



Citizens for Alternatives to Animals Research & Experimentation

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Public Comments to the National Toxicology Program
RE: NTP's rodent study on safety of cell phone radiofrequency radiation
March 12, 2018

These comments are submitted on behalf of Citizens for Alternatives to Animal Research & Experiments (CAARE) a non-profit organization dedicated to promoting research without animals.

As scientists, clinicians and public health officials continue to debate the results from the ten-year, \$25 million rat and mouse study carried out by the National Toxicology Program (NTP), it's essential to take this opportunity to reflect on the limited utility of animal studies to predict outcomes in human health and biology.

By now, in 2018, these deficiencies with animal tests are so widely acknowledged that the National Toxicology Program issued a "Roadmap to guide progress toward replacing animal use in toxicity testing" on January 30 of this year. The roadmap is the result of the partnership of sixteen federal agencies, asserting the need "to develop a strategic roadmap that offers a new framework for the safety testing of drugs and chemicals, which aims to provide **more human relevant toxicology data** while reducing the use of animals." [emphasis added] ¹

More human relevant toxicology data is at the crux of what is lacking in the present study under discussion by the NTP. The disparity in results between the two species, (rats and mice), along with the gender disparities observed in the rat study are impossible to extrapolate to human radiofrequency radiation exposure. These can be debated *ad infinitum*, with any number of possible theories that may explain them, but they remain only theories, with no solid guiding principles for extrapolating them to human exposure.

As stated in the *Handbook of Laboratory Animal Science* in 1994, "It is impossible to give reliable general rules for the validity of extrapolation from one species to another. [This] can often only be verified after the first trials in the target species [humans]. Extrapolation from animal models. . . will always remain a matter of hindsight." ²

This is why CAARE was shocked to learn that the NTP intends to continue the rodent studies. At the February 2 Telebriefing discussing the results of the latest round of testing, John Bucher, senior scientist stated that the NTP plans to continue rat and mouse exposure trials using a

“redesign of the exposure chambers.”³ This was confirmed in an email from Dr. Bucher, in response to CAARE’s question.

Given the severe limitations of animal tests, the availability of new technologies that perform better, and the announcement of the Strategic Roadmap to utilize more relevant human test methods, it is unjustifiable – indeed unconscionable – to allow more animals to die in the remaining NTP study.

CAARE is calling for the immediate termination of any plans to use an additional 300 animals to complete the NTP study. We understand that a budget may have been allotted for this purpose, and that it may render a portion of the study more “complete,” but given the consistent inconclusiveness of the animal data over the past ten years, there is no scientific explanation to believe that testing on an additional 300, or any number of animals will change that.

And considering that one of the three main goals of the Roadmap is “Promoting flexible approaches for establishing confidence in new methods,” this is an excellent place to demonstrate flexibility. Funding allotted for the remaining animal studies can be re-directed to studying existing tissue samples, which NTP states are in its possession. An excellent application of those tissue samples would be to utilize them to develop organoids or organs-on-chips. (See discussion further down.)

CAARE believes that terminating plans to use additional animals to continue the present NTP radiofrequency radiation study is strongly warranted by the following:

- 1. Results of animal experiments do not translate to human biology**
- 2. Animal data is frequently disregarded when assessing human health concerns**
- 3. The NTP animal study won’t change the current recommendations on cell phone usage**
- 4. Resources should be devoted instead to cutting-edge methods such as organs-on-chips and organoid models.**
- 5. It violates the intent of the Strategic Roadmap and other animal use policies**

We discuss each of these points further.

Results of animal experiments do not translate to human biology

Twenty-first century science has demonstrated repeatedly that animal data does not extrapolate with any confidence to human biology and medicine. This has resulted in the current trend to replace animal data with human-relevant data, as exemplified by the Strategic Roadmap.

Even so, there is a proclivity to depend on animal tests based on years of habit and other factors, such as the belief that it may be more applicable in some areas than others, or, that lacking other methods, the use of animals is a starting point. But this is without justification.

