SOUVENIR-CUM-COMPENDIUM

SVBBI-2023

VII ANNUAL CONVENTION OF SVBBI AND INTERNATIONAL SYMPOSIUM

on

Multiomics to One Health: Challenges and Way Forward in Biomedical Research

DECEMBER 14-15, 2023

Organized by

DIVISION OF BIOCHEMISTRY

ICAR- Indian Veterinary Research Institute
Izatnagar - 243122 (U.P.), India

&

Society of Veterinary Biochemists and Biotechnologists of India (SVBBI)

Concept of Hatchery Vaccination

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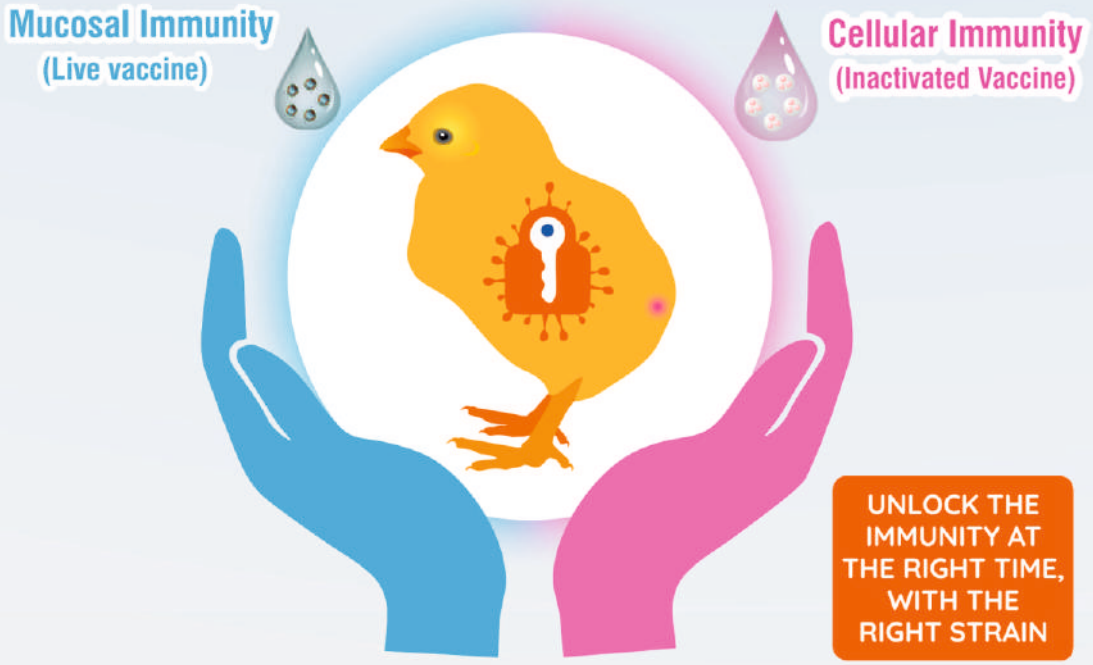
THE BEST IN ANIMAL HEALTHCARE

IBD-VIPx

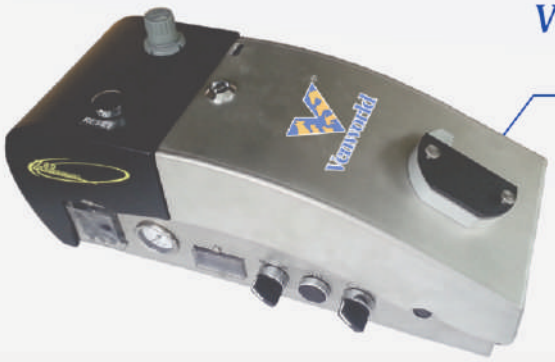
VENTRI'S IMMUNE PLUS COMPLEX

'CATCHING UP WITH THE FUTURE'



VH ND SHIELD

Protection against both endemic Genotype XIII and emerging Genotype VIIi circulating in India



Vaccinator can Accommodate VIPx and ND Shield

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SOUVENIR-CUM-COMPENDIUM

SVBBI-2023

VII Annual Convention

of

Society of Veterinary Biochemists and Biotechnologists of India (SVBBI)

and International Symposium

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Multiomics to One Health: Challenges and Way Forward in Biomedical Research

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ICAR- Indian Veterinary Research Institute

Izatnagar - 243122 (U.P.), India

https://www.ivri.nic.in

SVBBI-2023

VII Annual Convention of Society of Veterinary Biochemists and Biotechnologists of India (SVBBI)
and
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December 14-15, 2023

Published & Printed By

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We declare that editors compiled these abstract received in response to their invited lectures and not responsible for duplicity of the materials published previously or accepted for publication in English or any Other language.

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ORGANIZING COMMITTEE

SVBBI-2023

VII Annual Convention of Society of Veterinary Biochemists and Biotechnologists of India (SVBBI)

and

International Symposium

on

Multiomics to One Health: Challenges and Way Forward in Biomedical Research

December 14-15, 2023

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ICAR-IVRI, Izatnagar

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Message

I am pleased to know that that the Division of Biochemistry, ICAR-IVRI, Izatnagar is organizing the VII Annual Convention of Society of Veterinary Biochemists and Biotechnologists of India (SVBBI) and International Symposium on "Multiomics to One Health: Challenges and Way Forward in Biomedical Research" during December 14-15, 2023 at Izatnagar, Bareilly(UP).Biochemistry is important in veterinary medicine because it helps to understand the basic cellular function of animals, thereby improving livestock production and health. The scientific program and theme of the convention highlight the present advances in multiomic approaches in one health and the excellent line-up of plenary lectures and invited speakers from both national and international reputes.

I hope the galaxy of scientists, academicians, entrepreneurs and students attending this symposium would discuss the emerging problems relating to livestock health and production. I am confident that the recommendations would have a far reaching effect in the progress of veterinary research. I wish the SVBBI all success in organizing this scientific event.

I wish the symposium a grand success.

5th December, 2023
New Delhi

(Himanshu Pathak)

भारतीय कृषि अनुसंधान परिषद
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Message

It gives me immense pleasure to know that the division of Biochemistry of the ICAR-Indian Veterinary Research Institute (IVRI), Izatnagar is celebrating its Golden jubilee, and organizing the VII Annual Convention of SVBBI and International Symposium on "Multiomics to One Health: Challenges and Way Forward in Biomedical Research" on 14-15 December 2023 under the aegis of the Society of Veterinary Biochemists and Biotechnologists of India (SVBBI).

Biochemistry and Biotechnology are the two disciplines which have made a major impact on animal health, production and livestock product technology all over the world. These disciplines are the essence of fundamental research that has given a thrust for innovations in tackling the problems of diagnosis and control of diseases and for selection of desired traits. The conference theme will focus on new and innovative research areas in biomedical research, including genomics, proteomics, metabolomics, vaccinomics, gene editing, nanotechnology-based biosensing, and computational biology.

I sincerely believe that the conference will provide an appropriate platform for the academicians, biomedical researchers, scientists, professionals, industry partners, and entrepreneurs associated with the subjects of both Biochemistry and Biotechnology for scientific discussions, sharing knowledge and interactions to better understand and integrate multi-omics data with conventional biomedical research for improvement in animal health and productivity. I am sure, the recommendations emerging from the deliberations will have far-reaching implications in designing future research programmes in the country.

I wish the Symposium a great success.

7/12/2023

(J.K. Jena)

भाकृअनुप-भारतीय पशु-चिकित्सा अनुसंधान संस्थान
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इज्जतनगर-243 122 (उ०प्र०) भारत
ICAR-Indian Veterinary Research Institute
(Deemed University)
Izatnagar-243 122 (U.P.) India

डॉ त्रिवेणी दत्त
निदेशक एवं कुलपति
Dr Tiveni Dutt
Director-cum-Vice Chancellor

Message

The integration of multiomics and One Health approaches offers a more comprehensive and interconnected perspective on health and disease, encompassing genetics, environmental, and socio-economic factors acros species boundaries.Multiomics allows researchers to study the impact on the genomes, transcriptomes, and metabolomes of organisms and providing insights into how environmental changes may affect health. It can help to identify potentital zoonotic threat by studying the genetic makeup of pathogens, understanding their transmission dynamics, and assessing the risk factors associated with their spread. The recent advance in the biomedical research has been making tremendous impact on animal health, production and livestock product technology all over the world. Biochemistry and Biotechnology are the essence of basic and fundamental research and innovation that has given athrust for new innovations in tackling the problems of diagnosis and control of diseases and for selection of desired traits.

Division of Biochemistry, Indian Veterinary Research Institute, Izatnagar is celebrating its Golden jubilee celebration, and organizing the VII Annual Convention of SVBBI and International Symposium on "Multiomics to One health: Challenges and Way Forward in Biomedical Research" from 14th - 15th Dec. 2023 under the aegis of Society of Veterinary Biochemists and Biotechnologists of India (SVBBI). This symposium will create a platform for exchange of knowledge and innovation in the area of animal health with more emphasis on one health amound national and international delegates.

I extend a warm welcome to delegates from various corners of the country and around the world who will be attending SVBBI 2023. I hope that this conference will not only provide an opportunity to identify and design future research programs for out country but will also establish valuable linkages and platforms for collaborative research with our international counterparts. My heartfelt congratulations to the organizing team for their dedicated planning and execution of this international symposium.

I wish all participants and the organizing committee a resounding success at SVBBI 2023.

(Triveni Dutt)
7/12

ICAR-CENTRAL AVIAN RESEARCH INSTITUTE
Izatnagar-243122

Dr Ashok Kumar Tiwari
Director
ICAR-Central Avian Research Institute
Izatnagar-243122 (U.P.), India

Message

It gives me immense pleasure to know that the Division of Biochemistry of the ICAR-Indian Veterinary Research Institute, Izatnagar is organizing the VII Annual Convention of SVBBI and International Symposium on **"Multiomics to One Health: Challenges and Way Forward in Biomedical Research"** from 14th – 15th Dec 2023 under the aegis of Society of Veterinary Biochemists and Biotechnologists of India (SVBBI) on the Occasion of Golden Jubilee of the Division of Biochemistry.

Biochemistry and Biotechnology are the two disciplines which have made a major impact on animal health, production and livestock product technology all over the world. These disciplines are the essence of basic and fundamental research and innovation that has given a thrust for new innovations in tackling the issues in control of diseases and for selection of desired traits for enhancing production & productivity of livestock & poultry.

The conference will focus on new and innovative research areas in biomedical research, from traditional to high-end research, including genomics, proteomics, metabolomics, gene editing, nanotechnology-based biosensing, computational biology etc.

I am happy to note that in this conference academicians, biomedical researchers, scientists, professionals, industry partners and entrepreneurs will share a common platform for scientific discussions, sharing knowledge and interactions in order to better understand and integrate multi-omics data with conventional biomedical research for improvement in animal health and productivity. I am sure that the recommendations emerging from the serious deliberations will have far reaching implications in designing future research programmes in the country.

I wish this symposium a grand success.

(Ashok Kumar Tiwari)

SOCIETY OF VETERINARY BIOCHEMISTS AND BIOTECHNOLOGISTS OF INDIA (SVBBI)

Regd. No.22912/122

Dr. Bimal Prasanna Mohanty ARS, FNAAS

Asst. Director General, ICAR & President SVBBI
Krishi Anusandhan Bhawan II
Indian Council of Agricultural Research
Pusa, New Delhi 110012.
Tel. 011-25842178; M. 9230618153
E-mail: bimal.mohanty@icar.gov.in bimalmohanty12@gmail.com

Message

I am delighted that the Society of Veterinary Biochemists and Biotechnologists of India (SVBBI) is holding its **VII Annual Convention and International Symposium on 'Multiomics to One Health: Challenges and Way Forward in Biomedical Research'** on December 14-15, 2023 at the ICAR - Indian Veterinary Research Institute (IVRI), Izatnagar, Bareilly. The Annual Convention and International Symposium is being organized jointly by Division of Biochemistry, ICAR - IVRI and SVBBI. This also coincides with commemoration of Golden Jubilee celebration of Division of Biochemistry, established in the year 1972 during the 5th Five Year Plan.

The International Symposium on "Multiomics to One Health: Challenges and Way Forward in Biomedical Research" covers topics on thematic areas of Animal health and production, Rational drug design, New generation vaccines and diagnostics and Nutritional interventions to food security. This symposium aims to bring together scientists, research scholars, academicians from Biochemistry, Biotechnology and allied-disciplines as well as industry partners and entrepreneurs for scientific discussions, and knowledge sharing for better understanding of omics technology applications and its integration with conventional biology and life sciences for improvement in animal health (and productivity), human health and ecosystem health under the framework of One Health. The scientific deliberations and interactions will comprise lead talks, invited lectures, research paper presentations, abstract presentations etc.

A **Compendium** is being brought out to document the scientific deliberations. I am sure this document would be amalgamation of current science, emerging science, new ideas and innovations for future scientific advancements.

I extend my Best Wishes to the participants and the organizers for the grand success of the Convention and International Symposium.

December 4, 2023

(Bimal Prasanna Mohanty)

SOCIETY OF VETERINARY BIOCHEMISTS AND BIOTECHNOLOGISTS OF INDIA (SVBBI)

Regd. No.22912/122

Dr. Subhasis Batabyal
Professor
Department of veterinary Biochemistry
West Bengal University of Animal and Fishery Sciences
Secretary (SVBBI)

Message

Dear Esteemed Colleagues,

On behalf of the Society of Veterinary Biochemists and Biotechnologists of India (SVBBI), I extend mywarmest greetings to all participants, speakers, organizers, and sponsors of the International Symposium on "Multiomics to One Health: Challenges and Way Forward in Biomedical Research".

As the Secretary of SVBBI, I am immensely proud to present this symposium, a testament to our society's unwavering dedication to promoting excellence in veterinary biochemistry and biotechnology. Founded in 1995, SVBBI has consistently championed the advancement of these fields, fostering a vibrant community of researchers and practitioners committed to improving animal health and welfare.

The theme of this symposium, "Multiomics to One Health: Challenges and Way Forward in BiomedicalResearch," reflects the growing recognition of multiomics as a powerful tool for addressing the complex challenges of One Health. Multiomics, the simultaneous analysis of multiple layers of biological information, holds immense promise for unraveling the intricate mechanisms underlying animal diseases, zoonotic infections, and environmental health issues.

This symposium brings together a distinguished panel of experts from around the globe to share their insights and expertise in this rapidly evolving field. Their presentations will cover a wide range of topics, from the latest advancements in multiomics technologies to their applications in One Health research.

I am confident that this symposium will provide a stimulating platform for knowledge exchange, fostering collaboration and inspiring new directions in multiomics research. The discussions that emerge will undoubtedly contribute to the development of innovative solutions that will improve animal health, protect human health, and safeguard our environment. On behalf of SVBBI, I extend my heartfelt gratitude to all who have contributed to the success of this symposium. Your dedication and commitment to scientific excellence are truly inspiring.

I wish you all a fruitful and enriching experience at this symposium. May the discussions and collaborations that emerge lead to significant breakthroughs in multiomics research and pave the way for a healthier future for all.

(S. Batabyal)

Dr Manish Mahawar
Principal Scientist
Organizing Secretary, SVBBI-2023
ICAR-IVRI, Izatnagar-243122 (U.P.)

Message

It is the matter of immense pleasure that the Division of Biochemistry, ICAR-IVRI in association with SVBBI is organizing "VII Annual Convention of SVBBI and International Symposium on Multiomics to One Health: Challenges and Way Forward in Biomedical Research" from 14th-15th December, 2023. I am glad to share that the convention is coinciding with the Golden Jubilee year of the Division of Biochemistry, ICAR-IVRI that we will be celebrating along with the meeting.

I am sure that the meeting will offer abundant opportunity to interact with the expert personnel researching in various areas of biochemistry and biotechnology. The interaction with international delegates and industry participants will allow exploring future collaboration for basic as well as translational research.

On behalf of organizing committee SVBBI-2023, Division of Biochemistry and ICAR-IVRI, I extend warm welcome to all delegates. I am confident that the organizing team shall fulfil the expectations of participants.

(Manish Mahawar)

About Society of Veterinary Biochemists and Biotechnologists of India (SVBBI)

SOCIETY OF VETERINARY BIOCHEMISTS AND BIOTECHNOLOGISTS OF INDIA (SVBBI)

Regd. No.22912/122

About the Society

Visionary teachers and scientists in the field of Veterinary and Animal Biochemistry of the nation, including Late Prof. LN Singh, Late Prof. RK Srivastav, Prof. RL Prasad, Prof. Ashok Kumar, Prof. PC Bisoi and many more dreamt of a platform for overall development of the subject, Veterinary Biochemistry and Biotechnology. They convened the 1st formal meeting at IVRI in the year 1995 and decided to form a society named as "*Society of Veterinary Biochemists and Biotechnologists of India*" (SVBBI). The official registration process of the society was assigned to Prof. PC Behera, Dept. of Biochemistry, Faculty of Veterinary Science and Animal Husbandry, OUAT, Bhubaneswar, Odisha who with the help of other active and enthusiastic members, registered the society vide Regd. No.22912 / 22 of 2008 under Society Registration Act, 1960. Prof. RL Prasad was nominated as President, Prof. S Batabyal, Vice President, Prof. PC Bisoi, Secretary, Dr JK Ray, Joint Secretary and Prof. PC Behera, Treasurer as the Office Bearers of the society.

As per the mandate of SVBBI, the academic and research activities in the subject of Veterinary Biochemistry and Biotechnology have been updated and expanded through organization of National Conferences and Symposia for mutual sharing of advanced knowledge and Annual Conventions for formulating the future plan of actions. In this context, Prof. PC Behera, as Organizing Secretary, hosted the 1st Annual Convention and National Symposium at Bhubaneswar during 11-12 March, 2016. Thereafter, the Annual Conventions and National Symposia were organized in different states across the nation.

1. 1st, Bhubaneswar, 2016
2. 2nd, Bengaluru, 2017
3. 3rd, Hissar, 2018
4. 4th, Tirupati, 2019
5. 5th Mathura (E-conference), 2020
6. 6th, Jabalpur, 2022

On the request of the Director, IVRI, the 7th (VII) Annual Convention and International Symposium on 'Multiomics to One Health: Challenges and Way Forward in Biomedical Research' is being organized at IVRI, Izatnagar to coincide with the commemoration of the Golden Jubilee Year Celebration of Division of Biochemistry, IVRI, which was established in the year 1972.

In the Annual Conventions, the SVBBI has felicitated the retired Professors and Scientists in recognition of their significant contributions in the field of teaching and research. Further, the young members are being encouraged for teaching and research excellence by conferring the "Teacher of the Year" and the "Scientist of the Year" Award, since 2021.

The journey ahead for the SVBBI is filled with possibilities, and its success lies in achieving the society's mission and in safeguarding the professional well-being of its members. Although hosting conventions, symposia, conferences, and felicitations for retiring members and such other activities have been conducted in regular manner, many other features planned, like launching the society's website, publishing a journal, and honouring members with more awards, fellowships, and recognitions, are still pending to be implemented. The society aims to confer the Fellowship of SVBBI from the next year onwards.

Office Bearers

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Dr. B. P. Mohanty (President)

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Dr. S. V. Perumal (Member)

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SVBBI-2023 Organizers

Dr Triveni Dutt
Director-cum-Vice Chancellor
ICAR-Indian Veterinary Research Institute
Izatnagar-243122 (U.P.), India

Biography

Dr Triveni Dutt, Director ICAR-Indian Veterinary Research Institute (Deemed University), izatnagar is having 30 years of experience in research and extension including 23 years in teaching and more than 10 years in RMP in the capacity of Joint Director (Academic) and Joint Director (Extension Education) at ICAR-IVRI, Izatnagar. So far, he contributed significantly in 34 research projects including 11 external funded and 7 inter-institutional projects. Most significant achievements include development of an outstanding germplasm of synthetics crossbreed cattle strain ***"Vrindavani",*** faster growing crossbred pig variety - **"Landlly",** registration of local goat as ***"Rohilkhandi Goat"*** and local pig as ***"Ghurrah"*** and standardization of cost effective super ovulation protocols. He has developed **5 IP protected devices** for effective livestock management. Received 34 Copyrights of various ICT Tools besides published **222 research articles** in National (180) and International (42) journals, 22 books, 8 training manuals and 2 Policy papers.

He has been the recipient of the ICAR- Dr. Rajendra Prasad Puraskar for Technical Books in Hindi in Agricultural and Allied Sciences 2020; ICAR-Ganesh Shankar Vidyarthi Hindi Krishi Patrika Award for bi-annual magazine "Pashu Chikitsa Vigyan" , 2010; Best Krishi Vigyan Kendra Award (Zone IV), 2010; Appreciation award-2009 by Indian Society of Extension Education, New Delhi; Life Time Achievement Award-2017 by Society of Veterinary and Animal Husbandry Extension, Ludhiana.

He is Fellow of the National Academy of Agricultural Sciences and Fellow of Bioved Research Institute of Agriculture and technology, Allahabad (UP), India

Dr Sanjay Kumar Singh
Joint Director (Research)
ICAR-Indian Veterinary Research Institute
Izatnagar-243122 (U.P.), India

Biography

Dr Sanjay Kumar Singh, Principal Scientist & Joint Director (Research), ICAR-IVRI, Izatnagar has > 22 years of experience of research, teaching & training with expertise on Assisted Reproductive Technologies (ART), Primary Cell culture of utero-ovarian origin, Molecular Reproduction, Diagnostics for improving fertility and Management of infertility and obstetrical problems in livestock for enhancement of reproductive efficiency besides experience on IPR, Technology Transfer and Agri-business Incubation. He has obtained his graduation degree in Veterinary Science & Animal Husbandry from G.B.P.U.A.T, Pantnagar and subsequently Master and Doctoral degree in Veterinary Gynaecology & Reproduction from Deemed University, ICAR-IVRI, Izatnagar. After PhD, he joined Agricultural Research Services as Scientist in February, 2000 at ICAR-IVRI, Mukteswar. He joined main campus again on 11th May, 2005 and became Principal Scientist in 2015. He has handled a total of fourteen external funded projects (3 as PI and 11 as Co-PI) and eleven institute funded (3 as PI and 8 as Co-PI) & developed number of technologies which include Development of Goatpox & PPR vaccines; MAP based sELISA & Lateral Flow Assay for pregnancy detection in cows, 3D- bubaline endometrial cell culture system; 2D-culture of endometrial & ovarian cells in bovine, Formulated mineral & feed supplement for Cattle & buffalo, Cloned & characterized genes involved in pregnancy/cyclicity/ uterine infections in livestock, Tested phyto-principles, Insulin, Kisspeptin & devised many anti-luteolytic strategies to enhance fertility vis-à-vis infertility management etc. He has visited world class Laboratory of Prof. Richard Ivell & Ravinder Anand Ivell at University of Nottingham, UK under DBT-CREST Award for one-year research (2014-15) on the frontier area of Animal Biotechnology. Filed five patents (A live attenuated vero-cell based goat pox vaccine for protection against goat pox; Formulation of a mineral supplement for cattle and buffaloes reared under Wheat- Rice-Sugarcane farming system and two patents on synthesis of MAPs against the protein specific to pregnancy and their use in EPD) and awarded one patent on Goatpox vaccine. Associated as inventor/ team member for the Goatpox and PPR vaccine technology commercialization to many industrial houses and realized huge revenue to the institute (> 4 crores through license fee and/ or royalty). Also filed five designs related to reproduction and four of them have already registered at the Indian Patent Office of Design in addition to six copyrights on critical care of diseases, technology services app, AI etc. Received many awards/ fellowships & recognitions viz., DBT-CREST Award, ICAR Team research Award, ISVIB-Fellow, Nils Lagerlof Award, Young Scientist, Dr. S.N. Luktuke, CSIR-SRF, ICAR-JRF, SRF. Published >96 research papers (International-52 and National-43 journals), written many book chapters (11), published number of popular articles and leaflets and folders (>20), review (3), manuals (2), compendium (5) and guided masters (6) and PhD students (7) in addition to Member, Advisory committee of many students (>20). He is also postgraduate diploma on Technology Management in Agriculture (PGDTMA).

Dr S. K. Mendiratta
Joint Director (Academic)
ICAR-Indian Veterinary Research Institute
Izatnagar-243122 (U.P.), India

Biography

Dr S. K. Mendiratta, born on 8th Feb. 1965, joined ICAR - Indian Veterinary Research institute, Izatnagar (UP) as Joint Director (Academic) on 16th November, 2012. He has obtained his B.V.Sc. & A. H., M.V.Sc and Ph.D. degrees (in the discipline of Animal Products Technology) from Haryana Agricultural University Hisar (Haryana). He was awarded University Gold Medal for his M.V.Sc thesis and Senior Research Fellowship by CSIR for Ph. D. degree programme. After qualifying Agricultural Research Services, Dr Mendiratta joined as a Scientist (LPT) in ICAR-IVRI on 15th June 1992. Dr. Mendiratta has made outstanding contributions in research, teaching, extension and institute building activities during his illustrated career of more than 31 years. During his tenure as Incharge of Dairy Technology Section (> 6 year), Head of LPT Division, IVRI (>10 years), Scientific Coordinator of IVRI Deemed University (>2 years) and Dean College of Dairy Science and Technology, GADVASU Ludhiana, he has brought significant improvements in the outputs and received many recognitions. He has successfully completed 25 Institute and 6 externally funded research projects, generated more than 30 technologies, got three patents, and transferred 10 technologies to prospective entrepreneurs. He has guided 16 Ph.D. and 12 M.V.Sc. students in the capacity of Major Adviser and organized more than 25 training programmes as a co-ordinator. Dr Mendiratta has published over 240 research papers in reputed journals and written more than 45 reviews and technical articles, 50 invited lectures, 7 book chapters and edited and authored 6 books and manuals. Dr Mendiratta was awarded FAO and INSA fellowships for advanced trainings in USA and GERMANY in the years 2000 and 2005, respectively. He is a recipient of Best Teacher Award of IVRI Deemed University, ICAR Dr Rajendra Prasad Award for writing technical book in Hindi, Fellow NAVS and Fellow IMSA. He has received many recognitions and important assignments from different central and state agencies. These include Chairman/members of different Scientific panels of FSSAI, Member Expert Panels for MoFPI, SERB/DST and APEDA, President IMSA, Member BSMA, Member Institute Research Council for CIPHET, CARI and DPR, Member Institute Management Committee for NRC meat, Member Research Advisory Committee for NRC Pig, Member QRT for NRC Mithun and Member BOM for IVRI Izatnagar.

Dr Smt. Rupasi Tiwari
Joint Director (Extension Education)
ICAR-Indian Veterinary Research Institute
Izatnagar-243122 (U.P.), India

Biography

Dr (Mrs) Rupasi Tiwari, PhD in Extension Education is currently working as Joint Director, Extension Education at the ICAR-Indian Veterinary Research Institute, Izatnagar. She has more than 25 years of experience of research, teaching & extension and has made significant contribution towards the development of various e-learning systems and extension methodologies for effective technology transfer to the livestock owners and other stakeholders. She has handled 25 research projects, leading to the development of 20 multilingual Mobile Apps & 11 need based information/ expert systems apart from 30 educational videos for effective technology transfer & entrepreneurship generation. These systems have spread across 25 states of India and across > 100 countries in the world. Further, she has also organized large number of extension activities for various stakeholders including training, gosthies, interface meets, Farm School on AIR, Kisan Mela, Exhibitions etc & facilitated in development of startups. She has guided 24 PG students, taught 25 courses & published 175 research papers in national & international journals along with 213 other publications including books, training manuals/ e-manuals/ monographs, farm publications & delivered >100 lectures at various forums.

She is also the recipient of 6 ICAR Awards : ICAR-Bharat Ratna Dr. C. Subramaniam award for Outstanding teachers, ICAR-Swami Sahajanand Saraswati Extension Scientist Award, ICAR-Dr Rajendra Prasad Puruskar 2020 & 2021, ICAR-Ganesh Shankar Vidyarthi Hindi Krishi Patrika Puruskar 2010 & 2021, besides 27 various fellow awards and recognitions & 44 best paper awards from institutions/ professional societies for her outstanding work in research, teaching and extension.

Prof. (Dr.) Raghvendar Singh
Principal Scientist & Head
Convener, SVBBI-2023
ICAR-IVRI, Izatnagar-243122 (U.P.), India

Biography

Prof. Raghvendar Singh is Principal Scientist and Head, Division of Biochemisry and has been former Director (A) ICAR- Central Sheep and Wool Research Institute, Avikanagar, Rajasthan. Prof. Singh did his M.Sc. and Ph.D. in Biochemistry in the year 1987 and 1992 from GBPUA&T, Pantnagar and ICAR-NDRI, Karnal, respectively. Prof. Singh Joined Agriculture Research Service (ARS) as Scientist (Animal Biochemistry) in 1993 and was posted at ICAR-NRC on Camel, Bikaner and served as scientist to principal scientist in different capacities. In the year 2007, he was selected as professor and served Head, Department of Immunology and Defense Mechanism, Department of Biotechnology and Coordinator of Bioinformatics Infrastructure Facility (BIF) at Sardar Vallabhbhai Patel University of Agriculture and Technology (SVPUA&T), Modipurum, Meerut, Uttar Pradesh.

Prof. Singh is having 28 years of research and teaching experience in Animal Biochemistry, Biotechnology, Immunology. He is recipient of SAAR and NESA Fellows, Vigyan Pradeep Samman and Rajasthan State Appreciation certificate and awards. He is a life member of more than six Professional Bodies and presented more than 50 invited papers and scientific presentations in national and international conferences. Dr. Singh has more than 150 publications in international and national peer reviewed journals, authored many books, book chapters, technical bulletins and manuals. He has guided more than 15 doctoral and 20 master students. He has been invited as experts in various seminars, radio talks and served as reviewer of various peer-reviewed journals and he is member of academic council/boards of educational institutions, editorial board and chief editor of scientific annual reports, vision, newsletters and magazines.

Dr Mohini Saini
Principal Scientist
Chairperson SVBBI-2023
ICAR-IVRI, Izatnagar-243122 (U.P.), India

Biography

Dr Mohini Saini obtained her BSc (Honours School) Biochemistry degree in 1984 and MSc (Honours School) Biochemistry degree from Punjab University, Chandigarh in 1986. She started her career as Junior Biochemist in Dayanand Medical College & Hospital, Ludhiana, Punjab in 1986. From 1987 onwards she served as Research fellow in Ministry of Environment and Council of Scientific & Industrial Research projects on Biochemical toxicology and while working in these projects completed her Ph.D. from Panjab University, Chandigarh in 1992. She joined as ARS Scientist in 1993 and got posted in the Division of Biochemistry where she is serving as Principal Scientist till date. In between, she was placed in Centre for Wild Life during 2004 to 2014. She acted as Head, division of Biochemistry from June 2019 to Sept 2023.

She has more than 30 years of experience in teaching Biochemistry courses to PG students and conducting basic research related to animal health and production and have guided 10 MVSc and 12 PhD students for their thesis research as Major advisor and more than 100 students as Minor advisor. Her areas of research include Biochemical toxicology, Host-pathogen interactions, Biochemical basis of cancer, Molecular Biochemistry, Immunomodulation, Wildlife forensics, Gyps Vulture conservation etc. Her notable contribution included her research on Gyps vultures for which she received appreciation from Royal Society for Protection of Birds, UK. She has contributed 69 Nucleotide sequences and two whole genome sequences of animal viruses in GenBank/EMBL. She has one patent and published 150 research articles with 2358 citations, h-index 26 and i-10 index 57. She has been recognised as peer reviewer for research papers in National and International Journals & for theses from various Universities. She is also recognised as subject Matter Expert in various Selection and Promotion panels in ICAR and other Universities. She is Life Member for Society of Toxicology, India, Society of Immunology & Immunopathology India and a Member of Technical Advisory Committee of International Consortium “Saving Asia's Vultures from Extinction”.

Dr Praveen K. Gupta
Principal Scientist
Co-Chairperson, SVBBI-2023
ICAR-IVRI, Izatnagar-243122 (U.P.), India

Biography

Dr Praveen K Gupta received his veterinary graduate degree from G.B. Pant University of Agriculture & Technology, Pantnagar in the year 1988. He received his Masters and Doctorate degrees in Biotechnology in the year 1991 and 1998, respectively. In 1993, he joined Indian Council of Agricultural Research (ICAR) as research scientist and since then he is serving at ICAR-Indian Veterinary Research Institute, Izatnagar in the Division of Veterinary Biotechnology, Izatnagar.

He visited Institute for Animal Health, UK during 1999 under Indo-DFID, UK Transfer of Molecular Biology Techniques (TomBiT) programme and during 2008 as Commonwealth Academic Staff Fellow. He was visiting scientist during 2013 under Endeavour Research Fellowship at Queensland Alliance for Agriculture and Food Innovation (QAAFI), the University of Queensland, Australia. As faculty member of Animal Biotechnology, he has supervised more than 20 Masters and Doctoral students and has been conferred with Best Teacher Award by the Deemed University, ICAR-IVRI. He has authored more than 100 research articles in different peer-review high impact journals.

His areas of research interest are new generation vaccines and diagnostics for animal viral diseases. His current research interests include, marker vaccine development for infectious bovine rhinotracheitis (IBR) and classical swine fever (CSF). He received extramural grants for his research projects from different funding agencies including, DBT and NASF. He has been awarded one Indian Patent on IBR marker vaccine and the technology is with AgriInnovate India for commercialization.

Dr Praveen Singh
Principal Scientist
Co-Chairperson SVBBI-2023
ICAR-IVRI, Izatnagar-243122 (U.P.), India

Biography

Dr Praveen Singh did his master M.Sc. (Physics) with specialization in Electronics from University of Allahabad and obtained 2nd Rank in University in year 1988. He successfully cleared the Junior Research Fellowship (JRF) from Council of Scientific and Industrial Research in year 1992 and also passed the Graduate Aptitude Test in Engineering (GATE 1991). He joined his PhD (Physics) programme at Indian Institute of Technology Delhi, New Delhi in year 1992 under the supervision of Professor R K Puri and Professor T C Goel. He worked on magnetic materials embedded in epoxy resin for their application as coating on various surfaces including on fighter plane to absorb the radar signals enabling them to become invisible on screen. The PhD work resulted into highly cited publications leading to award of Japan Society of Promotion of Science (JSPS award) of value US $100000/- as collaborator for working at Professor Kiyoshi Toko's Lab in Kyushu University, Fukuoka, Japan for two years. Dr Singh got selected as Scientist in Indian Council of Agricultural Research (ICAR) through ARS 1994 and joined the service in 1996. He served initially at Central Arid Zone Research Institute Jaisalmer from 1996-1999. The thermal time requirements of date palm, pomegranate and ber were estimated in Thar Desert region of India. On transfer to Indian Veterinary Research Institute Izatnagar, Bareilly, he was placed in Biophysics and Electron Microscopy Section in year 1999. He worked on magnetic stimulation of fracture in very first project at IVRI. Later went on JSPS-Postdoctoral assignment to Japan from 2005-07, where he learnt the science and art of Biosensor, which he later propagated in IVRI Izatnagar through series of high value projects, National, International and Institutional. His significant contribution lies in development of Lateral flow devices for pathogens, Host pathogen interaction in relation to PPRV-SLAM homologous peptides for antivirals effects, Neuraminidase inhibitors for PPRV. He developed the biosensor surfaces termed as electronic nose for detection of explosive substances, TNT and protein antigen for animal diseases, Lateral flow assays for detection of Aglactia, Ciprofloxacin, PPR virus, Brucella, Babesia gibsoni, Japanese encephalitis, Mycobacterium avium subspecies paratuberculosis etc. Recently, he obtained a massive grant from DBT of 700 lakh for developing Integrated Magneto-Acousto-Dielectrophoresis based device for sorting of Bovine spermatozoa. He has more than 100 publications to his account and a citation base of approximately 2000. He is holding the post of principal scientist since March 2011 and Incharge, Central Instrumentation facility-Bioengineering at IVRI since September 01, 2015. Currently, he is also Chief Hostel Warden (Boys) of ICAR-IVRI Izatnagar.

Dr Manish Mahawar
Principal Scientist
Organizing Secretary, SVBBI-2023
ICAR-IVRI, Izatnagar-243122 (U.P.), India

Biography

Manish Mahawar completed his B.V.Sc & A.H. from CVAS Bikaner, Rajasthan in 1999. Subsequently, he earned MVSc and PhD from CVSc & AH Mathura and ICAR-IVRI Izatnagar, UP respectively. He then took couple of postdoctoral trainings at Albany Medical College, Albany NY and University of Georgia, Athens, GA, USA. He joined Division of Biochemistry, ICAR-IVRI as a senior scientist back in 2011 and subsequently promoted to Prinicipal Scientist in 2017. He has been recipient of Endeavour fellowship, Dept of Education and training, Australian Govt. and spent six months at the University of Queensland during 2016.

His research interest includes deciphering the roles of bacterial antioxidants including protein repair enzymes in stress survival and virulence. He has authored more than 30 papers including in JBC, Journal of Bacteriology, BBA and Scientific Reports. His research is supported by grants funded by DBT and NASF.

He has supervised/supervising more than 15 masters and PhD students and currently supervising an early career fellowship funded by DBT-Wellcome trust India Alliance.

Dr S.K. Bhure
Principal Scientist
Co-Organizing Secretary, SVBBI-2023
ICAR-IVRI, Bengaluru, Karnataka, India

Biography

Dr Bhure has worked for the last 19 years in the field of genetic characterization of camel breeds, molecular disease diagnosis, molecular cryoinjury to buffalo spermatozoa, and novel approaches for improvement of FMD vaccine. As a Principal Investigator, he has completed six research projects and had been collaborator in more than 10 research projects. Currently working on the combinatorial effect of peptide epitopes and inactivated Foot-and-Mouth Disease virus (FMD) on the duration of immunity; and also on long-term storage of inactivated FMD virus. The other research interests include the immunological memory against FMD virus, and host-pathogen interactions. He had published 63 research articles in different journals of national and international repute. Further, he had edited a coffee table book of IVRI, "My Journey since 1889" and had been editorial member of ICAR-IVRI Annual Reports. Dr Bhure is a faculty in the Division of Biochemistry, ICAR-IVRI, and is associated with post-graduate teaching and research. He has guided six MVSc and five PhD students for their research work.

Dr Ajay Kumar
Senior Scientist
Co-Organizing Secretary & Treasurer, SVBBI-2023
ICAR-IVRI, Izatnagar-243122 (U.P.) , India

Biography

Dr Ajay Kumar has graduated from College of Veterinary Science and Animal Husbandry, OUAT, Bhubaneswar, Odisha in the year 2003. He earned his Master in 2005 and PhD in 2009 from Division of Biochemistry, Indian Veterinary Research Institute, Izatnagar, Bareilly. He has worked on developing serodiagnostic for BHV-1 infection based on gE recombinant protein under DBT funded project. He also worked on key gene in BHV-1 responsible for development of latency. Based on his findings, he was awarded ICMR-Long Term International Fellowship for a period of one year in the year 2019 and joined College of Veterinary Medicine, Auburn University, Auburn, Alabama, USA in 2020. He worked there on HSV-1 and developed HSV-1 mutant by deletion of a gene responsible for virus latency. Later he also worked on Salmonella Typhimurium under DBT and DST funded projects wherein he developed mutant bacteria by deletion of couple of genes in bacterial metabolic pathway responsible for its colonization in mice and poultry. He has more than 20 research publications in peer reviewed journals from his research works and guided 5 MVSc and 4 PhD students and currently he is supervising 2 PhD students.

Dr Karuna Irungbam
Scientist
Co-Organizing Secretary, SVBBI-2023
ICAR-IVRI, Izatnagar-243122 (U.P.), India

Biography

Karuna Irungbam earned her Bachelor's degree in Veterinary Science and Animal Husbandry from CVS & AH, CAU, Selesih, Mizoram in 2007. And later obtained her MVSc degree in Veterinary Biochemistry from the College of Veterinary Sciences, GADVASU in 2009. Following her master's degree, she served as a teaching assistant in the department of Biochemistry at Veterinary College, KVAFSU, Bidar for more than a year. In 2011, she joined ARS services and was posted in the Division of Biochemistry, ICAR-IVRI Izatnagar, Bareilly, U.P. She did her PhD as in service candidate and obtained doctoral degree from Justus Liebig University Giessen, Germany with DAAD scholarship in 2020. She has gained experience in the area of lipidomics, metabolic alteration in NAFLD and cancer. Her research focus on understanding the role of altered lipid droplet-associated proteins, particularly perilipins, in disease processes. She has been awarded with DBT-Wellcome trust India Alliance Early Career fellowship funding in 2022 for period of 5 years. She has published 15 articles in international reputes journals such as Heptology, Cellular & Molecular Gastroenterology and Hepatology (CMGH), Laboratory investigation, and Cells. Being engaged in both research and education, Karuna has guided one MVSc student and is currently mentoring two MVSc students while actively teaching Veterinary Biochemistry to undergraduate and postgraduate students.

Dr Amir Kumar Samal
Scientist
Co-Organizing Secretary, SVBBI-2023
ICAR-IVRI, Mukteshwar, Uttrakhand, India

Biography

Amir Kumar Samal is presently a scientist in ICAR- Indian Veterinary Research Institute, Mukteswar Campus. He graduated in BVSc and AH from College of Veterinary Science, OUAT Bhubaneswar in 2005 and worked as a Veterinary Doctor in Govt. of Odisha from 2005 to 2012. He obtained his Masters in Animal Biochemistry from ICAR- Indian Veterinary Research Institute, Bareilly in 2011. He joined the ICAR in 2013 and worked as a scientist in ICAR-Central Marine Fisheries Research Institute in the field of marine aquaculture and chlorophyll based biodynamic forecasting of Indian marine fisheries resources. In 2018, he joined ICAR-IVRI and is working on biochemical aspects of host-parasite interaction. He is pursuing his Doctoral studies from ICAR- NDRI Karnal.

Dr Mukesh Kumar
Scientist
Co-Organizing Secretary, SVBBI-2023
ICAR-IVRI, Izatnagar-243122 (U.P.), India

Biography

Dr. Mukesh Kumar obtained his master's degree in Veterinary Biochemistry from College of Veterinary Science and Animal Husbandry, Anjora-Durg in 2011. He joined ICAR-IVRI in 2013 and started working on cancer biology. After working as a collaborator on a project investigating canine Matrix metalloproteinase (MMPs) as a potential xenogenic vaccine candidate to treat mouse mammary tumor, he continued as an independent researcher investigating the role of cholesterol and P2RX7 in cancer cell survival and metastasis. Subsequently, in 2018, he received the coveted Netaji Subhash ICAR International Fellowship and enrolled as a PhD student at the University of Copenhagen, joining the Danish Cancer Research Center (DCRC) as a guest scientist. Under the supervision of Associate Professor Dr. Elena Papleo, he pursued his PhD, "From -omics to structure: an investigation of the autophagy-core machinery in cancer." He explored the integrated approaches of structural bioinformatics and omics during his PhD to understand the core autophagy machinery in cancer. Currently, he is applying both in silico and in vitro based strategies to understand the molecular determinants of cholesterol-P2RX7 interaction and its role in pore dilation in order to utilize them as better therapeutic targets against cancer cells using small molecular inhibitors.

Dr Chandra Mohan S.
Scientist
Co-Organizing Secretary, SVBBI-2023
ICAR-IVRI, Bengaluru, Karnataka, India

Biography

Dr Chandra Mohan S, holds PhD in Animal Biochemistry and currently working as scientist at Foot-and-Mouth disease research laboratory, IVRI, Bengaluru. Has multidisciplinary exposure with optimum working knowledge in biochemical and molecular biological techniques. So far worked under six research projects as PI or Co-PI. Has authored 22 research articles with 14 in International and 8 in National peer reviewed journals. Has presented 33 abstracts in various National and international symposia or conferences. He has also imparted training to both national and international participants in the field of molecular biology techniques. Apart from research, he is actively involved in teaching post-graduate students.

Dr Meeta Saxena
ACTO
Treasurer, SVBBI-2023
ICAR-IVRI, Izatnagar-243122 (U.P.), India

Biography

Meeta Saxena completed her Master degree in Animal Biotechnology from Indian Veterinary Research Institute, Izatnagar in 1991 and joined ICAR Technical Services in 1997 at IVRI Izatnagar in Department of Biotechnology. She had a good hand in handling cell lines and primary culture there and other ongoing lab techniques. She later obtained her PhD in Animal Science from Rohilkhand University as inservice candidate in 2005, and joined Flow Cytomerty facility in Department of Biochemistry. She has experience in field of flow cytometry and molecular biology techniques. She is associated with lab project work and supports other researchers and students in their lab activities. She has been engaged in different workshops and training programme which were conducted in the department and institutes. She holds responsibilty in demonstrating practical classes of undergraduate and postgraduate students and taking master course.

Golden Jubilee Lecturers

GOLDEN JUBILEE ORATION

Dr Vineet K. Singh
Professor of Microbiology
Kirksville College of Osteopathic Medicine
A.T. Still University of Health Sciences
Kirksville, MO 63501, USA
*Correspondence: vsingh@atsu.edu

Dr Vineet K. Singh completed his BSc (Chemistry) in 1987 and MSc (Biochemistry) in 1989 at Banaras Hindu University, followed by a PhD (Biochemistry) in 1993 from the Indian Veterinary Research Institute (IVRI). He conducted his doctoral research under the supervision of Dr. T. More and studied cationic antibacterial proteins of buffalo polymorphonuclear cells. In 1993, he joined the Indian Council of Agricultural Research and served as a Scientist in the Division of Biochemistry and Food Sciences at IVRI until 1997. He pursued a postdoctoral research opportunity at Illinois State University, focusing on antibiotic resistance mechanisms in *Staphylococcus aureus* from 1997 to 2002. Subsequently, he briefly engaged in postdoctoral research work involving *Mycobacterium tuberculosis* before joining the Kirksville College of Osteopathic Medicine (KCOM), part of the A.T. Still University of Health Sciences (ATSU), as an assistant professor in 2004. Since 2012, Dr Singh has held the position of Professor of Microbiology at KCOM. Alongside teaching Microbiology and Infectious Diseases to ATSU medical and dental students, Singh remains committed to advancing his research on understanding stress tolerance mechanisms in the bacterium *S. aureus*.

