# 4<sup>TH</sup> ASIAN CONGRESS FOR ALTERNATIVES TO ANIMAL EXPERIMENTS (4ACAAE)





Non-animal Approaches: Concept, Validation and Regulatory Acceptance



7<sup>th</sup> Annual Meeting of the Society for Alternatives to Animal Experiments – India

# SOUVENIR AND BOOK OF ABSTRACT







अनुसंधान नेशनल रिसर्च फाउंडेशन Anusandhan National Research Foundation

विज्ञान एवं प्रौद्योगिकी विभाग Department of Science & Technology









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ISAAF





https://www.jamiahamdard.ac.in/4ACAAE/

#### About 4ACAAE

The 4<sup>th</sup> Asian Congress for Alternatives to Animal Experiments (4ACAAE) and the 7th Annual Meeting of the Society for Alternatives to Animal Experiments – India (SAAE-I) will be hosted in the historical city and capital of India – New Delhi, from December 12 to 14, 2024, after a successful 3rd ACAAE held in Jeju, South Korea, in 2022. The theme of the Congress would be "Non-animal approaches: concept, validation, and regulatory acceptance". The discussion will not only be on 3Rs but beyond. It is also envisaged to provide a forum at the Congress for the global community, especially in the Asian region, for deliberations and networking. The aim is to bring to fore the need for creation of a regulatory framework for the real-time adoption of alternatives/NAMs in drug and product development and toxicology. Representatives of the Japanese Society for Alternatives to Animal Experiments (JSAAE), Korean Society for Alternatives to Animal Experiments (KSAAE) and other Asian Societies will grace the Congress.

SAAE–I (<u>https://saae-i.org/</u>) is a registered Scientific Society working in the field of alternatives to animal experiments and supports efforts for development of alternatives and their acceptance in regulatory framework through policy change.

Jamia Hamdard, New Delhi is a university under Deemed to be University category established in 1989. It is one of the top-ranking universities in the field of pharmacy. It is also known for research in the field of toxicology, biotechnology, and pharmaceutical sciences. The Department of Medical Elementology & Toxicology, School of Chemical & Life Sciences, has a Centre for Research on Alternatives producing quality research in fruit fly (*Drosophila melanogaster*) and nematode (*Caenorhabditis elegans*) models. Jamia Hamdard hosted the first Conference of SAAE-I in 2018. We feel privileged to host the fourth Asian Congress in India combined with the seventh SAAE-I conference meeting and conference.

There will be a number of Pre- and Post-congress Workshops on selected topics from the field of alternatives and NAMs with partner institutions such as CSIR-Institute of Genomics & Integrative Biology (IGIB) – New Delhi (Zebrafish), Aligarh Muslim University (*D. melanogaster* and Reconstructed Human Epidermis- RHE), CSIR-Centre for Cellular and Molecular Biology (CCMB) - Hyderabad (Zebrafish), Department of Zoology and Advanced Centre for Cutaneous Biology and Regenerative Medicine, University of Kerala - Thiruvananthapuram, and others.

#### Topics

- Current developments in alternatives
- New approach methodologies (NAMs)
- Non-mammalian model organisms
- In vitro and in silico models and methods
- Organs-on-chip and molecules-on-chip
- Alternatives research beyond 3Rs
- Regulatory framework for alternatives
- AI and machine learning in alternatives
- Education and awareness modules

# **4<sup>TH</sup> ASIAN CONGRESS FOR ALTERNATIVES TO ANIMAL EXPERIMENTS (4ACAAE)**

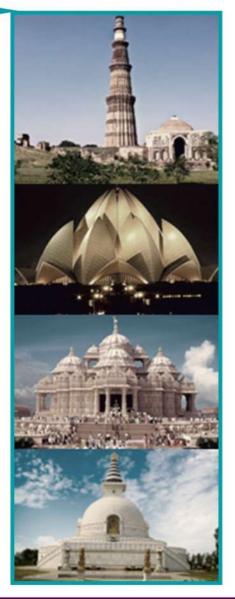




Non-animal Approaches: Concept, Validation and Regulatory Acceptance



### 7<sup>th</sup> Annual Meeting of the Society for Alternatives to Animal Experiments - India



### Topics

- $\Rightarrow$  Current developments in alternatives
- ⇒ New Approach Methodologies (NAMs)
- ⇒ Non-mammalian model organisms
- $\Rightarrow$  In vitro and in silico models and methods
- ⇒ Organs-on-chip and molecules-on-chip
- ⇒ Alternatives research beyond 3Rs
- ⇒ Regulatory framework for alternatives
- $\Rightarrow$  AI and machine learning in alternatives
- $\Rightarrow$  Education and awareness modules

PRE- AND POST-CONGRESS WORKSHOPS



!! Seeking proposals for sessions, 🦄 sponsorship and partnership !!



https://www.jamiahamdard.ac.in/4ACAAE/



SAAE - India Office Bearers: President - Prof. Y. K. Gupta; Vice Presidents - Prof. S. Raisuddin & Dr. Vijay Pal Singh; General Secretary - Prof. M.A. Akbarsha; Jt. Secretaries - Dr. A.B. Pant & Dr. Adip Roy; Treasurer - Dr. Kadalmani; International Advisors - Dr. Thomas Hartung, CAAT USA; Prof. Hajime Kojima, JaCVAM Japan; Dr. Christian Pellevoisin, MatTek France.

### 4<sup>th</sup> Asian Congress for Alternatives to Animal Experiments (4ACAAE) on Non-animal Approaches: Concept, Validation and Regulatory Acceptance

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7<sup>th</sup> Annual Conference of Society for Alternatives to Animal Experiments -India

### December 12-14, 2024 Jamia Hamdard (Deemed to be University) New Delhi 110062, India

### **PROGRAMME SCHEDULE**

Day 01 (December 12, 2024- Thursday)			
TIME	EVENT	VENUE	
8.30 am - onward	REGISTRATION	Hamdard Convention Centre	
10.00 - 11.30 am	INAUGURAL SESSION	Hamdard Convention Centre	
11.30 am - 12.00 pm	HIGH TEA/COFFEE BREAK & NETWORKING	Hamdard Convention Centre	
12.00 - 12.30 pm	PLENARY LECTURE SESSION	Hamdard Convention Centre	
12.30 - 12.45 pm	HSI- MASTERCLASS IN ANIMAL-FREE SAFETY ASSESSMENT (AFSA)	Hamdard Convention Centre	
12.45 - 1.45 pm	DR. DIPTI M. KAPOOR ENDOWMENT AWARD CEREMONY	Hamdard Convention Centre	
1.45 - 2.25 pm	LUNCH BREAK -	Hamdard Convention Centre	
2.30 pm onward	1) POSTER SESSION - I 2) MAtTek AWARD POSTER PRESENTATIONS	Hamdard Convention Centre	
2.30 - 3.00 pm	PLENARY LECTURE SESSION	Hamdard Convention Centre	
3.00 - 4.20 pm	KEYNOTE LECTURE SESSION	Hamdard Convention Centre	
4.20 -4.40 pm	TEA/COFFEE BREAK	Hamdard Convention Centre	
4.40 - 6.10 pm	SPONSORED SESSION - CENTRE FOR PREDICTIVE HUMAN MODEL SYSTEMS (CPHMS), CSIR-CENTRE FOR CELLULAR & MOLECULAR BIOLOGY (CCMB), HYDERABAD, INDIA	Hamdard Convention Centre	
6.30 - 7.30 pm	CULTURAL PROGRAMME	Hakeem Abdul Hameed Auditorium	
8.00 pm onward	CONGRESS DINNER	VIP Guest House Lawn	
	DAY 01 CLOSES		

Day 02 (December 13, 2024- Friday)			
TIME	EVENT	VENUE	
9.00 am - onward	REGISTRATION	Hamdard Convention Centre	
9.30 - 11.30 am	LAUNCHING OF THE ASIAN FEDERATION OF SOCIETIES FOR ALTERNATIVES TO ANIMAL EXPERIMENTS (AFSAAE)	Hamdard Convention Centre	
11.30 - 11.50 am	TEA/COFFEE BREAK	Hamdard Convention Centre	
11.50 am - 12.20 pm	SESSION FOR SPEAKERS FROM ASIAN COUNTRIES- I	Hamdard Convention Centre	

12.20 - 1.10 pm	SESSION FOR SPEAKERS FROM ASIAN COUNTRIES- II	Hamdard Convention Centre
11.00 am - 1.00 pm	POSTER SESSION -II	Hamdard Convention Centre
1.20 - 2.30 pm	LUNCH BREAK	Hamdard Convention Centre
2.30 - 4.00 pm	PARALLEL SESSION	Hamdard Convention Centre
	TECHNICAL SESSION 1 (ORGAN, TISSUE AND CELL-ON- CHIP)	
2.30 - 4.00 pm	PARALLEL SESSION PeTA -SPONSORED SESSION 1	Hakeem Abdul Hameed Auditorium
4.30 - 6.00 pm		Hamdard Convention Centre
	TECHNICAL SESSION 2 (IN VITRO SYSTEMS)	
4.30 - 6.00 pm	PARALLEL SESSION	Hakeem Abdul Hameed Auditorium
	PeTA - SPONSORED SESSION 2	
4.30 - 6.00 pm	PARALLEL SESSION	HILSR Ground floor
	TECHNICAL SESSION 3 (ADVANCES IN BIOTECHNOLOGY & TOXICOLOGY)	
6.00 - 6.30 pm	EC/GB MEETING OF SAAE - INDIA	Hamdard Convention Centre
DAY 02 CLOSES		

Day 03 (December 14, 2024 - Saturday)				
TIME	EVENT	VENUE		
9.00 am - onward	REGISTRATION	Hamdard Convention Centre		
9.30 -10.30 am	MatTek ORAL PRESENTATION AWARD SESSION FOR RESEARCH SCHOLARS AND POST-DOCTORAL FELLOWS	Hamdard Convention Centre		
9.30 - 10.30 am	ORAL PRESENTATIONS	Hamdard Convention Centre		
10.30 - 11.00 am	TECHNICAL SESSION 4 (IN VITRO AND MICROBIAL TECHNOLOGY)	Hamdard Convention Centre		
11.00 - 11.20 am	TEA/COFFEE BREAK	Hamdard Convention Centre		
11.30 am- 1.00 pm	PANEL DISCUSSION (TOPIC - VALIDATION AND REGULATORY SCENARIO AND ALTERNATIVES FOR DEVICE TESTING)	Hamdard Convention Centre		
1.00 - 2.00 pm	PARALLEL SESSION TECHNICAL SESSION 5 (IN SILICO SYSTEMS, AI AND ML IN ALTERNATIVES RESEARCH)	Hamdard Convention Centre		
1.00 - 2.00 pm	PARALLEL SESSION TECHNICAL SESSION 6 (ZEBRAFISH AND OTHER VERTEBRATES)	HILSR Ground Floor		
1.00 - 2.00 pm	PARALLEL SESSION TECHNICAL SESSION 7 (BIOENGINEERING, TISSUE ENGINEERING AND 3D BIOPRINTING)	Hakeem Abdul Hameed Auditorium		
2.00 - 2.30 pm	LUNCH BREAK	Hamdard Convention Centre		

2.30 - 4.30 pm	PARALLEL SESSION	Hamdard Convention Centre	
	TECHNICAL SESSION 8 (INVERTEBRATE ALTERNATIVES)		
2.30 - 4.30 pm	PARALLEL SESSION	HILSR - Ground Floor	
	TECHNICAL SESSION 9 (ANIMAL WELFARE AND OTHER ALTERNATIVES)		
2.30 - 4.30 pm	PARALLEL SESSION	Hamdard Archives Auditorium	
	TECHNICAL SESSION 10 (METABOLOMICS, BIOINFORMATICS, TOXICOINFORMATICS AND NETWORK PHARMACOLOGY)		
4.30 - 5.30 pm	VALEDICTORY SESSION AWARD DECLARATION AND DISTRIBUTION	Hamdard Convention Centre	
	Declaration of the Next Venue for the 5 <sup>th</sup> Asian Congress		
5.30 pm	TEA/COFFEE	Hamdard Convention Centre	
CURTAINS DOWN			

**Note:** Last-minute changes are possible. In case of revision, an advance notice will be sent and the revised programme will be uploaded on the Congress Portal.

Organizing Committee - 4ACAAE



### Prof. (Dr.) M. Afshar Alam Vice Chancellor Ph.D

### Jamia Hamdard

(Deemed to be University), 'A+' Category-NAAC Hamdard Nagar, New Delhi-110 062, India Tel. (O) +91-1126059662 / 3, (R) 011-29966650 Cell : +91-9891971999 Email: vc@jamiahamdard.ac.in ; aalam@jaimahamdard.ac.in Website: www.jamiahamdard.edu

6<sup>th</sup> December, 2024

#### MESSAGE

I am happy to know that the Department of Medical Elementology & Toxicology, School of Chemical & Life Sciences and Office of the Director, R&D Cell are hosting the 4<sup>th</sup> Asian Congress for Alternatives to Animal Experiments (ACAAE) and the 7<sup>th</sup> Annual Meeting of the Society for Alternative to Animal Experiments – India (SAAE-I) on December 12-14, 2024. It is a matter of pride that the global scientific community has selected Jamia Hamdard as the venue for this Congress.

I, on my behalf, and as the Head of the Institution welcome delegates from India and abroad to this Congress. I would also like to thank various sponsors, especially Hamdard National Foundation India-HECA for sponsoring the event.

I must appreciate the efforts of the Organizing Committee for preparing a very impressive programme encompassing plenary lectures, keynote lectures, invited lectures and oral and poster presentations.

I hope that after the deliberations at the Congress some recommendations will come out for the policymakers with respect to annual welfare, animal alternatives and intervention of technology to reduce and finally eliminate the use of animals in biomedical and toxicological research.

I wish all the success to the 4ACAAE Organizing Committee.

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Prof. M. Afshar Alam Vice Chancellor

### SOCIETY FOR ALTERNATIVES TO ANIMAL EXPERIMENTS-INDIA (SAAE-I)



National Centre for Alternatives to Animal Experiments Bharathidasan University, Tiruchirappalli 620024, India Registration No. SRG/Trichy/10/2019

Website: www.isaae-i.org; E.mail address: saae.india@gmail.com;

President	Vice-Presidents	<b>General Secretary</b>	<b>Joint Secretaries</b>	Treasurer
Prof. Y.K. Gupta	Prof. S. Raisuddin	Prof. M.A.	Dr. A.B. Pant	Dr. B. Kadalmani
	Dr. Vijay P. Singh	Akbarsha	Dr. Adip Roy	



#### **PRESIDENT'S MESSAGE**

I, as the President of the SAAE-India, and my own behalf welcome delegates to the 4<sup>th</sup> Asian Congress for Alternatives to Animal Experiments (4ACAAE) - Nonanimal Approaches: Concept, Validation and Regulatory Acceptance (December 12-14, 2024). The Congress will be held concurrently with the 7<sup>th</sup> Annual Conference of the Society. I thank the Vice Chancellor of Jamia Hamdard for accepting our request to host the Asian Congress. I also like to place on record deep sense of appreciation for Prof. S. Raisuddin, the Organizing Secretrary of 4ACAAE. He has been tirelessly working for making various arrangements for the 4ACAAE.

During the Congress there will be a wide-ranging discussion on animal welfare, animal alternatives and new approach methodologies (NAMs). The interest shown by Members of the Asian Societies working in the field of alternatives has been overwhelming. A number of sponsors from India and abroad have also come forward to support the Congress and its objectives. I thank them all.

I hope at the end of the Congress there will some meaningful outcome which will shape the future direction of research and regulation with regard to use of NAMs and non-animal approaches for the evaluation of safety and efficacy drugs, chemicals and devices.

Wishing all the best to the Organizing Committee of the 4ACAAE.

(Prof. Y.K. Gupta) PRESIDENT SAAE-I

### SOCIETY FOR ALTERNATIVES TO ANIMAL EXPERIMENTS-INDIA (SAAE-I)



National Centre for Alternatives to Animal Experiments Bharathidasan University, Tiruchirappalli 620024, India Registration No. SRG/Trichy/10/2019

Website: www.isaae-i.org; E.mail address: saae.india@gmail.com;

President	Vice-Presidents	<b>General Secretary</b>	<b>Joint Secretaries</b>	Treasurer
Prof. Y.K. Gupta	Prof. S. Raisuddin	Prof. M.A.	Dr. A.B. Pant	Dr. B. Kadalmani
	Dr. Vijay P. Singh	Akbarsha	Dr. Adip Roy	



#### **GENERAL SECRETARY'S MESSAGE**

Dec 04, 2024

Dear Prof. Raisuddin

It is nostalgic to recollect the several 'firsts' in of the history of Society of Alternatives to Animal Experiments-India happening at Jamia Hamdard. The first meeting of the Executive Committee of SAAE-I was held at Jamia Hamdard in 2018 when the byelaws of the Society were discussed and finalized. The first Conference of the Society was organized in 2018 at Jamia Hamdard, under your seasoned leadership, when several scientists from abroad were also present. Now the seventh conference is organized here, which is also the Fourth Asia Congress, but first time it is organized in India. More importantly, the Asian Federation of the Societies of Alternatives is going to be launched at Jamia Hamdard on December 13, 2024, which is another milestone. The Society thanks Jamia Hamdard-you, your colleagues and the Administration- for all good it has done for SAAE-I.

I wish the forthcoming events of SAAE-I and the Asian Federation of Societies for Alternatives for Animal Experiments all the very best.

Sincerely

Dx. Abarl

General Secretary, SAAE-I







(Deemed to be University)

**Prof. (Dr.) Suhel Parvez** (Humboldt Fellow & FRSB) Department of Medical Elementology & Toxicology Dean, SCLS, Jamia Hamdard Coordinator, DST-PURSE & STUTI



### MESSAGE

The Society for Alternatives to Animal Experiments (SAAE) was established in January 2019 at Bharathidasan University in Tamil Nadu with the mission of advancing humane and innovative scientific practices. Over the years, SAAE has grown into one of India's most dynamic scientific societies, bringing together a diverse community of researchers, educators, and policymakers. By fostering dialogue and collaboration, SAAE aims to accelerate the adoption of alternative methodologies that reduce or replace the use of animals in scientific research. Its annual meetings, hosted in different parts of the country, serve as a vibrant platform where scientists can share breakthroughs, exchange ideas, and enhance their understanding of alternative models in fields such as drug discovery, biomedical research, and toxicity testing.

At the forefront of this movement, our laboratory has made significant contributions by developing and refining a range of alternative research models. These include using the nematode *Caenorhabditis elegans*, the fruit fly *Drosophila melanogaster*, chick embryos, and advanced cell culture systems. These models represent ethical, cost-effective, and scientifically robust alternatives to traditional animal testing, offering valuable insights into biological processes and toxicological responses. Through rigorous experimentation and validation, our work not only meets global research standards but also contributes to the overarching goal of making science more sustainable, humane, and relevant to human biology.

This abstract book highlights the collective effort of researchers and contributors dedicated to advancing innovation in the field of alternative models. It captures the breadth and depth of ongoing research, highlighting the creative solutions and advancements being made to address critical challenges in regulatory acceptance, standardization, and interdisciplinary collaboration. These contributions reflect the global momentum towards adopting non-animal methodologies and the increasing recognition of their value in providing more accurate and human-relevant data.

We are deeply grateful to the many individuals who have contributed to this initiative—the authors who submitted their valuable research, the reviewers who ensured the quality of the content, and the organizers who made this event possible. We also extend our sincere thanks to the institutions and organizations that supported this endeavour, enabling us to provide a meaningful platform for knowledge exchange and collaboration. It is our hope that this abstract book serves as both a record of the progress being made and an inspiration for future innovations in the field of alternatives to animal experimentation. Together, we move closer to realizing a vision of science that is both ethically responsible and scientifically rigorous.

**Professor (Dr.) Suhel Parvez** Dean School of Chemical and Life Sciences



### **MESSAGE FROM THE CHAIR**

### ASIAN FEDERATION FOR SOCIETIES FOR ALTERNATIVES TO ANIMAL EXPERIMENTS (AFSAAE)

Dear colleagues and delegates,

Congratulations on the fourth Asian Congress (4ACAAE) kindly organized by the SAAE-I and welcome to all the Asian and other countries' colleagues!

In addition to the well as the well-organize scientific sessions, symposium/workshop, and satellite meetings as can be seen in the program, this fourth Asian Congress is particularly important because we are about the establish the Asian Federation of the Societies for Alternatives in Animal Experiments (AFSAAE). This is really a significant milestone for broadening the 3Rs concept in Asian countries, thus, this fourth congress should be a historically memorable one when we think about the past, present, and future of the 3Rs in Asian region. I remember that the idea to organize Asian colleagues in alternative research was first proposed by Dr. Hajime KOJIMA at JSAAE. This proposal has been increasing the number of endorsements, leading to the successful holding of four Asian Congresses for Alternatives in Animal Experiments (ACAAE) up to now; the first one was in 2016 in Japan by organized by Dr. KOJIMA/JSAAE, the second was 2018 in China by Dr. Shuangqing PENG/TATT-CSOT, third one was in 2022 in Korea by Dr. Kwang-Man KIM/KSAAE), and then this fourth one in 2024 is in India by Dr. Sheikh RAISUDDIN/SAAE-I.

Before the third Asian Congress in Korea in 2022, we started preparation meetings toward the establishment of the AFSAAE. Eventually, the four societies of KSAAE, JSAAE, SAAE-I and TATT-CSOT successfully reached an agreement of the establishment of the AF. These four founding societies have a key responsibility both in the organization/management of the AF and broadening the 3Rs concept in Asia with corporations with colleagues in other Asian countries, all of which are strongly expected to join the AF.

As you are all aware, there are region-specific issues in the Asian countries in broadening the 3Rs concept. Such issues are often rooted in the diverse cultural, legislative, and administrative contexts, which are different from the those in European and American regions where the 3Rs concept was first proposed. First sharing such issues, identifying the causes and then finding the solutions are strongly required as the mission of the AF. I believe such efforts may also contribute to the enrichment of the 3Rs concept over the world.

Finally, I wish the success of the fourth the Asian Congress through the communication among colleagues not only in Asian regions but also from western countries. Also, I especially would like to thank Indian colleagues and SAAE-I for the great contributions to this memorable congress!

Dr. Yasuyuki SAKAI Scheduled to become the chair of the Asian federation of the Asian Federation of Societies for Alternatives to Animal Experiments (AFSAAE)

Director of International Affairs, JSAAE Professor at Graduate School of Engineering, University of Tokyo, Japan





### **MESSAGE FROM THE SECRETARY GENERAL**

### ASIAN FEDERATION FOR SOCIETIES FOR ALTERNATIVES TO ANIMAL EXPERIMENTS (AFSAAE)

"I am extremely delighted to have this opportunity to declare the establishment of the Asian Federation of Alternatives to Animal Experiments, with the aim of developing and promoting the 3Rs (Replacement, Reduction, Refinement) of alternative methods to animal experiments in Asia. The Asian Federation is a new cooperative framework involving the academic organizations of TATT, SAAE-I, JSAAE, and KSAAE, which is expected to be a collaboration ground.

We held the first Asian Congress in Japan in 2016, the second in China in 2018, the third in Korea in 2022, and the fourth in India in 2024. Through the Asian Congress meetings, we have come to the idea of establishing the Asian Federation for further collaborations among scientists in this field of study to start this project of the Asian Federation.

The first Prep Meeting was held in May 2022. Since then, eight prep meetings have been held, and although it took nearly three years to make the final preparations, we have eventually achieved the establishment of the Asian Federation. I hope that this Asian Federation will promote the exchange of scientists in Asia and lead to new innovations.

We have just started the Federation. I see a light of innovation far away on the top of the mountain. We still don't know how to get there. But I believe the light will guide us if we don't miss its brightness.

Now, the time.

Let's hit the road."

Best regards,

Matt



#### Dr. Masato Hatao

Principal Scientist/Managing Director - Japan Cosmetic Industry Association (JCIA), Tokyo, Japan Secretary General Designate - Asian Federation of Societies for Alternative Animal Experiments (ASFSAAE)

### MESSAGE

First of all, I would like to sincerely congratulate the 4<sup>th</sup> ACAAE conference held in New Delhi, India, as the president of KSAAE.

I believe this conference is very meaningful in that it is beginning of "The Asian Federation of Societies for Alternatives to Animal Experiments".

Now the paradigm of safety assessment is changing significantly and interest in alternative testing methods to replace animal testing is increasing worldwide.

I am confident that this federation among Asian countries is truly necessary and will open a new era.

I hope cooperative system among Asian countries will continue well, and it can influence beyond Asia to the world in the future.

Lastly, I would like to express my deepest gratitude to the Indian scientists who prepare the conference.





Bae-Hwan Kim DVM, PhD, President of KSAAE Professor, Keimyung University

Baettwan Kim

### **Goodwill Message**



It is with great admiration and respect that I extend my warmest greetings to all participants, organizers, and supporters of the **4th Asian Congress of Alternatives to Animal Experiments (4ACAAE)** and the opening of **Asian Federation for Alternatives to Animal Experiments**. This gathering symbolizes a shared commitment to advancing ethical, innovative, and scientifically robust approaches in research and testing across Asia.

The pursuit of alternatives to animal experimentation is not just a scientific necessity but a profound ethical imperative. Through the

development of cutting-edge methodologies such as in vitro models, computational simulations, and organ-on-chip technologies, we can address global challenges in medicine, toxicology, and environmental science while fostering compassion for all living beings.

This congress provides a vital platform for knowledge exchange, collaboration, and the cultivation of shared values among nations. Our efforts will inspire future generations of researchers and policymakers to continue forging a path that balances scientific progress with humane practices.

May this event be a resounding success, inspiring meaningful dialogue and paving the way for transformative advancements in our shared mission.

With my heartfelt wishes for a productive and impactful congress,

Ki-Suk Kim, Ph.D Principal Research Scientist / Secretary General Korea Institute of Toxicology / Korean Society of Alternatives to Animal Experiments (KSAAE)





#### SOCIETY FOR ALTERNATIVES TO ANIMAL TESTING IN SRI LANKA (SAAT-SL)

Department of Physiology, Faculty of Medicine, Kynsey Road, Colombo 08, Sri Lanka Estd : February 2021

President: Dr. Kalpani Ratnayake Department of Cosmetic Science, Faculty of Health Sciences, CINEC Campus Tel: +94 771658384

kalpani ratnayake@cinec.edu. kratnayake1984@gmail.com

President-Elect: Dr. Prasadi N. De Silva UVA Wellassa University of Sri Lanka

Honorary Secretary: Dr. Varuni Gunathilake Faculty of Applied Sciences, University of Sri Jayewardenepura Tel: +94 714478801

#### varunig@sjp.ac.lk

Honorary Treasurer: Dr. Sachim Amaradekara Faculty of Science, University of Colombo Tel: +94 705694265

sachim@zoology cmb.ac.lk

Assistant Secretary: Dr. Banukie Jayasunya University of Sri Jayewardenepura

Assistant Treasurer: Dr. Ureshani Karunarathne University of Sri Jayewardenepura

Editor: Ms. Chedliya Ishak Human Genetics Unit, Faculty of Medicine, Colombo

Tel: +94 776854155

#### Executive Committee members:

Prof. Mangala Gunatilake University of Colombo

Prof. Sugandhika Suresh University of Sri Jayewardenepura

Prof. Swama Hapuarachchi University of Colombo

Dr. Mayuri Thammitiyagodage Medical Research Institute

Dr. Ramani Karunakaran Medical Research Institute

Dr. Saroja Weerakoon University of Colombo

Dr. Hasanthi Rathnadiwakara University of Colombo

Dr. Tharindi Prasadinie University of Peradeniya Message from the President of the Society for Alternatives to Animal Testing in Sri Lanka (SAAT-SL)



Dr. Kalpani Ratnayake President SAAT- SL

On behalf of the Society for Alternatives to Animal Testing in Sri Lanka, I extend my heartfelt congratulations to the organizers and participants of the 4<sup>th</sup> Asian Congress for Alternatives to Animal Experiments (4ACAAE). This prestigious event marks a significant step toward advancing ethical, innovative, and humane approaches in scientific research.

The collective efforts of experts, researchers, and policymakers at this congress exemplify the spirit of collaboration needed to promote the 3Rs; Replacement, Reduction, and Refinement and pave the way for groundbreaking innovations in alternatives to animal testing.

May this congress inspire meaningful discussions, ignite new partnerships, and lead to transformative progress in the field. Best wishes for a successful and impactful event!

Warm regards,

Matnayate

Dr. Kalpani Ratnayake President SAAT-SL



# **4ACAAE**



4th Asian Congress for Alternatives to Animal Experiments (4ACAAE)

Non-animal Approaches: Concept, Validation and Regulatory Acceptance 7th Annual Meeting of the Society for Alternatives to Animal Experiments - India

### December 12-14, 2024 | Jamia Hamdard (Deemed to be University) | New Delhi 110062, India

Patron Mr. Hammad Ahmed Chancellor, Jamia Hamdard

**Co-patron Prof. (Dr) M. Afshar Alam** Vice Chancellor, Jamia Hamdard

#### **Organizing Committee**

#### Chairman

Prof. S. Raisuddin, Dean, School of Chemical & Life Sciences

#### Co-chairman

Prof. H.A. Khan, Head, Department of Medical Elementology & Toxicology

Organizing Secretary Prof. S. Raisuddin

Joint Organizing Secretary Prof. Suhel Parvez, Department of Medical Elementology & Toxicology

**Treasurer** Dr. Nidhi, Department of Translational & Clinical Research

### WELCOME MESSAGE

I am very pleased to welcome delegates from various countries to the 4<sup>th</sup> Asian Congress for Alternatives to Animal Experiments (4ACAAE) - Non-animal Approaches: Concept, Validation and Regulatory Acceptance. It was a challenging task given the fact that response was very strong. A matching support I got from all the quarters be it at Jamia Hamdard or others from India and abroad. The Congress will be historic as there will be a Ceremony for Declaration of formation of the Asian Federation for Societies for Alternatives to Animal Experiments (AFSAAE). The declaration will be witnessed by not only Asian Society representatives but esteemed representatives of European and American organizations working in the field of alternatives.

I like to place on record my gratitude to the Vice Chancellor, Prof. M. Afshar Alam for his support throughout. Thanks are also due to the generous sponsorship, especially from the Hamdard National Foundation (HNF)-HECA, Anusandhan National Research Foundation (ANRF), DST, Government of India, Humane Society International (HIS), Doerenkamp Zbinden Foundation (DZF), People for Ethical Treatment of Animals (PeTA), and Japanese Society for Alternatives to Animal Experiments and from industry such as J-TEC, Amway, MatTek, NBIL, Eurofins Advinus, and MyCalpharm.

The Organizing Committee has prepared a packed scientific programme comprising of Plenary Lectures, Keynote Lectures, Invited Lectures and Oral and Poster Presentations, Panel Discussion and Special Sessions (PeTA and CPHMS). Scientifically it will be relishing.

Looking forward to meeting at the 4ACAAE.

