Annual Report
2023

भा.कृ.अनु.प. - राष्ट्रीय अश्व अनुसंधान केन्द्र, हिसार

ICAR-National Research Centre on Equines, Hisar

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Dr TK Bhattacharjya
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Sanjay Kumar, Taruna Anand, TR Talluri,
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About the Cover

The cover features Raj Prathama, a Marwari filly, with her surrogate mother, marking a historic achievement as the first in the country born through embryo transfer technology at ICAR-NRCE

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NRC on Equines
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HISAR CAMPUS
ICAR-National Research Centre on Equines
Sirsa Road, Hisar - 125 001, Haryana, India
Phone : 01662-282502, 275787
Email : nrcequine@nic.in

BIKANER CAMPUS
Equine Production Campus
ICAR-National Research Centre on Equines
Shiv Bari, Jorbeer, Bikaner - 334 001, Rajasthan, India
Phone : 0151-2942814

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2023

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ICAR–National Research Centre on Equines


ICAR-NATIONAL RESEARCH CENTRE ON EQUINES

With best compliments from

Dr. TK Bhattacharjya

Director
ICAR-National Research Centre on Equines
Sirsa Road, Hisar - 125001 (Haryana)

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Director's
Foreword

The ICAR-National Research Centre on Equines (ICAR-NRCE), established on November 26, 1985, in Hisar, Haryana, stands as a leading research institution under the Indian Council of Agricultural Research (ICAR) in the field of Animal Sciences exclusively aimed to improve the livelihood of landless and marginal equine farmers by offering diagnostic, advisory, and consultancy services as necessary to improve the equine health, productivity, and utility in agriculture and transportation in difficult terrains of India. These efforts have earned the international recognition worldwide and as a distinguished one among ICAR institutions within the country.

The institute has consistently focused on making significant achievements through basic, applied, and translational research. It has provided trainings and extension services, leading to advancements in equine health and production in India. The Centre's research projects cover areas such as vaccinology, diagnostics, therapeutics, immunology, pathology, parasitology and equine production. Emerging research areas include the development of new-generation vaccines, rapid diagnostic techniques, nanotechnological interventions, host-pathogen interaction studies, ethno-veterinary medicine applications for the direct benefit of stakeholders and the creation of a repository of microbes like viruses, bacteria, and bacteriophages for benefit of researchers and students. Significantly, vaccines namely Lumpi-ProVac^{ind} & ANCOVAX as well as a diagnostic kit for detection of coronavirus infection in animals: CAN-CoV were developed and released by the Centre in the recent past. Additionally, genetic studies on equine production and enhancement of reproductive efficiencies are being conducted. The Centre's research activities bridge the gap between basic biology and clinical applications, providing innovative translational research to improve equine health and welfare in the country. This annual report highlights various research achievements and efforts to produce commercially viable technologies and demand-driven research to benefit equine farmers.

The research and development activities of ICAR-NRCE are achieved through well-structured research programmes comprising 25 institute funded and 25 externally funded research projects, which also include inter-institutional collaborative research projects. ICAR-NRCE has been successful in getting external funding from almost all leading national funding agencies in the field of agricultural and biological sciences. ICAR-NRCE is actively involved in the surveillance of different equine diseases in the country and is of paramount importance for policy-making purposes and for implementing effective control measures for emerging and re-emerging equine diseases. In this direction, during current report of the year 2023, a total of 2280 equine serum samples from 9 states were tested for various diseases like Equine Infectious Anaemia (EIA), Equine Influenza (EI), *Trypanosoma evansi* (Trypanosomiasis), Equine Herpes Virus-1 (EHV-1), Piroplasmosis, Japanese Encephalitis (JEV), *Salmonella Abortus equi* and Brucellosis. The highest sero-prevalence was observed for equine *piroplasmosis* (22.67%) followed by EHV-1 (14.1 %), *Trypanosoma evansi* (1.53%), EI (1.31%) and JE (0.3%). Also a total of 457 samples were tested for various above mentioned equine diseases under diagnostic investigations and 12/53 cases were found positive for EHV1/4 and 4/64 for *T. evansi*.

Under contractual diagnostic services, a total of 7907 samples received from race courses, turf clubs, stud farm, riding schools, animal quarantine & certification services (AQCS) and other organized sectors during the year 2023 were tested for various notifiable and exotic diseases to check ingression of diseases from abroad and monitoring of elite horses in private sectors leading to generation of revenue amounting to Rs 70.15 lakhs. For surveillance of glanders, in year 2023, a total of 43,600 equine sera from 17 states/UT were collected and examined where a total of 117 glanders positive cases were reported in 10 states; however, around 50% of the samples and glanders positive cases were originated from Uttar Pradesh. Out of 193 sera from occupationally exposed humans (veterinary Officers, equine handlers, and laboratory workers) none were found positive. Also, in another study, Hcp1 protein showed quick and strong reactivity against glanders positive serums and IL-1b, IL-6, IL-17, MCP-1 cytokines were found to be significantly higher in glanders positive serum.

The recombinant bivalent constructs of EHV1 and EIV (Florida clade 1&2) by replacing virulence associated genes of EHV1 were generated to develop combined vaccine for EHV1 and equine influenza. Herpervirus, which is a major concern for equines, studies were conducted on standardization of a real-time PCR targeting glycoprotein B gene. The incidence rate for EHV-2, 5 and mixed infection were found to be 28.57%, 35.71% and 14.28% respectively.

Under the "National One Health Program for Prevention and Control of Zoonotic Diseases (NOHPCZ)", NRCE is actively involved in capacity building, strengthening of laboratory diagnostic facilities, inter-sectoral coordination and creating awareness about zoonotic diseases of public health importance by organizing training, workshop and webinar on various zoonotic diseases.

A one-step, probe-based real-time RT-PCR assay for rabies diagnosis was standardized and 50 (nos.) samples of saliva/ brain tissues were tested for rabies from suspected animals and 13 samples were positive through RT-PCR and direct fluorescent antibody testing. In another study the protocol for preparation of *R. equi* whole cell lysate was optimized and characterization of its immunogenic proteins was carried out. The whole cell lysate along with recombinant vapA proteins will be used for immunization study in mouse model and in foals. A fixed cell indirect ELISA using whole cell- *Streptococcus equi* and an RPA assay for sensitive and specific detection of the nucleic acids of *S. equi* are being standardized for diagnosis of strangles. Additionally, a multiplex Taqman qPCR assay intended to be used for *in vitro* detection of *Streptococcus* sps and *Streptococcus equi* subsps. *equi* in respiratory samples has been internally validated. In another study, we identified members of the

Stilbenes group (specifically resveratrol and pterostilbene) in extracts of *Aerva javanica* plant showing anti-proud flesh activity. In field cases of summer dermatitis in horses, it was found that the expression of anti-inflammatory cytokines IL-10 and TGF-b1 may be important targets for therapeutic purpose to control chronic infection.

In a study, barbamine and emetine were found to exhibit anti-trypanosomal activity against *Trypanosoma evansi* in axenic culture. Also, careful selection of adjuvants and delivery systems is being carried out for *T. equi* piroplasms. Simultaneously, *T. equi* antigen detection ELISA using TE/tEMA-2, GST cleaved recombinant proteins is being developed. A skin-specific bioink made from digested chicken/porcine skin and incorporated with polymers has been developed and analyzed for its structure, stability, and compatibility for therapeutic applications. Nanocarriers, like ZnO nanoparticles with flower-like morphology (ZnONFs) decorated with chitosan and hydroxychloroquine (CHCZnO NPs) is being developed as a more potent approach against coronaviruses.

As a significant achievement, for the first time in the country, live Marwari breed foals have been born through embryo transfer technology at the sub-centre EPC Bikaner. Also, zanskari mare was conceived with the transferred embryo and is due for foaling in the year 2024. The primary structure determination and physicochemical characterization of DSP-3, a phosphatidylcholine binding glycoprotein of donkey seminal plasma was carried out. In another study, it was found that addition of melatonin significantly increased T-AOC levels and reduced MDA levels indicating increase in antioxidant levels, reduction in oxidative stress and lipid peroxidation levels. The cryovial method for cryopreservation of stallion spermatozoa, vitrification of horse embryos and cryopreservation of semen from indigenous horses was also carried out. Additionally, detection of SNPs of MSTN gene among indigenous donkey breeds, creation of equine CNV databases were carried out. In a study, biochemical, dielectric and surface characteristics of freeze-dried donkey milk powder were studied. The institute carried out social media usage pattern studies among horse keepers and field veterinarians and it was found that ~ 71.60% of the respondents used social media for finding out news and events whereas 68.70% of the respondents used it for exchange of information.

During the year 2023, a total of 160 microbes were accessioned in the repository thereby the cumulative strength of NCVTC reaching to 2613. The important virus isolates accessioned in the NCVTC repository during the year included, Avian infectious bronchitis (n=2), Fowlpox virus (n=1), Marek's disease virus (n=2), Inclusion body hepatitis virus (aka Fowl adenovirus) (n=1), African swine fever virus (n=1), Bovine Corona virus (n=3), Rous Sarcoma Virus (RAV-1), Duck enteritis virus (n=1), PPR virus (Mutant strain) (n=1), Bluetongue virus (n=15), Sheeppox virus (n=3), Orf virus (n=2) and Lumpy skin disease virus (n=2). The evaluation of safety, immunogenicity and efficacy of Lumpi-ProVac^{Ind}was carried out. Also, miRNA profiling of lumpy skin disease virus infected primary lamb testicle cells was performed to find biomarkers as well as novel targets for therapeutic intervention against LSDV and in this study, miR-29a was found to be a novel biomarker. A novel HRMbased gap-qRT-PCR for identification and quantitation of the vaccine and field strain(s) of lumpy skin disease virus was developed. In another study, Hesperetin was found to block poxvirus replication by competitively inhibiting binding of the 5' cap of viral mRNA with eIF4E. The surveillance of SARS-CoV-2 in waste water in Hisar region was also carried out. Additionally, a novel sub-genotype AL1b of rabies virus in domestic animals across Haryana, India was identified.

In an attempt to identify and isolate bovine coronavirus from respiratory and enteric infections in cattle, the BCoVs were confirmed by RT-PCR. Isothermal "Recombinase Polymerase Amplification" (RPA) coupled with CRISPR Technology and LFA based assay for point-of-care detection of Porcine circovirus 3 (PCV3) in pigs was developed which could serve as a versatile POC platform for rapid detection of PCV3 nucleic acids in pigs. Also, an isothermal RT-RPA-CRISPR-LFA assay was developed for detection of SARS-CoV-2 nucleic acid targeting highly conserved genes of the virus. The mRNA vaccine candidate for SARS-CoV-2 has been evaluated in BALB/c mice and RPA-LFA assay for detection of EHV1 & 4 nucleic acids has been developed. Studies on bovine rota virus, which is a leading cause of calf mortality are also underway for diagnosis, isolation and characterization of viral strains. In the bacteriophage repository, physiological and genomic characterization of bacteriophage

encoded lysins was carried out to explore a promising antimicrobial agent against multidrug resistant strains of *Salmonella* and *E. coli.* During year 2023, a total of 209 bacterial cultures were processed, out of which 115 belonging to 28 taxa were accessioned into the bacterial repository, increasing the total collection of veterinary bacteria to 1857. Bacillus cultures were accessioned under anthrax diagnosis programme. Antibacterial activity of domestic donkeys' milk against *Escherichia coli* and *Staphylococcus aureus* was carried out. Under INFAAR surveillance project, a total of, 217 *E. coli* isolates were characterized and 69 *Staphylococcus* spp., were genotyped. Importantly, *Corynebacterium pseudotuberculosis* was isolated from a field case of caseous lymphadenitis in sheeps and goats. Additionally, from 109 collected samples, 7 nos. acid-fast bacilli were isolated in order to explore mycobacterial infections. CRISPR- Cas9 mediated gene editing in *Mycobacterium kansasii* is being pursued to explore using CRISPR-Cas9 approach to decipher the role of MCC genes in survival of *M. kansasii.*

During the year, scientists published 68 high impact research papers in international and national peer-reviewed journals. In addition, 7 popular articles, 1 technical bulletin, 6 compendium compilations, 2 book chapters, 68 training manual chapters, 30 research abstracts, 42 GenBank accessions and four technologies certified by ICAR.

The vision, guidance, and technical support provided periodically by the esteemed chairmen and members of QRT, RAC, and IRC experts have been instrumental in keeping NRCE on the right path and maintaining its focus. I would like to express my sincere gratitude to the Chairman and Members of the Publication Committee for producing this excellent annual report of the Centre, showcasing the significant achievements. I am deeply thankful for the unwavering support extended to this institute by Dr. Himanshu Pathak, Secretary of the Department of Agricultural Research and Education, and Director General of ICAR, as well as Dr. Joykrushna Jena, Deputy Director General (Animal Sciences). My appreciation also goes to the Assistant Director General, Dr. Ashok Kumar (Animal Health), and the Principal Scientists at ICAR Headquarters for their continuous support to NRCE. Finally, my heartfelt thanks go to the entire NRCE staff members for their wholehearted efforts and contributions to the progress of this premier institute.

I am deeply thankful to Dr. Himanshu Pathak, Secretary, DARE and Director General, ICAR for the unwavering support, direction and guidance in conducting research, education and extension for the benefit of equine and livestock farmers, and stakeholders in the country. I am also indebted to Shri Sanjay Garg, Additional Secretary, DARE and Secretary, ICAR and Ms. Alka Nangia Arora, Joint Secretary, DARE and Finance Advisor, ICAR for whole hearted support to the Institute & to aspirations of equine and other livestock farmers in the country. I extend my heartfelt thanks and gratitude to Dr. Joykrushna Jena, Deputy Director General (Animal and Fishery Sciences) and Dr. Raghvendra Bhatta, Deputy Director General (Animal Science) for extending whole hearted support to the Institute to carry out research and all the necessary activities to perpetuate for the ultimate benefit of the students, researchers, farmers and Industry working in the field of animal production and health. The vision, guidance, and technical support provided periodically by the esteemed Chairmen and members of QRT, RAC, and IRC committees have been instrumental in keeping NRCE on the right path and maintaining its focus and my heartfelt thanks go to all of them. My gratitude and thanks also go to Dr. Ashok Kumar, ADG (Animal Health), Dr. Amrish Kumar Tyagi, ADG (Animal Nutrition and Physuiology) and Dr. GK Gaur, ADG (Animal Production and Breeding) for unwavering support to NRCE. I am also thankful to Dr. Rajneesh Rana, Pr. Scientist (Animal Health), Dr. HK Narula, Pr. Scientist (AP&B), Dr. Barman, Pr. Scientist (AN&P), Sh. Pankaj Kumar, Dir. Admin. And other Officers of ICAR for continuous support and help being extended to the Institute for functioning smoothly and achieving success in the field of Animal production and health.

I take this opportunity to express my sincere appreciation and thanks to the Chairman and Members of the Publication Committee for untiring efforts for compiling, editing and bringing out the Annual Report-2023 in beautiful manner by showcasing all the Institute activities thoroughly for easy understanding of the Readers. Finally, my heartfelt thanks go to the entire NRCE team for their wholehearted efforts, commitment and contributions to the progress of this premier Institute for ultimate benefit of all the stakeholders in the country.

TK Bhattacharjya
(Director, ICAR-NRCE)

Executive Summary

Horses have been domesticated since prehistoric times and hold a special place in our history & culture. To cater to the needs of equine health and augment equine productivity in the country, Indian Council of Agricultural Research established National Research Centre on Equines (ICAR-NRCE) on November 26, 1985 at Hisar (Haryana). The strength that makes NRCE truly enduring and unique emanates from our commitment to improve health and productivity of equines. They are the basis of our growth and inspire us along every path. Our concerted efforts were directed to understand infectious diseases confronting equines to improve the sustainability of equine farming. ICAR-NRCE has contributed significantly in the area of diagnosis and control of equine infectious diseases. The Centre has developed diagnostics against various equine diseases like equine herpesvirus, equine rotavirus, equine influenza virus, Japanese encephalitis, equine infectious anemia, glanders, *Theileria equi* and *Trypanosoma evansi*, etc. In addition, vaccines for EHV1, equine influenza, and *Salmonella* Abortus equi have been developed by this centre. The Centre has also established National Centre for Veterinary Type Cultures (NCVTC) for acquisition, authentication, preservation, documentation and conservation of the microbial diversity of animal origin. The Centre has contributed significantly in conservation and characterization of Indian breeds of equines and even established nucleus herds of Marwari, Kathiawari, Zanskari and Manipuri breeds. The Centre made significant research contributions through 50 research projects, including 25 externally funded projects by DBT, DST, DRDE, DADF, ICAR extramural, inter-institutional collaborative research projects and WAHO during the year. This year, the institute is in limelight for achieving the new milestones like production of live foals through Embryo transfer in

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प्रागैतिहासिक काल से ही घोड़ों को पालतू बनाया जाता रहा है और वे हमारे इतिहास और संस्कृति में एक विशेष स्थान रखते हैं। देश में अश्व स्वास्थ्य की आवश्यकताओं को पूरा करने और अश्व उत्पादकता बढ़ाने के लिए भारतीय कृषि अनुसंधान परिषद ने 26 नवंबर, 1985 को हिसार (हरियाणा) में राष्ट्रीय अश्व अनुसंधान केंद्र (रा. अ. अनु. के.) की स्थापना की थी। रा. अ. अनु. के. को वास्तव में स्थायी और अद्वितीय बनाने वाली ताकत अश्वों के स्वास्थ्य और उत्पादकता में सुधार के लिए हमारी प्रतिबद्धता से उत्पन्न होती है। वे हमारे विकास का आधार हैं और हमें हर पथ पर प्रेरित करते हैं। हमारे ठोस प्रयासों का उद्देश्य अश्व पालन की स्थिरता में सुधार करने के लिए अश्वों में होने वाली संक्रामक बीमारियों को समझना था। भा. कृ. अनु. प. – रा. अ. अनु. के. ने अश्व संक्रामक रोगों के निदान और नियंत्रण के क्षेत्र में महत्वपूर्ण योगदान दिया है। केंद्र ने विभिन्न अश्व रोगों जैसे – अश्व हर्पीस विषाणु (ईएचवी), अश्व रोटाविषाणु, अश्व इन्फ्लूएंजा (ईआई) विषाणु, जापानी एन्सेफलाइटिस (जेईवी), अश्व संक्रामक एनीमिया (ईआईए), ग्लैंडर्स, थीलेरिया इक्वी और ट्रिपैनोसोमा इवांसी आदि के खिलाफ निदान विकसित किया है। इसके अलावा ईएचवी1, अश्व इन्फ्लूएंजा और साल्मोनेला एबॉर्टस के लिए टीके भी विकसित किए हैं। केंद्र ने पशु मूल की सूक्ष्मजीव विविधता के अधिग्रहण, प्रमाणीकरण, संरक्षण और दस्तावेजीकरण के लिए राष्ट्रीय पशु चिकित्सा प्ररूप संवर्धन केंद्र (एनसीवीटीसी) की भी स्थापना की है। केंद्र ने अश्वों की भारतीय नस्लों के संरक्षण और लक्षण वर्णन में महत्वपूर्ण योगदान दिया है और मारवाड़ी, काठियावाड़ी, जांस्करी और मणिपुरी नस्लों के नाभिक झुंड भी स्थापित किए हैं। केंद्र ने 51 अनुसंधान परियोजनाओं के माध्यम से महत्वपूर्ण अनुसंधान योगदान दिया, जिसमें डीबीटी, डीएसटी, डीआरडीओ, डीएडीएफ, आईसीएआर एक्स्ट्रामुरल, अंतर-संस्थान सहयोगात्मक अनुसंधान परियोजनाओं द्वारा 25 बाह्य वित्त पोषित परियोजनाएं शामिल हैं। बीते वर्ष के दौरान संस्थान भारत में पहली बार मारवाड़ी नस्ल में भ्रूण स्थानांतरण के माध्यम से जीवित बछड़ों का उत्पादन, भीमथाड़ी को भारत की 8वीं नस्ल के रूप में मान्यता

Marwari Breed for the first time in India, recognition of Bhimthadi as 8th breed of India and bagging the Breed conservation award for the Marwari Breed and 14 technologies of the institute being certified by the Council. The salient achievements of the Centre during 2023 are outlined below.

Surveillance and monitoring of equine infectious disease are one of the continuous service projects of the institute to monitor existing diseases as well as keeps vigilance on exotic diseases. In this direction, during current report of the year 2023, a total of 2280 equine serum samples from 9 states were tested for various diseases like Equine Infectious Anaemia (EIA), Equine Influenza (EI), *Trypanosoma evansi* (Trypanosomiasis), Equine Herpes Virus-1 (EHV-1), Piroplasmosis, Japanese Encephalitis (JEV), *Salmonella Abortus equi* and Brucellosis. The highest sero-prevalence was observed for equine piroplasmosis (22.67%) followed by EHV-1 (14.1 %), *Trypanosoma evansi* (1.53%), EI (1.31%) and JE (0.3%). Also a total of 457 samples were tested for various above mentioned equine diseases under DI and 12/53 cases were found positive for EHV1/4 and 4/64 for *T. evansi*.

Under contractual diagnostic services, a total of 7907 samples received from race courses, turf clubs, stud farm, riding schools, animal quarantine & certification services (AQCS) and other organized sectors during the year 2023 were tested for various notifiable and exotic diseases to check ingression of diseases from abroad and monitoring of elite horses in private sectors leading to generation of revenue amounting to Rs 70.15 lakhs. For surveillance of glanders, in year 2023, a total of 43,600 equine sera from 17 states/UT were collected and examined where in a total of 117 glanders positive cases were reported in 10 states; however, around 50% of the samples and glanders positive cases were originated from Uttar Pradesh. Out of 193 sera from occupationally exposed humans (veterinary Officers, equine handlers, and laboratory workers) none were found positive. Also, in another study, Hcp1 protein showed quick and strong reactivity against glanders positive serums and IL-1b, IL-6, IL-17, MCP-1 cytokines were found to be significantly higher in glanders positive serum.

