



## Conference Reports

# Special Symposium “Animal Alternatives and Sustainable Development” within the “International Conference on Ecosystem Conservation and Sustainable Development”

Ambo, Ethiopia, February 10-12, 2011

Ambo (also known as Hagere Hiwot) is a small town in the West Shewa region of the State of Oromia in Central Ethiopia, 112 km west of Addis Ababa. Ambo University, established in 1939, is a pioneering institution in the advance of scientific thought and practice in the fields of agriculture and rural development. The university now is making its impact felt in various other fields as well, including engineering and the health sectors. With its sprawling campus (8 colleges with 27 academic departments), more than 8000 students, large library, and modern laboratory equipment, Ambo is one of the prominent institutes of higher education in Ethiopia.

The International Conference on “Ecosystem Conservation and Sustainable Development” (ECOCASD 2011) was organized by the Department of Biology of Ambo University and was held on February 10-12, 2011. More than 300 scientists from Africa, Asia, Australia, North-America, and Europe attended. The Conference was organized primarily by Prof. Pavanasam Natarajan, Director-CATER, Department of Biology, but the President and Vice-Presidents of the University also contributed significantly to its success. The program was supported by TWAS-UNESCO (Italy), MGDC (India), DZF (Switzerland), I-CARE (Italy), ILRI, IWMI, HoA-REC/N, EFASA (Ethiopia), CTA (Netherlands), SABS (India), and IOBB (Uganda).

The importance of this international conference is reflected in the list of illustrious attendees at the inaugural function. In addition to the organizer, Prof. Natarajan, and the President of Ambo University, Dr. Mitiku Tesso, the audience included the President of the Federal Democratic Republic of Ethiopia, Mr. Girma Wolde-Giorgis, as well as three ministers, Mr. Tesgenu (Minister for Water and Energy), Mr. Mekonen (Minister of Education) and Dr. Urgessa (State Minister of Education). The presence also of the Second Secretary of the Indian Embassy, Mr. Subhash Chand, demonstrated the significance of the meeting to the country and to international relations.

The Special Symposium on “Animal Alternatives and Sustainable Development” took place on February 11 in the auditorium of the university, with no parallel sessions.

Prof. **Mohammad A. Akbarsha**, Director of the Mahatma Gandhi Doerenkamp Center (MGDC) and Gandhi-Gruber-Doerenkamp Chair (India), opened the symposium with an introduction about the Gandhian philosophy of non-violence in teaching and research in the life sciences. Prof. Akbarsha traced the history and present status of animal use in education, research, and testing. Outlining the contexts of animal use, he discussed the implications of animal use in education from the perspectives of curricular, pedagogic, environmental, legal, and ethical issues. He emphasized not only that digital alternatives, models, charts, mannequins, self-experimentation, field studies, etc. can replace animal use in education, but also that these alternatives offer a much better learning opportunity than does the dissection of animals.

With respect to animal use in pharmacology, toxicology, and cosmetics testing, Prof. Akbarsha quoted surveys that revealed the use of several million animals for these purposes each year. Providing a long list of prescription drugs that were accepted in animal testing during the preclinical stage but later were withdrawn from the market due to life-threatening side effects, he highlighted the difference between animals and humans with respect to drug and toxic chemical metabolism. He then introduced Russell & Burch’s 3Rs concept and listed various alternatives in this context.

Elaborating upon the laws of the European Union and the USA that require the reduction/replacement of animals in testing, Prof. Akbarsha briefly touched upon the evolution and role of the regulatory and validating authorities and organizations (i.e. FDA, OECD, ECVAM, ICCVAM, JacVAM, REACH, ZEBET, etc.) in the introduction of alternatives. He lauded the roles and contributions of CAAT, CAAT-Europe, the transatlantic think tank for toxicology (t<sup>4</sup>), DZF, FRAME, etc., as well as the scope and aims of tox21c, AXLR8, and the Human Toxicology Project Consortium. He went on to introduce the journals publishing articles on animal alternatives: ALTEX, AATEX, ATLA, Toxicology In Vitro, etc. Then he briefly introduced the World Congresses and other international meetings that discuss alternative



methods. He elaborated on the trends in *in vitro* toxicology and traced developments in the field that led to Integrated discrete Multiple Organ Co-Culture (IdMOC), Human Whole Blood Pyrogen Test (fever in the test tube), the 3D skin and cornea reconstruction models, hepatocyte spheroid culture, etc.