S. Kainell

Prof. S. Raisuddin Organizing Secretary - 4ACAAE





# ABSTRACTS

#### PLENARY LECTURES

#### Toward animal-free science globally

#### **Troy Seidle**

Vice President, Research and Toxicology, Humane Society International (HSI), Washington, District of Columbia, USA tseidle@hsi.org



Troy Seidle leads a global team of experts who work with governments, companies, research funding bodies, scientists, and public interest stakeholders to promote acceptance of modern, animal-free approaches to testing and research. With three decades experience in biomedical and toxicological science policy, he possesses extensive knowledge of current and emerging testing and research methodologies and of legal and regulatory frameworks across many different countries and sectors. Seidle's contributions to political negotiations led to an unprecedented 50% reduction in animal test requirements for pesticides and biocides in Europe, which earned HSI a LUSH Prize. He is the chief architect behind the Animal-Free Safety Assessment (AFSA) Collaboration, a forum for stakeholder dialogue and cooperation toward a common vision of a not-so-distant future without animal testing. He has provided expert testimony to governments across the globe and served on numerous high-level policy committees, including the Organisation for Economic Co-operation and Development, the European Union and the U.S. Environmental Protection Agency. Seidle started early as an animal advocate: At 18, he was the youngest person to serve on the Canadian Council on Animal Care and witness first-hand the plight of animals used in science.

PL-2

# Global progress through collaboration: The role of validation projects in advancing NAMs in toxicology and related sciences

#### Dr. rer. nat., Ing. Helena Kand'árová, ERT

Director of the Institute of Experimental Pharmacology and Toxicology, Member of the Executive Board – CEM Centre of Experimental Medicine SAS Institute of Experimental Pharmacology & Toxicology, Bratislava, Slovak Republic <u>helena.kandarova@savba.sk</u>



Dr. Helena Kand'árová is the Director and Senior Scientist at the Institute of Experimental Pharmacology and Toxicology at the Centre of Experimental Medicine, Slovak Academy of Sciences in Bratislava. She also serves as an Assistant Professor at the Institute of Biochemistry and Microbiology, Faculty of Chemical and Food Technology, Slovak University of Technology. With over two decades of experience, her research focuses on the development, validation, and regulatory implementation of in vitro methods to replace animal testing in toxicology, particularly using 3D reconstructed human tissue models. Dr. Kand'árová has played a pivotal role in

international projects that led to the adoption of alternative testing protocols into OECD Test Guidelines and ISO standards. She has authored more than 80 scientific papers and book chapters. Her contributions have been recognized with several prestigious awards, including the Doerenkamp-Zbinden Foundation Award (2021), the Björn Ekwall Memorial Award (2022), and the EUROTOX Lecture Award (2022). In addition to her research, Dr. Kand'árová holds leadership positions in various scientific organizations. She is the President of the European Society of Toxicology In Vitro (ESTIV), Vice-President of the Slovak Toxicology Society (SETOX), Chair of the Slovak National Platform for 3Rs (Replacement, Reduction, and Refinement of animal experiments) and member of EPAA MG. Her dedication to advancing humane and scientifically robust testing methods is reflected by her pro-bono work in the OECD expert panels and national committees on NAMs and medical device testing.

#### **KEYNOTE LECTURES**

# Toward the development of physiological in vitro liver tissues for toxicity testing and disease modelling

#### Yasuyuki Sakai<sup>1</sup>, Hiroshi Kimura<sup>2</sup>, Masaki Nishikawa<sup>1</sup>

<sup>1</sup>Department of Chemical System Engineering, Graduate School of Engineering, University of Tokyo, Tokyo 113-8656, Japan <sup>2</sup>Micro/Nano Technology Center, Tokai University, Hiratsuka 259-1292, Japan sakaiyasu@chemsys.t.u-tokyo.ac.jp



His current research topics are development of microphysiological systems (MPS), engineering of implantable 3D tissues/organs, and large-scale culture of stem cells. He has been placing particular importance on realization of good mass transfers and 3D organization of cells in vitro. During his research carrier, he published over 250 original publications. He got several scientific awards such as Young Investigator Award of Society of Chemical Engineers, Japan, Publication Awards of Society for Bioscience and Bioengineering, Japan and Japanese Society for Alternatives to Animal Experiments (JSAAE). He became a fellow of American Institute for Medical and Biological Engineering (AIMBE) from 2012 for his outstanding contributions to tissue engineering and cell-based assays. He is a Guest Professor at Technological University of Compienge, France from 2019. He is also a Fellow of Society for Chemical Engineers, Japan, from 2021 and a Member of the Engineering Academy of Japan from 2022. He is working as an Editorial Board Member of Biofabrication, Bio-Design and Manufacturing (BDM) and Frontiers in Toxicology (in vitro toxicology), and Health Engineering.

#### Abstract

Toward development of physiologically in vitro liver tissue for efficacy/safety testing and disease modeling, we need to integrate various pericellular microenvironments using latest technologies and knowledge; they are concerning stem cell-derived mature organ cells, 3D hierarchical organization of organ parenchymal cell, nonparenchymal cells and extracellular matrices, local communications via local paracrine and autocrine factors, good oxygen/nutrient supply, wastes removal, physiological culture medium, and mechanical stimulations etc. With such fundamental problem consciousness toward ultimate-physiological in vitro tissue models, we would like to introduce our latest investigations such as 1) new static heterogenic liver tissue culture and 2) microphysiological system (MPS) comprising liver and small intestine tissues supported by the MPS Project of Japanese Agency of Medical Research and Development (AMED). Those are both based on direct oxygenation of the cells using oxygen-permeable membranes to completely solve the unphysiological insufficient oxygen supply (Front. Toxicol., 2022). This problem has been historical, forgotten but still unsolved in current cell culture formats. Use of oxygen-permeable membrane easily resolves the problem and allows cells to show their unprecedented spontaneous organization, high functionality or enhanced organ-to-organ crosstalk (PNAS Nexus, 2024) through in vivo-like aerobic respiration and in vivo-mimicking hierarchical 3D coculture. In particular, 3D heterogenic multilayered tissue can be called as "Open Organoid" and are better compatible with MPS perfused formats for observing organ-to-organ interactions. In addition, we

would like to discuss the integration of such physiological in vitro cell culture models with the systems biology model (Front. Pharmacol., 2022) and/or agent-based model (Sci. Rept., 2022) toward understanding the systemic responses of humans.

KL-1

#### Learning by Doing: Gaining confidence in new approaches for regulatory decisionmaking and risk assessment

Kristie Sullivan, MPH Institute for In Vitro Sciences, Gaithersburg, Md, United States ksullivan@iivs.org



Ms. Sullivan joined the IIVS team in 2023 to lead its Education and Outreach programs which are designed to advance the IIVS mission. These programs include organizing workshops and trainings to advance acceptance and use of New Approach Methodologies (NAMs), engaging with toxicologists and stakeholders to develop educational materials and resources, and acting as an information resource to industry, government, and the animal protection community. Prior to joining IIVS, Ms. Sullivan served as Vice President for Research Policy at the Physicians Committee for Responsible Medicine, where she led a team charged with developing, promoting, and implementing New Approach Methodologies (NAMs) for the testing of drugs, chemicals, and other products. She has 20 years' of experience advancing NAMs through scientific scholarship, validation, policy advocacy, and outreach. Ms. Sullivan has served on a number of OECD committees related to specific animal testing and NAM issues, and has provided advice and training to state and federal agencies, including as a past member of the Pesticide Program Dialog Committee and *ad hoc* FIFRA Science Advisory Panel and Board of Scientific Counselors reviews. She is currently a member of the executive boards of the Society for the Advancement of Adverse Outcome Pathways, the Society of Toxicology In Vitro and Alternative Methods Specialty Section, and the American Society for Cellular and Computational Toxicology, of which she is a founding member, and the editorial board of Computational Toxicology and Frontiers in In Vitro Toxicology.

#### Abstract

The use of toxicological test methods that do not use animals, but rather rely on computational predictions or in vitro methods, (New Approach Methods, NAMs) is becoming more common worldwide. Currently, NAMs are available and accepted by regulatory agencies for many endpoints, including skin and eye irritation and skin sensitization. However, many more tools are available, including high-content and high-throughput assay batteries, assessment of absorption, distribution, metabolism, and excretion (ADME) behavior, physiologically-based kinetic modeling, mechanistically- driven testing strategies, and similar Integrated Approaches to Testing and Assessment (IATA). These approaches can be used within a Next-Generation Risk Assessment (NGRA) framework to assess the potential risks for systemic toxicity from chemicals and products. This presentation will discuss efforts to implement modern NAM tools into chemical risk assessment through case studies, validation, and good practices.

# Importance of standardization in animal replacement: Success story of local toxicity endpoints and challenges for systemic toxicity

#### **Christian Pellevoisin**

Scientific Director, MatTek, USA & CEO Urbilateria, France <u>cpellevoisin@urbilateria.com</u>



Christian Pellevoisin is a European Registered Toxicologist (ERT) with a PhD in neuroscience. He is Scientific Director for Mattek and a consultant on toxicology and the effectiveness of NAMs in meeting regulatory requirements. Over the last 20 years, he worked on the development, validation and application of alternative in vitro methods to animal testing. During his career he participated in projects leading to the introduction of in vitro models and methods for the evaluation of product safety and efficacy in various industries, including cosmetics, medical devices and drugs. As chairman of the French AFNOR commission for biocompatibility of medical devices and convenor of ISO/TC-194 working group 8, he has been involved in and directed projects leading to the publication of ISO standards for skin irritation and sensitization (ISO 10993-10, 10993-23, 11796). He is also a member of the OECD expert groups on skin sensitization and on define approaches. Strongly committed to the recognition of alternative methods to animal testing, he created the module "Tissue Engineering and predictive toxicology" at Sorbonne Science University to raise students' awareness to NAMs challenges. He is to the board of the Indian Society for Alternatives to Animal Experiments (SAAE India) and of Adebiotec, a French thinktank on biotechnologies. He authored and co-authored of several scientific publications and book chapters on in vitro methods for safety and efficacy testing.

#### Abstract

Twenty years ago, a new paradigm in toxicology began to take shape for replacement of the animal by in vitro, in silico and in chemico approaches. Alongside scientific and technological advances, organizations like OECD, ISO, or ICH have been instrumental in realizing this new paradigm in toxicology, by providing harmonized guidelines, test methods, and reporting frameworks that facilitate the adoption of alternative approaches and ensure their regulatory acceptance across different countries and sectors. These standardization bodies facilitate collaboration between academia, industry, and regulatory agencies to develop consensus-based standards that ensure reproducibility, comparability, and reliability. Local toxicity endpoints, such as skin and eye irritation, or skin sensitization have successfully transitioned to in vitro methods due to the development and validation of standardized test methods through international collaboration. However, replacing animal testing for systemic toxicity endpoints presents significant challenges due to the complex interactions between different organ systems. The development of new test systems, spheroid, organoid or organotypic 3D models of these organs are promising strategies, but standardization is still in progress. Despite these challenges, progress is also made through organon-a-chip technologies, advanced in silico models, and the integration of omics data. Standardization bodies such as OECD, ISO, and ICH will again play a crucial role in addressing the complex challenges of future regulatory acceptance of such approaches for systemic toxicology. The success of local toxicity endpoints demonstrates the critical role of standardization in advancing animal replacement methods. Continued international collaboration for the development of international standards and guidelines will be key to overcoming challenges in regulatory acceptance of non-animal-based systemic toxicity testing, ultimately establishing comprehensive, animal-free methods for reliable human toxicity prediction.

KL-3

#### Overview about recent European 3Rs activities with a focus on 3Rs Centers

#### Prof. Dipl.-Ing. Dr. Winfried Neuhaus,

Austrian Institute of Technology GmbH, Center Health and Bioresources Competence Unit Molecular Diagnostics, Giefinggasse, Wien, Austria; President, EUSAAT winfried.neuhaus@ait.ac.at



The research group of Prof. Neuhaus is working on the development and validation of in-vitro models for drug transport studies and disease modeling (stroke, traumatic brain injury, Alzheimer's disease, inflammation). They are also investigating relevance and role of different transport routes (passive transport, active transport/efflux mediated by ABC-transporters, pinocytosis), and the role of the microenvironment (cells such as astrocytes) and shear stress mediated by blood flow on barrier properties in physiologic and disease models. His group is also exploring in silico-in vitro-in vivo correlations of drug transport.

#### Abstract

The talk will give an overview about the current development of the 3Rs field in Europe. The development and growth of 3Rs centres in Europe is tightly linked to political activities. One of the lighthouse project is the COST Action IMPROVE. The COST Action IMPROVE was developed from a bottom-up approach initiated by members of European 3Rs centres and its network platform EU3Rnet. COST Actions are European projects that promote the formation of networks. This activity will be presented in detail with its possibilities, plans and instruments. The working groups focus on Quality and Translatability of Science, Implementation, Dissemination and Education, whereby ethics will be an integral part in the work of the single working groups. Currently, over 230 participants reflecting several stakeholder groups take part in the activities of this project. Cooperation and active involvement in the COST Action will be invited within the framework of this open bottom-up network approach, also with international partners. Further information can be found via following link: <a href="https://costimprove.eu/">https://costimprove.eu/</a>

KL-4

# DR. DIPTI M. KAPOOR ENDOWMENT AWARD ORATION BY THE WINNER OF 2024

# Advancing lung disease research: Innovative alternative-to-animal models for testing anti-cancer and anti-infection therapies

#### Dr. Prajakta Dandekar Jain

UGC Assistant Professor, Dept. of Pharmaceutical Sciences and Technology, ICT, Mumbai <u>pd.jain@ictmumbai.edu.in</u>



Dr. Prajakta Dandekar is a UGC Assistant Professor at the Department of Pharmaceutical Sciences and Technology, Institute of Chemical Technology, Mumbai, India. Her research team mainly focuses on polymeric nanocarriers for drug and gene delivery, developing preclinical cellular models for evaluating biopharmaceuticals, 3D cell models, and tissue engineering.

#### Abstract

Lung diseases, including cancer and infections, pose a serious global threat due to limited understanding of their underlying complex biological processes, which often result in high mortality rates. Lung cancer is a leading cause of cancer-related deaths, while lung infections such as COVID-19 have recently caused worldwide havoc. These challenges can be overcome through the development of newer therapeutics and therapies, the development of which currently relies on animal models that may not most accurately replicate these conditions. Thus, advanced *in vitro* models simulating these conditions are urgently needed to develop effective therapies. These models more accurately replicate human lungs and disease progression than the traditional 2D cell cultures and animal models, allowing for accurate understanding and faster screening of potential treatments. We have developed a spheroid model of lung adenocarcinoma using ultra-low attachment (ULA) plates to mimic the tumour microenvironment. This model was characterized by high-content imaging of epithelial-mesenchymal transition (EMT) markers and hypoxic conditions. Drug sensitivity was evaluated by imaging flow cytometry, which demonstrated the effectiveness of spheroids in replicating therapeutic responses. Migration and angiogenesis assays demonstrated enhanced proliferation and vasculature formation, respectively, imitating tumor behaviour. The model also exhibited trait of mesenchymal-epithelial transition (MET), providing valuable insights into the progression of lung adenocarcinoma for advanced biological research and drug development.

Furthermore, we have also developed a lung-on-a-chip (LoC) model using a microfluidic device to study COVID-19 infections and test newer therapies against this and similar deadly infections. This triple co-culture model, created by combining microfluidic systems and bioprinting, was designed to replicate the proximal lung environment, the primary site of COVID-19 and other infections. Characterisation with respect to cellular and disease-related biomarkers and validation trails with model drugs proved that the model was an excellent alternative-to-animal method for effective screening of anti-viral and anti-inflammatory therapies.

#### **INVITED LECTURES**

# Empowering Predictive Toxicology: Past, Present, and Future of Alternative Testing Methods in Korea

#### Seokjoo Yoon

Korea Institute of Toxicology, Daejeon, Republic of Korea President-elect, KSAAE 2-25-2026 sjyoon@kitox.re.kr



Dr. Seok Joo Yoon is a Principal research scientist of the Korea Institute of Toxicology (KIT). At the Korea Institute of Toxicology, he contributed to set up a research program in the toxicogenomics field. He worked at NCCT, US-EPA as a visiting scientist. He is also appointed Professor of human and environmental toxicology at the Korea University of Science and Technology (UST). He has authored or co-authored more than 100 publications. He was also served as the Director of the department of predictive toxicology and vice president of KIT. It is here that he led the research to develop alternatives to animal study, biomarkers, human pluripotent stem-cell-derived hepatocytes & cardiomyocytes, zebrafish for drug screening, 3D cell culture, organoid and in silico toxicology. He is also contributing as an expert in ISO TC229/WG3 (Nanotechnology).

#### Abstract

This talk covers Korea's transformative journey in developing and implementing alternative testing methods, marking a fundamental shift from conventional animal testing to advanced predictive toxicology approaches. The narrative encompasses three distinct phases: historical development, current status, and future prospects. Koreas commitment to alternative testing methods began taking shape in the early 2000s, initially focusing on adopting internationally validated methods. A watershed moment occurred in 2009 with the establishment of the Korean Center for the Validation of Alternative Methods (KoCVAM). This institution has since played a pivotal role in validating new testing approaches and aligning Korean practices with global standards. During this foundational period, Korean researchers made substantial contributions to developing novel in vitro methods, particularly in areas such as skin sensitization and eye irritation testing. Currently, Korea has emerged as an Asian leader in alternative testing methods, characterized by sophisticated regulatory frameworks and extensive international collaboration. The nation has successfully incorporated various OECD test guidelines while developing innovative approaches. The Korean cosmetics industry has been particularly progressive, driving significant advancements in non-animal safety assessment methodologies. The future trajectory of Korean predictive toxicology focuses on integrating cutting-edge technologies and methodologies. This includes the development of multi-organ chips, AI-powered prediction models, and advanced biomarker systems. The emphasis lies on creating comprehensive integrated testing strategies that combine multiple alternative methods to enhance predictive accuracy. Koreas vision extends beyond mere replacement of animal testing, aiming to establish more reliable, efficient, and ethical approaches to toxicological assessment. This forward-thinking strategy positions Korea to continue making significant contributions to global advancements in modern toxicology.

#### **Development and Distribution of Alternative Testing Methods and Educational** Materials

#### **Seung Hyeok Seok**

Seoul National University, Medical College, Seoul, Republic of Korea <u>lamseok@snu.ac.kr</u>



Prof. Seok's research group is involved in the development of Cancer Immunotherapeutic Treatment by Targeting Tumor-Associated Macrophage Macrophage, which makes up the large part of tumors, suppresses the function of CD8 T cells, greatly inhibiting the effectiveness of the immune checkpoint blockers currently in use. They also explore how tumor-associated macrophage suppresses CD8 T cells and develop a new booster-concept cancer immunotherapeutic drug that can maximize the effectiveness of existing treatments by converting M2 type macrophage to M1 type. To this end, they introduce a variety of transgenic mice, and at the same time, and aim for applicable pre-clinical research through efficient convergence with clinical studies. The other area of his research group is the development of a Method for Improving Tissue Regeneration by Macrophage Cell Therapy By using the great potential of macrophage in a process of tissue regeneration, his laboratory is trying to develop Macrophage Cell Therapy for many regenerative diseases.

#### Abstract

In this study, experts from various fields related to alternative animal testing methods in Korea were invited to standardize cutting-edge knowledge into textbooks, leading to the world's first development of a curriculum for alternative testing methods. To utilize this curriculum, we analyzed the current status of related education and educational material formats, along with an intensive analysis of alternative testing education conducted at universities, and graduate schools, using this as foundational data for target audiences and expansion of educational materials distribution. Simultaneously, we surveyed the awareness of domestic professionals and demands regarding alternative testing education curricula, establishing data on their past alternative animal testing course subjects, learning formats, educational material types, interest levels, preferred future course subjects, willingness to open and take courses, textbook utilization intentions, and perceptions about textbook distribution directions. Finally, we established implementation plans for textbook development and distribution through analysis of the 2023 Korea Chemical Management Association alternative testing educational materials development project results, pilot education, and evaluation. According to our research results, there were significant differences in educational content and methodological requirements between instructors and noninstructors, as well as between academia and industry, indicating the need for target-specific standard textbook development. Additionally, since currently developed educational materials primarily target graduate students at a basic level, it is necessary to consider creating advanced textbooks for practitioners and experts by selecting topics requiring in-depth coverage. Based on these educational materials, it is necessary to establish regular course curricula in academia while enabling industry professionals to complete the same educational curriculum online, thus maintaining nationwide alternative testing education standards as set by the government. Continuous discussions with industry are needed to clearly define the benefits granted to those who obtain alternative testing expert certifications. Above all, developing alternative testing experts ultimately depends on securing a pool of instructors capable of providing specialized education. Creating a national atmosphere where non-experts can establish courses with relatively low burden and promote undergraduate and graduate education based on these newly developed materials is considered the key direction for expanding educational material distribution.

# Zebrafish embryo toxicity Assay: Journey from water quality to natural product toxicity in Sri Lanka

#### D.P.N. De Silva

Department of Chemistry, Faculty of Science, University of Colombo, Sri Lanka. prasadi@uwu.ac.lk



Dr. D. Prasadi N. De Silva's research focuses on immunotherapeutics, transcriptomics, aquatic animal diseases, and the use of zebrafish as biomedical models. Dr. De Silva is a member of the Sri Lanka Veterinary Association and the Sri Lanka Association for Laboratory Animal Science. She has published extensively on topics such as zebrafish embryo toxicity testing, molecular identification of aquatic parasites, and immunological studies involving brown-bodied bamboo sharks. Her ongoing research includes developing immunotherapeutic agents based on shark-derived immunoglobulins. Completed projects involve molecular analysis of Koi Herpes Virus and pesticide toxicity testing in zebrafish models. Dr.. De Silva has received numerous accolades, including the Astron Scholarship for academic excellence in BVSc, the Japanese Government (MEXT) Scholarship, and a Best Presenter award at the Annual Research Conference in Tokyo. She has held academic positions since 2009, including roles at Uva Wellassa University and the University of Peradeniya. She also supervises undergraduate research, particularly in environmental toxicology and aquatic health. Dr. De Silva's work emphasizes multidisciplinary approaches, integrating molecular biology, immunology, and toxicology, contributing significantly to veterinary science and aquatic biomedical research.

#### Abstract

Zebrafish embryo toxicity testing (ZET) is a widely used alternative test for laboratory animal use. Transparency, convenience of observing embryonic development under the microscope, genetic similarity to humans, ability to obtain results in four days, less labour-intensive procedure and availability of whole genome database are some of the advantages of using this test. In Sri Lanka, first zebrafish embryo toxicity experiment was conducted in 2014 on determination of heavy metal toxicity for the water quality analysis. The ZET is widely used to assay ecotoxicity and in Sri Lanka, several pesticides particularly containing organophosphates were screened for their effects on aquatic environment. ZET was also successfully tested for the toxicity of metal-glyphosate complexes and the presence of fungicide residues. Moving on to water quality and environmental toxicity testing ZET turns new pages on determining toxicity of natural products particularly herbal extractions with implications on human health. The effects of the exposed chemicals on embryonic growth, their sublethal abnormalities and biodistribution can be detailed in the zebrafish embryo model. In Sri Lankan context, optimum climate and water quality parameters for brooders is an advantage and frequent awareness programs organized for the scientific community is a plus. However, lack of transgenic strains of disease models, uncertainty of the genetic make-up of the wild type and inbreeding complications are some issues to date. Emerging diseases and technical issues of making water quality sometimes cause complete loss of adult zebrafish and lack of live feed supply results in poor quality embryos. Despite all these, ZET is currently a widely used bio-assay for toxicity testing in Sri Lanka focusing on various aspects of its usage. Hence, development of computational model and novel strains using transgenic techniques and cross-breeding are undergoing to facilitate effective application of ZET as an alternative for lab animal toxicity testing in Sri Lanka.

# Cost-effective alternatives: Challenges and opportunities for implementing non-animal testing techniques in Sri Lanka

#### Dr. Kalpani Ratnayake

Faculty of Health Sciences, CINEC Campus, Malabe, Sri Lanka <u>kalpani.ratnayake@cinec.edu</u>



Dr. Kalpani Ratnayke is the head of the Department of Cosmetic Science. She has expertise in Biochemistry and natural products with a research interest in the formulation of cosmetic products, dermatology, pharmacology, chemistry of natural products, and food and nutrition.

#### Abstract

Cost-Effective Alternatives: Challenges and Opportunities for Implementing Non-Animal Testing Techniques in Sri Lanka explores the potential of replacing traditional animal-based research methods with innovative and ethical alternatives. While non-animal testing techniques such as in-vitro models, computational simulations, and organ-on-chip technologies hold significant promise, their adoption in Sri Lanka faces hurdles like limited resources, high initial costs, and a lack of infrastructure. Additionally, there is a need for increased training and awareness among researchers regarding validated alternatives to animal testing. Regulatory frameworks and institutional support for implementing alternative methods are still evolving. Despite these challenges, there is a positive trend, with academic institutions and research organizations initiating discussions on alternatives, driven by global movements to replace, reduce, and refine animal use. Embracing these alternatives not only aligns with international ethical standards but also promotes cost-effectiveness and scientific precision, paving the way for a progressive research landscape in Sri Lanka. Society for Alternatives to Animal Testing in Sri Lanka (SAAT-SL) is the main society that brings together all Sri Lankan researchers interested in alternatives to animal testing. SAAT-SL has conducted a number of workshops, webinars and certificate courses to encourage Sri Lankan researchers towards the alternative techniques to laboratory animal testing and "Lemna minor (Duckweed) and Allium cepa (Onion) as alternative tools for testing cytotoxicity and genotoxicity" workshop was one of the most popular one. The Allium cepa and Lemna minor models are promising alternatives, as they can be used to detect both cytotoxicity and genotoxicity with the presence of chromosomal alterations. It is a simple, economical and easy-to-perform test, which is considered to be an extremely efficient indicator of environmental pollution. The In-vitro porcine liver slice method is another alternative technique which we tried to popularize among Sri Lankan researchers to screen in-vitro hepatoprotective action. Likewise, efforts have been made to promote low-cost alternatives to overcome fund barriers which is the one of major challenges and it opens the door to the opportunity to practice the 3R concept in laboratory animal experiments. In addition, survey research based on "Evaluation of challenges and opportunities on alternative techniques to laboratory animal experiments among the researchers in Sri Lanka" was conducted. The findings of this research will help identify the challenges and opportunities for alternative techniques in Sri Lanka and support for arranging future activities of SAAT-SL including promoting awareness and education among researchers and stakeholders about the ethical and scientific benefits of alternatives.

#### Establishing the 'Replacement' journey in Sri Lanka

#### Prof. Mangala Gunatilake

Dept. of Physiology Faculty of Medicine University of Colombo, Sri Lanka <u>mangalag@physiol.cmb.ac.lk;</u> <u>mangalagunatilake@hotmail.com</u>



Professor Mangala Gunatilake (*BVSc, PhD, MLAS, FSLCVS, FELASA, FIUPS*) is attached to the Department of Physiology at the Faculty of Medicine, University of Colombo. She has made significant contributions to the field of laboratory animal science (LAS), ethics and animal welfare and alternatives. In 2011, she took a groundbreaking step by launching the first International Certificate course in Laboratory Animal Science in the Asian region, in collaboratory Animal Science (SLALAS), Professor Gunatilake spearheaded the introduction of the 'Replacement' concept to Sri Lankan researchers in 2014, with the support of international experts significantly impacting the LAS landscape in the country. Models introduced include the zebrafish embryo model, HET-CAM assay, IdMOC model, *in vitro* EpiDerm skin irritation test, and hydra model. In addition to her role at SLALAS, she is also the Founding President of the Society for Alternatives to Animal Testing in Sri Lanka (SAAT-SL) and currently, serves as the Founding Director of the 3Rs Centre in LAS.

#### Abstract

The concept of 'Replacement' within the 3Rs principle (Replacement, Reduction, and Refinement) was formally introduced to Sri Lankan researchers during the inaugural scientific conference of the Sri Lanka Association for Laboratory Animal Science (SLALAS) in January 2014. This initiative marked a pivotal shift in research practices, particularly for junior researchers who began to explore and adopt alternative methodologies. Key models for replacement introduced include the Zebrafish Acute Embryo Toxicity Test, HET-CAM Assay, IdMOC, and EpiDerm Models. Over the past decade, SLALAS has organized various activities (workshops and courses) that served as platforms for introducing several other replacement models, such as cell Hydra, C. elegans, Drosophila and Allium cepa, organoids etc. Additionally, the acceptance of artificial mouse and rat models, along with suture training models, has been well-received by trainees participating in these activities, showcasing their educational value and effectiveness in training. The incorporation of an Alternatives Module into the Postgraduate Certificate and Diploma courses in LAS further underscored the commitment to promoting the Replacement concept. The establishment of the 3Rs Centre in 2019 and the formation of the Society for Alternatives to Animal Testing in Sri Lanka in 2021 also reflect significant strides in this field. This journey towards embracing the Replacement concept has not only fostered a culture of ethical research but has also empowered researchers in Sri Lanka to explore innovative alternatives, ultimately enhancing the integrity and impact of scientific research in the region.

# In vitro NAMs: Gaps in scientific and regulatory validations and role of India in global context

#### Dr. Subrahmanyam Vangala

Co-Founder & CEO, Reagene Biosciences, India subrahmv@gmail.com



Dr. Vangala is currently Co-Founder and Chief Executive Officer at ReaGene Biosciences Private Limited (Bengaluru, India), and ReaGene Innovations Private Ltd (Hyderabad, India). He is an experienced pharma scientist and executive with more than 25 years of leadership experience with increasing responsibilities, at global pharma in the USA (Wyeth, JNJ, Purdue Pharma and Shire) and Contract Research Organizations in India (Sai Life and Advinus). His industrial experience was focused on, but not limited to, new drug discovery and development with expertise in DMPK, Bioanalytical, Clinical Pharmacology, and Toxicology. His Pharma experience produced more than 30 IND submissions, with at least six marketed molecules drugs including Zaleplon (short-acting sedative-hypnotic), Tygacil (4th generation tetracycline antibiotic against vancomycin and methicillin-resistant bacteria), Canagliflozin (targeted SGLT-2 inhibitor for type II diabetes), Tibsovo (targeted IDH1 inhibitor for refractory myeloid leukemia). Other areas he gained experience include predictive clinical drug-drug interactions, pre-formulation development, drug repurposing, specialty pharmaceuticals, generics, biologics/biosimilars, medical devices, pharmacogenomics, metabolomics and alternatives to animals in research.

#### IL-7

IL-6

#### Engineering the placenta: drug safety screening using organ-on-chip technology

#### **Deepak Modi**

*ICMR-National Institute for Research in Reproductive and Child Health* <u>deepaknmodi@yahoo.com</u>



Dr Deepak Modi is a Scientist at ICMR NIRRH. His lab is working on understanding the fundamental mechanisms of embryo implantation and early placentation. They are also studying how these processes go awry in women with infertility. They are also interested in determining the molecular basis of gonad development and cell fate decision-making within them. Dr. Modi is passionate about Understanding the biological basis of sexuality and gender. Currently, he is working on the concept of Placenta-on-Chip to make pregnancy safe.

#### Abstract

Placenta is the largest fetal organ and the first to develop. It is a key organ controlling the transfer of nutrients across the feto-maternal interface and regulate fetal growth. It also protects the fetus from immunological insults and yet acts a barrier towards infection. To study placental function various animal model systems have been used, however, the structural and functional makes extrapolation of all the observations to humans difficult. Hence there is a need of complementary human models to study placental functions. We have developed in vitro platforms that recapitulate the spatial and temporal kinetics of the human placenta to assess placental function. We have developed modular devices viz, a device for 3D organization of placental villi, and a device to mimic the placental barrier function. The results will be discussed.