दिलाने और मारवाड़ी नस्ल के लिए नस्ल संरक्षण पुरस्कार प्राप्त करने जैसे नए मील के पत्थर हासिल करने के लिए सुर्खियों में रहा है। संस्थान की 14 प्रौद्योगिकियों को परिषद द्वारा प्रमाणित किया या है। वर्ष 2023 के दौरान केंद्र की अन्य मुख्य उपलब्धियाँ नीचे दी गई हैं।

मौजूदा बीमारियों की निगरानी के साथ-साथ विदेशी बीमारियों पर निगरानी रखने के लिए अश्व संक्रामक रोगों की सर्वेलेंस और निगरानी संस्थान की निरंतर सेवा परियोजनाओं में से एक है। इस दिशा में, वर्ष 2023 की वर्तमान रिपोर्ट के दौरान, 9 राज्यों के कुल 2280 इक्वाइन सीरम नमूनों का परीक्षण विभिन्न बीमारियों जैसे कि ईआईए, ईआई, ट्रिपेनोसोमियासिस और ईएचवी-1, पायरोप्लाज्मोसिस, जेईवी, साल्मोनेला एबॉर्टस इक्वी और ब्रूसेलोसिस के लिए किया गया था। सबसे अधिक सीरो प्रिवेलेंस इक्वाइन पायरोप्लाज्मोसिस (22.67%) के लिए देखा गया, इसके बाद ईएचवी-1 (14.1%), ट्रिपैनोसोमा इवांसी (1.53%), ईआई (1.31%) और जेई (0.3%) का स्थान रहा। इसके अलावा डीआई के तहत उपरोक्त विभिन्न अश्व रोगों के लिए कुल 457 नमूनों का परीक्षण किया गया और 12/53 मामले ईएचवी 1/4 के लिए सकारात्मक पाए गए। पायरोप्लाज्मोसिस के मामले में 1/8 या 8/1य और टी. इवांसी के लिए 4/64 सकारात्मक पाए गए।

संविदात्मक निदान सेवाओं के तहत, रेस कोर्स, टर्फ क्लब, स्टड फार्म से कुल 7907 नमूने प्राप्त हुए। वर्ष 2023 के दौरान घुड़सवारी स्कूलों, पशु क्वारंटाईन और प्रमाणन सेवाओं (एक्यूसीएस) और अन्य संगठित क्षेत्रों में विदेशों से बीमारियों के प्रवेश को रोकने और निजी क्षेत्रों में विशिष्ट घोड़ों की निगरानी के लिए विभिन्न उल्लेखनीय और विदेशी बीमारियों का परीक्षण किया गया, जिससे 70.15 लाख रुपये का राजस्व उत्पन्न हुआ। ग्लैंडर्स की निगरानी के लिए, वर्ष 2023 में, 17 राज्यों/केंद्र शासित प्रदेशों से कुल 43,600 इक्वाइन सीरा एकत्र किये गये और जांच की गई, जहाँ 10 राज्यों में कुल 117 ग्लैंडर्स सकारात्मक मामले सामने आए। हालाँकि, लगभग 50% नमूने और ग्लैंडर्स पॉजिटिव मामले उत्तर प्रदेश से उत्पन्न हुए थे। व्यावसायिक रूप से संपर्क में आने वाले मनुष्यों (पशु चिकित्सा अधिकारी, अश्व संचालक और प्रयोगशाला कर्मचारी) के 193 सीरा में से कोई भी सकारात्मक नहीं पाया गया। इसके अलावा, एक अन्य अध्ययन में, एचसीपी1 प्रोटीन ने ग्लैंडर्स पॉजिटिव सीरम के खिलाफ त्वरित और मजबूत प्रतिक्रिया दिखाई और ग्लैंडर्स पॉजिटिव सीरम में IL-1β, IL-6, IL-17, MCP-1 साइटोकिन्स काफी अधिक पाए गए।

The recombinant bivalent constructs of EHV1 and EIV (Florida clade 1&2) by replacing virulence associated genes of EHV1 were generated to develop combined vaccine for EHV1 and equine influenza. Herpervirus, which is a major concern for equines, studies were conducted on standardization of a real-time PCR targeting glycoprotein B gene. The incidence rate for EHV-2, 5 and mixed infection were found to be 28.57%, 35.71% and 14.28% respectively. Under the "National One Health Program for Prevention and Control of Zoonotic Diseases (NOHPCZ)", NRCE is actively involved in capacity building, strengthening of laboratory diagnostic facilities, inter-sectoral coordination and creating awareness about zoonotic diseases of public health importance by organizing training, workshop and webinar on various zoonotic diseases.

A one-step, probe-based real-time RT-PCR assay for rabies diagnosis was standardized and 50 (nos.) samples of saliva/ brain tissues were tested for rabies from suspected animals and 13 samples were positive through RT-PCR and direct fluorescent antibody testing. In another study the protocol for preparation of *R. equi* whole cell lysate was optimized and characterization of its immunogenic proteins was carried out. The whole cell lysate along with recombinant vapA proteins will be used for immunization study in mouse model and in foals. A fixed cell indirect ELISA using whole cell-*Streptococcus equi* and an RPA assay for sensitive and specific detection of the nucleic acids of *S. equi* are being standardized for diagnosis of strangles. In a study conducted on serosurveillance for strangles in equines, a total of 90 field serum samples were screened for serum antibodies against *S. equi* from Haryana, Jammu and Kashmir, Rajasthan, Uttar Pradesh and Uttarakhand and found 28 samples positive with an overall prevalence rate of 31.1%. Additionally, a multiplex Taqman qPCR assay intended to be used for in vitro detection of *Streptococcus* sps and *Streptococcus equi* subsps. *equi* in respiratory samples has been internally validated. In another study, we identified members of the Stilbenes group (specifically resveratrol and pterostilbene) in extracts of *Aerva javanica* plant showing anti-proud flesh activity. In field cases of

ईएचवी 1 और ईआवी (फ्लोरिडा क्लेड 1 और 2) के पुन: संयोजक द्विसंयोजक निर्माणों को ईएचवी 1 के विषाणु संबंधी जीन प्रतिस्थापित करके ईएचवी 1 और इक्वाइन इन्फ्लूएंजा के लिए संयुक्त टीका विकसित करने के लिए तैयार किया गया। हर्पिस विषाणु, जो घोड़ों के लिए एक प्रमुख चिंता का विषय है, जीपीबी जीन को लक्षित करने वाले आरटी पीसीआर के मानकीकरण पर अध्ययन आयोजित किए गए थे। ईएचवी -2, 5 और मिश्रित संक्रमण की घटना दर क्रमश: 28.57%, 35.71% और 14.28% पाई गई। ''ज़ूनोटिक रोगों की रोकथाम और नियंत्रण के लिए राष्ट्रीय वन हैल्थ कार्यक्रम (एनओएचपीसीजेड)'' के तहत, रा. अ. अनु. के. क्षमता निर्माण, प्रयोगशाला निदान सुविधाओं को मजबूत करने, अंतर-क्षेत्रीय समन्वय और प्रशिक्षण आयोजित करके सार्वजनिक स्वास्थ्य महत्व के जूनोटिक रोगों के बारे में जागरूकता पैदा करने में सक्रिय रूप, विभिन्न जूनोटिक रोगों पर कार्यशाला और वेबिनार आयोजित किए गए।

रेबीज निदान के लिए एक-चरणीय, प्रोब आधारित आरटी पीसीआर परख को मानकीकृत किया गया था और संदिग्ध जानवरों से रेबीज के लिए लार/मस्तिष्क के ऊतकों के 50 (संख्या) नमूनों का परीक्षण किया गया था और 13 नमूने आरटी पीसीआर और फ्लोरोसेंट एंटीबॉडी परीक्षण के माध्यम से सकारात्मक थे। एक अन्य अध्ययन में आर इक्वि होल सेल लाइसेट की तैयारी के लिए प्रोटोकॉल को अनुकूलित किया गया और इसके इम्युनोजेनिक प्रोटीन का लक्षण वर्णन किया गया। पुन: संयोजक वैप-ए प्रोटीन के साथ संपूर्ण कोशिका लाइसेट का उपयोग माउस मॉडल और शिशुओं में टीकाकरण अध्ययन के लिए किया जाएगा। स्ट्रैंगल्स के निदान के लिए संपूर्ण कोशिका- स्ट्रेप्टोकोकस इक्वी और एस इक्वी के न्यूक्लिक एसिड के ओपटीमाइज़ और विशिष्ट पता लगाने के लिए एक आरपीए परख का उपयोग करके एक फिक्स्ड सेल अप्रत्यक्ष एलाईसा को मानकीकृत किया जा रहा है। घोड़ों में गलघोटूँ बीमारी के लिए सीरो-निगरानी की गई। एक अध्ययन में, हरियाणा, जम्मू और कश्मीर, राजस्थान, उत्तर प्रदेश और उत्तराखंड से एस इक्वि के खिलाफ सीरम एंटीबॉडी के लिए कुल 90 फील्ड सीरम नमूनों की जाँच की गई और कुल मिलाकर 28 नमूने सकारात्मक पाए गए। जिसकी प्रिवेलेंस दर 31.1% थी। इसके अतिरिक्त, एक मल्टीप्लेक्स टैक्मैन क्यूपीसीआर परख का उपयोग स्ट्रेप्टोकोकस और स्ट्रेप्टोकोकस इक्वी के इन विट्रो पता लगाने के लिए किया जाना है। श्वसन नमूनों में समानता को आंतरिक रूप से मान्यता प्राप्त हुई है। एक अन्य अध्ययन में, हमने एर्वा जावनिका पौधे के अर्क में स्टिलबेन समूह (विशेष रूप से रेस्वेराट्रोल और टेरोस्टिलबिन) के सदस्यों की पहचान की, जो एंटी प्राउड फ्लैष गतिविधि दिखा रहे हैं। घोड़ों में ग्रीष्मकालीन

summer dermatitis in horses, it was found that the expression of anti-inflammatory cytokines IL-10 and TGF-b1 may be important targets for therapeutic purpose to control chronic infection.

In a study, barbamine and emetine were found to exhibit antitrypanosomal activity against *Trypanosoma evansi* in axenic culture. Also, careful selection of adjuvants and delivery systems is being carried out for *T. equi* piroplasms. Simultaneously, *T. equi* antigen detection ELISA using TE/tEMA-2, GST cleaved recombinant proteins is being developed. A skin-specific bioink made from digested chicken/porcine skin and incorporated with polymers has been developed and analyzed for its structure, stability, and compatibility for therapeutic applications. Nanocarriers, like ZnO nanoparticles with flower-like morphology (ZnONFs) decorated with chitosan and hydroxychloroquine (CHCZnO NPs) is being developed as a more potent approach against coronaviruses. The 3D printed skin facilitated the attachment of cells to the scaffolds in the CAM assay, and wound healing was accelerated in animal studies with re-epithelization increased hydroxyproline and collagen contents, TNF-a, and IL-6 genes as compared to the control and the results suggest that 3D bioprinting is a suitable technology for generating bioengineered skin for therapeutic applications. Preparation and charaterisation of lipid-based adjuvants for preparing the adjuvated *Theileria equi* recombinant novel surface proteins immunogen as a vaccine delivery agent.

The Centre has been awarded with Breed Conservation Award -2023, Second prize in Institutional category by ICAR-NBAGR for the *in situ* and *ex situ* conservation and propagation efforts done at the Centre. The Bhimthadi (Deccani) breed of horse used to be referred as the horse breed at the verge of extinction along with the Chumarti and Sikang breeds. The Centre took the initiative to find out the present status of the breed, which was developed in the 17th-18th century during Maratha rule in the area. In the 11th meeting of Breed Registration Committee, chaired by honourable DDG (AS) on December 5, 2023, the Bhimthadi breed has been recognized as the 8th breed of indigenous horse in the

डरमेटाइटिस के क्षेत्रीय मामलों में, यह पाया गया कि एंटी-इंफ्लेमेटरी साइटोकिन्स IL-10 और TGF&-1 की अभिव्यक्ति क्रोनिक संक्रमण को नियंत्रित करने के चिकित्सीय उद्देश्य के लिए महत्वपूर्ण लक्ष्य हो सकती है।

एक अध्ययन में, बार्बामाइन और एमेटिन को एक्सेनिक कल्चर्स में ट्रिपैनोसोमा इवांसी के खिलाफ एंटी-ट्रिपानोसोमल गतिविधि प्रदर्शित करते पाया गया। इसके अलावा, टी. इक्वि पिरोप्लाज्म्स के लिए सहायक और वितरण प्रणालियों का सावधानीपूर्वक चयन किया जा रहा है। इसके साथ ही, टीई/टीईएमए-2, जीएसटी क्लीव्ड रीकॉम्बिनेंट प्रोटीन का उपयोग करके टी. इक्वि एंटीजन डिटेक्शन एलिसा विकसित किया जा रहा है। पचे हुए चिकन/ सूअर की त्वचा से बना और पॉलिमर के साथ शामिल एक त्वचा-विशिष्ट बायोइंक विकसित किया गया है और चिकित्सीय अनुप्रयोगों के लिए इसकी संरचना, स्थिरता और अनुकूलता के लिए इसका विश्लेषण किया गया है। फूल जैसी आकृति विज्ञान (ZnONFs) के साथ ZnO नैनोकणों जैसे नैनोकैरियर्स को काईटोसन और हाइड्रोक्सीक्लोरोक्वीन (CHCZnO NPs) से बनाया गया है, जिन्हें कोरोना वायरस के खिलाफ अधिक शक्तिशाली दृष्टिकोण के रूप में विकसित किया जा रहा है। 3डी मुद्रित त्वचा ने सीएएम परख में कोशिकाओं को स्कैफोल्ड से जोड़ने की सुविधा प्रदान की, और रीएपीथिलाईजेषन व हाइड्रॉक्सीप्रोलीन और कोलेजन अधिक पाया गया। टीएनएफ-α और आईएल-6 जीन की तुलना की गई व जानवरों में घाव भरने में तेजी आई। सुझाव है कि चिकित्सीय अनुप्रयोगों के लिए बायोइंजीनियर्ड त्वचा तैयार करने के लिए 3डी बायोप्रिंटिंग एक उपयुक्त तकनीक है।

वैक्सीन वितरण एजेंट के रूप में एडज्युवेटेड थीलेरिया इक्वी रीकॉम्बिनेंट नॉवेल सतह प्रोटीन इम्युनोजेन तैयार करने के लिए लिपिड-आधारित एडजुवेंट की तैयारी और चरित्र-चित्रण किया गया। केंद्र को इन सीटू और एक्स सीटू संरक्षण और प्रसार प्रयासों के लिए भा. कृ. अनु. प. - रा. प. आनु. सं. ब्यूरो द्वारा नस्ल संस्थान पुरस्कार -2023, संस्थागत श्रेणी में दूसरे पुरस्कार से सम्मानित किया गया है। भीमथाड़ी (दक्खनी) घोड़े की नस्ल को चुमरती और सिकंग नस्ल के साथ विलुप्त होने के कगार पर घोड़े की नस्ल के रूप में जाना जाता था। केंद्र ने क्षेत्र में मराठा शासन के दौरान 17वीं-18वीं शताब्दी में विकसित की गई नस्ल की वर्तमान स्थिति का पता लगाने की पहल की। 11वीं बैठक में 5 दिसंबर, 2023 को माननीय उप महानिदेशक (पशु-विज्ञान) की अध्यक्षता में नस्ल पंजीकरण समिति की बैठक में भीमथाड़ी नस्ल को देश में स्वदेशी घोड़ों की 8वीं

country and it has been assigned the Accession No. INDIA_HORSE_1100_BHIMTHADI_07008.

As a significant achievement, for the first time in the country, live Marwari breed foals have been born through embryo transfer technology at the subcentre at Bikaner. Also, Zanskari mare was conceived with the transferred embryo and is due for foaling in the year 2024. The primary structure determination and physicochemical characterization of DSP-3, a phosphatidylcholine binding glycoprotein of donkey seminal plasma was carried out. In another study, it was found that addition of melatonin significantly increased T-AOC levels and reduced MDA levels indicating increase in antioxidant levels, reduction in oxidative stress and lipid peroxidation levels. The cryovial method for cryopreservation of stallion spermatozoa, vitrification of horse embryos and cryopreservation of semen from indigenous horses was also carried out. Additionally, detection of SNPs of MSTN gene among indigenous donkey breeds, creation of equine CNV databases were carried out. In a study, biochemical, dielectric and surface characteristics of freeze-dried donkey milk powder were studied. The institute carried out social media usage pattern studies among horse keepers and field veterinarians and it was found that ~ 71.60% of the respondents used social media for finding out news and events whereas 68.70% of the respondents used it for exchange of information. A Mobile App named as "EquiCare: All-in-one mobile companion" is also developed by the ICAR-National Research Centre on Equines, Hisar and released on 26th November, 2023 on the foundation day of NRCE. It is a user friendly bilingual mobile App in Hindi and English language for equine owners, Veterinary officers, Animal Health department officials, students, industry professionals and other stakeholders.

NCVTC virus repository is being strengthened with the addition of viruses from different geographical locations of the country through the deposition/ collection of isolates and clinical samples from different animals and poultry. In this regard, thirty-six

नस्ल के रूप में मान्यता दी गई है और इसे परिगृहण संख्या India_HORSE_1100_BHIMTHADI_07008 प्रदान की गई है।

एक महत्वपूर्ण उपलब्धि के रूप में, देश में पहली बार बीकानेर के उपकेंद्र में भ्रूण स्थानांतरण तकनीक के माध्यम से जीवित मारवाड़ी नस्ल के बच्चों का जन्म हुआ है। इसके अलावा, जांस्करी घोड़ी का गर्भाधान स्थानांतरित भ्रूण के साथ किया गया था और वर्ष 2024 में बच्चा पैदा होने वाला है। गधे के सेमिनल प्लाज्मा के फॉस्फेटिडिलकोलाइन बाइंडिंग ग्लाइकोप्रोटीन डीएसपी-3 की प्राथमिक संरचना निर्धारण और भौतिक रासायनिक लक्षण वर्णन किया गया था। एक अन्य अध्ययन में, यह पाया गया कि मेलाटोनिन के अतिरिक्त टी-एओसी स्तर में काफी वृद्धि हुई और एमडीए स्तर में कमी आई, जो एंटीऑक्सीडेंट स्तर में वृद्धि, ऑक्सीडेटिव तनाव और लिपिड पेरोक्सीडेशन स्तर में कमी का संकेत देता है। स्टैलियन शुक्राणु के क्रायोप्रिजर्वेशन, घोड़े के भ्रूण के विट्रीफिकेशन और स्वदेशी घोड़ों के वीर्य के क्रायोप्रिजर्वेशन के लिए क्रायोवियल विधि भी अपनाई गई। इसके अतिरिक्त, स्वदेशी गधों की नस्लों के बीच एमएसटीएन जीन के एसएनपी का पता भी किया या। अश्व सीएनवी डेटाबेस का निर्माण किया गया। एक अध्ययन में, फ्रीज-ड्राई किया गधी के दूध का पाउडर की जैव रासायनिक, डाई-इलैक्ट्रिक और सतह विशेषताओं का अध्ययन किया गया। संस्थान ने घोड़ा पालकों और क्षेत्र के पशु चिकित्सकों के बीच सोशल मीडिया उपयोग पैटर्न का अध्ययन किया और यह पाया गया कि 71.60% उत्तरदाताओं ने समाचार और घटनाओं का पता लगाने के लिए सोशल मीडिया का उपयोग किया, जबकि 68.70% उत्तरदाताओं ने इसका उपयोग सूचनाओं के आदान-प्रदान के लिए किया। भा. कृ. अनु. प. - रा. अ. अनु. के. हिसार द्वारा ''इक्विकेयर: ऑल-इन-वन मोबाइल कंपेनियन'' नाम से एक मोबाइल ऐप भी विकसित किया गया है जो 26 नवम्बर, 2023 को स्थापना दिवस पर जारी किया गया है। यह घोड़े के मालिकों, पशु चिकित्सा अधिकारियों, पशु स्वास्थ्य विभाग के अधिकारियों, छात्रों, उद्योग के पेशेवरों और अन्य हितधारकों के लिए हिंदी और अंग्रेजी भाषा में एक उपयोगकर्ता-अनुकूल द्विभाषी मोबाइल ऐप है। यह ऐप उपयोगकर्ताओं को नस्लों, कृत्रिम गर्भाधान, गर्भावस्था निदान, प्रबंधन, पोषण, बीमारियों, नैदानिक सेवाओं, टीकाकरण के संबंध में घोड़ों के विभिन्न पहलुओं में अपने ज्ञान को बढ़ाने की अनुमति देता है।

एनसीवीटीसी विषाणु भंडार को विभिन्न जानवरों और पोल्ट्री के आइसोलेट्स और नैदानिक नमूनों के जमाव/ संग्रह के माध्यम से देश के विभिन्न भौगोलिक स्थानों से विषाणुओं को शामिल करके मजबूत किया जा रहा है। इस संबंध में, विषाणु वायरल आइसोलेट्स एनसीवीटीसी नेटवर्क इकाइयों से प्राप्त हुए थे। जमा

viral isolates were received as deposits from NCVTC Network units. The deposits included Avian infectious bronchitis (n=2), Fowlpox virus (n=1), Marek's disease virus (n=2), Inclusion body hepatitis virus (aka Fowl adenovirus) (n=1), African swine fever virus (n=1), Bovine Corona virus (n=3), Rous Sarcoma Virus (RAV-1), Duck enteritis virus (n=1), PPR virus (Mutant strain) (n=1), Bluetongue virus (n=15), Sheeppox virus (n=3), Orf virus (n=2) and Lumpy skin disease virus (n=2). The samples/ isolates were processed for authentication & accession of the different viruses. Four viruses were accessioned (VTCC AVA 381- 384), in the NCVTC repository which include Rous Sarcoma Virus (RAV-1), Duck enteritis virus (n=1), PPR virus (Mutant strain) (n=1) and African swine fever virus (n=1). Besides, the bulk production and preservation of 30 previously accessioned viruses (10 vials each) including NDV (n=11), fowl adenovirus (n=3), Infectious bronchitis virus (n=4), Bluetongue virus (n=4), Chicken astrovirus (n=2), Fowlpox virus (n=2), Swinepox virus (n=1) and Infectious bursal disease virus (n=3) was also completed. Furthermore, 21 previously preserved viruses including NDV (8), SWPV (1), LSDV (1), BTV (6), IBDV (3) and IBV (2) were revived and checked for their viability, which is an important activity of the virus repository. During the year 2023, a total of 160 microbes were accessioned in the repository thereby the cumulative strength of NCVTC reaching to 2613.

