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UCFM as the forerunner in the field of Laboratory Animal Science in Sri Lanka

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Animals have been models for humans since the dawn of medicine as per the first medical handbook '*Corpus Hippocraticum*' (around 400 B.C). In an era where no anaesthetic agents were identified, live animals such as dogs, pigs and monkeys were the subjects for vivisections to study human anatomy. Reverend Stephen Hales (1677–1761) inserted a brass pipe into a major blood vessel in the neck of a live horse through which a glass tube was sent and introduced the concept of blood pressure to the scientific fraternity (van Zutphen, 2001; Lewis, 1994; Felts, 1977). Similar to these, different animals have been the models for development of many fields of medicine especially for the benefit of humans. The objective of this article is to describe the role played by the Faculty of Medicine, University of Colombo (UCFM) as the forerunner in laboratory animal science in Sri Lanka to facilitate laboratory animal use in medical education, research, ethics and laboratory animal science education.

Use of animals in medical education in Sri Lanka

In order to facilitate understanding of certain physiological principles/ concepts, frogs, rats and rabbits have been used as animal models in medical education with live demonstrations for better understanding (van Zutphen, 2001). Trends in animal use in medical education in the past not only limited to the European world but in Asian countries as well. Sri Lanka was not an exception. Tracings preserved literature in the Department of Physiology, Faculty of Medicine, University of Colombo provide adequate evidence for the use of animals during physiology practical sessions in 1951 and 1952 in the oldest medical school, established in 1870 (Gunatilake, 2018ba. Changes in action potentials generated in the cardiac muscles in the presence of different ions and effects of changes in autonomic nervous system on the heart function in animals had been demonstrated by the late Physiologist, Dr H. D. W. Jansz using frogs, rats and rabbits. The effects of autonomic nervous system on the intestinal movements using dissected out parts of intestines of animals and keeping them in intestinal jar baths was among the demonstrations done in the department. The instruments used during these physiology practical sessions are items displayed in the department museum. Although, these animal-based teaching/learning methods showed physiological concepts and relevance between animals and humans, the use of animals in medical education was abandoned in 1987 with the changes in the medical curriculum in the faculty but with no substitute to explain the underlying physiological principle.

Establishment of the first animal house in Sri Lanka

Although the use of animals in medical education in the UCFM dates back to early 1950s, animals were kept in the Department of Physiology itself in the absence of a separate animal facility. This requirement was fulfilled by establishing the 'First Animal House/Facility' in the country in 1969 at the UCFM to facilitate not only medical education but also for research purposes. There had been a time where ruminants like goats and sheep were available in the faculty animal house for blood collection, which was performed routinely for several years until early 1990s. Blood have been used for the preparation of blood agar plates for microbiological investigations. Space limitations with further developments in the UCFM did not support housing of these ruminants any more since 1994. Currently this animal house has facilities to keep laboratory bred animal models like mice, rats, Guinea pigs, hamsters and rabbits and wild-caught Toque monkeys (Gunatilake, 2018a).

Faculty animal house as a research facility

With the establishment of the animal house, late Professor K. N. Seneviratne of Department of Physiology used the facility for his research work on isolated sciatic nerves of healthy and alloxan-diabetic rats in order to determine permeability of blood nerve barriers in diabetes and to understand why diabetic patients develop nerve damage (Seneviratne and Peiris, 1969; Seneviratne, 1972; Seneviratne and Weerasuriya, 1974). Not only the established researchers, but many undergraduate and postgraduate students also used faculty animal house to conduct their research projects since its establishment. Almost all undergraduates in the BSc Pharmacy degree programme from 2000 to 2015 have used the Sprague-Dawley rat model bred in the animal house except for one project. As per the available records, the research projects conducted at the animal house for specific degree programmes are given in Tables 1 and 2. Several other important research projects conducted at the animal house are given in Table 3. Publications made based on several research projects conducted at the animal house of UCFM are given in the reference list (Gunatilake *et al*, 1996; Gunatilake *et al*, 2003; Gunatilake *et al*, 2012; Herath *et al*, 2005; Ranasinghe *et al*, 2012; Samanmali *et al*, 2014; Delwatta *et al*, 2018; Wijewickrama *et al*, 2018).

