethics committees, and investigators considering plans to use nonhuman primates in research that exposes them to harm for the potential benefit of developing socially valuable biomedical knowledge.

Notes

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