The evaluation of safety, immunogenicity and efficacy of Lumpi-ProVac^{Ind} was carried out. Also, miRNA profiling of lumpy skin disease virus infected primary lamb testicle cells was performed to find biomarkers as well as novel targets for therapeutic intervention against LSDV and miR-29a was found to be a novel biomarker. A novel HRM-based gap-qRT-PCR for identification and quantitation of the vaccine and field strain(s) of lumpy skin disease virus was developed. In another remarkable study, evaluation of antibody- and cell-mediated immune responses following vaccination with a newly developed live-attenuated LSD vaccine (Lumpi-ProVac^{Ind}) was carried out. The detectable amount of anti-LSDV antibodies was observed at 1-2 months following

किये गए, एवियन संक्रामक ब्रोंकाइटिस (एन=2), फाउलपॉक्स विषाणु (एन=1), मारेक रोग विषाणु (एन=2), इंक्लूजन बॉडी हेपेटाइटिस विषाणु (उर्फ फाउल एडेनोवायरस) (एन=1), अफ्रीकी स्वाइन फीवर विषाणु (एन=1), बोवाइन कोरोना विषाणु (एन=3), रौस सारकोमा विषाणु (आरएवी-1), डक एंटराइटिस विषाणु (एन=1), पीपीआर विषाणु (म्यूटेंट स्ट्रेन) (एन=1), ब्लूटंग विषाणु (एन=15), शीपॉक्स विषाणु (एन =3), ओर्फ विषाणु (एन =2) और लम्पी त्वचा रोग विषाणु (एन=2) थे। विभिन्न विषाणुओं के प्रमाणीकरण और परिग्रहण के लिए नमूनों/ आइसोलेट्स को संसाधित किया गया था। एनसीवीटीसी भण्डारण में चार विषाणु परिग्रहित किए गए (वीटीसीसी एवीए 381-384), जिसमें रौस सारकोमा विषाणु (आरएवी-1), डक एंटरटाइटिस विषाणु (एन=1), पीपीआर विषाणु (म्यूटेंट स्ट्रेन) (एन=1) और अफ्रीकी सूअर विषाणु शामिल हैं। शेष विषाणु (हाल ही में प्राप्त प्राप्त किये गये) की पुष्टि के लिए प्रक्रियाधीन हैं। इसके अलावा, एनडीवी (एन=11), फाउल एडेनो विषाणु (एन=3), संक्रामक ब्रोंकाइटिस विषाणु (एन=4), ब्लूटंग विषाणु (एन=4), चिकन एस्ट्रो विषाणु (एन=2) और फाउलपॉक्स विषाणु (एन=2), स्वाइनपॉक्स विषाणु (एन=1) और संक्रामक बर्सल रोग विषाणु (एन=3) सहित 30 पहले से जुड़े विषाणु (प्रत्येक में 10 शीशियां) का थोक उत्पादन और संरक्षण भी पूरा हो गया। इसके अलावा, एनडीवी (8), एसडब्ल्यूपीवी (1), एलएसडीवी (1), बीटीवी (6), आईबीडीवी (3) और आईबीवी (2) सहित 21 पहले से संरक्षित विषाणुओं को पुनर्जीवित किया गया और उनकी व्यवहार्यता की जांच की गई जो कि एक महत्वपूर्ण गतिविधि है। वर्ष 2023 के दौरान कुल 33% रोगाणुओं को भंडार में शामिल किया गया जिससे एनसीवीटीसी की संचयी ताकत 3333 तक पहुँच गई।

लंपी प्रो वैक इंडिया की सुरक्षा, प्रतिरक्षाजन्यता और प्रभावकारिता का मूल्यांकन किया गया। इसके अलावा, एलएसडीवी के खिलाफ चिकित्सीय हस्तक्षेप के लिए बायोमार्कर के साथ-साथ नए लक्ष्य खोजने के लिए गांठदार त्वचा रोग वायरस से संक्रमित प्राथमिक मेमने अंडकोष कोशिकाओं की एमआईआरएनए प्रोफाइलिंग की गई और इस अध्ययन में, एमआईआर-29 को एक नया बायोमार्कर पाया गया। ढेलेदार त्वचा रोग वायरस के टीके और फील्ड स्ट्रेन की पहचान और मात्रा निर्धारित करने के लिए एक नया एचआरएम-आधारित गैप-क्यूआरटी-पीसीआर विकसित किया गया था। एक अन्य उल्लेखनीय अध्ययन में, नव विकसित लाइव-एटेन्यूएटेड एलएसडी वैक्सीन (लंपी परो वैक इंड़िया) के साथ टीकाकरण के बाद एंटीबॉडी और कोशिका-मध्यस्थ प्रतिरक्षा प्रतिक्रियाओं का मूल्यांकन किया गया। टीकाकरण के बाद 1-2 महीने में एंटी-एलएसडीवी एंटीबॉडी का पता लगाने

vaccination, with a peak antibody titer at 3 months. Upon stimulation of the 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-g levels upon antigenic stimulation of their PBMCs with LSDV antigen. This showed that the buffaloes also mount a potent antibody- and cell-mediated immune response following vaccination with Lumpi-ProVac^{Ind}. In another study, Hesperetin was found to block poxvirus replication by competitively inhibiting binding of the 5' cap of viral mRNA with eIF4E. The surveillance of SARS-CoV-2 in waste water in Hisar region was also carried out. Additionally, a novel sub-genotype AL1b of rabies virus in domestic animals across Haryana, India was identified and this highlights the rabies diversity. This study will help to fill crucial knowledge gap and to bolster rabies control programme in the state.

In an attempt to identify and isolate bovine coronavirus from respiratory and enteric infections in cattle, the BCoVs were confirmed by RT-PCR. An isothermal RT-RPA-CRISPR-LFA assay was developed for detection of SARS-CoV-2 nucleic acid targeting highly conserved genes of the virus. The mRNA vaccine candidate for SARS-CoV-2 has been evaluated in BALB/c mice and RPA-LFA assay for detection of EHV1 & 4 nucleic acids has been developed. Studies on bovine rota virus, which is a leading cause of calf mortality are also underway for diagnosis, isolation and characterization of viral strains. In the bacteriophage repository, physiological and genomic characterization of bacteriophage encoded lysins was carried out to explore a promising antimicrobial agent against multidrug-resistant strains of *Salmonella* and *E. coli*. During year 2023, a total of 209 bacterial cultures were processed, out of which 115 belonging to 28 taxa were accessioned into the bacterial repository, increasing the total collection of veterinary bacteria to 1857. Bacillus cultures were accessioned under anthrax diagnosis programme. Antibacterial activity

योग्य मात्रा देखी गई, 3 महीने में चरम एंटीबॉडी टाईटर आया। यूवी-निष्क्रिय एलएसडीवी एंटीजन के साथ परिधीय रक्त मोनोन्यूक्लियर कोशिकाओं (पीबीएमसी) की उत्तेजना पर टीकाकरण न किए गए पशुओं की तुलना में टीका लगाए गए पशुओं में सीडी8+टी कोशिकाओं की संख्या में उल्लेखनीय वृद्धि पाई गई। इसके अलावा, टीका लगाए गए जानवरों ने भी एलएसडीवी एंटीजन के साथ अपने पीबीएमसी की एंटीजेनिक उत्तेजना पर आईएफएन-ङस्तर में उल्लेखनीय वृद्धि देखी। इससे पता चला कि लंपी प्रो वैक इंड़िया के टीकाकरण के बाद भैंसों में एक शक्तिशाली एंटीबॉडी और कोशिका-मध्यस्थ प्रतिरक्षा प्रतिक्रिया भी विकसित होती है। एक अन्य अध्ययन में, हेस्पेरेटिन को ईआईएफ4ई के साथ वायरल एमआरएनए के 5' कैप के बंधन को प्रतिस्पर्धात्मक रूप से रोककर पॉक्सवायरस प्रतिकृति को अवरुद्ध करने के लिए पाया गया था। हिसार क्षेत्र में अपशिष्ट जल में SARS-CoV 2 की निगरानी भी की गई। इसके अतिरिक्त, पूरे हरियाणा, भारत में घरेलू पशुओं में रेबीज विषाणु के एक नए उप-जीनोटाइप AL1b की पहचान की गई और यह रेबीज की विविधता पर प्रकाश डालता है। यह अध्ययन महत्वपूर्ण ज्ञान अंतर को भरने और राज्य में रेबीज नियंत्रण कार्यक्रम को मजबूत करने में मददगार होगा।

मवेशियों में श्वसन और आंत्र संक्रमण से गोजातीय कोरोना वायरस की पहचान करने और उसे अलग करने के प्रयास में, आरटी-पीसीआर द्वारा बीसीओवी की पुष्टि की गई। सूअरों में पोर्सिन सर्कोविषाणु 3 (पीसीवी3) का पॉइंट-ऑफ-केयर पता लगाने के लिए सीआरआईएसपीआर (CRISPR) टेक्नोलॉजी और एलएफए (LFA) आधारित परख के साथ आइसोथर्मल ''रीकॉम्बिनेज पॉलीमरेज एम्प्लीफिकेशन'' (RPA) विकसित किया गया था, जो पीसीवी3 न्यूक्लिक एसिड अल्म का तेजी से पता लगाने के लिए एक बहुमुखी पीओसी प्लेटफॉर्म के रूप में काम कर सकता है। सूअरों में इसके अलावा, वायरस के अत्यधिक संरक्षित जीन को लक्षित करके SARS-CoV 2 न्यूक्लिक एसिड अल्म का पता लगाने के लिए एक आईसोथर्मल RT-RPA-CRISPR-LFA परख विकसित की गई थी। SARS-CoV 2 के लिए mRNA वैक्सीन उम्मीदवार का मूल्यांकन BALB/c चूहों में किया गया है और ईवीएच 1 और 4 न्यूक्लिक अल्म का पता लगाने के लिए RPA-LFA परख विकसित की गई है। विषाणु स्ट्रेन के निदान, अलगाव और लक्षण वर्णन के लिए गोजातीय रोटाविषाणु पर अध्ययन भी चल रहा है जो बछड़े की मृत्यु का एक प्रमुख कारण है। जीवाणुभोजी भंडार में, फिजियोलोजिक और जीनोमिक वर्णन द्वारा साल्मोनेला और ई. कोली के मल्टीड्रग-प्रतिरोधी उपभेदों के खिलाफ एक आशाजनक रोगाणुरोधी एजेंट का पता लगाने के लिए बैक्टीरियोफेज एन्कोडेड लाइसिन का लक्षण वर्णन किया गया था। वर्ष 2023 के दौरान, कुल 209 जीवाणु संस्कृतियों को संसाधित किया गया, जिनमें से 28 टैक्सा से संबंधित 115 को जीवाणु भंडार में शामिल किया गया, जिससे पशु चिकित्सा

of domestic donkeys' milk against *Escherichia coli* and *Staphylococcus aureus* was carried out. Under INFAAR surveillance project, a total of, 217 *E. coli* isolates were characterized and 69 *Staphylococcus* spp., were genotyped. Importantly, *Corynebacterium pseudotuberculosis* was isolated from a field case of caseous lymphadenitis in sheep and goats. Additionally, from 109 collected samples, 7 nos. acid-fast bacilli were isolated in order to explore mycobacterial infections. CRISPR- Cas9 mediated gene editing in *Mycobacterium kansasii* is being pursued to explore using CRISPR-Cas9 approach to decipher the role of MCC genes in survival of *M. kansasii.*

During 2023, the scientists of the Centre published 68 high impact original research articles in international and national refereed journals. A total of 7 popular articles, 1 technical bulletin, 6 compendium compilations, 2 book chapters, 68 training manual chapters, 30 research abstracts, 42 GenBank accessions and 14 technologies were certified by ICAR. The scientists of the Centre presented papers in 27 different national and international conferences, seminars or symposia, 47 expert/invited lectures were delivered by the scientists. During the current year, the center has inked MoU with research/cadmic institutions, commercialized 3 technologies, 4 technologies were developed and assessed and 5 new technologies were developed and filed 3 patents.

The Centre organized various activities under directives from Government of India. Yoga camp (13-21 June) to celebrate International Day of Yoga, Hindi Fortnight (14-26 September) to promote hindi, Sanitation Drive (2-16 October), Vigilance Awareness Week (31 October-5 November), Agriculture Education Day (3 December), National Productivity Week (12-18 February) were celebrated with great fanfare. Foundation Day of the Centre was celebrated on 26 November 2023 by organizing Scientists-Veterinarian Interface meeting. Other institutional activities organized were World Veterinary Day (30 April 2023) and National Science Day (28 February, 2023).

The center also offers paid consultancy and diagnostic services for important infectious diseases

बैक्टीरिया का कुल संग्रह 1857 तक बढ़ गया। बैसिलस संस्कृतियों को एंथ्रेक्स निदान कार्यक्रम के तहत शामिल किया गया। एस्चेरिचिया कोलाई और स्टैफिलोकोक्स ऑरियस के खिलाफ घरेलू गधों के दूध की जीवाणुरोधी गतिविधि पता की गई। इन्फार निगरानी परियोजना के तहत, कुल 217 ई. कोली आइसोलेट्स की पहचान की गई और 69 स्टैफिलोकोक्स को जीनोटाइप किया गया। महत्वपूर्ण रूप से, कोरायनीबैक्टीरियम स्यूडोट्यूबरकुलोसिस को भेड़ और बकरियों में केसियस लिम्फैडेनाइटिस के एक क्षेत्रीय मामले से अलग किया गया था। इसके अतिरिक्त, 109 एकत्रित नमूनों में से 7 माइकोबैक्टीरियम द्वारा संक्रमण का पता लगाने के लिए एसिड-फास्ट जीवाणुओं को अलग किया गया था। एम. कंसासी में एमसीसी जीन की भूमिका को समझने के लिए CRISPR-CAS 9 दृष्टिकोण का उपयोग करके माइकोबैक्टीरियम कंसासी में CRISPR-CAS 9 मध्यस्थ जीन संपादन का पता लगाया जा रहा है।

वर्ष 2023 के दौरान, केंद्र के वैज्ञानिकों ने अंतरराष्ट्रीय और राष्ट्रीय रेफरीड पत्रिकाओं में 68 उच्च प्रभाव वाले मूल शोध लेख प्रकाशित किए। 7 लोकप्रिय लेख, 1 तकनीकी बुलेटिन, 3 सार-संग्रह संकलन, 2 पुस्तक अध्याय, 68 प्रशिक्षण मैनुअल अध्याय, 30 शोध सार, 42 जेनबैंक परिग्रहण प्रकाशित किए गए, और 14 प्रौद्योगिकियों को आईसीएआर द्वारा प्रमाणित भी किया गया था। केंद्र के वैज्ञानिकों ने 27 विभिन्न राष्ट्रीय और अंतर्राष्ट्रीय सम्मेलनों, सेमिनारों या संगोष्ठियों में शोधपत्र प्रस्तुत किए, वैज्ञानिकों द्वारा 47 विशेषज्ञ/ आमंत्रित व्याख्यान दिए गए। बीते वर्ष के दौरान, केंद्र ने अनुसंधान/अकैडमिक के साथ समझौता ज्ञापन पर हस्ताक्षर किए हैं संस्थानों ने 3 प्रौद्योगिकियों का व्यावसायीकरण किया, 4 प्रौद्योगिकियों का विकास और मूल्यांकन किया, 5 नई प्रौद्योगिकियों का विकास किया और 3 पेटेंट दायर किए।

केंद्र ने भारत सरकार के निर्देशों के तहत विभिन्न गतिविधियों का आयोजन किया। अंतर्राष्ट्रीय योग दिवस मनाने के लिए योग शिविर (13-21 जून), हिंदी को बढ़ावा देने के लिए हिंदी पखवाड़ा (1-14 सितंबर), स्वच्छता अभियान (2-16 अक्टूबर), सतर्कता जागरूकता सप्ताह (31 अक्टूबर - 5 नवंबर), कृषि शिक्षा दिवस (3 दिसम्बर), राष्ट्रीय उत्पादकता सप्ताह (12-18 फरवरी) बड़े धूमधाम से मनाये गये। 26 नवंबर को वैज्ञानिक-पशुचिकित्सक इंटरफेस बैठक आयोजित करके केंद्र का स्थापना दिवस मनाया गया। आयोजित की गई अन्य संस्थागत गतिविधियाँ विश्व पशु चिकित्सा दिवस (30 अप्रैल) और राष्ट्रीय विज्ञान दिवस (28 फरवरी) थीं।

केंद्र घोड़ों के महत्वपूर्ण संक्रामक रोगों के लिए सशुल्क परामर्श और निदान सेवाएं भी प्रदान करता है। इस कार्यक्रम

of equines. Under this programme, 7907 equine serum samples were tested for various infectious diseases, including 3226 for equine infectious anemia and 3265 for glanders. The centre generated revenue of Rs 73.60 lakh from contractual diagnostic services and Rs 21.31 lakh from sale of farm produce.

के तहत, 7907 इक्वाइन सीरम नमूनों का परीक्षण विभिन्न संक्रामक रोगों के लिए किया गया, जिनमें 3226 इक्वाइन संक्रामक एनीमिया और 3265 ग्लैंडर्स के लिए शामिल हैं। केंद्र ने अपने आंतरिक स्रोतों से संविदात्मक निदान सेवाओं से 73.60 लाख रुपये और कृषि उपज की बिक्री से लाख 21.31 रूपये अर्जित किए हैं।

Introduction

ICAR-National Research Centre on Equines

Horses hold a unique and enduring importance across cultures and throughout history, embodying qualities of grace, power, and companionship that have shaped civilizations worldwide. From their early domestication over 5,000 years ago to their contemporary roles in sport, agriculture, and leisure, horses have left an indelible mark on human society. Culturally, horses symbolize strength and nobility. Their presence in ceremonies and rituals underscores their revered status in many cultures, where they symbolize prestige and honor. Economically, horses have been indispensable for agriculture, transportation, and warfare throughout history. Before mechanization, they were essential for plowing fields, transporting goods over long distances, and carrying soldiers into battle. Even today, in some parts of the world, horses remain crucial for herding livestock and navigating rugged terrain inaccessible to vehicles. In modern times, horses continue to play significant roles in sports and recreation. Equestrian events such as racing, show jumping, dressage, and polo attract enthusiasts and participants globally. These sports not only showcase the athleticism and agility of horses but also foster a deep connection between humans and animals, emphasizing trust, communication, and mutual respect.

Beyond their practical and cultural roles, horses provide therapeutic benefits through equine-assisted therapy and activities. Interacting with horses has been shown to reduce stress, improve emotional well-being, and enhance social skills, making them invaluable partners in therapeutic settings. Horses wield profound significance in the Indian subcontinent, cherished for their historical, cultural, and economic roles. To advance equine health and productivity, the Indian Council of Agricultural Research established the National Research Centre on Equines (ICAR-NRCE) in Hisar, Haryana, in 1985. This pioneering institution houses cutting-edge laboratories and facilities dedicated to research spanning equine health, equine virology, bacteriology, parasitology, immunology, pathology, medicine, biochemistry, biotechnology and more. The research activities are supported by centralized services such as animal and agriculture farms, experimental animal facility, microbial containment laboratory, AKMU cell, ATIC, library and Info-equine Museum. Complemented by the Equine Production Campus in Bikaner, Rajasthan, which focuses on breeding and management, these centers play pivotal roles in preserving and advancing prized breeds in the country. In addition, the institute also houses a national repository of microbes known as the National Centre for Veterinary Type Cultures (NCVTC). Established in 2005 at the ICAR-NRCE main campus in Hisar, this facility focuses on collecting and preserving microbes of veterinary significance derived from animals. Currently, the Centre operates through 15 network units located across the country.

MANDATE OF ICAR-NRCE

- Basic and strategic research on equine health and production
- To provide advisory and consultancy services and capacity development

OBJECTIVES OF ICAR-NRCE

- Generation of demand-driven technologies for equine health and production management.
- Capacity building for competitive equine power utilization in agricultural operations to serve the underprivileged under changing environment & socioeconomic scenario.