Advancing alternative models for toxicity assessment: In vitro pyrogen test and organon-a-chip approaches

#### Megha KB

Toxicology Division, Biomedical Technology Wing, Sree Chitra Tirunal Institute for Medical Sciences and Technology (Govt. of India), Poojapura, Trivandrum 695 012, Kerala, India meghakb997@yahoo.in, meghakb@sctimst.ac.in



Dr. Megha K B, Post Doctoral Fellow at Division of Toxicology, Biomedical Technology, Sree Chitra Tirunal Institute for Medical Sciences and Technology has awarded the Gold Medal for her presentation and research work entitled 'Development of ELISA kit for animal Pyrogen test' at the 5th Annual Conference of Society for Alternatives to Animal Experiments – India (SAAE-India 2022), held at CLIF, Karyavattom Campus, University of Kerala, during 8-9 December 2022.

#### Abstract

Pyrogenic contaminants pose a serious threat to biological and medical products, as they can trigger febrile responses, potentially leading to septic shock and death. To ensure the safety of parenteral drugs, it's critical to eliminate pyrogens. This study highlights the importance of a detection system for tracking IL-1 $\beta$  release, a key marker of inflammation, upon contact with specific agents. An indigenously developed ELISA method was used to quantify pro-inflammatory cytokine IL-1β. Pyrogenic induction was achieved using bacterial endotoxins (lipopolysaccharide (LPS), lipoteichoic acid (LTA)) and chemical pyrogens (phytohemagglutinin (PHA), trinitrophenol (TNP), and ketamine) in fresh pooled human blood, with cytokine release measured via sandwich ELISA. The study provides insights into IL-1β release following exposure to pyrogens, with a rapid increase observed 2 hours post-treatment. This research establishes a platform for reliable IL-1 $\beta$  detection using a stable blood supply, minimizing personal variation in in vitro pyrogenicity testing. Additionally, a rabbit model was used to study the pyrogenic response mechanism and biochemical parameter variations under endotoxin-induced inflammation, allowing picogram-level IL-1 $\beta$  detection—beneficial for pharmaceutical and healthcare applications. Another approach for testing the toxicity assessment is Organ-on-a-chip technology, which mimics the microphysiological system of the target organ site or area. To study inhalation toxicity by mimicking the complex structures and functions of human lung tissue in a controlled, miniature environment, hydrogel-based primary lung smooth muscle (PLSM) on a chip to study alveolar smooth muscle remodeling upon exposure to pollutants. These microfluidic devices are engineered to replicate key physiological processes, such as air-blood barrier interactions, essential for assessing how inhaled substances impact respiratory health. By allowing realtime observation of cellular responses to toxins, particulates, or chemicals, lung-on-a-chip models provide a more accurate, human-relevant alternative to traditional in vitro and animal testing methods. This technology has the potential to revolutionize respiratory toxicology, improving safety assessments for airborne contaminants in pharmaceuticals, environmental pollutants, and consumer products. Traditional biocompatibility testing often

relies on animal models and standard cytotoxicity tests, but newer approaches like in vitro pyrogen testing and organ-on-a-chip systems are transforming the field. These advanced techniques offer more precise, human-relevant data by simulating complex biological environments and cellular responses. This progress not only enhances the reliability of biocompatibility assessments but also aligns with ethical goals to reduce animal testing, paving the way for safer and more effective medical innovations.

#### 3D Organoids models in solid tumors for drug development and translational research

#### Fazal Khan

Sultan Qaboos Comprehensive Cancer Care & Research Center, Al Khoudh St, Muscat, Oman <u>f.khan@cccrc.gov.om</u>



Dr. Khan is a senior research scientist at the Sultan Qaboos Comprehensive Cancer Care & Research Center, Muscat, Oman. He has expertise in stem cell regenerative medicine, bioengineering, artificial tissue engineering, cancer therapy, and molecular diagnostics.

#### Abstract

Despite recent advances in cancer treatment and diagnostics, it remains the second leading cause of death worldwide. There are several factors contributing to this, such as the surging cost of anticancer agents, the complexity of clinical trials, and regulatory requirements. The most concerning factor is the successful translatability of preclinical in-vitro and in-vivo studies into clinical trials. The lack of identical models for human tumors not only affects the experimental results but also hampers drug approvals by regulatory agencies. The most popular 2-dimensional cell culture models oversimplify real human tumors and do not recapitulate all the essential aspects of solid tumors, such as cellular organization and inter-cellular interactions within the tumor microenvironment. Alternatively, three-dimensional organoids or tumproids derived from tumor cells or tumor tissues could bridge the gap. These organoids are identical to the tumors in terms of physical growth kinetics, cell-to-cell interactions, nutrient penetration, and tumor immune and extracellular microenvironment. Recently, the United States Food and Drug Administration (FDA) has removed the requirement for animal testing of new drugs, leading to a rapid expansion in the field of in-vitro drug testing using patient-derived organoids for novel therapeutic and personalized treatment development. Here, we share our recent data on organoid development using hydrogel and ultra-low attachment technologies. A platform has been established for the development of patient-derived organoids from fresh solid tumor tissues and malignant ascites. Organoids were successfully validated for their cellular and molecular features and pathological protein expression patterns in comparison with the patient's diagnostic details. Further organoids were evaluated for chemotherapy drug efficacy and proliferation at Sultan Qaboos Comprehensive Cancer Care and Research Center in Oman. This established platform will further be utilized to study ex-vivo patient-derived organoids for co-culture and cell death assays for the development of immune cell therapies and novel anticancer drug discovery.

# Chicken embryo model as a tool to unravel saroglitazar-induced teratogenic manifestations and its underlying mechanisms

Swarna Dabral<sup>1\*</sup>, Tarun Pant <sup>2,3</sup>, Vishnu Raj<sup>4</sup>, Imran Ahmd Khan<sup>5</sup>, Sabina Khan<sup>6</sup>, Suhel Parvez<sup>7</sup>, Nilanjan Saha<sup>8</sup>

<sup>1\*</sup>Maharishi Markandeshwar College of Pharmacy, Maharishi Markandeshwar deemed to be University, Mullana, Ambala, Haryana, India

<sup>2</sup>Department of Pediatrics, Medical College of Wisconsin, 8701 Watertown Plank Road, Milwaukee, WI, 53226, USA

<sup>3</sup>Department of Surgery, Division of Pediatric Surgery, Medical College of Wisconsin, 8701 Watertown Plank Road, Milwaukee, WI, 53226, USA

<sup>4</sup>Department of Biochemistry, School of Sciences, Jain (deemed to be University), Jayanagar, Bangalore, India

<sup>5</sup>Department of Chemistry, School of Chemical and Life Science, Jamia Hamdard, New Delhi 110062, India

<sup>6</sup>Department of Pathology, Hamdard Institute of Medical Sciences and Research, Jamia Hamdard, New Delhi 110062, India

<sup>7</sup>Department of Medical Elementology and Toxicology, School of Chemical and Life Science, Jamia Hamdard, New Delhi 110062, India

<sup>8</sup>Department of Centre for Clinical and Translational Research, School of Chemical and Life Science, Jamia Hamdard, New Delhi 110062, India swarna.dabral@mmumullana.org



Dr. Swarna Dabral is Assistant Professor and a dedicated pharmacologist at Maharishi Markandeshwar (Deemed to be University), Ambala, India. She has harnessed alternative models and cutting-edge bioinformatics techniques to shape her research and drive innovation in the field. Her passion lies in drug repurposing, a frontier that offers new possibilities for improving human health while reducing our reliance on animal experiments. Dr. Dabral also deeply engaged in exploring bioinformatics tools as a synergistic tool to accelerate biomedical research.

#### Abstract

The landscape of biomedical research is transforming driven by ethical imperatives to minimize reliance on mammalian pre-clinical research model. This shift has catalyzed the pursuit of alternative pre-clinical research models using lower sentient organisms. Among various models, the chicken embryos model has emerged as a simple, ethically sound and versatile system for screening drug safety in pregnancy. In the current study insilico and in-ovo system were integrated to predict the teratogenic influence of saroglitazar. Computational tools encompassing network analysis, molecular docking, and molecular dynamic simulations were employed to screen key targets crucial for embryonic development and its potential to interact stably with saroglitazar. The findings of the study indicated that saroglitazar was teratogenic in the developing embryos and impeded embryonic development corroborating the results of computational analysis. The study not only highlights the potential safety of saroglitazar, but also reflects the effectiveness of multimodal systems - integrating bioinformatics, computational chemistry and in-ovo model – as consolidated system to predict and validate drug induced teratogenicity, providing insights into development of safer drugs during pregnancy.

# Assessment of cytotoxicity, genotoxicity and mutagenicity of Yttria (Yttrium oxide) nanoparticles in V-79 cells

#### Alok K. Pandey, Durgesh Prasad Maurya

Nanomaterial Toxicology Laboratory, Drug and Chemical Toxicology Group (FEST Division) CSIR-Indian Institute of Toxicology Research, Vishvigyan Bhawan, 31, Mahatma Gandhi Marg, P.O. Box 80, Lucknow-226001, India alokpandey@iitr.res.in



Dr. Alok K Pandey is currently working as Senior Principal Scientist in Nanomaterial Toxicology Laboratory at CSIR-Indian Institute of Toxicology Research, Lucknow, India. He completed his PhD in 2007 from University of Lucknow in Environmental Sciences. His area of work includes Nanomaterial Toxicology and Genetic toxicology. His current research work involves mechanism of nanomaterial toxicity in *in vitro* and *in vivo* models using different cytogenetic techniques as well as cell cycle and apoptosis. The work is also being done towards ecotoxicological studies of the nanomaterials. His responsibilities include the course coordinator of one course in Academy of Scientific and Innovative Research, Scientist in-charge of advanced imaging facility and flow cytometry and participating in other institutional Committees. He has published more than 65 articles in International Peer reviewed Journals and five book chapters. He is also serving in more than 25 journals as Editor/Reviewer. So far, Dr. Pandey has trained more than 50 MSc/MTech Scholars as project trainees. He has supervised 5 PhD, students, 5 currently pursuing.

#### Abstract

Recently nanoparticle usage has increased to an extent to which humans and animals are certainly getting exposed. With the advancement in technology, the nanoparticles use is increasing very fast. Yttria in its nanoparticle form is used in a variety of applications such as phosphors, and garnet, and is sometimes mixed with other elements to make them chemically stable at room temperature. So, it is essential to evaluate the toxicity risk of Yttria nanoparticles, V-79 cells were used as a model system as inhalation is the major route of exposure. In this study, Yttria nanoparticles are found to be low toxic as in viability experiments it shows overall viability of more than 60% in most of the concentrations although these nanoparticles cause significant membrane damage when evaluated by LDH release assay. In this study, Yttria nanoparticles do not cause any significant ROS generation in a given concentration in V-79 cells. Internalization is evident in flow cytometry with the increase in SSC % and TEM micrograph. In the genotoxicity experiment, Yttria nanoparticles caused DNA strand breaks in single-cell alkaline comet assay as well as single-cell neutral comet assay; it also caused chromosomal damage and nuclear damage. In the mutagenicity test, Yttria nanoparticles cause a significant increase in mutant frequency. These results can be used as a safety and risk assessment of Yttria nanoparticles when used in any application where there is a chance of exposure to the human system.

#### In vitro models to study tumor microenvironment and cancer therapy

#### Bilal Bin Hafeez, Ph.D.

Department of Medicine and Oncology ISU, South Texas Center of Excellence in Cancer Research, School of Medicine, UTRGV, McAllen, TX 78504, bilal.hafeez@utrgv.edu



Dr. Hafeez's laboratory research focuses are 1) to establish new molecular targets/biomarkers for solid tumors which can be used for rationale design for the prevention and treatment of prostate and pancreatic cancers, 2) to develop new approaches to improve the immunotherapy for solid tumors using orthotopic syngeneic xenograft and genetic engineered mouse models of cancers, 3) Targeting ribosome biogenesis and tumor microenvironment for the treatment of pancreatic cancer, and 4) to understand the molecular mechanisms of Enzalutamide therapy failure and developing new prognostic biomarkers for metastatic castration-resistant prostate cancer (mCRPC). Dr. Hafeez has a research background in immunopharmacology, cancer therapeutics, and cancer biology. He previously established protein kinase C epsilon (PKCɛ) as a novel molecular target for prostate cancer development and metastasis using genetic and pharmacological approaches. His pre-clinical research against prostate cancer lead to the use of plumbagin (a novel and selective inhibitor of PKC epsilon) in phase I clinical trials against metastatic castration-resistant prostate cancer. Dr. Hafeez's laboratory is dedicated to understanding the molecular mechanisms of ribosome biogenesis dysregulation in solid tumors. Dr. Hafeez's lab is funded by NIH and CPRIT where he serves as PI. He also served as Co-Investigator on multiple NIH-funded grants and has published more than 70 high-impact cancer research articles in peer-reviewed journals.

#### Abstract

The tumor microenvironment (TME) is a complex and dynamic ecosystem that plays a critical role in tumor progression, metastasis, and therapeutic resistance. In vitro models provide a versatile and controlled platform to study the TME and evaluation of new cancer therapeutic strategies. Recent advancements in biomedical research developed promising in vitro model systems which are closely mimics to animal model system which include 2D monolayer cultures, 3D spheroids, organoids, and microfluidic-based tumor-on-a-chip technologies. These models mimic various TME components, such as hypoxia, extracellular matrix, stromal cells, and immune cells, to recreate tumor-stroma interactions and study drug responses. 3D organoids, derived from patient tumors, closely replicate the histological and genetic heterogeneity of cancers, making them particularly promising for personalized therapy studies. Meanwhile, microfluidic platforms enable real-time monitoring of cell behavior under physiologically relevant conditions, such as fluid shear stress and nutrient gradients. Despite their promise, in vitro models face limitations in fully capturing the complexity of systemic interactions present in vivo. Integration with computational modeling and co-culture systems is emerging as a strategy to address these challenges. By bridging the gap between simplified 2D systems and animal models, advanced in vitro TME models are poised to accelerate translational cancer research and the development of more effective therapies. However, further research is required to develop more alternative in vitro models to study TME and cancer therapeutics.

# Cholesterol and hair follicle health: Insights into alopecia from in vitro and organoid models

#### Nikhila Leemon, and Sreejith Parameseara Panicker

Dept. of Zoology and Advanced Centre for Regenerative Medicine and Stem Cell in Cutaneous Research (AcREM-Stem), University of Kerala

p.sreejith@gmail.com



Dr. Panicker and his research group work on dermatological research, cancer biology, molecular biology, biochemistry, and endocrinology. He has expertise in the observation and collection of Caecilians and *Rana Curtipus*. His team developed primary cell lines of hair follicle outer root sheath cells, keratinocytes, brain tumor cells, prostate cancer cells, osteoblasts, and epithelial cells.

#### Abstract

Hair follicles (HFs) are dynamic entities of ectodermal origin, essential for human identity, health, and selfperception. The lipid composition, especially cholesterol, is essential for heart failure biology, and disturbances in cholesterol homeostasis are increasingly associated with hair diseases, such as alopecia-a chronic inflammatory syndrome resulting in hair loss. This study investigates the regulatory function of cholesterol production in hair follicle stem cell (HFSC) activity, employing in vitro and organoid culture methods as alternatives to animal experiments. Our work utilized substances such as 7-dehydrocholesterol (7DHC), BM15766, simvastatin, and atorvastatin to regulate cholesterol production and evaluate their impact on HFSC function. Furthermore, scalp specimens from alopecia patients were tested to assess HFSC functionality inside the compromised tissue. Findings indicated a notable downregulation of HFSC marker genes, accumulation of sterol intermediates, and compromised stem cell properties in cells exposed to statins, resulting in diminished HFSC functionality. Mice undergoing cholesterol modulation exhibited compromised hair regrowth, ascribed to HFSC apoptosis. The findings were consistent in both in vitro and organoid culture methods, with statin administration in human hair follicle organoids prompting premature catagen phase, hence demonstrating disturbed hair development cycles. Transmission electron microscopy (TEM) examination of hair follicles revealed impaired cellular integrity, irregular melanin distribution, and disorganized keratin in statin-treated samples, resulting in weakened hair shafts susceptible to breakage. The results highlight the critical need of cholesterol management in preserving HFSC health and indicate that cholesterol-lowering medications may unintentionally affect HF integrity and regeneration, offering fresh perspectives on alopecia causation. This study clarifies the connection between cholesterol dysregulation, HFSC function, and hair follicle regeneration, enhancing our comprehension of alopecia while offering innovative alternatives to animal experimentation. Our research underscores the therapeutic promise of cholesterol regulation in alopecia, stressing the necessity for targeted lipid-based strategies that preserve HFSC viability. This in vitro and organoid-based methodology creates a scientifically sound, ethical, and pragmatic foundation for advancing research on alopecia and associated dermatological disorders.

# Generation of immune cells through in vitro cell culture methods

**Dr. Rizwanul Haque, Ph.D.** Department of Biotechnology, SEBES, Central University of Bihar rhaque@cub.ac.in



Dr. Rizwanul Haque received postdoctoral training for 9.2 years (2003-2012) at Penn State College of Medicine, Hershey, PA, USA. Dr. Haque has extensive experience in the area of immunology, particularly on stem cells to treat immune-mediated diseases. His breakthrough research and his background are significant for developing therapeutic immune cells to treat cancer potentially. Dr. Haque is currently working on cancer reactive T cells and dissecting the role of Telomere and Telomerase modulation by using novel and natural compounds to modulate liver and lung cancer as well as identifying novel epitopes for neo-antigen based vaccine development leading to the treatment of cancer and other infectious diseases. Dr. Haque has delivered more than 30 invited lectures and 40 selected lectures in national and international conferences all over the world. Dr. Haque has made significant scientific contributions, evident from his 60 publications in a variety of top-class scientific journals and his research has been cited more than 4000 times by other research groups worldwide, with an *h*-index of 24 and *i10*index of 37. Dr. Haque is serving as the PI and Co-PI in SERB, DST-FIST, ICMR, Bihar S&T, and in corporatefunded research projects and previously served as Co-PI in 5 research projects in the USA. Dr. Haque served as Head of the Department of Biotechnology from 2018 to Aug 2021. He also served as Head of the Centre for Biological Sciences (2012-2018) which included Biotechnology, Life Science, and Bioinformatics programs.

#### Abstract

Immune cells are a key pool of mammalian adaptive immunity and are composed of several different subsets of immune cells, each with a distinct function, such as T helper cells, cytotoxic T cells, and regulatory T cells. Recent advancements in induced pluripotent stem (iPS) cell technology and the progress in an in vitro system for gene incorporation are capable of generating iPS cells from patients without any surgical approach. In addition, like ESCs, iPS cells possess indefinite proliferative capacity in vitro and have been shown to differentiate into hematopoietic cells. Here, we present methods for the generation of T lineage cells from iPS cells by using an in vitro model system. By using in vitro 2D and 3D culture methods, we can differentiate a specific subset of T cells from iPS cells that can be used for the treatment of immune-mediated diseases. We stimulated iPS cells in vitro with a Notch ligand drives T cell differentiation from iPS cells, and the incorporation of the FoxP3 gene by gene transduction and then stimulation with Notch ligand results in iPS cells differentiating into Treg cells. To generate therapeutically active Tregs with an increased lifespan and hence greater therapeutic potential, we used retrovirusmediated transduction to introduce FoxP3 or FoxP3 with anti-apoptotic bcl-2 family molecule Bcl-xL linked by a 2A picornavirus self- cleaving peptide, into iPS cells. We found that iPS cells expressing both FoxP3 and Bcl-xL differentiated into functional regulatory T cells and had a long-term survival advantage over cells transduced with FoxP3 alone in iPS cells by using 2D and 3D culture systems. The data suggest a potentially novel approach to generate highly reactive T cells and Tregs for augmenting cellular immunotherapy for autoimmune disease.

IL-14

### IL-15

# Development of the accurate, accelerated, and affordable diagnostic kit (Real Tick) for rapid identification of re-merging tick-born zoonotic infections (Rickettsia & Orientia)

## Dr. Yaawar Bashir Mir

Department of Biotechnology, Sher-e-Kashmir University of Agricultural Sciences & Technology, Srinagar, India <u>ymir.dx@gmail.com</u>



Dr. Mir Yaawar is a dynamic microbiologist and versatile researcher with over a decade of experience in scientific research, teaching, and laboratory management. He holds a Ph.D. in Microbiology from Sher-i-Kashmir Institute of Medical Sciences (SKIMS), where his doctoral research focused on the development of a multiplex PCR method for the rapid diagnosis of Rickettsial infections. His academic journey includes a Master's and a Bachelor's in Microbiology from Guru Nanak Dev University. He has made innovative contributions to biotechnology and molecular diagnostics, with three notable patents to his name. These include the Infection-Free Bandage, a collagen-based bandage strip loaded with stem cell exosomes and phytomolecules for infectious wound healing, and the Heal-Exo-Fast cream for wound healing, a gelatin-chitosan polymeric scaffold loaded with calcium peroxide, nitrous oxide, and mesenchymal stem cell exosomes. He is also the developer of "INJQUANT," an Android application designed to quantify the extent of superficial tissue injury and the rate of healing. This application has gained significant traction, with a wide range of users relying on it for wound management solutions. Currently, Dr. Yaawar is working as a scientist and pursuing his postdoc in the Department of Biotechnology at Sher-e-Kashmir University of Agricultural Sciences and Technology of Kashmir. His research interests lie in infectious diseases, with a particular focus on understanding pathogen biology and host-pathogen interactions to advance the development of novel diagnostic platforms. His work aims to address critical challenges in the rapid detection and management of infectious diseases, leveraging molecular biology, bioinformatics, and advanced diagnostic technologies. His professional career spans esteemed institutions like the CSIR-Institute of Microbial Technology and SKIMS, where he has conducted pivotal research on infectious diseases and protein science. During the COVID-19 pandemic, he served as a nodal officer for genome sequencing of COVID-19 variants, earning him a state-level award for exemplary service. An accomplished academic, Dr. Yaawar has published extensively in international peer-reviewed journals and has presented his research at national and international conferences. Additionally, he is a skilled educator and mentor, recognized for his ability to communicate complex concepts effectively. With expertise in advanced laboratory techniques and bioinformatics tools, Dr. Yaawar continues to innovate in diagnostics and infectious disease research.

## Abstract

Rickettsial diseases can present a serious public health threat if misdiagnosed. The clinical signs often mimic other febrile illnesses, leading to improper treatment. Existing diagnostic methods, such as Weil-Felix, ELISA, and IFA, have limitations including low sensitivity, inability to detect early infections, and issues with antigenic variability. There is a need for a rapid, affordable, and accurate diagnostic assay that can detect infections caused by both Rickettsia and Orientia. The current study aimed to develop a multiplex PCR diagnostic assay that detects the three main groups of Rickettsial diseases: Spotted Fever, Typhus, and Scrub Typhus. A total of 200 patient samples meeting the inclusion criteria were collected and analyzed. Novel genetic markers for each Rickettsial group were identified: GltA, 17 kDa, and ompA for Spotted Fever; OmpB and PPiase for Typhus; and 47 kDa and 56 kDa for Scrub Typhus. Primers were optimized, and positive controls for each group were used. PCR confirmed the presence of 24, 12, and 34 positive samples for Spotted Fever, Typhus, and Scrub Typhus, respectively. A newly developed multiplex PCR method was optimized to detect bacterial DNA concentrations as low as 1 pM/µl for Spotted Fever and Scrub Typhus, and 200 pM/µl for Typhus. All samples that tested positive via conventional PCR were also positive with the multiplex PCR, showing 100% sensitivity. Sequencing confirmed the results. In response to the increasing complexity of co-infections, the multiplex assay was tested on mixed DNA samples from different fever groups. The assay successfully detected the distinct bands for all combinations, confirming its potential for co-infection detection. The next phase of this study is to patent this multiplex PCR method and

explore its commercialization as a diagnostic kit. This method has the potential to replace current molecular and serological tests and could be expanded to detect additional Rickettsial and Orientia species through further research.

# Serological & Molecular Identification of Measles & Rubella in Jammu & Kashmir

# Bashir Ahmad Fomda<sup>1</sup>, <u>Mehvish Saleem<sup>2,\* 3</sup>Sheeba Mushtaq</u>, <sup>4</sup>Mansoor Ahmed , <sup>5</sup>Aaqib Shakeel

<sup>1</sup> Professor & H.O.D Department of Microbiology SKIMS Soura Srinagar 190011 India <sup>2</sup>Scientist –C State Level Viral Research and Diagnostic Laboratory SKIMS Soura Srinagar 190011 India, <sup>3,4</sup> Research Assistant State Level Viral Research and Diagnostic Laboratory SKIMS Soura Srinagar 190011 India, <sup>5</sup> Multitasker & MLIS data Entry operator for MR Program <sup>1</sup>Email:bashirfomda@gmail.com; \*Email:mehvish.saleem@gmail.com



Presently, she serves as an ICMR/DHR Senior Research Scientist-C at the Viral Research and Diagnostic Laboratory (VRDL), Sher-I-Kashmir Institute of Medical Sciences, Soura, Srinagar. With over 16 years of experience in microbiology, she earned her Ph.D. in Microbiology from Vinayaka Mission University in 2011, focusing on "Urinary tract infections in diabetic patients and biodiversity of UTI microflora." As a senior research scientist, her work emphasizes on the development of cost-effective diagnostic tests for viral infections in resource-limited settings, implementing surveillance programs, and conducting national and international audits for laboratory quality assurance programs. I have secured ICMR extramural grants to develop a rapid diagnostic kit for identifying congenital cytomegalovirus in neonates. She has published over 15 research papers, contributed two book chapters, and authored one book in reputed journals. She holds memberships with the American Society for Microbiology as a Global Outreach Member and the European Society of Clinical Microbiology and Infectious Diseases (ESCMID) as a Young Investigator Member. Notably, she received the ESCMID Travel Award for presenting her research in Barcelona, Spain, and she also served as a peer reviewer for four international journals. Her other research interests include viral oncology, host-pathogen interactions, and antimicrobial resistance.

## Abstract

**Background:** The South-East Asia Regional Measles and Rubella (MR) Laboratory Network, established in 2003 as part of the WHO Global MR Laboratory Network, supports regional efforts for MR surveillance. Currently, the SL-VRDL at SKIMS conducts both serological and molecular detection of Measles and Rubella across districts in the Union Territories of Jammu & Kashmir and Ladakh. This program aims to further strengthen surveillance by testing the cases of fever and rash suspected of Measles or Rubella across Indian States.

**Material and Method:** Between July 1, 2021, and November 30, 2024, the SL-VRDL-SKIMS received 8,592 serum samples and 5,861 throat swab samples from the WHO regional zone of Jammu & Kashmir. Each sample was cataloged with a unique EPID ID, linking it to coded patient details. Serological detection of Measles and Rubella IgM antibodies was performed using the CDC-approved "EuroImmune Kit" following WHO algorithms. Throat swab samples underwent RNA extraction using the Qiagen Viral RNA Mini Kit and subsequent molecular detection. RT-PCR was performed to amplify a 634-nucleotide (nt) fragment of the Measles virus (MeV) nucleoprotein (N) gene using CDC Measles genotyping and Qiagen One-Step RT-PCR kits. Detection of Rubella virus (RuV) was conducted using the CDC Rubella End-point PCR kit and Invitrogen Superscript to amplify a 154-nt sequence in the RuV non-structural protein coding region.

**Results:** Of the 8,524 serum samples analyzed, 694 (8.14%) tested positive for Measles IgM, 7784 (91.3%) were negative, and 46 (0.53%) were equivocal. For Rubella IgM, 346 (4.05%) were positive, 24 (0.28%) were equivocal, and 7,596 (89.11%) were negative. Among the 5,861 throat swab samples processed for molecular testing, 75 (0.01%) were identified as Measles genotype positive between 2023 and 2024. Fifteen of these samples, sequenced at NIV Mumbai, were confirmed to belong to Measles genotype D8. No Rubella genotypes were detected.

IL-16

# Adsorption of Heavy Metals and Pharmaceuticals from Water: Application of Plant and Animal Waste

## Joydeep Dutta

Department of Zoology, School of Bioengineering and Biosciences, Lovely Professional University, Phagwara, 144411, PUNJAB

joydeep.dutta@lpu.co.in



He is the Head, Department of Zoology and Botany, School of Bioengineering and Biosciences, Lovely Professional University. His current research interests focus on nanotechnology in aquaculture, nanoparticles, and pesticide toxicity in vertebrate and invertebrate models.

## Abstract

The bioactive functional groups in chitosan, a naturally occurring polysaccharide, provide it significant adsorption properties. The study enhances adsorption qualities by modifying a variety of composite chitosan-based hydrogel beads with plant components such as lignin, peanut hulls, walnuts, and almond shells. The study employs techniques like SEM, FTIR, TGA, and XRD to characterize each composite, revealing significant improvements in pH resilience, mechanical stability, and thermal stability. With pseudo-second-order kinetics and a good match to Langmuir and Freundlich isotherms, batch adsorption studies verify that these hybrid chitosan beads are highly effective at eliminating pollutants such as dyes, heavy metals, and antibiotics. The results demonstrate the feasibility of these inexpensive, environmentally benign composite beads as a long-term wastewater treatment solution, pointing to a possible avenue for further development and use in environmental pollution prevention.

# Series of non-mammalian model organisms for toxicological testing and research on Xenobiotics

## **Anand Mohan**

School of Bioengineering and Biosciences, Lovely Professional University, Phagwara, Punjab <u>anandmohan77@gmail.com</u>



Dr. Anand Mohan started his career with his Master's degree in Biotechnology from IIT, Roorkee. He also qualified for CSIR JRF 3 times and SRF. Further, he did his research-level training at AIIMS, New Delhi as a Junior research fellow and earned his doctorate degree from Jamia Hamdard, New Delhi. Professor Anand Mohan has 2 years of Industry experience and 14 years of teaching and research experience. He has published almost 80 International and national research papers and 15 international book chapters in reputable publication houses like Springer, American Chemical Society, Royal Society, Royal Society of Chemistry, Elsevier etc. He has also got 3 patents and more than 15, 16 S rDNA sequence accession numbers in the NCBI database. He has supervised 10 doctoral students till now and more than 150 master's and bachelors' students. His major research field includes interesting themes in Biotechnology, Nanotechnology, Toxicology and Environmental Sciences. He has also held the posts of academic coordinator in the field of Entrepreneurship Development and NAAC Coordinators. He has also received awards for best researcher two times from Lovely Professional University.

### Abstract

With the advent of modern chemistry and synthesis techniques newer compounds are produced by numerous research organization all around the world. These compounds have to be tested not only for their effectiveness in the said parameter but also for their toxicity in surrounding environment. There are number of animals employed such as rodents, monkeys, and rabbits. The use of these mammalian animals requires stringent ethical clearance from various committees and testing on them also requires higher costs, labour intensiveness. Agrochemicals like herbicides, pesticides and other related chemicals have more exposure to their surrounding flora and fauna as compared to humans. Their testing on lower organisms present at farm sites, provides an excellent opportunity for testing them on groups receiving the highest exposure. These kind of tests are not only cost-effective but also require lower ethical considerations. The use of Drosophila melanogaster (common fruitfly), Esenia fetida (earthworm) and other common species are specifically advocated. The rationale for this is their high reproductive abilities, shorter life span, and easy culturing. Along with these abilities they represent major ecologically dominant invertebrate groups and have been utilized in testing for decades. They could be effectively used as predictive models in multiple types of testing. The current work discusses on importance of these models, challenges, and standard testing protocols.