Physiological and clinical significance of membrane branched chain fatty acids in the bacterium *Staphylococcus aureus*

Vineet K. Singh
Kirksville College of Osteopathic Medicine, A.T. Still University of Health Sciences, Kirksville, MO 63501, USA

Staphylococcus aureus is a Gram-positive bacterium and a major human pathogen with the ability to cause a range of mild superficial skin infections to serious deep tissue infections. Its ability to withstand environmental fluctuations and quickly develop resistance to antibiotics assists its persistence and limits infection control options. Adaptation to lower temperatures is a key determinant for the survival of *S. aureus* outside of the human host. Cytoplasmic membranes are important barriers between bacterialcells and the environment. It is essential for thebacteria to keep their membrane fluid, particularly at low temperatures, when it is likely to freeze, for the proper functioning of various membrane-associatedprocesses. *S. aureus* has a complex membrane fatty acid compositioncomprisedof straight-chain saturated fatty acids, unsaturatedfatty acids, and branched-chain fatty acids (BCFAs). BCFAs that account for about 55 to 65% of the total fattyacids, impose steric hindrance and interfere with the tight packing of the membrane fatty acids, and thusare the major determinants ofmembrane fluidity in *S. aureus*.

Branched-chain a-keto acid dehydrogenase (BKD) is a multimeric enzyme consisting of four polypeptides that catalyzes the early stages of BCFA synthesis. Our studies have shown that a BKD-deficient *S. aureus* has a significantly reduced levels of BCFAs (35% in the BKD-deficient strain compared to 63% in the wild-type bacterium) in its cytoplasmic membrane. We initially postulated that the pyruvate dehydrogenase (PDH) and/or thea-keto glutarate dehydrogenase (KGDH) enzymes were probably responsible for the residual membrane BCFAs in the BKD-deficient *S. aureus* due to the similarity of the individual components of these multimeric enzymes. Our studies, however, have shown that the BCFA content of a KGDH-deficient *S. aureus*is comparable to the BCFA content of the wild-type *S. aureus* suggesting that KGDH plays no role in BCFA synthesis. On the other hand, the BCFA content of a PDH-deficient *S. aureus* strain is much higher (80%) compared to the BCFA content of the wild-type *S. aureus*. Our subsequent studies have provided further insights that the media components contribute to residual BCFAs as BKD-deficient strains of *S. aureus* fail to grow in defined media with no known products of BKD catalyzed reactions. These findings further emphasize that the BCFAs are essential for the survival of *S. aureus*. Additionally, growth of the BKD-deficient mutant (with reduced membrane BCFA content) is progressively more impaired than that of wild-type *S. aureus* with decreasing temperature, to the

point that the mutant fails to grow at 12°C. Growth of the BKD-deficient mutant is markedly stimulated by 2-methylbutyrate, that is an intermediate past BKD catalyzed reaction in the BCFA biosynthetic pathway. BKD-deficiency leads to reduced adherence of *S. aureus* to the human lung epithelial cells, its decreased survival in mice, and a relatively less fluid membrane. On the other hand, lack of a functional BKD enzyme increases staphyloxanthin production and susceptibility of *S. aureus* to alkaline and oxidative stress conditions. Our studies have further shown that the BCFA levels have a role in the activity of two-component regulatory systems such as SaeR/S and overall toxin production in the bacterium*S. aureus*. More recently, we observed that a highly daptomycin-resistant *S. aureus* strain produces a significantly higher levels of BCFA (~78%) in its membranesuggesting that the BCFA levels may impact the functions of the membrane targeting antibiotics. Our findings thus suggest that BCFAs andthus the BKD enzymeare critical for survival and virulence and can be an attractive target to manage *S. aureus* clinical infections.

GOLDEN JUBILEE ORATION

Dr Yogesh Saini
Professor of Immunology and Toxicology
Population Health and Pathobiology, NCSU College of Veterinary Medicine
Raleigh, NC, 27606, USA
Correspondence: ysaini@ncsu.edu

Dr Yogesh Saini received his DVM (1996-2001) from College of Veterinary Sciences, Palampur (India) and MVSc in Veterinary Biochemistry (2001-2003) from College of Veterinary Sciences, Pantnager (India). Thereafter, he completed a Senior Research Fellowship (2003-2005) at Indian Veterinary Research Institute, Izatnagar (India). He received a dual-PhD (Genetics and Toxicology) at Michigan State University in 2009. After completing postdoctoral training (2010-2014) at the University of North Carolina (UNC), he was promoted to the rank of research instructor at Marsico Lung Institute. He then joined LSU/SVM in 2014 as a tenure-track Assistant Professor. In 2019, he received early tenure with promotion to Associate Professor. Most recently in August 2023, he joined NCSU/CVM as full professor in the department of Population Health and Pathobiology.

Multi-omics approach to identify molecular pathways of environmental lung diseases

Yogesh Saini
Population Health and Pathobiology, NCSU College of Veterinary Medicine, Raleigh, NC, 27606, USA

Ozone causes inflammatory responses in the airspaces of experimental animals and humans. The resident cells of the respiratory tract (i.e., epithelial cells and macrophages) respond to inhaled ozone in a variety of ways that affect their survival, morphology, and functioning. However, a complete understanding of the transcriptomic and proteomic changes in response to ozone exposure is still limited. Through transcriptome profiling, we aimed to analyze gene expression alterations and associated enrichment of biological pathways in three distinct cell type-enriched compartments of ozone-exposed murine lungs. We subchronically exposed adult male and female mice to 0.8 ppm ozone or filtered air. RNA-Seq was performed on airway epithelium-enriched airways, parenchyma, and purified airspace macrophages. Differential gene expression and biological pathway analyses were performed and supported by cellular and immunohistochemical analyses. While a majority of differentially expressed genes (DEGs) in ozone-exposed versus air-exposed groups were common between both sexes, sex-specific DEGs were also identified in all of the three tissue compartments. As compared with ozone-exposed males, ozone-exposed females had significant alterations in gene expression in three compartments. Pathways relevant to cell division and DNA repair were enriched in the ozone-exposed airways, indicating ozone-induced airway injury and repair, which was further supported by immunohistochemical analyses. In addition to cell division and DNA repair pathways, inflammatory pathways were also enriched within the parenchyma, supporting contribution by both epithelial and immune cells. Further, immune response and cytokine-cytokine receptor interactions were enriched in macrophages, indicating ozone-induced macrophage activation. Finally, our analyses also revealed the overall upregulation of mucoinflammation- and mucous cell metaplasia-associated pathways following ozone exposure.

Epithelial lining fluid (ELF) in the respiratory tract harbors a variety of proteins that influence homeostatic and stress responses in the airspaces. Exosomes, nano-sized extracellular vesicles, contain a plethora of proteins that vary in abundance and composition based on the prevailing conditions. However, the exosomal protein signatures contained within the ELF from ozone-exposed lung airspaces remain poorly characterized. To explore this, we hypothesized that ozone triggers the release of exosome-bound inflammatory proteins from various cells that reflect mucoobstructive lung disease. Exosome-bound proteomic signatures, as well as the levels of soluble inflammatory mediators in the bronchoalveolar lavage fluid (BALF), were determined 12–16 h after the last exposure of sub-chronic exposure paradigm. Principal component analyses of the exosome-bound proteome revealed a clear distinction between air-exposed and ozone-exposed mice, as well as between ozone-exposed males and ozone-exposed females. In addition to 575 proteins that were enriched in both sexes upon ozone exposure, 243 and 326 proteins were enriched uniquely in ozone-exposed males and females, respectively. Ingenuity pathway analyses on enriched proteins between ozone- and air-exposed mice

revealed enrichment of pro-inflammatory pathways. More specifically, macrophage activation-related proteins were enriched in exosomes from ozone-exposed mice. Cytokine analyses on the BALF revealed elevated levels of G-CSF, KC, IP-10, IL-6, and IL-5 in ozone-exposed mice. Finally, the histopathological assessment revealed significantly enhanced intracellular localization of mucoinflammatory proteins including MUC5B and FIZZ1 in ozone-exposed mice in a cell-specific manner indicating the cellular sources of the proteins that are ferried in the exosomes upon ozone-induced lung injury. Collectively, through multi-omic approach, we have identified compartment-specific transcriptomic changes, protein signatures in airspaces, and integrated biological pathways relevant to the human and animal environmental lung diseases..

Technical Session - I

Concepts of multi-omic approaches in animal health and production

LEAD PAPER

Dr Ravi Kumar Gandham, V.P.P.S.
Principal Scientist & Head
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ICAR-National Bureau of Animal Genetic Resources
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Dr Ravi Kumar works in the area of Livestock Genomics. He was trained at School of Informatics, IUPUI, Indianapolis and USDA in the field of Computational Biology and Genomics. He is a combination of being a computational and experimental biologist. He has handled several externally funded projects. He worked on host pathogen interaction to delineate molecular pathogenesis using high throughput genomics and proteomics approaches and assembled several genomes of pathogens. His major contributions in the field of livestock genomics include world largest high density SNP chip – IndiGAu and Platinum genome assembly of Tharparkar and Sahiwal.

Interactome analysis to understand the systems biology behind the phenotype of a disease condition

Ravi Kumar Gandham, V.P.P.S.
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Karnal-132001 (Haryana), India

The change in biological function is reflected by dysregulation of the transcriptome that not only involves mRNA, but also other RNA types such as lncRNAs , miRNAs and circRNAs. The interplay among them influences RNA metabolism and in turn the biological and pathological conditions. Different types of strategies are being employed to profile the global change in different types of RNA. Considering the fact that rRNA accounts for 85% of total RNA level, under-representation of mRNA , lncRNAs , miRNAs and circRNAs is inevitable when whole transcriptome sequencing (total RNA sequencing) is carried out. To overcome this caveat, two strategies are followed - 1. Isolating the mRNA using oligodT probes attached to beads as all the mRNAs have a Poly (A) tail, and 2. Using a Ribozero Kit initially, to eliminate rRNA from the total RNA before sequencing. However, both the strategies have their own limitations. In the former, though all the differentially expressed mRNA could be identified, identification of differentially expressed lncRNAs and circular RNAs is limited in this strategy. This is because some of lncRNA would not have a polyA tail and circular RNA are not polyA modified.This makes the later strategy an ideal choice, though, the Ribozero kits available in the market are only 60 - 70 % efficient. This strategy has enabled researchers to identify differentially expressed mRNA, lncRNA and circular RNA using appropriate tools for analysis from whole transcriptome data. It is noteworthy that for identifying differentially expressed miRNA, the chemistry for generation of miRNA- seq data is different from the above-mentioned method. These RNA are now commonly termed as competitive endogenous RNA (ceRNA). Further, by integrating genomic and proteomic mapping approaches, biological hypotheses can be formulated with increasing levels of confidence.

Peste des petits ruminants virus (PPRV), a member of the genus Morbillivirus within the family Paramyxoviridae, causes a devastating disease that affects the lives of sheep and goat. The host immune response to PPRV involves secretion of interferons and cytokines by natural killer cells, macrophages, dendritic cells, B cells, T cells, etc,. However, virus evades or modulates the immune response by altering the expression of several immune-related genes/proteins. Long non-coding RNAs (lncRNAs), messenger RNAs (mRNAs), and microRNAs (miRNAs) are all part of the competitive endogenous RNA (ceRNA) network that govern the host response. Transcriptome analysis identified mRNAs, lncRNAs, and miRNAs that are differentially expressed under PPRV infection. Interactome analysis on the long non-coding RNA, messenger RNA, microRNA, and proteome of PPRV-infected goat peripheral blood mononuclear cells revealed dysregulation of immune processes. Transcriptome of spleen and lung revealed a greater enrichment of immune response processess than in PBMCs. This indicated an initial balance between viral and immune response in PBMCs which further tilted in favour of virus replication/load, which must have resulted in the death of animals.

INVITED PAPER

Dr Amit Kumar
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Division of Animal Genetics
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Dr Amit Kumar has contributed significantly in teaching and research about genome wide genomics data analysis and its application in the quantitative genetics. He successfully completed more than 08 Institute and extra mural project. At present he is contributing as principal Investigator in World Bank funded CAAST-ACLH, project of NAHEP and deputed 49 PhD students and 22 faculties for overseas international training. Also, he is principal investigator of Network project of ICAR in Bioinformatics (CABin Scheme). He successfully guided six Master's and eight PhD students, respectively as major advisor. He coordinated to establish two Experiential Learning Unit (ELU) projects at Animal Reproduction and Livestock Product Technology division of Institute for improving entrepreneurship skill of BVSc students. He is officer in charge of the Bioinformatics centre of institute wherein proving Bioinformatics analytical support to PG/PhD students of institute. He established a Big data analysis laboratory for computational genomics work at Institute under aegis of NAHEP project. He has been trained at IOWA state University, USA and ETH, Zurich, Switzerland in area of Genome Wide Association Study and RNA Seq data of livestock. Dr Amit Kumar has published more than 150 research papers in reputed peer reviewed international and national journals with 18 h-index (i10-43). Further, he has published two book and 12 technical bulletin/manual.

Concepts of multi-omics approaches in animal health and production

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Since the beginning of the 2000s, the evolution of sequencing technologies and bioinformatics methods provided facility for sequencing the expressed fraction of the genome (or transcriptome) making transcriptomic analysis early in the 2000s itself and also in sequencing/re-sequencing of complete genomes of farm animals. The construction of maps of genetic markers for cattle, pigs and chicken began in the 1990s. Due to the immense development in sequencing technology accompanied by evolving bioinformatics tools capable of handling big data have paved way for both small and large research groups to generate draft genome sequences of any organism of interest. In last two decades an astonishing development in livestock genomic have taken place. First livestock species whose genome sequenced was chicken, which was released in 2004 tailed by updated version in 2017 (Genome Reference Consortium) and genome sequence of Aseel breed from India also sequenced by PDP in 2014. The release of whole genome sequences of major livestock species like cattle Btau_3.1 draft assembly in 2006 improved draft assembly Btau_5.0.1 in 2015 (The Bovine Genome Sequencing and Analysis Consortium), Horse genome EquCab2 in 2007 improved version EquCab3 in 2018 (Kalbfleisch et al. 2018), Pig assembly Sscrofa9 in 2009 with latest Sscrofa11.1 in 2017 (Swine Genome Sequencing Consortium), Sheep assembly Oar_v1.0 in 2010 with recent version Oar_v4.0 in 2015 (The International Sheep Genomics), Water buffalo assembly Bbu_2.0-alpha in 2011 from India (Tantia et al. 2011), Yak genome assembly version 1.1 in 2012 (Hu et al. 2012), Draft sequence of camel in 2012 (The Bactrian Camels Genome Sequencing and Analysis Consortium) and Goat genome CHIR_1.0 in 2013 updated version CHIR_2.0 in 2015. In decades, NGS has emerged as a revolutionary tool for genomics study involving both structural genomics (SNP beadchips, GBS) and functional genomics (RNA-seq). Utilization of NGS technologies in research investigations would provide radical insights in field of livestock genomics. With the massive development of high-throughput NGS technologies, the research of biological sciences has been entering era of holistic approach incorporating all omics studies in understanding a physiological process or biological process, production performance, disease resistance and exerting an increasing effect on the future development of various fields of biosciences *viz*. Structural genomics, epigenomics, comparative genomics, transcriptomics, metabolomics, metagenomics, oncogenomics and pharmacogenomics even beyond. High throughput genotyping can also be put to use in Copy number variation (CNV) detection. CNV refers to a 1 kb or larger DNA segment having variable numbers of copies in comparison with a reference genome. One approach for CNV detection is by comparison of the fluorescent signal intensity ratios of alleles at each SNP across the genome based on the Illumina BeadChip platform. Whole-exome

sequencing (WES) is a targeted sequencing method under NGS where only the exome regions are targeted for sequencing. Mostly only less than 1 to 2 percent of the entire genome represents actual coding regions for functional proteins. Whole-exome sequencing (WES) provides a cost-effective alternative to Whole genome sequencing for studies focusing on functional proteins. Epigenetic inheritance plays a crucial role in gene expression in early embryo development, tissue specific expression and in imprinting. Epigenomics are studied by using whole genome bisulfite sequencing (WGBS) which gives sequence methylation by comprehensive base-pair resolution and quantitative information at most genomic cytosine. Genome-wide gene expression study / transcriptomics/Global Gene Expression Analysis investigates the expression levels of all gene transcripts in a particular cell, at a particular time, and in a particular state. The gene Up-regulation and down-regulation result in diverge levels of proteins that induce phenotypic changes. Understanding the transcriptome is essential for interpreting the functional elements of the genome, revealing the molecular constituents of tissues and also for understanding development. With advent of genomics that addresses the same issues through the examination of the molecular signatures of all genes using high-throughput techniques. RNA-Seq aims to catalogue all species of transcript, including mRNAs, non-coding RNAs and small RNAs; to determine the transcriptional structure of genes, splicing patterns and other post-transcriptional modifications; and to quantify the changing expression levels of each transcript during development and under different conditions.

LEAD PAPER

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Dr Prasad Thomas is a graduate of KVASU, Kerala in 2005; with an MVSc (Animal Biotehnology) in 2007 from ICAR-IVRI, Uttar Pradesh and PhD in 2018, from the Freie Universität Berlin (FU-Berlin), Germany after receiving the ICAR-International Fellowship (2013-14). Presently Dr. Thomas is working as a Senior Scientist in the Division of Bacteriology & Mycology at ICAR-IVRI and heading the National Salmonella Center (Veterinary). During his career he was involved in both basic and applied research on various bacterial and zoonotic pathogens such as *Salmonella, E. coli, Brucella* and *Clostridium*. Dr Thomas has published more than 60 high impact articles in various peer reviewed journals. He has been working on NGS data analysis and bioinformatics for genomics, bacterial pan-genome analysis, virulence profiling, establishment of genotyping tools, understanding antimicrobial resistance mechanisms and development of diagnostics and vaccines. He was also involved in establishing genome based typing tools for epidemiological investigations in *Clostridia* and *Brucella.*

Delineating the population structure, antibiotic resistance, and virulence potential of *Salmonella*: A bioinformatics approach

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Salmonellosis constitutes a major food borne illness and poultry is recognized as the most common source for zoonosis. Typical Non typhoidal *Salmonella* (NTS) disease in immunocompetent individuals is a self-limiting gastroenteritis. On the other hand, serovars such as Enteritidis, Dublin and Typhimurium have been reported to cause fatal systemic disease forms. Humans are often infected by consuming *Salmonella* contaminated food of animal origin such as eggs, milk/milk products and meat. NTS are most prevalent in intensively reared poultry. In the poultry industry, among NTS serovars, serovar Typhimurium and Enteritidis are primarily known for producing clinical salmonellosis in very young birds. *Salmonella* Typhimurium infections with mortality rates ranging from 10% to as high as 80% are reported in young poultry. *Salmonella* Typhimurium strains infecting humans have been reported worldwide. Two *Salmonella* strains were recovered from birds belonging to different outbreaks and farms from Bareilly region of Uttar Pradesh. In both cases, the strains were isolated from liver observed for septicaemic manifestations (perihepatitis and swollen liver) during post-mortem examinations. Following species confirmations, conventional serotyping (White-Kauffmann-Le Minor scheme) identified both the strains (NSC0003 and NSC0015) as *Salmonella* Typhimurium. Earlier studies reported variations among *Salmonella* Typhimurium strains for virulence, host associations and zoonotic potential. In this study, we used a pathogenomic-based approach to infer the disease associated genotypes and virulence gene and antimicrobial resistance carriages among strains. To compare, we involved genome sequence data representing 398 global strains associated with clinical diseases in poultry. The completeness and quality of the genomes were assessed using the mash-based distance analysis and the CheckM tool. The number of virulence genes varied from 92 to 124 among the strains, with the study strains carried 116 (NSC0003) and 111 (NSC0015) genes. The entire plasmid-encoded fimbriae (*pef*) genetic locus (4 genes; *pef*A - D) and the invasin and complement-resistance gene *rck* were unique to the NSC0003 strain. Carriage of antimicrobial resistance genes varied from 1 to 24, with the study strains carrying 4 (NSC0015) and 9 genes (NSC0003). While comparing both the strains, NSC0003 carried genes conferring aminoglycoside resistance (*aadA2, ant(3'')-Ia*, *aph(3')-IIb*), chloramphenicol (*cmlA1*), trimethoprim resistance (*dfrA12*), macrolides (*mef(B)*), sulphonamide (*sul3*) and tetracycline (*tet(A)*). On the other hand, the strain NSC0015 harboured an unique quinolone resistance gene *qnrS1* and another *dfrA* variant (*dfrA14*), but lacked the *sul3, mef(B), cmlA1. aadA2, ant(3'')-Ia* and *aph(3')-IIb* genes identified in the NSC0003 genome. MLST analysis from genome sequence data indicated Sequence Type 19 (ST 19) for both the strains. Among all strains, the predominant ST identified were ST19 (283/398) followed by ST128 (48/398) and ST34 (30/398). ST 19 has been recognized as a major pathogenic clone associated with poultry in many European countries, Brazil and China, and are recognized for their high zoonotic potential. To conclude, the study indicate that the ST19 genotype could be a prominent genotype among Typhimurium strains in India and may serve as suitable strains for developing vaccine candidates for combating poultry salmonellosis.

ORAL PRESENTATION

OP_I.1 Renal transcriptome profiling reflecting anomalous molecular signatures associated with hyperuricemia in *Gyps bengalensis*

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Hyperuricemia mediated acute nephrotoxicity has been observed in white-rump vulture (*Gyps Bengalensis*) because of exposure to NSAID medicated carcasses. In the present study transcriptional profile was obtained from renal cells in association with hyperuricemia. The mRNA Library was prepared from isolated total RNA from kidney tissues of *Gyps Bengalensis* procured for post mortem analysis. The cDNA library was constructed after end repairing, dA tailing, adapter ligation and products enrichment followed by paired-end sequencing. A de novo assembly of the derived clean and high-quality reads were conducted using the Trinity. The transcripts so generated with length ≥500 bps were annotated and classified by a sequence similarity search against NCBI non-redundant protein database (Nr), Gene Ontology (GO) and KEGG database. BLASTX tool and unigenes were used to do the homolog-based search. The known miRNA profiling was done by homology-based BLAST from Chordata miRbase-21 and the novel miRNAs were predicted by MIREAP. Comparative differential expression of both mRNAs and miRNAs was done by De Seq R package. The unigenes were found to be up-regulated in vulture when the Log2 Fold change is greater than two else denoted to be down-regulated. Besides, the global differential expression picture, the individual FPKM values were used for comparing the expression urate transporter gene. Amongst the commonly expressed 32033 unigenes were found to be up-regulated, 57192 were found to be down-regulated whereas 85812 unigenes were found to remain neutral. The longest unigenes for urate transporters OAT10/SLC22A13, GLUT9/SLC2A9, ABCG2, ABCC4/MRP4, OAT1/SLC22A6andOAT2/SLC22A7 were found to be differentially expressed within individual species. The expression of URAT1/SLC22A12 and OAT3/SLC22A8 was found to be absent. Besides the said urate transporters, the expression of nucleoside transporters like ENT1, NPT1 and NPT4 were also examined. Amongst the commonly expressed annotated miRNAs 107 miRNAs were found to be up-regulated, 155 were found to be down-regulated whereas 829 unigenes were found to remain neutral. The present data will be helpful for investigating differential gene expression in nephrotoxic condition.

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OP_I.2 Carbonylated proteome unravels the molecular basis of cryoinjury to the spermatozoa

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During cryopreservation, oxidative damage to proteins occurs. Simple proteome analysis may not reflect the affected cellular functions, as most carbonylated proteins lose their functions. Choosing buffalo semen as a model, carbonylated proteins from fresh-extended (FESCL) and frozen-thawed (FTSCL) spermatozoa were enriched and then sent for proteomic analysis. Data were analysed with Proteome Discoverer (v2.2), PTM analysis was performed using SEQUEST, and affected functional activities were predicted using the FunRich tool (v3.0). The LC-MS/MS analysis of sperm proteins led to the identification of 415 carbonylated proteins, 151 specific to FESCL, and 405 specific to FTSCL. A total of 98 peptides have been precisely annotated that have oxidative modifications. The GO analysis of the carbonylated proteins was restricted to the selected pathways and processes relevant to the spermatozoa. In comparison to FESCL, more proteins in various cellular components, pathways, and processes in FTSCL were carbonylated. Carbonylated proteins are specifically linked to glycolysis, gluconeogenesis, oxidative phosphorylation, and fatty acid oxidation, thus hampering the energy requirement of spermatozoa. Spermatozoa's energy requirement is hampered by carbonylated proteins, which are specifically linked to glycolysis, gluconeogenesis, oxidative phosphorylation, and fatty acid oxidation. The study indicated that cryopreservation induces the carbonylation of selected sperm proteins. The findings provide significant evidence in favour of the theory that protein carbonylation is one of the major factors and molecular basis of cryodamage, which compromises the functions of frozen-thawed semen. The results can be utilised to explore substances that either suppress or limit the generation of reactive oxygen species (ROS) and particular carbonylated proteins associated with energy metabolism, free radical scavenging, cytoskeleton, plasma membrane,

capacitation, zona-pellucida binding, and sperm-oocyte interaction, and then correlate them with functional characteristics of semen. With cryopreservation becoming an inevitable tool, study becomes more relevant, and this experimental design would give better-quality frozen-thawed spermatozoa with good molecular profiles.

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OP_I.3 Global transcriptome profiling delineates in role of circular RNAs in modulating the vaccine response against Brucellosis in natural hosts

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The present study was undertaken to elucidate the genetic basis of vaccine response to Brucellosis *S19* vaccination in indigenous cattle calves. The study involved RNA-Seq analysis on whole transcriptome data on 10th-day after vaccination to elucidate the differentially expressed circular RNAs in the vaccinated animals as compared to unvaccinated controls. Three Tharparkar calves were vaccinated with the *S19* Brucella vaccine while three claves were mock vaccinated to act as control. After ribodepletion, the cDNA libraries were sequenced to generate 100 bp paired-end reads. After quality control, the reads were aligned to B*os taurus* genome assembly. SeekCRIT program was used to elucidate the differentially expressed circular RNA. A total of 32610 circular RNA events were observed in Tharparkar vaccinated versus control comparison. Among these, 31875 were circRNAs while 735 events included ciRNA. The maximum number of exons involved in single circular RNA was 50. A total of 3125 circular RNAs were differentially dysregulated ($p<0.05$) involving 1799 unique genes on comparison of vaccinated calves with unvaccinated control animals. These genes were involved in important pathways including the T-cell receptor signalling pathway; ubiquitin-mediated proteolysis; B-cell receptor signalling pathway; endocytosis; phosphatidyl-inositol signalling pathway; regulation of actin cytoskeleton; nucleocytoplasmic transport; and thyroid hormone signalling pathway. Overall, the dysregulated circRNAs were involved in modulating the different facets of innate and adaptive immune response post-vaccination. The study provides important insights into the role of circular RNAs in modulating the vaccine response in indigenous Tharparkar calves.

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OP_I.4 Analyzing the impact of homology arm length on site-specific CRISPR/Cas9-mediated gene knock-in

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CRISPR/Cas9-induced homology-directed repair (HDR) is gaining popularity among researchers as a valuable genome editing tool for gene knock-in to add a new phenotype and correct the existing one in a cell line or animal model. This study used CRISPR/Cas9 to target the thymidine kinase gene in Vero cells, leveraging homology- directed repair (HDR) for precise gene insertion. The research focused on evaluating the efficiency, accuracy, and specificity of the CRISPR/Cas9 system in facilitating site-specific integration at the thymidine kinase locus. The effectiveness of HDR depends on the length of the homology arm, although there is still much debate about the ideal length for an effective knock-in of the CRISPR/Cas9 gene. The experimental procedures included designing and providing guide RNAs, Cas9 protein, and donor DNA templates for Vero cells. Donor plasmids were constructed by cloning a prokaryotic vector backbone with a 2 kb heterologous eukaryotic expression cassette (EEC) containing the GFP gene and 2 kb homology arms (HA) at each end. After transfection, gene-knocked in Vero cells were selected by BrDU (5-bromo-2′-deoxyuridine). The total number of GFP-positive and - negative cells from the mixed cell population was counted, and the influence of the length of the homology arm was determined. Finally, the CRISPR/Cas9-edited Vero cells were selected by single-cell cloning and characterized by PCR, karyotyping, and Sanger sequencing. Quantitative PCR, sequencing, and phenotypic assays were used to evaluate the targeted integration and functional effect of the modified thymidine kinase gene. In this experiment, the data showed that the ideal HA length for inserting a heterologous 2 -kb gene is between 0.8 and 1 kb. Furthermore, the resulting GFP-positive cell clone has two Lox sites that could be useful for creating a new cell line by replacing the GFP cassette with other valuable genes by Cre recombinase.

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ORAL PRESENTATION

OP_I.5 Sequencing-free molecular identification of *Saprolegnia parasitica*

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`*Saprolegnia* is a genus of oomycetes or fungus like organisms, also known as water molds. These organisms can cause a disease known as Saprolegniasis in fish which is characterised by the presence of white or grey tuft of hyphae at the site of infection. The genus contains many species which were considered to be secondary pathogen but some species such as *S. parasitica* is highly virulent and can cause primary infection in fish. *S. parasitica* is associated with mass mortality in many farmed fishes and decline in population of wild fish and amphibians. Therefore, it is important to delineate the pathogenic strains from saprophytic ones. Conventionally, *Saprolegnia* species are identified through microscopic observation of unique sexual reproductive features which is time consuming, difficult and often not reliable. At present, molecular method of species identification is mainly done through amplification and sequencing of ribosomal DNA internal transcribed spacer (rDNA-ITS) region. However, sequencing by outsourcing further delays the process and also increases the cost. Considering the above points, we have developed a highly sensitive multiplex PCR that simultaneously amplifies rDNA-ITS region and a species specific site of hypothetical protein gene. In positive reaction, amplicons were visible as two distinct bands at ~750 bp and ~365 bp. In negative reaction, there is a single band only at ~750 bp. In this multiplex PCR, the amplicon at 750 bp served as internal amplification control and the other helps in species identification. Internal control makes it easier to rule out any false negative results so that correct interpretation can be done. The protocol is also highly sensitive with limit of detection, 0.016 ng of template DNA. As this protocol can identify *S. parasitica* in a single reaction without sequencing, it may be used as an easier, faster and cheaper molecular method for species identification.

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OP_I.6 miR-29a serves as a novel immunovirological marker and determines breed susceptibility to lumpy skin disease virus infection in cattle

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miRNAs have been implicated in regulating maturation, proliferation, differentiation, and activation of immune cells. In this study, we demonstrated a negative correlation between miR29a expression and IFN-γ levels during the acute phase of lumpy skin disease virus (LSDV) infection in cattle. In addition, in the LSDV sensitized cattle, a reduced miR-29a expression in stimulated peripheral blood mononuclear cells (PBMCs) negatively correlated with lymphoproliferation, higher levels of IFN-γ and an increased CD8+ T cell count. Furthermore, as compared to the sensitized crossbred cattle, PBMCs from sensitized Rathi (a native Indian breed) animals exhibited lower levels of miR-29a, along with enhanced lymphoproliferation and an increase in LSDV-specific CD8+ T cell counts. In conclusion, miR-29a expression was shown to serve as a novel biomarker during the acute phase of LSDV infection and may be used to predict the functionality of LSDV-specific CD8+ T cells in sensitized cattle. Also, the Rathi cattle mount a more potent cell-mediated immune response against LSDV as compared to the crossbred cattle.

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ORAL PRESENTATION

OP_I.7 A report on the whole genome analysis of terbinafine susceptible *Trichophyton indotineae* strains of canine origin isolated from India

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Dermatophytosis is a superficial mycotic disease affecting the keratinized structures present in animals and humans. Though this group of keratinophilic fungi includes a number of genera, the majority of infections in humans and animals are caused by *Microsporum* and *Trichophyton*. Among the *Trichophyton* genera, *T. mentagrophytes* is a zoonotic pathogen. It is a highly heterogeneous group of dermatophytes with a large number of reported strain variations. *T. mentagrophytes* ITS genotype VIII was identified as an endemic strain of human dermatophytosis in India characterized by terbinafine resistance in the majority of the strains. The point mutations in the squalene epoxidase genome confer the property of terbinafine resistance in these strains. Recently, this genotype has been recognized as a separate species and renamed as *T. indotineae*. In this study, we report the whole genome sequence analysis report of the two canine strains of *T. indotineae* isolated from Uttar Pradesh, India. The strains had a genome size of around 22 Mb with 6800 protein-coding genes. Both the strains were harboring a mitochondrial genome of size 24Kb. Phenotypically both the strains were susceptible to terbinafine which was supported by the absence of point mutations in the corresponding squalene epoxidase genes. Functional annotations based on the whole genome data of a single strain identified approximately 200 CAzymes and secretomes. The proteins identified include many lyases, keratinases, chitinases, subtilisins, and fungalysins. Whole genome-based comparative analysis of the two strains was carried out with the 65 *Trichophyton* genomes downloaded from the NCBI database and a species-specific clustering was observed. However, a phylogenetic analysis based on 12 *T. indotineae* whole genome data identified heterogeneity with two distinct clusterings. This study indicates the complex diversity existing among the different species and strains of dermatophytes and the need to monitor the genotypes of the predominant and emerging dermatophytes of both animals and humans.

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OP_I.8 Synthetic sperm membrane stabilizer enhances semen preservation and fertility of ram semen

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Cold-shock sensitivity of sperm depends on the type and proportion of lipids present in cell membrane. Higher cholesterol in sperm membrane provides more stability against cold shock while presence of lesser polyunsaturated fatty acids (PUFA) and higher saturated and monounsaturated fatty acids minimize oxidative damage to sperm membrane. Synthetic liposome could be used to enhance the cold shock resistance and minimize oxidative damage to sperm membrane by increasing the presence of these beneficial lipids in and around the sperm membrane. In the present study, a novel liposome was synthesized and assessed on preservation and fertility of ram semen. The liposome was characterized for physico-chemical properties, stability and it's membrane stabilizing effect both during liquid and cryopreservation of ram semen. Finally, it's effect on fertility rate was examined in liquid-preserved ram semen. The size of liposome ranged between 158 and 211 nm and zeta potential between -29.9 and -37.0 mV. It was found extremely stable while liquid stored at 2 - 5 °C. It reduced lipid peroxidation and enhanced both structural and functional integrity of sperm membrane as compared with the egg yolk in liquid-preserved ram semen. In presence of the liposome, sperm

motility and functional membrane integrity were increased in a dose-dependent manner following liquid preservation of ram semen. Similarly, post-thaw sperm progressive motility, acrosomal integrity, mitochondrial membrane potential and sperm cholesterol content were higher while sperm capacitation was lower in presence of it as compared with the egg yolk. The fertility rate of 48 h liquid-preserved ram semen was also higher in presence of the liposome as compared with egg yolk. In conclusion, the novel liposomal formulation was found superior to egg yolk as sperm membrane stabilizer both in liquid and cryopreservation of ram semen and hence could be used in ram semen extender in place of egg yolk to improve fertility rate of preserved ram semen.

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OP_I.9 Improved quality and fertilizability of cryopreserved *Bubalus bubalis* spermatozoa with the supplementation of methionine sulfoxide reductase A (MsrA) and protein iso-aspartate methyl transferase (PMIT)

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Spermatozoa undergo protein modifications due to the oxidative stress resulting from freezing-thawing that impair its functional life-span. With the increasing information on the oxidatively damaged proteins of cryopreserved semen; it is important to study the effect of supplementation of protein-repair enzymes to the semen extender on the quality of cryopreserve semen. Study reports the effect of supplementations of rPIMT and rMsrA on the quality of cryopreserved buffalo semen. Initially, the rPIMT were supplemented at different concentrations to the semen extender at 2.0 µg/ 80 million spermatozoa/ml concentration group showed a significant improvement in frozen- thawed semen quality compared to other groups. Further, a combined protein treatment study having the supplementation of rPIMT (2.0 µg/ 80 million spermatozoa/ml) plus rMsrA (1.5 µg/ 80 million spermatozoa/ml) improved the quality of frozen-thawed semen with increase of 9.73% progressive motility, 14.17 % viability, 14.34 % HOST positive sperms, 14.44 % acrosome integrity as compared to control group. The frozen-thawed semen of combined study has shown better functional attributes as compared to only rPIMT. The computer- assisted sperm analysis (CASA), inematics values have significantly increased in Group IV (alone 2 ug rPIMT/ml study) and also in group III of combined (2 ug/ml PIMT plus 1.5 ug/ml rMsrA) study. Further, the *in vitro* fertilizability of frozen-thawed spermatozoa supplemented with rPIMT in group IV is 2.32 times higher than the control group. In conclusion, an overall improvement in frozen- thawed progressive motility, HOST, acrosome-integrity, fertilizing ability has been observed upon supplementations of rPIMT plus rMsrA to tris-egg yolk-based extender during cryopreservation. This may be due to the repair of the oxidatively damaged proteins of seminal plasma by PIMT and MsrA supplementation. Further, study is needed to investigate the actual proteins repaired in the semen and correlate their role in improving the frozen-thawed semen quality.

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POSTER PRESENTATION

PP_I.1 Genomic characterization of *Stenotrophomonas* spp. Isolates from animals and associated environments

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The Genus *Stenotrophomonas* includes 25 species, frequently encountered in diverse animal and environmental settings. It is responsible for a significant increase in nosocomial infections and is known for its inherent resistance against multiple antibiotics. In the present study, four strains recovered from diverse sources (Endocardiogram gel used at a veterinary hospital, nasal swab from dogs, lung swab from leopards, and raw milk samples from cattle) were confirmed as *Stenotrophomonas* spp. based on cultural, biochemical, molecular, and by MALDI-TOF MS. All the strains were resistant to carbapenem, penicillin and beta-lactamase inhibitors, cephalosporin, and kanamycin. However, they had higher susceptibility to fluoroquinolones and sulfonamides. All four strains were subjected to whole genome sequencing following standard procedures. For accurate taxonomical classifications, we used two phylogenetic clustering-based approaches viz., gene-based (16S rDNA and 23S rDNA) and genome-based (ANI and TYGS). The analysis identified three strains representing the species *S. maltophilia* and one as *S. muris*. More than 40 antimicrobial resistance and virulence genes were identified within the strain genomes. Correlating their phenotypic resistance to carbapenems, we observed *blaL*1, *blaL*2, and aminoglycoside modifying enzymes in the genomes. Similar correlations were observed for three strains with the *aph(3'II)* gene for having phenotypic resistance for aminoglycoside (kanamycin) whereas the strain (PM20Lgb) lacking the gene was susceptible. The strains harboured RND and MFS efflux pump gene cassettes probably to counter the effect of antimicrobials. The study concludes that *Stenotrophomonas* spp. may be a potential pathogen in animal and their environment. Given their multidrug-resistant nature, therapeutic interventions are constrained, underscoring the need to accurately identify *Stenotrophomonas* spp. during routine diagnoses of clinical cases in animals.

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PP_I.2 Cloning and sequence analysis of HC58 complete cds of *Haemonchus contortus*: An Indian isolate

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Haemonchus contortus, a hematophagous parasite, causes high mortality and economic loss in small ruminants. Control strategies include vaccination, breeding worm-resistant sheep lines, and integrated management. Efforts are ongoing to develop recombinant protein-based vaccines. In the present study, HC58 mRNA complete cds sequence (AY948978.2) was retrieved from GenBank and expression primers were designed to clone the complete ORF of HC58. The cDNA was prepared from RNA of *H. contortus* was cloned into pET-32a(+) expression system. The recombinant clone of HC58 was sequenced and analyzed using nucleotide-Basic Local Alignment Search Tool (nBLAST) of NCBI and DNASTAR Lasergene software. The BLAST analysis shown that the newly generated sequence of HC58 is 94.64% identical to that of HC58 complete sequence of *H. contortus* Chinese isolate (AY948978.2) in GenBank. A closer comparison of our Indian isolate of HC58 gene when aligned with that published sequence of HC58 Chinese isolate (AY948978.2) revealed that our isolate of *H. contortus* differed by 40 nucleotide substitutions, majority of which were either between 91 to 114 nucleotide positions or between 655 to 676 nucleotide positions. Besides these, 03 nucleotide deletions in HC58 gene sequence of Indian isolate were also observed at 43 to 45 nucleotide positions. In addition, the sequence was also compared with cysteine proteinase mRNA partial cds (AF305964.1) from the Australian isolate and it was found identical up to 86.76%. Our sequence matching at the region of 183-258 and 277-318, with deletion of 18 nucleotides in the Australian isolate. The variations in the nucleotide sequence observed in the present study suggest that the HC58 gene of *H. contortus* Indian Isolate is genetically differing from that of Chinese isolate. The antigenic variations in HC58 of Indian isolate might be due to evolutionary process of the parasite in different geographical locations, which may even alter the pathogenicity of *H. contortus*.

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POSTER PRESENTATION

PP_I.3 Comparative seasonal adaptation of indigenous dairy cattle to heat stress

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Indigenous cattle breeds are reported to be better adapted to adverse temperatures under Indian climatic conditions. Hence the current investigation was carried out to compare the response of two indigenous dairy cattle Viz., Gir and Rathi to heat stress during summer and autumn seasons. Six apparently healthy lactating animals from each breed and total of twelve (12) animals in each season were selected for the study. The blood samples were analyzed for serum biochemical profile, oxidative status and HSP70 expression. Serum glucose and triglyceride concentrations were significantly lower while serum cholesterol and creatinine levels and the activity of ALT and AST were not altered during summer compared to autumn season in both the breeds. Serum calcium, magnesium levels were low while phosphorus levels were high in both cattle breeds during summer compared to autumn season. Gir has recorded significantly low Ca levels during summer. While no significant difference in lipid hydroperoxide (LHP) levels was observed, malondialdehyde (MDA) levels decreased significantly ($P<0.05$) during summer compared to autumn season. Total antioxidant capacity (TAC) and glutathione (GSH) levels as well as the activities of catalase, glutathione peroxidase and glutathione -s- transferase (GST) activities were significantly high and Superoxide dismutase (SOD) activity was significantly low during summer compared to autumn season. Significantly ($P\leq0.05$) higher levels of HSP70 was observed in Rathi compared to Gir during summer, could be an indicator of higher heat tolerance of Rathi cattle compared to Gir.

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PP_I.4 Comparative transcriptomics of chicken trachea infected with different clades of highly pathogenic Avian Influenza Cirus H5N1 reveals molecular basis of host response

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Due to its high mortality rate and zoonotic potential, Highly Pathogenic Avian Influenza Virus (HPAIV) subtype H5N1 is a significant public health concern. The virus has diverged into various clades, such as clades 2.2 and 2.3, each of which has unique genetic and antigenic properties. In this study the host response to different clades (clade 2.2 and clade 2.3) of HPAIV H5N1 infection was studied by comparative transcriptome analysis of chicken trachea experimentally infected with H5N1 of Clades 2.2 and 2.3. Our findings showed considerable variations in the expression of immune-related genes, and pathways between the clades. Compared to clade 2.3 virus infection, clade2.2 virus infection has a more muted immune response and fewer DEGs. Here, we found a total of 635 and 1226 differentially expressed genes (DEG) from clade 2.2 and clade 2.3 viral infections, respectively. We conducted a Gene ontology (GO) enrichment analysis and a pathway analysis to examine the impact of various clade infections on immune response. Functional analysis revealed the immune regulatory pathways were downregulated in both clades with more genes engaged in clade 2.3 infection. The cytokine-cytokine receptor interaction among the downregulated genes is significantly increased, according to KEGG pathway analysis. The interferons and interferon stimulated genes (ISG) profiles with respect to the two clades are significantly different. Also, we have significantly differentially expressed candidate genes that may contribute to the variations in pathogenecity between the two clades of viruses. These discoveries provide important insights on the molecular basis of H5N1 pathogenesis that are of immense use for the creation of more efficient prevention and control strategies against H5N1.

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POSTER PRESENTATION

PP_I.5 Development of an egg yolk-free semen extender for buffaloes

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Poultry egg yolk is commonly used in conventional semen extender for dilution and and subsequent cryopreservation of buffalo and cattle semen. However, preparation of semen extender using egg yolk is associated with several problems. The egg yolk has to be fresh as its shelf-life is just few hours. Extensive variability between one batches of egg to another leads unequal post-thaw motility and viability of spermatozoa. Egg yolk may be contaminated with several pathogenic microbes posing risk to its use in animal breeding. Separating egg-yolk from the egg albumin is also a cumbersome process on the day of semen collection. Further, egg yolk contains some unknown factors that inhibit respiration of spermatozoa thus diminish their motility. During semen cryopreservation with egg yolk-based semen extender, spermatozoa undergo capacitation like changes called cryo-capacitation and this process is also associated with an increase in protein tyrosine phosphorylation in spermatozoa that otherwise should occur during physiological capacitation. Egg yolk-free semen extender is available commercially in foreign countries. Thus, it has to be imported, but, it is bit expensive. To the best of our knowledge, no such egg yolk-free semen extender is available in India. The present study was conducted to investigate whether we can replace the egg yolk of semen extender with any of the milk components such as skim milk, milk caseins, whey proteins or their combinations. Of all the milk components studied, the whey protein-based semen extender was found to be very promising to serve as egg yolk-free semen extender [Patent application No. 202241015794 dated 22.03.2022] for cryopreservation buffalo semen as the post-thaw sperm motility and progressive motility of buffalo semen cryopreserved with this new semen extender were increased significantly ($P<0.05$) as compared to that with the control egg yolk-based semen extender. Further, this egg yolk-free semen extender could reverse significantly the protein tyrosine phosphorylation of sperm proteins that was observed in egg yolk-based semen extender.