SALIENT ACHIEVEMENTS

During the past 37 years, ICAR-NRCE has contributed significantly in the area of diagnosis and control of equine infectious diseases by developing state-of-the-art diagnostics and biologicals. Some of the major achievements and accolades of the Centre are enlisted below:

I. Development of diagnostics

The Centre has been recognized as the National Referral Centre for diagnosis of important equine infectious diseases by the Department of Animal Husbandry, Dairying &Fisheries, Ministry of Agriculture, Government of India. The Centre has developed and refined diagnostics against various equine diseases:

- HERP kit for field diagnosis of equine herpesvirus 1 (EHV1) infection.
- COFEB kit for diagnosis of *Theileria equi*.
- Monoclonal antibody-based diagnostic kit 'Equiherpes B-ELISA' for EHV1 antibody detection.
- A type-specific ELISA and real-time PCR for differentiation of EHV1and EHV4 infections.
- Complement fixation and r-protein-based ELISA for diagnosis of glanders.
- A monoclonal antibody-based sandwich ELISA and RT-PCR for detection of equine rotavirus (ERV) from faecal samples.
- RT-PCR and real-time RT-PCR based assays for typing and diagnosis of equine influenza virus.
- A recombinant antigen based-ELISA for detection of antibodies to *Theileria equi*.
- An indirect ELISA using whole cell lysate antigen and PCR for detection of *Trypanosoma evansi*.
- ELISA and RT-PCR for diagnosis of Japanese encephalitis.
- A recombinant protein-based indirect ELISA for sero-diagnosis of glanders and equine infectious anemia.
- Lateral flow assay based rapid diagnostic for *Theileria equi* infection.
- Lateral flow assay kit for glanders.
- Lateral flow assay kit for equine infectious anemia.
- Standardization of a nested (gB-nPCR) and real-time PCR (gB- qPCR) targeting gB gene for detection of EHV1 latency.
- Recombinant protein based Indirect ELISA for detection of JEV specific antibodies in horse and pig.
- Standardization of Multiplex PCR to differentiate *Streptococcus equi* subsp*. equi* and *zooepidemicus.*
- Lateral flow assay for rapid diagnosis of trypanosomosis using different *T. evansi* antigens.
- ELISA for detection of *T. evansi* antibodies in multiple animal species.
- Monoclonal antibody-based ELISA kit for detection of equine influenza (H3N8) antigen.
- Development of Recombinant antigens based indirect ELISA kit for detection of antiTrypanosoma evansi antibodies in animals
- Developed of recombinant nucleoprotein based indirect ELISA for SARS-CoV-2 antibody detection in canines
- Standardized RT-RPA-CRISPR based LFA assay for detection of SARS-CoV-2
- Developed isothermal "Recombinase Polymerase Amplification" (RPA) based assays for detection of Porcine circovirus 3 (PCV3) in pigs.

II. Development of vaccines and immuno-biologicals

- Inactivated EHV1 vaccine "Equiherpabort" using indigenous virus for prevention of abortions in mares.
- Updated equine influenza vaccine by incorporating recent virus strain {A/eq/Katra-Jammu.06/08 (H3N8)}.
- Bacterin and outer membrane protein-based vaccine for *Salmonella* Abortus equi.
- Monoclonal antibodies against EHV-1, equine rotavirus, equine influenza, Japanese encephalitis and *Trypanosoma evansi*.
- Inactivated EHV1 vaccine using montanide adjuvant (The modified vaccine is currently under trial in horses).
- Encapsulated phage formulation carrying *Salmonella* phages for therapeutic application in poultry.
- Developed of LSD vaccine (Lumpi-ProVacInd) to prevent Lumpy skin disease (LSD) in animals
- Developed of SARS-CoV2 vaccine (Anocovax) for animals
- Development of RPA-LFA assay for detection of EHV1 & EHV4 viruses

III. Characterization of equine pathogens

- Nucleic acid sequencing of HA, M, M1 and M2 genes of equine influenza virus (EIV) isolates from 2008 outbreak (A/eq/Jammu- Katra/08, /eq/Mysore/08 and A/eq/ Ahmedabad /09) revealed clustering of Indian and Chinese isolates in a separate cluster designated as "Asian clade" and vaccine updated accordingly.
- Sequencing of VP7 gene of equine rotavirus isolates indicated circulation of G10, G3 and G6 serotypes in India.
- Whole genome sequence analysis of Japanese encephalitis virus isolated from an equine indicated virulent strain of genotype 3 is causing the disease in equine.
- The *in-vitro* cultivation of *Trypanosoma evansi* and *Theileria equi* was successfully established.
- Experimental mouse models for equine influenza and equine herpesvirus-1 infections developed.
- Complete genome sequencing of two EHV1 isolates was carried out using NGS. The primary NGS data obtained covered up to 90 % of the genome.
- Sequence comparison of Indian EHV1 isolates with other published isolates revealed that Indian isolates are more closely related to EHV1 isolates (OH03 and VA02) from Japan (97.4- 98.8%).
- Phylogenetic analysis based on US segments classified our isolates into clade 5 along the reference isolates V592.
- Genotypic characterization of *Burkholderia mallei* isolates recovered from glanders outbreaks and currently circulating isolates are differing from the older Indian isolates.
- Whole genome sequencing of RVA/Horse-wt/IND/ERV3/2003, RVA/Horse-wt/IND/ERV2/2015, RVA/Horse-wt/IND/ERV4/2017, RVA/Horse-wt/IND/ERV6/2017 carried out.
- Development of multiplex taqman qPCR for *in vitro* detection and *Streptococcus equi* subsps. eq*ui* and other *Streptococcus sps.*

IV. Surveillance and monitoring of equine diseases in India

- India has gained OIE disease-free status for African horse sickness (AHS) in 2014 based on sero-monitoring data generated by ICAR-NRCE.
- Control of EIA in India was possible due to timely diagnosis and implementing a package of practices formulated by ICAR-NRCE.

- Effective control of the equine influenza outbreak of 1987 (involving 83000 equines) was done by implementing bio-security and development of effective vaccines. Similarly, a major outbreak of equine influenza that spread in 13 different states of India during 2008-09 and caused huge mortality and economic losses was timely diagnosed and controlled in collaboration with state animal husbandry departments.
- The National Action plan for control and eradication of glanders in India was drafted by ICAR-NRCE and the same has been implemented by the Ministry of Fisheries, Animal Husbandry and Dairying, Govt. of India in June 2019.

V. Establishment of nucleus herd of equines and characterization (phenotypic and genotypic) of Indian equine breeds

- ICAR-NRCE has initiated *in-situ* conservation programme in the form of developing an equine sanctuary at EPC, Bikaner where nucleus herds of different Indian horse breeds are being maintained which include Marwari horses from Rajasthan; Kathiawari horses from Gujarat; Zanskari ponies from Zanskar valley (Jammu & Kashmir) and Manipuri ponies from Imphal (Manipur). In addition, Large white (Halari) donkeys for conservation and improvement of donkeys and exotic Poitou donkey herd for production of superior mules are being maintained in the campus.
- Seven equine breeds, namely, Marwari, Kathiawari, Kachchhi-Sindhi, Spiti, Zanskari, Bhutia and Manipuri, have been characterized on the basis of their biometric indices and coat colour.
- Microsatellite marker based genetic diversity analyzed for proposing effectual population breeding and management strategies for future.

VI. Improvement in production potential of equines

- In order to conserve the germplasm of indigenous equine breeds, cryopreservation of semen of Marwari, Kathiawari, Zanskariand Manipurstallions and Halari & Poitou donkeys are being practiced.
- Artificial insemination using frozen semen has been perfected for production of superior quality horses, mules and donkeys.
- An eCG based sandwich ELISA has been developed for pregnancy diagnosis between days 35 to 120 of gestation in mares.
- Pregnancy diagnosis between days 14 and 18 post-insemination has been perfected using ultrasonography in donkeys and in mares.
- Donkey fibre has been used to produce carpets by mixing with sheep fibres (40:60).
- Studies on assessment of fertility related genes in stallions have been assessed. Expression of SPATA1, PLCz and CRISP3 fertility genes has been studied and established their correlation with DNA integrity and mitochondrial membrane potential of the stallion spermatozoa.

  Research initiated in the direction of treatment of Fibroblastic sarcoid, excessive growth of granulation tissue (proud flesh), Alopecia and Habronemiasis using herbal formulations.
- Developed fatigue cum fitness score card for working equines.
- Customized artificial vagina has been designed for collection and cryopreservation of equine semen and customised artificial vagina was also transferred to equine farmers/ breeders.
- Developed donkey milk-based products (Bathing soap, Body butter and Lip balm)
- Estrus synchronization protocols were optimized and methods of embryo recovery from mares and successful transfer of embryos to the surrogates were optimized

- SNP markers associated with fertility in indigenous breeds of horse were studied
- Compositional changes in Halari donkey milk during Lactation were studied
- New creep feeders were designed and fabricated for optimum growth of the foals
- development of protocols for embryo transfer in indigenous horses
- development of non-egg yolk based alternative semen extender for stallion semen
- development of customised novel cryodevices for vitrification of horse embryos
- development methods for cryovial preservation of stallion semen and thawing protocols
- Production of Marwari breed horse foals through embryo transfer technology for the first time in the country

VII. Utilization of equine energy in agricultural activities

- Single animal drawn matching plough, seed drill (two furrows) and harness have been designed and developed for donkeys and mules for agricultural operations like ploughing and sowing.
- Draught ability studies conducted on adult donkeys using conventional pneumatic two-wheel cart.

The technique of vermin composting of equine dung has been optimized for use in agricultural fields.

VIII. Services to farmers and equine breeder

- Disease diagnostic services for various infectious and non-infectious diseases to equine owners, breeders, state animal husbandry departments, police and army horses.
- Health certification for movement of equines within and outside the country to promote export of horses.
- Clinical and diagnostic (including pregnancy diagnosis) services for equine diseases.
- Artificial insemination to augment the production of superior quality horses, mules and donkeys.
- Provision of quality jacks and jennies to various states, breeding societies and farmers, for production of superior quality mules and donkeys.
- On site and online consultancy in equine health and production, including tollfree telephonic advisory at Hisar and Bikaner campuses for farmers and stakeholders.
- Training and supply of educational materials for equine management, production and health.
- Organization of health camps, awareness campaigns and farmers meet in different areas of the country.
- During the COVID-19 pandemic ICAR-NRCE served as a COVID-19 testing facility amongst one of the four institutes of ICAR

IX. Patents granted

- Nano-drug delivery for quinapyramine sulphate (Patent No.310429, Application, No.2560/DEL/2011, dated 06.09.2011).
- A method for preparation of a diagnostic kit for forecasting equine herpesvirus1 disease (Patent No. 55E4-1891278 dated 25.10.2003).
- A method for preparing complement fixation test based (COFEB) kit for diagnosis of Babesia equi infection of equines (Patent No. 196690 dated 31.07.2009).

  Recombinant *Tss*A protein for detection of antibodies against *Burkoholderia mallei* and uses thereof. Application No.3610/DEL/2015.
- A recombinant protein for diagnosis of glanders (Patent No: 296824, 2018).

- Polymeric metal nanocomposites and methods of synthesis thereof (Patent No. 411620, dated 16.11.2022).

X. Patents filed

- A highly sensitive kit for detection of antibodies against *Theileria equi* in serum of equids. Application No. 2763/DEL/2012dated 06.09.2012
- Polynucleo-desequence, process, composition and methods thereof. Application No. 2560/DEL/2011 dated 06.09.2011.
- Polynucleo-desequence, processes, composition and methods thereof. Application No. 1575/CHE/2010andPCT/IB 2011/052475.
- A recombinant haemagglutinin domain containing protein for the detection and diagnosis of glanders and method of preparation thereof. Application No. 1328/DEL/2010 dated 08.06.2010.
- Recombinant *Hcp*1 protein for detection of antibodies against *Burkoholderia mallei* in Equines. Application No.4120/DEL/2015.
- *Aerva javanica* extract for the treatment of exuberant granulation tissue and tumors in horses. Application No. 201811048899, dated 24.12.2018.(Provisional).
- Modified vaccine construct for EHV 1 and methods of preparing the same. Application No 202111000312, dated 05.01.2021.
- Monoclonal antibody-based immunoassay for detection of equine influenza (H3N8) antigen. Application No 202111004847, dated 04.02.2021.
- Mutated EHV-1 (TOH Strain) genome-based vaccine construct and method for preparation. Application No.202111057300, dated 09.12.2021.
- Recombinant nucleocapsid protein based indirect ELISA kit for detection of anti-SARS-COV-2 antibodies in canines. Application No. 202111057358, dated 09.12.2021
- Hydroxychloroquine/chloroquine zinc oxide nanoparticle formulation. Application No. 202111057698, dated 11.12.2021
- Hydroxychloroquine/chloroquine zinc oxide nanoparticle formulations. Application No. PCT/IB2022/062019, dated 10.12.2022.
- Recombinant antigens based indirect ELISA kit for detection of anti-*Trypanosoma evansi* antibodies in animals. Application No. 202211008619, dated 18.02.2022
- Development of a Novel Modified Attenuated Lumpy Skin Disease Virus (LSDV) For Use as Vaccine Application No.202211013092, dated 10.03.2022
- A novel vaccine formulation (Ancovax) to prevent SARS-CoV-2 infection in animals. Application No. 202211026023, dated 04.05.2022
- A method for encapsulation of bacteriophage cocktail against *Salmonella* sp for oral delivery in poultry. Application No. 202211050633, dated 05.09.2022
- Development of a Novel test to differentiate the vaccine and field strains of LSDV. Application No. 202211074538, dated 22.12.2022.
- Chitosan-Alginate-Zinc Artificial Skin Construct. Application No. 202311086503, dated 18.12.2023.
- miR-29a serves as a novel immunovirological marker to predict the functionality of immune response to Lumpy Skin Disease virus infection. Application No. 202311063758, dated 22.09.2023

National Centre for Veterinary Type Cultures

National Centre for Veterinary Type Cultures (NCVTC) initiated its activities in 2005 for conservation of the microbial diversity of animal origin. The activities comprise acquisition, authentication, preservation, documentation, and repository database management system of animal microbes. A network programme is in operation with 15 network units located in 9 different states viz., Haryana, Rajasthan Uttar Pradesh, Himachal Pradesh, Assam, Tamil Nadu, Gujarat, Kerala and Karnataka. These network units are contributing to conservation of animal microbial diversity in three specialized areas: veterinary microbes at ICAR-NRCE Hisar, dairy microbes at NDRI, Karnal and rumen microbes at NIANP, Bengaluru.

MANDATE OF NCVTC

- National repository of veterinary, dairy and rumen microorganisms and their identification, characterization and documentation.
- Distribution of microbes for teaching, research and development of new technologies

OBJECTIVES OF NCVTC

- Exploration and collection of microorganisms of animal origin/significance/relevance
- Central storage of animal microbes from existing culture collection centres, institutions and universities
- Characterization, documentation and digitization of microbial database of cultures of animal microbes
- Development of a National Microbial Gene Bank for conserving the biodiversity of animal microbes
- Conservation (both short-term and long-term) and utilization of microorganisms.

SALIENT ACHIEVEMENTS

During the past few years, ICAR-NRCE has contributed significantly to the area of conservation of microbial diversity and characterization of microbial pathogens. Some of the major achievements of the Centre are enlisted below

I. Veterinary Microbes

- First laboratory confirmed camelpox virus zoonosis.
- First isolation of BoHV-5 from cattle, Swinepox virus from pigs and Lumpy skin disease virus from cattle.
- First confirmatory report equine pythiosis in India
- First isolation of bacteria such as *Bordetella bronchiseptica* from horse, *Actinobacillus equilli* from foal, *Staphylococcus hyicus* from pig, Methicillinresistant coagulase negative *Staphylococcus sciuri* from goats, *Trueperella pyogenes, Exiguobacterium* spp. from pigs, *Nocardia otitidiscaviarum* from equine granulomatous pneumonia, *Moraxella* (*Branhamella) ovis* from ovine keratoconjunctivitis in sheep and *Mannheimia varigena* from buffalo.
- Whole genome sequencing of viruses such as SARS-CoV-2, BPXV, LSDV, NDV and Jaagsiekte sheep retrovirus, Avian nephritis virus, Chicken astrovirus and classical swine fever virus.
- Whole genome sequencing of bacteria such as *Pasteurella multocida* sub spp. *multocida* B:2 serotype, *Trueperella pyogenes, Bordetella bronchiseptica, Clostridium botulinum* isolate from horse*, Pasteurella multocida, Actinobacillus equuli* and *Salmonella* Gallinarum.
- Isolation of anaerobic bacterium *viz., Clostridium perfringens*, *Clostridium sordelli* and *Clostridium sporogenes* isolated from disease outbreak in brick-kiln ponies; Isolation of strains of genera *Gemella, Sphingomonas, Ochrobactrum, Rodentibacter, Gallibacterius. Shewanella* and *Aggregatibacter*.

- Isolation of Novel thermo tolerant bacteriophage from Ganga River water against *Klebsiella pneumonia and* Isolation and characterization of bacteriophages against mastitis causing *Staphylococcus aureus.*
- Development of bacteriophage cocktail to ameliorate *Pseudomonas aeruginosa* infections in Biofilms
- Development of phage delivery system for safe oral delivery of bacteriophages in poultry gut.
- Characterization of bacteriophages against ESBL producing bacteria for targeting biofilms in bovines
- Methodologies developed to successfully purify a positive sense-RNA virus (FMDV) from a virus mixture containing a negative sense-RNA virus (PPRV).
- Adopted CRISPR/Cas9-medited gene editing technology to generate knock out cell lines.
- First time demonstrated that MAPK interacting kinase I (MNK1, a host factor) regulates buffalopox virus (BPXV) replication at the level of protein translation initiation.
- Identified the role of heterogeneous nuclear ribonucleoprotein A1 (hnRNPA1, an RNA-binding protein) in regulating early translation to replication switch in SARS-CoV-2 life cycle.
- Evaluated the role of p38 mitogen-activated protein kinase (MAP Kinase) in buffalo pox virus replication
- Generated flexible Gateway ORF library of equine influenza virus to study protein-protein interactions.
- First time demonstrated *in vitro* and *in ovo* broad spectrum antiviral activity of emetine against RNA and DNA viruses (PPRV/NDV/BPX/BHV-1).
- Development of isothermal "Recombinase Polymerase Ampli_ication" (RPA) based assays for detection of Porcine circovirus 2 (PCV2) and 3 (PCV3).
- Development of LSD vaccine (Lumpi-ProVac^{Ind}) to prevent Lumpy skin disease (LSD) in animals.
- Development of SARS-CoV2 vaccine (Ancovax) for animals
- Developed recombinant nucleoprotein based indirect ELISA for SARS-CoV-2 antibody detection in canines
- Development of a novel HRM-based gap-qRT-PCR for identification and quantitation of the vaccine and field strain(s) of lumpy skin disease virus.
- Development of point-of–care diagnostic for detection of SARS-CoV-2 nucleic acids employing RPA-CRISPR-LFA assay

II. Rumen Microbes

- Isolation and characterization of tannin degrading bacteria such as *Streptococcus gallolyticus* from goat; fibre degrading bacteria *Ruminococcus flavefaciens*, *Prevotella* sp. and *Butyrivibrio* sp. from buffaloes and cattle; and nitrate reducing and cellulose degrading *E. coli* from buffalo.
- Isolation of rumen fungi such as *Anaeromyces* sp., *Orpinomyces intercalaris* and *Orpinomyces joyonii* from buffaloes; *Piromyces* sp. and *Neocallimastix sp.* from goats.

III. Dairy Microbes

- Preservation of dairy microbes, *viz*, *Lactobacillus* spp; *Lactococcus* spp; *Lactococcus lactis* ssp. lactis; *Lactococcus lactis*. ssp. cremoris; *Lactococcus lactis* ssp. diacetylactis; *Streptococcus thermophiles; Leuconostoc* sp; Bifidobacterium sp; *Bifidobacterium dentium; Bifidobacterium longum; Micrococcu* sp., *Kluyveromyces lactis* and *Saccharomyces bisporus*.
- Combination of *L. lactis* ssp Lactis-C12 and *Leuconostoc mesenteroides* subsp *mesenteroides* is very suitable for curd and butter milk preparation.

- Six *Lactobacillus* sp. having phytase degrading potential and strong antifungal activity have been isolated form milk-cereal fermented products (Rabadi samples).

  An amylytic strain of *Pediococcus acidolactici* isolated has potential as starter culture in preparation of milk cereal fermented products.