Table 1: Research projects conducted at the UCFM animal house by the BSc Pharmacy undergraduates from 2000 - 2015.

Title of the research project
1. The effects of captopril on blood
2. Potency determination of anti-snake venom serum I.P
3. Screening of CNS drugs for transdural targeted delivery system
4. Cerebrum targeted trans-scalper CNS drug delivery system
5. The effect of tea extract on paracetamol pharmacokinetics in rats
6. A simple bioassay to assess the therapeutic equivalence of generic versions of furosemide
7. A preliminary investigation on the effects of polpala on the structure and function of urinary tracts of rats
8. The effect of mosquito coils on reproductive parameters of rats fetal teratogenicity
9. The effect of coriander extract on paracetemol pharmacokinetics in rats
10. Screening of brain targeted oil-based CNS drugs
11. Investigation into chylomicron mediated hepatic bypass drug delivery via lymphatics
12. The effect of Tea polyphenols on drug protein interactions in rats
13. Evaluation of cashew nut plant gum as a prospective pharmaceutical excipient
14. Evaluation of anti-diabetic activity of leaves of *Adenathera pavonina* (Madatiya)
15. Hypoglycemic & Antihyperglycemic effects of *Aporusa lindleyana* (Kebella)

Table 2: Research projects conducted at the UCFM animal house for postgraduate degrees from 1984 to 2018.

Title of the project	Postgraduate degree	Animal model used
Molecular characterization of proteins from *Plasmodium inui* and the erythrocyte membranes of *Macaca sinica*	MSc	Torque monkey (*Macaca sinica*)
Effect of *Memordica charantia* on streptozotocin induced diabetes and an evaluation of toxic effect in normal animals-Rats	MSc	*Sprague- Dawley* rats
Disposition of tea polyphenols in rats	MSc	*Sprague- Dawley* rats
Mode of action of an ayurvedic anti-hypertensive	MSc	*Sprague- Dawley* rats
Drug Interactions	MSc	*Sprague- Dawley* rats
The effect of tea polyphenols on serum protein binding of caffeine	MSc	*Sprague- Dawley* rats
Effect of polyphenols extracted from tea infusion on the absorption of caffeine in rats	MSc	*Sprague- Dawley* rats
The diuretic activity of the ayurvedic drug *Gokshura Guggulu*	MSc	*Sprague- Dawley* rats
Comparison of effects of Paracetamol and Pas panguwa	MSc	*Sprague-Dawley* rats
Antibody prevalence to rickettsial and leptospirosis infections in Sri Lanka in rats	MSc	*Sprague- Dawley* rats
Investigation of diuretic activity of *Flueggea leucopyrus* (Katapila) on rats	MSc	*Sprague- Dawley* rats
Investigation of Diuretic activity of *Aporusa lindleyana* (Kebella) on rats	MSc	*Sprague- Dawley* rats
Investigation of the Diuretic activity of *Pongamia pinnata* on rats	MSc	*Sprague- Dawley* rats
Experimental analysis of the neural stem cell populations in the hippocampal and lateral ventricle regions of the mouse brain	MSc	*Mice*
Testing the effects of vitamin E and Angiotensin converting enzyme Inhabitation kidney preservation for transplantation	MPhil	Rabbits
Reproductive molecular biology using endometrial stromal cell cultures	MPhil	*Sprague- Dawley* rats
A Biochemical study of the oral Hypoglycemic activity of *Momordica chrantia*	PhD	*Sprague- Dawley* rats
An exploration into immunity in a natural malaria infection Plasmodium fragile in the toque monkey *Macaca sinica*	PhD	Toque monkey
Transmission blocking immunity to human *Plasmodium vivax* malaria	PhD	Toque monkey
Plasmodium vivax Asexual erythrocytic stage antigens	PhD	*Balb/c* mice
A study of factors modulating infectivity of sexual malaria *Plasmodium cynomolgi ceylonensis* in *Macca sinica*	PhD	Toque monkey
A pharmacodynamics evaluation of a few members of the *Cucurbitaceac* family	PhD	*Sprague- Dawley* rats
Oral hypoglycaemic activity of *Momordica Charantia:* Possible Mechanism of action & activity related fraction	PhD	*Sprague- Dawley* rats
Metabolism of caffeine: Effects of chemical constituents of tea	PhD	*Sprague- Dawley* rats
Testing renal function in rats and rabbits after envenomation with Venoms of Russell's Viper (*Vipear russelli*), Cobra (*Naja naja*) and Hump Nosed Viper (*Hypnale hypnale*)	PhD	*Sprague- Dawley* rats and rabbits
Expression of leptin and leptin receptors in the female reproductive tract and effect of leptin in endometrial cell function of rats	PhD	*Sprague- Dawley* rats
Exploration into merozoite surface Protein 1 vaccination in a natural simian host parasite system	PhD	Toque monkey
Studies on identification of genes Coding for Strain-specific protective immunity using *Plasmodium cynomolgi*/ Toque monkey system	PhD	Toque monkey
Production of polyclonal antibodies against phytoplasma causing Leaf wilt disease in coconut palms Development of diagnostic polyclonal and monoclonal antibodies against Weligama coconut leaf wilt causing phytoplasma production	PhD	New Zealand white rabbits and *Balb/c* mice
Biochemical activity of Ayurveda formulation *'Palkalyana Gritha'*	PhD	*Sprague- Dawley* rats
Antidiabetic activity of *Canarium zeylanicum* (Kakuna), *Osbekia octandra* (Heenbovitiya) & *Piper beetle* (Bulath) and isolation of active principals	PhD	*Wistar* rats
Determination of the sub-lethal nephrotoxic dose of Russell's viper (*Daboia russelii)* venom in Wistar rats	PhD	*Wistar rats*