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PP_I.6 Genetic parameter analysis of reproductive and longevity traits in Landlly crossbred sows

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Global swine breeding programmes are gaining popularity to genetically select better sows for reproduction and lifetime output, as they have a significant impact on sow productivity, welfare, and profitability. Accurate heritability estimates for variables associated with reproduction and longevity, as well as genetic correlations between the pair of traits, are required for the genetic improvement plan for these traits to be successful. Therefore, the present study was conducted with the objectives to examine the reproductive and longevity traits and estimating the genetic and phenotypic parameters for these traits in Landlly sows kept at a swine production farm, Izatnagar, Uttar Pradesh, India. The effect of non-genetic factors on different reproductive traits was estimated using SPSS software (version 16.0) while the Bayesian approach was utilized to estimate the genetic parameters for reproductive and longevity traits. The current study's result that the year and season of birth have a significant effect on reproductive characteristics generally suggests that management practices may be optimised to increase production performance. The heritability estimates for the reproductive and longevity traits were low which indicates that there is a very little additive genetic variance in these traits, and individual selection will not be helpful for improving them. Some traits, such as LSB_FF (litter size at birth during first farrowing) & LSW_FF (litter size at weaning during first farrowing) and LSB_SF (litter size at birth during second farrowing) & LSW_SF (litter size at weaning during second farrowing) had a moderate genetic correlation, suggest that indirect selection can be used to improve these pairs of traits.

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POSTER PRESENTATION

PP_I.7 Genome-wide elucidation of selection signatures within the population in Landlly crossbred pigs

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The commercial piggery sector holds immense potential in India and can meet global protein demand. However, there has been no exploration of the Indian pig population regarding selection signature studies. In India, the commercial piggery sector is primarily characterized by the prevalence of crossbred pig varieties. These varieties have been developed by leveraging the highly productive exotic germplasm within the framework of the AICRP on pig. This research program has been instrumental in advancing the productivity and profitability of pig farming in India. The crossbred varieties have been selectively bred to combine the desirable traits of different breeds, resulting in pigs that are well-suited to the Indian environment. In this study, we analyzed a crossbred pig population Landlly, developed at ICAR-IVRI to perform a selection signature analysis. The Landlly pig is a suitable breed for the Northern parts of India, as it inherits 75% of its genes from the Landrace pig breed and 25% from the indigenous Ghurrah pig breed. A total of 69 Landlly, 16 Landrace, and 11 Ghurrah pigs were genotyped using the Illumina PorcineSNP60 v2 BeadChip. The Landlly population was accessed for selective sweeps within the population using two approaches i.e., iHS & Tajima's D. Using multiple approaches enhances the effectiveness of selective sweep analysis, thus providing more robust results. The genomic segments that were under selection were used to search for overlapping genes and QTLs in the BioMart, ENSEMBL, and Pig QTL database in AnimalQTLdb, respectively. Various genes of economic importance were reported to be under selection in the Landlly pig population like NAV2 responsible for mammary function, U6 gene related to immunity, etc. This basic study can pave the way for future genomic studies in the Indian pig population. The regions reported under selection can be used as candidate genes for future breeding plans of the Landlly pig population.

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PP_I.8 DNA methylation profiles of indigenous (*Bos indicus*) and crossbred cattle (*Bos indicus* X *Bos taurus*) showing distinct heat stress response

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Epigenetic variations may potentially contribute to the stress response and adaptation mechanisms in animals. The zebu (*Bos indicus*) cattle adapted to tropical conditions whereas taurine cattle *(Bos taurus*) are adapted to temperate conditions, hence the native zebu cattle and its crossbred (*B indicus* X *B taurus*) show distinct responses to heat stress. The present study examined the genome-wide DNA methylation patterns for differences in the thermo-adaptability of Hariana (*B indicus*) and its crossbred, Vrindavani (*B indicus* X *B taurus*) cattle. The *in vivo* heat stress challenge was given to these two breeds of cattle. Physiological responses to heat stress, estimated values of Iberia heat tolerance coefficient (HTC) and Benezra's coefficient of adaptability (BCA) revealed better relative thermo-tolerance of Hariana compared to the Vrindavani cattle. Genome-wide DNA methylation patterns show marked differences between the two breeds. The comparative analysis indicated presence of differentially methylated CpGs (DMC) with both hypermethylated and hypomethylated in Hariana compared to the Vrindavani cattle. Further, we found genes that showed both differential methylation and differential expression that are involved cellular stress response functions. Taken together, the results revealed the involvement of DNA methylation in the regulation of heat stress response and long-term adaptation of *B indicus* cattle for higher environmental temperature.

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POSTER PRESENTATION

PP_I.9 Exploring epitranscriptomic regulation of proinflammatory cytokine production in SARS-CoV-2 infection

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Elevated level of inflammatory mediators, such as cytokines and chemokines—also known as hypercytokinemia or cytokine storm—is linked to a significant risk factor for the severity or mortality of COVID-19 patients; but the molecular mechanism behind this phenomenon is unclear. Epitranscriptomic modifications are RNA molecules' post-transcriptional changes that can significantly affect their structure, stability, and function. Even these modifications play important role in controlling the replication of viruses. This work showed that methylation (m6A modification) of viral RNA occurs when SARS-CoV-2 infects A549 cells and that m6A modification in the SARS-CoV-2 genome is a dynamic process (switch on off phenomenon). SARS-CoV-2 infection to A549 cells also induces production of proinflamamtory cytokines (IL-6 and TNF-α). The SARS-CoV-2 genome's m6A modification and the genes that produce proinflamamtory cytokines appear to be correlated, suggesting that the m6A modification has a function in inducing cytokines in the target cells. Furthermore, IL-6 was induced in A549 cells upon transfection of methylated RNA rather than unmethylated RNA, indicating that IL-6 induction is m6A-dependent. The levels of IL-1β and TNF-α were comparable in cells that received either methylated but not unmethylated RNA, which suggested that induction of IL-1β and TNF-α is m6A-indepenedent. DZNep (a small molecule chemical inhibitor of methylation) suppressed SARS-CoV-2 yield by inhibiting synthesis of viral RNA. It also suppressed the levels of m6A-modidfes SARS-CoV-2 RNA, concomitant with decreasing the levels of IL-6. In conclusion, this study provides novel insights on epitranscriptomic control of proinflammatory cytokines (cytokine storm) production in SARS-CoV-2 infected cells. The cytokine storm may be therapeutically managed by inhibiting cellular enzymes responsible for methylation (m6A) of SARS-CoV-2 RNA in the target cells.

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PP_I.10 Full genome analysis of *Rotavirus* strain isolated from buffalo in Gujarat provides evidence for a human-to-artiodactyle interspecies transmission event

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The *Rotavirus* (*RV*) has been recognized as the leading cause of viral diarrhea and acute gastroenteritis in humans and animals. *Rotavirus* A (*RVA*) produces new genome constellations by reassortment of its 11 segmented double stranded RNA genome that enable it to enlarge its host range or elude immune responses. The genome constellation of *RVA* Gx-P[x]-Ix-Rx-Cx-Mx-Ax-Nx-Tx-Ex-Hx represents the genotypes of VP7-VP4-VP6-VP1-VP2-VP3-NSP1-NSP2-NSP3-NSP4-NSP5/6, respectively. The potential origin of new emerging strains and interspecies transmission of this virus can be traced by combining classification system, whole genome sequencing and genotypic analysis. The present research work was carried out to characterize Group A *Rotavirus* of buffalo from Surat region of Gujarat by performing whole genome sequencing (WGS) using Illumina sequencing, followed by genotyping of RNA segments. *Rotavirus* cell culture isolate, already confirmed with G10P[11] genotype was preserved and was used for RNA extraction followed by cDNA conversion, library preparation, sequencing and data analysis by using various bioinformatics tools. WGS was performed and complete genotype constellation was determined to be G10-P11-I2-R2-C2-M2-A3-N2-T6-E2-H3 for the eleven gene segments, out of which six gene segments (VP6, VP1, VP2, VP3, NSP2, NSP4) resembled human like DS-1 *RV* genotype constellation, but *NSP5/6* gene had human like AU-1 like *RV* genotype, while, *VP4*, *VP7, NSP1* and *NSP3* gene had genotype of bovine/artiodactyle origin. The gene segments of the strains of this study showed similarity with either bovine (94.75%) or human (82.32% to 96.80%) *RVA*, pointing towards

their genetic relatedness. This analysis suggests a common link between human and animal or artiodactyle rotaviruses. Overall, the genotype analysis gave evidence of zoonosis due to interspecies reassortment between *RV* strains of bovine and humans.

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PP_I.11 Studies on residual effects of iron oxide nanoparticles on membrane fluidity and freezability of spermatozoa

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Objective of this research was to study the residual effects of in-house synthesized iron oxide nanoparticles after characterization on membrane fluidity and freezability of buffalo spermatozoa. The experiment was conducted on 18 ejaculates collected by AV method from 3 Murrah buffalo bulls. IONPs were synthesized from tetra-hydrate and hexa-hydrate of ferric chloride at 3:1 ratio with continuous nitrogen purging along with maintaining a constant temperature of 65° for 90 min synthesis process following recommended protocol. Sonication was performed for 20 min for maintaining homogeneity in solution and reduces agglomeration. Perchloric acid based peptization was done to retain the stability of IONPs in suspension. Characterization of synthesised NPs was done by DLS and TEM. Size of the synthesized NPs evaluated using TEM revealed average size of NPs of 11.006 with a range of 4.27-19 nm. The AAS was used to measure the concentration of IONPs and determined as 6492 ppm /1000µL. Ejaculates with ≥3^{+} mass motility, ≥500 m /mL of conc., volume ≥1.5 mL, and IPM ≥70% were considered for further processing. IPM, mass motility, sperm viability, abnormal sperm averages, HOST and Acrosome integrity were determined to be 80.2±1.48 (%) and 3.8± 0.23, 86.8± 1.83 %,8.7 ± 0.57 %, 86.4± 2.09 % and 82.8± 2.30% respectively. In each group containing 300 million spermatozoa different concentration of IONPs viz. Control (No IONPs, (Gr I)), 50 ppm (Gr II), 100 ppm (Gr III), 1000 ppm (Gr IV) was added. IONPs addition in Gr II-IV had no significant effect (p>0.05) on PTM, viability, HOS response, acrosome integrity and DNA integrity. Level of antioxidant enzymes, SOD and CAT were comparable to the Control. The study results revealed that addition of IONPs imparted no significant change in the quality parameters of

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PP_I.12 A pathogenomic-based approach for assessing the virulence potential of *Salmonella* Typhimurium strains associated with poultry in India

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Salmonellosis constitutes a major food borne illness and poultry is recognized as the most common source for zoonosis. Two *Salmonella* strains were recovered from birds belonging to different outbreaks and farms from Bareilly region of Uttar Pradesh. In both cases, the strains were isolated from liver observed for septicaemic manifestations (perihepatitis and swollen liver) during post-mortem examinations. Following species confirmations, conventional serotyping identified both the strains (NSC0003 and NSC0015) as *Salmonella* Typhimurium. Earlier studies reported variations among *Salmonella* Typhimurium strains for virulence, host associations and zoonotic potential. In this study, we used a pathogenomic-based approach to infer the disease associated genotypes and virulence gene carriages among strains. To compare the virulence genes, we involved genome sequence data representing 398 global strains associated with clinical diseases in poultry. The number of virulence genes varied from 92 to 124 among the strains, with the study strains carried 116 (NSC0003) and 111 (NSC0015) genes. The entire plasmid-encoded fimbriae (*pef*) genetic locus (4 genes; *pef*A - D) and the invasin and complement-resistance gene *rck* were unique to the NSC0003 strain. MLST analysis from genome sequence data indicated Sequence Type 19 (ST 19) for both the strains. Among all strains, the predominant ST identified were ST19 (283/398) followed by ST128 (48/398) and ST34 (30/398). ST 19 has been recognized as a major pathogenic clone associated with poultry in many European countries, Brazil and China, and are recognized for their high zoonotic potential. To conclude, the study indicate that the ST19 genotype could be a prominent genotype in India and may serve as suitable strains for developing vaccine candidates for combating poultry salmonellosis.

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POSTER PRESENTATION

PP_I.13 Epitranscriptomic regulation of *Buffalopox* virus replication

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In this study, we evaluated the antiviral effect of S-adenosylmethionine-dependent methyltransferase inhibitor DZNep against buffalopox virus (BPXV) replication. DZNep treatment reduced viral mRNA, DNA, and protein levels while other steps of BPXV replication were not affected. Further, by RNA-immunoprecipitation (RNA-IP) assay, we demonstrated that DZNep treatment blocks interaction of eIF4E with viral 5' cap of the viral mRNA, eventually resulting in reduced viral protein synthesis. In conclusion, BPXV replication may be epitranscriptomically regulated by suppressing the methylation of the viral RNA, and hence methylation may serve as a novel target for development of novel antiviral therapeutics against buffalopox.

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PP_I.14 Comparative proteome analysis of Bikaneri and Baisalmeri camel milk with seasonal variation

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Camel milk is regarded as one of the most valuable food sources for nomadic people living in arid and semi-arid regions due to its exceptional therapeutic qualities and high nutritional content. A total of 180 milk samples were collected from 60 Bikaneri and Jaisalmeri camels (30 each) during the summer, rainy and winter seasons for the proteomics study. Protein profiling of camel milk was done by LC-MS/MS. All samples were processed and analyzed with Proteome Discoverer (v2.4) against the Uniprot camel reference proteome database. A total of 704 protein groups were identified in the milk of Bikaneri and Jaisalmeri camels on the Uniprot *Camelus dromedarius* Database. There were 21 (3%) and 17 (2.4%) unique protein groups with 666 (94.6%) common proteins in the milk of Bikaneri and Jaisalmeri, respectively. In addition, 687 (97%) and 683 (98%) specific proteins were found in Bikaneri and Jaisalmeri camel milk, respectively. Breed variation was less observed because only 18 significantly expressed proteins were obtained in the milk of the Bikaneri and Jaisalmeri camel breeds.

Protein groups during various seasons were obtained as 631, 628 and 580 in Bikaneri and 600, 517 and 633 in Jaisalmeri camel milk during the rainy, summer and winter seasons, respectively. During the rainy season, 578 (88.5%) milk protein groups were found to be common in both breeds, namely Bikaneri and Jaisalmeri camels, while 53 (8.1%) unique proteins were found in Bikaneri milk and 22 (3.4%) in Jaisalmeri milk. 497 (76.7%) milk protein groups were found common in the summer season in both breeds, while 131 (20.2%) unique proteins were identified in Bikaneri and 20 (3.1%) were found in Jaisalmeri camel milk in the summer season. 569 (76.7%) milk protein groups were found common in the winter season in both breeds, whereas 11 (1.7%) unique proteins were identified in Bikaneri and 64 (9.9%) were found in Jaisalmeri camel milk in the winter season The camel casein components (CNs) were found as αs1-CN, αs2-CN, β-CN and κ-CN. Whey protein such as β-lactoglobulin was not found in the camel milk but α-lactalbumin was abundant.

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POSTER PRESENTATION

PP_I.15 Synergistic effect of carboxymethyl cellulose and glycerol during cryopreservation of ram semen

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Artificial insemination in sheep has not been widely accepted in the field condition till date due to very poor conception rate of cryopreserved semen after cervical insemination. Poor penetrability of ewe's cervix together with short post-thaw lifespan of cryopreserved sperm is considered responsible. Non-penetrating cryoprotectants such as ethylene glycol, polysaccharides, polyamines etc. act by reducing the freezing temperature of extender and thus minimizing extracellular ice crystal formation during semen freezing. In the present study, carboxymethyl cellulose (CMC) was evaluated as a non-penetrating cryoprotectant in cryopreservation of ram semen. Ejaculates from eight adult Malpura rams were pooled and diluted (800×10^6 sperm mL^{-1}) with TES-Tris-fructose-egg yolk extender having either 5 or 6% glycerol and supplemented with 0, 0.25, 0.5, 0.75 and 1.0% (w/v) CMC. Diluted semen was packaged into 0.25 mL French mini straws, cooled progressively to 5 °C inside a cold cabinet and then equilibrated for 22 h at 2 - 5 °C. Freezing was carried out at -25 °C min^{-1} up to -125 °C by using a cell freezer (Planer, Biomed R-204, UK) and finally the straws were plunged into liquid nitrogen. Post-thaw progressive motility was higher ($P<0.05$) in 0.75% CMC-treated group as compared with control. Overall, both pre-freeze and post-thaw sperm kinetics was comparable between the CMC-treated and control groups. Post-thaw sperm viability, acrosomal integrity and sperm with high mitochondrial membrane potential (hMMP) were relatively higher while sperm with high membrane cholesterol was significantly ($P<0.05$) higher in the presence of 0.25% CMC compared to the control. Sperm having hMMP and non-capacitated sperm were significantly ($P<0.05$) higher in presence of 5% glycerol than 6% glycerol. In addition, 0.5% CMC in combination with 5% glycerol resulted in higher functional membrane integrity than with 6% glycerol in the extender. In conclusion, 0.25 - 0.5% CMC together with 5% glycerol act synergistically during cryopreservation of ram semen causing improvement in post-thaw qualities of cryopreserved ram sperm.

Technical Session - II

Structural & Synthetic Biochemistry-Role in rational drug design

LEAD PAPER

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Dr Amit Kumar Pandey obtained his B.V.Sc and A.H degree from Odisha Veterinary College, OUAT, Bhubaneswar, Odisha. He received his M.Sc and PhD degrees from National Dairy Research Institute (NDRI), Karnal, Haryana and Indian Veterinary Research Institute (IVRI), Izatnagar, U.P., India respectively. He did his postdoctoral training from University of Nebraska-Lincoln (UNL), Nebraska, USA and University of Massachusetts Medical School (UMASS), Worcester, Massachusetts, USA. In 2011, he received the prestigious Ramalingaswami fellowship and joined Translational Health Science and Technology Institute (THSTI), as an Assistant Professor. He is currently working as Associate Professor, Infection and Immunology, THSTI, Faridabad. His area of interest includes understanding mycobacterial pathogenesis with major focus on understanding various mechanisms of antibiotic and disease persistence leading to AMR in tuberculosis. His lab is working on the hypothesis that targeting "persisters" would help shorten the therapeutic regimen and will reduce the frequency of AMR cases in tuberculosis. He has published many research papers on tuberculosis biology and has over 24 years of experience in working with several in-vitro and in-vivo animal models of tuberculosis infection.

Targeting "adaptability": Exploring novel therapeutic opportunities and strategies to shorten the current anti-TB regimen

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Tuberculosis remains one of the world's most important infectious diseases, with an estimated 11.0 million cases and 1.6 million deaths in the year 2021. Mycobacterium tuberculosis, the causative agent of tuberculosis, demonstrates immense plasticity with which it adapts to a highly dynamic and hostile host environment. This high adaptability phenotype is a consequence of the enhanced ability of the pathogen to neutralize different host-induced stressors. Consequently, the pathogen, by modulating its metabolic and growth rate, strikes a perfect balance between virulence and growth. These phenotypic changes also induce some degree of drug tolerance that necessitates prolonged exposure to anti-TB therapy during treatment. Depending on the drug susceptibility profile, the treatment duration can extend from 6 months to 2 years. Noncompliance associated with the lengthy treatment regimen increases the frequency of multidrug-resistant and extensively drug-resistant cases of tuberculosis. Identifying and targeting both the pathogen and host pathways critical for optimum pathogen adaptability could increase the effectiveness of antibiotics and shorten treatment duration. One of the major research interest of our lab is to identify and target proteins from both host and pathogen that critically modulate "adaptability" and influence longterm disease and antibiotic persistence in tuberculosis. Further, using animal model studies we have shown that these genes also modulate the host immune response in ways that favour the growth and survival of the pathogen inside the host.

INVITED PAPER

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Dr Awadh Bihari Yadav did his PhD from Central Drug Research Institute, Lucknow, India, one of premier institute in the country. His PhD work was on targeted delivery of drug-loaded microparticles for the development-targeted therapy for tuberculosis. He did his post doc from Royal College of Surgeons in Ireland. At present, he is working as Assistant Professor, Center of Biotechnology, University of Allahabad. His group is working on different areas for the development of targeted therapy for the treatment of lung inflammation, lung cancer, tuberculosis, diabetes and mastitis in dairy animals. He is using microparticles, nanocarriers and composite nanoparticles for targeted delivery of therapeutics protein, peptide, siRNA, shRNA and drug molecules for the cure of different disease. He has also successfully completed 6 extramural project funded by Department of Biotechnology, SERB, Department of Science and Technology, Govt. of India, organized 2 GIAN course and several short term training program sponsored by SERB and DBT. He is project coordinator for Design Innovation Center of University of Allahabad, Prayagraj.

Design and evaluation of a novel peptide based on lung splunc1 loaded nanoparticles against *S. aureus* biofilms: A promising approach to combat against antimicrobial resistance

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The emergence of threatening "superbugs" of drug resistant bacterial strain due undiscerning use of antibiotics, which has caused upsurge in the global prevalence of the disease, caused by antimicrobial resistance (AMR) strain of bacterium. Multifaceted strategies are being adopted by bacteria to evade killing and develop resistance against antibiotics, one of which is biofilm formation. Methicillin-resistant Staphylococcus aureus (MRSA) accounts for 64% of positive cases, and Mycobacterium tuberculosis accounts for 54% of multidrug-resistant (MDR) and 28% of extensive drug-resistant (XDR) cases, as per the WHO report.

The misuse and overuse of antibiotics have led to the development of antimicrobial resistance, which has now become a major threat to human health. Pathogenic bacteria have evolved various strategies to combat adverse conditions, including use of different antimicrobial drugs. One such strategy is the formation of biofilms. Biofilms are notoriously difficult to treat owing to their high resistance to antibiotics; therefore, there is a pressing need for new therapeutic approaches. In This study, we reporting a novel antibiofilm peptide for the treatment of biofilms caused by Staphylococcus aureus, a major pathogen responsible for chronic and recurrent infections. Therapeutic peptides are a class of molecules composed of short chains of amino acids that have gained attention for their potential in the treatment of bacterial biofilms. This study focuses on the design and evaluation of a novel antibiofilm peptide based on the SPLUNC-1(Short Palate, Lung, and Nasal epithelium Clone 1) protein, which is an innate immune protein known for its antimicrobial and antibiofilm properties. Their different physicochemical and antibiofilm properties were analyzed using various bioinformatics tools, and their effectiveness in disrupting in vitro antibiofilm activity was assessed. In this study, we found that peptides loaded nanoparticles to enhance efficacy of therapeutics peptide against biofilm. In this study, we found that peptide loaded nanoparticles efficiently disrupt *S. aureus* biofilms. We concluded this study with peptide loaded can be used to combat against AMR by disruption bacterial biofilm approach; this approach could help to develop new therapy for the treatments of disease where antibiotics become ineffective.

INVITED PAPER

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Dr. Ratan K. Choudhary is serving as an Assistant Professor at the College of Animal Biotechnology, Guru Angad Dev Veterinary and Animal Sciences University, Punjab. Dr. Choudhary holds academic degrees inBVSc (TNVASU, Chennai), MVSc (AGB, IVRI, Bareilly),and Ph.D. (Animal Sciences, University of Maryland, USA). His doctoral research aimed to characterize bovine mammary stem cellsand*in vivo* cellular manipulation for enhanced bovine lactation persistency. He has working experience atthe United States Department of Agriculture (USDA) facility for more than six years (2007-2012). He received two post-doctoraltrainings, one from theUniversity of Kentucky (2012-2013) and the other from the University of Vermont (2018-2020), USA.
He is working on two main areas, including Regenerative medicine – wound healing, cancer, mastitis. The long-term goal of his research is to develop stem cell therapies for pets and utilize stem cells as 'drug' in bovine mastitis. And, Veterinary diagnostics – early pregnancy detection in buffaloes, AFSV, canine mammary tumors, mastitis, and tracing recombinant Bovine Somatotropin Ab(Use).
He has published 03 international books as an editor, 02 mobile apps- including "Stem Cells: Basic Understanding and Clinical Applications, available on the Google App Store. He is author of >55 papers, 10 book chapters and > 60 presentations. He is the investigator of two extramural projects and co-investigator of three projects.

Canine stromal vascular fractions: Isolation and immunomodulatory properties

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Stromal vascular fractions (SVFs) encompass a diverse population of cells within adipose tissue, characterized by their stromal and vascular components. The stromal component comprises fibroblasts, preadipocytes, and immune cells, while the vascular component includes endothelial cells and pericytes. Due to the differentiation potential due to the presence of a multipotent and self-renewal population of stem cells, coupled with their immunomodulatory ability, SVFs provide therapeutic efficacy in treating many diseases. The impact of SVFs on immune cellsrelies on cell contact-dependent mechanisms and paracrine effects, resulting in the release of various soluble factors by the stem cells that regulate immune cells. Despite the widespread use of stem cells in cell therapy trials, the immunomodulatory roles of canine SFVs havenot yet beenthoroughly investigated. Here, we focus on analyzingthe immunomodulatory properties of SVFs to explore viable options incanine regenerative medicine.

Isolating SVFs involves harvestingstem cells with the highest viability and functionality. Standard methods for SVFs isolation include enzymatic digestion of adipose tissue followed by centrifugation, which separates the SVF from adipocytes. SVFs can also be harvested non-enzymatically by mechanical macerations. The non-enzymatic method of SVFs isolation is preferred due to rapid, safe, and GMP compliance that do not involve the use of reagents of animal origin and also due to 'minimal manipulation of cells' for therapeutic applications.

Our DBT-funded project explores utilizing the therapeutic potential of canine SVFs. Under this project, first, we established a method of harvesting canine SVFs from adipose tissue. About 10 g of periovarian fat was harvested from the female dog during the cesarean section, isolated SVFs, cultured in the complete medium of DMEM, and characterized. SVFs contain approximately 7 million cells per gram of periovarian fat. Cells, when harvested, had viability > 90%. The average perimeter and area of cells of canine SFVS when grown in culture (for3 days) were 274.3 ± 10.12 µm and 3743 ± 216.3 µm2, respectively. Flow cytometry and gene expression analysis results showed enriched expression of mesenchymal stem cell markers in SVFs. Next, we studied the proliferation kinetics and immunomodulatory potentials of canine SVFs using an *in vitro* model. PBMCs of

healthy and lymphoma dogs were isolated. Results showed that the coculturing SVFs with PBMCs increased the population doubling time in both the groups of PBMCs, and decreased (3.5-fold downregulation of *Ki67; $p < 0.05$)* cell proliferation in a ratio-dependent manner. Immunomodulatory properties of SFVs were evident by 44-fold down-regulation of the inflammatory cytokine tissue necrosis factor-alpha (*TNFA*) and 88-fold up-regulation (*p-value* = 0.008) of the anti-inflammatory prostaglandin-endoperoxide synthase 1 (*PTGS1*). In conclusion, SVFs showed immunomodulatory effects on the canine immune cells of lymphoma and healthy dogs, complementing their potential for therapeutic applications. Potential strategies to boost SVFs-mediated immunomodulation, to improve clinical outcomes are worth future investigation.

INVITED PAPER

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Dr. Chanchal Singh, an Associate Professor at the College of Veterinary Science, brings over fifteen years of teaching expertise in veterinary biochemistry. He completed his graduation from the West Bengal University of Animal and Fisheries Sciences, Kolkata, earning MVSc and Ph.D. degrees from IVRI, Izatnagar. Dr. Singh's impact transcends teaching; he's guided four postgraduates and one doctoral student while serving as Co-advisor to over 30 others, emphasizing his commitment to grooming future professionals. His focus on molecular and clinical biochemistry has yielded substantial contributions, highlighted by his leadership in managing SERB, DST and other extramural projects as Principal and Co-Principal Investigator, respectively. His extensive publication history comprises over 60 national and international research papers, book chapters, extension articles, and practical manuals, validating his substantive contributions and dedication to advancing scientific frontiers. Engaging globally, he's contributed as a visiting faculty at the University of Newcastle, Australia. He has garnered over ten prestigious awards for his research presentations at national conferences. Additionally, he has been actively involved in various departmental, college, and university level committees.

Advances in Organoid Technology: Implications for Animal Health and Drug discovery

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The advent of organoid technology has revolutionized biomedical research, offering promising avenues in understanding complex organ systems and disease mechanisms. Organoids, three-dimensional in vitro models derived from pluripotent stem cells or tissue-specific progenitors, faithfully recapitulate organ structure, function, and pathology. In recent years, significant strides have been made in advancing organoid technology, particularly in the context of animal health and drug discovery. One of the pivotal implications of organoids in animal health lies in their potential to model and elucidate intricate physiological processes and disease pathologies in various species. These miniature organ replicas offer a platform for studying species-specific responses to diseases, enabling tailored research models for veterinary medicine. From understanding developmental processes to simulating diseases such as cancer, infectious diseases, and genetic disorders, organoids serve as invaluable tools to bridge the gap between in vitro and in vivo studies, offering insights into animal-specific diseases that were previously challenging to investigate. Moreover, the application of organoids in drug discovery and development has garnered significant attention. These three-dimensional structures provide a realistic microenvironment for testing drug efficacy, toxicity, and metabolism, thereby reducing reliance on traditional animal models and expediting the drug screening process. The ability to generate patient-specific organoids holds promise in personalized medicine, facilitating the identification of optimal treatment strategies based on individual variations in drug response and disease susceptibility.Recent advancements in organoid technology have addressed several challenges, including enhancing the complexity and functionality of organoids, improving scalability, and implementing novel biomaterials and bioengineering approaches to mimic the native tissue microenvironment more accurately. Innovative techniques such as organoid-on-a-chip systems and incorporation of multiple cell types have further diversified the applications of organoids, offering platforms for high-throughput screening and disease modeling with enhanced physiological relevance. Nevertheless, despite the immense potential, several limitations and ethical considerations persist. Challenges related to achieving full organ functionality, reproducibility, and standardization across different organoid models remain areas of active research. In conclusion, advances in organoid technology hold immense promise for advancing animal health research and revolutionizing drug discovery. Continued innovation and collaboration among multidisciplinary fields are pivotal in addressing the challenges and maximizing the potential of organoids, thereby shaping the future of veterinary medicine and pharmaceutical development.

ORAL PRESENTATION

OP_II.1 Role of enzymes in acaricide resistance to deltamethrin and coumaphos in *Hyalomma anatolicum* ticks collected from Haryana

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In order to investigate the status of acaricide resistance and potential mechanisms of action resulting in resistance to widely used acaricides (deltamethrin and coumaphos), *Hyalomma anatolicum* ticks were collected from six dairy farms located in the Hisar and Charkhi Dadri districts of Haryana. The *H. anatolicum* tick larvae of the Charkhi Dadri isolates were found to be susceptible to both acaricides (100% mortality) by using the standard larval packet test. Four isolates showed level-I resistance, whereas just one isolate from Hisar showed level-II resistance to coumaphos. Additionally, one of the Hisar isolates (Kaimri) exhibited level-I resistance to deltamethrin. Biochemically, ticks with greater resistance factor (RF) against coumaphos had higher levels of enzymatic activity for the enzymes glutathione-S-transferase (GST), α-esterase, β-esterase, and mono-oxygenase; on the other hand, monoamine oxidase did not exhibit a consistent pattern. However, the RF showed a statistical significant correlation with GST only. Using napthyl acetate as the substrate, native PAGE analysis of *H. anatolicum* ticks revealed the presence of nine types of esterases (EST-1 h to EST-9 h). The presence of serine residue in the catalytic triad was indicated by the inhibition of esterases by PMSF in the inhibitory experiment. In order to identify potential mutations in resistant isolates of *H. anatolicum* ticks, the partial cds of the carboxylesterase and domain II of the sodium channel genes were sequenced. Nevertheless, no mutations were found in either gene, suggesting that elevated expression of detoxification enzymes may be a mechanism for resistance development in the current study.

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OP_II.2 Effect of reservatrol supplementation on serum metabolic profile, thyroid and cortisol hormone levels and expression of sirtuin 1 and 3 gene in indigenous piglets

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Poor metabolic capacity frequently found in indigenous piglets, is major factor affecting the sustainability of indigenous pig production. Sirtuins are the class of histone deacetylating enzymes which regulates the overall cellular metabolism of animals. Sirtuin 1 and 3are mainly involved in regulating nutrient sensing pathways, caloric restriction response of animals. Various nutraceuticals supplementations were found to affect the sirtuin gene expression in human and a mouse models. The present research was conducted to study the effect of reservatrol supplementation at 300mg/kg body weight for 30 days on the expression level of sirtuin 1 and 3 genes in blood samples of indigenous piglets (N=8) relative to control group (N=8) using de novo custom synthesized primers of sus scrofa species. Further various serum metabolic profile parameters and hormones T3, T4 and cortisol were studied and the results were compared between resveratrol supplemented group and control group of indigenous piglets. Gene expression results were correlated with serum metabolic profileand thyroid and cortisol hormone levels. The expression of sirtuin1 gene was found significantly higher ($P < 0.01$) and expression of sirtuin 3 gene was found significantly lower ($P < 0.01$) in resveratrol supplemented piglets relative to control group. Significantly higher values ($P < 0.01$) of total protein, albumin, globulin, calcium T3 and T4 hormones and significantly lower values ($P < 0.01$) of triglycerides, alkaline phosphatase, phosphorus, lactate dehydrogenases and cortisol were found in resveratrol supplemented piglets relative to control group. The values of total protein, albumin ,T3 and T4 were found positively correlated ($P > 0.05$) and triglycerides, phosphorus and alkaline phosphatase ($P > 0.05$) were found negatively correlated with sirtuin 1 fold expression level. Values of glucose ($P > 0.05$), BUN, triglyceride, ALP and lactate dehydrogenase ($P > 0.01$) were found positively correlated and values of calcium and albumin ($P > 0.05$) were found negatively correlated with sirtuin 3 fold expression level. Thus it is found that reservatrol supplementation is effective as nutraceutical option for improving the basal metabolic rate and metabolic capacity in indigenous piglets.

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ORAL PRESENTATION

OP_II.3 Exploring the potential of Histone deacetylase inhibitors as candidates for antiviral therapeutics against lumpy skin disease virus

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Lumpy skin disease (LSD) has caused devastating epidemics in the recent times in India. The molecular biology, replication and mechanism of the pathogenesis of LSDV in not well understood. In recent times, several host/epigenetic factors have been shown to be implicated in virus replication. However, the role of various cellular/epigenetic factors in LSDV replication is totally unexplored. We for the first time screened a small molecule chemical inhibitor library (comprising inhibitors targeting epigenetic machinery) and observed that the small molecule chemical inhibitors targeting HDAC (histone deacetylase) suppresses LSDV replication. The time-of-addition assay indicated that iHDACs inhibited DNA and protein synthesis. In addition, iHDAC also blocked LSDV induced cell contraction (which facilitates virus replication). In conclusion, HDAC (an epigenetic modifier) supports LSDV replication and may serve as potential target for development of antiviral therapeutics against LSD.

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OP_II.4 Amelioration of excision wounds by topical application of biofabricated gold nanoparticles in rat model

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In the simplified manner any breach in the continuity of skin is considered as wound. Antibiotic resistance is a big issue in wound healing which is a result of indiscriminate use of antibiotics. Both *Moringa olifera* and Gold nanoparticle (GNP) plays an important role in wound healing. The successfully synthesized *M. olifera* gold nanoparticles (MGNP) were characterized with the UV-VI spectroscopy, nano particle sizer and transmission electron microscopy. Spectroscopically, the MGNPs were characterized with the presence of a surface plasmon resonance at 521 nm, suggesting the synthesis of quasi-spherical gold nanoparticle. The nanoparticle sizer and transmission electron microscopy (TEM) analysis revealed the average size of synthesized MGNPs were 10- 60 nm. The wound healing potential of MGNP was evaluated on 42 healthy male wistar rats equally divided into three groups comprising 14 animals in each group. The animals of group I served as healthy control and treated with normal saline. The group II animals were treated with betadine and group III animals were treated with *M. olifera* biofabricated gold nanoparticle (MGNP). Treatment was done at first 5 days from wound creation. Tissue was collected from 7th days and 14th days from wound creation for hydroxyproline estimation, Real time based expression analysis and histopathology. Collagen has role in later stages of wound homeostasis and re-epithelisation process in reparative wounds as evident by the significant increase in Hydroxyproline value among the rats which is treated with biofabricated gold nanoparticle. The expression of Cox-2 gene was down regulated in MGNP treated group as compare to betadine treated group on both 7th and 14th days. In excision wound, group III showed faster healing compared with group I and II. In present study group III showed more development of all the normal cells including epidermis, dermis & hypodermis components as compare to other group. Epithelization was observed from the first day up to last of the experiment i.e. 14th day.

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OP_II.5 *In silico* identification of a novel antimicrobial peptide from rainbow trout haemoglobin

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Antimicrobial peptides (AMPs) play an essential role in the innate immune defence of various organisms, including fish. Understanding the AMP repertoire of rainbow trout (*Oncorhynchus mykiss*), an important commercial fish species, is crucial for enhancing disease resistance and promoting sustainable aquaculture. In this study, we identified potential AMPs in the rainbow trout haemoglobin sequence using various bioinformatics tools. The haemoglobin protein sequence was retrieved from the NCBI database. AntiBp predicted 24 antimicrobial peptides from the sequence with the SVM algorithm. Out of the 24 antimicrobial peptides, CAMPR3 predicted four antimicrobial

peptides to possess antimicrobial properties with the RF algorithm, while the remaining were considered non-antimicrobial. All four peptides were considered probable non-allergen and non-toxic, as predicted by AllergenFP and ToxinPred, respectively. As predicted by HemoPI, three peptides showed hemolytic tendencies, and only one peptide of 15 mer with the sequence "DKSVVKAFWGKISGK" was selected for further analysis as it did not show hemolysis. This peptide also showed antiviral and antifungal potential, as predicted by iAMPpred. SOPMA and PEP2D were used to predict the secondary structure of the peptide, and it was found that random coil was the most predominant structure of this peptide, and it also contained alpha helix and beta sheet.

Further, the properties of this selected peptide were predicted by PepCalc tools. The peptide exhibited a molecular weight of 1649.93 g/mol and a pI of 10.01. The peptide exhibited good water solubility and carried a net charge 3 at pH 7. The Amphiphilicity of the peptide was calculated through the HemoPI web server and was determined to be 1.44. These results indicate that these peptides can act as potential antibiotic substitutes, making them appropriate for further investigation and therapeutic application. In the future, this peptide will be synthesized and validated *in vitro* against bacterial and fungal pathogens for validation.

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OP_II.6 Experimental safety testing confirms that the NSAID nimesulide is toxic to Gyps vultures in India

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Population decline of Gyps vultures throughout South Asia were caused by unintentional poisoning by the various NSAIDs is being reported. Few of them were banned to use in veterinary practice to conserve the vulture population. However, many other which have not been safety tested available for veterinary use including nimesulide. In order to ascertain the toxicity, the safety-testing of nimesulide was carried out on Himalayan Griffons *Gyps himalayensis*. Two vultures were given a dose of nimesulide at maximum level of exposure orally by gavaging, with two controls dosed with benzyl alcohol. Blood samples were collected from all the birds at intervals (0, 2, 6, 12 and 24h) and serum/plasma was separated. Plasma samples were subjected to metabolite analysis and serum samples were used for analyzing biochemical parameters. In test group, the plasma nimesulide concentrations peaked after six hours, while serum uric acid concentrations increased steadily up until 24h post-treatment. Both the birds under treatment group were died after 24h, hence the further experiment was terminated. Systematic necropsy examination revealed deposition of chalky white crystals on the visceral organs. Histopathological and histochemical staining (De-Galantha staining) confirmed the presence of urate crystals in the visceral organs. The other serum biochemical parameters remained normal during entire period of the study. The control birds showed no adverse clinical or biochemical signs. Nimesulide is harmful to Gyps vultures, according to the study, and as such, its usage in veterinary medicine has to be banned in order to support vulture conservation efforts.

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OP_II.7 Drug repurposing to prevent the Lumpy Skin Disease Virus by obstructing viral RNA polymerase 30 (LSDV071)

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The lumpy skin disease virus is a dsDNA virus characterized by nodular skin of cattle. The outbreak allowed least time to react and develop conventional vaccines. The spread of viral outbreak can be controlled by selecting a suitable non-structural target molecule and screening of drugs through databases. With the advances in computing tools, the screening of thousands of drugs can be done merely in few days. The virus multiplies in host cell utilizing several transcription factors responsible for vital function. One such transcription factor of RNA polymerase 30 (Rpo30) play a significant role in viral replication and transcription and can be a potential drug target. Herein we generated an energy-minimized homology model of Rpo30 and used it for virtual screening against 748 antiviral compound libraries. The best five ligand-Rpo30 complexes were picked for further energy minimization via molecular dynamics (MDs), where

the complex with drug showing a minimum score characterized with stable hydrogen bonds and hydrophobic interactions with the catalytic site residues was selected. Our study identified some important ligands for development of remedial approach for treatment of lumpy skin disease post infection. The use of artificial intelligence and machine learning has significantly reduced duration of investigation.

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OP_II.8 Chitosan-STPP nanoparticles as innovative carriers for targeted and efficient delivery

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Chitosan nanoparticles stand as biomedical pioneers, finding applications in targeted drug delivery, imaging and diagnostics, showcasing their potential to revolutionize personalized medicine. In the pursuit of improved vaccines, Chitosan-STPP nanoparticles take the lead, promising precise antigen delivery and enhanced immune response, shaping the future of vaccine development.

Keeping this in consideration, we have developed a chitosan nanoparticle-based delivery system for the improvement of immune response to vaccine. Chitosan nanoparticles were synthesised through ionic gelation by dissolving chitosan in 1M acetic acid. Then 5 ml of Chitosan was used with the concentration of 1, 1.25, 1.42 and 2.8 to which 2 ml STPP with the concentration of chitosan 0.21, 0.28 and 0.43 mg/ml was added drop by drop in a magnetic stirrer for 1 hour. Based on the ratio of Chitosan:STPP (w/w) 12 different formulations were obtained (5:1, 3:1, 6:1, 7:1, 5:1, 4:1, 3:1, 2:1, 3:1, 10:1, 13:1, 6:1). Zeta analysis of the chitosan nanoparticles showed the size of (200-800 nm) and charge of (+37 to +45 mV). All 12 formulations with concentrations of (8.46, 9, 10, 10.21, 10.46, 10.75, 11.75, 12, 13, 20.46, 22, 23) µg/µl showed a cytotoxicity of 4-25 % at first 12 hrs of incubation on MDBK cells. Out of all the combinations, 3 nanoparticle formulations with concentrations of (8.46, 9, 10) µg/µl were selected based on the zeta size (250-350nm), charge (+43 to +47 mV) and cytotoxicity (4-10%). Intracellular uptake in MDBK cells was performed using FITC-conjugated chitosan nanoparticles. Previous studies also provided the morphological details of chitosan nanoparticles using transmission electron microscopy. Further investigation could reveal various properties which enhance the bioavailability and its effect on dose concentration and frequency of vaccine. These results thus indicate the promising role of chitosan nanoparticles as an innovative carrier for potential vaccine delivery systems.

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OP_II.9 Generation of a novel antimicrobial peptide through bio-engineering approach

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In recent days, antimicrobial peptides (AMPs) have garnered the attention of numerous researchers due to their potential applicability in various fields, including aquaculture. They are also gaining importance as potential alternatives to address public health issues caused by antibiotics and chemicals. However, the practical implementation of AMPs, particularly naturally occurring ones, is often impeded by their low stability, toxicity, and notably, higher production costs resulting from their lengthy sequence. In this study, we have developed a short and compositionally simple amphipathic peptide, RH12, which demonstrates potent antibacterial activity against bacterial fish pathogens. RH12 possesses a cationic nature and carries a net charge of +4.3. Molecular docking analysis revealed that the peptide exhibits a strong binding affinity to the virulence protein of bacteria. The synthesis of the peptide was accomplished using Fmoc-chemistry, and its antimicrobial activity was assessed against *Aeromonas sobria, A. salmonicida, A. hydrophila, Edwardsiella tarda, Vibrio parahaemolyticus, Pseudomonas aeruginosa, Escherichia coli, Staphylococcus epidermidis,* and methicillin-resistant *S. aureus*. RH12 displayed good antibacterial activity against the tested bacteria, as evidenced by the MIC and MBC values, which ranged from 0.98 to 500 µM and 8 to 600 µM, respectively. The peptide exhibited resistance to high temperatures and maintained its antimicrobial properties even in the presence of serum and salt, indicating its stability. Furthermore, the peptide demonstrated lower cytotoxicity even at higher concentrations. These findings suggest that bioengineering approach may be employed to generate effective short AMPs

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ORAL PRESENTATION

OP_II.10 Hesperetin blocks interaction of poxvirus mRNA with eIF4E without producing drug-resistant mutants.

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In this study, hesperetin was shown to inhibit the replication of multiple poxviruses, including buffalopox virus (BPXV), vaccinia virus, and lumpy skin disease virus (LSDV). Hesperetin mainly suppressed viral protein synthesis without affecting other steps of the viral life cycle such as attachment, entry, and budding. In a chromatin immunoprecipitation (CHIP) assay, we further demonstrated that hesperetin-induced reduction in BPXV protein synthesis is due to disruption of the binding of the 5' cap of viral mRNA with the cellular translation initiation factor eIF4E. The molecular docking and MD simulation studies, also confirmed binding of the hesperetin with the cap-binding pocket of eIF4E, in a similar conformation as m7GTP binds. In a BPXV egg infection model, hesperetin was shown to suppress the development of pock lesions on the chorio-allantoic membrane, as well as the associated mortality of the chicken embryos. Most importantly, long-term culture of BPXV in the presence of hesperetin did not induce the generation of drug-resistant viral mutants.