LANDMARK ACHIEVEMENTS SINCE INCEPTION

Year	Salient Achievements
1985	Foundation of NRCE, Hisar
1987	Detection of first outbreak of equine influenza in northern India
1989	Establishment of Equine Production Campus, Bikaner
1990	Import of Poitou donkey from France
1995	Cryopreservation of Jack semen for AI
1996	Establishment of a herd of Marwari horses
1996	Crystal structure of mare milk lactoferrin
1997	Release of inactivated equine influenza vaccine
2003	Award of Indian patent to HERP kit for diagnosis of EHV1 infection
2005	Establishment of National Centre for Veterinary Type Cultures (NCVTC)
2006	Collection and cryopreservation of stallion semen at farmers' door
2008	Release of 'Equiherpes B-ELISA' kit for EHV1 diagnosis
2008	Release of 'Pregmare kit' for pregnancy diagnosis in mares
2009	Establishment of a herd of Zanskari ponies
2010	Re-emergence of a case of Equine Infectious Anaemia (EIA)
2011	First report of Buffalo pox virus causing concurrent disease in cow, buffalo and human
2011	Whole genome sequencing of Japanese Encephalitis (JE) virus isolated from a horse
2011	Establishment of a herd of small, grey and large white indigenous donkeys
2012	Organisation of SAARC trainings on equine piroplasmosis under OIE twinning program
2012	Development of r-protein based ELISA for Equine Infectious Anaemia (EIA)
2012	Technique for Vermicomposting using equine dung optimized
2012	Quinapyramine sulfate nanoformulation developed against *Trypanosoma evansi*
2013	Establishment of ATIC and infoequine museum
2014	Development of r-protein based ELISA for diagnosis of *Burkholderia mallei*
2014	Development of r-HSP70 based ELISA for *Trypanosoma evansi* infection
2015	ICAR-NRCE conferred Sardar Patel Outstanding ICAR institution award
2015	Release of 'Equiherpabort vaccine' for prevention of EHV1 abortions in mares
2015	Release of r-protein based Theileria equi antibody detection kit
2015	Whole genome sequencing of classical swine fever virus
2016	Organisation of SAARC trainings on equine influenza and glanders under OIE twinning program

Year	Salient Achievements
2016	Methodology for isolation of RNA virus from mixed infection developed
2017	Establishment of a herd of Kathiawari horses
2018	Ecotourism started at Equine Production Campus, Bikaner
2018	Release of ELISA kits for EHV1/4 and LFA for equine piroplasmosis
2020	Japanese Encephalitis (JE) virus antibody test kit was released
2021	Technology commercialization and transfer on semen collection and cryopreservation in Equines; commercialization of prototype of AV for semen collection from Stallions
2022	Development of Recombinant antigens based indirect ELISA kit for detection of anti-Trypanosoma evansi antibodies in animals
2022	Development of LSD vaccine (Lumpi-ProVac^{Ind}) to prevent Lumpy skin disease (LSD) in animals
2022	Development of SARS-CoV2 vaccine (Ancovax) for animals
2022	Development of recombinant nucleoprotein based indirect ELISA for SARSCoV-2 antibody detection in canines
2022	Developed isothermal "Recombinase Polymerase Amplification" (RPA) based assays for detection of Porcine circovirus 3 (PCV3) in pigs
2023	Development of point-of–care diagnostic for detection of SARS-CoV-2 nucleic acids employing RPA-CRISPR-LFA assay
2023	Developed of a novel HRM-based gap-qRT-PCR for identification andquantitation of the vaccine and field strain(s) of lumpy skin disease virus
2023	Development of RPA-LFA assay for detection of EHV1 & EHV4 viruses
2023	Production of Marwari breed horse foals through embryo transfer technology for the first time in the country
2023	Development methods for cryovial preservation of stallion semen and thawing protocols
2023	Development of customised novel cryo-devices for vitrification of horse embryos
2023	Development of non-egg yolk based alternative semen extender for stallion semen
2023	Development of protocols for embryo transfer in indigenous horses

Summary of Expenditures under Unified budget and NCVTC including SCSP, NEH & TSP

Head-wise Details	2022-2023 (Rs in Lakhs)	2023-2024 (Rs in Lakhs)
Other charges including equipment's and recurring charges	686.54	1070.22
Establishment charges including LSP/PF, wages, OTA	1267.05	1261.04
Travelling allowances and HRD	18.47	22.78
Works	5.82	57.12
Equipment	55.17	140.87
Loan and Advances	00	7.50
Disaster Emergency fund - General	75.00	69.56
Disaster Emergency fund - Capital	20.00	0.00
Non-Scheme	100.00	25.00
Total	**2228.05**	**2654.09**

Summary of Revenue Receipts

Head-wise Details	2022-2023 (Rs in Lakhs)	2023-2024 (Rs in Lakhs)
Leave Salary & Pension Contribution	7.65	0.00
Sale of Farm Produce	2.18	19.12
Sale of Livestock.	20.40	0.21
Eco Tourism	2.83	4.06
Rents (Charges & License Fee)	3.17	2.89
Contractual Diagnostic Services	69.54	66.68
Sale of cultures	1.10	0.78
Sale of Vaccine	3.43	67.48
Candidates Tuition Fees, Diploma Charges /Training Fee etc.	3.51	2.33
Sale of machine tool/etc.	0	1.10
Interest on Short Term Deposit	5.82	4.84
Interest on short term deposit under DBT/DST Project/Award Money	1.95	2.19
Recovery Loans & Advances	6.38	6.70
Interest on Loans & Advances	4.18	2.71
Other Miscellaneous Receipts	14.19	15.69
Total	**146.33**	**196.7**

Revenue generation through contractual diagnostic services and consultancy

Under contractual diagnostic services, a total 7907 samples were received from racecourses, turf club, stud farm, riding schools, animal quarantine & certification services (AQCS) and other organized sector during the year 2023. These samples were tested for various notifiable and exotic diseases to check ingression of diseases from abroad and monitoring of elite horses in private sectors. A total of 3226 sera samples for EIA and 3265 samples for glanders were tested. Among exotic diseases, 396 swab samples from for Contagious Equine Metritis (CEM), 206 sera samples for equine viral arteritis (EVA), 195 sera samples for African Horse Sickness (AHS) and 217 sera samples for dourine were received from AQCS, Govt. of India, collected from imported equines.

All the samples were found negative for these exotic diseases. Revenue of about Rs. 70.15 lakhs were generated through contractual diagnostic service.

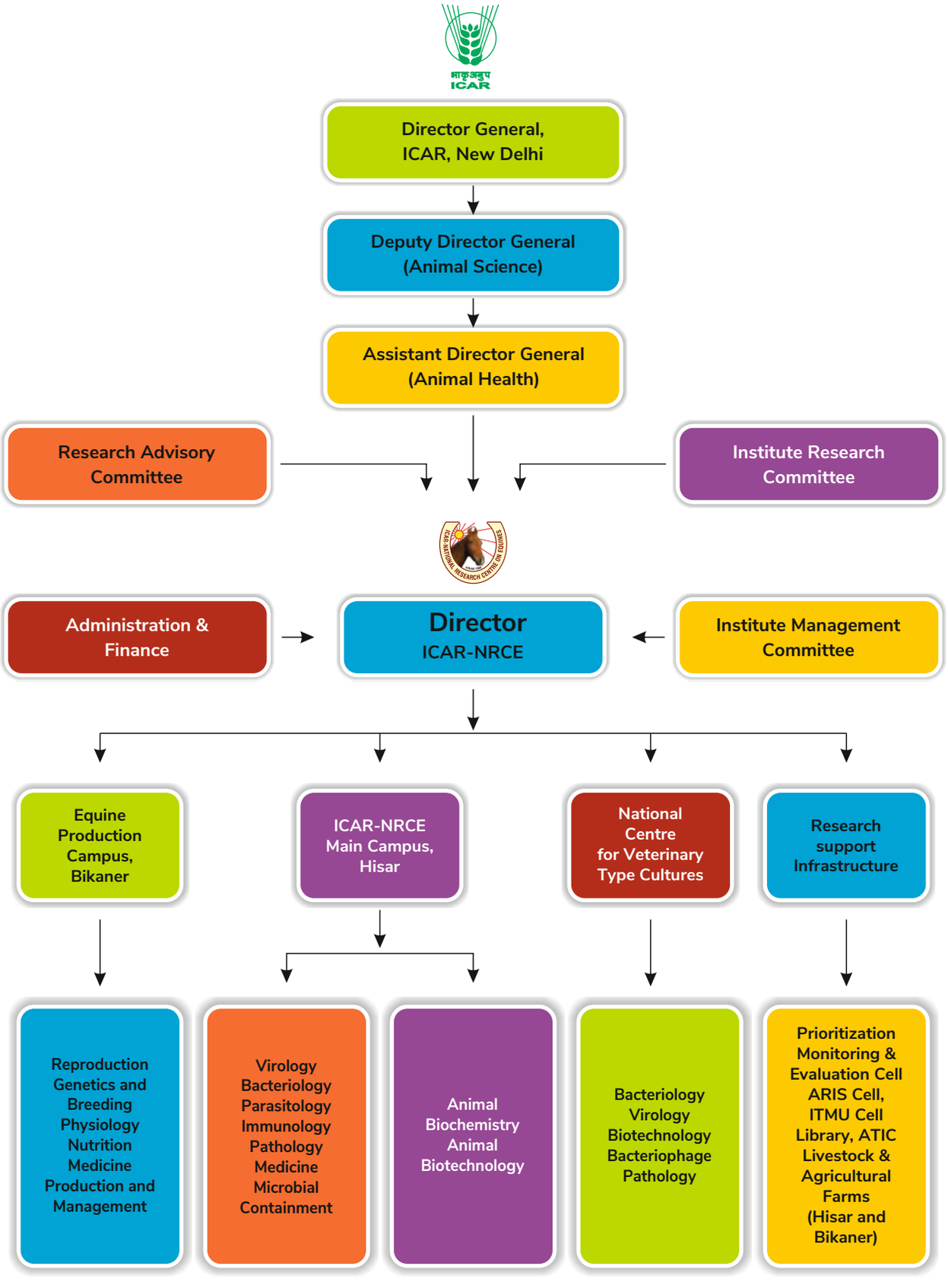
Number of samples tested and revenue generation through contractual diagnostic services (Jan-Dec.2023)

Diseases/infection diagnosis	Number of samples tested	Revenue (Rs.)
Equine Infectious Anaemia (EIA)	3226	2005700
Glanders	3265	2659150
Contagious Equine Metritis (CEM)	396	683400
Equine Herpes Virus-1 (EHV-1)	42	91600
Dourine	217	297100
African Horse Sickness (AHS)	195	240100
Equine Viral Arteritis (EVA)	206	438200
West Nile Fever (WNF)	4	8400
Theileria equi	71	46150
Babesia caballi	120	249000
Salmonella Abortus equi	30	210200
Trypanosomiasis	112	65100
Japanese Encephalitis	4	8600
Bacteriological Agent	2	3600
Equine Influenza (EI)	12	7800
Pregnancy diagnosis	5	1500
Total	**7907**	**7015600**

Staff position at ICAR-NRCE & NCVTC

Name of the post	ICAR-NRCE			NCVTC		
	Sanctioned	Filled	Vacant	Sanctioned	Filled	Vacant
Director	01	01	00	-	-	-
Scientific	23	13	10	10	07	03
Technical	25	18	07	01	-	01
Administrative	19	10	09	-	-	
Supporting	20	12	08	-	-	

Organizational SET-UP



Achievements

EQUINE HEALTH

Sero-surveillance and monitoring of emerging and existing equine infectious diseases in India

Surveillance and monitoring of equine infectious disease is one of the continuous service projects of the institute. The objectives of the project are to keep surveillance of important equine diseases, to investigate disease outbreak and its control. The project plays an important role in monitoring existing diseases as well as keeps vigilance on exotic diseases. During the year 2023, a total of 2280 equine serum samples from 9 states were tested for various diseases like Equine Infectious Anaemia (EIA), Equine Influenza (EI), *Trypanosoma evansi* (Trypanosomiasis), Equine Herpes Virus-1 (EHV-1), Piroplasmosis, Japanese Encephalitis (JEV), *Salmonella Abortus equi* and Brucellosis (Table 1). Total number of positive cases and sero-positive percentage are indicated in the Table 1. Highest seroprevalence were observed for equine *piroplasmosis* (22.67%) followed by EHV-1 (14.1 %), *Trypanosoma evansi* (1.53%), EI (1.31%) and JE (0.3%). None of the equines were found to be positive for equine infectious anaemia, brucellosis and *Salmonella Abortus equi*.

Sero-prevalence of important equine diseases among indigenous equines (Jan-Dec.2023).

State/UT	EIA	EI	Trypanosomiasis	EHV-1	Piroplasmosis	JEV	Sal. Ab. Equi & Brucellosis
Uttar Pradesh	894	894 (20)	894 (11)	894 (89)	894 (214)	894	894
Haryana	58	58	58	58 (12)	58 (14)	58	58
Jammu	192	192	192 (6)	192 (58)	192 (18)	192	192
Chhattisgarh	99	99	99	99 (13)	99 (3)	99	99
Madhya Pradesh	491	491	491 (9)	491 (114)	491 (144)	491 (6)	491
Andhra Pradesh	33	33 (6)	33 (2)	33 (6)	33 (2)	33	33
Tamil Nadu	72	72 (4)	72 (2)	72(12)	72 (9)	72	72
Uttarakhand	367	367	367 (5)	367 (15)	367 (107)	367 (1)	367
Rajasthan	74	74	74	74 (4)	74 (6)	74	74
Total	**2280**	**2280 (30)**	**2280 (35)**	**2280 (323)**	**2280 (517)**	**2280 (7)**	**2280**
Sero-prevalence (%)		**1.31**	**1.53**	**14.1**	**22.67**	**0.3**	

Number in parenthesis indicates sero-positive samples.

Disease investigation undertaken

During the year 2023, a total of 457 samples were tested for various diseases like Equine Herpes Virus, Equine Infectious Anaemia, Piroplasmosis, Equine Influenza (EI), Japanese Encephalitis, West Nile virus, *Trypanosoma evansi* (Trypanosomiasis), African Horse Sickness (AHS), Equine Viral Arteritis (EVA) and Rota virus under disease investigation is shown in Table.

Number of samples tested under disease investigation (Jan-Dec.2023)

Other Disease	DI	Positive
EHV1/4	53	12
Equine Infectious Anaemia (EIA)	209	0
Piroplasmosis	10	8
Equine Influenza (EI)	27	0
JE	32	0
WNV	23	0
T. evansi	64	4
African Horse Sickness (AHS)	20	0
Equine Viral Arteritis (EVA)	27	0
Rota virus	1	0
Total	**457**	**24**

Microbial isolations from samples

Microbiological analysis was carried out on 368 biological and environmental samples including nasal swab, tissue, abscess, aborted fetus, semen, water, feed, fecal etc. originating from Haryana, Uttar Pradesh, Jammu, Uttarakhand, Rajasthan, Punjab and Maharashtra yielded 100 bacterial isolates including *Klebsiella pnuemoniae* (n=40), *E. coli* (n=25), *Streptococcus equi* subsp *zooepedemicus* (n=21), *Burkholderia mallei* (n=6), *Streptococcus equi* (n=4), *Staphylococcus aureus* (n=3) and *Pseudomonas* (n=1).

Bacterial isolations from biological/environmental samples (Jan.-Dec. 2023)

Organism	No.	Site	Place
Burkholderia mallei	6	Nasal swab (3), Pus/Pus swab (3)	Rajasthan (4), Jammu (1), Uttarakhand (1)
Klebsiella pneumoniae	40	Nasal Swab (26), Wound swab (2), Postmortem – Liver, Colon, Kidney, Peritoneal fluid, Brain (5), Swab (2), Feed (1), Water (1), Soil (1), Env Sample (2)	UP (6), Haryana (7), Rajasthan (13), PB (7), UK (7)
E. coli	25	Stomach (1), Lung (3), Spleen (3), Heart (3), Liver (1), Kidney (1), Small & Large Intestine (3), Stomach Contents (1), Brain (1), Trachea (1), Abortion - Placental Tissue (2), Nasal Swab (1), Faecal (2), Vaginal Swab (2)	Rajasthan (12), Haryana (5), GJ (4), MH (2), UP (2)
Streptococcus zooepidemicus	21	Nasal swab (16), Eye Swab (1), Wound Swab (2), Soil (1), Vaginal Swab (1)	UP (2), Rajasthan (3), HR (4), HP (1), PB (1), UK (10)
Streptococcus equi	4	Nasal Swab (2), Wound Swab (1), Vaginal Swab (1)	PB (1), UK (2), UP (1)
Pseudomonas	1	Nasal Swab (1)	RJ (1)
Staphylococcus aureus	3	Nasal Swab (2), Postmortem - Brain (1)	UP (1), UK (2)

(H. Singha, Shanmugasundaram K, Nitin Virmani, Rajender Kumar, Sanjay Kumar, Sanjay Barua, Rajesh K Vaid, Ramesh Dedar, Anju Manuja, Balvinder Manuja, Yash Pal & T. K. Bhattacharya)

Glanders Surveillance Report : 2023

Glanders is a bacterial infection caused by *Burkholderia mallei*. The organism primarily infects equid species such as horses, donkeys and mules. Humans may acquire the disease through direct contact with infectious organism and prolonged contact with horses and mules infected with *B. mallei*. Re-emergence of glanders was detected in 2006 across seven states in India. Since then, continuous occurrence of glanders and the gradual spread of it to new territories were reported. To control and eradicate the glanders incidence, National Action Plan on Glanders in India was launched by the Department of Animal Husbandry & Dairying, Ministry of Fisheries, Animal Husbandry and Dairying, Government of India in 2019. This action plan has been framed for surveillance of the entire equine population reared in different management and animal husbandry practices following the conceptual framework of the OIE Terrestrial Code and the OIE Terrestrial Manual.

ICAR-NRCE is coordinating the glanders surveillance programme in collaboration with State Animal Husbandry Department. Two tier diagnostic system (ELISA and CFT) is being followed for the surveillance purposes. In addition, molecular methods (PCR, qPCR) and culture isolation is applied in biological samples collected from a glanders outbreaks. In the recent past, NRCE has trained laboratory personnel from different State Lab/RDDLS (Uttar Pradesh, Gujarat, Haryana, Rajasthan, Himachal Pradesh, Madhya Pradesh, Karnataka, Chhattisgarh, Punjab, Maharashtra, Bihar, Jammu, Kashmir, Uttarakhand and Tamil Nadu) on glanders diagnosis by ELISA developed at NRCE. This was found useful for setting up of network laboratory for rapid and efficient execution of surveillance activities. At present, State laboratories are using commercially available glanders ELISA kit manufactured by Genomix Diagnostic Pvt. Ltd. As per guidelines, net-work laboratories conduct initial screening by ELISA and positive samples are retested and confirmed by complement fixation test (CFT) and molecular methods at NRCE.

In 2023, a total of 44365 equine sera from 186 districts of 17 states/UT were collected and tested for glanders. Out of these, 18351 equine samples were screened by ELISA at 9 State Lab/RDDLS (Gujarat, Madhya Pradesh, Punjab, Maharashtra, Karnataka, Uttarakhand, Rajasthan, Jammu and West Bengal). Among these, Uttarakhand, Maharashtra/WRDDL, Punjab have significantly contributed by testing 5754, 4924 and 2838 equine samples, respectively.

In this year, 117 glanders positive cases were reported in 10 states. Glanders affected states include

Uttar Pradesh (n=46), Uttarakhand (n=20), Rajasthan (n=15), Punjab (n=9), Himachal Pradesh (n=7), Gujarat (n=7), Jammu (n=6), Maharashtra (n=4), Haryana (n=2) and Madhya Pradesh (n=1). It was found that around 50% of the samples and glanders positive cases were originated from Uttar Pradesh. State wise glanders surveillance data is shown in Table 5.

In zoonotic point of view, 193 sera from occupationally exposed humans (Veterinary Officers, equine handlers, and laboratory workers) were tested and none of them were found positive.

Glanders surveillance data (Jan-Dec.2023)

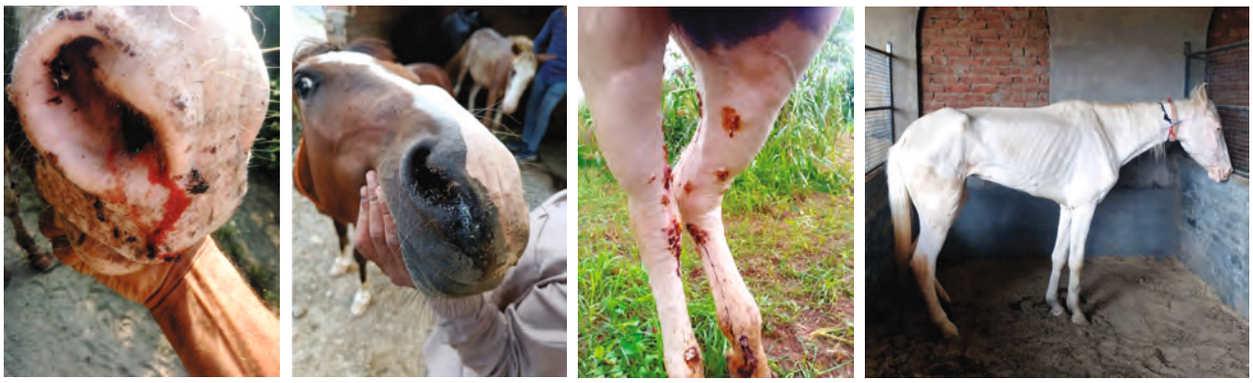
Sr. No.	State	No. of samples tested	No of samples tested at State Lab/RDDLs	No. of districts surveyed	Positive cases
1	Uttar Pradesh	18356	-	75	46
2	Haryana	702	-	14	2
3	Madhya Pradesh	508	600	21	1
4	Himachal Pradesh	2032	-	11	7
5	Punjab	44	2838	7	9
6	Uttarakhand	250	5754	5	20

7	Gujarat	40	820	9	7
8	Maharashtra	23	4924	5	4
9	Chhattisgarh	245	-	5	0
10	Tamil Nadu	73	-	3	0
11	Delhi	56	-	2	0
12	Rajasthan	1044	482	18	15
13	Jammu	2530	818	4	6
14	Kashmir	-	765	2	
15	Andhra Pradesh	82	-	1	0
16	Karnataka	5	1297	1	0
17	Kerala	24	-	3	0
18	West Bengal	-	53	2	0
	Total	**26014**	**18351**	**188**	**117**
	Grand Total	**44365**		**188**	**117**

Taking account of past five-year surveillance data, sample size has been significantly increased in this year. However, it was observed that only 12 states regularly participated in the glanders surveillance. On the other hand, irregular surveillance was observed in Southern and North-Eastern India. Therefore, pro-active participation of all State Animal Husbandry Department in the glanders surveillance programme is necessary to assess state wise sero-prevalence and to devise future strategies for control and eradication of glanders in India.