Dr. **Mukkura C. Sathyanarayana**, Adjunct Professor, MGDC, India, in his talk on “Animal protection laws, alternatives, and sustainable development,” underscored for the audience the very high numbers of frogs and lizards sacrificed for dissection and experiments in education: in India alone, three million frogs are utilized every year for teaching purposes. Prof. Sathyanarayana explained the absolutely essential role animal species play in the stability of an established ecosystem and the need for legal assurance of their rights: “The biodiversity conservationists must make it their mission to promote sustainable ecosystems and conservation of biodiversity by advancing the understanding, development, and implementation of animal protection laws.” He then addressed the status of animal protection regulations in different countries, focusing, in particular, on the “Prevention of Cruelty to Animals” Act (1960) of India. The statutory provisions associated with this Act are enforced in India by an independent Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA). Due to the binding guidelines of CPCSEA, investigators using animals in their research must provide ethical and scientific justification in their applications to the Institutional Animal Ethics Committees (IAEC) prior to undertaking screening and/or pre-clinical studies on animals.

Dr. Sathyanarayana then gave an overview of the legal efforts aimed at strengthening animal and wildlife protection worldwide – a clear indication that the protection of animal rights and the protection of wildlife are of significant concern in modern societies, as is the ethical evaluation of society’s actions with regard to animals and wildlife. He laid special emphasis on the animal protection law of Germany, enacted as early as 1930s, which reflected grave concern for the wellbeing of animals. During the lively discussion that ensued, Dr. Sathyanarayana concluded that use of the alternative methods in education can obviate the removal of animals from their natural habitats and thereby contribute to biodiversity conservation and sustainable development.

Next, Prof. **Krishna K. Sharma** from the Amphibian Biodiversity Research Laboratory, MDS University, Ajmer, Rajasthan, India, gave a lecture on the role of amphibians in the ecosystem and on current threats and conservation strategies. Prof. Sharma elucidated the role of amphibians in the ecosystem and their significant impact as secondary consumers in many food chains. Amphibians, as insectivores, also play an important role in containing insect-transmitted viral infections to man and plants (crops). Owing to their sensitivity in the larval stage and the usefulness of their decoded genomes, amphibians in early developmental stages are exploited, both as experimental material and as sensors for gathering information about the stress status of ecological sites. The amphibian population has declined notably over the past two decades; currently 30% of frog strains worldwide are threatened. Prof. Sharma explained that in addition to natural stressors such as infections and radiation, the human threat to the amphibians includes destruction

of the ecosystems on the one hand and, on the other hand, the over-exploitation of amphibians in laboratories for experiments and for teaching purposes. Prof. Sharma introduced alternative non-invasive methods for phylogenetic relationship and taxonomy using marker genes and sonographic techniques. He also demonstrated e-learning programs that can replace the dissection of amphibians for educational purposes. The encouraging results viewed digitally on-screen raised a lengthy discussion about the features and future of the digital alternatives to *in vivo* hands-on education in classical biological education.

Dr. **Surendra Ghaskadbi** (Agharkar Research Institute, Pune, India) proposed “*Hydra* as a simple alternative model for studying diverse biological phenomena and their evolution.” He described the phylogeny of *Hydra* as an early, thus simple, metazoan with an organized nervous system, and he presented ongoing research data and new findings on the molecular features of *Hydra*. The focus of the research consortium around Prof. Ghaskadbi lies in functional similarities between *Hydra* and vertebrates, particularly with respect to pattern formation in vertebrate embryonic development. In this regard, the Noggin-Protein is of central importance in vertebrates. As Dr. Ghaskadbi’s research revealed, the *Hydra* *noggin*-like gene shows functional conservation in vertebrates. He also described findings regarding the similarities between *Hydra* DNA repair abilities (Nucleotide Excision Repair) and those of higher vertebrates. Dr. Ghaskadbi’s research anticipates that *Hydra* could be developed as an alternative model system to *in vivo* as well as to *in vitro* testing of agents interfering with Noggin-related pathways, DNA repair capacity, and similar molecular correlations that could be expounded.