IL-18

IL-19

# Structure-based computational and experimental approaches in drug discovery: Exploring phytochemicals as novel inhibitors of MTH1

### Md. Imtaiyaz Hassan

Centre for Interdisciplinary Research in Basic Sciences, Jamia Millia Islamia, Jamia Nagar, New Delhi 10025, INDIA mihassan@jmi.ac.in



Dr. Md. Imtaiyaz Hassan is a distinguished expert in structural biology and drug discovery, currently serving as a Full Professor at the Centre for Interdisciplinary Research in Basic Sciences, Jamia Millia Islamia, New Delhi, India. With over 15 years of research experience, Dr. Hassan's work bridges experimental and computational techniques, focusing on the design of high-affinity, selective inhibitors with minimal side effects. His prolific academic output includes approximately 532 research articles published in prestigious journals, amassing over 19,000 citations and an impressive h-index of 65. He has completed over 20 research projects, supervised more than 30 PhD scholars, and guided 200+ MSc dissertations, highlighting his commitment to mentoring and scientific excellence. Dr. Hassan holds prominent roles in the scientific community, including Vice President of the Indian Biophysical Society, General Secretary of The Protein Society, and Joint Secretary of the Bioinformatics and Drug Discovery Society. He is also a life member of over 10 professional societies. Additionally, he serves as Associate Editor for journals such as the Journal of Biomolecular Structure and Dynamics, Oxidative Medicine and Cellular Longevity, and Journal of Alzheimer's Disease, alongside guestediting several leading subject journals. Recognized globally, Dr. Hassan has been honored as a Fellow of the Royal Society of Chemistry (UK) and the Royal Society of Biology (UK). For the past five years, he has consistently ranked among the top 2% of scientists worldwide. He exemplifies excellence in research, education, and leadership, pushing the boundaries of drug discovery while inspiring the next generation of scientists.

#### Abstract

MTH1 is a critical enzyme that safeguards cellular DNA integrity by hydrolysing oxidized nucleotides, thereby mitigating oxidative damage. Tumour cells, frequently subjected to heightened oxidative stress, heavily depend on MTH1 to maintain survival and proliferation, making it an attractive therapeutic target. This study employs an integrative approach combining structure-based computational drug discovery and experimental methodologies to identify phytochemicals with potential MTH1 inhibitory activity. Structure-based drug discovery utilizes the detailed three-dimensional structure of target proteins to design or identify molecules that bind with high specificity and affinity, accelerating the identification of lead compounds. Integrating this with experimental validation ensures a comprehensive understanding of the compounds' therapeutic potential, bridging the gap between in silico predictions and real-world efficacy. Using the IMPPAT library of natural compounds, molecular docking and virtual screening of ~18,000 molecules were conducted, followed by rigorous filtering based on Lipinski's rule, ADMET properties, and molecular dynamics simulations. Two potent phytochemicals, Vinburnine and Norstephalagine, were identified, exhibiting strong binding affinity, structural stability, and favorable pharmacokinetic profiles as MTH1 inhibitors. Additionally, dietary phytochemicals, including Thymoquinone (TQ), Baicalin (BC), Resveratrol (RV), and Quercetin (QT), were analyzed for their interactions with MTH1. Molecular docking revealed that these compounds interact with key residues in the MTH1 binding pocket, with QT showing the highest binding affinity  $(3.8 \times 10^7 \text{ M}^{-1})$ , followed by TQ, RV, and BC. Experimental validation confirmed the inhibitory effects of these phytochemicals on MTH1 activity and their cytotoxic potential against breast cancer cells. This study underscores the value of integrating computational and experimental approaches in drug discovery, demonstrating the therapeutic promise of natural compounds targeting MTH1 to manage oxidative stress and advance cancer treatment strategies.

IL-20

# Modulatory effects of Vinclozolin on Cytochrome P450 and its estimation using in-silico approach

## Haroon Habib Beigh

Faculty of Medicine Alatoo International University Bishkek Krygz Republic haroonhabib2@gmail.com



Dr. Beigh, MSc, Ph.D. Toxicology, is currently an associate professor at the Faculty of Medicine, AlaToo International University, Kyrgyz Republic. He has expertise in reproductive Toxicology, with a profound knowledge of in-silico techniques. His research interests are fungicide and their implication on the male reproductive system. He has an insight into drug safety which he gained while working in the pharmaceutical industry and is currently contributing to the field of Toxicology as a Researcher and educator as well looking forward to the Use of alternative animal methods in his research and education.

### Abstract

Cytochrome P450 family is distributed in all living populations in the world. There are more than fifty thousand Cytochrome P450 genes reported to date. It has been put forth that there are almost sixty cytochrome P450 genes in humans. We studied CYP 11A1, and CYP 17A1 (in- silico) which are considered to participate in steroid metabolism in testicular tissue. Molecular docking was done to check the binding efficacy of vinclozolin, a fungicide with CYP 11A1 and CYP 17A1 against standard estradiol. Enzyme structure was selected from the protein Data Bank for CYP 11A1, and CYP 17A1, standard and vinclozolin respectively. Schrodinger program Maestro 10.5 Molecular docking software has been used, PPW tool and Chem Draw 12.0 software were used to draw the structure of vinclozolin metabolites. Docking scores and binding scores were compared. Qikprop tool was used to determine the percent oral absorption. Due to the role of cytochrome P450 in androgenic activity, it is considered as a promising target for drugs. Results indicated possible interaction of fungicide Vinclozolin with CYP 11A1, and CYP 17A1 suggesting possible reprotoxic effects.

# Regenerating zebrafish - A novel model for effectual screening

IL-21

## Idris M

CSIR-CCMB, Hyderabad, India <u>idris@ccmb.res.in</u>



Dr. Idris is currently a senior principal scientist at CCMB, Hyderabad, India. His current research interests are concerned with understanding the complexity of developmental biology and neuroscience using alternate model animals such as zebrafish (Danio rerio), marine chordates (Ascidians) and echinoderms (Asterias sp). His research activity mainly focuses in understanding the mechanism of regeneration and degeneration in these model animals involving proteomics and transcriptomics approaches. Regeneration of appendages in zebrafish, nervous tissue in ascidians and arms in Asterias sp are the few important ongoing research activities in his lab. He is also interested in understanding the molecular and functional mechanism of neurodegeneration due to the triplet repeat expansion as like in spino cerebellar ataxia and Huntington's disease using zebrafish as the model animal.

#### Abstract

Our lab investigates the biomechanisms of tissue regeneration using alternative animal models, particularly zebrafish. As a vertebrate model, zebrafish exhibit remarkable regenerative capacity, particularly in the rapid and complete regrowth of amputated caudal fins. We leverage this system to examine the effects of small molecules, toxicants, and biomodulators in adult zebrafish across critical stages of regeneration, including dedifferentiation and redifferentiation. These studies provide valuable insights into the molecular and cellular processes underlying tissue regeneration and have potential applications in regenerative medicine and toxicology.

IL-22

## Heavy metal stress adaptation in fishes increases with their compromised health state

#### Nazura Usmani

Department of Zoology, Aligarh Muslim University, Aligarh, 202002, India nazurausmani@gmail.com



Professor Nazura Usmani had been associated with the teaching of Diploma in Pisciculture and undergraduate and postgraduate (Chemical Biology, Aquaculture and Fish Biology) courses in the Department of Zoology, prior to induction as a permanent Faculty in the Women's College (2006). Her research is focused on (a) the Identification of water bodies influenced by anthropogenic activities at different locations (b) Impact of xenobiotics primarily heavy metals on the health of various warm and cold-water fishes (as bioindicator) using biochemical, histological and molecular biomarkers (c) Immunotoxicity (d) Stress in research scholars. Her team has the credit of sequence submission metallothionein CDS sequence from Wild Walking Catfish Clarias batrachus obtained from a culture pond at NCBI. She has published over 40 papers in refereed journals including those in Frontiers in Nutrition, Molecular and Cellular Biochemistry, Environmental pollution, Scientific Reports (Nature), Chemosphere, Ecotoxicology and Environmental Safety, Comparative Biochemistry and Physiology Part C: Toxicology & Pharmacology, Archives of Environmental Contamination and Toxicology, Saudi Journal of Biological Sciences, Environmental Monitoring and Assessment, Proceedings of National Academy of Sciences, India, Section B Biological Sciences, Aquaculture Nutrition, Aquaculture Research, Journal of World Aquaculture Society., etc. with citations count 2700, H-index 25, and i10-index 32. She has supervised 4 Ph.D.'s, 2 undergraduate stem fellows and has a present team of 4 scholars. She qualified UPSC, Assistant Director Fisheries (2006). She was awarded the Stem Mentor Fellow Award Under US Department of State (OHIO State University, INDO-US collaboration STEM (2017). Her team was awarded Prof. Mahdi Hasan Memorial Award for best experimental design. She has taken training in Genomics and Molecular biology.

#### Abstract

The increase in metallothionein and glutathione peroxidase expressions are linked with increased oxidative stress. The heavy metals are found to elicit the mechanism of generating free radicals, thus increasing the body burden. The digestive system, which also gets exposed to such toxicants, may have an impact on its functioning. The digestive enzymes like amylase and trypsin were found to have regular activity in both carps and catfishes. Owing to the difference in anatomical structures, amylase and trypsin activity were recorded to be highest in the foregut (intestinal bulb) in carps followed by a gradual decline in mid and hindgut sub-sections. Catfishes had increased activity in the stomach, followed by a gradual decline in sub-sections. The variation in the serum bilirubin values in both carps and catfishes reflects upon the poor liver functioning. The majorly affected parameter was the protein concentration in muscle tissues of fishes. Increased proteolysis and lipid peroxidation are the reasons attributed to fishes exposed to toxic metals. The histopathological alterations were less due to single metal exposure in *Clarias batrachus*. The adaptability of organisms towards toxic environments have found to help them thrive in unsuitable aquatic bodies. The elicitation of metal-specific genes, i.e. metallothionein in both metabolically active and less-metabolic tissues, reflects the active defense state in fishes. It appears from the values that the fishes have successfully adapted themselves over some time and have mechanised itself to thrive well in toxic environments.

# 4D Scaffolds: Adding a new dimension to tissue engineering

## **Gopinath Packirisamy**

Department of Biosciences and Bioengineering, Indian Institute of Technology Roorkee, Roorkee -247 667, Uttarakhand, India. <u>gopi@bt.iitr.ac.in</u>



Dr. Packirisamy is a Professor at the Department of Biosciences and Bioengineering, IIT-Roorkee, India. His research expertise is in biomedical nanotechnology, drug delivery, biosensors, and tissue engineering. He has an excellent research record as many Indian patents are granted to him several others are filed.

## Abstract

With the growing number of literatures on the clinical importance of 3D scaffolds in tissue engineering, a number of them have been commercialized and have already made their way into cell culture laboratories around the world. Although such scaffolds support 3D cell growth, they barely recapitulate in-vivo dynamic native conditions in which the cell exists in an organism and because of such altered conditions (static 2D or 3D in-vitro culture conditions), they tend to lose their native physiology when cultured in. Such discrepancies in culture conditions contribute to a greater extent to the fact that in-vitro experiments do not always correlate well with in vivo results. Thus, to overcome this and enable cells to retain their native characteristics, the scaffold outlined here effectively mimics the dynamic in-vivo conditions of cells apart from supporting 3D growth and proliferation of cells. The hybrid multilayered scaffold will have a significant impact on tissue engineering research.

# Customized engineering and pharma applications of bioprinting

Piyush Padmanabhan, CEO Co-Founder & Director, Next Big Innovation Labs, Bengaluru, India piyush@nextbiginnovationlabs.com



Piyush Padmanabhan is the CEO and Co-Founder of Next Big Innovations Lab (NBIL), India. His core training has been in the domain of biotechnology and pharmaceuticals. His biggest achievement to date was learning, comprehending, and applying key engineering concepts and leading a team of engineers and designers to build not one but two fully operational 3D Bioprinters. These 3D Bioprinters are currently used at Next Big Innovation Labs for the development of 3D Bioprinted Skin.

# Studying neurological diseases in *C. elegans*: PTR-10, the homolog of human PTCHD1, plays a crucial role in neuroprotection

## Dr. Aamir Nazir

Senior Principal Scientist (CSIR) and Professor (AcSIR) CSIR – Central Drug Research InstituteLucknow, 226031, INDIA anazir@cdri.res.in



Dr. Aamir Nazir obtained his PhD from CSIR Indian Institute of Toxicology Research, Lucknow, and then went on for Postdoctoral training at Medical College of Georgia USA where he studied the Mechanism of Anesthetic action employing genetic model system C. elegans. Thereafter he obtained advance training from the Weizmann Institute of Science, Israel, the University of Nottingham UK, and the University of Freiburg, Germany. He joined CSIR-CDRI as a Scientist and established a research laboratory that focuses on the area of protein quality control employing functional genomics and epigenetics tools within the genetic model system C. elegans. He has established a very precious repository with more than 80 transgenic and mutant lines of the nematode which are used for addressing questions related to various aspects of neurobiology. His group holds the credit of discovering a novel circular RNA molecule called circ-Zip-2 in C. elegans which has relevance in the cause of Parkinson's disease. His group has also identified and functionally characterized novel peptide conjugates and a novel Insulin Degrading Enzyme termed "ce-IDE-1" in C. elegans with relevance to neurodegenerative diseases. Dr. Nazir has numerous accomplishments to his credit; he has been awarded with prestigious "Raman Research Fellowship", by CSIR, Govt of India, and the "India Distinguished Visiting Fellowship" by the University of Nottingham, UK. He is an elected Fellow of the Society of Applied Biotechnology, India; he serves on the Editorial board of several international journals and is associated with multiple scientific societies and other professional bodies. Dr. Nazir

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is an author on more than 80 scientific publications (h-index 33). The research work from his group has received wide recognition and has been well funded by the Council of Scientific and Industrial Research (CSIR), Department of Science and Technology (DST) and Indian Council of Medical Research, Govt. of India.

### Abstract

The role of glial cells in neuroprotection and the progression of neurodegenerative diseases is increasingly recognized, particularly in the context of aging. C. elegans offers precious genetics and neuronal circuitry towards studying the neuron-glia association. In C. elegans, the glia-enriched gene PTR-10 (human orthologue PTCHD1) plays a crucial role in maintaining neuronal integrity. This study explores PTR-10 and its downstream targets, basl-1 (orthologue of dopa decarboxylase, DDC) and daf-18 (orthologue of PTEN), to understand their roles in neurodegeneration and neuroprotection during aging. Using wild-type and transgenic C. elegans strains, we performed behavioral assays, lifespan studies, and assayed 6-OHDA-based injury models. Transcriptomic analysis of the PTR-10 knockout strain (RB1693) revealed significant downregulation of basl-1 and daf-18, both implicated in axonal regeneration pathways. Our results show that knockdown of basl-1 increases alpha-synuclein expression, shortens lifespan, and disrupts dopaminergic neuron function, all of which are associated with aging and neurodegeneration. Similarly, daf-18 knockdown also led to increased alpha-synuclein expression and reduced lifespan. PTR-10 expression itself declines with age exacerbating these effects and its absence impacts neuronal repair even in the presence of neuronal repair agents. In summary, PTR-10 and its downstream targets are critical in age-related neurodegeneration and neuroprotection. These findings highlight the potential of this axis for therapeutic strategies aimed at mitigating neurodegenerative diseases by promoting axonal regeneration and maintaining neuronal health thereby promoting healthy aging

IL-26

# Drosophila as a research model for studying neurodegenerative diseases

## Yasir Hasan Siddique

Laboratory of Alternative Animal Models, Section of Genetics, Department of Zoology, Faculty of Life Sciences, Aligarh Muslim University, Aligarh, 202002, Uttar Pradesh, India. <u>yasir\_hasansiddique@rediffmail.com</u>



Dr. Siddique is a Professor at Department of Zoology, Aligarh Muslim University, Aligarh, India. His team working on neurodegenerative disorders using transgenic Drosophila as a study model for Parkinson's Disease (expressing human alpha-synuclein) and Alzheimer's disease (expressing A $\beta$ -42). He also worked on the toxicity of various synthetic progestins and anti-cancerous drugs and on the protective effects of natural plant products (in vitro as well as in vivo) and proposed the mechanism of steroid toxicity and possible mechanisms of scavenging the free radicals by natural anti-oxidants.

### Abstract

*Drosophila melanogaster* is widely used as a research model due to the presence of about 50% homology with mammalian proteins. Nowadays there has been a worldwide effort to reduce the use of higher animals in research and testing. To study the pathogenesis of neurodegenerative diseases *Drosophila* has been established as a most successful model. The brain of *Drosophila* can be easily access and various cognitive parameters can also be performed easily which further makes this model ideal to study the cognitive dysfunctions associated with various neurodegenerative disorders. It has been also recommended by the European Centre for the Validation of Alternative Methods (EVCAM) for its use in research. This model is widely used for the study of various pathological processes associated with human neurodegenerative diseases.

IL-27

# Using C. elegans as a model system to understand molecular mechanisms of longevity assurance

## Arnab Mukhopadhyay

National Institute of Immunology, New Delhi arnab@nii.ac.in



His current designation is Staff Scientist VII at the National Institute of Immunology (NII), New Delhi. He completed his BSc (Botany, 1995, Presidency College Calcutta), MSc (1997), and PhD (2003) in Plant Molecular Biology from Delhi University South Campus followed by a postdoctoral stint (2003-2008) at UMASS Medical School, Worcester, Massachusetts, USA. Since 2009, his lab at NII has been studying various molecular mechanisms of aging and longevity assurance using *Caenorhabditis elegans* as the model system. He is an elected fellow of the Indian National Science Academy (INSA), The Indian Academy of Science (IASc), the National Academy of Science, India (NASI), and the Guha Research Conference Society (GRC). He is a recipient of the Ramalingaswami Re-entry Fellowship (2009-2014), the National Bioscience Award for Career Development (2016), and SERB-Science Technology Award for Research (STAR) award (2019), and the JC Bose National Fellowship (2023).

#### Abstract

Aging is a conserved biological process that afflicts every human being, often culminating in their death. Aging also predisposes us to many age-related diseases like type 2 diabetes, cancer, and neurodegenerative disorders that accelerate our demise, even at an early age. Considering the universal and fundamental nature of this process, and that the human race has pondered over it since time immemorial, both in art and in science, one would expect scientists to know a lot about what causes it. On the contrary, we still know relatively little about this process in terms of molecular mechanisms. Thus, it is important to understand the mechanisms of aging to design pharmacological interventions to delay or prevent the onset of the diseases of aging, if not to attain immortality. While immortality is a distant dream today, we do know of interventions, both genetic as well as non-genetic, that can delay aging, and increase life and health span, even in mammals. I will discuss our current understanding of the biology of aging and how a simple invertebrate research organism has been used to tease out molecular mechanisms of an intervention that dramatically enhances longevity and delays age-associated diseases.

IL-28

# Drosophila as an alternate animal model for the male reproductive toxicity assessment

# Ravi Ram Kristipati

Embryotoxicology Laboratory, Systems Toxicology Group (FEST), CSIR-Indian Institute of Toxicology Research, 31 MG Marg, Lucknow, 226001. India. raviram@iitr.res.in



Dr. Ravi Ram Kristipati obtained his Ph.D. in Zoology from the University of Mysore in 2002. Subsequently, he did his postdoc at one of the famous IVY league universities, Cornell University, USA, until 2008. In 2008, he joined the Council of Scientific and Industrial Research (CSIR), the premier research organization in the Country, as a Scientist. He is currently working as a Senior Principal Scientist at CSIR-Indian Institute of Toxicology Research, Lucknow. He is also a Professor at the Academy of Scientific and Innovative Research, an Institution of national importance. Dr. Ravi Ram's lab is focused on understanding the genetic basis of environmental chemical-mediated endocrine toxicity and associated complications, including fertility and diabetes by using a versatile Drosophila as a model. Dr. Ravi Ram has more than 50 research publications in highly reputed international journals, including Nature. His research work has been included in the guideline documents of regulatory agencies, such as the OECD and EPA, as evidence for the toxic impact of chemicals, even in lower organisms. His research is funded by grants from CSIR, ICMR, SERB, DHR, and DST. He is also an expert member of the toxicology task force of ICMR, New Delhi, and a fellow of the Society of Toxicology, India.

#### Abstract

Infertility is a significant global public health issue affecting one in six individuals worldwide. The rise in male infertility, in part, has been linked to exposure to environmental chemicals and pollutants, prompting the need to evaluate these xenobiotics for their effects on male fertility. Traditional reproductive toxicity assessments rely on animal models, such as rodents and non-human primates, but this approach faces significant challenges, including the toxicant load, regulatory limitations and ethical concerns, which hinder high-throughput testing. To reduce reliance on laboratory animals, there is a strong focus on developing alternative testing methods. However, as of now, there is no validated *in vivo* alternative for assessing male reproductive toxicity. To address this issue, my lab has been utilizing *Drosophila melanogaster*, an excellent invertebrate model that shares conserved reproductive processes and molecules with mammals. Our research has demonstrated that Drosophila effectively recapitulates the male reproductive toxicity phenotypes observed in mammals when exposed to a known male reproductive toxicant, di-butyl phthalate (DBP). The presentation will reflect on the potential of Drosophila as an alternate animal model for pre-screening chemicals for reproductive hazards and will provide essential insights into chemical-induced endocrine disruption.

# Caenorhabditis elegans a model for nanomaterial safety assessment

## **Aruna Satish**

Principal Scientist, Ecotoxicology Laboratory, Environmental Toxicology Group, FEST Division, CSIR-Indian Institute of Toxicology Research, Lucknow

aruna@iitr.res.in



Dr. Aruna Satish is a Principal Scientist at Environmental Toxicology CSIR-IITR, Lucknow, India. Her research specialization includes nano and ecotoxicology, xenobiotics & obesity, aging & neurodegenerative diseases.

### Abstract

Nanotechnology deals with particles a billionth of a meter (nanometer). These nano-size atoms/molecules have novel properties and functions because of their small size, shape, high surface area, charge, chemical properties, solubility, and degree of agglomeration. They are called nanomaterials (NMs). NMs possess infinite market potential globally and are extensively employed in various fields such as medicine, agriculture, energy, communications, materials, and electronics. The enormous usage of NMs increases environmental exposure. However, the consequences of exposure to NMs on health and the environment are poorly understood and warrant investigation. Towards this direction, we employ *Caenorhabditis elegans*, a free-living nematode, as an *in vivo* model for the safety evaluation of NMs, and the same will be discussed.

IL-30

# Developmental and neurological effects of arsenic on Drosophila melanogaster

Jawaid Ahsan Senior Assistant Professor Department of Biotechnology Central University of South Bihar, Gaya, Bihar jahsan@cub.ac.in



Dr. Ahsan is a senior Assistant Professor in the Department of Biotechnology, CUSB, Gaya, India. His areas of research interest include neuroscience, Drosophila neurobiology, and neurotoxicology. He has several awards and recognitions to his credit.

## Abstract

Millions of people in developing countries are affected by arsenic (As) toxicity and its prevalence. Arsenic's detrimental effects on humans have been amplified by an unacceptable level of exposure to food and drinking

water, the ongoing rise in industrial usage, and several other occupational conditions. Due to increased cellular absorption and the ability to cross the blood-brain barrier (BBB), inorganic arsenic (iAs) is extremely hazardous to living organisms in its trivalent form. Arsenic toxicity damages an organism's tissues and organs, resulting in skin cancer, circulatory system abnormalities, and central nervous system disorders. A competent model system is required to investigate the acute effects of arsenic on the brain, and cognition ability, and to assess any behavioral impairment. *Drosophila melanogaster* (common fruit fly or vinegar fly), with its short generation time, genomic similarities with humans, and its availability for robust behavioral paradigms, may be considered an ideal alternative model for studying arsenic toxicity. The present study helps to understand the toxic effects of acute arsenic treatment on the behavior, cognition, and development of Drosophila in a dose and time-dependent manner. We found that the exposure of fruit flies to arsenic significantly affected their life span, pupae size, locomotor abilities, cognitive functions, and neurobehavioral impairment. Hence, providing a better understanding of how arsenic toxicity affects the brain leading to acute behavioral disorders and neurological alterations, this study will lead to a better understanding of the mechanisms and reversal of the ill-effect.

IL-31

# Stop and use non-animal methods: How the SUN Project is illuminating the path forward

#### Savita Nutan

Founder, Medicine Without Cruelty London, UK savita@medicinewithoutcruelty.com



Savita is a passionate advocate for animal-free medical research. She has studied, trained and worked within the medical sciences and has seen firsthand how potential medicine tested on animals has kept medical research from moving forward. She hopes to educate and explore animal-free innovative technologies, as well as highlight animal cruelty facts, which can create real change.

#### Abstract

Animal testing is an outdated and ineffective practice that consistently fails to predict human responses, leading to scientific setbacks, delayed medical advancements, and immense, preventable suffering. Every year, millions of animals endure severe pain, stress, and death in laboratory experiments, despite the overwhelming evidence that animal testing produces unreliable results due to fundamental physiological and genetic differences between animals and humans. Pharmaceutical drugs deemed safe in animals repeatedly fail in human trials, and with a 92% drug failure rate, it underscores the urgent need for immediate change. The SUN Project is committed to eliminating these obsolete practices from science curricula by equipping educators with science modules centered on human-relevant, humane alternatives. This initiative introduces students to advanced technologies such as organ-on-a-chip systems, human cell-based testing, and computational models-methods that are not only ethically sound but also demonstrate unparalleled accuracy in predicting human outcomes. By integrating these cutting-edge tools into education, the SUN Project aims to replace cruelty with compassion and ineffective testing with effective, reliable scientific approaches. It is both unethical and scientifically irresponsible to continue relying on animal testing in educational institutions. Given the superior accuracy and ethical foundation of modern, human-focused methods, the time for modern science is now. Recent policy shifts, such as the FDA's recognition of non-animal testing methods as reliable, highlight the necessity for educational institutions to lead the way in adopting and teaching advanced, humane methods.

# Organoid models as intermediate disease modelling platforms

### Dr B Kiran Kumar

CSIR-Center for Cellular and Molecular Biology, ANNEXE II, Medical Biotechnology Complex, Uppal Road, Hyderabad, Telangana, India, bokarakiran@ccmb.res.in



Dr. Bokara Kiran Kumar is a Senior Scientist at the CSIR- Center for Cellular and Molecular Biology (CSIR-CCMB), Hyderabad. He completed his Ph.D. in Zoology at Andhra University, focusing on the toxic impact of sublethal lead exposure on crustaceans. His research spans neurodegenerative disorders, stem cell therapy, corneal regeneration using 3D bioprinting, and SARS-CoV-2 antiviral drug platforms. He has also explored lead toxicity in the brain and its effects on antioxidant systems. Dr. Kumar has held prestigious roles, including a Postdoctoral Fellow at Yonsei University, South Korea, where he worked on neural stem cell characterization. His accolades include the Young Scientist Award from the Indian Academy of Biomedical Sciences in 2023 and numerous memberships in scientific societies. He has contributed significantly to academia with over 50 research publications, book chapters, and patents in stem cell and molecular biology. He has mentored numerous students and researchers and collaborated with esteemed institutes like IIT Hyderabad and LV Prasad Eye Institute. His ongoing projects involve biomimetic hydrogels for corneal diseases and organoid models for biomedical applications. Dr. Kumar's work demonstrates a multidisciplinary approach to tackling significant biomedical challenges.

## Abstract

Organoids, three-dimensional cellular structures derived from stem cells, closely mimic the architecture and functionality of human tissues and organs. This innovative technology bridges the gap between in vitro cell cultures and in vivo models, offering an invaluable tool for understanding complex human diseases. By replicating the cellular microenvironment and tissue-specific interactions, organoids provide a unique platform for studying disease pathogenesis, drug responses, and patient-specific therapeutic interventions. Their scalability and reproducibility make them suitable for high-throughput drug screening and personalized medicine approaches. This platform holds promise for accelerating translational research, enabling more accurate disease modeling, and reducing dependency on animal models. As intermediate systems, organoids stand at the forefront of biomedical innovation, paving the way for a deeper understanding of human diseases and the development of targeted therapies.

## Illuminating lymphatic biology using zebrafish model system

Rudra N Das Shiv Nadar University <u>rudra.das@snu.edu.in</u>



Dr. Das is an assistant professor at the School of Natural Sciences, Shiv Nadar University, Noida. His major research interest is vascular biology, lymphatic system, developmental biology, regeneration, and genomics.

#### Abstract

Endothelial cells (ECs) build the elaborate lymphatic and blood vessel networks that are associated with every organ system of the vertebrate body. This omnipresence of endothelial cell-derived vasculature underscores their vital role in nearly every physiological process. While blood vessels have received significant attention, literature on the lymphatic system is severely limited. The well-known role of the lymphatic system includes fluid homeostasis, immune function, and lipid metabolism. Recent studies have uncovered several new functions of lymphatics, such as the regulation of organogenesis and organ-repair, strongly suggesting that the system offers a wealth of information in understanding vascular biology and greater efforts are needed to explore them. Challenges such as the lack of robust models, difficulties in identifying lymphatics, and experimental inaccessibility have contributed to the limited progress in lymphatic biology research. Thus, there is a need for model systems that allow for an integrated analysis of development, heterogeneity, and function. Zebrafish lymphatic system shares a high degree of conservation with its mammalian counterpart. The fish provides advantages in time-lapse live imaging and the ability to monitor changes in the same individual over developmental timescales. Furthermore, the fish is an excellent model to study tissue repair and regeneration. Over the years, zebrafish has emerged as a successful system to model several human diseases, owing to the ease of targeted mutagenesis and therapeutic compound screenings. Well-established transgenic techniques for overexpression/misexpression and CRISPR- mediated mutagenesis allow for the interrogation of gene function. Furthermore, studies of cellular diversity have been enabled by single cell RNA sequencing techniques that are fully optimized for fish tissues. Through this presentation, we explore how zebrafish are pushing the boundaries of vascular biology research. At my research group, we focus on identifying organ-specific lymphatic specializations and the functional significance of those. In this presentation, I will showcase key findings from our work, including the discovery of specialized blood vessels that are derived from lymphatic, a plasticity that was previously unrecognized (Das et.al., Nature, 2022). I will also discuss our exploration of lymphatics-associated with various other organs, such as the eye and the intestine.

#### IL-34

# Metabolomics in animals and alternatives to animal models: An emerging comprehensive screening tool for toxicological applications

## Mohana Krishna Reddy Mudiam, Ph.D.,

Institute of Pesticide Formulation Technology, Sector-20, Udyog Vihar, Gurugram – 122 016, Haryana State director@ipft.gov.in



Dr. Mudiam is a Director at the Institute of Pesticide Formulation Technology, Gurugram, India. His research areas include analytical method development and validation, metabolomics for toxicology, food Safety and phyto applications, xenobiotics residue analysis, environmental monitoring and biomonitoring, biosimilars characterization, regulatory affairs, untargeted contaminants analysis, pharmaceutical analysis including impurity profiling and forced degradation studies.