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OP_II.11 *In vitro* assessment of the anticancerous and antioxidant properties of aqueous extracts of *Bryophyllum pinnatum* flowers

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Several plant-derived compounds are currently employed successfully in cancer treatment, and many natural products have been tested against cancer. Despite enormous efforts towards this direction, several medicinal plants having numerous bioactive compounds still remained overlooked that may help towards developing novel cancer therapeutics. In present investigation *in vitro* assessment of the anticancer and antioxidant properties of aqueous extracts of *B. pinnatum* flowers was carried out. The antioxidant activity of *B. pinnatum* flower extracts was evaluated using 1,1-diphenyl-2-picrylhydrazyl (DPPH), ferric reducing antioxidant power (FRAP), ABTS assay, phosphomolybdate assay, and the cytotoxic effect of *B. pinnatum* flower on human breast adenocarcinoma (MDA-MB 231) cell line was studied using MTT assay. Furthermore, we determined the total phenolic and flavonoid contents of the extracts using the Folin-ciocalteu and aluminum chloride methods, respectively. The comprehensive results obtained from the current experiment elucidated the presence of diverse phytochemical compounds in the aqueous extract such as alkaloids, phenols, flavonoids , terpenoids, tannins etc. Total phenolic and flavonoids contents were found to be 69.69 ± 6.64 mg GAE/g and 175.671 ± 25.74 mg QE/g, respectively. DPPH radical scavenging activities (EC_{50} 46.21± 0.33) µg/mL, ABTS radical scavenging activities (EC_{50} 232.43±15.09), FRAP assay, and phosphomolybdenum assays also established significant antioxidant activity of the extract. The antiproliferative activity on MDA-MB-231 cells collectively indicated that the aqueous extracts of *B. pinnatum* flowers possess significant antioxidant and anti-proliferative activities; the IC_{50} was calculated to be 29.30± 0.58 µg/mL. These findings suggest that *B. pinnatum* flower extracts may have substantial potential as antioxidants and in inhibiting the proliferation of MDA-MB-231 cells *via* inducing apoptosis as observed in AO/EtBr, DAPI, Hoechst staining, reducing mitochondrial membrane potential and increasing intra cellular ROS production.

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ORAL PRESENTATION

OP_II.12 Coordinated interaction between Lon protease and catalase-peroxidase regulates virulence and oxidative stress management during Salmonellosis

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Lon, an ATP-Dependent Protease is essential for systemic *Salmonella enterica* serovar Typhimurium (ST) infection in mice. Proteomic comparison of ST wild-type and *lon* deletion mutant led to the recognition of a highly expressed catalase-peroxidase (KatG) protein product among five other protein candidates. Bacterial two-hybrid assay (B2H), showed that the catalytic domain of Lon protease potentially interacts with the KatG protein and causes proteolytic cleavage. The single and double *lon* and *katG* mutants revealed *katG* to be a positive modulator of both *Salmonella* pathogenicity Island-1 (SPI-1) and −2, whereas *lon* significantly affected the SPI-1 genes. Additionally, the deletion of *katG* and *lon* enhances ST susceptibility to exogenous H_2O_2. ST double deletion mutant, Δ*lon*Δ*katG* was more susceptible to survival defects within macrophage-like cells and exhibited meager colonization of the mouse spleen compared to the single deletion mutants. The findings reveal a previously unknown function of Lon and KatG interaction in Salmonella virulence. Taken together, our experiments demonstrate the importance of Lon-mediated regulation of KatG to cope with oxidative stress, intracellular survival and *in vivo* virulence of *Salmonella*.

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POSTER PRESENTATION

PP_II.1 qPCR analysis of goat β- defensin mRNA in different tissues of Osmanabadi goat

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Defensins are cationic, cysteine-rich motif containing antimicrobial peptides (AMPs) synthesized by leucocytes and epithelial cells. Expression levels of defensins vary in different tissues, highest being in those tissues that are constantly exposed to, and colonized by, microorganisms. Defensin peptides contribute to the innate immune response against a variety of bacteria, viruses and fungi. In the present investigation, goat β-defensin (GBD) expression in different tissues of Osmanabadi goat was studied using qPCR. It was observed that goat tongue epithelia had the highest level of GBD expression, followed by reticulum, rumen, omasum, kidney, spleen, liver and uterus. Minimal expression of GBD was observed in Osmanabadi tracheal tissue. To our knowledge, this is the first study to report expression of GBD in goat rumen.

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PP_II.2 Effect of plant based formulation on biochemical profile of *Rhipicephalus microplus* ticks

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Hemolymph of ticks is analogous to the vertebrate blood but it is aqueous clear fluid consisting of vital proteins, carbohydrates, lipids, hormones and hemocyte cells. The entire tick organs bath in the hemolymph and thus it acts as an important vehicle for transportation of nutrients, free or bound proteins etc. The synthesized protein precursors are released to the hemolymph for absorption by the oocytes through receptor mediated endocytosis. The impact of the treatment on haemolymph proteins of treated ticks was studied and a significantly ($p < 0.001$) lower quantity of total proteins in herbal formulation group as compared to DW treated ticks and fipronil treated group was noted. Comparative quantative protein profile of gut and ovary revealed no significant differences in the profile of gut protein amongst the different treated groups. A statistically significant ($p < 0.01$) reduction in ovarian protein of herbal formulation group in comparison to control and other treated group was recorded. Besides impact of protein content, the impact of treatment on key enzymes involved in digestion was studied. A significant ($p < 0.001$) reduction of cathepsin D enzyme level while Cathepsin B and Leucine Aminopeptidase enzymes showed no significant alteration in all the treated groups. The alkaline phosphatase assay revealed a significantly ($p < 0.001$) higher values in acaricide treated group in 12 and 24 hr post exposure however, significantly lower ($p < 0.05$) values in the formulation treated group as compared to control were observed.

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PP_II.3 Isolation, characterization and evaluation of lytic activity of bacteriophages against shiga toxin producing *Escherichia coli* (STEC) isolated from sewage environments of Bareilly

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Shiga toxin-producing *Escherichia coli* (STEC) is a significant foodborne pathogen with zoonotic potential, posing health risks from contaminated food sources. Use of antibiotics to treat STEC infections is controversial due to concerns about heightened toxin production and the risk of antimicrobial resistance. Bacteriophages, natural predators of bacteria, offer a promising alternative due to their specificity, safety, and self-replicating nature. Therefore, the main goal of this investigation was to isolate and characterize bacteriophages effective against STEC strains of bovine origin. Thirteen bacteriophages were isolated from IVRI farm wastewater and other sewage environments in and around Bareilly against indicator STEC isolates obtained from Division of Bacteriology and Mycology, IVRI, Izatnagar. After that host susceptibility study was done by spot test using double agar overlay method against 66 *E. coli* isolates obtained from the same division. Based on their lysis profiles, it was found that five phages had a wide range of lytic activity (22.72% to 33.33%) and the isolated bacteriophages showed clear plaques of varying diameters, ranging from small to large (1–2 mm). On morphological characterization via Transmission Electron Microscopy, the five isolated bacteriophages were identified as members of the order *Caudovirales*, out of which three phages belonged to the *Podoviridae* family with icosahedral capsid of diameters ranging from 32.87 nm to 91.16 nm and short non-contractile tails measuring about 7.51 nm to 39.23 nm.

The other two phages of the *Siphoviridae* family displayed icosahedral capsids measuring approximately 55 nm and long non-contractile tails ranging from 126.93 nm to 189.39 nm. Hence, *Caudovirales* members, with their broad lytic range, hold promise as potential bacteriophages. Exploring these phages and their products, like endolysins, can offer avenues for future research and applications as antimicrobials. Lytic bacteriophages can thus serve as alternatives to antibiotics, functioning not only in therapy but also as biocontrol agents and disinfectants.

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PP_II.4 Rosiglitazone suppresses BPXV replication by subverting host miR- 27a

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Buffalopox virus (BPXV) is a close variant of the vaccinia virus, the type-species of the genus Orthopoxvirus. BPXV causes pock-like lesions, primarily in domestic buffaloes (*Bubalus bubalis*), but cattle and humans can also be infected and hence is considered as a potential zoonotic threat. Recently, several host miRNAs have been identified to play crucial role in the replication of many viruses. We for the first time demonstrate that miR-27a, widely known as a direct target of peroxisome proliferator-activated receptor γ (PPAR-γ) supports BPXV replication and a small molecule rosiglitazone (an agonist of PPAR-γ) suppresses viral replication by subverting the proviral role of miR-27a to target PPAR-γ. Using virus step- specific assays, we show that rosiglitazone treatment results in reduced synthesis of viral DNA and proteins. Collectively we have demonstrated the antiviral efficacy of rosiglitazone against BPXV and propose that rosiglitazone could provide significant therapeutic value against BPXV.

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PP_II.5 Tobacco smoke induced oxidative stress in healthy indoor dogs

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Numerous health risks for non-smokers have been linked to passive smoking or ambient tobacco smoke exposure. Companion dogs, which are kept in close physical proximity to their owners, are exposed to tobacco smoke both directly through inhalation (second-hand smoke) and indirectly by ingestion (third-hand smoke) of smoke residues that have been contaminated on skin, clothes, and other surfaces. The purpose of this study was to assess how exposure to ambient tobacco smoke influenced healthy pet dogs' oxidative status.

In this investigation, twenty healthy canines were used. Dogs were divided into two groups: ETS (exposed to tobacco smoke; n = 10) and NETS (not exposed to tobacco smoke; n = 10) groups. Catalase, total antioxidant status, superoxide dismutase, thiobarbituric acid reactive compounds, reduce glutathione, iron, and copper levels were all assessed in the blood. A significant ($p< 0.01$) increment in the TBARS, GSH and Copper was observed in ETS group whereas the SOD and catalse activity and Iron concentration was significantly ($p<0.05$) in ETS than NETS group. Other serum biochemistry was altered non significantly except glucose that was observed significantly ($p<0.05$) lower in NETS group. The oxidant/antioxidant balance in companion dogs was shown to be disrupted by tobacco smoke exposure.

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POSTER PRESENTATION

PP_II.6 Site-directed ClpP mutation of *Leptospira* demonstrate unprecedented gain-of-function

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The pathogenic *Leptospira interrogans* is the causative agent of leptospirosis, an emerging disease with more than 1 million severe cases worldwide and 60,000 deaths annually. Although treatment with conventional antibiotics has saved millions of lives, excessive overuse and inappropriate prescribing have led to the emergence of antibiotic-resistant and persistent bacteria. Thus, to combat these, there is a need for new alternative targets in anti-microbial therapy. In this context, Caseinolytic proteases (Clp) have garnered considerable attention as targets for antibacterial action due to their direct relationship with bacterial viability and virulence. The complete genome sequence analysis of *L. interrogans* shows an array of genes belonging to the Clp system. The Clp system in leptospires is composed of two different ClpP isoforms (ClpP1 and ClpP2), forming an active hetero-tetradecameric assembly (LinClpP1P2). In this study, five mutants of LinClpP (LinClpP1^{E170D}, LinClpP1^{N172D}, LinClpP2^{IG_del}, LinClpP2S^{40AK41N}, LinClpP2^{Y62A}) targeting its critical hotspot residues were generated via site-directed mutagenesis. The functional activity of pure LinClpP mutant variants or its heterocomplex in association with a chaperone (LinClpX) or antibiotic acyldepsipeptide (ADEP1) was examined. The two mutants (LinClpP2^{S40AK41N} and LinClpP2^{Y62A}) displayed gain-of-function (GOF) in peptidase activity. The ADEP1-bound heterocomplex (LinClpP1P2^{S40AK41N} and LinClpP1P2^{Y62A}) showed 1.7 and 1.5-fold higher protease activity than ADEP1-bound LinClpP1P2. The deletion mutant (LinClpP2^{IG_del}) or its heterocomplex (LinClpP1P2^{IG_del}) displayed no activity. Similarly, the pure LinClpP1^{E170D} and LinClpP1^{N172D} could not cleave a model dipeptide. However, its heterocomplex (LinClpP1^{E170D}P2 and LinClpP1^{N172D}P2) showed 0.5-fold lower peptidase activity than the LinClpP1P2. Overall, this work provides an elaborative understanding of critical hotspot residues for unprecedented LinClpP activation and will pave several aspects of new antibacterial targets.

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PP_II.7 Deletion of *msr*s enhances sensitivity of *Ssalmonella* Typhimurium to oxidants

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Salmonella encounters various reactive oxygen species during host infection. Oxidative stress leads to the oxidation of methionine (Met) residues, resulting in the formation of methionine sulfoxide (Met-SO), disrupting protein function. Methionine sulfoxide reductase (Msr) catalyzes the reduction of Met-SO (free/protein-bound or '*R*'/'*S*' form) to Met. Therefore, Mrs(s) contributes to mitigating the adverse effects of oxidative stress. *Salmonella* Typhimurium possesses five Msrs (four in cytoplasm and one in periplasm). Here, we compared the susceptibility of *Salmonella* Typhimurium and its isogenic Δ5*msr* mutant strain to different oxidants. The Δ5*msr* mutant strain showed increased susceptibility to HOCl, chloramine-T, and paraquat. Interestingly, the Δ5*msr* mutant strain showed resistance to exogenous H_2O_2 as compared to the wild type strain of *S.* Typhimurium.

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PP_II.8 Exploring antiproliferative effects of methanolic fruit extract of *Solanum xanthocarpum* L. On breast cancer cell line

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The drugs currently available to treat cancer have detrimental side effects along with stern limitations regarding efficiency, safety and cost which made it crucial to look for novel therapeutic approaches from natural products. In the current study, methanolic extract of *S. xanthocarpum* was investigated for phytochemical constituents, antioxidant potential and cytotoxic activities. Screening for phytochemicals confirmed the presence of alkaloids, flavonoids, phenols, proteins and amino acids, reducing sugars, terpenoids, tannins, volatile oils, betacyanins, carbohydrates and

coumarins, etc., which could be responsible for biological activities shown by the plant. The extract was examined for total phenolic compounds and was calculated to be 127.78 ± 3.547 mg of GAE/g of extract, whereas the total flavonoid content was calculated to be 318.06 ± 14.289 mg of QE/g of extract. Furthermore, the *in vitro* antioxidant activity of extract was evaluated using free radical scavenging assays (DPPH, $ABTS^{+}$), reducing power assay (FRAP) and phosphomolybdate assay (total antioxidant activity). The extract showed EC_{50} of 60.1 ± 0.88 µg/mL for DPPH (for ascorbic acid, it was 39.00 ± 0.33) and 392.29 ± 3.93 µg/mL (for ascorbic acid, it was 44.89 ± 0.5) for ABTS radical scavenging activities, respectively. The antioxidant activity increased in dose-dependent manner when measured through FRAP and phosphomolybdate assays. Additionally, the study employed LC-HRMS for the identification of secondary metabolites present in the *S. xanthocarpum* fruit extract. Through this analysis, 30 secondary metabolites belonging to phenol, flavonoid and terpenoid categories have been identified. Moreover, the cytotoxic effects were assessed on MDA-MB-231 breast cancer cell lines, and the IC_{50} was calculated to be 24.19 ± 0.56 µg/mL. The apoptotic effects of the extract was evaluated using AO/EB, DAPI, Hoechst 33342 and PI double staining. It depicted significant effects in inducing apoptosis in the treated cells that were presented with condensed and fragmented nuclei, apoptotic and necrotic bodies. Mitochondrial membrane potential was assessed by Rhodamine 123, and decreased membrane activity was observed in extract-treated cells, which was marked by decreased green fluorescence. ROS quantification was carried out by H_2-DCF-DA staining, and increased ROS was marked by increased fluorescence intensity in treated cells. These results suggested that *S. xanthocarpum* exhibited strong potential as a chemopreventive agent for cancer.

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PP_II.9 Application of artificial intelligence and machine learning for prediction of Immune active peptide epitopes for Lumpy Skin Sisease Virus (LSDV)

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The outbreak of Lumpy Skin Disease virus (LSDV) took a toll of 2.4 million cattle in previous years. The virus is classified under Genus Capripoxvirus. The protein sequences from pathogen were subjected to analysis for prediction of prophylactic immune active peptides for LSDV. The protein sequences of all 156 putative genes were retrieved from the NCBI and screened for Antigenicity and allergenicity by VaxiJen v.2.0 and AllerTOP v. 2.0 respectively. Then subjected to physiochemical properties analysis by using Protparam tool. The selected fifty-one protein sequence screened for epitopes B-cell and T-cell (MHC-I and MHC-II) using IEDB server. The predicted epitopes also subjected for Antigenicity and allergenicity analysis. It was found that 52 B-cell epitopes and 299 T-cell epitopes were found to be antigenic and non-allergen. Toxicity analysis was done using ToxinPred tool. Water solubility was determined using peptide property calculator PepCalc.com. MHC I and II immunogenicity was determined using IEDB. Around 139 T-cell epitopes were found to be immunogenic. Prediction of IFN- γ cytokine secretion by immunogenic epitope was done using IFNepitope webserver to identify IFN-gamma inducing MHC class II binding peptides. Eventually, 89 T-cell and 47 B-cell predicted epitopes were clustered based on sequence identity.

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PP_II.10 Alpha synuclein (α syn) interacting with transmembrane domain of P2RX7

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Alpha synuclein (yn) is an intrinsically disordered protein (IDP), mostly found in presynaptic terminals of neurons and responsible for maintaining the synaptic transmission, neuronal differentiation, apoptosis and regulates dopamine synthesis. Mutations of Syn and their altered level cause neural degeneration and other associated diseases such as Parkinson's and Alzhiemer's diseases. According to recent studies, Syn can interact with the purinergic receptor P2RX7, inducing receptor activation and pore dilatation. However, it is unknown how exactly Syn interacts with P2RX7 and what potential binding sites there may be. We used a combinatorial strategy in our current study to answer these problems, utilizing docking, structural analysis, molecular modeling, and data curations. Using MODELLER software, we first rebuilt the human P2RX7 3D structure from the rat P2RX7 crystal structure (PDB ID 6u9W and PDB ID 6U9V). Later, using the panel of molecular docking tools and multiple strategies, finally predicted the docking of Syn (1-97aa) over P2RX7. We found that the N-terminal domain of Syn is amphipathic and interacts with the transmembrane

domain (TMD) of P2RX7. The Residue Interaction Network Generator (RING) web server was used to conduct the strict cut off analysis of the intermolecular interactions (ionic interaction: 4Å, hydrogen bond: 3.5Å, Van der Waals: 0.5Å, stacking: 6.5Å.-cation: 5Å, and disulphide bond: 2.5Å). The results indicate that the interactions between them are primarily driven by Van Der Waals forces, followed by hydrogen bond and one salt bridge interactions (between K34 in Syn and D9 of P2RX7). Our results are still preliminary, and to finally identify the molecular determinants of the Syn and P2RX7 interactions, more confirmation will be required through in-depth structure analysis combined with MD simulation, protein structure network formalism, and free energy based analysis in future.

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PP_II.11 Comparative computational analysis of immunogenic epitopes in the phospholipase A2 of Indian snake venom

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Snakebite envenomation (SBE) referred as a neglected tropical disease (NTD) by WHO has been reported to cause thousands of humans and animal death in tropical countries like India. However, the only effective treatment available till now is the animal sera derived polyvalent antivenoms. The Indian snake (major four families) venom component majorly found to consist of proteins belonging to family phospholipase A2. The present study reflects computational approaches to identify B- and T-cell epitopes for the phospholipase A2 proteins of the venom component of Indian Snakes. The nucleotide and amino acid sequences of serpentine phospholipase A2s were retrieved from the NCBI server followed by comparative sequence analysis with those of human, rat, large and small ruminants. Further, in sico analysis was conducted for analysis of variability, relative solvent accessibility, and three-dimensional structures prediction for all the retrieved sequences and the variations were observed. The molecular docking was carried out with different possible ligands using HADDOCK web server followed by molecular dynamics simulation. T cell and B cell epitopes were analysed by Epitope Prediction and Analysis Tools. Top Six linear B-cell epitopes, four discontinuous B-cell epitopes, top five T-cell epitopes have been predicted for genomic protein. Sequence variation has been observed in the epitomic components of different snake species. These results can help us to understand the potentially strong immunoactive component of venomic protein in different snake species. The identified B-cell and T-cell epitopes will be helpful in designing potent epitope -based diagnostics and vaccines against snake envenomation.

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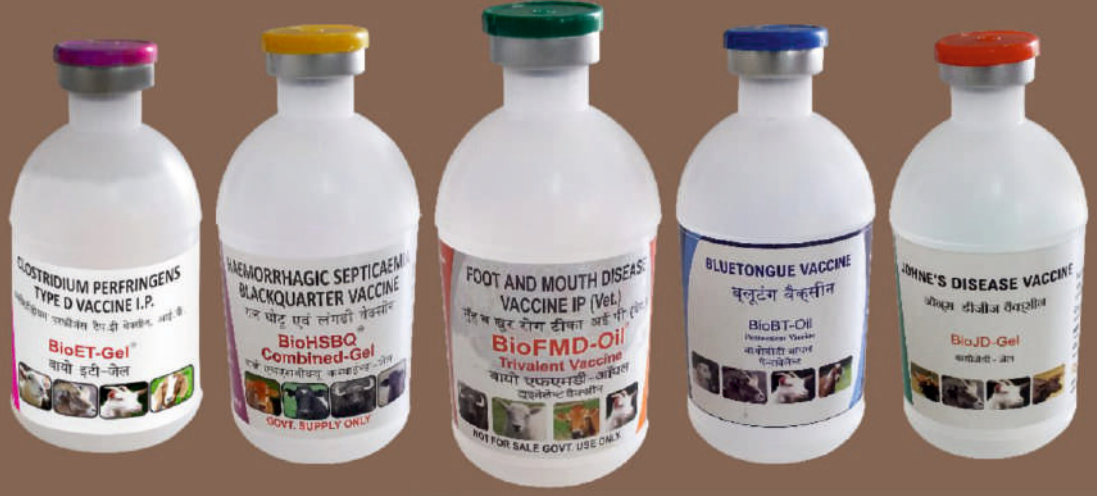

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Technical Session - III

Comprehending one health through host pathogen interaction

LEAD PAPER

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Dr Sachinandan De is an Animal Biotechnologist and has published more than 150 international peer-reviewed publications. He has developed five DNA and PCR-based technologies for Dairy and Animal Sciences and he has been awarded with two Indian patents. He is recipients of Fellow of National Academy of Agricultural Sciences (NAAS), National Academy of Veterinary Sciences (NAAS), Indian Society for Veterinary Immunology and Biotechnology (ISVIB), and National Academy of Dairy Sciences (NADS). He is also conferred with Lifetime Achievement Award (2022) by Society for Bioinformatics and Biological Sciences (SBBS). He was visiting scientist at Functional Genomics Laboratory, Uppsala University, Uppsala, Sweden (2008) and at Washington State University, Pullman, USA (2003). Dr De has research interests are in the areas of Animal Biotechnology, Animal Genomics, DNA and PCR based Genomic Technologies, Ruminant Immunology, Anti-microbial Resistant (AMR) Bacterial pathogen and their mitigation.

The mastitis pathogen of the dairy food chain: A special reference to *E. coli*

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The composition of microbiomes in bovine mastitis may vary according to different forms of mastitis (clinical, subclinical, and recurrent). This diversity comprises both contagious udder pathogens including *Staphylococcus aureus, Streptococcus agalactiae, Streptococcus dysgalactiae, Mycoplasma spp.,* and *Corynebacterium bovis,* and more commonly environmental pathogens such as *Escherichia coli, Klebsiella pneumoniae, Klebsiella oxytoca, Enterobacter aerogenes, Streptococcus dysgalactiae, and Streptococcus uberis* etc. Escherichia coli commonly present in organic matter, including bedding and manure infects the mammary glands through environmental contact. Recent studies suggest that mastitic milk contains a significant amount of antibiotic-resistant *E. coli*, specifically extended-spectrum beta-lactamase-producing *E. coli*. Particularly, multidrug-resistant (MDR) clones (normally defined as those resistant to three or more drug classes) are of great concern. Pathogenic *E. coli* from different resistomes (the total collection of antibiotic resistance genes) act as a potential key factor in disease complication, recurrence, complete damage to the udder, and transmission of the pathogen to the human food chain via milk and milk due to product processing loop holes. Molecular epidemiological studies have contributed considerably to our understanding of sources, transmission routes, and prognosis for many bovine mastitis pathogens and to our understanding of mechanisms of host-adaptation and disease causation.

The genomic diversity of the indicator *Escherichia coli* in bovine mastitis is huge. We relate it dynamic genetic make up and related to the isolates' antibiotic resistance and possible transmission to human. *E. coli* isolates of mastitis origin have a surprisingly large pan-genome, which harbors virulence genes, and antibiotic resistance genes.. We have analysed more than 700 *E.coli* isolates of bovine originbased on genomic and phenotypic correlation for a better understanding of the pathogen and to find ways to mitigate them. The antimicrobial susceptibility testing of these *E.coli* isolates showed 100% of isolates resistant to oxacillin, along with resistant to Erythromycin, and Ciprofloxacin. The isolates were susceptible to chloramphenicol (100%), and gentamycin (~90.00%).. The presence of ESBL genes such as blaSHV, blaCTX-M-1, blaTEM, blaOXA-1, and AmpC CMY-2, was observed in ~70.00% of *E. coli* isolates. A total of ~20% to 40% of *E. coli* isolates were found as multi-drug resistant (MDR) (resistance to ≥3 classes of drugs). Among the three Pathogenicity Islands (HPI, SerU, and pks island), only High Pathogenicity Island (HPI) was detected in ~30% of *E. coli* isolates. The abundance of *E. coli* estimated in the clinical mastitis, demonstrated the considerable pathogenic potential in the outflow of a dairy processing plant. Using *E. coli* as an example, this study demonstrates the importance of investigating common commensal pathogens with modern bioinformatics and genomic analysis in order to estimate the extent of genomic variation and resistance determinants with their clinical relevance.

We also propose a point of care diagnostics for a rapid, sensitive, and precise method to identify microbial

DNA coming from veterinary clinical samples, such as milk of cows affected with mastitis. We developed an amplification-free visual assay for rapid and sensitive detection of specific DNA from Escherichia coli, particularly uidA gene encoding for beta-glucuronidase, isolated in milk of cows affected with mastitis, based on multiple gold nanoparticles (AuNPs) captured on a magnetic microbead surface, leading to plasmonic signal enhancement, thereby improving overall sensitivity of the assay. This test can be performed in 1–1.5 h after post template DNA preparation and can be visualized with naked eyes without the use of any expensive equipment. The visual assay detects a minimum microbial load of 102 CFU/µl and can identify bacterial DNA as low as 1 pg. This test provides a precise detection tool for *E. coli* in clinical samples, such as milk of cows affected with mastitis, as a quick and user-friendly molecular detection approach. The work directed towards mitigation of isolated ESBL *E. coli* of bovine origin includes isolating bacteriophages from sewage.. Host range determination of ESBL-producing *E. coli* phages was done for 3 phages. One phage showed lytic activity against ~50% *E. coli* isolates. Around 60% of the phages had been adsorbed within 5 minutes in the presence of MgCl2 and 90% within 20 minutes. A single-step growth assay was performed for the determination of the latent period and burst size of the phage. The single-step growth curve of Phage A1 showed that the latent period of Phage A1 is ~35 minutes. The rise period is around 40 minutes, with a burst size of about 130 pfu per infected cell.

LEAD PAPER

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Dr Premanshu Dandapat is a graduate of BCKV, West Bengal in 1995; with an MVSc in 1997 and PhD in 2002, from the ICAR-IVRI and MVM (Bio-Security) from Massey University, New Zealand. Dr. Dandapat started carrier as Veterinary Pathologist followed by Assistant Rinderpest Investigation Officer under ARD Department, Govt. of West Bengal; followed by 5 years as Assistant Commissioner, Department of Animal Husbandry, Dairying & Fisheries, Government of India executing formulation and implementation of national policies on control and containment of important animal diseases, strengthening of diagnostic network in the country and handled several international aided projects. Dr. Dandapat served at the ERS, ICAR-IVRI, Kolkata from 2010 to 2023 and carried out basic and applied research on various zoonotic diseases and AMR. He is mainly engaged in research on animal and zoonotic tuberculosis in collaboration with several national and international organizations.

Tackling zoonotic tuberculosis in India: Need for one-health approach

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Tuberculosis is a chronic granulomatous disease of animals and human that results from infection with pathogenic members of the *Mycobacterium tuberculosis* complex *(*MTBC). Almost all mammalian species are susceptible to tuberculosis. The primary pathogens infecting animals (domesticated and wild) include *M. bovis* (bovine tuberculosis), *M. caprae* (caprine tuberculosis), *M. microti, M. orygis* and *M. pinnipedii*. Human infection with any member of the MTBC may result in zoonotic infection. *M. tuberculosis* and *M. africanum* are primarily pathogens of humans, but these too are known to infect animals. This disease has an enormous impact on human health, especially the well-being of communities that rely on livestock for their livelihood, and on livestock due to high mortality, reduced milk and meat productivity. Despite a well-recognized risk associated with tuberculosis, zoonotic TB is poorly monitored and continues to be an unaddressed global problem.

Studies on bovine tuberculosis in India have started in early 20th century and was considered as a rare disease until 1916 when Nield Cook in 1902 reported otherwise after screening thousands of carcasses in abattoirs. Recently India has an estimated 21.8 million bTB infected cattle which makes it a country with one of the largest infected herds in the world. In 2020 alone, India accounted for 26% and 34% of world's TB prevalence and death in humans, respectively. The cases of infection caused by MTBC members other than *M. tuberculosis* have been reported in our country from time to time. The consumption of raw milk and contacts with TB patients were found to be important determinants of zoonotic TB in human. The recent reports on isolation of *M. orygis* from several human patients, primarily with extrapulmonary or disseminated forms of the disease, is probably the largest reported series of *M. orygis* cases in humans from South Asia. Earlier and even in recent past either *M. bovis* in human or *M. tuberculosis* in animals alone or mixed infection along with *M. tuberculosis* has been reported by several researchers that demonstrate the zoonosis as well as the reverse zoonosis of TB in Indian settings.

Increasing evidence of regional and geographical variation in distribution of MTBC species are reported amongst livestock species. While *M. bovis* appears to be the predominant cause of cattle tuberculosis in Africa and much of Western Europe, *M. caprae* is the major cause of TB in cattle in central European countries. In South Asia including India, there is increasing evidence of *M. tuberculosis, M. orygis*, and *M. caprae* causing TB in cattle. Although tuberculosis is having lots of zoonotic importance, it's public health consequences in India have so far been scarcely investigated and often neglected. India being a high burden country for TB and the reports of tuberculosis infection in cattle making the situation even more alarming. Initiatives for proper surveillance of tuberculosis both in human and animal population should be considered a priority, especially in endemic settings. Strong multi-disciplinary action is required in India to estimate the scale of animal tuberculosis burden and its transmission at human-animal interface. Finally, One Health approach may be implemented in both animal and human health sectors to attain the Nation's End TB goal.

INVITED PAPER

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Dr Manish obtained B.V.Sc. & A.H. from Govind Ballabh Pant University of Agriculture and Technology, Pantnagar and PhD from University of Maryland, College Park, USA. Then he worked as Postdoctoral fellow from 2010-2012. Subsequently he joined as Assistant Professor in the Department of Biosciences and Bioengineering, IIT Guwahati in 2012 and promoted to in 2017 and Professor in 2021. His research interests includes, Molecular interaction of host-pathogen-vector of infectious diseases, Gene expression analysis of Spirochete, Leptospira interrogans and Borrelia burgdorferi, Development of vaccine against an outer membrane protein of Leptospira interrogans and Borrelia burgdorferi. His few Achievements and Fellowships includes, Avrum Gudelsky Veterinary Graduate Student Award for exemplary graduate research in Veterinary Medicine, College Park, Maryland, USA, Graduate Research Assistantship, University of Maryland, College Park, USA and Junior Research Fellowship (JRF) of Indian Council of Agriculture Research, New Delhi, India. He is the member of several associations including, Indian Association of Veterinary Public Health Specialists (IAVPHS), Indian Society for Veterinary Medicine (ISVM), American Society for Microbiology (ASM).

Deciphering the *Leptospira*-host interaction to develop various diagnostics and intervention strategies

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Leptospirosis, a widespread zoonosis prevalent in tropical regions, results from infection with pathogenic and intermediate *Leptospira* species. Afflicting farmers, veterinarians, and those in close animal contact, it registers over a million annual cases, often underdiagnosed due to diverse symptoms from jaundice to renal failure. Our study on leptospiral hypothetical outer membrane proteins (OMPs) identified a few potent serodiagnostic markers (LIC13341, LIC20035, and LIC11966) that can be detected by MAT-positive sera of humans and animals naturally infected with *Leptospira*. Moreover, these surface proteins, act as adhesins and facilitate leptospiral attachment to host tissues. LIC20035, a catecholamine-modulated virulent factor exclusively of pathogenic *Leptospira*, exhibits differential expression in the presence of stress hormones epinephrine and norepinephrine. Remarkably, ErpY-like (OspE-related protein Y) lipoprotein encoded by *LIC11966* enables *Leptospira* to evade host complement regulation by acquiring complement regulators (Factor H and I), activating alternative pathways, and intervening in the membrane attack complex. ErpY-like also disrupts host plasma coagulation by binding to fibrinogen.

Another virulence factor of *Leptospira* that has been extensively studied by our group is the caseinolytic protease (Clp) system (LinClpP). Our study focuses on understanding the molecular mechanism of the LinClpP system in maintaining cellular protein homeostasis in pathogenic spirochete. The Clp system is an antimicrobial therapeutic target, where over-activation of LinClpP results in growth inhibition and death due to the degradation of vital spirochetal proteins. We demonstrate that the novel antibiotic acyldepsipeptide (ADEP) dysregulates the LinClpP proteostasis, and may be exploited in addressing the looming menace of antimicrobial resistance. Additionally, the inherent challenges in employing a conventional reverse genetic approach to study leptospirosis, have spurred our investigation into the endogenous CRISPR-Cas system in pathogenic *Leptospira*. *L. interrogans* encodes both CRISPR-Cas I-B and I-C systems. The CRISPR-Cas I-B interference machinery of *Leptospira* when overexpressed in *E. coli*, tends to assemble into the cascade complex and impart target plasmid interference. In the future perspective, we aim to harness the leptospiral CRISPR-Cas as an endogenous genome editing tool.

INVITED PAPER

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Dr Niraj K Singh has been working as Associate professor, College of Veterinary & Animal Sciences (Kishanganj), Bihar Animal Sciences University, Patna, since June 2023. Previously, he was working as Scientist in College of Animal Biotechnology, Guru Angad Dev Veterinary and Animal Sciences University (GADVASU), Ludhiana, during Oct 2011 to June 2023. Dr Singh received his B.V.Sc & AH from Maharashtra Animal & Fishery Sciences University (MAFSU), Nagpur and MVSc & Ph.D in Animal Biotechnology from ICAR-Indian Veterinary Research Institute (IVRI), Izatnagar. He has published 28 research papers in peer reviewed journals (all above NAAS rating 8), 2 review articles, 4 book chapters, 3 popular articles and 5 practical manuals. Recently, he has honoured by National Academy of Veterinary Sciences (NAVAS) as member fellow. His areas of work are molecular virology, vaccine development, diagnostic and *in silico* viral host pathogen interactions (Codon usage bias).

Codon deoptimization approach: A new window for viral vaccinology

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The genetic language of DNA to the twenty-letter language of amino acids in proteins is mediated via 64 triplets codons. Out of 64 codons, 61 encode 20 amino acids and 3 are stop signals (stop codons). Except for tryptophan and methionine, the genetic codes for remaining amino acids are degenerate due to more than one codon (synonymous codons) that can encode the same amino acid. Synonymous codons are not equally used or randomly, but some are repeatedly preferred over others, to code for a given amino acid. This phenomenon of preference for the use of a specific codon in the pool of synonymous codons is referred as codon usage bias (CUB). CUB functions as a code inside the genetic code or the second genetic code and is present in many organisms. Within a single gene as well as between functionally related genes, the biased frequency of synonymous codons (CUB) fluctuates. The causes for the existence of CUB in organisms are fascinating. Mutations especially in the second or third nucleotide of an existing codon of a protein-coding gene, that exchange one synonymous codon for another, do not change the amino acid specified by new, modified codons nor the peptide primary sequence. Such silent mutations during evolution without any functional consequences cause synonymous codon usage biases.

Mutational pressure and natural selection are two major factors determining CUP. In addition, dinucleotide frequencies, geographical distribution, and evolutionary processes also have some effect on CUP in many viral pathogens. As the study of CUP of viruses and their hosts reveals more information about overall survival, fitness, evasion from the host immune system, and evolution, the evaluation of CUP can be useful to evaluate the viral host-pathogen interactions study.

The CUP is also useful for the development of codon deoptimized viral vaccines. Many codon deoptimized live-attenuated viral vaccine has been developed in the last decade including vaccines against Influenza, enterovirus, respiratory syncytial virus, foot-and-mouth disease, etc. Codon deoptimized viruses containing suboptimal codon composition can be used as vaccines with the following advantages: (i) novel gene manufacturing technologies enable the rapid generation of codon deoptimized viruses (ii) codon deoptimized genes in vaccine candidates express identical protein/antigen, and (iii) as the large number of changes (mutation) introduced in codon deoptimized vaccine candidates, revert to virulent pathogen due to gradual nucleotide sequence mutations is improbable. Due to the above-mentioned facts, several research teams have been in the process of developing codon deoptimized live-attenuated vaccines for viral pathogens.

INVITED PAPER

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Dr. Ravindra, PhD (IVRI), Postdoc (Univ. Chicago, NIH, USA), is the Founding Dean at Xavier University School of Veterinary Medicine, Aruba Campus. As a Principal Investigator, he secured funding from prestigious organizations such as NIH, DBT, DST, CSIR, and RBI. His extensive publication history in top-tier journals, including Nature Communications and Nature Medicine, encompasses virology, molecular sciences, and nutraceutical-based supplements. Recognized with over fifty honors, including Fellow of the Society of Immunology and Immunopathology (FSIIP) and awards such as "Outstanding Scientist in Animal Biotechnology," "Best Postdoc Award," and "DBT Ramalingaswami Fellowship, Scientist, Young Scientist " Dr. Ravindra earned appreciation from Prime Minister Narendra Modi for his significant contributions to the country's COVID-19 vaccination program. Beyond academia, he is a successful entrepreneur and a marathon runner, exemplifying his diverse impact on science and society.

One-health approaches to tackle rising antimicrobial resistance (AMR)

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Abstract
Implementing effective measures to counter the growing threat of antimicrobial resistance (AMR) requires a comprehensive "One Health" approach. This strategy integrates efforts across human health, animal health, and environmental sectors, aiming to curb the misuse and overuse of antimicrobials. Collaborative engagement among healthcare professionals, veterinarians, and environmental experts, coupled with surveillance, monitoring, and public awareness initiatives, can forge robust strategies. This talk emphasizes the need for coordinated, multidisciplinary approaches to preserve antimicrobial efficacy as well as protecting public health, animal welfare, and the environment from AMR consequences. Furthermore, it explores innovations, research advancements, and the challenges hindering the implementation of One Health strategies.

ORAL PRESENTATION

OP_III.1 Unidirectional protein recruitment is essential for viral latency

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Kaposi's sarcoma associated herpesvirus (KSHV, HHV-8) is associated with several human malignancies. During latency the viral genomes reside in the nucleus of infected cells as large non-integrated plasmids, known as episomes. All KSHV infected cells express LANA, and LANA is essential for viral latency. LANA binding to the viral episomes is critical for both replication of the viral genomes during latency, and for tethering the viral episomes to the cell chromosomes during cell division. Directional recruitment of protein complexes is critical for proper function of many nuclear processes. To test for recruitment directionality between LANA and cellular proteins we directed LANA via catalytically inactive Cas9 (dCas9) to a repeat sequence to obtain easily detectable dots. Then, recruitment of nuclear proteins to these dots can be evaluated. Using this protein recruitment assay we found that LANA recruits its known interactors ORC2 and SIN3A. Interestingly, LANA was unable to recruit MeCP2, but MeCP2 recruited LANA. Both LANA and histone deacetylase 1 (HDAC1) interact with the transcriptional-repression domain (TRD) of MeCP2. Similar to LANA, HDAC1 was unable to recruit MeCP2. While heterochromatin protein 1 (HP1) which interacts with the N-terminal of MeCP2, was able to recruit MeCP2. Forcing MeCP2 dimerization via the tandem epitopes of SunTag, allows LANA to recruit MeCP2 in infected cells. We propose that available interacting domains force this recruitment directionality. Knock-down of MeCP2 by shRNA, as well as cells derived from Rett syndrome and express a mutant MeCP2 (T158M), dramatically reduced the ability of LANA to support viral latency. Therefore, this unidirectional recruitment of LANA by MeCP2 identified MeCP2 as a critical factor for KSHV viral maintenance.

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OP_III.2 Comparative Analysis of m6A Modifications in Peste des Petits Ruminants Virus (PPRV) RNA Reveals Strain-Specific Epitranscriptomic Profiles: Sungri-96 vs. Izatnagar-94

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This study aimed to investigate m6A modifications in Peste des Petits Ruminants Virus (PPRV) RNA, specifically comparing the Sungri-96 and Izatnagar-94 strains using a variety of analytical techniques. The confirmation of m6A presence in PPRV RNA via dot blot assays initiated comparative analyses between these strains. Notably, the Sungri-96 strain displayed lower m6A methylation levels, starkly contrasting the significantly higher levels observed in the Izatnagar-94 strain, consistently supported by ImageJ quantification. Furthermore, Northern blot analysis indicated m6A presence in viral RNA from infected cells but not in artificially transcribed RNA, suggesting a potential cell-specific nature of m6A modifications. The application of m6A-meRIP sequencing offered critical insights into the specific enrichment of m6A modifications within key viral genes, notably highlighting heightened m6A levels in the matrix (M) and fusion protein (F) genes. Additionally, quantification of total viral RNA concentration showcased substantial differences between the strains, with Sungri-96 exhibiting higher RNA levels compared to Izatnagar-94. High-Performance Liquid Chromatography (HPLC) analysis revealed contrasting m6A methylation levels: Sungri-96 displayed lower levels (3.83%) while Izatnagar-94 showed notably higher levels (21.49%). These findings collectively emphasize the distinct epitranscriptomic profiles between the Sungri-96 and Izatnagar-94 strains, indicating potential implications for viral behavior, replication, and interactions with host cells. Understanding these disparities in m6A modifications could provide valuable insights into viral pathogenesis and behavior. The identification of cell-specific m6A modifications in infected cells suggests a nuanced regulatory mechanism within the viral RNA. Furthermore, the specific enrichment of m6A in key viral genes suggests potential functional implications in viral replication and host responses. These results serve as a basis for further research into the functional consequences of these epitranscriptomic differences, offering prospects for the development of targeted interventions and therapeutic strategies to mitigate the impact of PPRV infections.

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ORAL PRESENTATION

OP_III.3 Molecular docking studies on NCR domain of conglutinin protein expressed from the blood and liver tissue of indigenous pantja goats as a marker of disease resistance

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The research pertaining to the elucidation of innate components of the immune system in livestock species is gaining pace. Innate immunity is an important arm of the immune system that helps in the recognition of various pathogen-associated molecular patterns (PAMP) through pattern recognition receptors (PRRs). Conglutinin is a calcium-dependent collagenous C-type lectin that acts as soluble PRR which helps in the recognition of pathogens. Initially recognized in bovines, Conglutinin is now recognized in other domestic and wild herbivores. Conglutinin is primarily synthesized in the liver. Its expression level can be directly correlated with the disease-resistance trait of the animals. Pantja is a newly registered goat breed recognized for its similarity with deer in their morphological characteristics and is commonly found in the hot and humid climate (Tarai region) of Uttarakhand. The Pantja goats are hardy and resistant to most of diseases and infections. The present study is aimed at the molecular investigation of the Conglutinin protein and its role in disease resistance in indigenous Pantja goats. The expression of Conglutinin was seen in liver tissue and blood and the full-length gene encoding Conglutinin (CGN1) was amplified. The neck and carbohydrate recognition domain (NCRD) of Conglutinin was cloned, sequenced, and analyzed for its structure. A change in single nucleotide and single amino acid at position 76 was found when compared with the sequence of another goat breed suggesting the uniqueness of Pantja in the context of disease resistance. On secondary structure prediction by Phyre2 server for protein modeling, prediction and analysis it was found that there were equal presence of alpha helix and beta sheets with only 1% disordered structure. The expression of Conglutinin was also reported in blood so the quantification in blood using qPCR can be a good indicative of immune status of the animal.

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OP_III.4 Detection of virulent determinant genes of *Pasteurella multocida* isolated from different animal species

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Pasteurella multocida is a Gram-negative bacterium responsible for acute diseases in various animal species which in turn leads to a great economic loss to farmers. Various host and pathogen factors results in outcome of infection. The important pathogen related factors includes capsules, lipopolysaccharides and various virulence associated genes. Therefore, the current work was undertaken to detect the virulence associated genes among *P. multocida* isolates. About 200 *P. multocida* cultures of VTCC repository were examined. The cultures did not grow on MacConkey's agar and showed positive reaction to catalase and oxidase test. The DNA of each bacterial culture was amplified using *P. multocida* specific PCR and capsular serotype specific PCR. The cultures were belonged to Cap A (115), B (70), D (5), E (3), F (7) serogroups of *P. multocida*. The detection of 21 virulence genes was carried out individually using PCR and their distribution was as follow *ompH* (99%), *exbD* (99%), *nanH* (99%), *sodA* (99%), *oma87* (98.5%), *exbB* (98.5%), *tadD* (98.5%), *ompA* (97%), *hsf-1* (96%), *pmHAS* (95%), *sodC* (92%), *fimA* (92%), *hsf-2* (93.5%), *plpB* (88%), *pfhA* (84%), *hgbA* (80.5%), *hgbB* (75%), *tbpA* (71%) and *ptfA* (65.5%). The *toxA* and *nanB* were absent among all the cultures under study. The distinct association between each virulence genes along with each capsular type was observed which implied some unique information like *tbpA* is mostly associated with serogroups A and B, *pmHAS* is mostly associated with serogroups A, D and F, *pfhA* is mostly found in serogroup B, D, E and F, *tad D, ompA, omph* and *oma87* are usually associated with all serogroups. Gene *ptfA* was found to be associated with serogroup D than any other serotypes, *fimA* was found to be associated with serogroups B, D and E.

Some of the virulence genes like *ompH, oma87, ExbD, ExbB, nanH, sodA, tadD* were present regularly with more than 95%, irrespective of capsular types which will be helpful in providing cross-protective immunity in future development of vaccine.

