The Rise of *In-Vitro* Toxicity Testing

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The Conventional Rule

Toxicity studies examine what happens when a chemical is absorbed into the body, usually via the mouth, but sometimes also through the skin or the lungs. The aim is to find out if the substance has toxic effects. The most common tests includes acute, subacute and chronic toxicity tests and many different substances are tested in this way, including all drugs, agricultural chemicals, pesticides, and some cosmetics and their ingredients. Theoretically, toxicity profiling in animals would be most useful if the test model responded in a fashion that was identical to the human. However, such is seldom true in practice, even when the route of administration and vehicle used are identical to the clinical use because deposition (pharmacokinetics) can vary dramatically among species, and even between strains. Therefore, most toxicity tests are performed in two species - a rodent and a non-rodent - to ensure that any unexpected adverse effects are not overlooked before new chemical entities (NCE) are introduced into man. The sequence of toxicity testing proceeds from the simple to the complex. This is because toxicity testing is desirable at an early stage in the development process but large quantities of drug samples are usually not available at that time, limiting the advancement of medical science during the last 100 years which largely depended on research with animals. Animal studies have provided the scientific knowledge that allows health care providers to improve the quality of life for humans and animals by preventing, treating diseases and disorders, and easing pain and suffering.

The Movement

Use of animals in scientific research and testing has raised controversy and criticism for long. So, the issue of alternatives to the use of animals in research and testing is indeed not new, and in many ways continues to be contentious. The first protests for animal protection began in England during mid-nineteenth century, in which activists opposed all forms of animal research. The protests gained momentum during the seventies, and many believe that the work of Peter Singer entitled *Animal Liberation* in 1975, revived the call for animal protection. A few of the presentations are highlighted here. Alan M. Goldberg (John Hopkins University, Baltimore) spoke on 'The science of alternatives', which detailed the three Rs, i.e. replacement, refinement and reduction of alternative and humane science. While exploring the societal expectations of animal use in science, Goldberg addressed the importance of the three Rs. The Bologna Declaration of 1999 signed by 29 European countries was a watershed event for the issue of animal use and this declaration paved the way to the alternate animal testing protocol, viz. the three Rs.

The concept, developed by Russell and Burch advocates that, animal testing protocols conform to the three Rs. Replacement of animal testing by alternative methods is the most radical of the methods proposed, although a differentiation is made between absolute and relative replacement. Refinement on the other hand, is the subtle approach which advocates the reduction of incidence or severity of distress experienced by laboratory animals. Reduction entails obtaining precise information with animals through the use of well-designed, well conducted, reliable experiments that do not involve endless repetition of the same tests.

The Verdict

With the advent of modern era the understanding of the principles and mechanisms of the biomedical sciences blossomed significantly with the development of cell and tissue culture techniques. By the 1960s, the development of cell

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culture methods to grow animal and human cells and tissues on plastic showed promise in the survival of cells outside intact organisms. With further refinement of the culture conditions and insights into the requirements for maintaining viability of organs, tissues, and cells in an artificial environment, a transformation in cell biology was underway.

A variety of scientific disciplines began to appreciate the breakthroughs realized with these new techniques. Soon after that, in the 1970s, toxicologists adapted cell and tissue culture methodologies as pathways for revealing mechanistic toxicologies the mechanisms of actions of toxicants. Cell culture broadened the scope of cell and cancer biology and other toxicologists came to realize that cells grown *in vitro* also retained the properties of their organs of origin. Perhaps these surrogate "tissues" grown in plastic could mimic animal tissues and organs in their responses to toxic insults.

Rowan used the term 'alternative' to refer to those techniques or methods that replace the use of laboratory animals altogether, reduce the numbers of animals required, or refine an existing procedure or technique to minimize the level of stress endured by the animal. Thus the term *in vitro toxicology* is generally referred to the handling of tissues outside of intact organ systems under conditions that support their growth, differentiation, and stability. Since the seminal experiments established the validity of cell culture techniques in toxicology, the methods have proven fundamental for understanding and developing critical procedures in cellular and molecular biology, as well as in pharmacology, genetics, reproductive biology, and oncology. Cell culture technology has improved dramatically as a result of interest in the methods and through its broad applications.

The concept of alternative techniques is now widespread throughout the scientific community. This is due largely to regulations and standards which require consideration and support to alternatives. However, the field of alternative study particularly *in vitro* toxicology has evolved into a respected discipline and is attracting competent and motivated scientists around the world. Although the normal rate of progression of any scientific discipline is determined by progress within the scientific community, some areas have received more encouragement than others. Specifically, the public objections and disapproval of animal testing have forced academic institutions, industrial concerns, and regulatory agencies to direct research initiatives toward the development of alternative methods of toxicity testing.

References

1. Animals (Scientific Procedure) Act 1986, HMSO, London.
2. Caldwell, J., *Toxicol. Lett*., 1992, 64-65, Spec. No. 651-659.
3. Eaton D & Klaassen C, "Principles of Toxicology" in C Klaassen (ed), Casarett and Doull's Toxicology: The Basic Science of Poisons (5th ed), New York: McGraw-Hill, 1996.
4. Frank a. Barile , "Toxicology Testing In Vitro",Principles of Toxicology Testing, CRC Press, Taylor & Francis Group,2007
5. Rowan A, Of Mice, Models, and Men: A Critical Evaluation of Animal Research , Albany: State University of New York Press, 1984
6. Singer, P., Animal Liberation: A New Ethic for Our Treatment of Animals, Random House, New York, 1975.
7. Weiss, B., The Role of Animals in Biomedical Sciences, New York, 1983, pp. 82-91.
8. Zbinden, G., *Drug Saf*., 1990, 5 (Suppl. 1), 58-64.

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Indian Prime Minister awarded FAO Agricola Medal

FAO Director-General Jacques Diouf conferred the UN organization's highest award, the Agricola Medal, on Indian Prime Minister Manmohan Singh in recognition of his contribution to agricultural development and the reduction of hunger and poverty. India is the world's second largest agricultural producer after China, with farming employing over 60 percent of the population and accounting for 18.5 percent of GDP. The flow of credit to Indian farmers had almost doubled in the last four years. Dr Diouf told Mr Singh that he had shown "exemplary vision and resolve" in promoting the growth of Indian agriculture. Horticultural production is set to double by 2012 while plans are underway to increase national Rice, Wheat and Pulses production by 20 million tons, thus significantly improving domestic food security.

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