Table 3: Other important research projects conducted at the UCFM animal house from 2003 to 2018.

Title of the project	Animal model used
Investigation on wound healing properties of *Aloe barbadensis* miller	*Sprague-Dawley* rat
Polpala: its effect on the structure and functions of the urinary tract	*Sprague-Dawley* rat
Suitable animal model for local Leishmania parasite	Mice, rats and hamsters
Effect of *Cinnamomum zeylanicum* (Ceylon cinnamon) on blood & Glucose & lipid levels in rats	*Sprague-Dawley* rats
Reference values for selected physiological, haematological, and biochemical parameters of *Sprague-Dawley* rat model	*Sprague-Dawley* rats

Development and publication of ethical guidelines for the use of animals in research in Sri Lanka

Performance of surgeries on three dogs in 2005 created an ethical issue in the country[4] and lack of ethical guidelines for animal-based research was considered as a draw back. The request made in 2008 to UCFM Ethics Review Committee (ERC) which was the only internationally recognized ERC functioning in the country at that time accepted the challenge for the development of a set of comprehensive guidelines. The draft set of guidelines was discussed at a workshop organized by the ERC in 2008 with the participation of around 80 stakeholders from different universities, research institutions and other relevant government organizations. These draft guidelines were developed into a comprehensive set of guidelines in a few subsequent meetings and published in 2009 (*Ed.* Dissanayake *et al*, 2009). Researchers and reviewers in the universities and research institutions in the country are using this set of guidelines for obtaining/granting ethics approval before conducting animal-based research. As the time has come to identify the gaps in this set of guidelines-such as ethics in the use of animals in laboratory animal science education and the use of vertebrate embryos as toxicity models, and to do suitable amendments with more details, Sri Lanka Association for Laboratory Animal Science (SLALAS) undertook this task in 2018.

Initiation of laboratory animal science (LAS) education in Sri Lanka

Absence of organized educational programmes in LAS was another drawback faced by the researchers who enter into the field. Therefore, a 2-week intensive course was developed according to guidelines of the Federation of European Laboratory Animal Science Associations (FELASA) in collaboration with the Utrecht University, The Netherlands in 2011. This was the 'First international Certificate course in LAS' conducted in the Asian region. LAS experts from Utrecht University were the principal resource persons in the course. This course was considered as the basis for organization of similar courses in Asian countries such as India in 2013 and 2015 and in Vietnam in 2016. These courses were conducted with the financial assistance from International Council for Laboratory Animal Science (ICLAS) and Laboratory Animals Limited (LAL), UK.