Dr. **Mardas Daneshian** from the Center for Alternatives to Animal Testing in Europe (CAAT-Europe) at the University of Konstanz, Germany presented the success story of the *in vitro* pyrogen test (IPT) as an alternative method for replacement of the rabbit pyrogen test. He explained the nature of pyrogens – fever-inducing agents – and their interaction with the human immune system. He pointed to the necessity of testing the pyrogenic burden in injectable drugs, and a critical discussion of the *in vivo* rabbit pyrogen test and the *in vitro* *Limulus* amoebocyte lysate test (LAL) followed, which focused especially on ethical problems and methodological shortcomings. Dr. Daneshian introduced the *in vitro* pyrogen test principles of the second generation, which are based on measuring pro-inflammatory cytokine release from relevant human cell types and organs, i.e. monocytes and whole blood.

As the IPT involving human whole blood is a robust and sensitive pyrogen reporter system quantifying interleukin-1 $\beta$  (IL-1 $\beta$ ) as a functional readout, it was validated and included in the European Pharmacopoeia. This “fever in a test tube” approach could save the lives of millions of rabbits, since the pharmaceutical industry uses 400,000 rabbits per year for pyrogenicity safety testing. Furthermore, this method could be advanced by development and validation of cryopreserved human blood, thus standardizing the immune reactivity as a robust reference system. As the IPT system reveals a high degree of transferability, it could be successfully adapted to evaluate pyrogenic contamination on medical devices and materials and to an air-collecting



**Fig. 1: Dr. Mardas Daneshian, Prof. Mohammad A. Akbarsha, Prof. Rallapalli Ramamurthi, Prof. Krishna K. Sharma, Dr. Surendra Ghaskadbi (f.l.t.r.).**

system for evaluation of air-borne pyrogens. In addition, the combination with albumin-coated beads opens the possibility of detecting pyrogens in immunomodulatory or interfering drugs and also detection of pyrogens in the femtomolar range. During the subsequent discussion, Dr. Daneshian explained the reasons for the establishment of the lipopolysaccharide of *E. coli* strain O113 as the international standard reference system and described the complexity of the human immune system and the role of the cytokine network in it.

**Dr. Hossein Hosseinkhani** of the School of Biomedical Engineering at the National Yang Ming University, Taipei, Taiwan, thrilled the audience by introducing new developments in the biomedical area involving the most recent knowledge on three-dimensional (3D) *in vitro* cell culture applications in microfluidic chips. Dr. Hosseinkhani described the goal of his research as the integration of biology, nanotechnology, and material science to mimic organs and tissues as 3D cell cultures *in vitro* in a manner that simulates conditions, with the goal of reducing animal testing and providing human-relevant functional systems for research and testing. His system involves a novel type of collagen-based hydrogels that provide a scaffold for cell growth comparable to the conditions in human organs. The cell-laden hydrogels can be infused with molecules of the extracellular matrix, and distinct concentrations of second messengers harmonize the cells in their differentiation and aggregation. This very promising technology provides cell clusters that mirror the physical, chemical, and biological properties of organs *in vivo*. The enthusiastic discussion that followed gave Dr. Hosseinkhani the opportunity to point out the importance and urgency of further adaptation and development of the technology he and

his team are working on. Such constructs have the potential to replace both present *in vivo* and *in vitro* tests, delivering a more physiological and functional approach for basic research as well for safety testing and quality assurance.

The audience were mesmerized by the on-screen demonstration of the digital alternatives, *ProFrog Dissector*, *Dissection Works*, *PhysioEX*, etc., by Prof. Krishna K. Sharma and Dr. Mukkura C. Sathyanarayana.

The Chairperson, Prof. Rallapalli Ramamurthi, Former Vice-Chancellor, Sri Venkateswara University, Tirupathy, India, and General President of the Indian Science Congress Association 2007-08, in his concluding remarks threaded all the talks together into a cohesive story and elaborated upon the promising future for alternatives in the scientific scenario.

This special symposium covered many aspects of alternative methods and held the attention of the audience. The scientific exchange opportunities, the very inspiring and constructive atmosphere of the conference, the cordiality of Ethiopians, and the inviting features of the country all combined to make this event a fruitful, valuable, and unforgettable experience for all participants. Special mention should be made of the fact that most participants, including those from countries other than in Africa, were exposed to the concepts and practices of animal alternatives for the first time. To that extent, the special symposium was a novel venture of finding a link between animal alternatives and sustainable development, and of offering an audience its first “taste” of animal alternatives.

Mardas Daneshian, CAAT-Europe, Germany  
Mohammad A. Akbarsha, MGDC, India