## Abstract

The animals plays a vital role in the toxicology research to assess the safety of chemicals. Due to the use of large number of animals for toxicological experiments has raised alarming attention and made it necessary to find alternatives to it. The introduction of 3R principles has able induce momentum to find and use the alternatives to animals for toxicological testing, some of them approved by the regulators. The toxicology testing is an essential tool to assess the impact of chemicals on various organisms including humans. However, conventional toxicological assays require large number of animals for number of toxicological assays, which are time-consuming and tedious. In recent times, metabolomics has emerged as the fastest-developing tool in various research fields and has becoming as an emerging toxicological screening tool to assess the chemical safety with minimal use of animals and alternatives to animals. The metabolomics ideally deals with the evaluation of metabolic perturbations in the chosen biological samples at defined temporal conditions using multi-platform analytical techniques. The techniques like LC/GC-MS/MS and NMR were found to be versatile tools for performing metabolite identification and quantitation along with chemometrics. In recent times, quantitative metabolomics has gained importance in understanding the accurate variations in biological systems. Metabolomics finds applications in drug discovery and development for toxicological assays, phytopharmaceuticals development and even predicting the mechanistic insights about the contaminant action on various model organisms to understand their impact on human health. In my talk, I will be able to elaborate on the importance of metabolomics in chemical safety with the use of various model organisms including animals, and alternatives to animals.

# A digital platform for animal research

Dr. Umesh Prasad IT College, Lucknow, India uprasad.itcollege@yahoo.com



His expertise is in the area of health informatics with special reference to application of IT tools for risk assessment of exposure to chemicals in occupation settings.

IL-36

# Network Pharmacology: Next generation alternative in biomedical research

### **Anoop Kumar**

Department of Pharmacology, Delhi Pharmaceutical Sciences and Research University (DPSRU), Govt. of NCT of Delhi, New Delhi, India





Dr. Anoop Kumar is currently an Assistant Professor in the Department of Pharmacology, Delhi Institute of Pharmaceutical Education and Research (DIPSAR), Delhi Pharmaceutical Sciences & Research University (DPSRU), New Delhi. Recently, he has also been included in top 2% list of scientists released by Stanford. Earlier, he worked as an Assistant Professor and Head in the Department of Pharmacology and Toxicology, National Institute of Pharmaceutical Education and Research (NIPER), Raebareli, and as an Officiating Head and Associate Professor in the Department of Pharmacology, ISF College of Pharmacy, Moga, Punjab. He has also worked as a Research Scientist in the Medical Affairs and Clinical Research Department of Sun Pharmaceutical Industries Limited, Gurugram, and Translational Health Science Institute (THSTI), Faridabad. He has authored more than 120 research and review articles and 20 book chapters in International Journals and publishers of repute. His current h-index is 26, and i10-index 64 with citations of 2763. He has also filed 04 Indian patents. He has guided 42 postgraduate research students and 02 PhD students. He has also edited 05 International books. He is also an Associate editor and editorial board member of various International Journals of Elsevier, Wiley, and Springer Nature. He has also delivered more than 50 expert talks in International and National Conferences. He has also worked as Member Secretary of the Institutional Animal Ethics Committee (IAEC) and Animal House In-charge at ISF, Moga. Currently, he is working as member secretary of human ethics committee approved by CDSCO and Treasurer of ISPOR India Chapter. His lab also gets funding from DST SERB SRG (30 lakhs), ICMR Mission Project (1.3 crores), ICMR Investigator Initiated Project (65 lakhs & 1.2 crores), National Innovation Foundation (NIF)-India (10 lakhs) and IIT-Delhi (10 lakhs). His research interests are in the following areas: 1) Drug

IL-35

repurposing 2) Meta-analysis of clinical trials 3) Network Pharmacology 4) Pharmacoeconomic studies 5) Signal analysis in Pharmacovigilance.

## Abstract

Computational biology, bioinformatics, artificial intelligence, and big data science, medical and life science research have entered the era of big data. Globally researchers have moved from "Reductionist theory" to a "System theory" from a single isolated research mode to a multi-faceted and systematic one. Network pharmacology is an emerging discipline, that integrates system biology, multidirectional pharmacology, computational biology, network science, and other related disciplines. The advancement in high throughput experimental technologies, and computing methods represented by big data and artificial intelligence have also effectively promoted the development and wider application of network pharmacology. Therefore, Network Pharmacology is also considered as Next generation alternative in biomedical research. This expert talk will cover the basics of network pharmacology along with principles and processes with suitable case studies. The advantages as well as limitations will also be covered along with glimpse of work done so far in our lab at DPSRU, New Delhi.

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# Insights from flies: Drosophila as a powerful model to study interactions and intricate pathways of reproduction

Snigdha Misra UPES, Dehradun snigdha.misra@ddn.upes.ac.in



Dr. Snigdha Mishra is an avid fly researcher. While working on Drosophila melanogaster, a widely acclaimed invertebrate model for genetics, she received her Ph. D from CSIR- Indian Institute of Toxicology Research and later joined the Wolfner lab at Cornell University, Ithaca, NY, USA, to work on male-derived reproductive proteins and how they modulate the physiology and behavior of females, post-transfer. Dr. Mishra has publications in journals of international repute, including Elife, BMC Biology, PLoS Genetics, Toxicological Sciences, Scientific Reports, Phil Trans Royal Sci B and likewise. Dr. Mishra is currently an Assistant Professor at the School Of Health Sciences and Technology, UPES Dehradun, where she is heading the fly lab as a part of the Model Organism Platform (MoP facility) at UPES. Her lab addresses questions on mechanistic insights into reproductive and metabolic ailments employing fruit flies as the model of research.

#### Abstract

Interactions between sperm and the female reproductive tract (FRT) are essential for the fertility and reproductive success of the mating pair. These interactions not only help sperm move through the FRT but also involve crosstalk in the form of signals between sperm and the cells lining the female reproductive tract. These signals are important for sperm to reach the oviduct, trigger ovarian secretions, and undergo changes that help them become capable of fertilizing an egg. However, very little is known about the details of the FRT components, how they change the sperm, or how they affect sperm movement, survival, and ability to fertilize an egg. To identify such female components, we examined the spatial and temporal characteristics of the association of seminal proteins with sperm before and after their transfer to the FRT and disabled two secretory tissues in the FRT (spermathecal secretory cells (SSCs) and parovaria (female accessory glands) using Hr39 mutations. We showed that the female components modulate priming of sperm with seminal proteins, post-mating. And that, the secretions from SSCs and parovaria regulate the gradual release of sperm-associated seminal protein, Sex peptide (SP), which is important for the long-term persistence of its effects on mated females' behavior and physiology.

# **PeTA Session I**

# Accelerating the application of non-animal approaches in toxicity testing of pesticides in India

## Dr Ankita Pandey

Senior Science Policy Advisor People for the Ethical Treatment of Animals (PETA), India

#### Abstract

Over the past decade, the scientific community has made substantial advancements in developing reliable, humanrelevant testing approaches that are poised to replace and reduce tests on animals. Regulatory agencies worldwide are recognizing the importance of using data from these testing approaches in chemical evaluations to better protect human health and the environment. In 2023 updates to its guidelines for chemical pesticides and biopesticides, the Central Insecticides Board & Registration Committee (CIB&RC) accepted the use of nonanimal approaches including in vitro and in silico approaches, read-across, and waivers based on a weight-ofevidence (WoE) approach to meet data requirements for all categories of pesticides. Transitioning the current testing paradigm to a more robust risk assessment framework through the application of scientifically fit-forpurpose non-animal methods was discussed in the 'Webinar Series on the Use of New Approach Methodologies for the Risk Assessment of Pesticides' and during the regulatory session on pesticides conducted at the 6<sup>th</sup> International Conference of Society for Alternatives to Animal Experiments in India (SAAE-I) in 2023. The webinar series and SAAE session identified challenges to the regulatory uptake of non-animal approaches in India, including 1) lack of global harmonization, 2) lack of clarity on the acceptance of non-animal approaches, and 3) continued reliance on an animal-intensive check-box toxicity testing approach. This session aims to bring together Indian regulators, policymakers, and industry stakeholders to discuss critical needs within the current regulatory framework to support increased uptake of non-animal approaches. The session will convene diverse stakeholder perspectives, providing an opportunity to gather comprehensive feedback and shape targeted policy initiatives to advance the use of non-animal approaches in toxicity testing.

**Experts-1** 

# Using non-animal approaches for toxicity testing of pesticides: challenges and opportunities

### Dr. SK Jain

Senior Consultant (Medical Toxicology), Central Insecticides Board and Registration Committee, Ministry of Agriculture and Farmer's Welfare

# Abstract

The global shift towards non-animal methods has been fuelled by the need for more efficient, reliable, and fit-forpurpose testing approaches as well as animal welfare considerations. As regulatory decision makers evaluate how to apply the best science to protect human health and the environment across a diversity of chemical exposures, there is an increasing need to rely on non-animal approaches that provide more efficient and relevant insights into potential toxicities. Through its recent revision of guidelines, the Central Insecticides Board and Registration Committee (CIB&RC), Ministry of Agriculture and Farmer's Welfare has taken steps to adopt non-animal approaches, and there are remaining opportunities to further accelerate the pace of acceptance and implementation of such approaches for regulatory decision making. The talk will provide insight into the current pesticide regulatory testing landscape in India and globally, will outline regulatory challenges and opportunities towards adoption of new approaches that can improve pesticide toxicity evaluation, ensure international harmonization, and replace testing on animals.

**Experts-2** 

# Non-animal approaches in pesticide toxicity assessment: Industry perspective

## Mr Rajesh Dhawan

Head-Regulatory Affairs Group, CropLife India Senior Regulatory Specialist, Syngenta

## Abstract

Historically, pesticide hazard and risk assessment have relied on implementing cost and time-intensive tests on animals for active ingredients and formulations. The science underpinning pesticide toxicity testing methods has advanced remarkably, and the ability to implement modern non-animal approaches in assessing risks of such chemicals, with reduced reliance on animals, has dramatically increased. Globally, efforts are being made by the crop protection industry to implement fit-for-purpose methods for regulatory decision-making to better protect human health and the environment. It has thus become important to consider how to efficiently incorporate these scientific advancements in the Indian regulatory text to ensure global harmonization and ensure best use of time and resources. The talk will provide a snapshot of industry experience on the application of non-animal approaches, current regulatory barriers towards their successful implementation, and discuss further opportunities to accelerate their use for pesticide toxicity assessment.

# **PeTA Session II**

# Revolutionizing pyrogen testing: Non-animal approaches in India

### Dr Rohit Bisht,

Scientist-Pharmaceuticals and Medical Devices People for the Ethical Treatment of Animals (PETA), India

### Abstract

Pyrogen testing to detect the presence of fever-causing substances is an integral part of safety testing for vaccines, blood-derived products, medical devices, and other products. Historically, pyrogen tests have used animals, including the Rabbit Pyrogen Test (RPT) and the horseshoe crab-blood-based Limulus Amoebocyte Lysate (LAL) test, also called the Bacterial Endotoxin Test (BET). These tests have scientific issues that pose problems in light of the increasing complexity of drugs and medical devices, and their production processes. Non-animal methods are available that can replace the RPT and the BET/LAL. These methods, the recombinant Factor C (rFC) assay and the Monocyte Activation Test (MAT), still require product-specific evaluation to varying degrees as indicated in an increasing number of regulatory standards and other guidance. There is no formal process that ensures regulatory testing requirements modernize as quickly as new, reliable and human-relevant testing approaches are developed. The primary objective of PETA India's initiative is to collaborate with regulators, policymakers, and stakeholders to advance and implement non-animal pyrogen testing methods in alignment with global trends and the state-of-the-science. By fostering partnerships with these stakeholders, PETA India facilitates the exchange of knowledge and accelerates the adoption of innovative technologies by building consensus on the most appropriate and feasible steps toward this objective. Voices from broader scientific and policy sectors have helped identify a common set of challenges that all parties agree must be addressed before non-animal pyrogen testing can be routinely used and accepted for regulatory purposes in India. With this in mind, PETA India is coordinating opportunities for these stakeholders to inform a prioritized set of strategic goals to address these challenges in a way that is relevant to, and feasible for, these stakeholders.

**Experts-1** 

# Advancing Responsible Science through Sustainable Endotoxin Testing

## Sanjeev Singh,

Territory Sales Manager Charles River Laboratories India Pvt. Ltd

## Abstract

Charles River Laboratories offers optimized Endosafe® bacterial endotoxin testing solutions to continue to make mandatory safety testing faster, easier, and more reliable. As a result of our commitment to innovation the 3Rs, and sustainability, we've refined the next addition of our Endosafe bacterial endotoxin detection portfolio by introducing Endosafe Trillium, a robust recombinant cascade reagent (rCR) assay designed for sustainable progress. Trillium (rCR) detects and quantifies natural environmental endotoxins as well as endotoxin standards (RSE & CSE) by simulating the natural LAL enzymatic cascade through an optimized formulation and composition of three critical recombinant proteins (Recombinant Factor C, Recombinant Factor B, and Recombinant pro-clotting enzyme). This proprietary matrix demonstrates assay superiority in accuracy, comparability, robustness and has been proven to ensure its equivalency to LAL. In addition to being 100% animal-free, Endosafe Trillium rCR offers a host of benefits that will transform endotoxin testing program. This presentation will examine the benefits of adopting non-animal pyrogen testing methods in India, showcasing real-world applications, regulatory aspects.

- A kinetic chromogenic method, it is compatible with existing incubating absorbance plate readers.
- 3-Factor enzymatic cascade simulates the same cascade reaction as traditional LAL.
- Eliminates the potential for  $1,3-\beta$ -D-glucans, reducing the risk of false positives.
- Quantitative range: 0.001 to 100EU/mL
- Primary validation package includes protocols, summary report, and implementation support.

We are dedicated to constantly identifying technologies that reduce animal use while ensuring patient safety.

# **Experts-2**

# Sustainable BET testing by pyrogene rFC assay

## **Pratik Suthar**

Channel Manager- India and Middle East Lonza, India

#### Abstract

India's rapidly growing pharmaceutical and biotech industries are increasingly seeking efficient, ethical, and regulatory-compliant testing methods. Lonza, a global leader in Pharma and Biotech, is pioneering non-animal pyrogen testing platforms designed to meet the rigorous safety and efficacy standards required by regulatory bodies. These methods, including the monocyte activation test (MAT) and recombinant Factor C assays, offer reliable, accurate alternatives to traditional animal-based pyrogen and endotoxin testing methods, such as the rabbit pyrogen test and LAL test. In India, adopting non-animal pyrogen testing methods has gained significant traction as the industry shifts towards more ethical practices and aims to comply with international regulatory requirements. Lonza's innovative testing platforms provide significant advantages, such as faster results, lower costs, and improved reproducibility, while ensuring that pharmaceutical products, biologics, and medical devices meet high safety standards. This presentation will highlight the scientific principles behind Lonza's non-animal pyrogen testing methods, discuss their application in India's pharmaceutical and biotech sectors, and explore the regulatory landscape surrounding their adoption. Lonza's solutions represent a critical step towards a more ethical, cost-effective, and globally competitive future for India's pharmaceutical industry.

# **ORAL PRESENTATIONS**

**OP-1** 

# Evaluation of in-vitro hepatoprotective activity of *Cyanthillium cinereum* (Vernonia cinerea) against ethanol-induced hepatotoxicity

# <u>Pramuk Shiyan Adithya Rathnasooriya<sup>1</sup></u>, WGN Kaushalya<sup>1</sup>, HMDSN Rupasingha<sup>1</sup>, TS Suresh<sup>2,3</sup>, WMKM Ratnayake<sup>4</sup>, DVD Hemalika<sup>5</sup>, WAS Saroja Weerakoon<sup>6</sup>, KAAU Karunarathna<sup>3,7</sup>

<sup>1</sup>Department of Medical Laboratory Sciences, Faculty of Allied Health Sciences, University of Sri Jayewardenepura, Sri Lanka.

<sup>2</sup>Department of Biochemistry, Faculty of Medical Sciences, University of Sri Jayewardenepura, Sri Lanka <sup>3</sup>Center for Plant Materials and Natural Product Research, University of Sri Jayewardenepura, Sri Lanka. <sup>4</sup>Department of Cosmetic Sciences, Faculty of Health Sciences, CINEC Campus, Sri Lanka. <sup>5</sup>Department of Chemistry, Faculty of Natural Sciences, The Open University of Sri Lanka, Nawala, Nugegoda, Sri Lanka. <sup>6</sup>Faculty of Indigenous Medicine (FIM), University of Colombo, Sri Lanka and National Ayurveda Teaching Hospital, Colombo, Sri Lanka

<sup>7</sup>Department of Basic Sciences, Faculty of Allied Health Sciences, University of Sri Jayewardenepura, Sri Lanka.

shiyanadithya1999@gmail.com

## Abstract

Background: Cyanthillium cinereum has gained recognition for its potential therapeutic properties, particularly in treating liver-related disorders in the traditional medicinal system of Sri Lanka. Objectives-The study aims to determine the in-vitro hepatoprotective activity of 70% isopropyl alcohol (IPA) extract of C. cinereum against ethanol-induced hepatotoxicity. Methodology- The whole plant of C. cinereum was collected from Balangoda, Sri Lanka (Longitude: 80. 70266388888889 and Latitude: 6. 6334144) and authenticated (Accession number 3124). The IPA: water (70:30 v/v) extract of the dried plant materials was obtained by the maceration method and evaporated under vacuum to yield a dark green semi-solid (7.9 %). In-vitro hepatoprotective effect of IPA extract against ethanol-induced hepatotoxicity was assessed using porcine liver tissue obtained from a registered slaughterhouse, Ja-Ela, Sri Lanka. Accordingly, thin slices of liver tissues were exposed to the different concentrations of plant extract (200, 500, 1000, 2000, 3000, 4000, 10000 µg/mL) and absolute ethanol (15M) and incubated at 37 °C for 2 h (n = 3). The buffer solution was used as the negative control while absolute ethanol was used as the positive control. After the incubation, the spent media was assayed for aspartate transaminase (AST), alanine transaminase (ALT), and lactate dehydrogenase (LDH). Total enzyme activity was determined in the supernatants prepared by homogenizing the liver slices (n=5). Finally, the percentage cytotoxicity was calculated. Results and Discussion-The percentages of cytotoxicity of ALT, AST, and LDH in the positive control were  $42 \pm 1$ ,  $51 \pm 2$ , and  $59 \pm 2$  respectively, and they were significantly reduced when liver slices were co-exposed to ethanol with plant extract. The highest significant reduction was observed at 10000  $\mu$ g/mL compared to the positive control (19 ± 2.5, 17 ± 2.7 and 26 ± 3.2 against ALT, AST, and LDH respectively, p<0.05). Conclusion- IPA extract has the potential to act as a strong hepatoprotective agent against ethanol-induced hepatic damage.

OP-2

# In vitro alternative assays performed at Intox, an Aragen Life Sciences Company

## Yogini Soman and Jyoti Mane

INTOX, Pune, India

# Abstract

Historically information about the toxicity of a substance gained through rodent and other animal studies. These animal studies however take years to conduct and expensive due to the costs associated with housing and care of the animals. Additionally, there are sometimes limitations to the type of information these studies can provide. While adverse events such as weight change, tumour development, the appearance of lesions, etc can be observed in the animals, the studies often do not reveal the underlying pathophysiological mechanism behind such changes. Given the limitations associated with animal testing, there has been interest in developing new approaches (New Approach Methods: NAMs) which are faster, less expensive, and more informative. Of course, before any NAM is used to assess safety, it is critical that the validity and reliability of the method has been established. NAMs present an opportunity to: reduce the number of animals used in testing; refine the methods still requiring animals so they are less stressful to the animals; and to replace animal testing whenever possible. At Intox, validated alternative testing approaches (as listed below) are routinely employed in early

discovery stage to determine toxicity liability for series of compounds in short time and help identify right development candidate that can be tested in Phase I clinical trial.

- 1. Vegasoft for assessing genotoxicity potential using SAR and QSAR models.
- 2. Mini-Ames for genotoxicity potential

3. Mini-hERG to understand cardiovascular liability

4. In vitro Photox, to determine phototoxic potential

5.In vitro MNT to determine clastogenic and aneugenic potential

6.In vitro CA to determine clastogenic potential

7. Short-time exposure In vitro test to evaluate eye hazard potential

8. In vitro skin irritation by reconstructed human epidermis test to determine skin irritancy potential and hazard identification in accordance with UN GHS

9. In vitro skin corrosion study by transcutaneous electrical resistance test to determine the degree of corrosive potential

10. In vitro Cell gene mutation assay to determine mutagenic potential

The future of biological research and regulatory science is anticipated to be heavily reliant on NAMs as these technologies continue to advance. Intox will concentrate on the validation and sophistication of the models, providing a more accurate, efficient, and ethical method of testing.

OP-3

# Telomerase modulation in female breast cancer patients of arsenic affected region of Bihar

# Archana Chaudhary<sup>1</sup>, Nandani Kumari<sup>3</sup>, Manish Kumar2 , Lalit Mohan1, Rizwanul Haque3 , <u>Md.</u> <u>Margoob Ahmad<sup>1</sup></u> \*

<sup>1</sup>Department of Pharmacology, IGIMS-Patna,

<sup>2</sup>Department of Surgical Oncology IGIMS-Patna

<sup>3</sup>Department of Biotechnology, Central University of South Bihar, Gaya, Bihar, India



Dr. Md. Margoob Ahmad has received a Ph.D. Degree in Biotechnology from Jamia Millia Islamia, New Delhi, and M.Sc. in Biotechnology from Devi Ahilya University Indore. Dr. Ahmad has extensive experience in the areas of Molecular Oncology, Molecular Pharmacology, and Molecular Diagnostics. Dr. Ahmad is actively involved in research work. In the past, he was also involved in teaching of Graduate and Post Graduate students Biotechnology students at IMS Ghaziabad as an Assistant professor. In addition, he served as a Molecular Biologist at Quest Diagnostic Gurgaon, as well as a Research Scientist at Blood and Bone Marrow Transplant Research Center at Dharamshala Cancer Hospital Delhi. Dr. Md. Margoob Ahmad has made significant scientific contributions, evident from his 12 publications in a variety of top-class scientific journals and his research has been cited for more than 270 times by other research groups worldwide, with an h-index of 07 and i10- index of 6. He is serving as the PI and Co-PI in 4 Research projects such as the Indian Council of Medical Research (ICMR), Bihar Council on Science & Technology, Patna (BCST) & The Central Council for Research in Unani Medicine (CCRUM) Delhi.

# Abstract

Breast cancer seems to be the most frequent type of cancer worldwide at the moment, accounting for the majority of cancer-related deaths in women. The key to a favorable prognosis is early diagnosis and prevention. The capacity for infinite cell division, which renders cancer cells immortal, is one of the defining characteristics of the enzyme telomerase. Several recent studies in an arsenic-exposed human population found exposure was associated with altered telomere length, which is also attributed to enhanced chromosomal instability and cancer. The state of Bihar is an endemic belt for arsenic contamination in groundwater and the increased incidences of younger females with breast cancer in the state of Bihar has been a major challenge. The etiology of cancer incidences in

this area has not been revealed properly till now. Hence, the present study is an approach to decipher the root cause of the cancer incidences in the Gangetic basin of Bihar and its association with arsenic. Altogether, 20 female breast cancer patients were identified from OPD at the State Cancer Institute of IGIMS, Patna, Bihar, and both tumor and adjacent tissue along with blood samples were collected for the study at the time of surgery. The expression analysis of telomerase and protein was done through RT-PCR and western blotting. Arsenic concentration was tremendously high in 65% of female breast cancer along with upregulation of telomerase expression in that patient as confirmed by expression our studies. Our findings also revealed the significant association of telomerase overexpression in arsenic-affected areas of Bihar.

OP-4

# Towards understanding the anticancer role of gut microbial metabolites

# Dr. Anil Kumar

National Institute of Immunology, New Delhi, India



He is a scientist at the National Institute of Immunology, New Delhi. His expertise is in microbiome and human cancer. Several patents have been granted to him. CSIR Technology Award 2012, Merck Millipore India Innovation Award 2012, Young Scientist Award from IUMB-2006, Young Investigator Award from HUPO-2004, and several others are to his credit.

**OP -5** 

# Comprehensive evaluation of the anti-melanogenic and antioxidant properties of the benzoquinone derivative compound in in vitro and in vivo models

## <sup>1</sup>Parvathy CA, Akash Anilkumar U<sup>1</sup>, Sasidhar BS<sup>2</sup>, Sreejith P<sup>1</sup>

<sup>1</sup>Department of Zoology, Advanced Center for Regenerative Medicine and Stem cell Research in Cutaneous Biology (AcREM Stem), University of Kerala <sup>2</sup>CSIR-National Institute for Interdisciplinary Science and Technology, Thiruvananthapuram parvathyca125@gmail.com

## Abstract

This study investigates benzoquinone derivative NIIST 7 a novel compound with melanin inhibition and antioxidant properties, evaluating its efficacy in both in vitro and in vivo models. Zebrafish were maintained under controlled conditions, and protein content was quantified using BCA assays. The study evaluated the anti-melanogenesis and antioxidant effects of the benzoquinone derivative NIIST7 in vitro using A375 melanoma cells and in vivo with zebrafish embryos. The MTT assay showed that NIIST7 was non-toxic up to 250  $\mu$ M, with a 60% reduction in tyrosinase activity at 200  $\mu$ M. NIIST7 effectively decreased melanin content in IBMX-induced cells and exhibited minimal toxicity in zebrafish models, demonstrating significant melanin suppression. Additionally, NIIST7 displayed antioxidant activity, effectively scavenging free radicals and reducing apoptosis in zebrafish larvae. These findings suggest NIIST7 is a promising candidate for anti-melanogenic and antioxidant therapies. The study on the benzoquinone derivative NIIST7 revealed its potential as an effective melanin inhibitor and antioxidant. In vitro, NIIST7 significantly reduced melanin synthesis in A375 melanoma cells, and in vivo, it exhibited dose-dependent melanin inhibition in zebrafish embryos without notable toxicity. The compound demonstrated strong antioxidant activity, comparable to ascorbic acid, with an IC50 of 15.89  $\mu$ M. DCFDA and AO staining confirmed that NIIST7 effectively reduced oxidative stress and apoptosis in larvae. These findings suggest NIIST7 is a promising candidate for treating

hyperpigmentation and related skin disorders, warranting further clinical investigation and mechanistic studies.

# Environmental pollutant octyl phenol-induced neurotoxicity in the adult zebrafish model

## Kaunava Roy Chowdhury, Vir Vikram

School of Pharmaceutical Sciences, CT University, Ludhiana, 142024

## Abstract

Octylphenol (OP), a toxic xenobiotic classified as an endocrine disruptor, poses significant risks by interfering with the endocrine systems of diverse organisms. Derived primarily from the degradation of OP ethoxylates, widely used as industrial surfactants, OP contamination threatens aquatic ecosystems and human health. Its detrimental effects include reproductive and developmental toxicity, neurotoxicity, and carcinogenic potential. At the cellular level, OP induces the activation of astrocytes and microglia, heightens inflammatory cytokine production, and exacerbates oxidative stress by increasing reactive oxygen species (ROS) generation, leading to pronounced neurotoxic effects and cellular damage. Given the escalating environmental and health concerns, effective management of OP exposure remains a critical challenge. This study aims to explore OP-induced neurotoxicity in an adult zebrafish model by employing an integrative approach. Through behavioral analysis, biochemical assays, and molecular estimations, we seek to unravel the mechanistic underpinnings of OP toxicity and its impact on neurological health. This investigation not only enhances our understanding of OP's neurotoxic pathways but also contributes to the broader effort of mitigating its long-term environmental and public health risks.

## **OP-7**

**OP-6** 

# Tailoring gelma-guar gum bioink for skin tissue engineering via extrusion-based 3D bioprinting

### Lakshmi Rajan Menon, Ratnesh Jain, Prajakta Dandekar

Institute of Chemical Technology, Nathalal Parekh Marg, near Khalsa College, Matunga East, Mumbai-400019, India

lakshmi.menon@nano-medicine.co.in

#### Abstract

This study was focused on refining GelMA synthesis, optimizing GelMA-based bioink for extrusion-based 3D bioprinting, and evaluating its efficacy for skin tissue applications. GelMA-based bio inks offer significant potential for wound healing and regeneration of skin tissue. Incorporation of guar gum into the bioink enhanced its viscosity and controlled the gelation kinetics, while borax effectively cross-linked both GelMA and guar gum, creating stable constructs with improved structural integrity. GelMA with controlled functionalization was synthesized using syringe pumps for addition of methacrylic anhydride and NaOH, followed by sterile filtration and lyophilization. Quality checks included DOF estimation via 1H NMR or TNBS Assay, determination of cross-linking efficiency through FTIR, and determination of rheological properties using a rotational rheometer. The bio ink consisted of 10% (w/v) GelMA and 1.5% (w/v) guar Gum, with lithium phenyl-2,4,6-trimethylbenzoylphosphinate (LAP) for photo cross-linking and 0.5% (w/v) borax as a chemical crosslinker. Pre-printing evaluations included rheometric analysis, cross-linker optimization, and syringeability tests. Printing parameters such as pressure, speed, and nozzle size were optimized based on shape fidelity and infill density. Post-printing assessments covered degradation, swelling, SEM, mechanical strength, and cytotoxicity of the crosslinker using MTT assays. Biocompatibility was confirmed through live/dead assays on Human Adult Dermal Fibroblasts (HADF). Quality control ensured consistent GelMA synthesis with a DOF of 50±2% and reliable rheological behaviour. Filtration and lyophilization preserved GelMA's properties. Optimized crosslinking improved stability and cell viability. Printing showed good shape fidelity and stability. Live/dead assays indicated cell proliferation from day 7. The GelMA-Guar gum bioink demonstrated excellent printability, mechanical strength, degradation kinetics, and biocompatibility, making it a promising candidate for skin tissue engineering. These findings highlighted the potential of GelMA-Guar gum bio-ink for advancing skin tissue engineering through improved printability, mechanical strength, and biocompatibility.

# **3D** tumor spheroid-nk co-culture model: A novel alternative approach to animal studies in cancer immunotherapy

Ramya VVS, Suresh Poosala

Oncoseek Bio Pvt Ltd., VCR Park, Visakhapatnam – 530002 ramya@oncoseekbio.com

#### Abstract

The use of natural killer (NK) cells in cancer therapy presents a promising alternative to traditional animal models for studying immune responses against tumor. This research investigates the application of a 3D tumor spheroid co-culture model utilizing two cancer cell lines, ES-2 (ovarian cancer) and NCI H441 (lung cancer), to assess NK cell cytotoxicity in a more physiologically relevant environment. The study aims to evaluate the expression of programmed cell death ligand 1 (PD-L1) on these tumor cell lines and examine the effectiveness of NK cells in targeting PD-L1-expressing tumors. Natural killer (NK) cell-mediated cytotoxicity can be precisely evaluated by co-culturing NK cells with cell tracker-labelled tumor spheroids, followed by live confocal microscopy imaging to assess cancer cell death, where fluorescence intensity from spheroid staining serves as a quantifiable indicator of NK cell efficacy within a 3D environment. By demonstrating the effectiveness of NK cells in eradicating cancer cells within a 3D model, this research contributes to the development of alternative methods that prioritize ethical considerations while still yielding relevant biological insights. The findings underscore the potential of NK cell co-culture models as robust alternatives for studying cancer immunotherapy and advancing our understanding of immune interactions in a human-like context.