When many countries moved away from ether inhalation to anaesthetize smaller laboratory animals such as mice and rats, during experimental procedures, Sri Lankan researchers were using anaesthetic ether, which is irritable to living creatures. The first course on Applied Physiology of pain, analgesia, anaesthesia and euthanasia for laboratory animals conducted in 2017 at the UCFM as an activity organized on behalf of SLALAS in collaboration with the Comparative Biology Centre, Newcastle University provided an opportunity for researchers to learn about this subject area under the expertise of Emeritus Professor Paul Flecknell. During this course, the international experts demonstrated how an anaesthesia machine could be used in laboratory animal anaestheisa for the first time in Sri Lanka. The American Physiological Society (APS), ICLAS, LAL and the Asian Federation of Laboratory Animal Science Associations (AFLAS) financially support this course (Gunatilake, 2018b).

Need for an organization for LAS in Sri Lanka

Sri Lanka was among few other Asian countries where a separate organization was not available to discuss research findings and also the new trends in the field. Therefore, with the objective of bringing researchers in the field under one umbrella, an association named 'Sri Lanka Association for Laboratory Animal Science (SLALAS)' was established in December 2012 with its base at the Department of Physiology of UCFM. From the inception, SLALAS conducted workshops and courses at different universities and research institutions to disseminate knowledge and skills in LAS. Further, the annual scientific sessions of SLALAS with the participation of international experts from America, European and Asian-Oceanian countries provided an opportunity for researchers to share their research findings and to learn new things (Gunatilake, 2013; Proceedings of SLALAS, 2014; Gunatilake *et al*, 2014).

Inaugural Scientific sessions of SLALAS held in January 2014 was considered as an important landmark in the field of laboratory animal science in the country. It gave an opportunity to introduce formally, the alternative concept of the 3Rs principal in LAS (Replacement, Reduction and Refinement). Hence, the dissemination of knowledge on validated alternative models to replace animals in toxicity testing and research was possible for the first time in Sri Lanka with the support of international experts. These models included zebrafish embryo, IdMOC (Integrated discrete multi-organ co-culture) and *in vitro* EpiDerm skin models (Gunatilake, 2014).

New trends in LAS initiated at UCFM

University Grants Commission of Sri Lanka recognized the modular based curriculum that is parallel to the world trend, for postgraduate certificate and diploma courses in LAS. These

courses are at Sri Lanka Qualifications Framework levels 7 and 8 and have the features of an education and training framework, such as modularization, harmonization, mutual recognition and quality assurance. After running these courses for several years, the future plan is to obtain accreditation of these courses by 'FELASA', which is the only organization, exist at global level.

The 3Rs principle introduced by UK scientists, Russell and Burch in 1959 is the basis of LAS (van Zutphen, 2001). Until 2016, there was no national Centre to support specially the 'Replacement' concept of 3Rs principle and hence, the initiatives were taken and the 'Sri Lanka Centre for 3Rs' was established in 2019 at the UCFM. Our aim is to develop this Centre as a resource-providing Centre in the field of LAS in collaboration with the 3Rs Centres existing currently at global level in the years to come. Furthermore, Society for Alternatives to Animal Testing in Sri Lanka (SAAT-SL) was established at the UCFM in February 2021 to promote the 'Replacement' concept in the country.