As one example, scientists have been working to create an artificial kidney for at least 15 years but have been thwarted by the problem of how to keep the blood flowing smoothly through the artificial device without clotting. A recent breakthrough occurred using a new computer simulation developed by a group of U.S. researchers. The research team, recipients of Quantum Awards from the National Institute of Biomedical Imaging and Bioengineering (NIBIB) utilized a computer simulation of blood flow to provide an accurate model.

The *in silico* model has offered a viable and superior alternative to animal models, as explained by one of the lead researchers, Shuvo Roy, Ph.D., professor of bioengineering at University of California, San Francisco, who stated: "To do that in animal studies is time consuming, expensive, and at some level you never know if it is going to work out—because animal blood is not the same as human blood." ⁴

Here we see a phenomenon that science might suggest is impacted by only physical, rather than biological forces, but these two are inextricably merged, and the deeper we probe biology, the more relevant, and the more species-precise we learn that these difference are.

These biological disparities form the basis of why testing on animal species will never provide accurate answers for human health challenges. They are leading the way for the twenty-first century trend to move away from animal tests, as demonstrated by the U.S government's Tox21 program, initiated in 2008 to decrease reliance on animals for chemical safety testing. ⁵ Tox21 sparked the current NTP Strategic Roadmap which furthers those goals.

Last week, March 7, 2018 the Environmental Protection Agency (EPA) announced the release of its draft document to develop a strategy for advancing alternatives to animal testing, as required under the 2016 Toxic Substances Control Act (TSCA). According to the report, which is still in draft form, the EPA hopes to eventually and completely eliminate chemical testing on vertebrate animal species, which includes mammals, fish, birds, amphibians and reptiles.

This acknowledgement of animal testings' sweeping deficiencies is not unique to the U.S. According to a report from the UK, issued in January 2018 by the BioIndustry Association and the Medicines Discovery Catapult, it is essential to "humanize" drug development to enhance productivity in drug development. The reports states that current high rate of failure in developing successful new drugs exists because scientists are relying on animal experiments, which give poor results, rather than studying human data.

"Discovery must start with biological targets derived from patient data and samples, which create candidate drugs that are highly selective for proven human disease targets in well-defined patent subgroups, not animal models," said Chris Molloy, chief executive of the MDC. ⁶

Over and over again, we are seeing scientists and governing agencies acknowledge that animal tests are blatantly insufficient. It strains credulity to understand why the NTP plans to use, and kill, another 300 animals, for this futile pursuit.

Animal data is frequently disregarded when assessing human health concerns

Despite the widespread reliance on animal experiments, data obtained from them is frequently discounted when making assessments for human health and safety.

To address the question of risk from cell phone usage, the National Cancer Institute posts on its webpage a detailed Fact Sheet that discusses only human data.⁷ The fact sheet uses information from a dozen human epidemiological studies, including three large-scale studies that involved hundreds of thousands of individuals. Many of these studies span decades. Some information about studies on human cells is also included.

The *Wall Street Journal* reported in response to the NTP study that Otis Brawley, chief medical and scientific officer for the American Cancer Society, stated the results will have no impact on what he tells people about cell phone safety. “The evidence for an association between cellphones and cancer is weak, and so far, we have not seen a higher cancer risk in people,” he said. “But if you’re concerned about this animal data, wear an earpiece.”

Concerns about cell phone safety continue to be generated by confusing results from animal tests. A 2017 Norwegian human study demonstrated that cell phone use by pregnant women was not linked to any adverse effects on the child’s neurodevelopment. Dr Eleni Papadopoulou, the lead author from the Norwegian Institute of Public Health, noted that: “The concern for harm to the fetus caused by radio frequency electromagnetic fields, such as those emitted by mobile phones, is mainly driven by reports from experimental animal studies with inconsistent results.”⁸

The NTP rodent study won’t change the current recommendations on cell phone usage

As with so many animal studies, the findings of the NTP study are of ambiguous significance. Tumors showed up in numbers that may or may not have statistical significance. Confounding the data even further, the results showed up only in male rats, without explanation. Senior Scientist, Dr. John Bucher, who conducted the February 2 Telebriefing, corroborated this by his statement that “Everything is inconclusive.”