## **OP-9**

# Employing the silkworm (*Bombyx mori*) as an alternative model organism for drug development and safety evaluation

## Atsushi Miyashita, Masaki Ishii

Laboratory of Antifungal Immunobiology, Teikyo University Institute of Medical Mycology, Tokyo, Japan <u>atmivashita@main.teikyo-u.ac.jp</u>

### Abstract

Replacing traditional rodent-based experimental systems with models using lower-order organisms not only helps reduce research and development costs but also addresses societal demands regarding animal ethics. We have employed the silkworm (Bombyx mori) to develop various model systems and have worked on research and development for pharmaceuticals and food products. For instance, by infecting silkworms with pathogens that infect humans to create infection models, we have screened antibacterial drugs by administering unknown samples and determining their therapeutic effects. This method has led to the discovery of promising compounds such as lysocin E, nosokomycin, and ASP2397. Currently, we are developing infection models using Mucor, which causes severe fungal infections, to explore antifungal agents. Silkworms inoculated with Mucor spores die from the infection over time, and antifungal drugs used in human clinical settings have demonstrated life-extending effects. Pathological tissue sections revealed hyphal invasion in tissues, which resembled the pathological findings seen in human clinical cases. Quantitative evaluation of the pathogenicity of four Mucor strains based on LD50 values showed variability among the strains, with pathogenicity strength corresponding to genomic structures. These results suggest that using the silkworm model, we can discover compounds that exhibit therapeutic effects against Mucor and elucidate the mechanisms of Mucor pathogenicity. Moreover, we are expanding our research to use silkworms as a testing system for the functionality and safety evaluation of foods in addition to pharmaceuticals. In this presentation, I will report on the latest outcomes of these projects.

# **OP-10**

# CiPA validated iPSC-derived cardiomyocytes: A powerful tool in Drug Safety and Discovery

**Amit Khanna, Vijay Konala More S., Dhepe S., Reddy V, Bhanushali P, Khanna A** *Yashraj Biotechnology, Plot No C113, MIDC, Navi Mumbai, Maharashtra 400705* 

## amit.khanna@yashraj.com

## Abstract

Drug-induced cardiotoxicity remains a critical concern for the pharmaceutical industry, regulatory bodies, and healthcare providers. Cardiotoxicity manifests in various forms, including electrophysiological, contractile, and structural disturbances, leading to severe clinical outcomes such as proarrhythmic, heart failure, and cardiomyocyte damage and loss. These toxic effects may occur acutely, or develop chronically. Humaninduced pluripotent stem cell-derived cardiomyocytes (hiPSC-CMs) present a promising in vitro model for predicting cardiotoxicity across different toxicity profiles. We have developed YBLi Cardio, an hiPSC-CM model derived from the YBLi001 line, generated from a healthy donor using Sendai virus technology. Differentiated cardiomyocytes showed a seeding efficiency of over 80% with a unique cocktail mixture optimized for cardiomyocyte culture. YBLiCardio was validated for both acute and chronic toxicity assessments, including the Comprehensive in vitro Proarrhythmia Assay (CiPA), which provides an alternate proarrhythmic risk assessment rather than relying solely on clinical QT interval measurements. To assess the predictive power of YBLiCardio, we conducted CiPA testing at Innovitro GmbH with all 28 compounds from the CiPA panel across four concentrations. The effects were measured as dynamic beat pattern changes using Electric Field Potential (EFP) on the CardioExcyte platform. YBLiCardio cells responded to all compounds as expected based on HESI CiPA reference data, confirming their suitability for in vitro cardiac safety testing and in vitro proarrhythmic risk assessment for new drug candidates.

# **ORAL MatTeK AWARD PRESENTATION**

# Design, fabrication, and computational analysis of a multi-organ-on-chip device for the lung-liver-kidney axis model

### Joseph Xavier, Anil Kumar PR

Sree Chitra Tirunal Institute for Medical Sciences and Technology, Trivandrum, Kerala <u>josepxvr@gmail.com</u>



Joseph Xavier is a dedicated bioengineer specializing in toxicology, microfluidics, and organ-on-a-chip technologies. Currently pursuing a PhD with a CSIR-Direct SRF fellowship at Sree Chitra Tirunal Institute for Medical Sciences and Technology, he focuses on developing advanced organ-on-a-chip platforms for disease modeling and evaluating nano-based therapeutics. With a Bachelor's degree in Biotechnology and Biochemical Engineering from the University of Kerala and a Master's in Technology in Biotechnology from Maulana Azad National Institute of Technology Bhopal, Joseph brings a strong foundation to his research. His academic journey has been marked by excellence, including a Commonwealth Split-Site Fellowship from the UK Foreign, Commonwealth, and Development Office (FCDO) at Imperial College London, where he contributed to the development of airway disease models. Proficient in cutting-edge techniques like microfluidic fabrication, toxicology, molecular biology assays, and nanoparticle synthesis and characterization, Joseph excels in both innovation and interdisciplinary collaboration. His work has been recognized through publications in high-impact journals and prestigious awards. Beyond his research, Joseph actively mentors students manages laboratory operations, and collaborates on projects that bridge the gap between science and technology. Passionate about driving progress in biomedical research, he remains committed to improving healthcare solutions by integrating engineering and biology.

#### Abstract

In the scenario of the increasing trend of respiratory diseases globally, it is essential to have a reliable and rapid therapeutic measure to combat these diseases. Conventional static cell culture systems of drug screening lack the dynamic in vivo conditions and complexity of multi-organ interactions. Animal models are the next reliable test system, but animal-based platforms don't represent human physiology and are not completely reliable. Hence, this study aims to develop a multi-organ-on-chip (MOoC) platform representing lung, liver, and kidney tissue compartments as an alternative in vitro dynamic test platform for drug screening. The MOoC device comprises cell-specific culture chambers representing a different organ, which are connected sequentially by microchannels. Each chamber is separated from the microfluidic channel by a membrane to create a vascular-tissue interface. The dynamic flow conditions were simulated using COMSOL Multiphysics software. The MOoC chips were fabricated by micro-milling of clear acrylic sheets. The tissue compartments accommodate a membrane-bound tissue chamber. This assembly allows controlled cell seeding and connected cell interactions. The dynamic flow conditions were maintained using a peristaltic pump through microchannels. The flow dynamics were validated by flow visualization experiments and molecular diffusion across the membrane by fluorescent tracers. The MOoC was seeded with representative cell type and maintained under perfusion. The cell viability and morphology were analyzed by live/dead and actin filament staining, confirming its potential for in vitro toxicity and pharmacology studies. Further validation of this system with a disease model will be a promising alternative to animal experiments, advancing the development of human-relevant, reliable testing systems.

# OMA-2

OMA-1

# Impact of fluoxetine an antidepressant on female fertility through zebrafish (*Danio rerio*) as an experimental model

Teja Mothe\*, Mehdi Ali Mirza, S Seethalakshimi

Department of Pharmacology, ESIC Medical College and Hospital, Hyderabad, India



Teja Mothe is a Medical Graduate from 2010 batch of Gandhi Medical College, Secunderabad, Hyderabad, Telangana. After completing his Graduation, he worked as Medical Officer in various departments including emergency and causality, Medical and Surgical ICUs, Pediatric wards. Apart from this, he has worked in Bioavailability and Bioequivalence (BA/BE) centers which carries clinical trials for generic drugs. This has built his passion and interest in making Pharmacology his career. In the year 2023, he joined as Postgraduate in Department of Pharmacology at the prestigious Employees State Insurance Corporation (ESIC) Medical College and Hospital, Sanathnagar, Hyderabad under the Ministry of Labor and Employment that serves insured patients under the ESIC Act. His thesis deals with a drug trial on patients suffering from major Depression, in collaboration with Psychiatric Department. He is also interested in non-clinical research, specially dealing with animals of lower phyla such as zebrafish.

# Abstract

**Background:** Fish and other aquatic life are exposed to industrial waste, including drugs and their byproducts, which harm the environment, wildlife, and public health. Fluoxetine a Selective-Serotonin-Reuptake-Inhibitor is reported in water bodies, wastewater treatment plants, and fish. This study investigates fluoxetine's impact on zebrafish fertility, given that antidepressants are known to affect fertility in humans.

**Methodology:** Five adult male and female zebrafish pairs were segregated in 10-litre tanks at 28°C, with a 14-hour light and 10-hour dark cycle. They were fed high-protein diet thrice daily. To confirm the fertility each-pair was first kept in separate chambers and initial set of eggs were discarded. In Cycle-1, pairs were separated for five-days with daily water change, and then spawned on Day-6. The total number of eggs were counted immediately and the number of live eggs (clear and transparent considered live and the cloudy, opaque-eggs were considered dead) was recorded 12-hours post-fertilization under 20x entomology-microscope. In Cycle-2, males and females were exposed to fluoxetine for five-days. The 3.2µg/L drug was added daily after water change. Spawning was repeated on Day-6. The eggs-count and live-eggs were recorded. In Cycle-3, fish were tested without fluoxetine similar to Cycle-1. A total-number of eggs and live-eggs were counted to observe any delayed effects. The mean-number of eggs-generated and the cumulative live-eggs were compared among the cycles.

**Results:** All five-pairs were fertile. Mean eggs-count for Cycles 1, 2 and 3 were  $231(\pm 14)$ ,  $427(\pm 57)$ , and  $442(\pm 31)$ , showing a significant(p<0.01) increase from Cycle-1 for both Cycles-2 and 3. The cumulative live eggs significantly (<0.01) decreased across cycles, at 47% (543/1156), 33% (704/2134), and 29% (641/2212) respectively.

**Conclusion:** Fluoxetine affects serotonin levels, which could explain the increase in egg-production and decrease in egg-viability. These findings also highlight the One-Health approach, emphasizing the shared environmental impacts of pharmaceuticals on human, animal and ecological health.

OMA-3

# Mechanistic insights into PFHxS-induced structural modulation and conformational alterations in serum albumin

# Suramya<sup>1</sup>, Shivani A. Muthu<sup>2</sup>, Chanchal Chauhan<sup>2</sup>, Afnaan Qureshi<sup>2</sup>, Basir Ahmad<sup>2</sup>, Sheikh Raisuddin\*

<sup>1\*</sup>Molecular Toxicology Laboratory, Department of Medical Elementology and Toxicology, School of Chemical and Life Sciences, Jamia Hamdard, New Delhi-110062, India

<sup>2</sup>Protein Assembly lab, Department of Medical Elementology and Toxicology, School of Chemical and Life Sciences, Jamia Hamdard, New Delhi-110062, India suramya sch@jamiahamdard.ac.in



She is a dedicated Ph.D. scholar in toxicology with a background in Forensic Science, having graduated with top honors and received the Gold Medal from SHUATS, Prayagraj, Uttar Pradesh, where she also earned the prestigious Laxmi Rao Memorial Award for the best girl student in the field of Forensic Science. With a strong academic foundation, including qualifying for UGC NET and holding a Senior Research Fellowship, her work centers on environmental health, toxicology, and reproductive health. Her research focuses on the effects of environmental contaminants like PFAS on reproductive systems and metabolic disorders such as PCOS. She is also actively involved in teaching forensic science to master students. Her approach to research work is multidisciplinary, combining toxicology, molecular biology, protein biochemistry, nutrition, and computational tools.

#### Abstract

Perfluorohexanesulfonate (PFHxS), a man-made industrial chemical, is classified under the perfluoroalkyl substances (PFAS). PFAS group of compounds are also characterized as endocrine-disrupting chemicals (EDCs) in view of their disruptive effects on the endocrine system or they mimic hormones. EDCs interact with proteins and nucleic acids thereby affecting cellular processes and modulating a wide range of biological functions. Human serum albumin (HSA) is a major multidomain alpha-helical plasma protein that transports both exogenous and endogenous ligands. HSA is known to exist in N, B, F, and E isomeric forms depending on pH. In this study, we analyzed the binding of PFHxS on different isomers of HSA using fluorescence quenching and molecular docking. We also demonstrated the effect of binding on the conformation, stability, and ligand-binding activity of HSA isomers. The effect of PFHxS binding on the secondary, tertiary conformations, and exposed hydrophobic surfaces of isomers was examined by far-UV circular dichroism and fluorescence spectroscopy. We observed that both the secondary structure and tertiary structure of the F isomer were markedly altered whereas other isomers showed comparatively lesser effect. However, hydrophobic surfaces of isomers were found to be unaffected by PFHxS binding. PFHxS was found to modulate the unfolding pathway and stability of the protein. We also studied the co-binding of therapeutic agents such as Diazepam and Ofloxacin and found that PFHxS altered the drug-binding ability of HSA. These results demonstrate the interactive potential of PFHxS with HSA and its likely effect on critical biological pathways. Moreover, the significance of this study lies in the potential insights into the toxicological mechanisms of PFHxS and its impact on HSA function, which can be helpful in proposing strategies to mitigate the adverse effects of PFHxS on human health.

OMA-4

# Development of a synergy-based combination from traditional Unani medicine against drug-induced nephrotoxicity *in vitro* and *in vivo* zebrafish model

Naveen Reddy Penumallu<sup>1,2</sup> Sageer Abas<sup>1</sup>, Md Reyaz Alam<sup>1,2</sup>, Mohammad Ahmed Khan<sup>1,2</sup>, Anuja Krishnan<sup>3</sup> Sayeed Ahmad<sup>\*1,4,5</sup>

<sup>1</sup>Centre of Excellence in Unani Medicine (Pharmacognosy and Pharmacology), School of Pharmaceutical Education and Research, Jamia Hamdard, New Delhi, India-110062. <sup>2</sup>Department of Pharmacology, School of Pharmaceutical Education and Research (SPER), Jamia Hamdard,

New Delhi, India-110062. <sup>3</sup>Virology Laboratory, Department of Molecular Medicine, SIST, Jamia Hamdard, New Delhi 110062, India

<sup>4</sup>Department of Food Technology, SIST, Jamia Hamdard, New Delhi 110062, India

<sup>5</sup>Department of Pharmacognosy, SPER, Jamia Hamdard, New Delhi 110062, India

# naveenrpenumallu\_sch@jamiahamdard.ac.in



Naveen Reddy Penumallu is a Senior Research Fellow at the Centre of Excellence in Unani Medicine (Pharmacognosy and Pharmacology), funded by the Ministry of AYUSH, and a Ph.D. scholar in Pharmacology. His research focuses on chronic kidney disease, with expertise in preclinical model development, including zebrafish and advanced cell culture systems, to investigate nephroprotective agents. He has also contributed to development of rare disease models during his research internship at Dr. Reddy's Institute of Life Sciences, Hyderabad. He holds an M. Pharm in Pharmacology from JSS College of Pharmacy, Mysore, and a PG Diploma in Regulatory Toxicology. Naveen has substantial exposure to consultancy projects for industries, systematic literature review reporting, and meta-analysis. His technical expertise encompasses *in vitro* and *in vivo* pharmacology, genotoxicity studies and developmental toxicity.

### Abstract

**Purpose/Aim:** Chronic Kidney Disease (CKD) is a global health challenge that requires innovative therapeutic solutions. This study aims to develop a nephroprotective synergy-based combination (SBC) from the basis of Unani medicine and supported by ethnopharmacological evidence.

**Methods:** We conducted high-throughput screening of botanical extracts, to assess nephroprotective activity. The most effective SBCs were optimized and tested for nephrotoxicity, inflammation, oxidative stress, and apoptotic markers in HEK 293 cell lines. Key biomarkers including KIM-1, NGAL, NF- $\kappa$ B, TNF- $\alpha$ , TGF- $\beta$ , and Cystatin C, were analysed. Metabolite profiling of the SBC was performed using LC-MS, and efficacy was further evaluated against fluoride and drug-induced nephrotoxicity in zebrafish models. Additionally, the nephroprotective and antioxidant effects of SBC were assessed on fluoride and drug-induced nephrotoxicity in Zebrafish. Functional renal markers, antioxidant enzyme activities, and serum levels of blood urea nitrogen, uric acid, creatinine, and electrolytes were measured.

**Results:** A 1:1 SBC demonstrated significant synergistic nephroprotective effects, reducing inflammatory and oxidative stress markers while enhancing cellular protection. SBC co-administration significantly lowered serum BUN and creatinine levels, reduced excretion of proteinuria, and increased body weight, pH, serum total protein, calcium, sodium, and antioxidant enzyme activities compared to nephrotoxic controls.

**Conclusions:** The SBC developed in this study, shows promise as a nephroprotective agent for CKD management. Preliminary findings support its ethno-medicinal use and suggest further evaluation in rodent models to determine the best SBC formulation. This research highlights potential CKD treatments and the value of high-throughput screening in cell culture and zebrafish models, potentially advancing clinical applications.

# MatTek AWARD POSTER PRESENTATIONS

PMA-1

# Macromolecular Crowding Mediated Development of Cell-derived Matrix Scaffold: Drug Resistance to 3D in-vitro Disease Modelling

# Bitan Guchhait, Subhadeep Roy

National Institute of Pharmaceutical Education and Research, Kolkata, West Bengal 700054, India <u>g.bitan1998@gmail.com</u>

# Abstract

Cancer-associated fibroblasts (CAFs) are critical to tumour growth and treatment resistance in the tumour microenvironment (TME). Activated fibroblasts, CAFs release cytokines and growth factors that boost tumour cell growth, angiogenesis, and immunity. TGF- $\beta$  signalling increases Extracellular matrix (ECM) synthesis, including collagen and fibronectin, creating a supportive environment for tumour cells. CAF-produced ECM is stiffer and denser, allowing tumour invasion and limiting medication penetration, causing therapy resistance. McCoy, adult dermal, and mouse embryonic fibroblast cells need long incubation periods to form a strong ECM layer. We investigated macromolecular crowding phenomena (volume exclusion and dense molecular environment) to tune ECM secretion and target specific ECM types. The introduction of

macromolecular crowders can lead to rapid ECM deposition. Herein, we assessed the potential of macromolecular crowders, such as Ficoll 400, PVP 40, PVP 360, and carrageenan, to produce cell-derived matrices and identify their ECM secretion pattern. We have identified fibronectin, collagen and laminin content in the presence of variable macromolecular crowder to identify its secretion pattern. Further, we extended our study to develop decellularised cell-derived matrix development and culturing MDA-MB-231 cells to understand the role of differential cell-derived matrix in drug resistance (Doxorubicin). How differentially produced ECM and variable matrix stiffness can contribute to cellular migration, invasion, and epithelial to mesenchymal transition and drug resistance is the primary area of our present research. It will help to develop an ECM-derived 3D in-vitro drug screening.

PMA-2

# BMSC and triple-negative breast cancer cell homing: Development of stable 3D invitro EMT model

## Vaishnavi Madasu, Subhadeep Roy\*

National Institute of Pharmaceutical Education and Research, NIPER-Kolkata Chunilal Bhawan, 168, Maniktala Main Road Kolkata – 700054 [West Bengal] vaishnavi.madhasu@gmail.com, \*subhadeeproy.good@gmail.com

### Abstract

Breast cancer is the second most prevalent metastatic cancer among women. A pivotal biological mechanism in breast cancer metastasis is the epithelial-mesenchymal transition (EMT), particularly displayed in triplenegative breast cancer models such as MDA-MB-231 cells. During this process, epithelial cells lose their polarity, break down cell-cell connections, and transform into mesenchymal cells. This transformation represents a potential pathway through which tumor cells acquire characteristics that facilitate tumor metastasis and progression. In our study, we aimed to investigate the induction of EMT in MDA-MB-231 cells through co-conditioning and transwell multicompartmental co-culturing with Rat BMSC. TGF-B1 and conditioned media derived from rat bone marrow stem cells (BMSCs). Our findings demonstrated significant phenotypic alterations in the MDA-MB-231 cells, characterized by an upregulation of mesenchymal markers such as vimentin, TWIST and SNAIL alongside a marked downregulation of the epithelial marker E-cadherin. The morphological analyses indicated notable changes in cellular architecture that are known to enhance tumor cell motility and invasive potential. The TGF-B1 treatment resulted in a fibroblast-like morphology and increased migratory capabilities, altered oxidative defence mechanism (SOD, GSH, Catalase, Protein Carbonyl), confirming the role of EMT in promoting aggressive tumor characteristics. The findings underscore the crucial role of co-conditioning and co-culture in developing stable EMT induction in in-vitro disease model. It offers valuable insights into the mechanisms driving breast cancer aggressiveness and potential therapeutic targets.

### PMA-3

# Making pregnancies safe: A high throughput device to generate placental spheroids for assessing drug toxicity

### Pratik Rasal, Anshul Bhide, Deepak Modi

ICMR- National Institute for Research in Reproductive and Child Health, Molecular and Cellular Biology laboratory, Jehangir Merwanji Street, Parel, Mumbai- 400012 <u>Prasal308@gmail.com</u>

### Abstract

Placenta is a critical organ for pregnancy that helps in exchanges of gases, nutrients and also functions as an endocrine organ for maintaining pregnancy. Therefore, the proper functioning of the placenta during pregnancy is crucial and any disruptions to the placenta function can cause pregnancy related disorders including teratogenicity. During pregnancy, the use of most medications is restricted for their unknown effects on placenta and causing teratogenicity. However, many women need to take drugs due to pre-existing conditions like tuberculosis epilepsy, diabetes and sickle cell Anaemia. However, the restriction on the use of these drugs posies a challenge as the pregnant women cannot take the drug and suffer from their disease leading to significant morbidity and mortality. Animal models are used to predict safety of drugs in humans. But historical experiences have shown that in pregnancy the animal data often do not accurately predict the drug afety/toxicity in humans. This is largely because of extreme structural and functional difference between

human and animal placenta. Thus, Alternate model to assessing effects of drugs on placenta can be a surrogate measure of drug toxicity/safety in pregnancy, Herein we have developed a device to develop cell spheroids in a high throughput manner to permit drug testing in a rapid manner. Using this device we have generated spheroids of placental cells and tested cytotoxicity of selected anti-tuberculosis drugs. In addition, we have also used these spheroids to test the invasive & amp; migratory functions of the trophoblast cells in vitro. The data will be presented.

# PMA-4 In-silico evaluation of gastrodin neuroprotection against bisphenol A-induced ADHDlike symptoms

## Mohd. Anas Saifi, Huma Khan, Md. Jawed Akhtar, Sheikh Raisuddin

Department of Medical Elementology & Toxicology, School of Chemical and Life Sciences, Jamia Hamdard, New Delhi-110062

anasm9810@gmaill.com

## Abstract

**Background:** Bisphenol A (BPA) is a widely used endocrine-disrupting chemical known to interfere with neurological development and function, contributing to the onset of attention deficit hyperactivity disorder (ADHD)-like symptoms. Gastrodin, a bioactive compound derived from Gastrodia elata, has demonstrated neuroprotective, anti-inflammatory, and antioxidant properties in various neurodegenerative conditions. This study investigates the potential of Gastrodin to mitigate BPA-induced neurotoxicity and ADHD-like behaviors through in silico methods.

**Methods:** Using computational approaches, molecular docking simulations were conducted to identify possible interactions between Gastrodin and key molecular targets involved in ADHD pathophysiology, including dopamine receptors, brain-derived neurotrophic factor (BDNF), and oxidative stress-related enzymes. Additionally, we employed network pharmacology and pathway analysis to explore the mechanistic pathways through which Gastrodin may exert its neuroprotective effects against BPA-induced ADHD-like symptoms.

**Results:** The in-silico findings suggest that gastrodin forms stable binding interactions with critical proteins involved in neurotransmitter regulation, cellular protection, and inflammatory response, such as dopamine receptors (DRD1, DRD4, and DRD3). Gastrodin was predicted to enhance neuroplasticity receptor (BDNF), and reduce oxidative stress and inflammation, thereby counteracting the adverse effects of BPA exposure on brain function. These molecular interactions align with the known behavioral outcomes of ADHD, including hyperactivity, impulsivity, and cognitive impairments.

**Conclusion:** The in-silico analysis supports the hypothesis that gastrodin may offer neuroprotective effects against BPA-induced ADHD-like symptoms by modulating key molecular pathways. This computational study provides a foundation for future in vitro and in vivo investigations to further elucidate the therapeutic potential of gastrodin in the management of ADHD and environmental toxin-related neurodevelopmental disorders

# PMA-5 The effect of diethyl hexyl phthalate on myelin sheath in early developmental stages of zebrafish larvae

Garima Jindal<sup>1</sup>, Anuradha Mangla<sup>1</sup>, Padmshree Mudgal<sup>2</sup>, Sheikh Raisuddin<sup>1</sup> <sup>1</sup>Department of Medical Elementology & Toxicology, Jamia Hamdard, New Delhi – 110062 <sup>2</sup>Department of Biochemistry, Daulat Ram College, University of Delhi, Delhi-110007 garima.jindal\_sch@jamiahamdard.ac.in

## Abstract

Diethyl hexylphthalate (DEHP), an endocrine disruptor, has become a major concern for environment and human health. It is used as plasticizer in many daily used items because it gives the flexibility and softness to the plastic. It does not make any chemical bond with the plastic and easily leaches out from the products into the food and environment. Several studies have documented the toxicity of DEHP. Although studies have been conducted on the neurotoxicity of DEHP, there is a lack of knowledge regarding its toxic effects on myelin sheath. To fulfill this gap, this study assesses how DEHP affects the myelin sheath in zebrafish larvae at early development stage. Zebrafish is a promising alternative model for early developmental studies. We have investigated the neurobehavioral, histopathological and molecular changes in the zebrafish larvae at 5 days

post fertilization after exposure to two different doses of DEHP (50 and 100 ppm). Cuprizone was taken asthe positive control. Behavioral scototaxis assay showed greater number of exposed larvae in the dark area as compared to the control group larvae which were found more in the light area. Histological data revealed an increased cell death in DEHP treated groups. The expression of myelin sheath-related proteins was decreased in DEHP-exposed larvae. In summary, this study shows the toxicological potential of DEHP in affecting the myelin sheath during the developmental stages of zebrafish larvae.

PMA-6 RNA based anti-dengue vaccine to elevate CTL response: Design and Immunological response prediction

## Adib khan<sup>1</sup>, Manu Sharma<sup>1</sup>, Mairaj Ahmed Ansari<sup>1,2\*</sup>

<sup>1</sup>Department of Biotechnology, School of Chemical and Life Sciences, Jamia Hamdard, New Delhi-110062 <sup>2</sup>Centre For Virology, School of Interdisciplinary Sciences and Technology, Jamia Hamdard, New Delhi-110062

drmansari@jamiahamdard.ac.in

### Abstract

Dengue virus comprises of four different serotypes (DENV1-4) responsible for the disease, effecting around five million individuals with more than 5000 deaths globally and alone in India 2,89,235 cases and 485 deaths in year 2023 and 32091 cases with 32 deaths till 30th June 2024. Developing an effective vaccine faces significant challenges due to the virus's complex life cycle, serotype diversity, and antibody-dependent enhancement (ADE). Dengvaxia and TAK-003 are licensed vaccines, but due to their ineffectiveness against all the serotypes, ADE and safety among children remain a significant challenge. Vaccinomics is in-silico study which provides the major driver for the development, administration, and monitorization of vaccines. In-silico methods reduce the dependence on animal study, making them cost effective, enhanced precision, and integration of large data. This study focuses on designing an RNA-based vaccine candidate using reverse vaccinology/ vaccinomics with the help of bioinformatic/ immunoinformatic tools. 168 NS5 sequences from all four serotypes (53 DENV1, 69 DENV2, 38 DENV3 and 14 DENV4) isolated in India and the conserved regions were identified. B and T cell epitopes were predicted and assessed using Clustal omega, IEDB and other bioinformatic tools. Three B cell, forty-five T cell epitopes (38 MHC1, 7 MHC2) was predicted, the selected epitopes were linked, refined and validated, showing a high-quality model. Population coverage of 99.53% (India) and 98.3% (World). Docking predicted good immune response to develop a prophylactic candidate vaccine. The protein-based antigens were codon optimized for predicting RNA sequence to develop RNA based vaccine. Further validation is needed. Simulation base bioinformatics studies will predict the immunocompetent vaccine candidate, hence could reduce the animal use for prophylactic efficacy evaluation.

**PMA-7** 

# An in vitro model mimicking skin rigidity to study diabetic wounds

## Ranjita Rajesh Save<sup>1</sup>, Kanchan Chitnis<sup>1</sup>, Abhijit Majumder<sup>2</sup>

<sup>1</sup>Ramnarain Ruia Autonomous College L. Napoo Road Matunga East Mumbai Maharashtra 400019 <sup>2</sup>Indian Institute of Technology Bombay. <u>ranjitasave@ruiacollege.edu</u>

## Abstract

Diabetes is a silent pandemic, with approximately 537 million adults living with the condition as of 2021; this number is projected to reach 783 million by 2045. The majority of these cases are classified as type 2 diabetes. Diabetes is associated with significant complications such as nephropathy, neuropathy, cardiomyopathy, and retinopathy. One of the severe consequences is the development of chronic wound ulcers, which can lead to trauma, amputations, and even death. Over the past two centuries, various animal models, particularly mouse models, have been used to study wound healing. Larger animal models are often not cost-effective and raise many ethical concerns. Additionally, there are significant limitations in accurately replicating human diseased skin models, especially in wound healing, as mouse skin is hairy and thick, and healing occurs primarily through contraction. Hence, there is a need to develop an in vitro diabetic wound healing model. Towards this goal, in this work, we have developed a diabetic in vitro model incorporating mechanical cues in the form of substrate rigidity. It is known that skin rigidity changes from healthy skin to wound, and this changed rigidity significantly influences cellular behavior. Hence, we have created polyacrylamide (PAA) gels mimicking skin rigidities on which Keratinocytes acclimatized to the physiological glucose concentration were subsequently

cultured. Finally, we used insulin to mimic diabetic cells and studied the effect of substrate rigidity on cellular functions such as proliferation, migration, and ROS generation. We envisage that this model can be used for drug studies in diabetic wound healing to better predict without harming animals.

#### PMA-8 Neuroprotective mechanism of 4-hydroxyisoleucine and alpha-mangostin in multiple sclerosis: Evidence from in-silico analysis

#### Zuber Khan<sup>1</sup>, Sidharth Mehan<sup>1</sup>, Ghanshyam Das Gupta<sup>2</sup>

<sup>1</sup>Division of Neuroscience, Department of Pharmacology, ISF College of Pharmacy, Moga, Punjab, India <sup>2</sup>Department of Pharmaceutics, ISF College of Pharmacy, Moga, Punjab, India t <u>zuber3491@gmail.com</u>

#### Abstract

Background: Multiple sclerosis (MS) is an autoimmune inflammatory disease affecting the central nervous system, causing demyelination, oligodendrocyte loss, glial scar formation, and neuronal injury. We explore the role of EB-mediated neuronal degeneration in MS development and progression. We use 4hydroxyisoleucine (4-HI) amino acid from fenugreek seeds and Alpha-Mangostin (AMG), a xanthone derivative from Garcinia mangostana, to investigate the potential of these compounds to mitigate ethidium bromide-induced MS using in-silico approaches. Methods: Molecular docking simulations were conducted using computational techniques to investigate the possible interactions of 4-HI with important molecular targets involved in the pathogenesis of MS, such as IGF-1 and GLP-1. Additionally, the study analyzed the interactions of AMG with key signaling molecules like c-JNK, P38MAPK, Erk-1/2, and STAT-3 to understand their mechanistic roles in MS. Results: The in-silico findings suggest 4-HI and AMG form stable binding interactions with critical proteins involved in neurogenesis, synaptogenesis, glial cell repair, myelination, and neuroinflammatory response, such as IGF-1, GLP-1, c-JNK, P38MAPK, Erk-1/2, STAT-3. 4-HI and AMG were predicted to promote neuroplasticity, mitigate oxidative stress, and suppress neuroinflammation, counteracting the detrimental effects of ethidium bromide exposure on brain functions. Conclusion: The insilico analysis supports the hypothesis that 4-HI and AMG may offer neuroprotective effects against mutagen ethidium bromide-induced MS-like symptoms by modulating key molecular pathways. Computational study provides a foundation for future in vivo investigations to elucidate further the therapeutic potential of 4-HI and AMG in managing MS.