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OP_III.5 miRNA profiling of lumpy skin disease virus infected primary lamb testicle cells

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In this study, miRNA profiling of cells infected with the lumpy skin disease virus (LSDV) was conducted for the first time. As compared to the mock-infected cells, LSDV-infected primary lamb testicle (LT) cells revealed dysregulation of 64, 85, and 85 miRNAs at 12 hours post-infection (hpi), 48 hpi and 72 hpi, respectively. While some of these miRNAs were found to be specifically dysregulated at a particular time point following LSDV infection, others were commonly dysregulated across all three time points. The analysis of the differentially expressed miRNA-mRNA interaction networks, Gene ontology analysis of the predicted targets, and KEGG analysis of the highly enriched pathways revealed several cellular factors/pathways involved in protein/ion/enzyme binding, cell differentiation, movement of subcellular components, calcium reabsorption, aldosterone synthesis and secretion and melanogenesis. Some selected upregulated (oar-mir-379-5p, oar-let-7d, Chr10-18769, Chr2_5162 and oar-miR-493-5p) and downregulated (ChrX-33741, Chr3_8257 and Chr26_32680) miRNAs were further confirmed by quantitative real-time PCR. Besides understanding virus replication, virus-host interactions and disease pathogenesis, these miRNAs and their cellular targets may serve as biomarkers as well as novel targets for therapeutic intervention against LSDV.

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OP_III.6 Comparative transcriptomics approach for the identification of host response of different livestock to Delta and Omicron variants of SARS-CoV-2 using *ex-vivo* infection of lungs explant culture

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The causative agent, severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) has a zoonotic source and hence, reverse zoonosis increases the risk and rate of SARS-CoV-2 infection. Both experimental and natural infections of SARS-CoV-2 in different animal species provide useful information on viral host range and pathogenicity. In particular, transcriptome analysis can help us in generating new knowledge of intracellular signaling pathways that regulate the infection and pathogenesis of SARS-CoV-2, generating new information about its biology. As the pandemic continues to evolve, SARS-CoV-2 infection in animals will be expanding. Therefore, in this study we examined and compared the response of buffalo, sheep, goat, and pig to delta and omicron variants of SARS-CoV-2 by transcriptomics at three different time points i.e, 6hpi, 12hpi, and 24hpi by infecting lungs explants culture ex-vivo. We identified, total 2904, 197, 6821, and 777 DEGs (p.value <0.05. log2FC -1/+1) at 6hpi, 1153, 1313, 7536, and 1111 DEGs at 12hpi and 5823, 1818, 7007, and 844 DEGs at 24hpi, in buffalo, sheep, goat, and pig respectively with SARS CoV-2 delta variant infection. On the other hand with SARS CoV-2 omicron variant infection total 55, 1548, 7042, and 499 DEGs at 6hpi, 5719, 297, 6399, and 3718 DEGs at 12hpi, and 1590, 3205, 4854, and 733 DEGs at 24hpi were identified in buffalo, sheep, goat, and pig respectively. Delta virus induced more number of DEGs at 6 and 24hpi whereas the omicron virus induced more number of DEGs at 12hpi. It was observed that more number of pathways were activated after delta virus infection as compared to omicron. Following pathway analysis it was found that the differential regulation of genes associated with coronavirus-disease COVID-19 was stronger in sheep, goat, and buffalo, which are intermediately susceptible, as compared to pig which is reported to be resistant to SARS CoV-2. Also the gene count associated with various pathways were more in buffalo, sheep, and goat compared to pig. The pathways associated with cytokine-cytokine receptor interaction, metabolic pathways, Chemokine signaling pathway, MAPK signaling pathway, Viral protein interaction with cytokine and cytokine receptor, B cell receptor signaling pathway, etc were also found to be

activated however the extent of up regulation/down regulation was different for the different virus variants. This study has helped elucidate the molecular responses underlying susceptibility and resistance to SARS-CoV-2 in different livestock species.

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OP_III.7 *Salmonella* Typhimurium modulates lipid biogenesis in murine macrophages for its early survival and multiplication

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Salmonella Typhimurium (STM), a gram negative, facultative, zoonotically important intracellular anaerobe causes nontyphoidal salmonellosis (NTS) which leads to major economic loss not only in veterinary but also in medical field. Although different drugs have been tested against these bacteria, the emergence of antimicrobial resistance drives for searching of new therapeutics against STM. Lipid biogenesis is the accumulation of lipid droplets in host cells occurred, after bacterial infection for its survival and replication. So this study was carried out with objective of effect of *Salmonella* Typhimurium on lipid biogenesis in mouse macrophages. For this, mouse macrophages were infected with STM (MOI: 10:1) for 2 hrs, 6 hrs and 24 hrs. Further LDs from infected macrophages were quantified with Oil Red O and Nile Red staining. Visualization of LDs was performed by fluorescent and confocal microscopy. In addition, Real time PCR was done to evaluate the gene expression level of one important lipogenesis gene such as Fatty acid synthase (FaSn) and one vital lipolytic gene such as Adipose triglyceride lipase (ATGL). It was found that there was significant increase ($p \leq 0.01$) in lipid biogenesis at 6 hrs and significant decrease ($p \leq 0.01$) in lipid droplets accumulation at 24 hrs in wild *Salmonella* Typhimurium infected macrophages. FaSn transcript level was found significant increase at 6 hpi and then significant decrease at 24 hpi while the gene expression level of ATGL was found significant increase at 6 hrs which was further increased at 24 hrs in *Salmonella* infected macrophages than uninfected cells. Further, fluorescence intensity was observed higher at 6 hrs and lower at 24 hpi in *Salmonella* infected macrophages than uninfected cells. So this study can be concluded that lipid biogenesis in murine macrophages occurs in time dependant manner following *Salmonella* Typhimurim infection. Therefore, this pathway can be targeted for development of new therapeutics against *Salmonella* in nearest future.

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OP_III.8 Heme binding motifs (HBMS) in putative cytoskeletal heme binding proteins of *haemonchus contortus* (Putative-Cyto*Hc*HBPs)

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Haemonchus contortus is economically the most important parasite of the small ruminants causing billions of dollars of annual loss. Rapid and widespread emergence of drug resistance against all the known anthelmintics and non-availability of a viable vaccine against the parasite calls for urgent exploration of novel molecular targets against the parasite. As the parasite is a heme auxotroph, understanding the molecular mechanism of heme metabolism in the parasite may provide possible novel therapeutic and/or molecular targets against the parasite. To compensate the lack of *de novo* heme biosynthesis, the parasite must acquire heme from its soil-born nutrients in the free-living stage and from the host blood in the parasitic stage. Heme, however, is a double-edged sword. Although it is an essential prosthetic group in multiple proteins serving various crucial oxygenic and other biological processes, free heme is highly toxic owing to its idiosyncratic molecular structure. Therefore, the parasite must possess suitable protein molecules not only as transport and carrier molecules for its acquisition and distribution, but also for its detoxification, especially during the flooding of free heme during hematophagy. As an initial step to understand such molecules, we identified a total of 571 putative heme binding proteins of *H. contortus* (putative *Hc*HBPs) in its blood feeding stage using a coupled method of isolation by hemin-agarose chromatography and peptide mass finger printing. Interestingly, nearly three-fourth of the putative *Hc*HBPs proteins fall into defined groups of functional significance, including 31 (around 5.42%) cyctoskeletal and associated proteins. Although various mammalian cytoskeletal proteins have been found to bind heme for influencing heme metabolism and, conversely, their physiological function being modulated by heme binding to them, no such information is available for cytoskeletal

proteins in parasites, especially, in *H. contortus*. Therefore, an effort was made to understand the structural basis of heme binding by these putative cytoskeletal *Hc*HBPs and their functional significance using multiple bioinformatic approaches. While phylogenetic analysis clustered the putative Cyto*Hc*HBPs into five clusters of known functional significance [such as, tubulin β, Tubulin α, actin actin domain containing proteins (actin actin DCP), intermediate filament DCP and keratins), proteins with no sequence homology to each other were assigned a miscellaneous group for the convenience of analyses. Estimation of HBMs without considering their solvent accessibility revealed the domination of Y-based class VII (XXXXYXXXX) motif, followed by the H- based class V (XXXXHXXXX) and the C-based class I (XXXXCXXXX) motifs in all the groups of Putative-Cyto*Hc*HBPs. Computation of multiple protein sequence alignment using conserved domain and local sequence similarity information (COBALT) of the putative Cyto*Hc*HBPs revealed moderate to high conservation of the HBMs in the clustered paralogs of functional significance but, obviously not in the miscellaneous putative Cyto*Hc*HBPs. The paucity of C+H/Y- based Class II (XXXXCXXXH and XXXXCXXXY), CP-based class III (XXXXCPXXX), CP+H/Y- based class IV (XXXXCPXXH and XXXXCPXXY) HBMs, and heme responsive motifs (HRMs) XCPXXX and XCPXXH, and the absence of the contracted HRMs (CKCH and CXCH) in the putative CytoHcHBPs indicates towards their possible non-involvement in heme mediated gene regulation. In contrast to the general assessment of all possible HBMs, estimation of the solvent accessible transient HBMs, using the web-based SeqD-HBM algorithm along with Weighted Ensemble Solvent Accessibility (WESA) predictor tool, revealed the complete domination of H- based class V (XXXXHXXXX) HBMs on the surface landscape of all the groups of Putative-Cyto*Hc*HBPs and their high conservation in the clustered paralogs of tubulins, actin actin DCPs, keratins, and IF DCPs.

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OP_III.9 Fibrinogen binding ErpY-like protein of *Leptospira* interferes in the coagulation of host blood plasma by inhibiting fibrin polymerization.

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The ability of pathogenic *Leptospira* to survive and proliferate within a host is contingent upon its capacity to evade and counteract the host's immune response while disseminating to various host tissues. The conserved ErpY-like lipoprotein, which is displayed on the outer membrane surface of *Leptospira*, plays a multi-faceted role in enhancing the bacterium's pathogenicity. The recombinant form of this protein, known as rErpY-like protein, contributes to *Leptospira*'s virulence by interacting with various host factors, including host complement regulators. This interaction enables the bacterium to evade the host complement system, further augmenting its pathogenicity. Moreover, rErpY-like protein exhibits a strong binding affinity to soluble fibrinogen. The present study demonstrates that rErpY-like protein interferes with the clotting process in platelet-poor citrated plasma of bovine and human origin in a concentration-dependent manner. Supplementing 20% diluted bovine or human plasma with rErpY-like protein (1 - 4 µM) completely inhibits blood coagulation via the extrinsic, intrinsic, and common pathways. The rErpY-like protein significantly increases the time it takes for blood to start clotting, reduces the density of the blood clot, alters the viscoelastic properties of the blood clot (storage and loss modulus), and decreases the average rate of blood coagulation. The rErpY-like protein also significantly increases the Michaelis constant (K_m) value for thrombin-catalyzed fibrin formation from 3.2 ± 0.7 µM without rErpY-like to 19.7 ± 2.7 µM with rErpY-like, without affecting the maximum rate of fibrin formation (V_{max}). Interestingly, rErpY-like protein inhibits thrombin-catalyzed fibrin formation in a dose-dependent manner, with an half maximal inhibitory concentration (IC50) of 2 µM. These findings provide strong evidence for the anticoagulant activity of ErpY-like lipoprotein and its important role in leptospiral infection.

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OP_III.10 Role of Malate synthase in the survival of *Salmonella* Typhimurium against nutrient and oxidative stress conditions

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Malate synthase (MS) is a signatory enzyme of the glyoxylate shunt. We have previously observed Δ*ms* strain showed defective colonization in poultry spleen and liver. In current study we attempted to investigate the role of *ms* in the survival of *S.* Typhimurium under nutrient and oxidative stress conditions. We assessed the activity of *ms* promoter in a reconstituted system. Following exposure of stationary phase cells to HOCl, we observed 1.6 folds increase in β

galactosidase activity indicating upregulation of promoter activity o f *ms* under carbon starvation and oxidative stress conditions. By Western blot analysis, we observed about 20 fold increased expression of MS when *S.* Typhimurium was cultured in M9 minimal media with acetate in comparison to supplementation with glucose. We also observed 3.82 folds upregulation of *ms* in *S.* Typhimurium following incubation with neutrophils. Thus, our data clearly demonstrate that *ms* contributes to the survival of *S.* Typhimurium under oxidative stress conditions *in vitro* as well as in neutrophils. By OxyBlot analysis, higher degree of carbonylation was observed in oxidant exposed MS with respect to its pure form. This indicates the greater tendency of MS to be susceptible to oxidation. We exposed late stationary phase cells (deprived nutrients) of both WT and Δ*ms* grown in LB to varying concentrations of HOCl. Following two hours of incubation we observed that Δ*ms* was more than 35 and 10 folds more susceptible to 1.5 mM (p <0.001) and 3 mM (p <0.05) HOCl respectively. We evaluated the susceptibility of Δ*ms* in neutrophils and macrophages. The Δ*ms* strain was significantly (p < 0.01) more susceptible to neutrophil mediated killing. In macrophages, Δ*ms* strain showed approximately 2.72 and 2.2 folds decrease in invasion and replication as compared to WT after 2 and 24 hours post infection. Thus, our data demonstrate that MS contributes to the virulence of *S.* Typhimurium by aiding its survival under carbon starvation and oxidative stress conditions.

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OP_III.11 First report of Sero-molecular detection of *Bartonella henselae* infection among veterinarians in India and assessment of risk factors

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Zoonotic bartonellosis caused by *B. henselae* remains grossly underreported and underdiagnosed in warmer countries, including India. In this study, 532 clinical samples collected from 278 humans, including 70 Veterinarians, were screened for *B. henselae* infection by PCR targeting *ribC* gene of the pathogen and IFAT for anti-*B. henselae* IgG antibodies. Moreover, risk factors associated with human bartonellosis were also analyzed based on a questionnaire survey. On screening blood samples, 7.48% of samples tested positive for the pathogen in *ribC*-PCR, while 12.58% of serum/plasma samples tested positive in IFAT. Compared to the general public, veterinarians had a significantly higher risk of contracting bartonellosis. Among veterinarians, males were at higher risk (1.535 times) than females, and those who received cat scratches and tick/flea bites within a year were at significantly higher risk than unexposed people. Compared to healthy, those with disturbed liver functions showed higher seropositivity for bartonellosis. This study seems to be the first in India to assess *B. henselae* infection among veterinarians and analyze the associated risk factors. The observed high positivity of this hidden threat in humans has unravelled its public health significance. It warrants a realistic assessment of the prevalence and risk analysis of bartonellosis among high-risk groups in India, including endocarditis and hepatic peliosis cases, emphasizing the importance of a comprehensive "one health" approach in addressing this neglected zoonosis.

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OP_III.12 *Salmonella* Typhimurium increases glucose uptake in chicken derived macrophages

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*Salmonella*Typhimurium is a facultative anaerobe having the ability to remain in an anaerobic environment, which is required for pathogenesis and virulence in hosts. It has been shown that *Salmonella* grown anaerobically attaches itself to host cells and enters them more readily than *Salmonella* grown aerobically. There is a progressive drop in oxygen partial pressure along the stomach-rectum route. This suggests that for STM to survive in the lower intestine, such as the ileum and colon, it must use a variety of electron acceptors to change its metabolism from an aerobic to anaerobic state.Following an infection with wild type *Salmonella,*the relative glucose absorption by the chicken derived macrophages was assessed using the fluorescent glucose analog compound 2-NBDG. It was discovered that an increase in glucose absorption in infected cellsthat is 1.8 times than that of uninfected cells after two hours infection. However, the *Salmonella*infected cells began to exhibit a 20–30% reduction in glucose absorption after 3 hours of infection. That is 0.8 times the uninfected cells. This is due to the defence mechanism of the host system to supress the growth of bacteria within the chicken macrophage cells.

Keywords: *Salmonella*, 2-NBDG, oxygen, infection, macrophages

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ORAL PRESENTATION

OP_III.13 Trivial role of periplasmic methionine sulfoxide reductase (MsrP) in stress survival and virulence of *s.* Typhimurium

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Among others, Cys and Met residues are highly prone to oxidation. Repair of oxidized Met (Met-SO) residues to Met by methionine sulfoxide reductases (Msrs) play a key role in the infection process of bacterial pathogens including *S.* Typhimurium. As per the location, two types of Msrs, cytoplasmic and periplasmic are present in *S.* Typhimurium. Periplasmic Msr (MsrP) might play crucial role in defending the host generated oxidants. We constructed a Δ*msrP* mutant strain. Δ*msrP* (mutant strain) grew normally in *in-vitro* media. In comparison to *S.* Typhimurium, Δ*msrP* mutant strain showed mild sensitivity oxidants *in vitro* and almost similar levels of protein carbonyls (a marker of protein oxidation) as compared to *S.* Typhimurium strain. Interestingly, the Δ*msrP* mutant strain depicted mild defect in mice spleen and liver. Our data revealed a minor role of MsrP in defending oxidative stress and colonization of *S.* Typhimurium.

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OP_III.14 Repurposing of diflunisal in attenuating *Staphylococcus aureus* mastitis by reducing bacterial burden and inflammatory markers in mammary gland of mice

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Staphylococcus aureus is the one of most common causes of intramammary infection (IMI) in dairy cattle. The traditional antibiotic treatments are discouraged due to the possible development of drug resistant bacterial strains and antimicrobial residue in milk that need prolonged milk withdrawal period. In last few decades, the repurposing potential of various 'off-target' drugs for antimicrobial therapy has been explored in various infectious diseases. In the current study, we evaluated the efficacy of a U.S. Food and Drug Administration-approved, non-steroidal anti-inflammatory compound diflunisal (DIF) in limiting *S. aureus* mastitis in mouse model. In this study, intramammary inoculation of bovine mastitis origin S. aureus successfully established the IMI in mice. The therapeutic efficacy of DIF was evaluated on the basis of bacterial burden and histopathological changes of mammary tissues and anti-inflammatory efficacy was assessed by measuring the concentrations of myeloperoxidase (MPO), nitric oxide (NOx), NF-κB and expressions of inflammatory cytokines targeting NF-κB signaling pathway in *S. aureus* induced mastitis. The bacterial burden ($\log_{10}$CFU) in mammary tissues was markedly ($p<0.05$) lower in DIF group as compared to disease control group. The histopathology showed moderate fibrosis with infiltration of macrophages and neutrophils with comparatively lower inflammation and congestion in DIF group as compared to disease control group. The mean concentrations of MPO, NOx and NF-κB in mammary tissue homogenate of DIF group were significantly lower ($p < 0.05$) from disease control group. The DIF treatment attenuated ($p < 0.05$) the mRNA expressions of IL-1β, TNF-α and NF-κB in mammary tissues, while the level of IL-6, iNOS and TLR2 expressions in mammary tissue were unaffected ($p > 0.05$) by the DIF treatment when compared with disease control group. It is concluded that DIF may be potential therapeutic agents against mastitis; the DIF conferred protection against *S. aureus* IMI by reducing bacterial burden and inflammation potentially by attenuating the NF-κB signaling pathway. The findings of this study can be used as a starting point for the development of new medications as the adjunctive therapy for mastitis.

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POSTER PRESENTATION

PP_III.1 Alterations in anti-inflammatory and antioxidative biomarkers due to *in vitro* exposure of *Berberis aristata* in Concanavalin-A stimulated chicken splenocytes

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The global demand for poultry and poultry products has been steadily increasing, playing a crucial role in enhancing food and nutritional security. However, there are potential challenges related to chronic stress and inflammation, recognized as significant threats in the poultry industry. Phytogenics offer a diverse range of benefits, including antioxidant, immunomodulatory, anti-inflammatory and anti-microbial properties. Additionally, these exhibit anti-stress effect, enhance gut health and act as natural growth promoters. This shift towards phytogenic additives reflects a proactive approach to address and mitigate the adverse effects of stress and inflammation in poultry farming. *Berberis aristata* is a potential phytogenic from North Himalayan region, commonly known as 'Daruharidra'. In the present study aqueous root extract of *Berberis aristata* (BAE) was prepared and subjected to different biochemical tests and GCMS analyses to detect the presence of various phytoconstituents. BAE was further subjected to different *in vitro* antioxidative and anti-inflammatory assays. To evaluate immunomodulatory potential, lymphocytes proliferation assay (LPA) was conducted by exposing chicken lymphocytes to maximum non-cytotoxic dose of BAE. Alteration in expression of selected genes (IL-1β, IL-2, IL-4, IL-6, IL-10, iNOS, Nrf-2, NF-κB1) due to *in vitro* exposure of BAE was examined through qRT-PCR analysis in Concanavalin-A stimulated chicken splenocytes. Various *in vitro* assays exhibited significant antioxidant, anti-inflammatory and immunostimulatory potential of BAE. The expression level of IL-10, an anti-inflammatory cytokine was upregulated while pro-inflammatory cytokines showed significant downregulation. Increase in Nrf-2 is indicative of its antioxidant potential. Overall, the present study indicates potential antioxidative, immunostimulatory and anti-inflammatory properties of BAE. *Berberis aristata* could be used as a potential phytogenic to support overall health and help in management of different inflammatory conditions in poultry. However, suitable *in vivo* studies should be conducted along with identification of active ingredients in the plant bioresource.

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PP_III.2 Emergence of Coagulase-nagative *staphylococci* bearing penicillin and methicillin resistance-encoding genes isolated from bovine mastitis in North-24 Parganas District, West Bengal

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Coagulase-nagative *staphylococci* (CoNS) are regarded as emerging zoonotic pathogens and considered as the most predominant group of opportunistic pathogen causing human and animal infections. It is also associated with bovine intra-mammary infections worldwide. Similarly, human infection associated with methicillin resistant CoNS has been reported by several studies. Of note, there are limited published reports on penicillin and methicillin resistant CoNS as species level from bovine mastitis samples. Thus, the aim of the present study was to detect *blaZ mediated penicillin* and *mecA mediated methicilling resistance among* CoNS (n=48) isolated from bovine mastitis milk (subclinical and clinical, n=132) in eight randomly selected villages from North-24 Parganas district in West Bengal. Of 48 CoNS isolates, 30 were *Staphylococcus haemolyticus*, 10 were *Staphylococcus epidermidis* and rest 8 were *Staphylococcus scuiri*. All the isolates are confirmed based on PCR based detected of *soda* (for *S. haemolyticus*) *Rdr* (for *S. epidermidis*) and *Gap* (for *S. scuiri*) genes. The *blaZ* gene was detected in 60.4% (n=29) of all CoNS isolates whereas presence of *mecA* gene was detected 22.9% (n=11). All *mecA* carrying CoNS were also positive for *blaZ* gene. Further Antibiotic

sensitivity test was carried out *in vitro* for their sensitivity to ten different antibiotics revealed their absolute penicillin resistance. However, about 89.3% and 81.2% isolates were sensitive to Gentamicin and Enrofloxacin respectively. The methicillin resistance encoding gene (*mecA*) is located on mobile genetic elements -*Staphylococcal cassette chromosome mec* (*SCC mec*) which may be transferred among different species of *Staphylococci*. Therefore, carriage of penicillin and methicillin resistance-encoding genes by CoNS species in cattle indicates a significant public health threat. So, in view of "One Health" perspective, detailed molecular epidemiological studies are required to consider for mapping the spread of antimicrobial resistance of CoNS between livestock and community.

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PP_III.3 H3K27-me3 inhibition induces YTHDF2-mediated decay of m6A-marked SARS-CoV-2 transcripts

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Emerging evidence highlight the role of epigenetic modification in virus infection. In this study, inhibition of H3K27 methylation (H3K27-me3) by UNC1999 (H3K27 histone methyltransferase inhibitor) was shown to block SARS-CoV-2 replication, as evidenced by reduced levels of viral mRNA and protein, particularly the nucleocapsid (N) protein. The level of H3K27-me3 negatively correlated with N6-Methyladenosine (m6A) levels on SARS-CoV-2 mRNA during the course of viral life cycle. m6A modifications of SARS-CoV-2 RNA which were predominantly present in **s**ubgenomic "N" transcripts, promoted recruitment of YTHDF2 and decay of the viral transcripts. The transcriptional silencing of SARS-CoV-2 "N" transcripts by UNC1999 was further evident by the reduced number of viral factories in UNC1999-treated cells. The transcriptome analysis revealed a complex interplay among various epigenetic players (methylase/demethylase) involved in m6A modifications. Furthermore, no UNC1999-resistant SARS-CoV-2 mutants could be observed upon long-term sequential passage (P=50) of the virus in the presence of UNC1999. In conclusion, by integrating molecular virology, transcriptomics and functional analyses, we for the first time demonstrated that inhibition of H3K27-me3 induces m6A-mediated decay of SARS-CoV-2 transcripts, particularly the **s**ubgenomic N transcripts which are highly enriched in m6A sites. We propose that inhibition of methylation by UNC1999 may provide therapeutic effect against SARS-CoV-2 without inducing an antiviral drug-resistant phenotype.

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PP_III.4 p38-MAPK is prerequisite for the synthesis of SARS-CoV-2 protein

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The inhibition of p38 mitogen-activated protein kinase (p38-MAPK) by small molecule chemical inhibitors was previously shown to impair severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) replication, however, mechanisms underlying antiviral activity remains unexplored. In this study, reduced growth of SARS-CoV-2 in p38-α knockout Vero cells, together with enhanced viral yield in cells transfected with construct expressing p38α, suggested that p38-MAPK is essential for the propagation of SARS-CoV-2. The SARS-CoV-2 was also shown to induce phosphorylation (activation) of p38, at time when transcription/translational activities are considered to be at the peak levels. Further, we demonstrated that p38 supports viral RNA/protein synthesis without affecting viral attachment, entry, and budding in the target cells. In addition, we demonstrated that long-term culture of SARS-CoV-2 in the presence of p38 inhibitor SB203580 does not easily select resistant viral mutants. In conclusion, we provide mechanistic insights on the regulation of SARS-CoV-2 replication by p38 MAPK.

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POSTER PRESENTATION

PP_III.5 Sensitive to Antimicrobial Peptides A role against stress survival and virulence of *Salmonella enterica* serovar Typhimurium

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Innate immunity is the first line of defense against bacterial pathogens. For successful colonization in the host, bacterial pathogens must need to overcome innate immune responses. Antimicrobial peptides (AMPs) and oxidants constitute an important part of innate immunity. AMPs kill bacteria by forming pores in the inner membrane. In gastrointestinal tract lumen AMPs were freely synthesized, where enteric pathogens like *Salmonella* colonize. Thus *Salmonella* has evolved several resistance mechanisms to defend from the actions of AMPs. The multicomponent SapABCDE is one of such system that provides resistance against AMPs. We analyzed the role of *sapA* in *S.* Typhimurium against AMPS, neutrophils and virulence of *S.* Typhimurium in mice. To study the role of SapA, first, we have constructed *sapA* gene deletion strain (Δ*sapA*) in *S.* Typhimurium. Δ*sapA* strain showed hypersensitivity to insect AMPs such as melittin and mastoparan as compared to the WT strain of *S.* Typhimurium. Further, Δ*sapA* mutant showed more than hypersensitivity to neutrophils as compared to the WT strain of *S.* Typhimurium. Besides, Δ*sapA* strain showed attenuation in mice. Based on our study, it appears that *S.* Typhimurium stress survival and virulence depend on SapA.

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PP_III.6 Structural study of Fnr protein and its interaction with *narG* promoter of *Salmonella* Typhimurium

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Fumarate-nitrate-reductase regulator (Fnr), an iron-sulphur metallo-protein, is heterodimeric in its active form and has DNA-binding property. It senses the change in ambient oxygen tension in its microenvironment and modifies the activities of various genes and operons that are responsible for its pathogenicity and virulence by binding to their promoters in order to activate and deactivate them. One among them is - *NarX/L*, a two component regulatory system that regulates the expression of the *narGHJI* (nitrate-reductase A) operon. These activated regulators activate the downstream operons- *narGHI* and *napABC*. Therefore, for *Salmonella* to establish colonization in the gut, *NarGHI* reductase is crucial. It is reported that Fnr and NarL binds to -35 bases and -200 bases upstream of the *narGHI*'s 5'-initiation site, respectively and integration host factor interaction with -120 site is necessary for *narGHI* activation in case of *Escherichia coli*. However, not much study was conducted with respect to structure and role Fnr protein of *Salmonella* Typhimurium and its regulation. We cloned the *fnr* gene of *Salmonella* Typhimurium into the pRham N-His SUMO Kan vector, expressed and purified the protein by Ni-NTA affinity column chromatography. The secondary structures of the purified protein was studied by using Circular Dichroism-spectroscopy in different buffer systems with different pH and by bioinformatics tools. For Fnr protein interaction with *narG* promoter, we identified the *narG* promoter region using BDGP and BPROM web servers, and cloned the predicted promoter region into pUC19 vector replacing its *lac* promoter. The functionality of the promoter was confirmed by its ability to express beta-galactosidase required for blue white screening. Electrophoretic Mobility Shift Assay was also performed for the protein-promoter interaction using a modified detection system. We confirmed interaction between Fnr protein and *narG* promoter of *Salmonella* Typhimurium by the band shift of interacted complex compared to uninteracted promoter.

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POSTER PRESENTATION

PP_III.7 Cloning, expression and purification of transmembrane domain of P2RX7

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The purinergic receptor P2RX7 is activated by elevated extracellular ATP levels during chronic inflammation, which causes transmembrane pore dilatation and non-selective accumulation of multivalent cations, ultimately induces necrosis. However, constitutive expression of this receptor with low amplitudes is also required for cellular proliferation and immunity. P2RX7 is made up of three domains: cytoplasmic (CTD), transmembrane (TMD), and external (ETD). The TMD is a key component of the machinery that causes the pore to dilate when P2RX7 is activated; however, this domain has not yet been investigated to learn more about the process underlying the pore opening. Our present research work was aimed to clone and express the human P2RX7 transmembrane domains for downstream studies. The primer for partial human P2RX7 gene was designed and amplified from an already available full length recombinant P2RX7-pET32A plasmid, followed by the cloning of the TMDs in the pET32A vector using DH5-E Coli bacterial host. The rP2RX7-TMD protein containing 6X His tag was expressed in the BL21-E Coli bacterial host using 0.75mM IPTG for 6 hours and was later purified using the Ni-NTA purification column upon 8M urea lysis. The recombinant protein was confirmed by running in SDS-PAGE. Now in future, The purified P2RX7-TMD proteins will be used for downstream studies to understand the mechanistic reasons behind the pore dilation.

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PP_III.8 Dysregulated long non-coding RNAs (lncRNAs) influence immune response in PPRV infected/vaccinated sheep and goat

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Peste-des-petits ruminants virus causes PPR in small ruminants, a disease with high mortality and severe morbidity resulting in huge economic burden on farmers. It is an epitheliotropic as well as lymphotropic virus with transcendent immune evasion mechanisms. PPRV piggybacks on PBMC and travels across the bloodstream. Genome-wide expression studies have emerged as significant means to decipher host-virus interactions. Our previous findings have highlighted the differential expression of mRNAs and miRNAs in modulating immune signaling during PPRV infection. In this study, differential expression of long-noncoding RNA (lncRNA) across the PBMC subsets; T-helper cells ($CD4^+$), T-cytotoxic cells ($CD8^+$), monocytes ($CD14^+$), B-lymphocytes ($CD21^+$) and natural killer cells ($CD335^+$) cells on day 0 and day 5 of Sungri/96 vaccinated sheep and goats and in $CD4^+$, $CD8^+$, $CD14^+$ and $CD21^+$ subsets on day 0 and day 9 of goats infected with virulent PPRV strain Izatnagar/94 was evaluted. The number of differentially expressed lncRNAs (DElncRNA) was highest in the $CD21^+$ cells of vaccinated goats and $CD335^+$ cells of vaccinated sheep whilst the highest number of DElncRNA in infected goats was found in $CD14^+$ cells. Comparative analysis of cis-target immune process genes in vaccinated sheep and goat revealed enriched immune response in both the species on triggering different immune response pathways. On comparing the goat infected with goat vaccinated discrete clustering of vaccinated subsets as against the infected was observed. Functional annotation of the genes involved in immune response in both vaccinated and infected uncovered pathways involved in response to external biotic stimulus, defense processes, regulation of viral life cycle, immune system processes, etc. These results predict a clear-cut difference in immune response to PPRV infection and vaccination.

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POSTER PRESENTATION

PP_III.9 Analyses of nucleotide and codon usage pattern of Foot-and-mouth disease virus (FMDV) genome to elucidate information about molecular evolution of FMDV and adaptation to the host

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Foot-and-mouth disease (FMD) is a highly contagious disease of cattle and other cloven-hoofed animalscaused by FMD virus (FMDV), a small, non-enveloped virus that contains a single stranded positive-sense RNA genome of about 8500 nucleotides.Due to the redundancy of the genetic code each organism show preference for certain codons over othersa phenomenon termed as codon usage bias.Codon usage patterns an essential information in revealing evolutionary relationships between species as well as host-pathogen coevolution and adaptation of pathogens to specific hosts.The main forces that drive this bias from equal usage are mutational biasesdue to nucleotide compositional constraints and translational selection. In this study whole genome sequence of 73 FMDV isolates from India (Serotype O 31, serotype A 17 and Asia1 25) were downloaded from NCBI and analysed for different codon usage indices like base composition, synonymous codon usage (RSCU), effective number of codons (ENC), ENC–GC3 plots and codon adaptation index (CAI)to quantitate codon bias. Nucleotide content distribution and composition of FMDV genome reveals natural selection in shaping the codon usage patterns in FMDV as C + G at the synonymous codon third position (GC3%) is 1.8 times higher than AT3%. Analysis revealed that A3 ($r = 0.83$, $P < 0.01$), C3 ($r = 0.94$, $P < 0.01$), T3 ($r = 0.97$, $P < 0.01$), G3 ($r = 0.87$, $P < 0.01$) and GC3 ($r = 0.78$, $P < 0.01$), have significant positive correlations with the set of full-length gene sequences (A, C, T, G and GC) indicate nucleotide content influence FMDV codon usage patterns. These results validate that in addition to natural selection, nucleotide contents can also play a role in synonymous codon usage patterns.FMDV has higher ENC value (51.46±0.32) indicates a lower codon usage preference and lower gene expression. ENC–GC3 plots revealed in addition to the mutation pressure, translation selection also influences the codon usage bias of FMDV. CAI revealed selection pressure from hosts may influence the codon usage pattern of FMDV and the translation resources of cattle are more efficiently utilized by virus than sheep and pig. Correlation analysis between CAI and ENC revealed mutation pressure may be more preferred than translational selection in cattle than goat, sheep and pig. RSCU value indicates preference for G/C nucleotide over A/T nucleotide which impacts the codon usage for translation of viral proteins being enriched with G and C. This information is essential for codon optimization studies which involves replacing rare codons with frequent ones to enhance the translation efficiency of

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PP_III.10 Prevalence and molecular characterization of MAP in cattle and buffaloes in Haryana

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Chronic diarrhea and weakness have been recurrent issues in dairy breeds of buffaloes and cows, prompting suspicion of *Mycobacterium avium* subspecies *paratuberculosis (MAP)* infection. This study employed a multi-test regimen to ascertain the prevalence of MAP in the bovine population through blood, milk, and fecal samples. Screening tests included indigenous Enzyme-Linked Immunosorbent Assay (iELISA) and fecal microscopy, while confirmation tests comprised fecal culture and IS900 Polymerase Chain Reaction (PCR) as a model test regimen. Positive animals identified by PCR were further bio-typed using IS1311 PCR-RE to determine the MAP biotype. Fecal microscopy and fecal IS900 PCR revealed individual positivity rates of 27.1% and 4.34%, respectively, resulting in a combined positivity of 28.2%. Plasma ELISA and blood IS900 PCR indicated positivity in 43.9% and 3.03% of samples, yielding an overall positivity of 46.9%. MAP culture and fecal microscopy identified positivity in 9.60% and 25.9% of samples, contributing to an overall positivity of 33.6%. Furthermore, MAP culture and plasma ELISA detected positivity in 11.29% and 53.22% of samples, leading to a 58.06% overall positivity. Milk ELISA exhibited a higher positivity rate at 72.4% compared to plasma ELISA (41.3%). Fecal PCR (IS900) and blood PCR (IS900) detected positivity in 5.3% and 3.5% of samples, respectively, resulting in an 8.9% overall positivity. The study provided insights into the limitations of diagnostic tests, considering variable sensitivities, and the importance of a multiple-test regimen in screening and confirming cases of non-treatable diarrheic bovines suffering from MAP infection within the constraints of a one-time sampling opportunity.

POSTER PRESENTATION

PP_III.11 Antimicrobial potential of green synthesized silver nanoparticles against multi-drug resistant *Pseudomonas aeruginosa* and *Acinetobacter baumannii*

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The rising tide of antibiotic resistance in microbial pathogens has ignited the quest for novel antimicrobial solutions. The remarkable efficacy of metal nanoparticles as antimicrobial agents has thrust this field into the spotlight, paving the way for extensive exploration. In the present study, silver nanoparticles (AgNPs) synthesized by green chemistry using three plant extracts *viz.* guava leaf extract (GLE), peach leaf extract (PLE), and cannabis leaf extract (CLE) were evaluated for their antimicrobial potential against *Pseudomonas aeruginosa* (42 strains) and *Acinetobacter baumannii* (15 strains). All the strains under study were also subjected for their susceptibility to conventional antibiotics by disc diffusion method. AgNPs were successfully synthesized by green chemistry as indicated by spectrophotometric, FE-SEM and Zetasizer analysis. All the *P. aeruginosa* strains and *A. baumannii* strains under study were found to be multi-drug resistant (MDR). The AgNPs synthesized by cinnamon leaf extract (AgNP-CLE) were found to be most effective in terms of the diameter of the zone of inhibition produced. The minimal inhibitory concentration (MIC) of AgNPs for *P. aeruginosa* and *A. baumannii* was calculated to be 0.0625 mg/ml and 0.0312 mg/ml while the minimal bactericidal concentration (MBC) was 0.125 mg/ml and 0.0312 mg/ml, respectively. The AgNP-CLE were found to be non-toxic *in vitro* on Vero cells. The storage of the AgNPs for up to six months did not result in their aggregation as no significant difference was observed in their antibacterial potential. Formulation of an ointment containing AgNP-CLE produced an equivalent zone of inhibition similar to AgNP-CLE under *in vitro* testing. The green synthesized AgNPs can serve as a potential alternative to conventional antimicrobials for topical or local applications, especially against drug-resistant pathogens.

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Technical Session - IV

Augmenting animal health through new generation vaccines and diagnostics

LEAD PAPER

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Dr Naveen Kumar obtained his B.V.Sc. and M.V.Sc. degrees from College of Veterinary Sciences, Bikaner. He completed his Ph. D. in 2006 from CCS HAU Hisar while being a DAAD scholar at Friedrich Loeffler Institute, Insel-Riems, Germany. He worked as a postdoc at Emory University, Atlanta, USA from 2006-2011. Dr Naveen is a renowned virologist who has made some basic and fundamental contributions in the area of virus-host interactions and antiviral drug development, exemplified by high impact publications in top class Journals in the field of virology, which includes Journal of Virology, PNAS-USA, Antiviral Research, Clinical Microbiology Reviews, Molecular Biology and Evolution etc. He has published >100 papers with over 3200 citations and an "h" index of 31. He is the inventor of India's first lumpy skin disease vaccine. He also developed ANCOVAX- a vaccine to prevent coronavirus (SARS-CoV-2) infection (COVID-19) in animals. He is the recipient of Award of excellence in Agriculture Research, for development and commercialization of LSD vaccine; Commonwealth Professional Fellowship (UK); Postdoc fellowship (Emory University, Atlanta, USA); DAAD fellowship (DAAD Bonn, Germany) and Gold Medal in MVSc by RAU Bikaner. He is United Nations (WOAH/FAO)-designated expert on Lumpy Skin Disease and is the section Editor of "Virulence".

Vaccination against Lumpy Skin Disease: Current state of the art

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Lumpy skin disease (LSD) is a notifiable disease caused by the LSD virus (LSDV), which belongs to the *Capripoxvirus* genus under the family *Poxviridae*. The disease in cattle is characterized by fever, enlargement of lymph nodes, edematous swelling of the legs, and the development of multiple nodules on the body surface, including the lining of the respiratory and gastrointestinal tracts. The affected animals undergo a drastic reduction in milk production, pregnant cattle may abort, and bulls may become sterile. The recovered animals tend to have permanent damage to their skin, lowering the commercial value of their hide. The LSD results in high morbidity and mortality in cattle which serve as the principal host for LSDV infection. Since 2019, LSD has been reported in most Asian countries and has caused economic devastation to the livestock industry.

The current pool of available nucleotide sequences divides the LSDV into three major phylogenetically distinct lineages (clades), Clade 1.1 includes classical Neethling type field strains and are restricted to Africa. Clade 1.2 includes Kenyan/KSGP type field strains which are widely distributed in Asia and Africa including India. The majority of the recombinant LSDV strains are grouped under clade 2.5 which are predominantly circulating in Southeast Asia including Russia, China, Vietnam, Thiland and Taiwan. The minor populations of clade 2.1, 2.2, 23. and 2.4 is present in Russia and Kazakhstan. Previously it was assumed that these recombinant strains were emerged naturally in the field. However, later it was confirmed that this was the result of the vaccine spillover and generated due to mishandling of the multiple LSDV strains in the cell culture in laboratory in Kazakhstan, and later, via the vaccine, injected into the animals in Kazakhstan. From Kazakhstan, these recombinant vaccine-like strains spread to Russia and China. Nevertheless, natural recombination events in capripoxviruses have never been demonstrated. So far these recombinant strains have never been reported in India.

Irrespective of the nature of the LSDV strain used, homologous live-attenuated LSD vaccines prepared using a specific LSDV strain, provide a complete protection against other LSDV strains in cattle. Capripoxviruses which include LSDV, goatpox virus (GPV) and sheepox virus (SPV) are genetically quite similar and offers some degree of cross reactivity to each other. Whereas some of SPV/GPV vaccine strains provide partial protection, others do not provide any significant protection against LSD in cattle. Therefore, World Organization of Animal Health (WOAH) recommends that a particular heterologous LSD vaccine candidate must be independently evaluated before its use against LSD in cattle. In 2021, due to the unavailability of a proper (homologous) LSD vaccine, India authorized goatpox (heterologous) vaccine against LSDV in cattle. However, later, experimental trials and several evidences from field suggested that the goatpox vaccine (Uttarkashi strain) does not provide any significant protection against LSD in cattle. Therefore, India developed a homologous live-attenuated LSD vaccine (Lumpi-ProVac^{Ind}). The vaccine has been extensively tested in the field and has been found to be 100% safe and highly efficacious in cattle against LSD. The vaccine has been commercialized and is likely to replace the existing practice of vaccinating cattle with goatpox vaccine (heterologous vaccine).

INVITED PAPER

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Dr Aparna Chaudhari has research and teaching experience of about thirty years in the area of fish genetics and biotechnology at the ICAR-Central Institute of Fisheries Education, Mumbai (Deemed University). She has served as the Head of FGB Division from 2015 to 2022, and is currently in-charge of the PME Cell of the Institute. Her research interests include genetic engineering for various applications including vaccine and biosensor development, molecular interventions for captive maturation and morphotype manipulation of commercially important shrimp species, etc. Molecular genetics and functional genomics are also areas of her interest. Lately, her work is focused on identifying SNP and microsatellite markers associated with high body weight in the indigenous catfish species *Clarias magur* that is being selected for growth. She considers the WSSV DNA vaccine for shrimp developed in her lab as her most important contribution. At present, preclinical trials in rodents have been completed on the scaled up product produced in a 20 L fermenter, and the vaccine was found safe for use. The data has been submitted for regulatory clearance for field trials. She has led several multi institutional research collaborations, published over 75 research papers in peer reviewed journals, 4 book chapters and compiled several training manuals. She has also published two patents.

Integrating one-health concept into shrimp dna vaccine design and delivery

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Shrimp culturedominates global aquaculture with 9.4 million tonnesproduced in 2022 (FAO, 2023). With farmed shrimp production >0.9 million tonnes in 2022, India is the fourth major exporter, mainly exporting to USA, China, Japan, EU and South East Asia. Of the 166 crustaceans being cultivated globally, about 40 are being cultured in India, but since the last decade, the exotic decapod *Peneaus vannamei* constitutes 95% of the total production. The major threat to shrimp aquaculture is its vulnerability to pathogens, including parasites, fungi, bacteria, and viruses. Of these, the white spot syndrome virus (WSSV) that caused a global epidemic in mid-1990s is the most devastating in terms of quick spread and debilitating financial losses. Itreportedly causes about two-thirds of the annual economic loss, ~US$ 1 billion and a 10% loss to the shrimp industry globally. Losses in India amount to ~US$ 240.67 million. WSSV caused the shift from *Penaeus monodon* to *P. vannamei* as the favoured culture species, since the latter comes with specific pathogen free (SPF) certification. Nevertheless, cultured shrimp contract white spot or other viral diseases during grow-out. While it is easy to see that 'one-health' should be best achieved by keeping culture systems as close to natural habitats as possible, this strategy would fail to meet the growing demand for food. In this scenario, vaccination is considered a healthier alternative to therapy and is more economical. Shrimp being invertebrates lack the well-developed adaptive immunity and rely largely on innate immune mechanisms. Hence, vaccines dependent on host response are not very effective in shrimp. RNA interference/ antisense RNA approach that directly targets critical viral transcripts and prevents propagation *in vivo* is an attractive option. DNA vaccines designed to express RNAi molecules work in preventive and therapeutic modes. However, use of DNA molecules poses ecosystem biosafety concerns and here arises the need for a vaccine design and delivery strategy that ensures 'one-health'. The only DNA vaccine approved by USFDA for use in salmonids is Apex IHN that provides protection against the infectious hepatic necrosis virus. This vaccine is delivered by injection at least 60 days before harvest by which time it is almost cleared from the fish body. At CIFE, we have designed a shrimp WSV DNA vaccine that provides 70% protection against viral challenge. This talk will describe the strategy developed for biosafe delivery of plasmid DNA into shrimp, *in vivo* persistence and protection, decontamination of vaccine solution, investigations into possibilities of environmental transmission, preclinical trials of the up-scaled product in a mammalian model, and the requirements for regulatory approval of DNA vaccines.