The resulting coverage in the news media attested to the inconclusiveness of the outcomes. Announcing “Cell phone radiation study finds more questions than answers,” and “Why the Largest Study Ever on Cellphones and Cancer Won’t Settle the Debate,” a range of major news outlets expressed the confusion and ambiguity of this major study.

The *Wall Street Journal* reported: “U.S. researchers spent nearly two decades to design and carry out a definitive study on the health effects of cellphone radiation. The final results, released Friday, are likely to fuel rather than dispel the long-running debate about the planet’s most ubiquitous electronic device.”⁹

CNN quoted Dr. Bucher: “In our complete evaluation, we again had a lower level of certainty that small increases in the numbers of male rats with tumors in the brains were associated with

exposures to cell phone radio-frequency radiation. These findings are termed 'equivocal evidence of carcinogenic activity,' meaning it was unclear if the tumors were related to the exposures." ¹⁰

Due to their lack of conclusiveness, none of the NTP animal tests have had any impact on industry recommendations for cell phone use. According to a statement issued by the Cellular Telecommunications and Internet Association (CTIA) which represent the US wireless communications industry:

"NTP issued partial results of its rat study in June 2016 and since then the Federal Communications Commission, the Food and Drug Administration, the World Health Organization, the American Cancer Society and numerous other international and US organizations and health experts **have maintained their long-standing conclusion that the scientific evidence shows no known health risk due to the (radio-frequency) energy emitted by cell phones.**" ¹¹

Devote research resources into cutting-edge methods such as organs-on-chips and organoid models to derive human-relevant data.

Two advanced cell culture models have been emerging over the past 5-10 years that use human cells and tissues to develop highly-relevant human organ and tissue models: Organs-on-chips and 3-dimensional organoids. These both show excellent potential to deliver results for biomedical research that is unprecedented. Indeed, many have already gone beyond merely the potential phase and have delivered proof-of-concept results.

Organoids, often developed from induced pluripotent adult stem cells, have allowed researcher to generate structures known as mini-brains, small cellular formations which grow on a biological substrate and self-assemble using innate genetic cellular instructions to form 3-dimensional models of the developing human brain. Organoids have been used by a number of scientists to model the precise mechanisms by which the Zika virus disrupts normal neurodevelopmental processes. ^{12 13 14} This would be an excellent technology to test cell phone radiation on human organoids.

Organs-on-chips are micro-engineered biological systems that combine human cells, exposed to natural forces and factors to create a micro-environment that simulates natural physiology. This further equips them with the ability to simulate the interaction of different cells and tissues.

The Wyss Institute for Biologically Inspired Engineering, affiliated with Harvard University, has been at the forefront of developing organs-on-chips. Researchers there have created sophisticated organ chip models for lung, intestine, heart, and kidneys. Several of these have been further refined into disease models for cancer, asthma, pulmonary thrombosis, muscular dystrophy and others.

More recently, the Wyss Institute has enhanced its organs-on-chips with embedded electrodes that measure real-time assessment of electrical activity of the living cells. This provides them with a precise gauge of tissue health and differentiation.

“These electrically active Organ Chips help to open a window into how living human cells and tissues function within an organ context, without having to enter the human body or even remove the cells from our chips,” describes Don Ingber, PhD, Founding Director of the Wyss Institute. “We can now start to study how different tissue barriers are wounded in real time by infection, **radiation**, drug exposure or even malnutrition, and how and when they heal in response to new regenerative therapeutics.” [Emphasis added] ¹⁵

At the annual meeting of the American Association for the Advancement of Science, February 2018, organs-on-chips featured prominently as one of the brightest advancements poised to supersede animal models. Robert Urban PhD, head of Johnson & Johnson Innovation told conference attendees how using organ chips make it possible “to demonstrate how the molecular biology of toxicology is actually taking place. **You can understand pharmacokinetics and other features you can’t understand from animals. This is a timely addition and hopefully a replacement for laborious, ill-predictive animal models.**” ¹⁶

Scientists at the Imperial College of London have developed a ‘Liver-on-a-Chip’ that can accurately mimic Hepatitis B infection (HBV). This new liver chip model is over 10,000-fold more susceptible to HBV infection than other models and accurately mimics what happens in an infected patient.