**PMA-9** 

#### Implementation of reduction and replacement under 3Rs in antivenom quality testing

#### Alka Sahu, Avni Blotra, Karthikeyan Vasudevan

Centre for Cellular and Molecular Biology, Hyderabad <u>alkasahuak@gmail.com</u>

#### Abstract

Polyvalent antivenom raised against the venom of the big four snakes stands as the only available treatment for snakebites in India. The process of antivenom purification post hyperimmunization of the horses, traditionally involves four phases namely plasma stage, bulk stage, formulated bulk stage, and formulated product. Preclinical efficacy of polyvalent antivenom is tested by estimating the antivenom's ability to rescue lethality in mice. Although animal experimentation has been used as the gold standard for testing efficacy, it is associated with pain, distress, and lethality. During the manufacturing process, antivenom quality testing is a critical component and is performed at each stage of product development. Each phase requires a significant number of animals, with usage totalling up to 8,000 animals annually per manufacturing unit. These evaluations, as outlined by WHO standards, involve both in vivo and in vitro tests to determine the neutralization efficacy of antivenoms against venom toxicity. However, growing ethical concerns and WHO's endorsement of the 3R principle (Replacement, Reduction, and Refinement) have highlighted the need for alternatives to animal testing. In this study, we have designed a cassette of in vitro assays to assess diverse aspects of venom activities and its neutralisation by antivenom. These assays evaluate both binding and functional neutralization of venom toxins, providing a comprehensive assessment of antivenom efficacy. Our aim is to derive an average efficacy score from the in vitro assays and demonstrate correlation between in vitro and in vivo scores. This approach not only aligns with WHO's push for animal replacement but also has the potential to substantially reduce animal usage in the early phases of testing. By integrating in vitro methodologies with existing protocols, we aim to establish a pathway for minimizing animal dependence

while maintaining rigorous quality standards for antivenom production. This shift represents a critical step toward ethical and sustainable practices in antivenom testing.

#### PMA-10 Ecotoxicological profiling of atrazine: A novel approach using *Hydra magnipapillata* as an alternative animal model

### Angel Mary Arulanandu, Ankit Dilaware, Pradheena Vairaganthan, Kadalmani Balamuthu\*, Akbarsha Mohammad Abdulkader\*\*

National Centre for Alternatives to Animal Experiments (NCAAE), Bharathidasan University, Tiruchirappalli - 620024. India.

\*Professor, Department of Animal Science, Bharathidasan University,

\*\*Founder and Chair, Mahatma Gandhi – Doerenkamp Centre (MGDC), Bharathidasan University. angelmary.bio@gmail.com

#### Abstract

Atrazine (ATZ), a commonly used herbicide in aquatic ecosystems, is recognized as an endocrine disruptor with potential ecological consequences. In this study, Hydra magnipapillata was employed as an alternative model organism to assess the toxicity of ATZ, addressing the ethical concerns surrounding traditional animal testing. The LC50 of ATZ was initially determined through morphological assays and based on these results, sub-lethal concentrations (6.25 and 3.12 µg/mL) were selected for 24 and 48-hour exposures. The feeding assay indicated that ATZ treated hydra captured fewer Artemia compared to controls, suggesting compromised feeding efficiency. Since feeding is closely linked to reproduction, a 14-day reproduction assay was conducted which revealed a significant reduction in budding rates in ATZ exposed hydra, indicating of impairment reproductive function due to feeding deficiency. To explore the underlying cause of these impairments, regeneration assays were performed, which revealed a dose dependent delay in tentacle regrowth, suggesting impaired tissue repair. A battery cell complex assay using toluene blue highlighted significant cellular damage in tentacle cells responsible for prey immobilization. Additionally, Acridine Orange staining revealed green fluorescence punctuates in ATZ treated hydra, confirming cellular damage leading to apoptosis. In conclusion, the study underscores the ecological risks of ATZ in aquatic environments and demonstrates the utility of Hydra magnipapillata as an effective alternative model for toxicity assessment, emphasizing the need for careful consideration of herbicide usage in aquatic habitats.

**PMA-11** 

## Understanding dysregulation of human endometrial regeneration by endocrine disruptors: Use of regeneration in Hydra as a model

### Pradheena Vairaganthan., Ankit Dilaware., Angel Mary Arulanandu., Kadalmani Balamuthu\*., Akbarsha Mohammad Abdulkader\*\*

National Centre for Alternatives to Animal Experiments (NCAAE), Bharathidasan University, Tiruchirappalli- 620024, India.

\*Professor, Department of Animal Science, Bharathidasan University,

\*\*Founder and Chair, Mahatma Gandhi – Doerenkamp Centre (MGDC), Bharathidasan University <u>dheenagandh@gmail.com</u>

#### Abstract

Researcher utilizes many models for regeneration studies. This project aims to explore a novel approach to study aspects of human endometrial regeneration using Hydra as an alternative animal model to study the effect of endocrine disrupting chemicals. While regeneration varies across species, some conserved pathways exist across phyla. Invertebrates like Hydra are models for whole-body regeneration, whereas vertebrates like zebrafish focus on specific organs. Both Hydra and the human endometrium exhibit strong regenerative capacities and share similar signaling pathways, such as Wnt, JNK, and Notch. Wnt signaling is key for head regeneration in Hydra so also for homeostasis in the endometrium. The PI3K/AKT/mTOR pathway remodels the endometrium during menstruation, which also ensures regeneration of the endometrial lining following menstruation, interacting with apoptosis and Hydra's head-specific genes such as Wnt. JNK is crucial for Hydra's tentacle formation and it is also involved in endometrial regeneration during proliferative and secretory phases. Notch signaling activates Wnt3 and  $\beta$ -catenin in Hydra regeneration which also regulates endometrial stem cell proliferation and angiogenesis. Additionally, VEGF and FGF signaling are vital for both

endometrial and Hydra tissue regeneration. These highlight key similarities between these two systems despite differences in regeneration types: morphallactic in Hydra and physiological in the human endometrium and, still, many pathways related to regeneration are not yet explored. As they share key similarities in quick regeneration, injury signaling, early wound response, wound healing properties without scar formation, directing cells to the wound healing site, and apoptosis, thereby create appropriate cellular and tissue patterns. Therefore, regenerative property of hydra and human endometrium can be related. By analysing the molec ular changes occurring in Hydra's signaling pathways during regeneration, we can correlate as to how similar processes occur in human endometrial regeneration. This project aims to correlate regeneration processes in both contexts and motivates to explore the basic mechanisms underlying regeneration and the dysregulation caused by interruption of signaling pathways by endocrine disruptors in the human endometrium, which affect the normal regeneration process. Use of Hydra as an alternative model offers a valuable approach, reducing reliance on traditional in vivo animal models for regeneration studies.

#### **PMA -12**

## Developmental and vision toxicity of pyriproxyfen in zebrafish larvae, a competent alternative model for vision toxicity evaluation

#### Sumit Choudhary<sup>1</sup>, Nida Parveen<sup>2</sup>, Padmshree Mudgal<sup>3</sup>, Sheikh Raisuddin<sup>1</sup>

<sup>1</sup>Department of Medical Elementology & Toxicology, School of Chemical & Life Sciences Jamia Hamdard, New Delhi, India <sup>2</sup>Department of Paramedical Sciences (Optometry), School of Nursing Sciences & Allied Health, Jamia Hamdard, New Delhi, India <sup>3</sup>Daulat Ram College, Delhi University, Delhi, India <u>sumitchoudhary0120(@gmail.com</u>

#### Abstract

Pyriproxyfen (PPF) is a juvenile hormone analog that is widely used in agriculture to manage a variety of insect populations. It is acknowledged as an effective hygienic insecticide. Officially known as 2-[1-methyl-2-(4phenoxyphenoxy) ethoxy] pyridine, this substance is a member of a class of pyridine-based pesticides that are widely used to control mosquitoes and agricultural insect pests, especially in areas where drinking water supplies could be contaminated. Despite its widespread use, little toxicity data on ecologically relevant species are available on how it affects them. Investigation on its toxic effects, particularly in fish is desirable. In order to close this knowledge gap, the current study was undertaken to investigate any potential harm that PPF may cause to the development of zebrafish (Danio rerio) embryo opthalmoception. Zebrafish is a renowned alternative model for toxicity and safety evaluation. Embryonic growth was studied at different concentrations of PPF such as 2.0, 2.5, 3.0, 5.0, and 10 parts per million (ppm) over a span of 10 days post-fertilization (dpf). The survivability rate was evaluated at the above doses, and the results showed that 50% of larvae were not able to survive at concentrations above 3 ppm. We also evaluated developmental toxicity by observing multiple indicators such as the presence of edema, heart size, and instances of scoliosis, which are key signs of developmental well-being. In addition to morphological evaluations, we examined the effects of PPF (2 ppm and 2.5 ppm) on visual capabilities through eye examinations. This process involved measuring oxidative stress levels in the larval eyes, using markers such as reactive oxygen species (ROS) and lipid peroxidation (LPO). We also assessed the functionality of key enzymes, including catalase (CAT) and glutathione S-transferase (GST), which are vital for detoxification and cellular defense. Oxidative stress was particularly evident at the highest test concentration (2.5 ppm), as indicated by elevated lipid peroxidation (LPO) levels and significant alterations in antioxidant enzyme activities, including CAT and GST (P < 0.05). Furthermore, we performed behavioral tests to determine how PPF exposure affects the color-sensing abilities of the larvae, offering insights into possible functional consequences for vision arising from the treatments. This comprehensive strategy enabled a detailed examination of the influence of PPF on both physiological development and sensory function in the embryos. The findings of the current study revealed that exposure to higher concentrations of PPF can have detrimental effects on the early developmental stages of zebrafish and their vision capacity. This suggests that PPF may disrupt critical processes during the initial growth phases of these aquatic organisms. Furthermore, our investigation proved that zebrafish or its larvae can be used for the assessment of chemically induced vision toxicity

#### **POSTER PRESENTATIONS**

Poster-01

#### Bioengineering an elastic membrane to recapitulate in vivo alveolar basement membrane stretching properties for in vitro lung tissue development

**Pooja Pandurang Sawant, Priyanka Bishnoi, Ratnesh Jain, Prajakta Dandekar** *Institute of Chemical Technology, Nathalal Parekh Marg, Matunga, Mumbai, 400019 pooja.sawant@nano-medicine.co.in* 

#### Abstract

The development of an in vitro alveolar basement membrane (ABM) is essential in lung tissue engineering and regeneration. The ABM is a critical component of the lung architecture, which provides structural support and facilitates gaseous exchange between alveoli and capillaries. Replicating the ABM in vitro is significant for creating realistic lung models that mirror the structural, mechanical, and biochemical properties of the native lung environment. This study presents the development of an elastic, biomimetic membrane that replicates the properties of the ABM. Given the ABM's complex composition and dynamic behavior, a membrane was synthesized using semisynthetic and synthetic biocompatible polymers. The membrane was fabricated using electrospinning, a technique capable of creating nanofibrous structures that closely resemble the fibrous nature of the native ABM. The fabricated membrane exhibited controlled pore size distribution and elasticity like the native ABM. In vitro studies demonstrated the membrane's ability to support the attachment, proliferation, and growth of alveolar epithelial cells, showcasing its potential as a scaffold for lung tissue engineering. The effect of mechanical stretching, a physiological condition of the ABM caused by respiratory movements, were also investigated on cells cultured on the membrane. By applying controlled mechanical stretching, we studied the influence of mechanical forces on alveolar cell behavior. This biomimetic membrane holds promise for more realistic co-culture lung models.

#### Poster-02

## Development and validation of a static, in vitro disease model for Age-related macular degeneration: Making preclinical evaluations more relevant

#### Pradnya Mahendra Salve, Devashree Nagesh Jahagirdar

Institute of Chemical Technology (ICT), Matunga, Nathalal Parekh Marg, Mumbai, Maharashtra-400019 <u>bit24pm.salve@phd.ictmumbai.edu.in</u>

#### Abstract

Age-related macular degeneration (AMD) is a major cause of blindness worldwide. The increasing presence of aging population in many countries has resulted in greater than 20% of the population being afflicted with this condition. The current treatments for AMD include intraocular injections of anti-VEGF (vascular endothelial growth factor) agents, occasionally combined with other treatment options like photodynamic therapy. These therapies result in inflammatory adverse events such as sterile intraocular inflammation and post-injection endophthalmitis. Thus, there is still a pressing need for developing alternative and non-invasive therapeutics that may be better accepted by the patients. However, the complexity of the disease pathophysiology makes it difficult to mimic this disease in animals and traditional in vitro platforms to support preclinical drug development. Thus, our investigation focused on the development of co-culture model of retinal epithelial and endothelial cells to recapitulate AMD, in a static trans-well-based system. A disease induction cocktail was optimized for inducing cytokine storm, reactive oxygen species, and esterified lipid deposition to imitate drusen formation. This disease inducing composition was selected based on its IC50 value in the target cells, along with other pathophysiological parameters like increase in the intracellular ROS generation, lipid deposition etc. Inflammatory cytokines were analysed for confirming disease induction, as compared to the native cells. Further, bevacizumab, a known VEGF inhibitor, was used to arrest disease-like situation in the model, to enable the model validation. Treatment with bevacizumab decreased the lipid deposition and reduced ROS generation in the developed model. Thus, results indicated the ability of our coculture model to mimic AMD-like characteristics and reduction in disease-like parameters upon treatment with bevacizumab, the proven therapeutic against AMD. This confirmed the potential of our co-culture model for assessing newer therapeutic agents against AMD.

Poster-03

## Selected zinc salts as adjuncts to methotrexate exhibit pro-immune features in rheumatoid arthritis treatment: an in-silico approach

#### Mohammad Hasan<sup>1</sup>, Mohammad Ali Khan<sup>2</sup>, Shahzad Ahmad<sup>1</sup>, Haider Ali Khan<sup>1\*</sup>

<sup>1</sup>Department of Medical Elementology & Toxicology, School of Chemical & Life Sciences, Jamia Hamdard, New Delhi-110062

<sup>2</sup>Department of Biochemistry, School of Chemical & Life Sciences, Jamia Hamdard, New Delhi-110062 <u>haktox@jamiahamdard.ac.in</u>

#### Abstract

Methotrexate (MTX) is a first line disease-modifying anti-rheumatic drug (DMARD) used for the treatment of rheumatoid arthritis (RA). Its mechanism involves to inhibit key enzymes of folate metabolism and purine biosynthesis, which reduces inflammation and modulates autoimmune responses. Recent studies suggest that zinc compounds, such as zinc aspartate and zinc citrate, may serve as adjunctive therapies to counter MTX's immunosuppressive side effects by enhancing immune flexibility. Zinc is known to influence a range of proteins involved in immune function and inflammation. Presently, an in silico studies using molecular docking through InstaDock software was conducted to explore the interactions between MTX and its active metabolites such as methotrexate polyglutamate4 (MTXPG-4), methotrexate polyglutamate5 (MTXPG-5), 4deoxy-4-amino-N10-methylpteroic acid (DAMPA) and critical proteins involved in the anti-RA mechanism including thymidylate synthase (TS), reduced folate transporter (RFC1), proton-coupled folate transporter (PCFT), bifunctional purine biosynthesis protein ATIC, and dihydrofolate reductase (DHFR). Additionally, interaction zinc compounds viz. zinc aspartate, zinc citrate and their metabolites (aspartate and citrate ions respectively) with the proteins involved in immune function and inflammation including promyelocytic leukemia zinc finger (PLZF), B-cell lymphoma6 (BCL6), growth factor independence1 (GFI1), Blymphocyte-induced maturation protein1 (BLIMP1), thymus leukemia T-cell lineage-determining transcription factor (ThPOK), tumor necrosis factor- $\alpha$  converting enzyme (TACE), and matrix metalloproteinases (MMP1, MMP2, MMP7, and MMP10), as well as antioxidant enzymes superoxide dismutase1 (SOD-1) and superoxide dismutase2 (SOD-2) examining the potential for immune modulation and antioxidant activity without interfering with MTX's therapeutic targets. The preliminary findings reveal that zinc aspartate and zinc citrate interact favorably with these immune-related proteins, suggesting possible roles in enhancing immune flexibility, reducing inflammation, and promoting antioxidant defense in the context of MTX therapy.

Poster-04

## Enhancing preclinical drug screening with tumoroid models: the once seek bio approach

#### Reshma S\*, Ramya VVS, Nikita Naik, Suresh Poosala

Oncoseek Bio Pvt. Ltd, Visakhapatnam, India reshma@oncoseekbio.com

#### Abstract

The drug screening process using tumoroid models has gained significant attention due to its ability to mimic the complex 3D architecture, cellular heterogeneity, and microenvironment of human tumors. These models are established from patient-derived tumor tissues, cancer cell lines, or stem cells and cultured in a 3D environment that closely resembles actual tumors. In the screening process, tumoroids are exposed to various concentrations of potential therapeutic compounds, and their response is assessed using assays that measure cell viability, proliferation, apoptosis, and molecular changes. Advanced techniques such as immunohistochemistry and gene expression analysis further elucidate the drug's mechanism of action and potential resistance pathways. The importance of tumoroid models lies in their enhanced physiological relevance, leading to improved predictability of drug efficacy in preclinical studies, reduced reliance on animal testing, and their ability to support personalized medicine by allowing patient-specific drug screening. Additionally, their compatibility with high-throughput screening accelerates drug discovery. At OncoSeek Bio, we provide tumoroid and organoid models for various drug screening processes, making preclinical testing more efficient and significantly reducing the need for animal usage. Tumoroid models thus offer a more accurate and efficient platform for identifying effective cancer therapies, bridging the gap between in vitro studies and clinical outcomes.

Poster-05

## Performance evaluation of the RHE/IL-18 assay conducted on SkinEthic<sup>™</sup> RHE for identifying contact sensitizers compared to Local Lymph Node Assay

#### Kirti Tendulkar, Bipin Trada, Sanjaya R. J., Rina Gokani, Manish Rachchh

Accuprec Research Labs Pvt. Ltd., Opp. Zydus Pharmez, Changodar - Bavla Highway, Nr. Matoda Pstatiya, Ahmedabad, Gujarat 382213

am.bt@accuprec.in

#### Abstract

Allergic contact dermatitis is a significant occupational illness, accounting for 10% to 15% of all diseases. To assess the sensitization potential of compounds, the murine local lymph node assay (LLNA) has been widely used. In alignment with updated regulatory guidelines and the 4R's principle (Replacement, Reduction, Refinement, and Responsibility), LLNA test has been replaced with in vitro models. The objective of our study is to evaluate the performance of RHE/IL-18 assay using SkinEthic<sup>TM</sup> RHE tissue model for identification of skin sensitizing chemicals for regulatory purposes. A total of six samples were tested according to the RHE/IL-18 protocol, and the final assay outcomes were analysed to determine the most accurate prediction model. The study focused on IL-18 release levels and the assay performance compared to LLNA method. Results showed no significant performance differences across both methods, demonstrating that the SkinEthic<sup>TM</sup> RHE model is a viable alternative LLNA model. The study concluded that while the prediction model has been refined, further testing of additional substances is needed to gather more data and establish optimal criteria for using the SkinEthic<sup>TM</sup> RHE test system in this assay.

Poster-06

## Performance evaluation Monocyte Activation Test conducted on THP-1 cells for identifying pyrogenic contaminants compared to Rabbit Pyrogen Test

Shilpa Gupta, Bipin Trada, Sanjaya R. J., Rina Gokani and Manish Rachchh Accuprec Research Labs Pvt. Ltd., Ahmedabad, Gujarat <u>sd1.pt@accuprec.in</u>

#### Abstract

Pyrogen testing is considered a critical component in the product development and quality assurance of sterile pharmaceutical and medical products for their compliance with regulatory standards. The testing methods available for the detection of the pyrogen include in vivo Rabbit Pyrogen Test (RPT), Limulus Amebocyte Lysate test and Monocyte Activation Test (MAT). This study compares the efficacy of the in vitro Monocyte Activation Test with the conventional in vivo Rabbit Pyrogen Test (RPT) for the detection of pyrogenic contaminants in sterile pharmaceutical and medical products. The primary objective was to assess the reliability, sensitivity, and specificity of the MAT in relation to the established RPT. Experimental evaluation was conducted using both methods, and the outcomes were analyzed for their correlation and consistency. The results demonstrated that while the RPT effectively detected pyrogens, the MAT exhibited comparable sensitivity and specificity. Additionally, the MAT offered significant advantages in terms of rapidity and ethical considerations, as it eliminates the need for animal testing. Statistical analysis showed a strong correlation between the two methods, supporting the potential of the MAT as a viable alternative to the in vivo test in regulatory settings. In conclusion this research highlights the promise of the MAT in refining pyrogen testing practices, with the potential to improve both the efficiency and ethical standards in quality control for pharmaceutical and medical products. Further validation and standardization of the MAT are recommended to promote its broader acceptance and application in regulatory contexts.

#### Poster-07

#### Making pregnancies safe: A high throughput device to generate placental spheroids for assessing drug toxicity Pratik Rasal, Anshul Bhide, Deepak Modi

*ICMR-* National Institute for Research in Reproductive and Child Health, Molecular and Cellular Biology laboratory, Jehangir Merwanji Street, Parel, Mumbai- 400 012 *Prasal308(@gmail.com* 

#### Abstract

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#### 4ACAAE- SOUVENIR & ABSTRACT BOOK

Placenta is a critical organ for pregnancy that helps in exchanges of gases, nutrients and also

functions as an endocrine organ for maintaining pregnancy. Therefore, the proper functioning of the placenta during pregnancy is crucial and any disruptions to the placenta function can cause pregnancy related disorders including teratogenicity. During pregnancy, the use of most medications is restricted for their unknown effects on placenta and causing teratogenicity. However, many women need to take drugs due to pre-existing conditions like tuberculosis epilepsy, diabetes and sickle cell Anaemia. However, the restriction on the use of these drugs posies a challenge as the pregnant women cannot take the drug and suffer from their disease leading to significant morbidity and mortality.

Animal models are used to predict safety of drugs in humans. But historical experiences have shown that in pregnancy the animal data often do not accurately predict the drug afety/toxicity in humans. This is largely because of extreme structural and functional differences between human and animal placenta. Thus, Alternate model to assessing effects of drugs on placenta can be a surrogate measure of drug toxicity/safety in pregnancy, Herein we have developed a device to develop cell spheroids in a high throughput manner to permit drug testing in a rapid manner. Using this device we have generated spheroids of placental cells and tested cytotoxicity of selected anti-tuberculosis drugs. In addition, we have also used these spheroids to test the invasive & amp; migratory functions of the trophoblast cells in vitro. The data will be presented.

Poster-08

#### Risk Assessment of S017-622 by Salmonella Reverse Mutagenesis Assay

#### Neha Topno<sup>1,2</sup>, Navodayam Kalleti<sup>1</sup>, Anil Kumar Meena<sup>1</sup>, S. K. Rath<sup>1,2</sup>

<sup>1</sup>Division of Toxicology and Experimental Medicine, CSIR-Central Drug Research Institute, Lucknow 226031, Uttar Pradesh, India

<sup>2</sup>Academy of Scientific and Innovative Research, Ghaziabad 201002, Uttar Pradesh, India <u>neha.topno@gmail.com, navodayamk@gmail.com, anilhtox@gmail.com, skrath@cdri.res.in</u>

#### Abstract

Several new chemical entities are discovered that have the potential to cure diseases, but there can be potential toxicity of these molecules which can hinder its development as a successful drug molecule. There are different stages of non – clinical drug development of which, the genotoxicity assessment is one of them. Globally, all regulatory authorities require data for evaluation of genotoxic potential of a new chemical entity or new drug candidate as a part of safety evaluation data, before getting permission to initiate the clinical studies. In the regulatory studies, the Salmonella reverse mutagenesis assay is one of the gold standard in vitro tests, which is an alternate to animal model system available to decipher the mutagenicity of the molecule. In our study we tested the mutagenicity of CDRI compound, S017-622. The revertants obtained in the S017-622 treated plates in all the tester strains were comparable with that of vehicle treated controls both in the presence or absence of S9 mix. The compound was tested at 10, 100, 333, 1000, and 3333 ug/plate. It was found that the S017-622 was nonmutagenic compound.

Poster-09

#### Lung Organoids: A cutting-edge approach to infection and disease modeling

#### Alna Kuriyickal, Nandhakumaar R, Yogesh Sardana, Kiran Kumar Bokara

CSIR-Centre for Cellular and Molecular Biology, Hyderabad, Telangana, 500007, India <u>alnakuriyickal@ccmb.res.in</u>

#### Abstract

For decades, research has primarily relied on traditional 2D cell cultures and animal models, while valuable they often fall short in mimicking the intricate biological processes that underpin human physiology and diseases. As a transformative solution, we can turn to the recently emerging concept of organoids. While all organoid systems are invaluable, we chose to focus on the development of Lung Organoid models due to the critical role the lung plays in sustaining life. Lung organoids derived from human induced pluripotent stem cells (iPSCs) exhibit a 3D architecture that enables the formation of functional cellular structures, tissue-specific behaviors and dynamic intercellular interactions which can be employed to advance both disease research and therapeutic discovery. Differentiation medium containing growth factor such as FGF10, Wnt3a, and BMP4. After 3-4 days, the cells are transferred to a 3D culture system with additional factors like FGF7 and EGF to induce branching. Over 2-3 weeks, lung progenitors develop into organoids with branching structures resembling early-stage lung tissue. Upon stage specific characterization Surfactant protein B and Pro-Surfactant Protein C confirmed the presence of type-2 alveolar cells and MUC5AC expression confirmed

the presence of tracheobronchial secretory cells. Other markers such as SOX2 (Basal progenitors), NKX 2-1 (Alveolar Epithelial Marker), ACE2 (Tracheal Epithelial cells) and EpCam (Ciliated Bronchiolar Cells) were also used. The fully characterized mature Branching Lung Organoids were successfully infected by a known titre of SARS-CoV-2 virus to generate an infection model. The expression of spike protein along with ACE2, SPB and Pro-SPC suggested cell type dependent infection, thus generating a more physiologically relevant model for studying COVID-19 pathogenesis and drug testing. Leveraging these capabilities alongside CRISPR-Cas9 approaches will help us generate models with specific mutations, such as for CFTR.

#### Poster-10

#### Development of midbrain organoid through IPSC route for disease modelling

#### Renuga Devi, Hemasri Kilaru, Kiran Kumar Bokara\*

*CSIR-Centre for Cellular and Molecular Biology, Hyderabad, Telangana, 500007, India.* <u>\*bokarakiran@ccmb.res.in</u>

#### Abstract

One of the major hurdles for studying neurological diseases is the lack of human-like model systems. Recent studies demonstrated the use of human-derived cells to develop specific organ system (organoids) in-vitro. Organoid model system not only help us in understanding the development but also it paves the way for disease modelling and ultimately for new drug developments. Among the different brain regions, midbrain is considered to be one of the important regions regulating the sensory-motor control. Overexpression of alphasynuclein (SNCA) in the midbrain leads to a neurodegenerative condition named Parkinson's Disease (PD). Our present study optimized the development of midbrain organoids through human induced Pluripotent Stem Cells (hiPSCs) route. Briefly, the h-IPSC colonies were dissociated into single cells and were plated in ultralow attachment plates in specific media. After 48 hours, the embryoid bodies were formed and chosen for Neuro-ectodermal expansion with appropriate growth factors and inhibitors. After 26 days, the neuroectodermal stage organoids were maintained in differentiation medium. The cultures were maintained until 2 months. The stage specific characterization has been done with specific markers at embryoid bodies (EBs) formation using tubulin (ectoderm), AFP (Endoderm) and a-SMA(Mesoderm), Neuroectodermal (NE) differentiation stage using LMX1A and MAP-2, and Human Midbrain Organoids (hMOs) stage using Tyrosine hydroxylase and FOXA2. The thoroughly characterized hMOs are used for modelling PD like condition invitro. Studies are in progress to establish a PD organoid model utilizing the CRISPR-Cas9 system. This approach would be using homology-directed repair (HDR) to knock-in a Tet-on regulatable promoter with a reporter gene in iPSCs differentiating into neuronal stage. This enables us to mimic the overexpression of SNCA observed in PD condition. The hMOs could be used as an intermediate platform for disease modelling and drug testing which would minimize animal testing.

#### Poster-11

#### Nanoparticles of silver and titanium can increase temozolomide sensitivity against human glioblastoma: an in vitro study on apoptosis and inflammation

#### Sakshi Goswami, Yeshvandra Verma, SVS Rana

Department of Toxicology, CCS University, Meerut- 250004 U.P. sakshigoswami8909@gmail.com

#### Abstract

Temozolomide (TMZ) is one of the alkylating drugs that are preferably employed to treat human glioblastoma (GBM). However, its immune escape and resistance by GBM limit the survival of patients. Present in vitro study was undertaken to investigate whether AgNPs or TiO2NPs can enhance the sensitivity of GBM to TMZ through apoptotic and/or inflammatory responses. Human glioblastoma U87MG cells were exposed to different concentrations of TMZ, AgNPs and TiO2NPs after determining respective IC50 values, for 24h at 37°C under 5% CO2 atmosphere. Thereafter, biomarkers of LPO (malondialdehyde); apoptosis (caspase 3) and inflammation (TNF- $\alpha$  and IL-12) were assayed. Results on LPO, apoptosis and inflammation indicate that combined treatments with TMZ and AgNPs and TiO2NPs significantly increased the sensitivity of GBM to TMZ. It is suggested that over expression of drug efflux transporters, increased apoptosis and inflammation with nanoparticles. Gene expression changes induced by AgNPs and TiO2NPs may finally contribute in enhancing the sensitivity of GBM to TMZ.

Poster-12

#### Ursolic acid ameliorates 3-Nitropropionic acid-induced Huntington's disease-like symptoms in adult zebrafish: In-vitro and In-vivo evidence

#### Geetanjali Singh, Arti Singh

School of Health Science and Technology, University of Petroleum and Energy Studies, Dehradun, Uttarakhand geetanjali.130263@stu.upes.ac.in

#### Abstract

Huntington's disease (HD) is a hereditary disorder characterized by chorea and dementia. HD has also been linked to mitochondrial dysfunction and oxidative stress. 3-Nitropropionic acid (3-NP), a synthetic mitochondrial respiratory complex II inhibitor, is used to produce HD-like symptoms in zebrafish model. Ursolic acid (UA), a plant-based triterpenoid has been used as a neuroprotective agent, is able to decrease the effect of oxidative stress and improve balanced mitochondria. Material and Methods In vivo tests employed adult zebrafish, randomly grouped and pre-treated with UA (50 and 100 mg/kg; i.p) and tetrabenzene (TBZ; 3 mg/kg; i.p.) for three days, followed by 3-NP (15 mg/kg; i.p.) for five days. Comprehensive assessments encompassed behavioural, biochemical, molecular, mitochondrial, and histopathological analyses. Results In vivo zebrafish studies, where UA pre-treatment ameliorated behavioural deficits, reduced oxidative stress markers, quelled neuroinflammation, and enhanced mitochondrial function compared to 3-NP-treated zebrafish. Additionally, UA downregulated GSK-3β, MAPK14 expression while upregulating AMPK levels and improved mitochondrial and neuronal morphology, as demonstrated by TEM, H&E and Nissl staining. Conclusion Our research indicates that Ursolic acid has the potential to lower neurotoxicity and may provide neuroprotective benefits; yet, additional investigation is required to fully elucidate the underlying mechanisms.