INVITED PAPER

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Dr Sachin earned his BVSc & AH in 2004 from College of Veterinary and Animal Sciences, Parbhani, Maharashtra and MVSc in 2006 from IVRI, Izatnagar. He graduated from University of Maryland (UMD), USA in 2010. He joined as Assistant Professor in 2012 in the Department of Biosciences and Bioengineering, Indian Institute of Technology Guwahati. He was promoted to Associate Professor in 2016 and professor in 2021. His achievements & fellowships includes, F.M. Burnett Award from Indian Society for Veterinary Immunology and Biotechnology, ICMR- Dr. J.B. Srivastav Oration Award, NASI SCOPUS Young Scientist Award and Inspire faculty fellowship award, Department of Science and Technology, Govt. of India. He has published more than 165 publications with a total Impact Factor 654. He has supervised (and supervising) more than 20 PhD and 17 Masters Students. He is the recipient of many grants from various funding agencies.

Understanding the role of alpha-synuclein in the Japanese Encephalitis virus infection: A plausible biomarker

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Japanese encephalitis virus (JEV) stands as a prominent vector-borne zoonotic pathogen, displaying neurotropism and eliciting Parkinson's disease (PD)-like symptoms among most symptomatic survivors. A characteristic feature of PD is aggregation of mutated α-synuclein (α-syn) that damages the dopaminergic neurons. Considering this link between JEV-induced PD-like symptoms and α-syn pathogenesis, we explored the role of α-syn in JEV infectivity in neuronal cells. Our investigation revealed significant increase in endogenous α-syn expression in JEV-infected cells. Additionally, treatment with exogenous α-syn (Exoα-syn) led to a substantial reduction in JEV replication, suggesting its anti-JEV effect. Furthermore, Exoα-syn treatment led to the upregulation of superoxide dismutase 1 (SOD1) and reduction in reactive oxygen species (ROS). The results were validated by endogenous α-syn-silencing that decreased SOD1 level and raised ROS level in neuronal cells. Similarly, the SOD1 inhibition via LCS-1 also intensified ROS and JEV infection. Overall, our results suggest that α-syn exerts an anti-JEV effect by regulating protein involved in oxidative stress inside neuronal cells.

INVITED PAPER

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Dr Chetan Meshram is a senior scientist in the Division of Virus Research and Therapeutics at CDRI. He has 15 years of experience studying zoonotic, mosquito-borne, and respiratory viruses. His major expertise is in generating reverse genetics of RNA viruses, host-viral interaction to identify targets for therapeutics development, development of animal models for infectious disease, antiviral drug screening, development and evaluation of vaccine candidates, and development of diagnostic tools. His earlier appointments included staff scientist at the University of Alabama at Birmingham and Oklahoma State University, USA. At CDRI, his research focuses on developing a reverse genetics platform for JEV and chikungunya as a tool to develop high throughput and cost-effective antiviral screening platforms.

Reverse genetics approaches for the development of new vaccines against viral infections

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Viral diseases constitute a majority of infectious diseases in animals and humans. Animals act as intermediate or amplifying hosts for many viral infections in humans before spill-over events in humans. The SARS-CoV2 pandemic in the recent past and recurrent outbreaks of the Nipah virus present stark reminders for pandemic preparedness in the form of diagnostics, vaccines, and therapeutics. Reverse engineering of viral genomes to develop newer vaccine platforms provides an attractive alternative to conventional vaccine development technology. In my talk, I will discuss the strategies and methods to develop reverse genetic technology for RNA viruses using the Japanese Encephalitis virus as an example, exploitation of the RGS platform to attenuate viral pathogens by mutating viral genome, and use of RGS in studying viral pathogenesis or viral replication kinetics in vivo. In addition to vaccines, therapeutics development is essential in tackling widespread outbreaks or pandemics. One of the bottlenecks in developingsmall-molecule antivirals against many viral human and animal diseases is the unavailability of optimized, validated, and high-throughput functional antiviral screening assays. Viruses expressing reporter protein (GFP, Nluc, etc.) have been used to develop high-throughput, cost-efficient, and reliable in vitro and in vivo antiviral screening assays and paved the way in antiviral drug discovery efforts. I will discuss one example where chikungunya virus expressing GFP has been used to develop a flow cytometry-based chikungunya virus (CHIKV) infection inhibition assay to screen antiviral compounds to develop small molecule compounds against CHIKV infection. In conclusion, generating infectious clones using reverse genetics provides a promising platform for developing new vaccine candidates and high-throughput antiviral drug screening assays against viral diseases.

ORAL PRESENTATION

OP_IV.1 Evaluation of stimulatory responses of Infectious Bursal Disease immune-complex vaccine on chicken peripheral blood mononuclear cells (PBMCs)

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Vaccination is the prime protection strategy for the vast poultry sector of our country from economically important and immunosuppressive virus infections. Infectious Bursal Disease (IBD) is one of the major immunosuppressive virus infections of young chicken, inflicting high mortality and secondary complications. Though routine vaccination with live and killed vaccines is being followed, IBD outbreaks are regular in India. Hence other vaccine designs like, immune-complex based vaccines have been designed, so that chicks can be vaccinated at very early age even in presence of maternal antibodies and offers adequate protection. Immune complexes are reported to target antigen presenting cells, resulting in early maturation of B-cells. Hence the present study has been designed to explore the stimulatory effect of IBD immune-complex vaccine in chicken peripheral blood mononuclear cells (PBMCs), so that the effect of vaccine on lymphocyte proliferation can be evaluated. Three different IBD-Icx vaccines (VAC1, VAC2, and VAC3) were used at two dilutions (1X and 10X doses). The protection percentage of vaccines was evaluated by challenge test as 55% for VAC1, 85% for VAC2 and 100% for VAC3. For the stimulation test, anticoagulated blood was collected from healthy chicken, PBMCs isolated and cells were added to a 96-well tissue culture plate, with each well receiving 100µL of the PBMCs suspension at a concentration of $2x10^{6}$ cells/mL. For positive control, the cells in triplicate wells were stimulated with 100µL mitogen, Phytohemagglutinin-M (PHA-M; 15-20µg/mL) while negative control wells were added with RPMI-1640 growth medium. After 72hrs incubation cells were subjected for lymphocyte proliferation test. At 1X dose, SI of VAC1 and VAC2 were significantly reduced comparing to that of mitogen well (1.264±0.359) while VAC3 (1.35±0.012) showed SI comparable to that of mitogen group. However, the SI with 10X dose in different groups were more than the mitogen control with the values VAC1 (1.303±0.0359), VAC2 (1.4±0.0420) and VAC3 (1.921±0.138). The results indicate the strong stimulatory effect of Icx vaccines in PBMCs which is comparable to that of mitogen.

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OP_IV.2 Development of recombinant protein-based Chemiluminiscence assays for COVID-19 serodiagnosis

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The novel coronavirus (COVID-19) has spreaded worldwide, resulting in growing numbers of infected individuals and increased mortality numbers since late 2019. Diagnosis of COVID-19 involves laboratory tests. Laboratory-based molecular assays for detecting SARS–CoV-2 in respiratory specimens are the current reference standard for COVID-19 diagnosis, but serologic immunoassays are rapidly emerging. Studies suggest that the majority of patients develop antibody response in the second week after onset of symptom. Therefore, accurate sero-diagnosis using highly specific recombinant protein of SARS–CoV-2 for differentiation of COVID-19 from other corona virus infections prevailing in the same locality is imperative for proper treatment. Serologic tests may be more useful in patients or animals with later complications of disease, when Reverse transcription-PCR may be falsely negative due to a decrease in viral shedding with time. As a result, this test was proposed to identify SARS–CoV-2 seropositivity in dogs as a model animal spp. In cases where molecular testing was negative but there was a strong epidemiological relationship to COVID-19 infection, the test for antibody response in dogs may be useful in making a presumptive diagnosis of COVID-19 illness. Keeping in view the vulnerability of dogs to COVID-19, probable direct danger to human spread, and non-availability of any sero-diagnostic assay for animals there was an urgent need of a test that is cost-effective, simple, and suited for large-scale testing and surveillance. In this study work has been done for evaluation of applicability of truncated recombinant SARS Co-V2 S protein for detection of SARS Co V2 antibodies. For expression of multi-epitopic recombinant protein for development of Chemiluminiscence assays for COVID-19 Sero-diagnsois the S protein was divided in various epitopic regions. Region 2 from 868n to 1893n was our prime target for expression as it contains RBD (Receptor Binding Domain) along with small flanking regions on both N & C terminal end. The target gene was amplified from field isolate of

Delta SARS CoV-2 with the primers with NcoI and XhoI sitesand was cloned for Prokaryotic expression in pET32 A vector. Fusion protein of 37.926 KDa + 20.6 kDa (Tag in pET32A) 58.526 kDa i.e. ~59kDa size was expressed and confirmed by dot blot and western blot analysis. Chemiluminescence assay was standardised and cut-off value for the test is decided by the reading of the known negative samples. Checker board analysis for finalizing the concentration of protein, dilutions of primary and secondary antibodies was done. Chemiluminescence assay was done using recombinant protein with primary antibody dilution at 1:2580 and secondary antibody dilution at 1:16000. 12 out of 89 samples were found to be positive by Chemiluminescence assay. The inter-assay %CV was 9.3% while intra-assay %CV was 4.45% and is in the acceptable limit. The developed test can be used for seromonitoring of the canine samples for presence of SARS CoV 2 antibodies.

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OP_IV.3 Calcium phosphate particles coated bio-mineralized FMD virus for the generation of thermostable vaccine candidate

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The crucial component of FMDV vaccine is the intact viral antigen (146S particles), and any degradation of the intact viral antigen would lead to a significant loss in the potency of the FMDV-vaccine. Since the 146S particles are thermolabile in nature, in important component of FMDV-vaccination programme is the maintenance of continuous and fault-less cold chain of the vaccine from the site of production until the end use. However, maintenance of fault-less cold chain is not so easy task owing to the lack of resources and logistics. Therefore, thermostability of FMDV vaccine is essentially required to tackle the need of fault-less/continuous cold-chain requirement. Among the different biogenic mineral Calcium Phosphate (CaP), an important component of bones & teeth, is of interest because of its unique biocompatibility and adjuvant properties. Recently, it has been shown that CaP can be introduced on the surfaces of some viruses in the presence of high concentration of calcium ions. Therefore, it has been hypothesized that CaP coated FMDV antigen can provide enhanced thermostability to the FMD vaccine, and hence, such possibility was explored in the current research endeavour. The sequence-encoding the calcium-binding peptide (CbP) was introduced into the capsid coding region of FMDV serotype O vaccine strain. The genetically defined recombinant FMDV serotype O encoding the CbP was rescued in cell culture through reverse genetics approach. The recombinant virus was characterised through 2D-MNT assay and sequencing of the capsid coding region. Both the recombinant CbP-tagged and parental FMDV were grown in cell culture and inactivated using BEI. The CbP-tagged inactivated antigen was coated with CaP microparticles and coated antigen was incubated at 37°C for 3-days in parallel with non-coated parental FMDV serotype O antigen. After incubation at 37°C the amount of left-over antigen was determined by antigen-ELISA. From the initial analyses, it could be concluded that the CaP coat could provide an advantage of thermostability to the FMDV serotype O.

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OP_IV.4 Validation of electrochemical biosensor based handheld device for the measurement of blood glucose in lactating buffaloes *(Bubalus bubalis)*

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The experiment was conducted on 56 healthy lactating Murrah buffaloes, irrespective of their lactation number and stage. Blood samples were collected in an anticoagulant-added sodium fluoride bulb and in clot activator plain tubes by jugular venipuncture. Also, glucose concentration was measured by a handheld device by puncturing the small branches of the ear vein and the results were recorded. The estimation of blood glucose was done by standard laboratory enzymatic method (GOD-POD) immediately after separation of plasma and serum on an automated Falcon (ARK Diagnostic) chemistry. Intra-assay and inter-assay coefficient of variance was also calculated. Statistical Analysis**:** The One-way ANOVA followed by Duncan's Mean Range Test (DMRT) and linear regression analysis was carried out. All the analysis was carried out in the statistical R software. Results & Conclusions: There is no significant difference between the mean values of Plasma Glucose measured by the GOD-POD method and by the Biosensor. But, the regression analysis showed the blood glucose in plasma do not have a

significant correlation (15.94%, p=0.24) with that measured by biosensor. The glucose measured in serum is significantly less than in plasma and measured by the biosensor. The electrochemical handheld device method is minimally invasive, cost effective, but not accurate method for measurement of blood glucose in lactating buffaloes. It may give the approximate estimate of blood glucose levels which may help in diagnosis of hypoglycaemia and prognosis of Ketosis. As blood glucose levels are declining rapidly in blood serum, it is advisable to use fluoride-added preservatives anticoagulant tubes & not to use a serum for the estimation of blood glucose. Also, it is advisable to use branches of the ear vein instead of puncturing skin to measure blood glucose by glucometer in buffaloes. In future more sensitive and accurate species-specific glucometers can be manufactured and can be validated for use in buffaloes.

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OP_IV.5 Evaluation of the immune responses in buffaloes vaccinated with a live-attenuated Lumpy Skin Disease Vaccine (Lumpi-ProVac[ind])

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Since 2019, Lumpy Skin Disease (LSD) has suddenly spread in many Asian countries, including India. LSD primarily occurs in cattle. However, recent LSD outbreaks in India have also revealed significant morbidity and production losses in buffaloes. This has raised concerns about the role of buffaloes in the epidemiology and transmission of LSD and necessitates the inclusion of buffaloes in the mass vaccination program for the prevention and control of the disease in the country. There is no significant data on the immune response in buffaloes following vaccination with the LSD vaccine. In this study, we evaluated antibody- and cell-mediated immune responses following vaccination with a newly developed live-attenuated LSD vaccine (Lumpi-ProVac[Ind]). The detectable amount of anti-LSDV antibodies was observed at 1-2 months following vaccination, with a peak antibody titer at 3 months. Upon stimulation of the peripheral blood mononuclear cells (PBMCs) with the UV-inactivated LSDV antigen, there was a significant increase in CD8+ T cell counts in vaccinated animals as compared to the unvaccinated animals. Besides, vaccinated animals also showed a significant increase in IFN-γ levels upon antigenic stimulation of their PBMCs with LSDV antigen. In conclusion, the buffaloes also mount a potent antibody- and cell-mediated immune response following vaccination with Lumpi-ProVac[Ind].

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OP_IV.6 Deletion of ethanolamine utilization transcriptional regulator of *Salmonella* Typhimurium

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Salmonella Typhimurium (STM) is a food borne, facultative intracellular pathogen of zoonotic importance and a leading cause of acute gastroenteritis. Chemical and nutrient signalings are basic for all cellular processes, including interactions between the mammalian host and the gut microbiota. Ethanolamine (EA) is an essential component of all cell membranes and has great signaling activity within mammalian cells by modulating inflammatory responses and intestinal physiology. Ethanolamine can serve as a carbon and nitrogen source for bacteria in the host intestine as well as within epithelial cells wherein bacteria grow. EA is derived from the membrane phospholipid, phosphatidylethanolamine and it is particularly prevalent in the gastrointestinal tract because of erosion of epithelial cells, components of food and turnover of gut microbiota. Deletion of important anaerobic regulator fnr (Fumarate and nitrate reductase regulator) of STM resulted in significant upregulation of genes responsible for EA utilization in STM. Genes encoding for EA metabolism are clustered in the *eut* operon comprised of 17 genes that code for transport and catabolism of EA and expression of this operon is regulated by EA utilization transcriptional regulator

(EutR). Therefore, EutR is being targeted for its deletion from bacterial genome to compromise the bacterial metabolic pathways for its utilization. For this, 1.5 kb upstream and 1.5 kb downstream of EutR gene was amplified and one ampicillin resistance gene (800bp) was also amplified for positive selection and all these three PCR products were ligated as per Gibson assembly protocol. Ligated mixture was amplified and cloned in a suicide vector PDS132 that contains sucrase gene (Sac B). Presence of this plasmid makes bacteria sensitive to sucrose in LB agar. Recombinant vector was transformed in *E. coli*, and then electroporated in STM for removal of EutR gene through homologous recombination.

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OP_IV.7 Seamless genome editing in *Salmonella* Typhimurium: harnessing as9 and the lambda red recombinase system for enhanced efficiency

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The objective of this experiment was to assess the effectiveness of scarless genome editing in *Salmonella* Typhimurium (STM) using a combination of the CRISPR-Cas9 system and the lambda red recombinase system. Many existing genome editing techniques in *Salmonella* Typhimurium are known to be less efficient, time-consuming, resource-intensive, and result in scars at the edited genomic site. In our experiment, we aimed to enhance the efficiency of scarless gene editing by combining CRISPR-Cas9 and the lambda red recombinase system. For the current experiment, we first designed single-guide RNA (sgRNA) targeting our gene of interest, in this case, *arcA* in STM, using the CHOPCHOP software. Additionally, we utilized a plasmid that expresses both the Cas9 protein and the lambda red recombinase enzyme. The Cas9 protein was employed to introduce a double-stranded break, and homologous repair was facilitated by the lambda red recombinase enzyme. Furthermore, we cloned the sgRNA into the pTargetF plasmid via inverse PCR and used donor DNA containing homologous arms of 400 base pairs upstream and downstream of the *arcA* gene to facilitate homologous repair. After transforming the plasmids and donor DNA, we screened 64 colonies for mutants using colony PCR. Interestingly, all 64 colonies screened (100% efficiency) turned out to be knockout mutants of the *arcA* gene. In conclusion, the combination of the CRISPR-Cas9 and lambda red recombinase system demonstrated exceptional efficiency in scarless genome editing in STM. This approach proved to be highly efficient, minimally resource-intensive, and resulted in successful knockout mutants, highlighting its potential for streamlined genome editing in this context.

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OP_IV.8 Diagnostic evaluation of hepatic dysfunction in canine

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The present research work was done between September 2021 to April 2022 at Dr. I. P. Singh Veterinary Clinical Complex and Trauma Centre, College of Veterinary and Animal Sciences, G.B.P.U.A.&T, Pantnagar U.S. Nagar (Uttarakhand) with an aim to diagnose hepatic dysfunction in dogs presented in clinics. Eighteen dogs presented with varied symptoms such as fever, anorexia, vomition, emaciation, polydipsia, polyuria, dullness, diarrhoea, icterus and nervous signs indicating hepatic affections were selected irrespective of their age, breed and sex and were further subjected to haematological, biochemical, radiographic and ultrasonographic examination. Findings of haematological examination revealed significant decrease in Hb, PCV, TEC, platelets, lymphocytes whereas there was significant increase in neutrophils in dogs with hepatic dysfunction as compared to healthy dogs. Findings of biochemical parameter revealed significant increase in total bilirubin, ALT, AST, GGT, ALP whereas there was significant decrease in total protein, albumin, A: G ratio, blood glucose of dogs with hepatic dysfunction as compared to healthy dogs. Radiographic and ultrasonographic examinations unveiled abnormalities like ground glass appearance, hepatomegaly, and specific liver textures aiding in diagnosis of hepatic dysfunction. From this study it was estabilised that a combination of hematobiochemical studies, radiography, and ultrasonography gives

subtle understanding of the complicated nature of hepatic diseases in dogs, thereby enhancing veterinary diagnostic techniques and strengthening overall management strategies.

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OP_IV.9 Deletion of all *msrs* renders *Salmonella* Typhimurium susceptible to oxidative stress and abets its colonization in mice

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Salmonella is resilient to and survives host inflammatory response. To defend host-generated oxidants *Salmonella* makes use of primary antioxidants and protein repair enzymes. Proteins are most susceptible to oxidative damage. Methionine (Met) residues are highly prone to oxidation and get converted to Met sulfoxide (Met-SO). This compromises the protein functions and subsequently cellular survival. However, methionine sulfoxide reductases (Msrs) enhance cellular survival under stress conditions by reducing Met-SO to Met. *Salmonella* has five Msrs which reduce Met-SO (free/ protein bound), and 'R'/ 'S' types. In this study, we generated a pan *msr* gene deletion (Δ5*msr*) strain in *S.* Typhimurium. The Δ5*msr* mutant strain shows initial lag in *in vitro* growth. Further, the Δ5*msr* mutant strain shows high levels malondialdehyde (MDA), protein carbonyls, and protein aggregation. On the other hand, the Δ5*msr* mutant strain exhibits lower levels of free amines. Further, the Δ5*msr* mutant strain is highly susceptible to neutrophils and shows defective fitness in spleen and liver of mice. The results of current study suggest that the deletions of all *msrs* render *S.* Typhimurium highly prone to oxidative stress and attenuated virulence.

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OP_IV.10 MsrP-A secondary deterrent of oxidative stress in *Salmonella* Typhimuirum

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Periplasmic methionine sulfoxide reductase (MsrP) is a repair protein present in the periplasm of many gram negative bacterial periplasm including *Salmonella* Typhimurium (*S.* Typhimurium), the causative agent of major gastrointestinal diseases in human. MsrP is functionally similar to its cytoplasmic counterparts viz. MsrA, B, C and Bis C. Msrs play a critical role in oxidative stress survival of bacterial pathogens by virtue of its repairing property of oxidized methionine (Met-SO) residues when in contact with the host generated oxidants. Owing to its localization, we hypothesised MsrP might play a very important role in defending host generated oxidants. We conducted a series of *in vitro* and *in vivo* experiments, to assess the role of MsrP, if any, in the survival of *S.* Typhimurium against oxidative stress and colonization in poultry and mice. First, we generated an *msrP* gene deletion strain (Δ*msrP*) in *S.* Typhimurium. Following that we analyzed the growth pattern of WT and Δ*msrP* strains of *S.* Typhimurium in LB or M9 minimal essential media. No difference in growth was observed for both the strains in the tested media. Under exposure to HOCl, the Δ*msrP* strain was about 15 folds ($p < 0.05$) more susceptible to HOCl as compared to WT strain of *S.* Typhimurium., however, this effect was not evident in the rapidly dividing cells. Further, Δ*msrP* mutant strain did not show hypersusceptibility to macrophages and neutrophils. However, in live animal model experiments, moderate attenuation of Δ*msrP* mutant strain was observed in the spleen and liver of mice and poultry at early stages of infection. Therefore, the results of present investigation suggest that *msrP* plays a secondary role in the survival of *S.* Typhimurium under oxidative stress and in virulence.

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ORAL PRESENTATION

OP_IV.11 Optimization of droplet digital PCR for detection and quantification of rabies virus in cell culture

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A reverse transcriptase droplet digital PCR (RT-ddPCR) was optimized as an alternative method to fluorescent antibody test (FAT) for quantification of rabies virus in the cell culture.. Initially 55ºC annealing temperature of glycoprotein gene primers was optimized by RT-PCR as well as gradient RT-ddPCR. Subsequently, five 10 fold dilutions of rabies virus ranging from $10^{5.6}$ to $10^{1.6}$ fluorescent focus forming unit (FFU) /mL were tested by RT-ddPCR for determination of copy number in each dilution. An excellent correlation (r = 0.999) was observed between the FFU and copy number of glycoprotein gene of rabies virus. The copy numbers of glycoprotein gene determined by ddPCR were found 10-fold higher than the FAT titer. According to the results of the present study, for making one fluorescent focus forming unit (FFU) in the BHK-21 cells, at least 10 infectious rabies virus particles are required. Based on the results of the present study, RT-ddPCR can be used as an alternative method for quantification of rabies virus in cell culture with very high sensitivity

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OP_IV.12 Th1 cytokines were crucial for post-challenge protection of bovine calves immunized with radiation-attenuated live *Trypanosoma evansi*

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The unicellular eukaryote *Trypanosoma evansi* (Phylum Sarcomastigophora) infects many species of domestic and wild vertebrates. The slender leaf-like flagellate is transmitted by haematophagous flies. The infection results in a chronic debilitating disease, 'Surra', in livestock with high morbidity and mortality. We studied the cytokine response in experimental bovine calves following immunization with a radiation-attenuated *T. evansi* and challenged with the virulent parasites. Two to three-month-old male Vrindavani calves (n=4) were immunized with γ-ray attenuated *T. evansi*, horse isolate ($n = 6\times10^7$) through subcutaneous route on days 0, 21, and 60 and challenged with virulent *T. evansi*, horse isolate ($n = 1\times10^4$) on day 82. The animals were further challenged with *T. evansi*, dog isolate on day 112. A control group of unimmunized calves (n=4) were also challenged with the same number of parasites following the same schedule. The expression patterns of the IFN-γ, TNF-α, IL-2, IL-12, and IL-17 were studied by real-time PCR. The cDNA, used in the qPCR, was synthesized from the total RNA extracted from the PBMCs of the experimental calves at different time points. An upregulation of IFN-γ (6.80±0.5), TNF-α (8.81±1.46) and IL-12 (27.7±1.79) transcripts was noted on day 67 PI in the immunized calves, whereas the upregulation of IL-17 (2.81±0.98) and IL-2 (3.60±1.43) transcripts was noted on day 27. The immunized calves had significantly ($p<0.005$) high-level expression of all cytokine transcripts following the challenges than the controls. The immunized calves showed maximum upregulation of IFN-γ (3.06±1.1), TNF-α (8.13±1.6), IL-12 (1±0.4), IL-2 (0.75±0.8) and IL-17 (0.76±0.7) transcripts on day 90 PC I. The highest expression of IFN-γ (1.38±1), TNF-α (1.9±1.2), IL-12 (0.77±0.3) and IL-17 (6.2±1.3) in the control group was recorded on day 90 and IL-2 (0.99±1) on day 85. A downregulation of Th1 cytokines noted PC II in the immunized calves; the highest expression of IFN-γ (2.15±1.1) was recorded on day 114. The upregulation of IL-17 (1.08±1.2) was maximum among the cytokines on day 114. The higher upregulation of the Th1 cytokine transcripts in the immunized calves was crucial for protection.

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POSTER PRESENTATION

PP_IV.1 Polymerase chain reaction based diagnostic test using lSDV p32 gene

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The viral diseases have always posed challenge before researchers for their prompt diagnosis and treatment plan. One such viral disease Lumpy Skin Disease (LSD) of dairy animals attracted attention of several researchers due to its epidemic in the country of India and neighboring countries. The causative agent of this disease, Lumpy Skin Disease Virus (LSDV) comes under family Poxviridae, genus Capripox virus with other related viruses namely, Sheep Pox and Goat Pox virus. The prompt treatment plan could be initiated with early diagnosis of the disease. The clinical diagnosis of LSDV relies on ELISA, Indirect Fluorescent Antibody Test and Agar Gel Immuno Diffusion Assay. The virus confirmation can be performed by electron microscopy, virus isolation and Immunohistochemistry, isothermal gene amplification, Polymerase Chain Reaction (PCR). Since, PCR being most preferred method of disease diagnosis, OIE has recommended primers and protocol which can be followed in laboratory. Therefore, the present study was envisaged to investigate an alternate PCR based diagnostic strategy with improved sensitivity and specificity. The gene p32 has been selected in the studies, which codes for antigenic structural protein. The gene length of 969 bp has been utilized for designing of primers SB01 and SB02. The SB01 was found to be capable of screening Capripox viruses. The primer set SB02 was designed to amplify LSDV p32 gene, with the objective to differentiate LSDV with GPV and SPV. To validate the functionality of the developed method 24 samples were included in the study. At the annealing temperature of 66 °C, the primer set SB02 detected 18 scab samples as positive for LSDV and 2 skin scraping samples along with GPV, SPV and CPV negative. The method showed high sensitivity and specificity. The limit of detection (LOD) was observed to be 17.78 pg.

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PP_IV.2 Optimization of serological assay for diagnosis of Brucellosis in dogs

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Brucellosis in dogs is an important bacterial zoonotic disease which can spread rapidly in dogs kept in a confined environment. Canine brucellosis is primarily caused by infection with *B. canis*. Considering the significance of studying the occurrence of this underrecognized zoonosis, the study was designed to optimize Agar Gel immunodiffusion assay (AGID) for detection of *B. canis* infection, followed by comparing the same with a commercially available ELISA. The AGID assay was optimized by employing the hot saline extract (HSE) antigen from MEX-51 strain of *B. canis*. Varying concentrations of borate buffer and sodium azide were used to optimize the assay. The concentration of HSE antigen to be used in the test was standardized using hyperimmune serum raised against *B. melitensis* raised in rabbit. The central wells of the agarose gel were loaded with 30 µL of hyperimmune serum while the surrounding 6 wells were loaded with 30 µL each of two-fold serially diluted antigen. The plates were incubated at room temperature in a moist chamber and the precipitation line was examined after 24-48 hours. The concentration of the antigen at which clearest line was observed was taken as the standard. On screening 312 randomly collected canine sera samples, a seropositivity of 3.65% was observed by AGID, while the seropositivity was 9.55% by ELISA. The overall seropositivity calculated with the samples positive by at least one serological test was 12.08%. The concordance between AGID and ELISA was 89.04%. The relative sensitivity and specificity of AGID when compared with ELISA were 11.76% and 97.2%, respectively. The AGID assay had high specificity and can be employed as a screening assay to establish disease free status, however, the poor sensitivity is a deterrent is using the assay as a screening test to identify true positive infection.

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POSTER PRESENTATION

PP_IV.3 A comparison between imidazole gradient and pH gradient methods using IMAC for purification of recombinant Non-Structural 1 (NS1) protein of Japanese Encephalitis virus: A step towards development of robust diagnostics

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The higher pandemic potential of emerging and re-emerging zoonotic pathogens poses a significant challenge to public health; therefore, their timely diagnosis is very important. With the advancements in recombinant DNA technology, the utilization of specific recombinant proteins in the development of diagnostic tests offers a high level of diagnostic sensitivity and specificity. Additionally, the method used for protein purification substantially affects the purity, yield, and concentration of the recombinant protein obtained. Therefore, the present study was undertaken to compare two methods of recombinant protein elution, namely imidazole gradient and pH gradient, using Immobilized Metal Affinity Chromatography (IMAC) for protein purification. After successful expression and bulk production of the recombinant NS1 protein of the Japanese Encephalitis virus (JEV), it was purified by IMAC using the Imidazole gradient method and the pH gradient method for protein elution under denaturing conditions, followed by dialysis in decreasing concentrations of urea in PBS. The concentration of protein was measured using Bradford reagent. The rNS1 protein eluted by the imidazole gradient method had a concentration of 700 µg/ml, whereas with the pH gradient method it was 1 mg/ml. The storage life of recombinant protein was assessed by re-checking the protein concentration after 15 and 30 days of storage at -80ºC. The concentration of rNS1 protein eluted with the imidazole gradient was 700 µg/ml after 15 days of storage, as recorded previously, while it was reduced to 600 µg/ml from 1 mg/ml with the pH gradient method. Similarly, storage for one month further reduced the concentration of protein purified by the pH gradient method to 350 µg/ml, which revealed the low stability and shorter storage life of recombinant protein purified by the pH gradient method. Further studies are needed to know the exact reason behind the low stability of proteins purified using the pH gradient method. Albeit our study concludes that the stability of recombinant proteins after purification must be analysed for the development of robust diagnostics.

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PP_IV.4 Development of reverse transcription recombinase polymerase amplification (RT-RPA) assay for rapid detection of porcine *Sapelovirus*

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Porcine Sapelovirus infection have been confirmed in pigs worldwide, mostly asymptomatic, however it can also result in severe gastrointestinal, respiratory, neurological, or reproductive problems or failure, and considered as the emerging pathogen of porcine species. Recombinase polymerase amplification (RPA) is an isothermal technique which employs three enzymes for amplification and is noteworthy due to its simplicity and quick amplification. The reverse transcription recombinase polymerase amplification (RT-RPA) assay for detection of Sapelovirus was developed and optimized for field-based detection. The assay was developed by targeting 5´UTR region of PSV genome and was optimized for reaction time, temperature, primer and MgOAc concentration. The analytical sensitivity and specificity of assay was determined. The assay was evaluated on 85 porcine faecal samples collected from field. This assay was also optimized for visual dye-based detection format and lateral flow strips-based detection (in combination with probe). The developed assay works at constant temperature of 35°C for 20 minutes with forward primer concentration 20pm, reverse primer concentration 5pm and MgOAc concentration of 14mM. The assay was highly sensitive and detect up to 283 copies and 28 copies with PicoGreen dye. The developed assay indicate 25 samples positive out of 85 samples and showing positivity rate as 24.7%. In present study, the RT-RPA isothermal assay was developed for detection of PSV which is rapid, highly sensitive, specific, works at low, constant temperature and does not require any high-end instrument. The developed assay can be considered a potential diagnostics for pen-side testing of PSV.

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POSTER PRESENTATION

PP_IV.5 Characterization of genetically modified Foot and Mouth Disease Virus (FMDV) Asia1 mutated at the SAP domain of L-protease in BHK-21 cell

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Considering the economic importance of foot and mouth disease (FMD), India implements a biannual vaccination-based control program as the inactivated oil adjuvanted vaccine confers protective immunity for 6 months. Generating a stable avirulent but immunogenic FMD virus (FMDV) candidate is a prerequisite for developing attenuated FMDV vaccines with long-term immunity. Accordingly, we targeted the L-protease (L-pro), a non-structural protein of FMDV involved in the virulence and host range, through PCR site-directed mutagenesis of the FMDV pAsia1 clone. As mutant FMDV could not be rescued following complete or partial deletion of the L-pro, two pAsia1 mutant plasmid clones were constructed by deleting two adjacent amino acids of SAP domain of L-pro: SAP1 clone at position 55-56 isoleucine, arginine to serine, glycine, and clone and SAP2 clone by targeting position 57-58 glutamine, leucine to leucine, valine. Mutant viruses such as FMDV pAsia-SAP1 and FMDV pAsia-SAP2 were rescued following transfection; propagated till 20 passages and the genetic stability of the mutations were confirmed by nucleotide sequencing. The growth kinetics of the two mutant viruses were compared with that of parent FMDV Asia1 by end point dilution titration method, plaque assay, and virus copy number by real-time virus quantification. The mutant viruses showed increased virus titer of 0.1 to 0.3 $\log_{10}TCID_{50}mL$ compared with the parent FMDV Asia1 in the BHK21 cells. The plaque morphology of the mutants was comparable with that of parent virus. The copy number of FMDV pAsia-SAP1 and FMDV pAsia-SAP2 was higher than that of the parental virus at 16 hours post-infection. It is concluded that the mutations at the SAP marginally increased the replication rate of FMDV Asia1 *in vitro*. The reduction in the virulence needs to be tested *in vivo*.

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PP_IV.6 Cloning and expression of Lumpy Skin Disease Virus ORF154 for the development of diagnostic assay

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We recently developed and commercialized a live-attenuated vaccine candidate Lumpi-ProVac^{Ind} using a local LSDV strain (LSDV/Cattle/2019/India/Ranchi). The whole genome sequencing of vaccine strain revealed several mutations, including a major deletion of 801 bp in 5' inverted terminal repeat region (ITR). This unique molecular signature is aimed to develop a test for differentiation of vaccinated and infected animals The truncated fragments of ORF 154 were successfully PCR amplified, cloned in pET22b vector and expressed through heterologous *E. coli*. hosts. The purified products react with sera from infected animals and is under validation for differentiation of vaccine and field LSDV strains.

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PP_IV.7 Nucleic acid hybridization probe based assay for differentiation of wild type and glycoprotein E (gE) deleted Bovine herpesvirus-1

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Infectious bovine rhinotracheitis/Infectious pustular vulvovaginitis is a contagious disease affecting cattle of all ages caused by bovine herpes virus 1 (BoHV-1) and is controlled and prevented mainly by vaccination. Conventional vaccines protect cattle from IBR infection but affect the control program adversely as they also render seropositive status to the herd and vaccinated and infected animals cannot be distinguished. In such a scenario, marker vaccines are developed along with suitable companion diagnostic tests to differentiate infected from vaccinated animals (DIVA). A glycoprotein E (gE)-deleted infectious bovine rhinotracheitis (IBR) marker vaccine candidate has already been developed in our laboratory. Although allowing differentiation between naturally infected and vaccinated cattle, live gE-deleted marker vaccine virus is shed following vaccination and can establish latency similar to wild-type BoHV-1. The present study was designed to develop a nucleic acid hybridization probe-based assay that can be done routinely at the

farm level. Wild-type and gE-deleted Bovine herpesvirus-1 was propagated using a Bello cell bioreactor and characterized. The DNA isolated from the respective cell culture supernatant was employed for the assay. Colloidal gold nanoparticle solution was synthesized using the citrate reduction method and characterized by transmission electron microscopy and Zetasizer. Alkyl thiolated nucleic acid hybridization probes specific to the structural gene gE and non-structural gene DNA polymerase were designed, synthesized, and then conjugated on colloidal gold nanoparticles. The conditions for hybridization reactions were optimized as 5 µl of DNA isolated from infected cell culture supernatant 62ºC for DNA polymerase and 60ºC for gE probe. Under optimized reaction conditions, the AuNP-DNA polymerase probe gave a positive reaction (pink) for both wild-type and gE-deleted BoHV-1 and the AuNP-gE probe gave a positive reaction only for the wild-type BoHV-1. The AuNP-gE specific probe could successfully differentiate the wild type and gE-deleted BoHV-1.

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PP_IV.8 Development of a companion diagnostic indirect ELISA using prokaryotic expressed gE glycoprotein of bovine herpesvirus-1

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Infectious bovine rhinotracheitis (IBR), caused by bovine herpesvirus-1 (BoHV-1) is a WOAH notified disease of cattle and buffaloes causing economic loss to dairy industry due to severe reduction in milk production, reduced fertility and abortions. In a country like India, due to its socio-economic reason, the only alternative for the control of the disease is vaccination. Conventional vaccines protect cattle from IBR infection, at the same time the herd obtains a seropositive status as vaccinated and infected animals cannot be distinguished. This affects the control program adversely and restrict export of products as per WTO. To overcome these difficulties marker vaccines have been developed along with a companion diagnostic test to differentiate infected from vaccinated animals (DIVA). Marker vaccines against IBR are based on the deletion of glycoprotein E (gE) from the BoHV-1 genome and vaccination of cattle with these marker vaccines will not induce antibodies against gE while infection with the field virus will induce a positive antibody response against gE. In the present study, we developed an ELISA companion diagnostic test for a gE deleted IBR marker vaccine. The gE gene of BoHV-1 from an Indian isolate was codon-optimized for expression in *E. coli*, synthesized chemically and cloned into pUC57 vector. The recombinant pUC57-gE was characterized using nucleotide sequencing, restriction endonuclease digestion and PCR. The codon-optimized gE gene was PCR amplified and cloned into pET-SUMO-TA cloning and expression vector (Invitrogen). The recombinant plasmid was transformed into *E. coli* BL21 expression host and recombinant gE protein was expressed by inducing with IPTG. The expressed gE was characterized in SDS-PAGE and western blot by probing with IBR-specific hyper immune sera. The recombinant protein was expressed in bulk, purified using Ni-NTA affinity chromatography and its reactivity was checked in indirect ELISA using IBR-specific antibody. The purified gE was coated in Maxisorb ELISA plates (Nunc) from 50 ng to 1000 ng per well and positive to negative (P/N) ratio was determined using negative and positive IBR-specific sera with different dilutions (from 1:20 to 1:640). The coating antigen concentration (100 ng/well) showing P/N ratio more than 2 was selected for considered positive. The developed assay was applied to known IBR positive and negative serum samples. There was good correlation in ELISA reactivity with IBR positive and negative sera. The developed assay has potential to be used as companion diagnostic test along with IBR marker vaccine in cattle.

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POSTER PRESENTATION

PP_IV.9 Evaluation of sugar stabilizers on the thermostability of inactivated Foot and Mouth Disease Virus serotype O by PaSTRy assay

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Foot-and-mouth disease (FMD) is an acute, infectious disease of cloven-footed animals causing significant economic losses of INR 20000 crores annually. Biannual vaccination is employed in India to control FMD, which offers immunity for a period of 6 months. This inactivated foot-and-mouth disease (FMD) vaccine includes entire viral particles, also known as 146S antigen which is thermolabile, and requires stringent cold storage during manufacturing and transportation. It might be possible to construct a thermostable FMD vaccination by using physical stabilizing agents to stabilize 146S antigen and circumvent this bottleneck. In the present study, sugar stabilizers namely sucrose, sorbitol, and glycerol were used at different concentrations (5%, 10%, 15%, and 20%) on inactivated purified 146S and PEG precipitate of FMDV serotype O and evaluated their stabilizing effect by using PaSTRy (Particle Stability Thermal Release Assay). All three sugars have shown an upward trend of stabilizing effect as in a concentration-dependent manner in terms of difference of T_r values (ΔT_r). Sucrose and glycerol showed higher T_r when compared to sorbitol. Based on the melt curve graph results, easy availability, and affordability, sucrose 10% was selected over the glycerol to stabilize FMDV 146S particles for storage of PEG precipitated virus. From this study, it is concluded that sugars can indeed act as stabilizers, and sucrose 10% has potential to stabilize 146S antigen and further evaluation is warranted using bulk antigen by 146S quantification method.

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PP_IV.10 Expression of Immunodominant Regions of E2 from an Indian Isolate of Classical Swine Fever Virus.

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Classical swine fever (CSF) is an economically important contagious fatal disease of domestic pigs and wild boars. This disease is caused by classical swine fever virus (CSFV), a member of the Pestivirus genus within the Flaviviridae family. Upon infection, three proteins of CSFV, namely E2, Erns and NS3 induce detectable antibodies. Since E2 is a major glycoprotein that produces neutralizing antibodies and provides protective immunity, it is widely used as a marker for measuring vaccine efficacy and antibody titer. In the present study, immunodominant regions of E2 glycoprotein from Indian field isolate of CSFV were expressed in E. coli. The 336 amino acid long N-terminal ectodomain (full-E2) and the 207 amino acid (aa 173-380) long C-terminal immunodominant region (partial-E2) were expressed and purified as 54kDa and 24kDa recombinant proteins, respectively. Both full-E2 and partial-E2 recombinant proteins were also characterized using MALDI-TOF-TOF analysis as CSFV-E2 structural proteins. The yield of purified full-E2 and partial-E2 recombinant protein was 26 mg and 84 mg/ liter culture, respectively. For raising hyperimmune sera against these proteins, chickens and rabbits were immunized with 200µg purified protein and boosted three times with 100µg purified protein intramuscularly. The sera collected one week after last booster were analyzed for CSFV-specific IgG antibody response using purified CSFV as coating antigen in ELISA. The results revealed that full-E2 induced better antibody response in both rabbits and chicken as compared to partial-E2. This study also indicated that these recombinant E2 proteins can be used as diagnostic antigen in ELISA.

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Technical Session - V

Nutritional interventions to ensure food security

LEAD PAPER

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Dr Manju Roy has a total experience of 20 years in teaching, research and extension activities. She has guided 05 undergraduate and 01 Ph.D student as major advisor and 54 students as member advisory committee. She is recipient of University Best Teacher Award in 2015. Currently she is working as principal Investigator of one research project funded by State planning commission Chhattisgarh. She has published 75 research papers in National and International journals of good repute and published four books for students of undergraduate and Post graduate programme.

Lipidomics signature and its implications on meat quality and health concern

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Metabolomics is a rapid developing area to provide valuable information about diseases, of which one aspect is lipidomics, the content and function of whole lipids in the cell or tissue in biologic systems. Over the latest decade, lipidomics has been extensively developed to give robust strength to the qualitative and quantitative information of lipid molecules derived from physiological animal tissues and edible muscle foods. Cellular lipids are comprised of a plethora of unique structures that contain many hundreds of thousands (or more) distinct lipid molecular species). Lipids belongs to various classes consist of cholesterol, glycerol-based phospholipids, and ceramide-based sphingolipids. Dysfunction of lipids metabolism was found to be associated with pathogenesis of many diseases, such as diabetes, Alzheimer's disease, dementia, hypertension, and various types of cancers. The full analysis of lipid species and their biological roles with respect to health and diseases is an object of lipidomics research. adipose tissue is no longer considered a merely energy reservoir, but also a source of various signalling molecules, adipokine and Fas with proinflammatory properties that can modulate immune cells or activating autophagy. Hence, it can be concluded that lipidomics is a branch of metabolomics, and ought to be understood as the study of the structure and function of the complete set of lipids (the lipidome) produced in a given cell or organism, as well as their interactions with other lipids, proteins and metabolites.

The global meat industry is a continuously growing sector with an ever-increasing demand. During meat production, an understanding of the direct correlation between metabolites and biological phenotypes that vary according to breed, feeding, and storage conditions can be effectively used to improve meat quality and taste characteristics. Lipid analysis, many tools including gas chromatography (GC), HPLC, TLC, MS techniques played important roles in elucidating lipid structures, identifying new lipids, and quantifying lipid abundance. Clinical lipidomics is a new extension of lipidomics, which aims to investigate metabolic networks through quantifying the complete spectrum of lipidomes in cells. Clinical lipidomics is also emphasized to integrate genomics, proteomics, metabolomics, and phenomics. The development of clinically useful lipid biomarkers requires consistent research methodology. The lipidomics disciplines successfully encompass a comprehensive and high-throughput understanding of meat composition, nutritional value, and safety with a combination of biochemical and mechanical mechanisms. Many putative lipid biomarkers following computational approaches and possible metabolism pathways enriched by bioinformatics provide valuable suggestions on food safety and health concerns regarding their potential during the treatment with preservatives, fermentation, aging, and storage. However, challenges remain due to the complexity of meat lipidome, the nature of key intermediate lipid-metabolites, and their evaluation concerning the quality and nutritional value of the final product.