Glanders affected districts in India (Jan-Dec 2023)

State	Positive case	Distt. / Place
J&K	6	Reasi, Jammu
Maharashtra	4	Nandurbar, Aurangabad, Satara
Madhya Pradesh	1	Indore
Punjab	9	Hoshiarpur, Bathinda, Ludhiana, Amritsar, Jalandhar
Rajasthan	15	Bikaner,Sikar, Dholpur, Jhunjhunu, Jaipur
Uttarakhand	20	Rishikesh, Dehradun, Srinagar Garhwal
Uttar Pradesh	46	Prayagraj, Hardoi, Kasganj, Azamgarh, Barabanki, Mathura, Bahraich, Gonda, Meerut, Agra, Sitapur, Ghaziabad, Bulandshahr, Lakhimpur-Kheri, Muzaffar Nagar, Aligarh
Gujarat	7	Surat
Himachal Pradesh	7	Solan, Kullu
Haryana	2	Gurugram, Mahendargarh
Total	**117**	**40-Districts of 10 States**



Representative photographs of glanders affected equines

(H. Singha, Shanmugasundaram K & T. K. Bhattacharya)

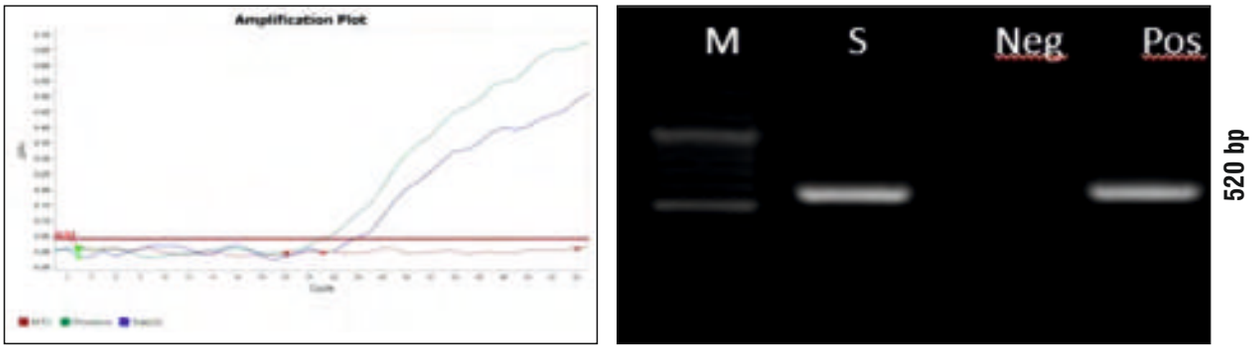
National One Health Programme for Prevention and Control of Zoonoses (NOHP-PCZ)

ICAR-National Research Centre on Equines (NRCE), Hisar is one of the Regional Coordinators under **"National One Health Program for Prevention and Control of Zoonotic Diseases (NOHPCZ)"** funded by National Centre for Disease Control, Ministry of Health and Family Welfare, Govt. of India. In this context, NRCE is actively involved in capacity building and strengthening of laboratory diagnostic facilities and inter-sectoral coordination and creating awareness about zoonotic diseases of public health importance by organizing training, workshop and webinar on various zoonotic diseases. The overall objectives of the programmes are :

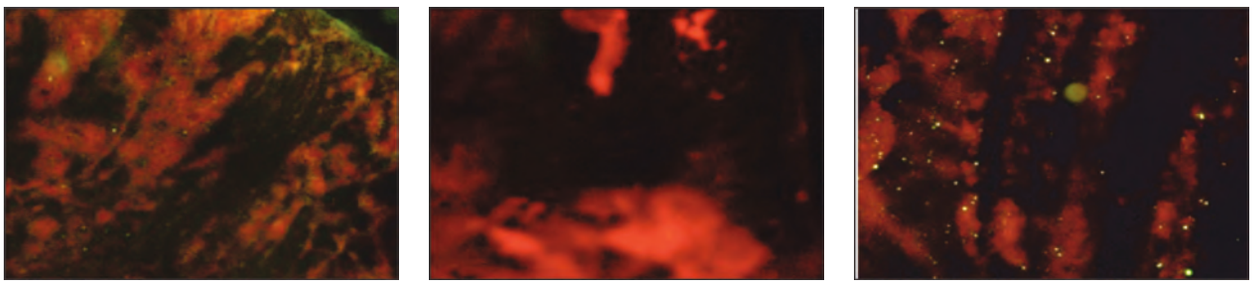
- Establish an inter-sectoral coordinating mechanism at National, State and District Level by utilizing the existing surveillance system (IDSP) to detect early warning signals of impending outbreaks for timely and effective public health actions.
- Facilitate sharing of relevant information within stakeholders for taking appropriate actions.
- Development of Laboratory capacity for diagnosis of Zoonotic diseases.
- Capacity building and creating awareness among health and veterinary professionals about Zoonotic Diseases of Public Health Importance (ZPHI).
- Formation of state-level and district level zoonotic committees
- Activities such as Information, Education and Communication for spreading awareness among target population for all ZPHI.
- Mapping of different diagnostic laboratories available in the state and working on zoonotic diseases

Rabies operational research

One step RT-qPCR assay and Direct fluorescent antibody test were standardized for rabies virus detection. A total of 50 samples including saliva, brain tissues were tested for rabies from suspected animals and out of which 13 samples were positive through RT-PCR and Direct Fluorescent Antibody testing.



TaqMan PCR | Conventional PCR



Clinical Sample | Negative Control | Positive Control

Fig. DFAT ASSAY

(H. Singha, Shanmugasundaram K, Riyesh T, Naveen Kumar & Rajender Kumar)

Immune responses and host-pathogen interaction analysis in *Burkholderia mallei* infected equines

Glanders is a fatal infectious and contagious disease of equids caused by *Burkholderia mallei* (*B. mallei*), a Gram-negative, coccobacillus, non-motile and facultative intracellular bacterium. No vaccine and specific treatment regime are available against glanders. Recent progress in *B. mallei* research identified type 6 secretion system as a major virulence determinant required for pathogenesis. Some of the effector molecules of this secretion system have shown promising results as a potential vaccine at the pre-clinical stage.

Previous findings emerged from experimental infection with *B. mallei* wild-type strain and immunization with live attenuated strain in mouse and non-human primates demonstrated that activation of early innate immune response by monocytes, macrophages, natural killer (NK) cells, antibodies and pro-inflammatory cytokines plays a critical role in controlling of the *B. mallei* infection. However, the immune response elicited by primary hosts (equids) upon natural infection is not known.

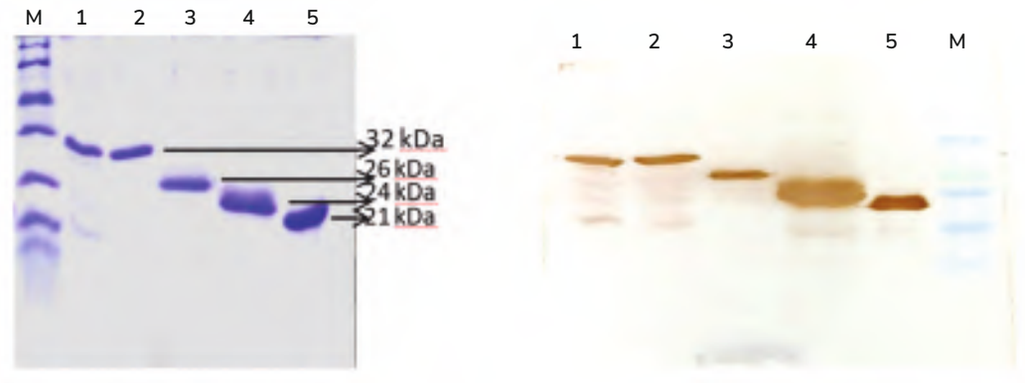
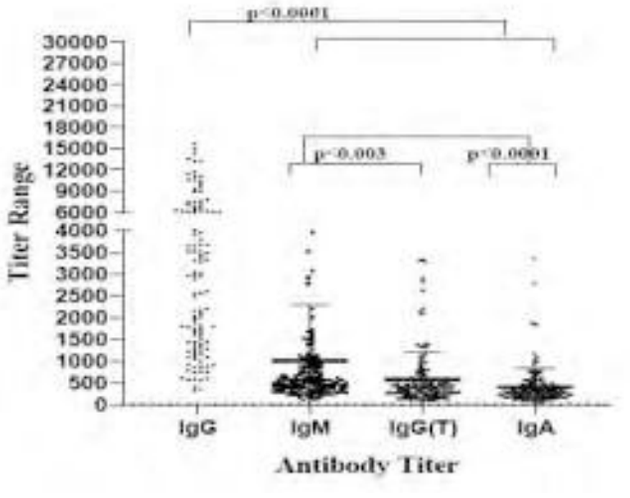


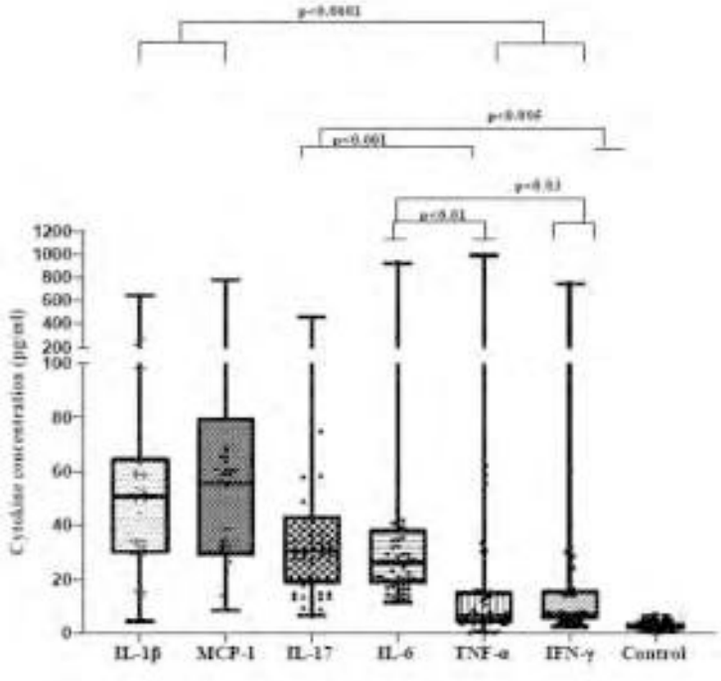
Fig. SDS-PAGE and wester blotting using recombinant *B. mallei* proteins. Lane 1 = PilA-HcP1-TssN-BipD, Lane 2 = BpaB-BpaC-BMAA0553, Lane 3 = TssB, Lane 4 = TssA, Lane 5 = Hcp1

Five recombinant proteins showed molecular weight of 21 kD, 25 kD, 26 kD and 32 kD for Hcp1, TssA, TssB, PilA-Hcp1-TssN-BipD and BpaB-BpaC-BMAA0553 respectively. Purity of the recombinant proteins were visualized by running 12.5% SDS-PAGE and Coomassie brilliant blue staining. No other co-purification was observed in the SDS-PAGE. Specific reactivity of recombinant Hcp1, TssA, TssB, PilA-Hcp1-TssN-BipD and BpaB-BpaC-BMAA0553 protein to *B. mallei* infected horse serum was determined by western blot. Among the five proteins, Hcp1 protein showed very quick and strong reactivity against glanders positive serums. These proteins were explored for IgG isotyping.



Immunoglobulin (Ig) responses (IgM, IgG, IgG (T) and IgA) to recombinant *B. mallei* proteins namely Hcp1, TssA, TssB, PilA-HcP1-TssN-BipD and BpaB-BpaC- BMAA0553 were assessed in 151 glanders positive equids by ELISA. The study showed that *B. mallei* infected equids generate strong IgG > IgM>IgG (T) and IgA antibody responses to recombinant Hcp1, TssB and TssA. Comparative analysis of immune response showed that Hcp1 was more potent immunogen in eliciting antibody response, while TssA, TssB were moderate responder PilA-HcP1-TssN-BipD and BpaB-BpaC- BMAA0553 showed minimal antibody responses.

Serum cytokine concentration of IL-1β, IL-6, IL-17, MCP-1, TNF-α, and IFN-γ were measured in 60 glanders positive equines and 10 healthy equines. These 60 glanders positive serum were selectively chosen from the panel of 151 serum used for antibody isotyping. The study revealed that IL-1β, IL-6, IL-17, MCP-1 cytokines were significantly higher in glanders positive serum.



(H. Singha, Pooja & Shanmugasundaram K)

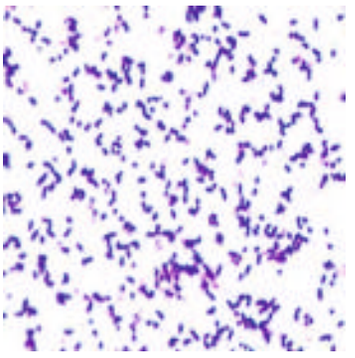
Development and evaluation of immunotherapy and vaccine constructs against *Rhodococcus equi* infection to protect foals from pneumonia

Rhodococcus pneumonia, a severe and frequently fatal form of intracellular bacterial infection in foals, is caused by the Gram-positive *Rhodococcus equi*. This respiratory pathogen mainly affects foals of less than six months of age, resulting in suppurative bronchopneumonia and pyogranulomatous lesions in the lungs. Besides, invasion of *R. equi* in the colonic mucosa leads to severe diarrhoea in foals. Conventional vaccine strategies, like live-attenuated and killed vaccines were ineffective in providing reliable protection against R. equi infection. On the other hand, contemporary vaccine approaches, including DNA plasmid vaccines, genetically attenuated vaccines, subunit vaccines, and e-beam inactivated bacterial vaccines, have yielded mixed results in terms of safeguarding foals from the pathogen. Therefore, the project was undertaken to evaluate immunotherapy and vaccine constructs to protect foals from *R. equi* infection.

The isolation and identification of *R. equi* from clinical samples such as nasal swabs of infected foals were done on nutrient agar medium. Gram's staining confirmed the Gram-positive coccobacilli. Further confirmation was performed using Polymerase Chain Reaction (PCR) targeting the *VapA* gene.

A

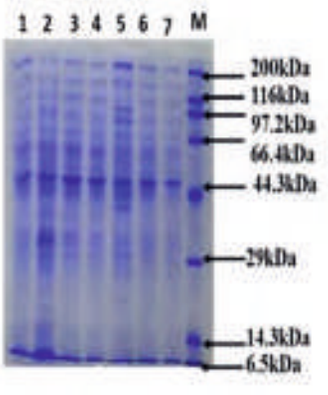
Agarose gel showing *VapA* (550 bp) positive *R.equi* (A).



B

Gram positive coccobacilli (B)

The different protocols for preparation of whole cell lysate from vapA positive *R. equi* strain 5890 were standardized. SDS lysis buffer was found as the most effective method for the purpose as this protocol yielded the highest concentration of proteins. Dialysis was performed to remove salt from the protein solution, and protein concentration was estimated using the Bradford method.



A

B C

Fig. SDS-PAGE analysis showing whole cell lysate protein *of R.equi* after dialysis (A) Western blot analyses of whole cell lysate protein of *R. equi* against *R.equi* positive serum (B) and negative serum (C)

Western blotting analysis was performed to validate the immunogenicity of the *R. equi* whole cell lysate using known positive and negative serum. *R. equi* cell lysate showed strong reactivity with positive control serum, confirming its immunogenic nature. Notably, no antigen-antibody reactions were observed against negative control serum.

In conclusion, this study optimized the protocol for preparation of *R. equi* whole cell lysate and characterized its immunogenic proteins. The whole cell lysate along with recombinant vapA proteins will be used for immunization study in mouse model and foals.

(H. Singha, Sonia, Shanmugasundaram K & Ramesh Dedar)

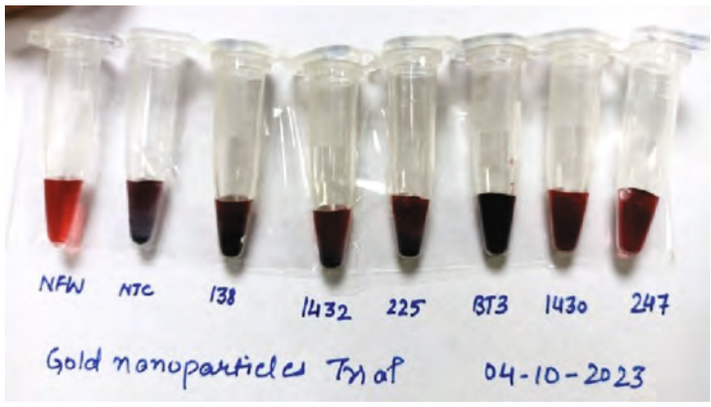
Development of diagnostics for strangles in equines

Strangles, caused by *Streptococcus equi subsp. equi*, is the most frequently diagnosed infectious disease of horses worldwide. The diagnosis of *S. equi* infection has traditionally relied on cumbersome tests like isolation and biochemical characterization of the organism. *S. equi* persists in chondroids, or possibly as a biofilm on mucosal surfaces, and can intermittently shed from carrier animals into the environment allowing transmission to naive individuals. The lack of clinical signs in persistently infected carriers emphasizes the need for effective testing procedures for identification of persistently infected carriers. Specific and sensitive tests are needed to detect serum antibodies against *S. equi* to identify the carrier animals and also differentiate from *S. zooepidemicus.* At the genomic level the two subspecies, *S. equi* and *S. zooepidemicus*, are very closely related sharing a large portion of the genome, therefore only few areas are suitable to be chosen for assays enabling differentiation between them. Scanty research in India despite the high occurrence of strangles necessitates the development of specific immunological and molecular diagnostic assays.

Serological and molecular assays for diagnosis of Strangles

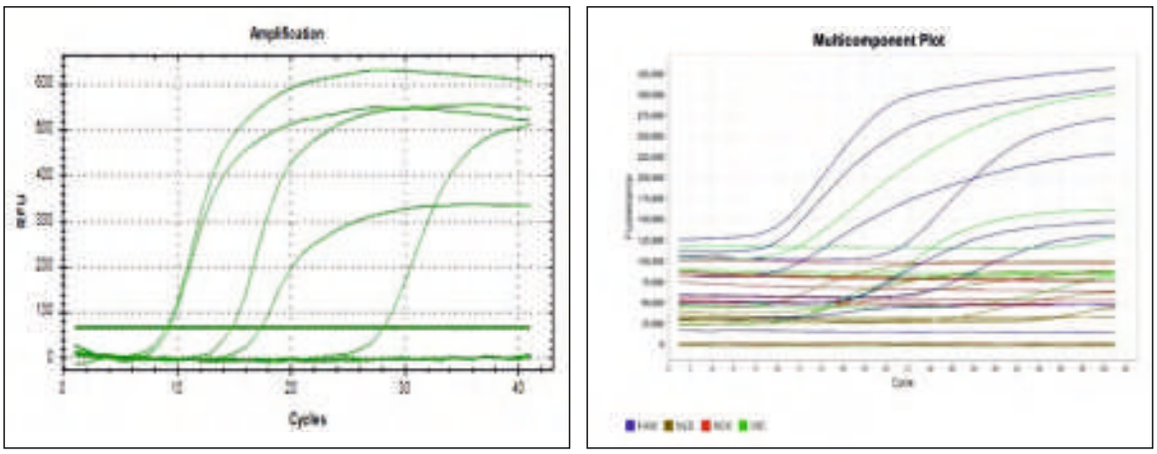
a) **Indirect ELISA to detect serum antibodies against *Streptococcus equi* :** To detect the antibodies a fixed cell ELISA was developed using whole bacterial organism. *Streptococcus equi* from continuously growing cultures were plated, using different bacterial dilutions. Indirect ELISA was developed using these antigen coated plates to detect serum antibodies. Validation studies for fixed cell ELISA were performed using commercial antibody detection kit as gold standard. The assay is being further standardized to give specific results with high sensitivity. The assay is being standardized to give specific results with high sensitivity.

b) **Recombinase Polymerase Amplification (RPA):** Recombinase polymerase amplification is highly sensitive and selective isothermal amplification technique, operating at 37–42°C, with minimal sample preparation and capable of amplifying as low as 1–10 DNA target copies in less than 20 min. Ready to use kit: Twist Dx-Twist AMP Basic kit was used for the detection of *Streptococcus equi.* The assay has been standardized for detection of the nucleic acid of *Streptococcus equi* in laboratory conditions. The amplicons have been conjugated with gold nanopaticles to make it visually detectable assay in field conditions. The assay is being further standardized for sensitive and specific detection of the nucleic acids of *S. equi.*



Gold Nanoparticles and RPA Products for coloriform assay

c) **Multiplex taqman qPCR for *in vitro* detection of *Streptococcus sps* and *Streptococcus equi* subsps. *equi* :** A multiplex taqman qPCR assay intended to be used for *in vitro* detection of *Streptococcus sps* and *Streptococcus equi* subsps. *equi* in respiratory samples (nasopharyngeal swabs, guttural pouch washes etc) for specific detection of the organisms. The assay has been internally validated by two laboratories at ICAR-NRCE and NCVTC, Hisar.



Amplification plots (qPCR) for differential diagnosis of *S. equi* and *S. zooepidemicus*

c) **Sero-surveillance for strangles in equines:** A total of 90 field serum samples were screened for serum antibodies against *S. equi* from Haryana, Jammu and Kashmir, Rajasthan, Uttar Pradesh and Uttarakhand. A total 28 samples were positive with an overall prevalence rate of 31.1%.

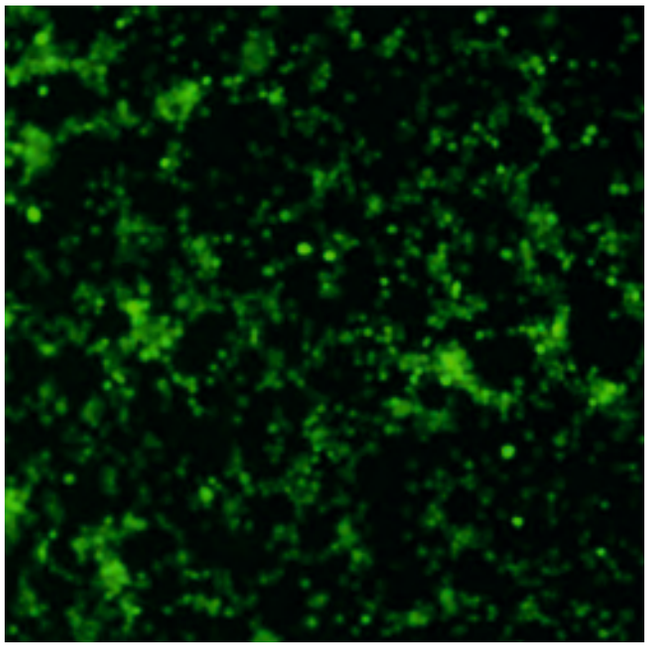
d) **Point- of- Care (POC) diagnostic kit for rapid detection of *Streptococcus equi* :** Whole genome sequencing of four field isolates (two each of *S. equi* and *S. zooepidemicus*) has been done. *In silico* analysis of the sequences initiated to identify unique sequences of diagnostic importance. Two genes coding for unique proteins of S. equi have been amplified, purified and cloned. A 22 KDa recombinant protein expressed. Further research work is in progress.