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References

1. Delwatta S L, Gunatilake M, Baumans V, Seneviratne M D, Dissanayaka M L B, Batagoda S S, Udagedara A H, Walpola P B (2018). Reference values for selected haematological, biochemical and physiological parameters of Sprague-Dawley rats at the Animal house, Faculty of Medicine, University of Colombo, Sri Lanka. *Journal of Animal Models and Experimental Medicine,* 1(4): 250-254.
2. Felts J H (1977). Stephen Hales and the measurement of blood pressure. *North Carolina Medical Journal*. 38(10): 602–3.
3. Gunatilake M, De Mel T, Fernando C, Angunawela P, de Tissera A, Welihinda J (1996). Time of onset and mechanism of nephrotoxicity after envenoming rats with Russell's viper venom and cobra venom. *Journal of the Ceylon College of Physicians*. 29(1): 29-35
4. Gunatilake M, Jayakody R L, Angunawela P, de Tissera A (2003). Direct nephrotoxic effects produced by venoms of Sri Lankan cobra, Russell's viper and hump nosed viper. *The Ceylon Journal of Medical Science.* 46(2): 61-66.
5. Gunatilake M, Lokuhetty M D S, Bartholameuz N A, Edirisuriye D T, Kularatne M U, Date A (2012). Polpala *(Aerva lanata)*: Its effects on the structure and function of the urinary tract. *Pharmacognosy Res.* 4(4): 181-188.
6. Gunatilake M. President's Message and SLALAS Inaugural workshop. In. *SLALAS Newsletter.* 2013;1(1).
7. Gunatilake M, Busquet F, Akbarsha M A (2014). Alternatives Initiative in Sri Lanka: Pre- and Post-Conference Workshops at the Inaugural Scientific Conference of the Sri Lanka Association for Laboratory Animal Science. *Altex.* 31(2): 224-226.
8. Gunatilake M (2018a). History and development in laboratory animal science in Sri Lanka. *Animal Models and Experimental Medicine,* 1(1): 3-6. https://doi.org/10.1002/ame2.12003.
9. Gunatilake M (2018b). Report on First Certificate Course on Applied Physiology of Pain, Analgesia, Anaesthesia and Euthanasia for laboratory animals held in Sri Lanka. *Adv Physiol Educ.* 42(2): 263-266. doi:10.1152/advan.00019.2018.
10. Guidelines for Ethics Review of Research Proposals involving Animals in Sri Lanka. (2009). *Ed: Dissanayake D H W, Gunatilake M, Thureisingham C and Jayamanne N M M.* Forum of Ethics Review Committees of Sri Lanka and Ethics Review Committee, Faculty of Medicine, University of Colombo.
11. Herath H M D R, Gunatilake M, Lokuhetty D, Wijayabandara J (2005). A preliminary investigation on the effects of polpala (*Aerva Lanata*) on the structure and function of urinary tract of rats. *The Ceylon Journal of Medical Science.* 48(2): 33-41.
12. Lewis O (1994). Stephen Hales and the measurement of blood pressure. *Journal of Human Hypertension.* 8(12): 865–71.
13. *Proceedings of the Inaugural Scientific Conference of the Sri Lanka Association for Laboratory Animal Science (SLALAS)*. 2014.
14. Ranasinghe P, Perera S, Gunatilake M, Abeywardane E, Gunapala N, Premakumara S, Perera K, Lokuhetty D, Katulanda P (2012). Effects of *Cinnamomum zeylanicum* (Ceylon cinnamon) on blood glucose and lipids in *Sprague-Dawley* rats. *Pharmacognosy Res.* 4(2): 73–79.
15. Samanmali B L C, Gunatilake M, Guneratne R D, Perera T R K (2014). Hypoglycaemic effect of methanolic extract of *Aporusa lindleyana* leaves on rats. J. Natn. Sci. Foundation Sri Lanka. 42(2): 129-135.
16. Seneviratne K N, Peiris O A (1969). The effects of hypoxia on the excitability of the isolated peripheral nerves of alloxan-diabetic rats. *J Neurol Neurosurg Psychiatry.* 32(5): 462-469.
17. Seneviratne K N (1972). Permeability of blood nerve barriers in the diabetic rat. *J Neurol Neurosurg Psychiatry.* 35(2): 156-162.
18. Seneviratne K N, Weerasuriya A (1974). Nodal gap substance in diabetic nerve. *J Neurol Neurosurg Psychiatry.* 37(5): 502-513.
19. *SLALAS Newsletters* – 2014;2(1&2), 2015;3(1&2), 2016;4(1) and 2017;5(1).
20. *van* Zutphen L F M (2001). Introduction. In: *Principles of Laboratory Animal Science*, 2nd Edition, Ed: van Zutphen L F M, Baumans V, Beynen A C. Elsevier Science B.V. Amsterdam, The Netherlands. pp 1-10.
21. *Wijewickrama* E S, Kurukulasooriya I, Gunatilake M, Amarathunga A H P, Gnanathasan A, Gawarammana I, Isbister G K (2018). Determination of the sub-lethal nephrotoxic dose of Russell's viper (*Daboia russelii)* venom in Wistar rats. *Toxicon*, 152: 43-45. *https://doi.org/10.1016/j.toxicon.2018.07.023*