INVITED PAPER

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Dr Sunil Ekanath Jadhav is a Principal Scientist (Animal Nutrition) at the Centre of Advanced Faculty Training, Animal Nutrition Division, ICAR-Indian Veterinary Research Institute, Izatnagar. He obtained BVSc & AH degree from the Krantisinh Nana Patil College of Veterinary Science, Shirwal, Maharashtra (1993-1998) and MVSc (Animal Nutrition) degree from College of Veterinary Science, Parbhani, Maharashtra (1998-2000). He qualified PhD (Animal Nutrition) degree from the ICAR-Indian Veterinary Research Institute, Izatnagar (2001-2005). He served in Defence Research and Development Organization, as scientist B & C during 2005-2012. He was selected as Senior Scientist (Animal Nutrition) and joined IVRI, Izatnagar in 2012 and promoted to Principal Scientist in 2018. He is having 20 years of research experience and 11 years of teaching experience. He has handled 12 research projects including NASF, AICRP, and NAE. He has guided six MVSc and three PhD students. He has published 70 research papers in peer reviewed journals and authored six books. He is a recipient of SRF for PhD degree of IVRI and National Technology Day Award and Technology Group Award of DRDO and Best Research Worker award of KNPVET Alumni Association. He is a Life Member of five professional societies. He is an Editor of 'Animal Nutrition and Feed Technology' journal of the prestigious Animal Nutrition Association of India. He is a former Executive Body member of Animal Nutrition Society of India, Karnal. He is a fellow of Animal Nutrition Association. He has acquired international exposure at the University of Florida, Gainesville, USA in the area of gut health and immunonutrition.

Importance of antioxidant trace minerals in amelioration of heat stress in animals

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The existing and projected climate change and global warming threats are believed to increase the earth temperature and thus heat stress to the animals in coming years is anticipated. Higher ambient environmental temperature is the potent stress inducer in variety of biological systems including domestic animals. Heat stress affects feed intake and balances of nutrients, hormones, antioxidants, and various minerals in animals. Heat stress increases free radical production where the antioxidant trace mineral levels may get reduced due to the partitioning of minerals towards the synthesis of molecules involved in antioxidant defence. Moreover, there is an increased excretion of nutrients due to higher body metabolism induced by heat stress. Hyperthermia disturbs efficiency of nutrient absorption and utilization in animals due to blood flow diversion from the gut towards the extremities, leading to hypoxia of intestinal cells resulting in leaky gut conditions which predisposes animals to bacterial infections or endotoxemia. Under heat stress condition the requirement of trace minerals involved in antioxidant defence such as zinc, selenium, copper and manganese is believed to be higher. Selenium is an essential ultra-trace element involved in antioxidant defence, immunity, and body metabolism through thyroid hormones. These functions are carried out through several selenoproteins like GPx, SePP, ID-II and TRx. Zinc is an important trace mineral that have multiple functional roles in health, reproduction, and growth. It acts as a cofactor for several metalloenzymes and many transcription factors require Zn for their structural integrity and function. Zn is involved in nutrient and nucleic acid metabolism. Zn protects the lipids, proteins, and cellular membrane from reactive oxygen species through Zn-containing superoxide dismutase enzyme and metalloprotein. Other trace minerals copper and manganese are also essentially involved in stress alleviation and health improvement during stress conditions.

INVITED PAPER

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Dr Anuradha Kumari received her B. Tech. degree in Dairy Technology from Indira Gandhi Krishi Vishwa Vidyalaya University, Raipur, Chhattisgarh, India and her M. Tech and Ph.D degrees in Dairy Chemistry from ICAR-National Dairy Research Institute, Karnal, Haryana, India. She had worked for approx. one year as a Senior officer at Sabarkantha District Cooperative Milk Producers' Union Ltd. (AMUL), Gujrat. She had also worked for one year as a Senior Research Fellow in ICAR-CIPHET, Ludhiana. She had worked as an Assistant Professor in Dairy Chemistry Department for about 3 and 7months at GADVASU, Ludhiana. Currently working as an Assistant Professor in Dairy Chemistry Department at Bihar Animal Sciences University, Patna. Her area of expertise includes whey protein ingredients enriched functional foods, development of low-calorie sweetened products and their detection. She has published 13 research papers in peer reviewed journals, 3 review articles, 10 book chapters, 5 popular articles and 3 practical manuals. She has also edited one book published by Springer. She was the student reviewer of e-learning content for B.Tech students. She has presented her research work at various national and international seminars and conferences and received several awards for best poster. She is the life member of Association of Food Scientists and Technologists (India).

Application of high-performance liquid chromatography for the analysis of low-calorie sweeteners

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In the present era, several health-related complications including obesity, diabetes, cardiovascular attacks, and many more are prevalent due to excessive consumption of sugar, a sedentary lifestyle, and lack of physical exercise. The growing awareness of health and fitness has led to the development of low-calorie sweetened products. A search for alternatives that have low or zero calories has motivated technologists to ponder over substitutes. A sugar substitute is a food additive that duplicates the effect of sugar in taste but usually has less food energy. Several artificial low-calorie sweeteners are available these days.Low-calorie sweeteners can be categorized as bulk sweeteners or intense sweeteners. Bulk sweeteners confer body and texture to food products and are fully metabolized by the body, providing a substantial part of energy. Thus, referred to as nutritive sweeteners examples are sorbitol, mannitol, xylitol, isomalt, etc. However, intense sweeteners like aspartame, acesulfame-K, saccharin, neotame, stevioside etc. have very high sweetness potency anda very low amount is required to sweeten the food. These are commonly not metabolized by our body and are excreted as such, thus referred to as non-nutritive sweeteners. As the very low level of intense sweetener required for the use in the food products, the food matrix would interfere with its analysis process. Its determination in the food is an analytical challenge. So, a highly sensitive and precise method is required for the analysis. Several methods like High-performance liquid chromatography with evaporative light scattering detection (HPLC-ELSD) or HPLC coupling with electrospray ionization mass spectrometry (HPLC/ESI-MS) etc. have been used but these are very expensive and complex for routine analysis. It was found that when the intense sweeteners were extracted and isolated with the solid phase extraction (SPE) method using a C18 cartridge before HPLC analysis, it removed the complex food matrixes hindrance during analysis by removing the unwanted peaks. Which eases the determination and identification of the peak of interest during analysis. The intense sweeteners aspartame and neotame were analysed in different food products like flavoured milk, yoghurt, ice cream and cake by HPLC method followed by SPE, the recovery for the method was 96-98% and the limit of detection was about 1.25 mg/l and 0.25 mg/l, respectively for aspartame and neotame. The precision of the method in terms of relative standard deviation was found to be within the 2% limit, indicating the repeatability of the method.

ORAL PRESENTATION

OP_V.1 Effect of *Tinospora cordifolia* leaf extract on expression of genes associated with regeneration of pancreatic islets in experimentally induced diabetic rats.

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Diabetes mellitus results from an absolute/relative absence of insulin due to either an insufficient number of functional insulin-producing pancreatic β cells or decreased sensitivity of peripheral tissues. Modern diabetes therapeutic strategies aim to device possible approaches for replenishing the β cells by (1) replacement of islets by transplanting cadaver derived β cells/islets from embryonic stem cells/induced pluripotent stem cells and/or (2) induction of endogenous regeneration of β cells. Endogenous pancreatic β cell regeneration could be achieved via exogenous natural sources such as medicinal plant extracts. The present study aimed to decipher the relative expression of genes involved in pancreatic β cell regeneration by supplementation of aqueous leaf extract of *Tinospora cordifolia.* For the study, twenty-four male Wistar rats were randomly divided into the three groups (n=8) viz., Group I: Normal control, Group II: Diabetic control (Streptozotocin induced) and Group III: Diabetic rats fed with aqueous extract of *Tinospora cordifolia* leaves. The expression of genes associated with pancreatic β cell regeneration viz. *Reg1α* and *Reg* 3β were studied by quantitative real time PCR assay in the pancreatic tissues. The results revealed an up-regulation of R*eg1α* gene expression up to 1.81 folds in Group III than Group II rats. The expression of *Reg* 3β gene was also found to be 9.9-fold higher in Group III than Group II rats. Thus, the results suggest that the aqueous leaf extract of *Tinospora cordifolia* can potentiate the process of β-cells' regeneration in the pancreas of experimentally induced diabetic rats by induction of genes *viz. Reg* 1α and *Reg* 3β that promote islet cell growth and survival. Further studies on transcriptomic analysis of gene regulation and validation of *Tinospora cordifolia* leaf extract could prove useful in the development of a more efficient and safer therapeutics in the management of Diabetes mellitus.

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OP_V.2 *In-silico* docking analysis to determine the effect of flavonols on insulin signalling mechanism

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Diabetes mellitus remains to be one of the most important metabolic disorders affecting a wide range of human and animal populations. In recent years, plant derived medications have found an immense use in the management of diabetes mellitus as they are considered as a safer alternative to allopathic medication. Many dietary polyphenols and phenolic compounds have been shown to regulate the expression of genes involved in insulin secretion in β-cells and insulin signalling mechanisms. With the advent of molecular docking tools, it is now possible to visualize the interactions between such plant-based compounds and effector proteins at an atomic level, enhancing our understanding and aiding in the selection of specific compounds for *in vivo* efficacy studies. The present study was designed to assess the interactions between three plant flavonols viz., quercetin, kaempferol & myricetin and two proteins associated with glucose homeostasis *viz*. GLUT-4 and PPAR-γ by *in-silico* docking analysis. Of the three flavonols, only kaempferol was found to have highly specific interactions with specific amino acid residues (viz., Tyrosine and Arginine) of PPAR-γ. Conversely, all the three flavonols were found to possess favourable hydrophilic/hydrophobic interaction with GLUT-4. The amino acid residue LYS 266 of GLUT-4 was found to interact with all the three flavonols, suggesting its possible role in potentiating the action of the flavonols. Thus, it can be hypothesised that the anti-diabetic potential of plants rich in these flavonols might be associated with alteration in the expression levels of GLUT-4. Further analysis on the drug-like properties of these flavonols followed by *in vitro* and *in vivo* studies may prove the potential of these compounds to be developed into an effective anti-diabetic therapeutic agent.

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ORAL PRESENTATION

OP_V.3 Unveiling citrate synthase enzyme as potential biomarker in developing a rapid method for monitoring of chilled and defrosted buffalo meat in supply chain

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With the globalization of meat trade, there is an increment demand for delivering premium quality of meat at distant locations. As chilled meat (CM) is very difficult to distinguish from the defrosted/frozen-thawed meat (FTM) visually while still it is in the supply chain so an improved method is required to develop to monitor the meat to meet regulatory compliance and also to satisfy consumer's needs. So, a study was conducted to develop a simple and rapid method for monitoring of CM and FTM for authentication. The method is based on extraction of mitochondrial citrate synthase (CS) enzyme (biomarker) from meat express juice (MEJ) and identification on SDS-PAGE and confirmation by Western Blotting (WB) and Enzyme Linked Immunosorbent Assay (ELISA). In the method, different substrates and developers used were also standardized, and for the robustness, it was validated for recovery study, linearity and precision study. Further, the developed method was used for stability study of CS enzyme on repeated freezing-thawing, storability and thermal history analysis. Results indicated that the method is very simple, easy and showed a very good precision in repeatability and reproducibility study. The CS enzyme showed very good stability on repeated freezing-thawing, and thus, CS has greater potential to acts a biomarker for authentication of chilled and defrosted meat to meet the regulatory compliance.

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OP_V.4 Proximal part of the magnum is a better choice for establishing contamination-free epithelial cell culture in chicken

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Magnum epithelial cells are used for different purposes such as studies of avian embryonic developmental biology, transgenesis, oncology, for production of biopharmaceuticals, for assessing environmental impact and host pathogen interaction. However, the establishment of magnum epithelia culture gets impeded due to high rate of contamination. Magnum is known to synthesize and secrete most of egg albumin during egg formation and provides a protein and moist rich environment favourable for microbial growth. Thus, epithelial cells isolated from magnum proper are severely contaminated even before proper establishment of the culture. Published report indicates that microbial diversity is lower in proximal part of magnum compared to the other parts. Hence, we hypothesize that proximal part of magnum would be a better choice to overcome the problem of contamination during the establishment of magnum epithelial cell culture. About 2 cm long piece of tissue was taken from the proximal part of magnum, chopped thoroughly and digested with 0.125% Trypsin for 15-20 min in room temperature. Cells were filtered with double muslin cloth or 100µM cell strainer. Finally, cells were plated in growth medium on 35 mm dish and on T25 flask. Medium was replaced in every 2 days. In about 4 days, cells started appearing in the tissue culture vessel and eventually, formed large colonies by day7 of plating. Morphologically, the cells look elongated or spindle shaped similar to that of fibroblasts. We have frozen some vials of these cells and separately continue to propagate them. We have also attempted multiple times to establish epithelial cell culture with magnum proper but have failed due to heavy contamination. Unlike those attempts of culture set up with magnum proper tissues, the current culture of epithelial cells isolated from proximal part of magnum remains free from contamination. Thus, proximal part of magnum is a better tissue for establishment of magnum epithelial cell culture.

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ORAL PRESENTATION

OP_V.5 Kisspeptin promotes follicular development through its effects on modulation of P450 aromatase expression and steroidogenesis in sheep ovarian follicles

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The present study was conducted to ascertain whether the role of kisspeptin in promoting *in vitro* development of preantral follicles was through the regulation of P450 aromatase gene expression and steroidogenesis in sheep. Accordingly, the cumulus cells and oocytes were collected from different development stages of preantral follicles grown *in vivo* and cultured *in vitro* in TCM199B (Group I), TCM199B + KP (10 µg/mL) (Group II) and Standard medium + KP (10 µg/mL). To measure the steroid (Estradiol-17β; E2 and Progesterone; P4) synthesis through ELISA, spent culture medium was collected separately from the same *in vitro* groups. E2 synthesis in the spent medium collected from all the three groups showed an increasing trend from PFs' exposed to respective culture media for 3 min to 2-day culture stage but decreased thereafter till 6-day culture stage. This is followed by a sharp increase in E2 concentration in the spent medium collected after *in vitro* maturation. However, P4 synthesis in group III followed increased pattern as the development progressed from PFs' exposed to culture medium for 3 min to *in vitro* maturation stage. The steroid production was observed at all stages of *in vitro* development in altered supplemented conditions. The steroid synthesis in the spent medium was highest in the 6 day cultured PFs' in Standard medium + KP matured *in vitro* for 24 h. Therefore, supplementation of kisspeptin along with other growth factors promoted steroid production in cultured preantral follicles far better than in other media.

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OP_V.6 Characterization and *in-vitro* evaluation of antidiabetic activity of collagen hydrolysate recovered from buffalo hide

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Collagen hydrolysate (CH) from buffalo hide was extracted from buffalo hide by enzymatic hydrolysis (alcalase) for 8 h at 55 °C and pH 8.0 under different enzyme substrate ratios of 1:50, 1:100 and 1:200 (v/w) and the corresponding CH samples thus obtained were referred as CH50, CH100 and CH200, respectively. One commercial collagen hydrolysate (CHM) was kept for comparison purpose. The yield of CH200 was significantly ($P<0.05$) higher (13.15%) than CH50 and CH100 (7.51 and 9.82%, respectively. Degree of hydrolysis was significantly ($P<0.05$) lower for CHM (11.52%) compared to other three CHs. 2, 2-diphenyl-1-picrylhydrazyl (DPPH) and 2, 2′-azino-bis (3-ethylbenzothiazoline-6-sulfonate) (ABTS) radical scavenging activities of CH200 was significantly ($P<0.05$) higher (52.72 and 68.93%, respectively) than CH50, CH100 and CHM (746.43 and 63.02%, 45.31 and 66.33% and 39.37 and 63.21%, respectively). Sodium dodecyl sulphate–polyacrylamide gel electrophoresis (SDS PAGE) revealed the molecular weight of CHs to be between 1.06-3.5 kDa. Scanning electron microscopy (SEM) showed that the particle size of CH200 sample was relatively smaller than the CH50 and CH100 whereas CHM possessed smallest particle size. Fourier transformed infrared (FTIR) results indicated that amide I of CH200 and CHM were noticed at higher wavenumber (1634.79 & 1636.24 cm^{-1}, respectively) having higher amplitudes than CH100 showing comparatively greater loss of triple helical structure in CH200 and CHM. *In-vitro* antidiabetic activity of different CHs showed non-significant ($P<0.05$) difference in α-amylase inhibitory activity but DPP-IV inhibitory activity was significantly ($P<0.05$) higher (72.27%) for CH200 than CH50, CH100 and CHM at 5 mg/mL concentration. Our findings indicated that the alcalase @200 units/g of buffalo hide at 55 °C and pH 8 for 8 h could be used to get better CH yield. CH thus obtained exhibited higher DH, antioxidant and antidiaabetic activity which might potentially serve as a natural

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ORAL PRESENTATION

OP_V.7 Assessment of the free radical scavenging activity and anti-oxidant effects of baicalein flavonoid against LPS-induced injury in goat isolated pulmonary artery: An *in vitro* study

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The present study was conducted to assess the free radical scavenging activity and antioxidant effects of baicalein against LPS-induced injury in isolated pulmonary artery of goat. Goat pulmonary arteries were isolated from goat lung obtained from slaughter house and rings prepared in MKHS solution. Rings were placed in 24 well plates containing DMEM (Dulbecco's modified eagle medium) supplemented with 10% foetal bovine serum. Injury was induced in by adding lipopolysaccharide (LPS) at the dose 50 µg/mL and for the treatment of injury, baicalin (200 µM) was added separately in LPS containing DMEM medium. Plates were incubated at 37 ^{0}C, & 5% Co_2 for 24 hours. Further, tissues were collected and processed to estimate oxidative stress parameters. DPPH and NO free radical scavenging activity of Baicalein was determined. In our result findings, DPPH free radical scavenging activity and IC50 value of baicalin at concentration of 20 µg/ml, was 84.3±3.22 and 7.48 µg/ml, respectively. However, NO free radical activity and IC50 value of baicalein at concentration of 20 µg/ml, was 80.3±4.38, and 9.73 µg/ml, respectively. Further, antioxidant effects of baicalin were assessed against LPS-induced injury in goat pulmonary artery. Baicalein significantly enhanced GSH level which was decreased in LPS group. MDA level was significantly decreased with baicalein treatment. However, catalase and SOD activity were not improved with baicalein treatment in comparison with LPS injury group. In conclusion, the current investigation reveals that baicalein has free radical scavenging activity as well as anti-oxidant potential against LPS-induced injury in goat pulmonary artery by increasing the level of reduced glutathione and decreasing MDA generation.

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OP_V.8 Blood haematological, biochemical and antioxidant status of preweaned indigenous calves treated with *Tinospora cordifolia* and *Asparagus racemosus*

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The aim of this study was to determine the effect of Giloy (*Tinospora cordifolia*) and Shatavari (*Asparagus racemosus*) supplementation on blood biochemical and antioxidant status, in growing pre-weaned indigenous calves. Eighteen growing pre-weaned indigenous calves were randomly allocated into three groups having six calves in each groups and fed for 90 days. Feeding regimen was similar in all three groups.The treatment groups were supplemented with 50 mg of Giloy/kg body weight (BW) (T-1) and 50 mg of Shatavari/kg BW (T-2). Standard ration was supplemented to control group. Peripheral blood samples were collected at 0, 30, 60 and 90 days post-treatment and analyzed for haematological attributes, biomarker of energy, lipid metabolism, protein metabolism, liver and kidney function, antioxidant status, oxidative stress and immune response.RBCs count was significantly higher ($P<0.05$) in Shatavari supplement group whereas, Hb concentration and PCV values were higher in Giloy supplemented calves. Plasma globulin level was significantly ($P<0.05$) higher in Giloy supplemented calves. The plasma cholesterol, triglyceride and lipid peroxide (LPO) levels were significantly lower ($P<0.05$) in Giloy supplemented groups. However, beta hydroxyl butyric acid (BHBA) concentration was lowest ($P<0.05$) in Shatavari supplemented group. Treatment did not exert any effect on plasma glucose and non-esterified fatty acid (NEFA) concentrations. Giloy and Shatavari supplementation did not show any effect on the biomarkers of liver functions and kidney function. The superoxide dismutase (SOD) activity and total antioxidant status (TAS) were higher in Shatavari supplemented group. Adding Giloy and Shatavari to the diet increase ($P<0.05$) plasma total immunoglobulin concentrations. In conclusion, dietary supplementation of Giloy and Shatavari in pre-weaned indigenous calves improves antioxidant status and immunity without any adverse effect on liver and kidney functioning.

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ORAL PRESENTATION

OP_V.9 Nutrient relationships in finger millet grains and their significance for health

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Finger millet grain is suitable for consumption in different forms depending on consumer preference and is a rich source of calcium. However, understanding the nutrient relationships is a prerequisite for development of nutrient rich varieties along with yield. To understand nutrient relationships, 88 grain samples of released cultivars, elite breeding lines and land races used in breeding program were analysed for protein, oxalic acid, phytic acid and minerals using standard AOAC methods. Kjeldahl total nitrogen was determined for Protein content (Pr) estimation. Oxalic acid was using HPLC. Mineral content was determined using microwave digestion and atomic absorption spectrometry. An aliquot of digested solution was used for phosphorus determination using Barton's reagent. Phytic acid content was determined using kit based enzymatic assay. The data was analysed using SPSS ver 16.0. The range for different nutrients in 100 g sample was: protein, 4.9-14.5 g; oxalic acid, 18.2-204.3 mg; phytic acid, 0.42-1.39 g; iron, 1.42-4.81 mg; zinc, 1.48-3.01 mg; calcium, 193.9-392.8 mg; magnesium, 122-239 mg; and phosphorus, 182-497 mg. Protein content was significantly positively correlated with Iron (r =0.645), Zinc (r =0.595), Calcium (r =0.432) and Magnesium (r =0.359); while oxalic acid had significant positive correlation with phosphorus (r =0.244); and phytic acid with calcium (r =0.312). There was no correlation between phosphorus content and Fe, Zn, Ca, Mg and protein content. This is encouraging to breed better varieties with high mineral content, high protein content and low phytic acid content without compromising on grain yield. Moreover, Ca:P molar ratio was in the range of 0.33-1.4 with a mean of 0.87. High Ca:P ratio in habitual diets was found to improve bone mineral density and reduced incidence of fractures in elderly. Among the cereal and millet grains Ca:P ratio is highest in finger millet due to its rich calcium content and is a suitable food grain for better bone metabolism.

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OP_V.10 Assessment of phytochemical properties, antioxidant and antimicrobial activity of certain ethnomedicinal plants

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The use of plant based natural products in the treatment and prevention of various diseases and health enhancement due to nutritive and pharmacological benefits has lead to the significant attention of the scientific community and the general public nowadays. *Murraya koenigii, Centella asiatica, Amaranthus viridis and Pisidium guajava* have been commonly used in India especially in the north eastern region in the treatment and prevention of various diseases. The aim of the present study was to determine the phytochemical properties, antioxidant and antimicrobial activity of the hydroethanolic and ethanolic extracts of the selected plants. Phytochemical analysis showed presence of steroids, saponin, glycosides, triterpenes, alkaloid, falvonoid, total phenol, tannin in all the selected plants. Antioxidant activity of both the extracts of plants were analysed by DPPH method and observed that all the plants have antioxidant activity. The ethanolic extracts of all the three plants except *Amaranthus viridis* showed sensitive response against both gram positive (*Staphylococcos aureus*) and gram negative bacteria (*Escherichia coli*) which indicates that it has antimicrobial activity.

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POSTER PRESENTATION

PP_V.1 Curcumin protects copper-induced oxidative status, growth performance, genotoxic potential and histoarchitectural anomalies in fish *channa punctatus* (Bloch, 1793)

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Curcumin (CUR) has been described for its anti-inflammatory, antioxidant, anti-carcinogenic, and anti-tumor activities along with its role in the prevention of histological injury due to exposure to xenobiotics. The present study shows the protective effects of curcumin against Cu-induced toxicity in fish. Healthy *Channa punctatus* was segregated into six groups. Group I served as a control, group II was exposed to 3 mg/L CUR; Group III to the Environmentally relevant concentration of copper (ERCC) i.e., 0.85 mg/L; Groups IV, V and VI were simultaneously co-exposed to ERCC, and three different CUR concentrations 1, 2, and 3 mg/L, respectively for 2, 4, 6 and 8 weeks. After completion of each exposure period, Cu accumulation, growth performance, and various hematological and biochemical parameters were measured. In group III, SOD, CAT, GR, and LPO were significantly ($p<0.05$) increased; decreased GSH activity; increased histoarchitectural changes in the liver and kidney, elevated reactive oxygen species (ROS) and Micronucleus (MN) frequencies in blood. In groups IV, V, and VI, significantly ($p<0.05$) decreased activities of SOD, CAT, GR, and LPO; and elevated GSH; recovery of histoarchitectural changes in the tissue sample while the reduction in ROS and MN frequencies were observed. This study establishes the protective role of CUR against Cu-induced hepatotoxicity and nephrotoxicity in *C. punctatus*.

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PP_V.2 Green synthesis, characterization and assessment of the antioxidant and antibacterial potential of phytochemically embellished silver nanoparticles

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Currently, drug resistance has been emerged due to the over use of the antibiotics and drugs in treating infectious diseases. The synthesis of nanoparticles using plants has received a lot of attention recently as a low-cost and environmentally friendly technique. The present study focused on the green biosynthesis of silver nanoparticles (AgNPs) using Azadirachta indica (*A. indica*) leaf extract. The antioxidant and antibacterial properties of *A. indica* leaf extract and synthesized AgNPs were assessed. The findings revealed that the synthesized AgNPs from *A. indica* leaf extract showed a prominent surface plasmon resonance (SPR) peak around 400–450 nm, were crystalline in nature, and had a shape that was spherical with size ranges from 55 nm to 88nm. Biochemical fingerprinting of aqueous extract of *Azadirachta indica* by High resolution liquid chromatography and Mass spectrometry (HPLC-MS) analysis revealed the presence of alkaloid, flavonoids, phenolic compounds which have potent antioxidant and antibacterial capabilities that protect cellular stress by quenching of free radicals. The phytofabricated AgNPs showed a higher antioxidant activity (IC50 0.37) as compared with the *A. indica* leaf extract (IC50 0.85). They also exhibited remarkable antibacterial activity at levels of 37.50and 65.63µg/ml and the protein leakage assay demonstrate that the phytochemicals of *A. indica* resulted in the leakage of essential proteins from the bacterial cells by damaging the bacterial cell membrane against both Gram-positive, methicillin-resistant (*S. aureus*), and Gram-negative (*E. coli*) bacteria, respectively. Therefore, the resulting phytofabricated AgNPs could be used as a promising antioxidant, antibacterial and may be used as an alternative to antibiotic drugs, exhibiting better effect on multidrug resistant bacteria.

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POSTER PRESENTATION

PP_V.3 Hypoglycemic effect of Curcuma leucorrhiza in Streptozotocin induced diabetic wistar rat

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The hypoglycemic effects of Curcuma leucorrhiza Roxb. was investigated in streptozotocin (STZ) induced diabetic Wistar rat. The rhizomes of the plant were shade dried, ground to powder and extracted by using ethanol. Ethanol extract of Curcuma leucorrhiza Roxb. was orally fed at the dose of 100mg/kg and 200mg/kg b.w. daily to STZ induced diabetic rats. The level of serum glucose was reduced significantly in the rats treated with metformin, 100mg/b.wt.kg-1 and 200mg/b.wt.kg-1 extracts of Curcuma leucorrhiza Roxb. The blood glucose level were estimated on 0, 3, 5, 8, 15,18 and 21 days of treatment. The blood glucose level among the rats treated with ethanol extract of Curcuma leucorrhiza Roxb. rhizome also significantly decreased ($p<0.01$) as the treatment progressed, similar to the group treated with metformin. The percent decreased on treatment with dose of 100mg/kg b.w. were 21.73%) on 1st day, 30.13%) on 3rd day, 40.12%) on 5th day, 49.34% on 8th day, 52.50% on 15th day, 57.31% on 18th day and 64.89%) on 21st day. In the case treatment with 200mg/kg b.w. the percent decreased were 4.41% on 1st day, 32.12% on 3rd day, 44.30% on 5th day, 51.90% on 8th day, 56.40% on 15th day, 60.67% on 18th day and 63.72% on 21st day of treatment. Thus, the extract at a dose rate of 100 mg and 200 mg is seen to have same hypoglycemic effect with a 21 days' regimen and is comparable with the standard treatment under reference. Treatment with the ethanol extract also increased the expression of GLUT-4 gene in heart tissue.

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PP_V.4 Effect of dietary supplementation of *Achyranthes Aspera* powder on growth performances, serum biochemistry and gut health of Broiler Chicken.

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The antibiotic resistance is a global health issue and now a days consumer demand for antibiotic free products has risen significantly. The phytogenic feed additives are considered as viable alternative to antibiotics in poultry. Therefore, the present study was conducted to evaluate the effect of feeding diets supplemented with *Achyranthes Aspera* on the performances of broiler. Two hundred day old chicks will randomly divide into five treatment groups, having five replicates with 8 birds each. The group were distributed as; group T-1 control offered basal diet and group T5 offered basal diet with antibiotic whereas group T-2, T-3, T-4 were offered basal diet supplemented with *Achyranthes Aspera* @ -0.5,1.0,1.5% respectively on dry weight basis. Duration of experiment was 42 days. The result indicated that there was a non - significant ($P \geq 0.05$) linear increase in body weight gain in broiler chicken over control with increase in level of *Achyranthes Aspera* supplemented from 0.5% to 1.5% . No significant ($P \geq 0.05$) effect was found in feed consumption of birds among various treatment groups including control and antibiotic. There was no significant ($P \geq 0.05$) differences among various carcass traits between control and *Achyranthes Aspera* supplemented groups. Serum total protein, triglycerides, uric acid, calcium and phosphorous content of broilers supplemented with various levels of *Achyranthus Aspera* did not differ significantly. The results revealed that the lactobacillus count was significantly ($P>0.05$) high in 1.5 per cent *Achyranthus Aspera* powder supplemented group and least lactobacillus count was recorded in the antibiotic supplemented group. Total coliform count was minimum in the 1.5 % *Achyranthus Aspera* powder supplemented group and highest was recorded in control group. Based on the results, it may be concluded that *Achyranthus aspera* plant powder supplementation at 1.5 % level in broiler diet for improved growth performances, beneficial gut lactobacillus count and reduced pathogenic *E.coli*, count in broiler chicken.

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POSTER PRESENTATION

PP_V.5 Unraveling Vitamin D as an antiviral agent against Newcastle disease virus

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Vitamin D (Vit-D) or calcitriol, is renowned for its role in promoting strong bones and teeth, as well as its ability to modulate inflammation and support the immune system. Additionally, it has been reported to be highly potent antiviral agent against various viruses, including hepatitis C, SARS-CoV-2, and infectious bursal disease. However, the underlying mechanisms behind the mode of action are still unclear. Newcastle disease virus (NDV) belongs to the *paramyxoviridae* family and causes infectious diseases in numerous avian species. It has been shown to be highly pathogenic for its host, with a 100% mortality rate. In the present study, we are focused to explore Vit-D as an antiviral agent against NDV infection. Our aim was to determine its *in vitro* and *in ovo* potential to inhibit the viral production and explore the mechanism(s) of inhibition. We hypothesized that Vit-D rich diet for the birds could offer a solution to curtail the disease and prevent it from spreading. The results suggested that Vit-D, when post-treated, remarkably inhibits NDV production in chicken embryo fibroblast (DF-1) cells. The anti-NDV effects were also observed in a concentration dependent manner. Furthermore, *in ovo* studies, the NDV titer in allantoic fluid exhibited a substantial reduction after administering Vit-D to 9-day-old chicken embryos. Currently, we are identifying the proteins associated with Vit-D modulated antiviral pathways, and determining their role in virus infection. Lastly, we concluded that Vit-D could effectively curtail the NDV infection which suggested the interplay between the vitamin D and NDV.

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PP_V.6 Effect of feeding variable dietary supplemental levels of vitamin E and ascorbic acid on growth, immune response and serum biochemical parameters of growing turkey poults

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An experiment was undertaken to evaluate the response of turkey poults to dietary levels of supplemental vitamin E and ascorbic acid on growth, immune response and serum biochemical parameters. An eight weeks (0-8wks) feeding trial was conducted involving two levels of supplemental vitamin E (50 and 100 mg/kg) each with two levels of supplemental ascorbic acid (100 and 200 mg/kg) in a 2×2 factorial experiment. Day old turkey poults (n=128) were randomly distributed in 16 groups of 8 chicks each and each of such diet was offered as mash *ad libitum* to four replicated groups of eight chicks each kept in battery brooder cages. Results indicated that the body weight gain was found significantly ($P \leq 0.05$) higher at 50 mg/kg vitamin E than 100 mg/kg vitamin E during all growth phases. Significantly ($P \leq 0.05$) lower and better FCR was recorded at 100mg/kg ascorbic acid than 200 mg/kg ascorbic acid during 5-8 and 0-8 wks of age. The cellular and humoral immune response as well as immune organs weight did not differed significantly due to different dietary treatments. Significantly ($P \leq 0.05$) higher total protein was recorded at 200mg/kg ascorbic acid than 100mg/kg ascorbic acid. The uric acid concentration was found significantly higher at 50mg/kg vitamin E than that recorded at 100mg/kg vitamin E. The serum cholesterol concentration was found significantly ($P \leq 0.01$) higher in a dietary combination of 100mg/kg vitamin E with 200mg/kg ascorbic acid in the diet. The Serum ALT activities was found significantly ($P \leq 0.05$) higher at 50mg/kg vitamin E than 100mg/kg vitamin E. Based on the results it was concluded that a dietary combination of 50mg/kg vitamin E with 100mg/kg ascorbic acid was found adequate for optimum growth, immunity and serum biochemical parameters of growing turkey poults during 0-8wks of age.

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POSTER PRESENTATION

PP_V.7 Evaluation of biochemical profile on non-descript goat during periparturient period under agro-climatic condition of Mizoram

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A study was carried out to evaluate the effect of periparturient period on non-descript goat reared under agro-climatic condition of Mizoram, a North-eastern state of India. Six numbers of apparently healthy does of almost same age in their advanced pregnancy was used as experimental animals; whereas equal numbers of clinically healthy non-pregnant does of similar age group was used as control for the current investigation. Blood sample was collected from each animal of the experimental group through jugular vein puncture on -30day, -15day, 0day, +15day and +30day from kidding; while for control group, blood samples were collected at monthly interval from each animal during the study period. Results obtained in the study revealed substantial alteration in biochemical profile of the does during periparturient period. The level of metabolic parameters was also considerably altered by periparturient period. The glucose concentration gradually increased from -30day prepartum till the day of kidding Conversely, the concentration of total protein decreased progressively with length of pregnancy during prepartum period, while it increased again in postpartum. The globulin concentration recorded in our study also exhibited variation similar to that of total protein. In the present study, control group showed low A:G ratio than the treatment group. The triglyceride was significantly lowest on day of parturition. The total cholesterol was found be highest on +30day postpartum. The HDL was significantly lowest on the day of kidding. Alteration in the levels of LDL were found be non-significant during the study period. The BUN level was lowest on -15day prepartum and highest on +30 day postpartum. Similarly, uric acid was found to be significantly lowest on 0 day and highest on +30 day postpartum. There was numerical variation in the concentration of total bilirubin, direct bilirubin and indirect bilirubin. The activities of enzymes viz. SGPT, GGT, ALP and CK-MB did not vary significantly during the study period. The postpartum AST activity measured in our study was higher than that of the prepartum period. The LDH level was found to rise around kidding and during early lactation as compared to prepartum period. It was significantly highest on +15day postpartum and lowest on -30day postpartum. The activity of LDH also enhanced during periparturient period as compared to non- pregnant does.

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PP_V.8 Effect of ascorbic acid and Zinc oxide nanoparticles on lipid metabolites and meat quality in broiler birds.

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Broiler chickens are susceptible to stress during land transportation. Stress conditions alter the physiological and biochemical status of broiler chicken. The present study was conducted to determine the effect of transport stress on serum lipid profile and meat quality. A total of six weeks old 120 broiler birds of Sonali breed were divided into four groups. Each group comprises of 3 replicates with 10 birds in each. Group I was considered as control and not transported. Group II, III, IV were transported for a distance of 90km. Group III and IV received the treatment of Ascorbic acid and Zinc oxide nanocomposites respectively. Body temperature of birds and meteriological data, (Environmental temperature, humidity and Temperature humidity Index) were recorded. Blood and thigh muscle samples were collected before and after the transport. Results revealed increased ($P<0.05$) rectal temperature, heterophil/lymphocyte ratio and muscle pH, whereas decreased percentage drip loss and meat colour was recorded in broiler birds after transportation. Treatment of ascorbic acid and zinc oxide nanoparticles prevent the deterioration of meat quality due to transport stress. Increased total cholesterol, triglyceride and LDL cholesterol and phospholipid ($P<0.05$) was observed in group II birds as compared to group III and IV. The present study elucidates the alleviation of transport stress in broiler chicken using phytofabricated zinc oxide nanocomposites.

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POSTER PRESENTATION

PP_V.9 Comparative evaluation of A1 and A2 milk casein hydrolysates feeding on diabetes related parameters in male W istar rats

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Many reviews and epidemiological studies have reported the negative effects of A1 milk on human health. This was reportedly attributed to the different variant of -casein in the same, which releases BCM7, an opioid that has been implicated in many illnesses, including heart diseases, Type 1 diabetes and autism. The present study was undertaken to draw an understanding about the difference in effects of A1 and A2 milk derived casein hydrolysates (A1CH and A2CH) feeding on diabetic related parameters in animal model. A 60 days trial in male Wistar rats (6wks old), where diabetes was induced on 51st day using streptozotocin (STZ), followed by sacrifice on 61st day. During the feeding trial, rats were fed with respective casein hydrolysates @800mg/kg b.w. along with the basal diet. Body weight and blood glucose level was recorded on weekly basis during the feeding trial and estimation of diabetes related parameters such as plasma Insulin, C-peptide and HbA1c was conducted after sacrificing the animals. Analysis of Insulin and C-peptide level in the plasma of rats collected at the termination of study revealed that A1CH or A2CH feeding did not offer any significant ($p>0.05$) effect on these two. The A1 and A2 CHs neither enhanced insulin and C-peptide level nor decreased the same in diabetic animals. It was likely that the significant difference ($p<0.05$) in the levels between control and STZ groups was due to the drug use. HbA1c levels in the plasma of rats estimated using ELISA at the end of study exhibited similar trend as that of C-peptide and Insulin levels. The results of the present study most likely suggest that A1 or A2 casein hydrolysate feeding did not cause significant effect on the diabetes related parameters.

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PP_V.10 Combination effect of essential oils' active components against high priority pathogens *P. aeruginosa* and *S. aureus*

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Increasing drug resistance among range of pathogens have emerged as the serious threat to global health. Various alternatives have already been evaluated for the potential therapeutic alternative of antibiotics, among which essential oil components have been one of the most explored alternatives. Active components of essential oils have well known and reported for their antimicrobial activity. In this study, we evaluated the antimicrobial activity of essential oil components including eugenol, carvacrol, cinnamaldehyde and thymol against *P. aeruginosa* and *S. aureus*, which have been categorized as the high priority pathogens in Indian Priority Pathogen List (IPPL) released by Department of Biotechnology, Government of India. In this study, eugenol was found to be effective against targeted pathogens with MIC of 0.25% (v/v) and cinnamaldehyde with the MIC of 0.031%-0.062% (v/v). Thymol exhibited MIC of 0.5% (v/v) for *S. aureus* and 0.062%-0.125% (v/v) for *P. aeruginosa* while, carvacrol exhibited MIC of 0.031%-0.062% (v/v) for *S. aureus* and 0.062%-0.125%(v/v) for *P. aeruginosa*. the combination effect of these components was analyzed by checkerboard method, in which carvacrol and thymol exhibited FIC index of 0.75 for *S. aureus* and 0.5625 for *P. aeruginosa*. Eugenol and thymol combination exhibited FIC index of 0.75 for *S. aureus* and 0.625 for *P. aeruginosa* indicating partial synergistic activity between them. These findings suggest that the combination of EO components can serve as promising alternative therapeutic against resistant infections.

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Technical Session - VI

Chemistry to omics: Role of biochemistry in clinical diagnosis

LEAD PAPER

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Dr Devendra S. Chauhan is involved in research on molecular biology of mycobacteria for more than 20 years. His research is focused on multiple aspects of mycobacterial infections especially on leprosy and tuberculosis. He received Young scientist award by Society of Immunology and Immunopathology (SIIP) in 2004, Young scientist award by Hon'able Dr APJ Abdul Kalam in 2008 and Best meritorious award awarded by International Conference of Leprosy in 2008. He was visiting scientist for France under ICMR-INSERM Fellowship in 2004 and for training at Louisiana State University, USA under ICMR-International fellowship in 2010. He has published more than 150 research publications and attended more than 40 conferences. He has been granted one Indian Patent and he has filed four Indian Patents.

Role of dna finger printing methods in clinical diagnosis of tuberculosis:

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Mycobacterium tuberculosis is one of the most successful bacterial pathogens in the history of mankind. The prevalence of tuberculosis in developed countries is lower than to developing countries. Most important advancement in the field of tuberculosis research has been the development of molecular techniques that allows the identification of sources and tracking of individual strains of *M.tuberculosis*. Before the introduction of molecular typing methods, there was little to aid distinction between individual strains of *M. tuberculosis*. Important DNA fingerprinting techniques to differentiate M.tb are :

A: Random Amplified Polymorphic DNA (RAPD):
RAPD also referred to as arbitrary primer PCR is a DNA fingerprinting method which is easy to use, rapid and needs one day to finish the experiment.

B: Restriction based Methods:
(a) Simple Restriction Analysis or Pulsed Field Gel Electrophoresis:
Simple restriction endonuclease digestion patterns of DNA from *M. tuberculosis* and other mycobacteria were initially used to trace the transmission route and to determine similarities and divergences.

(b) Ribotyping:
It is known that ribosomal gene region comprise of conserved sequences during evolution interspersed with divergent sequences in prokaryaote as well as in eukaryotes.

(c) Direct Repeat Probes:
Fingerprinting methods using direct repeat probe(s) have been observed promising for *M. tuberculosis* isolates which have low copies of insertion sequences.

(d) IS 6110 RFLP:
This discipline of molecular epidemiology of tuberculosis began with the identification of IS6110, a novel mycobacterial insertion sequence which formed the basis of a reproducible genotyping technique for *M. tuberculosis*. This method is now firmly established, but is still expensive, labour-intensive and only applicable using viable culture material.

(e) Polymorphic guanine-cytosine-rich repetitive sequence restriction fragment length polymorphism typing:
The identification of a PGRS present in multiple chromosomal clusters enabled the development of a second RFLP typing system. This has been shown to have a discriminatory power close to that of IS6110 typing, even in isolates with low copy numbers of IS6110, making it an ideal secondary typing system.

(f) Spoligotyping:
Spoligotyping, which interrogates a DR sequence comprising a repetitive 36-base-pair element separated by short nonrepetitive sequences, is one such PCR based technique. This method is based on the evaluation of the presence or absence of 43 spacer DNA sequences between the 36 bp direct repeats (DRs) in the genomic DR region.

(g) Mycobacterial interspersed repetitive unit variable number of tandem repeats and other polymerase chain reaction-based techniques (MIRU-VNTR):
Mycobacterial interspersed repetitive units (MIRUs) are an example of such elements. They are a specific class of VNTR that have been identified at 41 different loci in the genome of *M. tuberculosis*. PCR amplification across each MIRU, therefore, generates fragments of different sizes from different strains.

DNA Microarray / Chip based methods:
Using functional genomics approach (Microarray Technology), we have developed our own indigenous DNA chips for early diagnosis of MDR/XDR tuberculsosis as well as paucibacillary cases of leprosy. These DNA chips can predict the gene responsible for efflux out the drug.

INVITED PAPER

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Dr Dhanjit Das did his M.V.Sc in 2003 and PhD in 2006 from ICAR-Indian Veterinary Research Institute, Izatnagar. His research interests are Neuro-developmental and Neuro-psychiatric disease modeling using induced Pluripotent Stem Cells (iPSCs), Genetics of Intellectual Disability and Biochemical and molecular study in mitochondrial disorders. His group is presently engaged in diagnosis of different genetic disorders by cytogenetic and molecular techniques. The main focus of research is on neuro-developmental disorder in humans. In this area, we evaluate the suspected cases of fragile X syndrome patients for presence of FMRP protein in their peripheral leucocytes and the diagnosis is confirmed by analysis of CGG repeat analysis in FMRP gene. Studies have been undertaken to evaluate the presence of mutations in MECP2, CDKL5, FOXG1 gene in case of Rett syndrome. Spectrum of mutations including novel mutation have been identified in various genetic disorder Development of induced Pluripotent Stem Cells (iPCSs) based cellular model for various neurodevelopmental and neuro-psychiatric disorders is also one of the important research area of our lab. In this aspect, we have successfully developed number of various patient specific cell lines namely 22q11.2del, 15q11.2-13.1del, 17p13.1del etc. These iPSCs were successfully differentiated to neurons in order to develop cellular model for specific genetic conditions. Other area includes, studying the role of tissue resident stem cells in causation of various reproductive disorders such as Endometriosis. The research findings have been published in national and international high impact factor journals. More than 40 research paper have been published till today. He has been awarded with Senior Research Fellowship–Indian Council of Agricultural Research (ICAR), Junior Research Fellowship–Indian Council of Agricultural Research during 2001-2003 and Indo-US fellowship by IUSSTF to conduct advance research at University of Pittsburgh, USA during 2013–October 2014.

Clinicaland molecular biochemistry in human disease diagnosis: disease modelingusing induced pluripotent stem cells (iPSCs)

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Biochemical and molecular biomarkers provide a tremendous potential in disease diagnosis including human and animals. These markers have become increasingly important in clinical practice, as they can help physicians to make accurate diagnoses, monitor disease progression and evaluate the effectiveness of treatments. One of such human genetic disordersknown as mitochondrial disorder in which biochemical test such as serum lactate levels, estimation of mitochondrial enzyme activities has been used for diagnosis. A group of patients with suggestive of mitochondrial disorders based on clinical and serum lactate levels were recruited. Definitive diagnosis was made through estimation of mitochondrial enzymes activity in muscle biopsy and buccal swab samples. Further, genetic confirmation was carried out using whole exome analysis.