(Balvinder Kumar, R.K. Vaid, Anju Manuja, Shanmugasundarm K & H. Singha)

Development of a combined vaccine for EHV1 and equine influenza

Generation of recombinant bivalent constructs of EHV1 and EIV (Florida clade 1&2) by replacing virulence associated genes of EHV1: The recombinant genetic construct - EHV1ΔA/B with immunodominant gene of EIV Florida clade 1 and clade 2 sub-lineage was generated. For this, the GOI-KAN cassette having immunodominant gene and kanamycin resistance gene was prepared and inserted the cassette by replacing virulence associated gene from the mutant construct of EHV1 employing En Passant mediated Red1 mutagenesis and subsequently, the KAN was removed from the confirmed Red1 cones by Red2 mutagenesis. The KAN sensitive clones were selected by replica platting and further confirmed by cPCR of the deleted regions and inserted gene cassette. The genomic structure of the generated recombinant construct was confirmed by *HindIII* restriction enzyme digestion analysis in comparison with in-silico digestion profile.

Regeneration of recombinant virus: For production of recombinant virus from the generated recombinant construct, recombinant plasmid constructs was purified from the clones and transfected into RK-13 cells. The cells expressing GFP was observed, and foci of viral plaques were obtained on day one after transfection. The plaques of the regenerated virus were visible on the second day. Three days after transfection, P1 virus was passaged in the suitable cells and observed complete CPE at 96hrs. Subsequently, recombinant virus was passaged, and complete CPE was observed at 48hrs after adaptation in cell culture. The recombinant virus was bulk cultured and further works on analyses of plaque morphology and intra and extracellular growth pattern are under way.



Recombinant virus of EHV1 with immunodominant gene of EIV Clade 1

(Nitin Virmani, B.C. Bera & Taruna Anand)

Studies on prevalence of EHV2 and EHV5 infection in equines in India

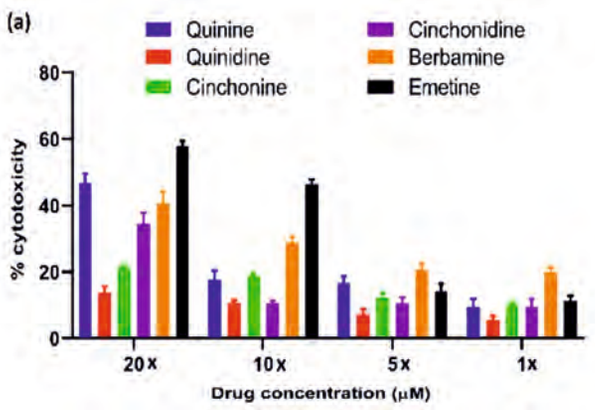
Herpesviruses are a major concern for equines and lots of studies have been conducted on various aspects of EHV1&4, however, the mild respiratory pathogens such as EHV2 and 5 are not well studied in Indian context. A realtime PCR standardized targeting the glycoprotein B sequence was developed previously and utilized for studying their prevalence. The incidence of EHV-2 and EHV-5 were evaluated from total 42 nasal samples from equids. The Real time multiplex PCR assay identified 12 (28.57%) nasal samples positive for EHV-2 while EHV-5 in 15 (35.71 %) equids. Coinfections with EHV2 and EHV-5 both were detected in 06 (14.28%) samples.

(Nitin Virmani, B.C. Bera & Taruna Anand)

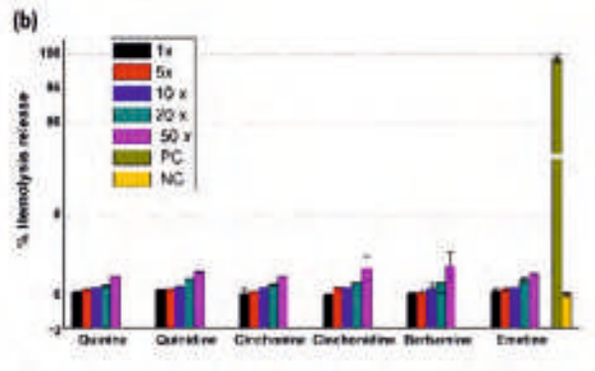
Identification and evaluation of alkaloids for anti-trypanosomal activity against *Trypanosoma evansi*

Trypanosoma evansi is one of the causative agents of animal trypanosomosis, also known as 'Surra' and represents a major limitation to livestock productivity that bring about severe economic losses especially in Asia, Africa, and South America. Limited number of available chemical drugs, incidents of growing drug resistance, and related side effects encouraged the use of herbal substitutes. Alkaloids are naturally occurring organic nitrogen-containing compounds and constitute a major class of secondary metabolites produced by plants and have been reported for myriads of pharmacological activities. Alkaloids play an essential role in both human medicine and in an organism's natural defence. Therefore, six alkaloids from two groups of alkaloids- quinoline and isoquinoline were selected to investigate the antitrypanosomal activity against *Trypanosoma evansi* in axenic culture.

Quinine, quinindine, cinchonine, cinchonidine, berbamine and emetine showed potent trypanocidal activities with $IC_{50/24\ h}$ values 6.631±0.244, 8.718±0.081, 16.96±0.816, 33.38±0.653, 2.85±0.065, and 3.12±0.367 μM, respectively, which was comparable to the standard anti-trypanosomal drug, quinapyramine sulfate (20 μM). All six alkaloids showed dose dependent cytotoxic effect on horse PBMCs (Fig. 1a) and quinine, berbamine and emetine showed selectivity index more than 5, based on ration of CC_{50} to IC_{50}. All alkaloids exhibited non-hemolytic behaviour and are hemocompatibile on blood cells up to 50x concentration of their IC_{50} (Fig. 1b). From all the compounds barbamine and emetine found more effective from all the studies carried out.



a : *In vitro* cytotoxicity on horse PBMCs.



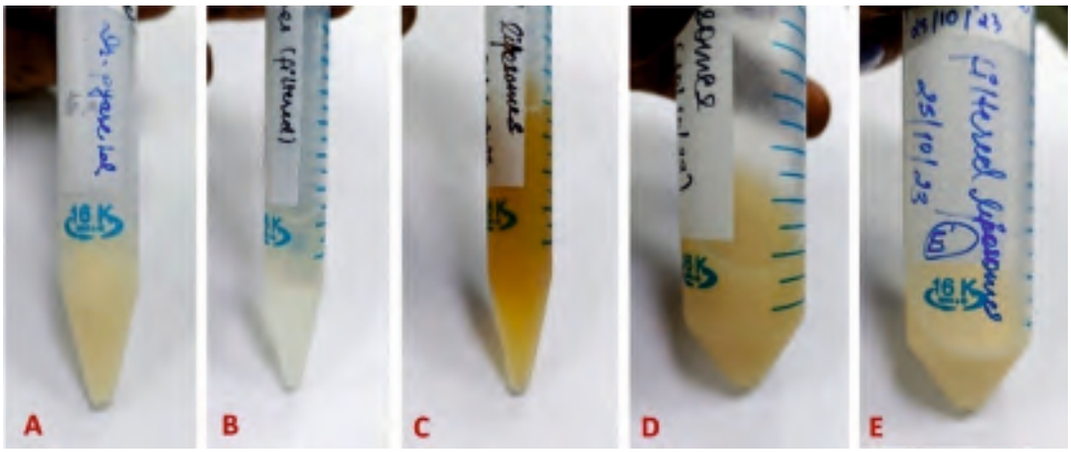
b : *In vitro* % hemoglobin release from horse blood cells at different concentrations of quinine, quinidine, cinchonine, cinchonidine, berbamine and emetine.

(Rajender Kumar, Ruma Rani & Sanjay Kumar)

Preparation of novel adjuvanted *Theileria equi* recombinant surface proteins immunogen as a vaccine delivery agent

Tick-borne pathogens (TBP) are a major source of production loss and are a concern in livestock across the globe. *Babesia bigemina*, *Theileria annulata* and *Theileria equi* are major TBP of bovines and equines, with different host types (i.e., exotic and native breeds of animals). Theileriosis and babesiosis causes major economic losses due to death, cost of treatment, and reduced production of meat, milk, and skin products. These parasites -*Theileria* sp and *Babesia* sp hide in red blood cells and causes them to stick to blood vessels, enabling the parasite to avoid host immunity and causing acute infection and death. Serological surveys conducted indicated that 30 - 60% of cross bred cattle/indigenous equines were positive for antibodies to *T. annulata*/*T. equi* piroplasms, all over India, except in Himalayan regions. Current control measures for theileriosis include chemotherapeutics, acaricides, and live vaccines (*T. annulata*), which have inherent limitations and are non-sustainable, while no effective vaccine is currently available to protect against *T. equi* infection. Effective, safe, and sustainable vaccines are needed, to boost our production and to save the precious livestock. To address this, careful selection of adjuvants and delivery systems needs to be considered in the beginning of the vaccine development process.

Keeping these points in view we initiated research work for preparation of lipid-based adjuvants by incorporating our candidate antigen. Lipids (lecithin : Cholesterol) were dissolved in organic solvent and evaporated in the form of dried thin film at the bottom of the flask. The resultant lipid film was hydrated with a buffer solution having *Theileria equi* recombinant surface proteins loaded with QuilA or MPLA adjuvant. The resultant solution was sonicated for 30 minutes at 4°C and thereafter filtered. As such five different novel adjuvants were prepared and stored at 4°C for further validation.



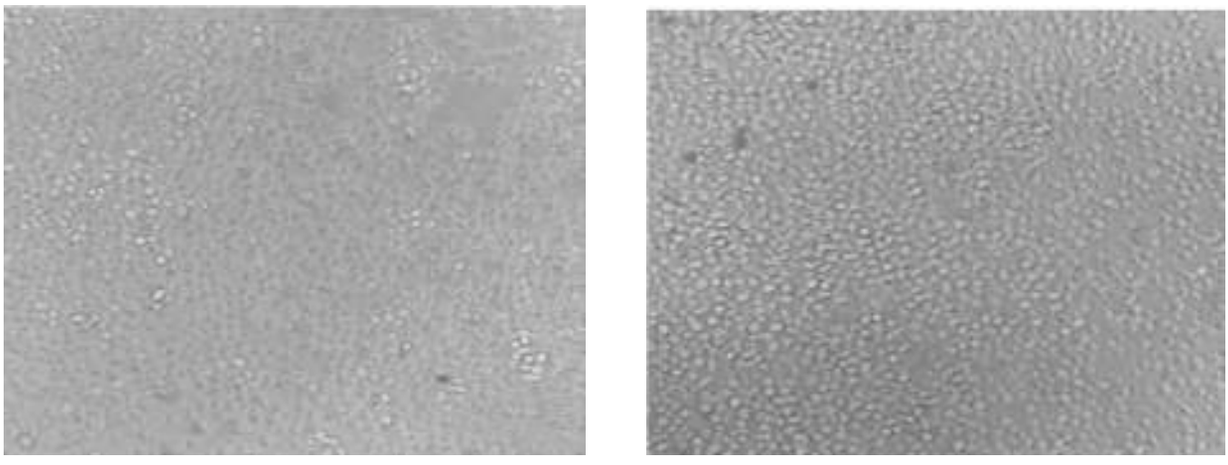
Different parameters were analyzed - PSA (Particle size analyzer), PDI (Polydisparity Index) and ZETA potential (MALVERN) and FTIR (Fourier Transfer Infrared Spectrophotometry). The values are in acceptable limits and adjuvanted proteins were stable. Further validation is under progress.

(Sanjay Kumar, Rajender Kumar, Diksha Panwar, Simran, Geetanjali & Mamta Tirdia)

Monoclonal antibodies raised against *Theileria equi* EMA-2 surface protein

Equine piroplasmosis, caused by *Theileria equi*, is an economically important tick-borne protozoan disease of horses in many regions of the world. The disease is characterized by fever, anemia, and icterus. Complete clearance or prevention of *T. equi* infection by drug therapy is not currently possible. The parasite is usually demonstrated during the acute phase of the infection by Giemsastained blood smears. However, horses that survive the primary infection are lifelong carriers of *T. equi*, and it is much more difficult to demonstrate the presence of parasites in these carrier animals, thus necessitating the use of a more sensitive test to detect carrier animals. Hence, there is need to develop *T. equi* antigen detection ELISA.

Approximately 100 to 200 μg of TE/tEMA-2, GST cleaved recombinant proteins were inoculated intraperitoneally into BALB/c mice with an equal volume of TiterMax Gold adjuvant. The mouse with the highest antibody titer was selected to be the spleen donor. Antibody titre was monitored by ELISA. Spleen cells (1.25×10^8 cells) were fused with 10^7 Sp2 myeloma cells, and hybridomas were cultured in RPMI-1640 medium supplemented with FBS, hypoxanthine, aminopterin, and thymidine in 96-well plates. Two to three weeks after fusion, screening for antibody-producing hybridomas was performed with undiluted supernatants by ELISA. Hybridoma-producing MAb wells were identified and cloned three times by limiting dilution.



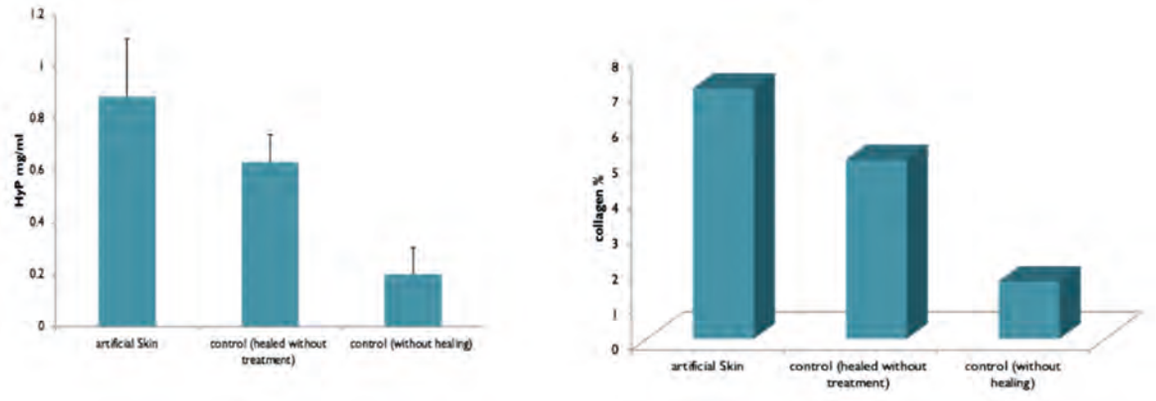
Positive *Theileria equi* EMA-2 antibody secretary hybridoma clones

T. equi positive serum samples (ELISA, RPP > 80) were obtained from the horses reared at NRCE, Hisar and EPC, Bikaner campus and samples received at diagnostic facility. *T. equi* negative serum samples collected from eight *Theileria*-free horses (ELISA, RPP < 10). These samples will be utilized for developing antigen detection ELISA. All serum samples were stored at −20°C until use.

(Sanjay Kumar, Rajender Kumar, Ruma Rani, Simran & Geetanjali)

Skin extracellular matrix specific bioink for 3D printing of skin

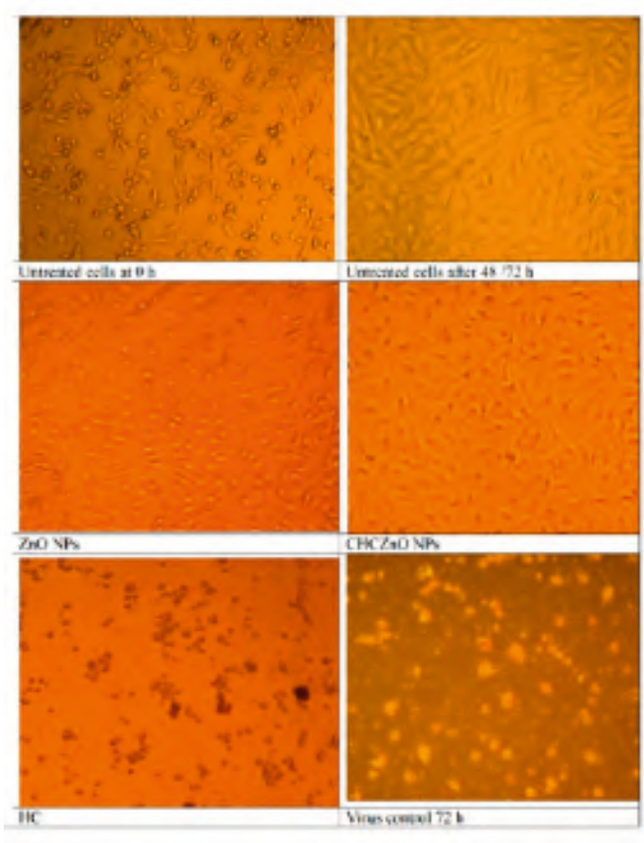
Recent advancements have been made in developing artificial skin that mimics human skin to overcome the limitations of allogenic skin replacement, such as graft rejection and the risk of infections. Artificial skin made with 3D printing technologies that include all epidermal and dermal components is a promising perspective in tissue engineering. A skin-specific bioink made from digested chicken/porcine skin and incorporated with polymers has been developed and analyzed for its structure, stability, and compatibility. The 3D printed skin showed excellent mechanical properties and proliferative ability. Histological and immunohistochemical methods were used to analyze the structure and function of the printed skin in vitro, in ovo, and in vivo. The 3D printed skin facilitated the attachment of cells to the scaffolds in the CAM assay, and wound healing was accelerated in animal studies with reepithelization increased hydroxyproline and collagen contents, TNF-α, and IL-6 genes as compared to the control. These results suggest that 3D bioprinting is a suitable technology for generating bioengineered skin for therapeutic applications.



(Anju Manuja, Balvinder Kumar, Riyesh T. & Meghnad Joshi)

Synergistic interplay of zinc-chitosan nanoparticles and hydroxychloroquine

Zinc ions can impede protein synthesis crucial for various stages of the viral life cycle. Zinc ionophores like (hydroxy)chloroquine can elevate intracellular zinc levels, inhibiting viral replication. However, both zinc and (hydroxy)chloroquine can be harmful to the host. Nanocarriers, like ZnO nanoparticles with flower-like morphology (ZnONFs) decorated with chitosan and hydroxychloroquine (CHCZnO NPs), can mitigate these adverse effects. Chitosan, a cationic polymer, was chosen for its biocompatibility, biodegradability, and ability to enhance drug transport across cell membranes. The formulation's properties were assessed, including size, shape, surface charge, and chemical interactions. Cytotoxicity, biocompatibility, and efficacy against bovine and pneumo-enteric coronaviruses were evaluated in embryonated chicks, showing promising results compared to ZnONFs/hydroxychloroquine alone. The study also delved into the lysosomotropic effect of the formulations on Vero cells infected with buffalo coronavirus. ZnONFs exhibited effective cellular uptake of zinc ions, showcasing enhanced synergy between chitosan, zinc oxide nanoparticles, and hydroxychloroquine in inhibiting bovine coronavirus. Chitosan, a cationic polysaccharide polymer, rapidly binds to the negatively charged cell membrane, facilitating the transport of zinc into cells in conjunction with hydroxychloroquine. Considering the recognized potential of hydroxychloroquine and zinc in combating viral infections, the successful delivery of this combination presents a novel strategy for developing a more potent approach against coronaviruses.



Photomicrographs (400X) of effects of formulations on African green monkey kidney cells (Vero cells) after 72 h of treatment.

(Anju Manuja, Balvinder Kumar, Riyesh T., Yash Pal & Minakshi Prasad)

Therapeutic efficacy of resveratrol in suppressing proud flesh and seasonal dermatitis in horses

Proud flesh develops when the natural wound healing process becomes disrupted, leading to excessive granulation tissue growth. This condition is commonly seen after injuries to the lower limbs of horses. In our previous reports, we demonstrated that an extract from *Aerva javanica* leaves effectively inhibits this excessive granulation and restores normal wound healing in horses. Through mass spectrometer and HPLC analysis in this study, we identified members of the Stilbenes group (specifically resveratrol and pterostilbene) in all extracts of *Aerva javanica* showing anti-proud flesh activity. We further demonstrated the anti-proud flesh properties of commercially available resveratrol. Our findings showed that topical application of resveratrol led to the gradual suppression of granulation and restored the normal healing process of wounds (27 out of 30 horses). Complete wound recovery occurred within 5–9 weeks, with the timeline varying based on wound size in horses. Taken together, we report for the first time the presence of members of the Stilbenes group (resveratrol and pterostilbene) in *Aerva javanica*, which has potential as a valuable therapeutic option for managing proud flesh in equine injuries. In our earlier study we also found that *Aerva javanica* extract successively suppressed the seasonal allergic dermatitis (Summer itch/insect bite hypersensitivity in horses). After identification of resveratrol and pterostilbene as active ingredients of the leaf extract of *Aerva javanica* we have successfully treated 20 horses suffering from seasonal dermatitis in horses by using resveratrol.

(Ramesh Kumar Dedar, Ram Kumar, Naveen Kumar, Mukesh Kumar Berwal , Shirish D Narnaware, T.R. Talluri, Jitender Singh, S.C. Mehta, & T.K. Bhattacharya)

Gene expression studies of clinically important cytokines in summer dermatitis in horses

Summer dermatitis is an extremely itchy, seasonal allergic dermatitis of horses caused by midge (*Culicoides* spp.) bites. It is also known as insect bite hypersensitivity, summer eczema, or sweet itch. Clinically, summer dermatitis is characterized by lesions that are typically found on the ventral midline, along the dorsal midline, especially at the bases of the tail and mane, and in more severe instances, also on the face, ears, and legs. The present study was therefore carried out with following objectives: (1) To study gene expression of pro-inflammatory cytokines (IL-4, IL-5, TNF-α) in skin biopsy of summer dermatitis affected horses. (2) To study gene expression of some antiinflammatory cytokines (IL-10, TGF- β1, TGF-β2) in skin biopsy of summer dermatitis affected horses. Horses (n=12) which were having history of repeated occurrence of itching and alopecia in summer season, and found negative for the presence of mites, fungal infection and bacterial infections were selected for this study. Control healthy horses (n = 12) were chosen randomly from the same stables with no history of clinical signs of skin diseases. The most common clinical signs of summer dermatitis were itching, scab, alopecia on mane, back and tail. The microscopic examination H&Estained biopsy slides of summer dermatitis affected horses have hyperkeratosis, usually associated with spongiosis. Acanthosis was present in epidermis of all affected horses. Infiltration of lymphocytes, histiocytes, plasma cells and fibroblasts were also observed in dermis. Results of the study indicated that in field cases of summer dermatitis in horses, horses bears chronic lesions of inflammatory changes caused by allergy, inflammatory reaction and chronic trauma due to itching. Expression of anti-inflammatory cytokines IL-10 and TGF-β1 may be important targets for therapeutic purpose to control chronically affected cases of summer dermatitis.

