



Workshop 2.6 Humane endpoints

Lecture

Impediments to humane endpoints in infectious disease research

Karl Andrutis¹ and Alicia Karas²

¹Tufts University Cummings School of Veterinary Medicine, Division of Teaching and Research Resources, North Grafton, USA;

²Tufts University Cummings School of Veterinary Medicine, Clinical Sciences-Anesthesiology, North Grafton, USA

Infectious disease studies can cause varying degrees of pain or distress from exposure to infectious agents or toxins and subsequent morbidity and mortality. Although the consideration of alternative methods prior to performing animal studies is mandated by United States regulatory authorities, many scientists continue to view this requirement as intrusive and not beneficial to their work. Another view might be that the development, evaluation, and implementation of alternative methods are a critical part of the refinements made in any scientific endeavour where methods are modified to produce the highest quality results. Refinements in infectious disease studies consist primarily of assessment strategies and selection of humane endpoints. This

begins with an understanding of the research's impact on the animal. Although animal models are used as biological systems, many scientists seem unable to consider the impact of research methods on the animal and their effects on the results. Why is it difficult for some scientists to recognise and understand the full effects of an experimental manipulation on an animal? Why are alternative methods not a priority for scientists? What can the regulatory agencies do to further encourage and enforce the use of alternative methods? How does an IACUC resolve these issues? Identifying the barriers to the development, evaluation and implementation of alternative methods may assist in greater application of alternatives.



Lecture

Workshop humane endpoints in animal experiments for biomedical research

Coenraad Hendriksen¹, Klaus Cussler² and David Morton³

¹Netherlands Vaccine Institute, Bilthoven, Netherlands; ²Paul-Ehrlich-Institute, Langen, Germany; (3) Univ. of Birmingham, Birmingham, UK

Animal well-being in an experimental study is often related to the endpoint being used. Therefore, one of the major targets in refinement strategies is replacement of potential painful or stressful endpoints by earlier, humane endpoints. This strategy has been adopted by the scientific community as well as by regulatory bodies, such as European Pharmacopoeia, and by organisations such as WHO and OECD. Humane endpoints can be defined as the point at which an experimental animal's pain and/or distress can be terminated, minimised, or reduced by actions such as killing the animal humanely, terminating a painful procedure, or providing treatment to relieve pain and/or distress (CCAC, 1998).

This workshop will present the outcome and will discuss the conclusions and recommendations from the 2nd International

Conference on the Use of Humane Endpoints in Animal Experiments for Biomedical Research that was held as a satellite meeting to the 5th World Congress, August 20-21. The objective of the conference was to review progress made since the first International Conference that was held in Zeist, November 1998. Important issues that were addressed are the recognition and assessment of adverse effects in animals and the determination, validation, implementation and acceptance of humane endpoints. Furthermore, new techniques, new approaches and new strategies using non-invasive methods were presented. Other key-issues were the training of observers and the use of recently developed remote sensing devices, such as telemetry and biophotonic imaging. The conference was initiated and organised by the Working Group on Humane Endpoints (HELP).

Lecture

Urinary biomarkers as non-invasive toxicological endpoints – recent advances

Raymond Poon

Environmental Health Science Bureau, Health Canada, Ottawa, Canada

Urinary biomarkers as humane endpoints were reviewed seven years ago (Poon and Chu, Proceedings of the International Conference on Humane Endpoints in Animal Experiments for Biomedical Research, Nov. 1998, Zeist. Hendriksen and Morton, eds., pp 85-88). Since then, many new urinary biomarkers associated with a broad range of toxicity have been reported. 4-Hydroxynonenal, a lipophilic aldehyde, is identified as a biomarker of lipid peroxidation induced by chemicals such as TCDD, while 8-isoprostaglandin F₂ α is a specific marker of free-radical catalysed peroxidation of arachidonic acid. Elevated urinary metallothionein-1 is correlated with exposure to heavy metals. Creatinuria has been repeatedly observed to associate with testicular injury. Increased Clara cell protein (CC16) is associated with lung damage following ozone exposure. Kidney injury molecule-1 (KIM-1) is a novel urinary biomarker of prox-

imal tubular damage. Sucrose excretion has been used to measure the intestinal permeability change following drug induced gastrointestinal damage. Microalbuminuria has been shown in our laboratory to be a sensitive indicator of kidney tubular dysfunction in rodents. NMR and MS based proteomic and metabolomic studies have identified new proteins and metabolites in urine that are associated with organ toxicity. Thus, parvalbumin alpha is recognised as a biomarker of skeletal muscle toxicity, the metabolite N-methylnicotinamide is a potential biomarker of peroxisome proliferation. Many metabolite profiles have been documented that characterise toxic expressions of specific chemicals and pharmaceuticals. Validating and incorporating these new target and mechanism specific endpoints in toxicological studies will enhance the power of detecting toxicity, and reduce the number of animals used.