Another group of disorder such as neuro-developmental and neuro-psychiatric disorder in which molecular analyses were carried out for definitive diagnosis. One group of neuro-developmental disorder known as autism spectrum disorder and Rett Syndrome have been analyzed for presence of genetic mutations in candidate genes. Numbers of pathogenic mutations have been identified in genes like MECP2, CDKL5, FOXG1 etc. Similarly, number of genetic mutations and copy number variations (CNVs) have been identified in a group of neuro-psychiatric condition known as adolescent onset Schizophrenia.

Understanding the pathogenesis of these disorders is utmost important for cellular therapy as well as development of targeted drug therapies. Therefore, neurons from the affected individuals are best suited cellular model to study the effect of these mutationsand testing novel drugs. It is generally impossible to obtain live neurons from an affected patient to study. Hence, induced Pluripotent Stem Cells (iPSCs) have been utilized for development of cellular model. The double advantage of this iPSCs technology is that, the cells would not only carry the particular gene mutation, but also the same genome as the patient. We have generated a couple of well characterized iPSCs line for neurodevelopmental disorders (22q11.2del, 15q11.2-13.1del, STXBP1 mutation) and neuropsychiatric disorder (17p13.1del). These iPSCs lines have been differentiated to cortical neurons. Functional analysis (such as calcium imaging) as well as structural analysis of iPSCs derived neurons such as dendritic length and arborization, dendritic spine type and numbers have been analyzed in patient cell lines compared to a control. Overall, we have demonstrated that these iPSCs lines could further be used either for cellular therapy or drug development study.

INVITED PAPER

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Dr Shalini Sharma completed her B.V.Sc. (2002) and M.V.Sc. (2004) degrees from College of Veterinary Sciences, Bikaner, India, and her Ph.D. from University of Tennessee, Knoxville, Tennessee, USA (2011). She worked on understanding immunity and immunopathology to acute viral infections during her doctoral research work. Thereafter, she served as a postdoc from 2011 to 2014 at St. Jude Children's Research Hospital, Memphis, Tennessee, USA, where she worked on immunity and immunopathology to heterologous viral infections. Since 2014, she has been working as an assistant professor at Lala Lajpat Rai University of Veterinary and Animal Sciences, Hisar, Haryana, India. Her area of interest is infectious diseases, with a focus on understanding cellular immune response against selected pathogens of domestic livestock. Dr. Shalini has made some basic and fundamental contributions in the area of host immune responses to viral and bacterial infections, exemplified by high impact publications which includes Nature, Science translational medicine, Cell Host Microbes, PNAS-USA, Journal of Virology, Journal of Immunology, Plos Pathogens, Antiviral Research, Clinical Microbiology Reviews, Molecular Biology and Evolution etc, with an impact factor of up to 69.5. She has published >50 papers with an "h" index of 24 and over 3000 citations. She has been conferred with various Awards/Honours including, DST SERB-TARE fellowship, EFIS (European federation of immunological societies) fellowship to attend 5th international conference on CMV and immunosenescence. Currently she is serving as PI in NASF and DST-SERB TARE grants.

Facs in veterinary disease diagnosis

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A fluorescent antibody tagged to the cell surface antigens of cells, can be excited by the laser, and the emitted light is recorded by a second detector system located at a right angle to the laser beam. Fluorescent activated cell sorters (FACS) are flow cytometers that have the capacity to sort fluorescent-labelled cells from a mixed cell population. The dedicated flow cytometry is yet to be introduced into commercial veterinary diagnostic laboratories; however, laboratories in several academic institutions have now accepted the technology and offer a limited service. On the other hand, haematology analysers that incorporate flow cytometric technology have gained widespread use.

Evaluating the percentage and number of CD4 and CD8 T lymphocytes in the peripheral blood by flow cytometry is a very accurate method of assessing the integrity of the cellular immune system. This immune-phenotyping procedure is referred to as lymphocyte subset analysis. A very well-known example of application of flow cytometry comes from the enumeration of CD4 T cells to evaluate the progression of acquired immunodeficiency syndrome (AIDS) in human immunodeficiency virus (HIV) infected human patients, and is the most commonly performed application of flow cytometry in human diagnostic laboratories. In companion animals, peripheral blood T cell subset analysis is typically used to assess immune competence, the effect of drugs on the immune system, and progression of infectious disease. The feline retroviral infections, feline immunodeficiency virus (FIV) and feline leukaemia virus (FeLV), both target the cellular immune system and cause decreases in CD4 T cells and the CD4:CD8 ratio. The decline in CD4 counts or CD4:CD8 ratio, analogous to HIV-AIDS, can be used to assess the progression to FIV-AIDS in field cases. Similarly, Equine infectious anaemia virus (EIAV) is another member of the lentivirus subfamily, together with HIV and FIV. Horses that are asymptomatic carriers of EIAV have a significantly decreased percentage of CD5 and CD4 T lymphocytes in the circulation.

In an experimental model of canine visceral Leishmaniasis, the percentage of peripheral blood CD4 cells is significantly increased following successful treatment for infection, and thus clinical monitoring and progression are efficiently evaluated through flow cytometry. The percentage of CD4 cells is also inversely proportional to the host's subsequent infectivity for the disease vector. Decreased CD8 count and increased CD4:CD8 ratio associated with active canine systemic lupus erythematosus (SLE) reverted with successful immunotherapy. Decrease in granulocyte count and marked lymphocytosis (by Flow cytometric analysis) in infected cattle compared to clinically healthy cattle has been shown as an hallmark in suspected cases of Theleriosis in cattle. Immunophenotyping by flow cytometry has been shown to accurately diagnose cases of acute and chronic leukaemia in the cat, dog, cow and horse. Flow cytometry, using fluorescent-tagged antibodies to detect cell-bound complement and/or immunoglobulin, shows good sensitivity for detection of immune-mediated anaemia and thrombocytopenia. We have observed diminished representations of $CD4^{+}CD62L^{+}$ and $CD4^{+}CD44^{+}$ cells in peripheral blood of chronically diarrhoeic MAP sero-positive cattle suggesting that downregulation of these adhesion molecules might play a role in dysregulated immune responses relating to defective antigen clearance in MAP sero-positive animals.

Practical considerations have limited the development of flow cytometry in veterinary medicine, largely due to the substantial cost of the machine and the restricted availability of reagents and the typically low number of leukemic animals treated at veterinary hospitals at any one time prevents the batching of samples to improve cost efficiency. Furthermore, the sophisticated methodology requires a highly trained technician to prepare and run the samples, and a specialist diagnostician to analyse, interpret, and synthesize data produced by flow cytometry and routine clinic-pathological testing (e.g. cell morphology, full blood examination).

ORAL PRESENTATION

OP_VI.1 Lipidomics approach to investigate canine mammary tumor

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Altered lipid metabolism is crucial for the genesis of cancer. In addition to Warburg effect, the cancer cells display definite lipid reprogramming *via* uptake of large quantities of lipids through enhanced lipogenesis, β-oxidation and cholesterol (CHO) production. Both systemic and intratumoral lipid metabolism play pivotal roles in tumor development as well as progression, hence investigating the changes in lipid metabolism and storage in canine mammary tumor (CMT) stands crucial. In the present study, the intratumoral non-polar lipid profile in canine mammary tissues (both healthy and tumor tissues, (n=4-11) was generated using Thin Layer Chromatography (TLC) and semi-quantified by ImageJ software. Furthermore, the levels of total lipids, triacylglycerols (TAGs) and cholesterol (CHO) in these tissues were quantified in colorimetric assays. In addition, serum biochemical parameters including, serum triglycerides, cholesterol, lipase, total proteins and glucose levels were also estimated. Results revealed significantly higher levels of TAGs, total lipids, and cholesterol in tumor tissues as compared to the healthy ones ($p<0.05$), indicating abnormal accumulation of lipids in the tumor microenvironment. Further, the TLC based analysis confirmed the presence of different neutral lipids representing CHO-esters, methyl-esters, TAGs, oleic acid and CHO with a significant increase in the amount of TAGs and CHO-esters in CMT tissues as compared to the healthy ones ($p<0.05$). Likewise, serum levels of TAGs, CHO and glucose as well as activity of lipase were also concomitantly increased significantly ($p<0.001$) in CMT dogs as compared to normal dogs. The levels of total proteins in these dog sera were found unaltered. These findings are suggestive of a critical role of lipids in cancer progressiveness that paves a way for further investigation.

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OP_VI.2 Nanoiron formulation in mitigation of iron deficiency anemia in rat model

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Iron plays a significant role in various biological functions like hemoglobin/myoglobin synthesis, DNA synthesis, electron transport system and many other anabolic processes. Its deficiency in the diet can lead to several clinical problems like weakness, exercise intolerance, retarded growth, and lethargic conditions. Usually, the oral supplementation of iron possesses limited bioavailability and is associated with several health problems. To overcome this, we synthesized IONP, IONP-Asc, and INOP-FA to improve the biodistribution, safety, and therapeutic efficacy in experimentally induced iron deficiency anemic (IDA) rat models. Our results showed IONP is more efficient in treating IDA than ferrous sulphate. Further, the IONP-Asc is found to be more stable than IONP-FA. Also, IONPs do not exhibit any toxicity during hemolytic assay and the histological examinations showed no organ toxicities of IONPs. IONP-Asc restored better hematological and biochemical changes as compared to IONP-FA in the IDA model. This study shows that INOP-Asc can be a safe therapeutic agent in treating IDA.

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OP_VI.3 Tear film as an easily accessible bio fluid for biomarker detection in ocular surface diseases of dogs

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Tear based biomarkers were explored for ocular surface diseases of significance and hence are an important tool for the easy and non-invasive detection of many such diseases in pets. Keratoconjunctivitis sicca (KCS) commonly known as dry eye disease is a cause of major ocular surface pathologies in dogs. They can lead to prolonged corneal surface inflammation, corneal opacity and in extreme cases corneal ulcer or perforation may occur leading to loss of vision. The techniques for identification of tear proteins can vary from immunoblotting which is a semi-quantitative method, ELISA for quantitative detection and tear fluid proteomics using Mass Spectrometry for biomarker mining.

Matrix-assisted laser desorption/ ionisation time of flight tandem mass spectrometry (MALDI-TOF MS) was used to characterise peptide components of tear collected from dogs suffering from early and late stages of Keratoconjuctivitis sicca (KCS). Tear samples were obtained from healthy dogs, dogs suffering from early and late stages of KCS (n=6) and were pooled into two aliquots (n=3). One dimensional SDS PAGE (12%) was used to separate intact tear proteins (30 µg in each well) into different bands. Two aliquots of three major bands 15Kda, 24 Kda and 66Kda were cut, each fraction was trypsin digested and analysed by MALDI TOF/TOF MS to characterize the protein components in each band. Total number of proteins identified in normal tear was 359, 228 in early KCS and 238 in late KCS. Differential expression of various inflammatory biomarkers in tear fluid was detected post corneal grafting with porcine small intestinal submucosa in dogs. Tear fluid cytokines MMP-9 and IFN-gamma was found upregulated through ELISA and immunoblotting in tear fluid of KCS affected dogs. The present study suggested the potential of tear fluid for biomarker discovery for potential ocular surface diseases in dogs.

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OP_VI.4 Assessment of somatic cell count and lactate dehydrogenase in the milk of cattle and buffaloes

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During mastitis, cell damage leads to an increase in the activity of lactate dehydrogenase (LDH), which can be used as an indicator of mastitis. However, there is no reliable data available about the levels of LDH in the milk of healthy animals. The availability of such values would help anyone to adjudge the status of healthy animals as well as those suffering from udder infestations. The study was conducted to carry out an analysis of LDH levels in milk and their correlation with somatic cell count (SCC). For this, milk samples were collected from 176 quarters of cattle and 200 quarters of murrah buffaloes from the dairy herds of the Lala Lajpat Rai University of Veterinary and Animal Sciences, Hisar, Haryana. Out of 176 cattle milk samples, 68 were from Sahiwal, 28 from Haryana and 80 from the Hardhenu breed. The milk samples were subjected to somatic cell count. The skim milk was separated by centrifugation of milk samples at 2000 x g for 10 minutes, followed by its collection after puncturing the cream layer. The lactate dehydrogenase activity was measured from acellular milk with the help of a clinical analyzer. Results showed that with an increase in the somatic cell count, LDH levels also increased from 224.77±7.48 to 377.67±20.28 in cattle and from 547.35±13.05 to 1233.00±357.00 in buffaloes. Thus, the LDH levels can also be used as a marker for clinical mastitis, which can be further studied with a larger number of samples.

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OP_VI.5 Flow cytometricimmunophenotyping of PBMCs and altered expression of adhesion molecules in *Mycobacterium avium* subspecies *paratuberculosis*(MAP) seropositive diarrhoeic bovines

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MAP, (*Mycobacterium avium*subspecies*paratuberculosis*), infection causes chronic, persistent granulomatous enteritis resulting in long-standing diarrhoea. Clinical cases of diarrhoea are treated with classical anti-diarrhoealsin the clinics; however, a significant number of animals do not respond to therapy. This state of non-responsiveness to anti-diarrhoeals could be due to immune-incompetence. Cellular immune responses play a major role, with little or no role of humoral immune responses in protection againstmycobacterial infections. Thus we undertook the study to estimate hosts cellular immune responses in MAP suspected natural cases of diarrhoeic cattle. Although the expression of CD62L and CD44 on bovine T cells has been studied in severalinfections in cattle, but, majority of these studies have been done in experimental model systems. No reports are available on regulation of adhesion molecules on overall peripheral blood mononuclear cells (PMBCs) in MAP seropositive diarrhoeic bovines in natural clinical settings.Blood and fecal samples were collected from 14 suspected, chronically diarrhoeic, cachexic and emaciated

cattle from stray cattle shed (Gaushala). MAP status was ascertained by iELISA (plasma), PCR and acid fast staining (fecal). Blood and fecal sampling was also done from 5 apparently healthy, non-diarrhoeic cattle (from gaushala). For flow cytometry measurement of the PBMCs, $1x10^5$ cells were plated in round bottom 96 well plates. Samples were incubated with CD4-RPE, CD8-FITC, CD44-FITC and CD62L-FITC purchased from BIO-RAD Laboratories, for 30 min. All samples were collected on the cytoFLEX flow cytometer (Beckman coulter), and data were analyzed using FlowJo software (BD biosciences). We observed diminished representation of $CD4^+CD62L^+$ and $CD4^+CD44^+$ cells in peripheral blood of chronically diarrhoeic MAP sero-positive cattle suggesting that downregulation of these adhesion molecules might play a role in dysregulated immune responses relating to defective antigen clearance in MAP sero-positive animals. High expression of CD44 represents activated leucocytes, thus decrease in the frequencies of $CD4^+CD44^+$ cells in the CD4 fraction could mean that lesser proportion of activated cells are circulating in the peripheral blood in MAP seropositive animals. Overall our data suggests dysregulated peripheral cellular immune responses (which are scarcely studied in bovines), in MAP seropositive animals.

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OP_VI.6 Serum biochemical markers as indicators of severity in pyometra-affected bitches: Insights from clinical and histopathological examination

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Pyometra is the most commonly documented reproductive disorder in intact bitches. The objective of this study was to illustrate the diagnostic and prognostic importance of key serum biochemical parameters in pyometra affected bitches. Based on the vital clinical parameters, pyometra bitches were classified into pyometra with SIRS (P-SIRS+, n= 28) and without SIRS (P-SIRS-, n= 24). Surgical treatment was performed in 25 bitches. Retrospective categorization of uteruses were done into normal (n=12), CEH (n=3), mild to moderate CEH-P (mCEH-P, n=9), severe CEH-P (sCEH-P, n=6) and atrophic pyometra (AT-pyometra, n=7) based on histopathology. A group of age matched (n=12) bitches without pyometra served as control. Significantly higher concentration of BUN and creatinine were estimated in AT-P than other groups. Similarly aspartate amino transferase (AST) concentration was significantly higher in AT-P. Alanine aminotransferase concentration and alkaline phosphatase (ALP) was significantly higher in sCEH-P. ELISA of SLPI revealed significantly high concentration in the sCEH-P and AT-P bitches than control. Similarly endometrial transcript expression of SLPI also revealed significant up regulation in mCEH-P, sCEH-P and AT-P relative to the control. The mean BUN concentration (mg/dL) was significantly higher in the P-SIRS+ than the P-SIRS- and control. Serum SLPI concentration was significantly greater in the P-SIRS+ bitch than the P-SIRS- bitch and control. The endometrial expression of SLPI was abundantly upregulated in the P-SIRS+ whereas moderate upregulation observed in the P-SIRS- relative to the control bitch. The mean creatinine concentration (mg/dL) was also significantly higher in the P-SIRS+ bitch compared to the control. The bitches were retrospectively classified into survivor and dead. A significant higher BUN and creatinine concentration observed in the animal that died later than those survived. The serum concentration and endometrial expression of SLPI was significantly increased in the bitch that died compared to those survived. In conclusion BUN, creatinine, ALT, AST, and ALP, exhibit significant variations across different grades and severities of pyometra affected bitches. Significant upregulation of SLPI in the peripheral circulation and in the endometrium of non-survivor bitch suggesting its possible association with the prognosis of pyometra led SIRS in

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OP_VI.7 Role of Hepcidin as a biomarker for anaemia in pregnant bitches

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Hepcidin, has emerged as the master regulator of systemic iron balance and plays a critical role in development of anemia. Elevated hepcidin levels inhibit intestinal iron absorption and macrophage iron recycling, leading to iron-restricted erythropoiesis and anemia. In canines, pregnancy related anemia is frequently encountered due to an increased demand of micro-minerals especially iron both by the pregnant animal and the growing fetus. The present study aimed at studying the association of maternal serum iron levels and serum hepcidin in pregnant bitches while also studying the expression pattern of canine hepcidin gene in pregnant bitches. This longitudinal study involved twenty female dogs, divided into two groups: 10 healthy, non-pregnant adults as the

control group, and 10 pregnant females in mid- and late-gestation as the experimental group. Physio-clinical examinations were performed to assess the health status of the animals. Blood samples were collected for hematological and biochemical analyses. The results indicated that pregnancy stages did not significantly affect the animals' physiological parameters. However, during mid- and late-gestation, there was a significant reduction in the hematological profile Liver function parameters, were significantly lower in the experimental group during mid- and late-gestation. Anemia profiling indicated increased serum iron levels in both stages of gestation, while transferrin saturation and total iron binding capacity showed a marked increase during late-gestation. Serum hepcidin levels were significantly lower in late-gestation. Furthermore, molecular analysis of the hepcidin gene revealed its conservation in all canine samples, and qPCR analysis showed a significant decrease in hepcidin gene expression during late-gestation. In conclusion, serum hepcidin has the potential to serve as a diagnostic marker and possible alternative for extensive anemia profiling during pregnancy in canines.

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OP_VI.8 Study of lipid pattern changes in obese dogs

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The study was undertaken to investigate the differences in serum fatty acid composition between normal and obese dogs. The blood samples were collected from dogs that were brought for vaccination to the Small Animal Outpatient Unit of Madras Veterinary College, Tamil Nadu Veterinary and Animal Sciences University. Prior consent from pet owners of all the animals that participated in the study was taken and they were informed regarding authorizing the collection of materials and use of the data in publications. Initially, the dogs were weighed and then subjected to body condition scoring (BCS). As per this system, those animals with BCS scores of 4 to 6 are classified as ideal weight and those with BCS scores above 8 are considered obese. Those animals that had BCS 6-8 were omitted for the study, to have a clear demarcation between the groups. So, a total of 24 dogs were grouped into two groups. Using BCS scores, 12 dogs of ideal weight with an equal number of male and female dogs were put into a group and the remaining 12 obese dogs with an equal number of male and female dogs were pooled in another group. Free fatty acids in serum were estimated using gas chromatography. The saturated fatty acid hexadecanoic acid (palmitic acid) levels were significantly higher in obese dogs when compared to normal dogs. The unsaturated fatty acid docosahexaenoic acid (Cervonic acid) levels were significantly lower in obese dogs when compared to normal dogs. These findings are in conjunction with similar results obtained in human subjects.

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OP_VI.9 Assessment of Hematological Changes in Donor Cow Blood Stored in CPDA-1 Bags for Transfusion: A Case Study

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The current study's goal is to evaluate the haematology of cattle blood stored in plastic bags containing CPDA-1. 100 milliliters of whole blood from six healthy, mature female cows were donated. This blood was then put in a bag together with 14 milliliters of CPDA anticoagulant, an anticoagulant designed to preserve and store human whole blood for an extended period of time. The bags were stored between 2°C and 6°C for 42 days. Hemoglobin concentration, packed cell volume (P.C.V.%), red blood cells, leukocytes, platelets, MCH, MCHC, and MCV, among other hematologic parameters, were measured at the following intervals: days 0 (shortly after collection), 7 (7 days later), 14 (shortly after 14 days), 21 (shortly after 21 days), 28 (shortly after 28 days), 35

(shortly after 35 days), and 42 (shortly after 42 days). Up until Day 35, the TEC was constant. However, hemolysis was the cause of the sudden rise in Hb concentrations from Day 35 to Day 42. Day 35 to Day 42 saw a change in the percentage of PCV, while Days 7 to 42 saw a considerable decline in MCH and MCHC. The WBC levels significantly decreased between Day 21 and Day 42. The platelets were incredibly unstable on Day 7, when they started to diminish. Donor blood can be preserved in CPDA-1 blood bags to facilitate blood transfusions in an emergency since the necessary haematological features can be maintained for up to 25–30 days at 4ºC. There is still room to grow the blood bank beyond veterinary use.

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OP_VI.10 Kisspeptin-Mediated Regulation of Buffalo Ovarian Granulosa Cells: Implications for Reproductive Control and Therapeutic Strategies

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Kisspeptin (Kp) is a pivotal factor in controlling the reproductive system, especially in buffalo ovaries. Our research was designed to delve into the impact of Kp on granulosa cells (GCs) and their activities, with a specific focus on how it affects the production of estradiol (E2) and progesterone (P4), as well as the expression of genes associated with cell proliferation, apoptosis and steroidogenesis. We isolated buffalo ovarian granulosa cells (GCs) from 6-10 mm follicles. These cells were treated with different Kp concentrations (10, 50, and 100 nM) for 48 hours, with a control group receiving no Kp. Estradiol (E2) and progesterone (P4) levels in the culture medium were then measured using ELISA. Our findings indicate that Kp at concentrations of 50 nM and 100 nM significantly increased the production of estradiol (E2) while decreasing progesterone (P4) production at 100 nM Kp. Furthermore, when GCs were treated with 100 nM Kp, we observed the activation of genes related to gonadotropic receptors (FSHR and LHR), steroid synthesis (STAR, 3βHSD, CYP19A1), and cell proliferation (PCNA). Notably, Kp at this concentration also stimulated the proliferation of GCs, as evidenced by a substantial increase in the expression of BCL2 (5.0-fold) and PCNA (94.9-fold) genes. Additionally, we detected elevated levels of p-ERK1/2, a signaling molecule, in Kp-treated GCs. In summary, our study demonstrates that Kp at 100 nM enhances E2 production through the activation of ERK1/2, STAR, and CYP19A1, while concurrently promoting GC proliferation by regulating the expression of BCL2 and PCNA. Future research focusing on Kp antagonists may uncover the underlying mechanisms responsible for Kp-induced steroid synthesis in GCs. This insight holds promise for developing therapeutic strategies to control ovarian function and address related disorders.

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OP_VI.11 Effect of *Murraya koenigii* Extract Supplementation on Diabetes in Experimental Diabetic Rats

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Many species of plants and herbs are known to act as anti-diabetic agents, but only a few of them have been investigated. *Murraya koenigii* (family, Rutaceae), popularly known as curry leaf is a medicinal plant that grows throughout the greater parts of India and South East Asia. This species is known to possess anti-inflammatory, antidysenteric, antioxidant, antidiabetic and diverse pharmacological properties. Several studies also shown that curry leaf decreased blood glucose significantly in different animal models. The effect of daily oral administration of aqueous extract (600 mg/kg b.wt.) and methanol extract (200 mg/kg b.wt.) of *Murraya koenigii* Spreng leaves for a period of eight weeks was studied on blood glucose and plasma insulin level in experimentally induced diabetes in rats. Blood glucose levels of diabetic rats treated with aqueous and methanol extracts of *Murraya koenigii* Spreng showed significant reduction ($P<0.05$) as compared to diabetic control groups. Plasma insulin showed significantly high on 43rd and 58th days of treatment in aqueous and methanol extracts of *Murraya koenigii* treated groups. This suggests that the hypoglycemic effect may be mediated through stimulating insulin synthesis and/or secretion from the beta cells of pancreatic islets of Langerhans.

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POSTER PRESENTATION

PP_VI.1 Epidemiological Insights into Canine Renal Insufficiency: A Decade-Long Study at Bihar Veterinary College's Veterinary Clinical Complex, Patna (2012-2023)

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One of the main causes of disease and mortality in dogs is renal insufficiency. The objective of this study was to determine the incidence of renal insufficiency in dogs at the Bihar Veterinary College's Veterinary Clinical Complex in Patna between May 2012 and November 2023. Age, gender, and season were among the epidemiological variables examined to clarify the trends related to this illness. Eligible subjects for this study included canines that showed up to the clinic with clinical signs of renal problems, such as changes in urine production (dysuria, anuria, and urinary incontinence), increased frequency of urination, vomiting, and abdominal discomfort. Kidney function testing definitively established the diagnosis of renal insufficiency. Over the course of this ten-year study, 5,700 dogs visited the Veterinary Clinical Complex of Bihar Veterinary College in need of veterinary care. 10.96% of these instances had creatinine levels between 2-5 mg/dL, 4.59% had levels between 5-10 mg/dL, and 6.29% had values above the 10 mg/dL threshold. It was observed that male dogs had significantly higher susceptibility to renal insufficiency than female dogs, and that older dogs ($p<0.05$) were more likely to have the illness. Seasonal fluctuation revealed a sharp rise in incidence of renal failure higher being in post-monsoon and lower in winter season. Considerable reductions in packed cell volume, considerable drops in haemoglobin levels, and a dip in total erythrocyte count were observed in the haematological profiles of the affected canines. Moreover, there was a notable decrease in total protein level relative to the healthy control group, and a significant increase in serum urea nitrogen, creatinine, and phosphorus levels ($p<0.05$). Thus, it may be inferred that changes in the canine body's biochemistry and haematology can serve as diagnostic markers for kidney insufficiency in dogs, along with the predisposing factors viz. age, sex and season.

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PP_VI.2 Pregnanediol-3α-glucuronide in Buffalo Urine: A Surrogate for Serum Progesterone Levels

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Pregnanediol-3α-glucuronide (PdG) is a steroid hormone metabolite that is elevated in the urine and serum during pregnancy due to the increased production of progesterone by the placenta. In the present study, PdG levels were analysed in serum and urine samples from synchronized breeding Murrah buffaloes (n=40) selected from the herd maintained at ICAR CIRB, Hisar on days 0 (day of estrus), 10, 20, 30, 35, 45, and 65 to quantify PdG concentrations-using immunoassay and high-pressure liquid chromatography (HPLC) simultaneously. Pregnancy was confirmed in 17 buffaloes, on day 30 using ultrasonography. On day 0, there was non-significant ($p<0.05$) difference in the mean PdG levels values of both serum and urine in pregnant buffalo (21.66±1.88 ng/dl,15.32± 0.94 ng/dl) and nonpregnant buffaloes (19.03±1.03 ng/dl, 13.07± 1.08 ng/dl). However, from day 30 onwards the values of PdG in both serum and urine were significantly higher ($p<0.05$) in pregnant buffaloes (30^{th} day 72.05 ± 2.18 ng/ml and 49.32 ± 1.45, 35^{th} day 60.65 ± 2.10 ng/ml and 54.38 ±2.09, on 45^{th} day 83.08 ± 1.45 ng/ml and 62.76 ± 3.11 and on 65^{th} day were 95.01 ± 1.56 ng/ml and 72.32± 3.26 ng/ml respectively) than in non- pregnant buffaloes (30^{th} day 32.76± 1.75ng/ml and 28.34 ± 1.33 ng/ml, 35th day 28.50 ± 1.02 ng/ml and 22.09 ± 2.19 ng/ml and on 45^{th} day were 21.09 ± 3.07 ng/ml and 26.00 ± 1.09 ng/ml respectively). PdG concentration in pregnant animals increased sharply till day 65 when sampling was terminated. Based on the results it may be concluded that PdG in buffalo urine could be used as a potential marker for developing a non-invasive method for reproductive assessment of dairy animals. Therefore, present study has shown that measuring PdG levels in urine and serum can be used as a surrogate for serum progesterone levels for diagnosing and monitoring animal pregnancy.

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POSTER PRESENTATION

PP_VI.3 Hematobiochemical findings in clinical cases of colitis in horses

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In this retrospective study we have analysed haematological and biochemical data of 5 clinical cases of colitis in horses those could not be survived. Out of these 4 cases occurred during the use of parental injections of Ceftriaxone and salbactum combination. While one mare suffered from colitis, whose foal was being treated for with Azithromycin and Rifampicin since last 20 days. Severe abdominal pain, rolling, flank watching, watery foul smelling diarrhoeas, increased heart rate, respiration rate were most common signs in these cases. Blood was taken immediately after appearance of clinical signs. Total leukocyte count was observed severely decreased with increased in neutrophils count, haemoglobin, packed cell volume and total erythrocyte count were found significantly increased. In the biochemical data serum total protein and albumin were found significantly decreased. Significant increase was observed in the serum levels of SGPT, SGOT and ALP. Findings of colitis cases were in agreements with the previously reported studies reported decrease in leucocytes count with neutrophilia. In colitis due to severe inflammation in colon, large amounts of leukocytes migrate towards the inflammatory site in the colon and that leads to sudden decrease in total leukocyte count. Findings suggested that use of broad spectrum antibiotics especially ceftriaxone and salbactum combination in horses should be used with extreme care.

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PP_VI.4 Cloning, Prokaryotic Expression and Partial Characterization of *Ovis aries* C ysteine-rich secretory protein-1

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Cysteine-rich secretory proteins-1 (CRISP-1) is an acidic glycoprotein of epididymal origin which is reported to bind to sperm surface and inhibit capacitation in epididymal sperm. In the present study, the putative mature peptide of ovine CRISP-1 protein was expressed in *E. coli*, purified and partially characterized. The cDNA corresponding to T^{23} - C^{242} peptide fragment of ovine CRISP-1 protein was cloned in to pET32b(+) expression vector. The recombinant vector containing the CRISP-1 gene was transformed into *E. coli* DH5α. The recombinant plasmid isolated from positive transformants was used to transform *E. coli* BL21(DE3) and protein expression was carried out by induction with 1 mM IPTG at 37 °C for 4 h. The recombinant protein was expressed as insoluble inclusion bodies which was purified by Ni-NTA affinity chromatography using pH gradient. Further purification of the protein was carried out by elution from SDS-PAGE gel following zinc sulphate negative staining. The partially purified recombinant CRISP-1 protein was analyzed by SDS-PAGE and appeared as a distinct band of approximate molecular weight 43.8 kDa. The activity of the purified CRISP-1 protein was assayed on sperm motility and capacitation. The ovine CRISP-1 protein caused significant inhibition of sperm motility at 5 µg/ml and the activity was lost after heating the protein at 100 °C for 5 min. The recombinant CRISP-1 protein also exhibited decapacitating activity and significant reduction of capacitation was demonstrated at 2 µg/ml.

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PP_VI.5 Oxidative status transitions in strongyle infected goats

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The present study was aimed to investigate the alteration in haemato-biochemical and oxidative stress biomarkers in goats naturally infected with strongyle. Twelve adult goats naturally infected with strongyle with egg per gram count in between 500 to 3100 and twelve healthy goats having zero EPG count were selected from Amanala goat farm, Jabalpur (M.P.) for blood samples analysis. Haematological findings showed that naturally infected goats exhibited microcytic hypochromic anaemia and lymphopenia. Serum biochemical investigation reveals significant

(P <0.05) decreased of serum total protein (TP), albumin and calcium level as compared to non infected control animal. A significant elevation observed in ALT (Alanine aminotransferase), AST (Aspartate aminotransferase), ALP (Alkaline phosphatases), ACP (Acid Phosphatase), total cholesterol, blood urea nitrogen and creatinine of infected goats while no significant difference were observed in the serum electrolyte level of inorganic phosphorus (Pi), potassium (K), sodium (Na). In oxidative stress markers Superoxide dismutase serum concentration was decreased, whereas lipid peroxidation (MDA) and catalase (CAT) level was increased in the infected animals than control. The oxidative stress markers were also altered, with a significant decline in total antioxidant capacity (TAC) and reduced glutathione (GSH) levels. In conclusion, it has been demonstrated that strongyle infection in goats resulted in haemato-biochemical alteration with stimulation of oxidative stress, thus can be the good indicators pertaining to worm load in animals.

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PP_VI.6 Influence of mustard oil supplementation in diabetic dogs

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Mustard oil contains a higher amount of unsaturated fatty acids and is well known for its medicinal utility. Long-term mustard oil consumption has been associated with prevention of dyslipidemia, coronary artery diseases, atherosclerosis, colon cancer and hyperglycemia. The hematological, serum biochemical and serum fatty acid changes in diabetic dogs treated with insulin and mustard oil were investigated in this study. Six healthy dogs were treated as healthy group. Prior consent from pet owners of all the animals that participated in the study was taken and they were informed regarding authorizing the collection of materials and use of the data in publications. Twelve diabetic dogs that came for treatment at Madras Veterinary College clinics were divided into two groups with six dogs in each group. Among the two groups of diabetic dogs one group was treated with insulin alone and the other group was treated with insulin along with oral administration of mustard oil. Insulin was administered subcutaneously at the dose rate of 0.2 to 1 U/kg body weight once daily. Mustard oil was administered at the dose rate of 0.25g per kg body weight. The study was carried out for a period of sixty days. There was a reduction in blood glucose, HbA1c levels and an increase in body weight in mustard oil-treated diabetic dogs. This indicates the anti-hyperglycemic activity of mustard oil. The unsaturated fatty acids like linoleic and linolenic acids were significantly higher and the levels of saturated fatty acids viz. myristic, palmitic, stearic arachidic and behenic acids were significantly lower in the serum of diabetic dogs treated with insulin and mustard oil when compared to diabetic dogs treated with insulin alone. The beneficial role of mustard oil is due to the higher amount of unsaturated fatty acids present in mustard oil. Thus, this study demonstrated that unsaturated fatty acid supplementation possess beneficial effects on insulin sensitivity and is likely to reduce the risk of type 2 diabetes.

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PP_VI.7 Evidence of lumpy skin disease virus infection in camels

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Countries in the Indian subcontinent are currently facing a deadly epidemic of lumpy skin disease (LSD). LSD is primarily a disease of cattle. Buffaloes may sometimes develop mild illness; however, other domestic animals are considered resistant to LSD. We confirmed the LSDV infection in camels as evidenced by skin nodules on the body surface of the affected camels, isolation of LSD virus (LSDV) and amplification of LSDV-specific gene segments from the skin nodules (PCR), nucleotide sequencing of the viral genome and, demonstration of anti-LSDV antibodies in serum. Phylogenetic analysis based on nucleotide sequencing of ORF011, ORF012 and ORF036 revealed that the virus (LSDV/Camel/India/2022/Bikaner) is related to the historical NI-2490/Kenya/KSGP-like field strains which are predominantly circulating in the Indian subcontinent. This is the first report wherein LSDV has been to infect camels.

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POSTER PRESENTATION

PP_VI.8 Protein profiling of saliva in delayed ovulatory condition of jersey crossbred cows

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The production potential of crossbred cattle is limited by inherent reproductive issues such as delayed ovulation and a low conception rate, which results in significant financial loss for farmers. Therefore, it is critically necessary to focus research efforts on the development of a suitable diagnostic assay for detecting delayed ovulation in Jersey crossbred cows. The goal of the current study is to investigate salivary protein profile during estrus in Jersey crossbred cows with delayed ovulatory condition. In this study, ten numbers each of normal cyclical (Group I) and delayed ovulation (Group II) Jersey crossbred cows presented to Large Animal Gynaecology Ward at Madras Veterinary College Teaching Hospital, Chennai for treatment and artificial insemination, were selected. Estrus detection was carried out by owner's history, rectal palpation and ultrasonography. The saliva samples were collected during estrus for total protein estimation by Bradford assay and protein profiling using SDS-PAGE and mass spectrometryas per standard protocol. The salivary protein concentration was numerically higher in group I compared to group II. The SDS-PAGE electrophoresis of salivary proteins revealed that the proteins of molecular weight 15, 20, 23, 27, 70 and 84 kDa was predominantly expressed during estrus in both the two groups of crossbred cows, which was further identified as estrus specific or marker proteins. These proteins were sequenced using liquid chromatography mass spectrometry (LC-MS), revealing 41 proteins exclusively expressed in the estrus phase. The present findings suggest that the proteomic approach employed to identify proteins in saliva during the estrous cycle may serve as a valuable tool for screening delayed ovulation in Jersey crossbred cows.

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PP_VI.9 Comparative study of Lipid Droplet-Associated Protein, perilipin2 in Canines and Humans

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Intracellular lipid droplets (LDs) are vital organelles that store neutral lipids, act as source for energy, manage oxidative stress, and coordinate cellular signaling. A key protein family associated with LDs is the perilipins, consisting of 5 members (PLIN1 to 5) with perilipin 2 (PLIN2) being the most prominent having an important role in LD homeostasis. The aim of the study is to evaluate the differences between canine and human PLIN2 in order to identify structural and functional similarities. With the help of bioinformatics tools and information available in various databases, the genomic data for PLIN2 in both species were curated, revealing location, transcript variants, exons, and introns. Multiple sequence alignment of mRNA results demonstrate 85% identity between canine and human PLIN2 showing well defined conserved regions which might be essential for structural and functional roles. Furthermore, amino acid composition analysis revealed similarities in charge residues, stabilizing and destabilizing residues. Despite their structural similarities (72% common secondary structure elements), a 3D structure comparison indicates a significant differences in structural conformation. Physicochemical properties, including stability indices, were found to be distinct, possibly influencing their interactions with lipid droplets and other signaling molecules. In conclusion, dissecting the functional role of PLIN2 will offer a more comprehensive understanding of LD biology and also hold promise for the development of treatments for LD-related disorders, across the species.

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POSTER PRESENTATION

PP_VI.10 Perilipin 2 deletion impairs LDs metabolism and cancer progression

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Lipid droplets (LDs) are specialized subcellular organelles responsible for storing neutral lipids and play a crucial role in preserving cellular lipid homeostasis. LDs are coated with various proteins, with the perilipin-ADRP-TIP47 (PAT) family being the predominant structural proteins. This family comprises of five members, with perilipin 2 (PLIN2) playing a pivotal role in the formation and stability of LDs. PLIN2 plays a role in the development and progression of several cancers, including breast cancer. However, the precise biological function of LD-associated PLIN2 in cancer remains a subject of research. In present study, we utilize the MDA-MB 231 cell line, a triple-negative breast cancer (TNBC), to investigate the deletion of PLIN2 gene on LD metabolism and its potential impact on cancer growth and progression. The PLIN2 gene was deleted from MDA-MB 231 using the CRISPR Cas9 genome editing method. The gene deletion was confirmed both at mRNA, and protein levels, exhibiting a significant reduction in PLIN2 expression. Furthermore, PLIN2 KO cells showed reduced LD storage, increased lipid hydrolysis, and also up regulation of the autophagy marker LC3B. Deletion of PLIN2 led to an increase in PLIN3 expression, suggesting a compensatory mechanism for PLIN2 loss. The loss of PLIN2 had a non-significant impact on cancer cell proliferation and migration which might be due to the compensatory increase in PLIN3 levels. To conclude, our data shows an intricate role of PLIN2 in LD dynamics, further detail study is warranted to fully comprehend its potential implication in cancer progression.

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PP_VI.11 The influence of select dietary trace mineral levels on hematobiochemical parameters, liver and kidney function attributes and serum hormones in male goats under thermoneutral and heat stress conditions

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The study was carried out to assess the effect of select dietary trace mineral (TM) levels under thermoneutral [control group TC; 69.75 THI (thermal humidity index)] and heat stress (HS; 85.47 THI) conditions for a period of 40 days on hematobiochemical parameters, liver and kidney function attributes and serum hormones in male goats. For this, 22 male goats (21.35±0.57 kg BW) were distributed into four groups. Goats in TC-TM1 and HS-TM1 were fed a diet containing Se, Zn, Cu and Mn at 0.19, 34.36, 8.82 and 36.13 ppm, respectively. The dietary levels of Se, Zn, Cu and Mn in HS-TM2 were 0.49, 54.36, 18.82 and 56.13 ppm and in HS-TM3 it was 0.79, 74.36, 28.82 and 76.13 ppm, respectively. Blood samples were collected at the end of the experiment. There was no effect on serum glucose, total protein, albumin, globulin, urea nitrogen and creatinine. However, blood Hb was lower ($P<0.05$) and serum ALT and AST were higher ($P<0.05$) in HS-TM1 as compared to other groups. Serum hormones namely, insulin and T_4 were comparable among the groups while serum T_3 was higher ($P<0.05$) in HS-TM2 and serum cortisol was higher ($P<0.05$) in HS-TM1 group compared with other groups. It is concluded that feeding higher levels of Zn (61.33 ppm), Cu (18.82 ppm), Mn (56.13 ppm) and Se (0.511 ppm) was beneficial for mitigating heat stress by improving serum T_3 and hepatic health and lowering serum cortisol levels in male goats under heat stress conditions.

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POSTER PRESENTATION

PP_VI.12 Studies on Blood Biochemical Profiles of indigenous free ranging donkeys (*Equus asinus*) in and around Hassan, Karnataka, India

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Donkeys have been close companions to humans for the millennia, and have been used as working animals all over the world. The free ranging donkey is a very important animal resource of nomads in around Hassan district, Karnataka. Donkeys living in the sub-tropical region are subject to the effects of various environmental factors that can alter their physiological and clinical parameters, which is particularly significant in free ranging indigenous donkeys. Very little has been done to establish reference values and there is paucity information in this regard for free ranging donkeys and mules in India. Present study reports the plasma biochemical profiles of indigenous donkeys in and around Hassan, Karnataka, India. The glucose level (mg/dl) ranges from70 to 85 with a mean value of 80.5±3.42, total cholesterol (mg/dl) from 23 to 85 with mean of 80.6±4.03, triglyceride (mg/dl) from 48 to 74 with mean of 62.6±9.32, HDL-Cholesterol (mg/dl) from 41 to 49 with mean of 46.2±3.34, creatinine (mg/dl) from 0.6 to 1.3 with mean of 0.79±0.08, total protein (gm/dl) from 6.0 to 7.5 with mean of 6.75±0.72, albumin (gm/dl) from 1.7 to 4.2 with mean of 2.96±0.29, Urea (mg/dl) from 28.17 to 70.16 with mean of 45.7±4.63, total bilirubin (mg/dl) from 0.3 to 0.6 with mean of 0.4±0.03 and direct bilirubin level (mg/dl) ranges from 0.6 o 1.3 with a mean value of 0.3±0.08. Plasma enzymes levels in this study was SGPT (U/Ltr) from14.2 to 16.33 with mean of 14.91 ±0.26, SGOT (U/Ltr) from 232.2 to 326.6 with mean of 280.3±12.13 and ALP level (U/Ltr) ranges from 258.0 to 386. 81 with a mean value of 340.6±18.14. Data generated may be of use as baseline values for blood biochemical profile of apparently healthy indigenous donkeys under Hassan Agro-climatic conditions.

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PP_VI.13 High fat diet decreases the transcription of BKCa channels and increases that of Gi protein in late pregnant rat uterus

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Hyperleptinemia and leptin insensitivity are two important features of obesity. In the present study, we studied the effects of high concentration of leptin and a high fat diet on β-adrenergic signalingcomponents in rat uterus. Wistar rats were subjected to a diet comprising 40% fat and 1.25% cholesterol for six weeks, both prior to and following mating, spanning the entire pregnancy period. The results revealed a significant elevation in cholesterol levels in both serum and uterine tissues, while triglyceride levels remained unaffected. Notably, rats exposed to the high-fat diet exhibited liver fatty degeneration.The high-fat diet markedly increased the mRNA expression of the Gi protein, a response comparable to that induced by 0.3 µMleptin. Conversely, the high-fat diet had minimal impact on the expression of the Gs protein. Furthermore, the high-fat diet led to a reduction in the transcription of BKCaα and BKCaβ channel subunits, mirroring the effect observed with 0.3 µMleptin.In summary, our findings suggest that high-fat feeding upregulatesGi protein expression and downregulatesBKCaα and BKCaβ channel subunits, aligning with the effects of leptin. This implies that hyperleptinemia and obesity may contribute to relaxant dysfunction in the late pregnant rat uterus.

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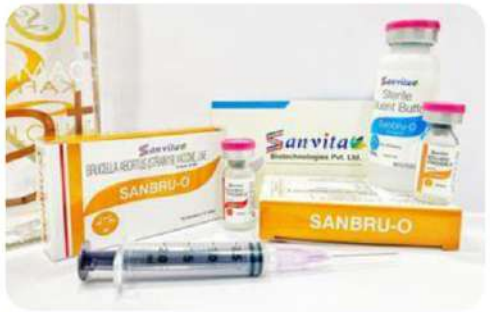

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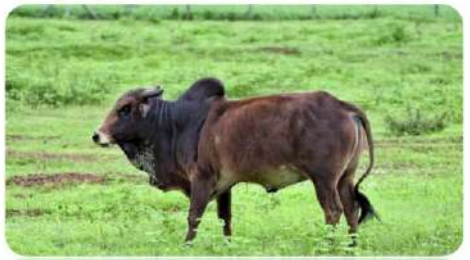

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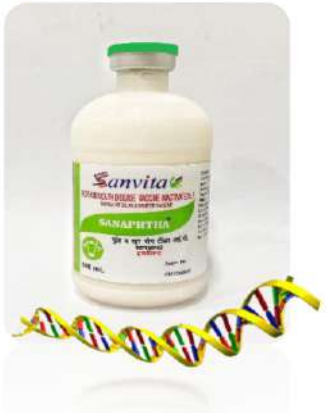

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