Poster-13

#### Exploring alternative models of traumatic brain injury

#### Igra Mazahir, Suhel Parvez, Sheikh Raisuddin

Molecular Toxicology Laboratory, Department of Medical Elementology and Toxicology, School of Chemical and Life Sciences, Jamia Hamdard, New Delhi, India *iqramazahir sch@jamiahamdard.ac.in* 

#### Abstract

Traumatic brain injury (TBI) is notably high affecting up to 69 million people worldwide per year with an estimated annual incidence of 811 to 1507 cases per 100,000 people depending on the region. TBI models in animals have been developed. We investigated the neurobehavioral, biochemical and histopathological changes in controlled cortical impact (CCI) rat model of TBI to evaluate protective effect of a neuroprotectant,  $\alpha$ -lipoic acid (ALA). However, I wondered if there were another paradigm of traumatic brain injury? It was revealed that the fly (Drosophila) model of TBI can provide unique insights into human TBI because the fly TBI model enables us to analyze many animals throughout their lifespan. Drosophila TBI models have demonstrated key features of mammalian TBI, including temporary incapacitation, disorientation, motor deficits, activation of inflammation, and autophagy responses observed immediately after injury. To demonstrate the induction of injury, ten-day-old transgenic flies received an injury of increasing angles from a modified high impact trauma (HIT) device where angle-dependent increase occurred in acute neurological behaviour. It was also noted that several measures of oxidative stress in Drosophila can be correlated with TBI. Although Drosophila model does not fully mimic all phenomena involved in post-TBI secondary injury in mammals, further experiments with Drosophila models and other animal models could elucidate the mechanisms involved in post-TBI secondary brain injury. Another cellular model involves mechanical transection and cell stretch injury. An impact device with 28 stainless steel blades has been used to induce mechanical damage on cultured rat cortical neuron cells. The cutting device produced uniform incisions in the cell layer of the tissue culture plate. Although the application of TBI models in the study of brain injury has made some progress, there are still some insuperable shortcomings. The brains of commonly used TBI model animals (especially rodents) are physiologically similar to human brains. Both types of TBI models have advantages and disadvantages. Therefore, different types of in vitro and in vivo TBI models should be combined, when studying a new treatment or drug, to simulate different pathobiological reactions caused during injury. Cross-validation in this way can make the experimental results more robust and reliable, and reduce the false positive neuroprotective effect of some drugs or treatments. Our study will adopt such a strategy to study protective effects of ALA against TBI.

## Unraveling the molecular impacts of bisphenol A on ADHD: An in silico toxicoinformatics approach

#### Huma Khan, Mohd. Anas Saifi, Shahzad Ahmed, Garima Jindal, Sheikh Raisuddin\*

Department of Medical Elementology and Toxicology, Jamia Hamdard, New Delhi. dr.huma3092@gmail.com

#### Abstract

**Background:** Bisphenol A (BPA), a common environmental contaminant found in plastics, has raised concerns due to its potential role in neurodevelopmental disorders, particularly ADHD. BPA is known to disrupt endocrine function, but its precise molecular mechanisms in ADHD remain unclear. In silico toxicoinformatics offers a powerful approach to simulate and predict molecular interactions, providing valuable insights into the potential impact of BPA on ADHD.

Objective: This study uses in silico methods to investigate BPA molecular interactions with key biological targets involved in neurodevelopment and behavior, with a focus on identifying pathways relevant to ADHD.

**Methods:** Using molecular docking, virtual screening, and network analysis, we modelled BPA interactions with dopamine receptors and neurodevelopment-related proteins. We integrated data from toxicology databases and gene expression studies to map disrupted pathways involved in ADHD. Pathway enrichment analysis was conducted to identify the most significantly affected biological processes.

Results: In silico simulations revealed that BPA binds strongly to dopamine receptors, suggesting significant disruption of hormonal and neurotransmitter systems. The results showed greater interactions of BPA and its metabolites in comparison to respective known inhibitors of those enzymes. The promising docking scores of the proteins of PPI networks of selected proteins involved in ADHD pathophysiology compared to their respective known ligands have also shown the contribution of BPA to ADHD pathophysiology through PPI network. BPA was predicted to interfere with dopamine receptor-mediated signaling, crucial for attention and behavior regulation, and impact pathways related neurogenesis. to Conclusion: This study demonstrates that BPA may contribute to ADHD through multiple entry points. Many close interactions between BPA and specific proteins were found through molecular docking including Van der Waals, hydrophobic, polar, and H-bonding interactions. KEGG pathway analysis, GO, and other enrichments exhibited the various biological, cellular, and molecular functions involved in the PPI network of selected proteins.

Poster-15

Poster-14

## A new approach on determining lethal concentration of cobra venom using zebra fish embryos

#### Madhura Pramod Ghatkar, Mrunal Ghag Sawant, Mrs. Swati Mumbarkar

Haffkine Institute for Training, Research and Testing, Acharya Donde Marg, Parel, Mumbai-400012 <u>madhughatkar1@gmail.com</u>

#### Abstract

Zebrafish is one of the emerging and highly utilized model organisms used for multiple studies such as regeneration and toxicology-based assays. Cobra venom is a potent neurotoxin and cardiotoxin which leads to physiological and anatomical changes in snake envenomation. Snake venom components include phospholipases A2, metalloproteinases, three-finger toxins, and L-amino acid oxidase, in current study, the acute toxicity of Cobra venom was determined by using multiple concentrations and its effects on the embryos were observed until 50% mortality. All other surviving embryos were observed till 96 hours. The test was only conducted till the Cobra venom reached 50% of mortality which varied among time points 24, 48, 72, and 96 hours. The physiological and anatomical effects were also assessed for all exposed embryos. Multiple anatomical changes were observed within the embryo and developed larvae at various concentration which is due to the toxic nature of Cobra venom. This novel approach will enable us to conduct venom research using Zebrafish as a model organism rather current animal model of choice i.e., mice or rats. It may also eventually help us to replace the use of rodents for venom studies and to inculcate one of the 3 Rs i.e., Replacement if it is a success.

#### Poster-16

#### Hydra: an emerging alternate model for assessing toxicity of contaminants

#### Afifa Parveen, Aasia Razi, Smriti Dhara, Anam Ali Khan

Department of Medical Elemtology and Toxicology, School of Chemical and Life sciences Jamia Hamdard, 110062.

*Email-afifaparveen33@gmail.com, aasiarazi02@gmail.com, smritidhara140@gmail.com, anamkhan3845@gmail.com* 

#### Abstract

Pharmaceutical contaminants, including antibiotics, antidepressants, and painkillers, are increasingly prevalent in aquatic ecosystems due to wastewater discharge, posing significant risks to non-target organisms. These emerging pollutants (EPs), which also encompass personal care products and industrial chemicals, persist in the environment and disrupt aquatic ecosystems through teratogenic, reproductive, and behavioural effects. Understanding their toxicological impact on non-target species is essential for environmental risk assessments and regulatory frameworks. This study employs the Hydra attenuate regeneration assay, a robust tool for aquatic toxicity testing, to evaluate the effects of few pharmaceuticals such as gemfibrozil, ibuprofen, naproxen etc that are identified in the effluents of a major wastewater treatment plant (WWTP). Hydra, a freshwater enidarian with remarkable regenerative capacity and sensitivity to pollutants, serves as an ideal model organism due to its simple anatomy and well-characterized molecularpathways. Exposure to these pharmaceuticals led to significant bioaccumulation, impaired regeneration, reduced reproductive success, and altered behaviours. Key molecular mechanisms of toxicity included reactive oxygen species (ROS) generation, oxidative stress, DNA damage, and disruption of critical pathways such as Wnt signalling, which are essential for regeneration. Advanced techniques, including comet assays, acridine orange staining, and real-time PCR, revealed pollutant-induced oxidative stress, modulation of antioxidant enzyme activities, and upregulation of stress- responsive genes. The study also investigated the impacts of specific pollutants like Bisphenol A (BPA) and cobalt on Hydra's regenerative and reproductive biology, revealing concentrationdependent effects, DNA damage, and apoptosis via mitochondria-mediated pathways. These findings emphasize the ecological risks of pharmaceutical contamination in water bodies and highlight the potential of Hydra as a model organism for environmental toxicity assessment. Hydra's sensitivity to pollutants and ethical advantages over vertebrate models make it an invaluable tool for studying the molecular mechanisms of EP toxicity and advancing environmental regulatory practices.

Poster-17

#### Non-Mammalian Model Organism

#### Fiza A Shaikh, Zaid Sayeed

School of Pharmaceutical Education and Research, Jamia Hamdard, New Delhi <u>fizaansarulhaque2001@gmail.com</u>

#### Abstract

The reliance on animal models in preclinical research has been instrumental in advancing biomedical science. However, escalating ethical concerns, regulatory compliances, and the inherent limitations of mammalian systems in accurately predicting human outcomes accentuate the need for innovative alternatives. Nonmammalian models, specifically invertebrates such as cockroaches, silkworms, and Caenorhabditis elegans (C. elegans), have emerged as effective substitutes, offering distinct advantages for preclinical investigations. Cockroaches, well known for their robust innate immune mechanisms and exceptional resilience, serve as a remarkable tool in toxicological and antimicrobial studies. Similarly, Silkworms, with their well-characterized immune system and scalability, have demonstrated efficacy in infection modelling and pharmacological screening. Likewise, C. elegans, a genetically tractable nematode with conserved biological pathways and a short lifecycle, facilitates high-throughput analyses in neurodegenerative, metabolic, and developmental disorders. The adoption of these models confers numerous advantages, including alleviation of ethical dilemmas, reduced experimental costs, expedited timelines, and diminished regulatory barriers. Furthermore, their utilization aligns with the principles of the 3Rs (Replacement, Reduction, Refinement), promoting ethical and sustainable research practices. This poster illustrates the rationale for employing non-mammalian models in preclinical research, highlights their comparative advantages over conventional mammalian systems, and highlights recent breakthroughs in the field. By embracing these alternatives, the scientific community can advance ethically conscious, cost effective and methodologically innovative approaches to address the complexities of biomedical research while reducing dependence on preexisting traditional vertebrate models.

Poster-18

#### Standardization of botanicals: A necessary step towards alternative testing

#### Zainab Mantasha\*, Madiha Ashfaque

Department of Pharmacognosy and Phytochemistry, SPER, Jamia Hamdard, New Delhi-110062, India <u>zainab.dziam@gmail.com</u>

#### Abstract

Botanical standardization is a significant step in pharmaceutical research because it ensures the consistency, purity, and reliability of plant-based chemicals utilized in medication discovery and development. Standardized botanicals are now essential for obtaining precise and reliable outcomes as the scientific community moves more and more toward non-animal, alternative testing techniques. This abstract explores the significance of botanical standardization in enhancing the reliability of in vitro and in silico testing, facilitating safety and efficacy assessments, and meeting regulatory requirements. It outlines the methodologies involved in standardizing botanicals, including the identification of key active ingredients, quantitative analysis, and adherence to good manufacturing practices (GMP). Furthermore, the challenges associated with the complex chemical composition of plants, environmental variability, and extraction methods are discussed. Advancements in analytical technologies, such as HPLC, LC-MS, and NMR, are highlighted as tools for improving standardization processes. By ensuring the consistent quality of botanicals, standardization plays a pivotal role in supporting the transition to ethical, efficient, and accurate alternative testing paradigms, ultimately contributing to the development of safer and more effective therapeutic agents. **Keywords:** Botanical Standardization, In Silico Testing, Good Manufacturing Practices (GMP), Safety

Poster -19

#### **Organ-On-Chip: A novel approach to toxicological evaluation**

#### Bushra Ali, Sheikh Raisuddin

Department of Medical Elementology and Toxicology, School of Chemical and Life Sciences, Jamia Hamdard, New Delhi-110062 ali12.bushra@gmail.com

#### Abstract

The question comes up whether and how organ-on-chip (OoC) can be useful as tools in the innovating arena of toxicological hazard and risk assessment. OoC models are increasingly proving useful in providing opportunities to study complex biological systems in a controlled in vitro environment. Clearly, as compared to simple single-cell type in vitro assays, OoC provide added complexity. Both the evolutionary and the revolutionary approach aim to transform toxicology and pharmacology from being predominantly based on animal testing to relying on other relevant approaches. One of the major benefits of OoCs is their great potential to recapitulate human physiology. This is especially relevant with the use of intact tissues instead of individual cells, enabling interaction between cells through direct contact, and between cells and extracellular components such as the extracellular matrix. A principal advantage of OoC over animal models is the possibility to introduce human-specific targets and thus identify compounds that exert their toxicological or pharmacological effect. Even in case, an OoC has some degree of predictivity, this does not necessarily lead to a reduction in the number of experimental animals, but may rather add another layer of knowledge on safety or reduce the attrition rate in drug discovery. Therefore, in case the use an OoC does not lead to reduction in animal use, the OoC still has merit in a better-founded safety profile or a reduced attrition rate. In the next phase of our study, we will examine the challenges and potential advantages of using Organ-on-Chip (OoC) technology for evaluating the safety and effectiveness of toxicological and pharmaceutical aspects, as well as the necessary actions to accomplish the future target.

#### Poster-20

#### A human iPSC-CM model (YBLiCardio) for predicting cardiotoxicity and drug safety

Reddy V.<sup>1</sup>\_More S.<sup>1</sup>, Dhepe S.<sup>1</sup>, Gossmann M.<sup>2</sup>, Lickiss B.<sup>2</sup> Linder P.<sup>2</sup>, Bhanushali P<sup>1</sup>, Khanna A.<sup>1</sup> <sup>1</sup>Integrated Drug Discovery & Development, Yashraj Biotechnology Ltd. Navi Mumbai. <sup>2</sup> innoVitro GmbH, Jülich, Germany *vijay.konala@yashraj.com* 

#### Abstract

Myocardial contractility (inotropy) is an essential property of cardiac function and must be maintained at constant physiological level. Non-cardiac drugs causing unintended contractility effects can lead to adverse cardiac events including contractile dysfunction, induction of arrhythmic events, and heart failure, limiting the utility of novel innovative treatments. Cardiac contractility evaluation using human induced pluripotent stem cell-derived cardiomyocytes (hiPSC-CMs) has recently attracted much attention as a preclinical cardiotoxicity predictive model. Most studies on this were conducted under spontaneous beating conditions and involved video-based analyses. Cardiac contractility is known to be influenced by beating rates; accordingly, beating rate measurement is recommended to accurately analyze the effects of drugs on cardiac contractility. We present a human pluripotent stem cell-derived cardiomyocyte model that allows for the prediction of both acute and chronic adverse effects on human cardiomyocyte function. For this purpose, we adopted a protocol for efficiently differentiating human pluripotent stem cells into fully functional human cardiomyocytes, referred to as YBLiCardio. This process utilizes a defined, culture medium in combination with small molecules and can be efficiently scaled from a 96well microplate format to larger formats. The resulting cardiomyocytes exhibit the expression of cardiac-specific markers such as Troponin, MLC and Alpha actin and display electrophysiological properties that validate their status as functional and mature cardiomyocytes. These induced pluripotent stem cell-derived cardiomyocytes prove invaluable in their efficiency of identifying cardiotoxicity hazards for a wide range of substances, including pharmaceuticals and non-pharmaceutical compounds.

Human-induced pluripotent stem cell-derived cardiomyocytes (hiPSC-CMs) present a promising *in vitro* model for predicting cardiotoxicity across different toxicity profiles. We have developed YBLiCardio, an hiPSC-CM model derived from the YBLi001 line, generated from a healthy donor using Sendai virus technology. Differentiated cardiomyocytes showed a seeding efficiency of over 80% with a unique cocktail mixture optimized for cardiomyocyte culture. YBLiCardio was validated for both acute and chronic toxicity assessments, including the Comprehensive in vitro proarrhythmia Assay (CiPA)

#### Poster-21

## Computational insights into bisphenol A and its analogs interactions with PCOS-related ovarian toxicity pathways

#### Mehjabeen Javed, Suramya, Humaira Naaz Bhutto, Shaesta Shahid, Sheikh Raisuddin\*

\*Molecular Toxicology Laboratory, Department of Medical Elementology and Toxicology, School of Chemical and Life Sciences, Jamia Hamdard, New Delhi- 110062, India <u>mehjabeenjaved sch@jamiahamdard.ac.in</u>

#### Abstract

Polycystic ovary syndrome (PCOS) is a multifactorial endocrine disorder affecting ovarian function, with growing evidence implicating environmental contaminants such as Bisphenol A (BPA) and its analogues in its pathophysiology. This study employs computational techniques to investigate the molecular interactions of BPA and its structural analogues with key pathways associated with PCOS-related ovarian toxicity. Using molecular docking simulations, we examined the binding affinities of BPA and its analogs to critical proteins involved in ovarian function, such as androgen receptors (AR), insulin-like growth factor-1 receptor (IGF-1R), and aromatase (CYP19A1). Our findings reveal that BPA and its analogs exhibit strong interactions with these targets, potentially disrupting hormonal signaling, steroidogenesis, and metabolic processes. The structural insights provided by this analogs. These results enhance our understanding of the mechanistic role of BPA in ovarian dysfunction and PCOS, offering a foundation for further experimental validation and risk assessment. This computational framework underscores the value of in silico approaches in elucidating environmental toxin interactions with disease pathways.

Poster-22

## Exploring aristolochic acid-induced nephrotoxicity through *in Silico* assessment of key biomarkers

#### Suraj Kumar, Suramya, Sheikh Raisuddin\*

\*Molecular Toxicology Laboratory, Department of Medical Elementology and Toxicology, School of Chemical and Life Sciences, Jamia Hamdard, New Delhi, 110062. sk727713(@gmail.com

#### Abstract

Aristolochic acid (AA), a natural compound found in Aristolochia plants, is well-known for its nephrotoxic and carcinogenic properties, posing significant health risks globally. This study employs an *in-silico* approach to

investigate the molecular interactions of AA with key biomarkers involved in kidney injury, inflammation, and oxidative stress. Computational techniques, including molecular docking and virtual screening, were used to identify potential binding affinities and interaction sites of AA with target proteins such as kidney injury molecule-1 (KIM-1), nuclear factor-kappa B (NF- $\kappa$ B), and antioxidant enzymes like superoxide dismutase (SOD) and catalase (CAT). Results highlight AA's strong affinity for KIM-1 and its potential to upregulate pro-inflammatory pathways mediated by NF- $\kappa$ B, contributing to inflammation and oxidative damage. Furthermore, the analysis suggests that AA disrupts the activity of key antioxidant enzymes, exacerbating oxidative stress in renal tissues. These findings provide valuable insights into the molecular mechanisms underlying AA-induced nephrotoxicity and identify potential therapeutic targets to mitigate its effects. This in silico framework lays the foundation for future experimental validation and drug discovery efforts aimed at addressing AA-related kidney toxicity.

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### 4<sup>th</sup> Asian Congress for Alternatives to Animal Experiments (4ACAAE) on Non-animal Approaches: Concept, Validation and Regulatory Acceptance

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7<sup>th</sup> Annual Conference of Society for Alternatives to Animal Experiments -India

> December 12-14, 2024 Jamia Hamdard (Deemed to be University) New Delhi 110062, India

#### PLAN OF PRE- AND POST-CONGRESS WORKSHOPS/SATELLITE CONFERENCES

	Dates	Venue	Theme/topic	Coordinator(s)
1	November 19-23, 2024	Kerala University, Trivandrum	Alternative animal models – <i>C. elegans</i> and <i>Drosophila</i>	Dr. Sreejith Parameswara Panicker Email: <u>p.sreejith@gmail.com</u>
2	December 5 - 7, 2024 (Hybrid Mode)	Mahatma Gandhi Medical Advanced Research Institute, Pondicherry	International Conference and Workshops on New Approach Methodologies (NAMs) of Pharmacology and Toxicology (A Satellite Programme to 4th ACAAE & 7th SAAE- I and, Jamia Hamdard, New Delhi, December 12- 14, 2024)	Dr. Veni Subramanyam Email: <u>venis@mgmari.sbvu.ac.in</u>
3	December 10-11, 2024	CSIR-IGIB, New Delhi	3D Bioprinting	Dr. Vijay Pal Singh Email: <u>vpsinghigib@gmail.com</u>
4	December 16-17, 2024	Jamia Hamdard, New Delhi	Leveraging <i>in vitro</i> and <i>in silico</i> toxicology and computational	Dr. Mohan Kamthan & Dr. Shahzad Ahmad Faculty – Dr. Fazal Khan, Sultan Qaboos Comprehensive Cancer Care &

			biology in the risk	Research Center, University Medical
			assessment	City, Muscat, Oman
				Email:
				mohan.kamthan@jamiahamdard.ac.in;
				satox@jamiahamdard.ac.in
5	December	Jamia	C. elegans and other	Prof. Suhel Parvez
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	2024	New Delhi	a model for	Email: sparvez@jamiahamdard.ac.in
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			disorders	
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	26-30,	University,	bioprinting	3
	2024	Calicut	1 0	Email: <u>divyacuk@gmail.com</u>
7	December	СТ	Zebrafish model in	Dr Vir Vikram Sharma
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	2024	Ludhiana	research	Email: virvikram76@gmail.com
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	9-10,	New Delhi	pharmacology – an	
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#### Guidelines

- 1. Workshops/Events will be conducted as part of 4ACAAE & 7SAAE-I.
- 2. Workshops/Event brochures will be advertised on the Congress website/circulars.
- 3. Due coverage will be given to the Society (SAAE-I). Its logo will be placed on all Workshop/Event material.
- 4. Up to 30 participants may be allowed. However, depending on facilities more participants may be allowed.
- 5. Moderate registration fee may be charged in order to encourage participation of young research students and post-doctoral researchers.
- 6. The Coordinator of the Workshop/Event will be free to apply for grant and sponsorship keeping the Society in loop.
- 7. A 10% portion of the amount of Registration Fee shall be shared with the Society for its corpus.
- 8. Invitation will be sent to the President, Vice President(s), General Secretary, Joint Secretaries and Treasurer of the SAAE-India and Organizing Secretary 4ACAAE.

(Prof. M.A. Akbarsha) General Secretary SAAE-I (Prof. S. Raisuddin) Organizing Secretary 4ACAAE & 7SAAE-I

### SOCIETY FOR ALTERNATIVES TO ANIMAL EXPERIMENTS-INDIA (SAAE-I)



#### C/o National Centre for Alternatives to Animal Experiments

#### Bharathidasan University, Tiruchirappalli 620024, India

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26.11.2024

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#### **Dear Friends**

It gives immense pleasure to notify that

Dr. Prajakta Dandekar Jain UGC Assistant Professor Dept of Pharmaceutical Sciences and Technology Nanomedicine Research Group ICT Mumbai

Has been selected by the duly appointed Committee of Juries as winner of Dr. Dipti Kapoor Endowment Award 2024. Among his many contributions, the outstanding ones are "Development of alternative-to-animal models: organ-on-chip models for drug development and testing; and designing of MITO Bioprinter". Needless to say that the other five contestants did contribute commendably but Dr. Prajakta's contribution has an edge over the others'. While congratulating her for this wonderful feat and thanking her for honoring SAAE-I by presenting herself as a candidate, let's welcome her to Delhi on 12<sup>th</sup> Dec 2024 to receive the award and talk about her contribution to Alternatives/NAMs.

Sincerely

General Secretary, SAAE-I

## 4<sup>TH</sup> ASIAN CONGRESS FOR ALTERNATIVES TO ANIMAL EXPERIMENTS (4ACAAE)





Non-animal Approaches: Concept, Validation and Regulatory Acceptance



7<sup>th</sup> Annual Meeting of the Society for Alternatives to Animal Experiments – India

## JAMIA HAMDARD US ALUMNUS BEST POSTER AWARD

The Award

Dr. Bilal Bin Hafeez, School of Medicine, South Texas Center of Excellence in Cancer Research, University of Texas Rio Grande Valley, Texas, USA has Instituted the Award to be given as 1<sup>st</sup>, 2<sup>nd</sup> and 3<sup>rd</sup> Best Poster Awards at 4ACAAE.

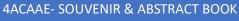
## Eligibility

Only Jamia Hamdard Ph.D. Scholars and Post Graduate students who will participate in the 4ACAAE.

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A Certificate along with Cash Prize will be presented to the Winners in the Valedictory Session on 14/12/2024.

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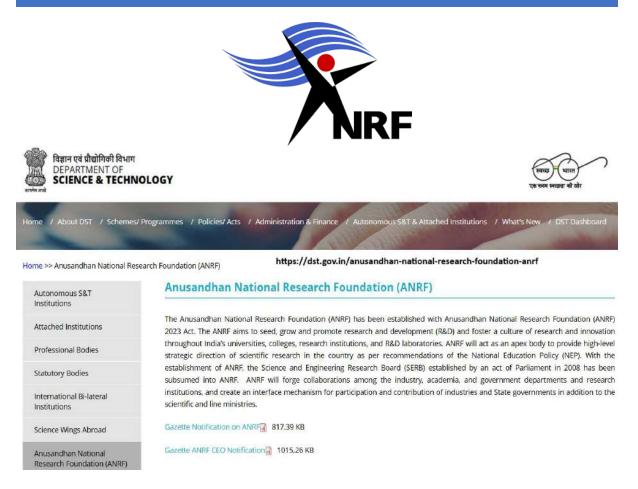
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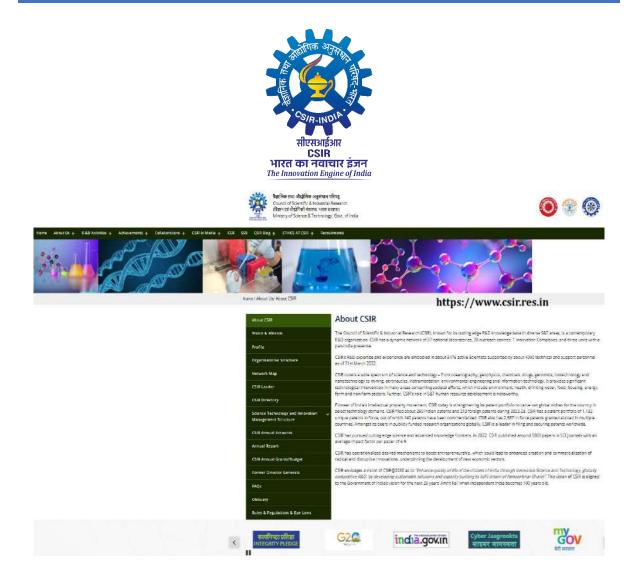
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The Japanese Society for Alternatives to Animal Experiments

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Purpose of the activity	An It's It's Annal Wasting of the Agenese South for Adventions to Advent Experim 日本動物実験代替法学会 第37回大
The Japanese Society for Alternatives to Animal Experiments (JASAL) is an academic organization that conducts research, development, education, surveys, etc. with the aim of promoting and	2024年 11月29日>12月1日 ・ライトキューフ字都会 ・第日 東 Electer
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suffering), and Replacement (replacement with alternative methods that do not use animals) *see	日本動物実験代替法評価センタ
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footnote), which are the international principles for the proper conduct of animal experiments. The 3Rs principles were proposed by Russell and Birch in 1959, and are the basic principles for conducting animal experiments that were adopted as the Bologna Declaration at the 3rd International Conference on Alternatives in 1999. These principles have been adopted in recent years not only in the laws and guidelines of each country related to laboratory animal welfare, but also in international standards and guidelines.







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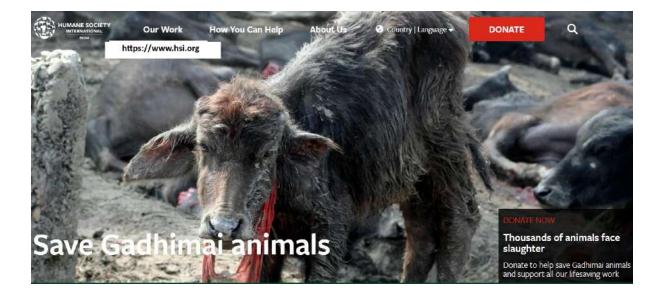
### Doerenkamp-Zbinden Foundation for Animal-Free Research

We advocate for alternatives to animal testing and promote innovative solutions to advance animal-free research and development.



Our work About Us





# **Peta** INDIA





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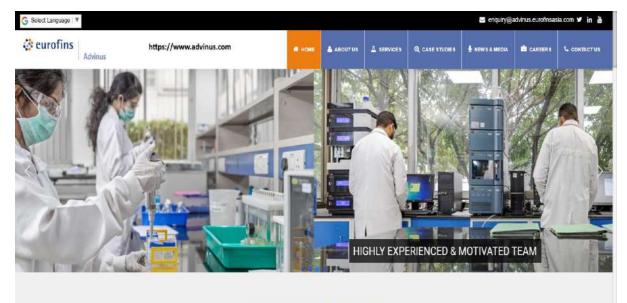


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#### Society for Alternatives to Animal Experiments – India (SAAE-I)



The genesis of the Society for Alternatives to Animal Experiments-India (SAAE-I) could be traced back to the establishment of Mahatma Gandhi-Doerenkamp Centre (MGDC) with funding from Doerenkamp-Zbinden Foundation, Switzerland, in 2009, at Bharathidasan University, Tiruchirappalli, Tamil Nadu, with the aim of fostering the 3Rs movement in India. In 2016, the MGDC became the National Centre for Alternatives to Animal Experiments (NCAAE) with the operational fund coming from the University Grants Commission of Government of India. Soon it was felt that an exclusively academic endeavour will not be able to translate the vision of a country embolden with non-animal methods in education, biomedical science and toxicology, and so the supporters of non-animal methods of India come together and establish a national society. Thus was born the Society for Alternatives to Animal Experiments-India (SAAE-I), head-quartered at NCAAE, Bharathidasan University, Tiruchirappalli, Tamil Nadu in July, 2018. The first Annual Conference was conducted at Jamia Hamdard, New Delhi, in December, 2018, accompanied by the First Executive Committee and General Body Meeting, SAAE-I which is now a Registered Society. It is one of the most regular, vibrant scientific societies in India with regular annual meets in different parts of the country, which provide platform for all scientists CROs, Industry and NGOs to remain connected to the domains of 3Rs/Alternatives/NAMs to meet and deliberate about shifting to non-animal methods and evolve appropriate strategies. The SAAE-I promotes humane education and research by way of introducing alternatives to the use of animals which will promote the internationally accepted "Concept of 3R's"- or "Alternatives" - Reduction, Refinement and Replacement - of use of animals in education, research and testing. The SAAE-I subscribes to the Asia Congress, and hosts the Fourth Asia Congress. It has moved into MoU with Japanese Society for Alternatives to Animal Experiments (JSAAE), and intends to enter into MoU with such other International Societies. SAAE-I is a party to the Asian Federation, which is coming up now. The Society is working for establishment of a Validation Centre for Alternatives in India.

#### **Executive Council of SAAE-I**

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Asian Federation of Societies for Alternatives to Animal Experiments (AFSAAE) Declaration of Formation on December 13, 2024 at 4ACAAE Jamia Hamdard, New Delhi, India.