According to Marcus Dorner, PhD, one of the study leaders: “Studying chronic viral infections and developing drugs often requires animal experiments, which might not always result in data, which are translatable to humans. **Using advanced tissue engineering models to study these infections in their natural human host environment not only results in directly usable data for human applications but furthermore also reduces the need for animal experiments.**” ¹⁷

Research at the Charles Stark Draper Laboratory in the U.S. has demonstrated that using its organ chips, scientists can observe neutrophils [white blood cells] penetrate cells and tissues in real time, indicative of an inflammatory response. Animal testing cannot show this. ¹⁸

The potential for organs-on-chips to elucidate precise mechanisms of intricate physiology is unprecedented and makes them a perfect methodology for assessing damage from cell phone radiofrequencies on human tissue and resulting potential damage. NTP should direct future resources for studying cell phone hazards into methods that are likely to give answers rather than perpetuate failed animal models.

Conclusion

A burgeoning amount of evidence points to the widespread failure of animal tests to simulate human biological mechanisms. To continue additional animal tests as part of the NTP study is to propagate a failed methodology that is also inhumane, resulting in unjustifiable animal suffering and death. It also violates the intent of the Strategic Roadmap recently published by NTP.

Is it scientifically valid to conclude that the NTP radiofrequency animal studies are and will continue to be inconclusive. To kill more animals at this point violates the *International Guiding Principles for Biomedical Research Involving Animals* which states that “The use of animals for scientific and/or educational purposes is a privilege that carries with it moral obligations and responsibilities for institutions and individuals to ensure the welfare of these animals to the greatest extent possible.” It further states that “Animals should be used only when necessary and only when their use is scientifically and ethically justified.”¹⁹ We do not believe that this threshold can be met by the continuation of the NTP study.

CAARE and our thousands of supporters urge the NTP and the National Institute of Environmental Health Sciences to terminate the animal studies and move on to superior, human relevant safety analysis.

Thank you for the opportunity to submit these comments.

Sincerely,



Barbara Stagno
President

¹ Roadmap to guide progress toward replacing animal use in toxicity testing, Jan 30, 2018, NIH press release at <https://www.nih.gov/news-events/news-releases/roadmap-guide-progress-toward-replacing-animal-use-toxicity-testing>

² Handbook of Laboratory Animal Science , Volume II: Animal Models Svendsen and Hau (Eds.) CRC Press 1994 p6.

³ Telebriefing: NTP draft conclusions for radiofrequency radiation studies in rats and mice <https://www.nih.gov/news-events/news-releases/telebriefing-ntp-draft-conclusions-radiofrequency-radiation-studies-rats-mice>

⁴ National Institutes of Health, “Computer simulation addresses the problem of blood clotting,” February 9, 2018, <https://medicalxpress.com/news/2018-02-simulation-problem-blood-clotting.html>

⁵ NIH News Release; *NIH Collaborates with EPA to Improve the Safety Testing of Chemicals, New Strategy Aims to Reduce Reliance on Animal Testing*, February 14, 2008. <https://www.nih.gov/news-events/news-releases/nih-collaborates-epa-improve-safety-testing-chemicals>

⁶ [Biotech groups urged to focus more on humans in drug tests](https://www.ft.com/content/4ae80030-fb8f-11e7-a492-2c9be7f3120a) Financial Times, Jan 17, 2018, <https://www.ft.com/content/4ae80030-fb8f-11e7-a492-2c9be7f3120a>

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