(Kalpna Godara, R.K. Dedar, T. Rao, Narender Singh, S.C. Mehta & T.K. Bhattacharya)

EQUINE PRODUCTION

Physical Characteristics of Bhimthadi

The predominant coat colour of Bhimthadi breed is Chestnut. However, other coat colours such as Roan, Bay, Dark Bay, Grey, Skew Bald are also found. White markings on the fore and hind limbs are common. Most of the horses typically have either Star, Star and strip, Snip, Blaze and Strip & snip. The Head is carried well over neck and is clearly defined. The Head is medium in size and the forehead is flat. Ears are relatively larger in size. They are carried far away from the head. A typical curvature is seen on one side of the ears in the breed. The height at withers ranges from 114 to 140 cm and adult body weight ranges from 148 to 328 kg. These horses are used by the nomads for transportation during migration and in traditional sports.

(S C Mehta & Sachin D Sorate)

Endurance Analysis using SNP markers

Endurance is a trait of significant importance in horses. Eight SNP markers associated with endurance racing were tilized for the study in five indigenous breeds of horses viz. Marwari, Kathiawari, Kachchhi -Sindhi, Manipuri and Zanskari. A total of 509 samples were genotyped for the SNP loci BIEC2-977605, BIEC2-1022884, BIEC2-11782, BIEC2755604, BIEC2-755603, BIEC2-363958, BIEC2-620109 and MSTN. One SNP marker (BIEC2-1022884) was monomorphic in Indian breeds, though it is polymorphic in exotic breeds. The SNP (BIEC2-11782) was not in Hardy-Weinberg equilibrium. The Minor Allele Frequency (MAF) and Polymorphic Information Content (PIC) were low for SNP loci BIEC2-11782 and BIEC2_977605 but the alleles were still segregating in the indigenous horses covered in the study. Hence, out of 8 markers 7 can be used in the indigenous horses for SNP genotyping for analysis of endurance potential in Indian Horse breeds.

Table 1: Status of SNPs associated with endurance racing in indigenous horse breeds (N=509)

SNP Marker	MAF	Ho	He	PIC	C_2	P-HWE
BIEC2_755603	0.47	0.43	0.50	0.37	1.143	0.565
BIEC2_755604	0.49	0.48	0.50	0.37	0.066	0.968
BIEC2_363958	0.24	0.31	0.36	0.30	0.364	0.833
BIEC2_11782	0.10	0.10	0.17	0.16	7.285*	0.026
BIEC2_977605	0.03	0.12	0.11	0.11	0.148 (3 alleles)	0.971
BIEC2_620109	0.23	0.26	0.35	0.29	4.253	0.119
MSTN	0.14	0.25	0.25	0.22	0.063	0.963
BIEC2_1022884	Monomorphic					

***P<0.05**

Fertility Analysis using SNP markers: Fertility is another trait of tremendous importance in horses. In all 293 samples of five indigenous breeds viz. Marwari, Kathiawari, Kachchhi Sindhi, Manipuri and Zanskari were genotyped at PLCz1, PLCz1-2, PLCz1-3, FKBP6-1 and FKBP6-2 SNP loci which were known to associated with stallion fertility. The SNP FKBP6-1 was not in HWE in the studied samples. The MAF and PIC was low for at PLCz1 and PLCz1-3 loci; and PLCz1-2 locus had higher frequency of G allele (0.78) which is beneficial for the stallion fertility. SNP PLCz1-3 affects the protein expression level of PLCz1. The frequency of AA/AG/GG at FKBP6-1 was 0.45/0.22/0.33 and that of AA/AC/CC at FKBP6-2 was 0.32/0.43/0.25. The AAAA combination which is associated with Impaired Acrosome Reaction was not found in studied stallions (Table 2). The association of above SNPs has also been studied with semen parameters and the results are presented in Table 3.

Table 2. Status of SNPs associated with stallion fertility in indigenous horse breeds (N=293)

SNP Marker	MAF	Ho	He	PIC	C_2	P-HWE
PLCz1	0.80	0.09	0.15	0.13	4.654	0.098
PLCz1-2	0.22	0.26	0.34	0.28	4.084	0.129
PLCz1-3	0.09	0.18	0.17	0.15	0.039	0.981
FKBP6-1	0.45	0.22	0.49	0.37	10.89**	0.005
FKBP6-2	0.32	0.43	0.50	0.37	1.093	0.579

***P<0.05**

Table 3. Association of SNPs with stallion fertility parameters. (N=18)

SNP Marker	Geno type	N	NS crotal Circum-feren (cm)	Seminal Volume (ml)	Progre-ssive Motility (%)	Live-Dead (%)	Acrosome Integrity (%)	DNA Integrity (%)	MMP (%) ce
PL Cz 1	CC	1	32.73	39.48	88.24	90.79	92.33	80.12	81.64
	C T	1	36.71	22.64	67.21	73.94	75.29	80.12	66.47
	TT	1	31.27	61.58	75.85	80.15	78.86	79.06	66.25

PL Cz 1-2	GG	1	31.43	40.9 9 a	77.29	81.88	82.09	80.94	70.70
	GT	2	28.74	107.8 2 b	77.31	80.76	73.84	72.74	61.24
	TT	2	33.06	69.92a,b	73.33	78.07	78.05	79.24	59.80
PL Cz 1-3	AA	1	30.78	61.60	77.42	81.89	80.10	79.44	67.75
	A G	4	33.55	26.51	76.37	80.53	84.12	81.42	72.49
	GG	0	-	-	-	-	-	-	-
FK BP6-1	AA	3	32.66	37.47	73.77	77.98	81.75	81.56	67.08
	A G	2	34.57	44.40	72.14	77.85	80.55	83.16	70.27
	GG	5	29.59	55.40	76.14	79.69	75.62	77.24	63.09
FK BP6-2	AA	4	28.94	59.80	71.30	76.39	71.97	72.2 7 a	59.13
	A C	1	32.16	56.70	79.70	83.98	83.73	82.1 7 b	72.04
	CC	2	32.39	21.97	75.01	78.69	84.44	82.7 3 b	70.93

The figures with different superscripts differ significantly (P<0.05)

Association of SNP Markers with Endurance Racing (Rewal Chaal)

In all 479 samples were genotyped for the seven polymorphic SNPs in Indian breeds. The performance of 77 K-Sindhi horses in endurance racing (Rewal) was analysed. SNP BIEC2_755603 and BIEC2_755604 are linked and have almost same phenotypic values in Rewal also. The heterozygous genotypes (B-03, B-04,B-82, B-05,B-09) had higher phenotypic values in endurance as well as in Rewal. SNP B-09 (DMRT) was not found associated in endurance but was found associated in Rewal Chaal. It is well known for lateral gait and ambling gait. SNP B-82 has 12 haplotypes and C allele is more common.

Thus, these markers can be used in Rewal Chaal also.

SNP Marker (Loci)	N	Genotypes	Frequency	Phenotypic Value (Speed-km/hr)	Variance explained	Comments
BIEC2_755603	64	AA/AC/CC	23-26-15	34.13/**35**/34.40 (-0.87/0/-0.60)	1.8%	Linked
BIEC2_755604	67	CC/CT/TT	21-30-16	34.10/**35.07**/34.44 (-0.97/0/-0.63)	2.3%	Linked
BIEC2_363958	72	GG/GT/TT	45-24-3	34.18/34.54/35.00 (-0.36/0/+0.46)	0.6%	
BIEC2_11782	70	CC/CT/TT	61-9-0	34.28/**34.89**/- (-0.61/0/-)	0.6%	12 haplotypes: C allele
BIEC2_977605	60	GG/AG/CG	53-3-4	34.55/**35.67/35.75** (-1.12/0/+0.08)	1.6%	3 alleles
BIEC2_620109 (DMRT)	72	CC/CT/TT	32-38-2	34.61/**34.25**/30.50 (+0.36/0/-3.75)	5.2%	Significant
MSTN	62	CC/CT/TT	4-16-42	33.75/34.00/34.86 (-0.25/0/+0.86)	2.1%	

(S C Mehta & T R Talluri)

RKVY : Conservation Marwari (Malani) indigenous breed of horse through AI

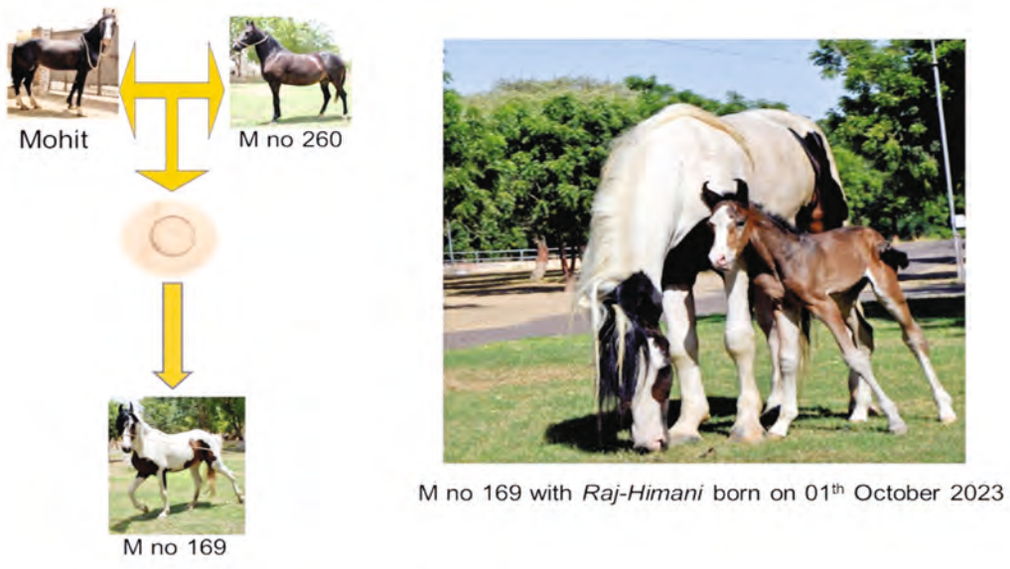
In an attempt to create country-wide network for the conservation and propagation of equines through the training on equine production with special emphasis on artificial insemination, six training programme during the period January 23-25, January 31February 1, February 13-15, February 20-22, February 27-March 1 and March 13-15, 2023 were organised successfully for the Veterinary Doctors of the Rajasthan State Animal Husbandry Department. All 33 districts of the state were covered by giving the training to 68 doctors posted in them.

(S C Mehta, T R Talluri & J Singh)

Production of live Marwari breed horse foals through embryo transfer technology for the first time in the country

The protocols for estrus synchronisation and flushing the mare for recovery of embryos were standardised. In the current year, a total of 50 flushings were made to recover 35 embryos with a recovery rate of 70%. Live foals were born through embryos conceived either from fresh or frozen semen. This is the first time in the country that the live Marwari breed foals have born through embryo transfer technology. Both the foals are healthy. DNA parentage test also confirmed the biological mother and sire status.

***Raj-Prathma*: India's First Marwari filly born through Embryo Transfer Technology**

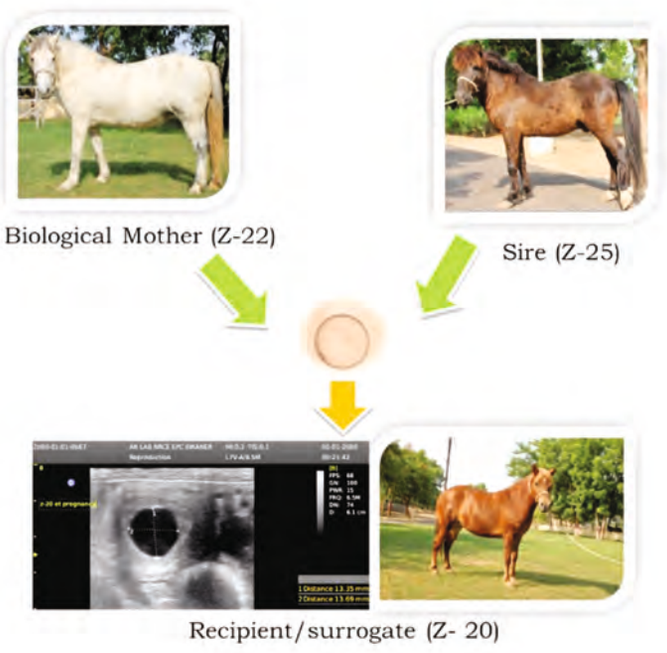


***Raj-Himani*: India's First horse foal produced through the combination of frozen semen and Embryo transfer technologies**

(T.R. Talluri, Yash Pal, R.A. Legha, R.K. Dedar & T.K. Bhattacharya)

Establishment of pregnancy in Zanskari mare through embryo transfer

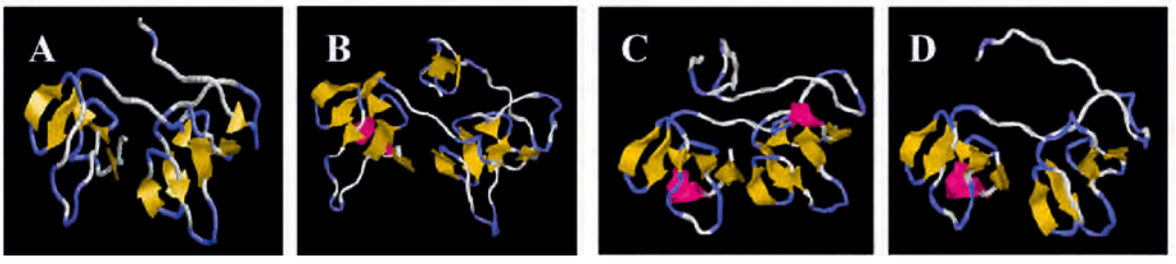
Attempts were made to recover embryos from Zanskari mares in the current year. A total of 6 attempts were made and recovered 4 embryos. Three embryos ere successfully vitrified and one embryo was transferred to estrus synchronised surrogate mare. The mare was conceived with the transferred embryo and due for foaling in the year 2024.



(T.R. Talluri, Yash Pal, R.A. Legha, R.K. Dedar & T.K. Bhattacharya)

Primary structure determination and physicochemical characterization of DSP-3, a phosphatidylcholine binding glycoprotein of donkey seminal plasma

Major proteins of the seminal plasma in a variety of mammals such as bovine PDC-109, equine HSP-1/2, and donkey DSP-1 contain fibronectin type-II (FnII) domains and are referred to as FnII family proteins. To further our understanding on these proteins, we carried out detailed studies on DSP-3, another FnII protein of donkey seminal plasma. High-resolution mass-spectrometric studies revealed that DSP-3 contains 106 amino acid residues and is heterogeneously glycosylated with multiple acetylations on the glycans. Interestingly, high homology was observed between DSP-1 and HSP-1 (118 identical residues) than between DSP-1 and DSP-3 (72 identical residues). Circular dichroism (CD) spectroscopic and differential scanning calorimetric (DSC) studies showed that DSP-3 unfolds at ~45 C and binding of phosphorylcholine (PrC) – the◦ head group moiety of choline phospholipids – increases the thermal stability. Analysis of DSC data suggested that unlike PDC-109 and DSP-1, which exist as mixtures of polydisperse oligomers, DSP-3 most likely exists as a monomer. Ligand binding studies monitoring changes in protein intrinsic fluorescence indicated that DSP-3 binds lysophosphatidylcholine (Ka = 1.08 × 105 M− 1) with ~80-fold higher affinity than PrC (Ka = 1.39 × 103 M− 1). Binding of DSP-3 to erythrocytes leads to membrane perturbation, suggesting that its binding to sperm plasma membrane could be physiologically significant.



Three dimensional structural models of DSP-3 (A), DSP-1 (B), HSP-1 (C) and PDC-109 (D). The structures were generated by computational modelling using the I-TASSER program available online (http://zhanglab.dcmb.med.umich.edu/I-TASSER) using the reported crystal structure of PDC-109 (pdb code: 1h8p) as the template.

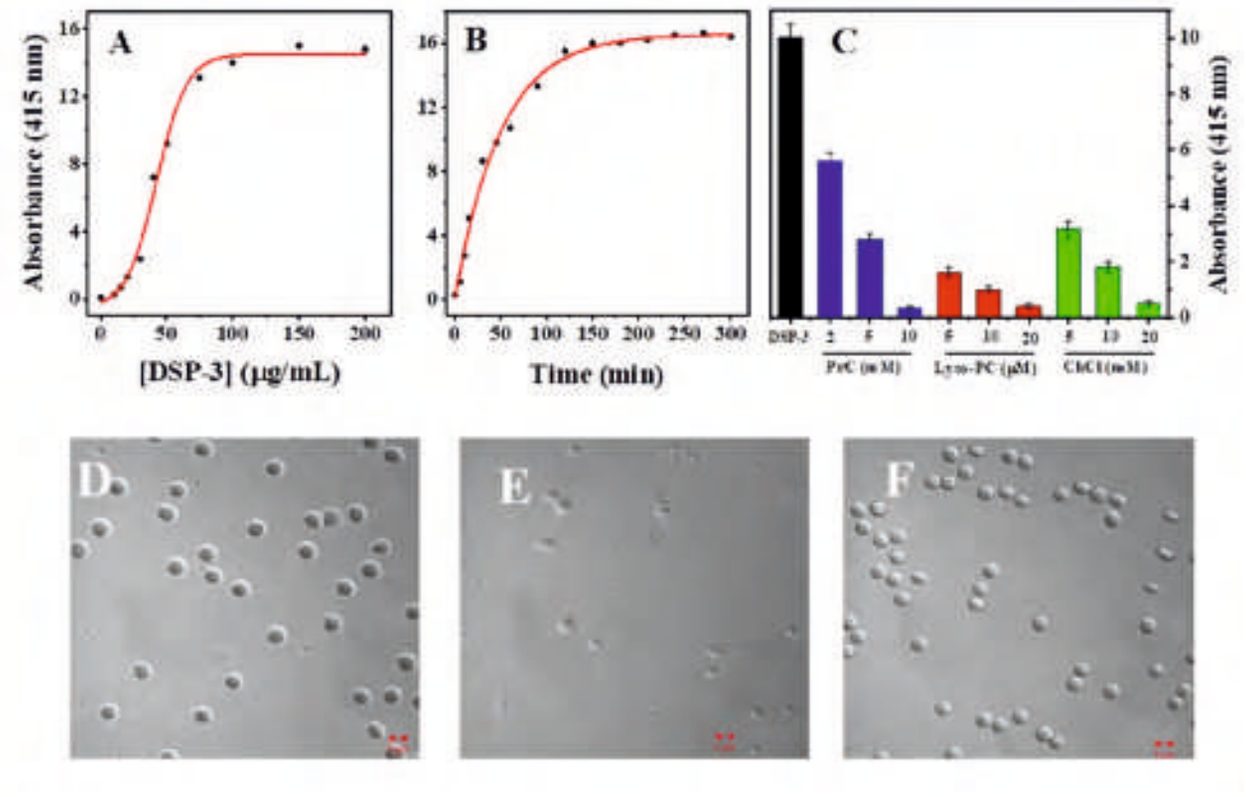


Fig. Effect of DSP-3 on human erythrocyte membrane. (A) Effect of increasing the concentration of DSP-3 on erythrocyte lysis. (B) Kinetics of erythrocyte lysis induced by DSP-3. The protein concentration in each sample was 100 μg/mL. (C) Erythrocyte lysis induced by DSP-3 alone and upon pre-incubation with different concentrations of Lyso-PC, PrC and choline chloride. Absorbance at 415 nm was measured to detect the haemoglobin released upon cell lysis. (D-F) Microscopic images of human erythrocytes under different conditions: (D) in TBS buffer alone, (E) upon incubation for 60 min with 100 μg/mL DSP-3 and (F) upon incubation for 60 min with 100 μg/mL DSP-3 that was pre-incubated with 20 mM PrC. Scale bar = 5 μm.

(T.R. Talluri & Musti J Swamy)

Effect of melatonin supplementation to the semen extender on cryopreserved stallion semen parameters

In the current study, the stallion semen was supplemented with different levels of melatonin (1mM, 1.5 mM and 2mM) and its effect on seminal parameters and oxidative parameters was evaluated during cryopreservation. Addition of Melatonin significantly improved freezability and cryosurvival rate of stallion spermatozoa as revealed by increased seminal parameters. Maximum beneficial effect of addition of Melatonin on cooled and post-thaw semen was observed at dose rate of 2mM. Addition of Melatonin significantly increased T-AOC levels and significantly reduced MDA levels indicating increased in antioxidant levels and reduction in oxidative stress and lipid peroxidation levels.

Effect of melatonin supplementation on post thaw seminal parameters in Marwari stallions

Group	Progressive Motility	Viability	HOST	DNA Integrity	Acrosome integrity
C	29.91a±1.11	57.14±3.16	36.2±1.56	84.21a±0.21	83.25±1.31
T1 (1m M)	34.44ab±1.62	58.98±3.62	36.75±1.13	84.34b±0.34	85.53±1.22
T2 (1.5m M)	38.33ab±1.87	60.84±3.25	38.2±1.21	87.25c±0.19	85.81±1.15
T3 (2m M)	42.44b±1.86	61.17±3.31	39.98±1.16	89.44d±0.34	87.12±1.41

Effect of melatonin supplementation on oxidative paramteres in Marwari stallions

Group	T-AOC	MDA
C	1.21a±0.32	6.96a±0.73
T1 (1m M)	2.29b±0.38	4.75a±0.53
T2 (1.5m M)	3.12c±0.29	3.66ab±0.51
T3 (2m M)	3.6d±0.31	2.49c±0.29

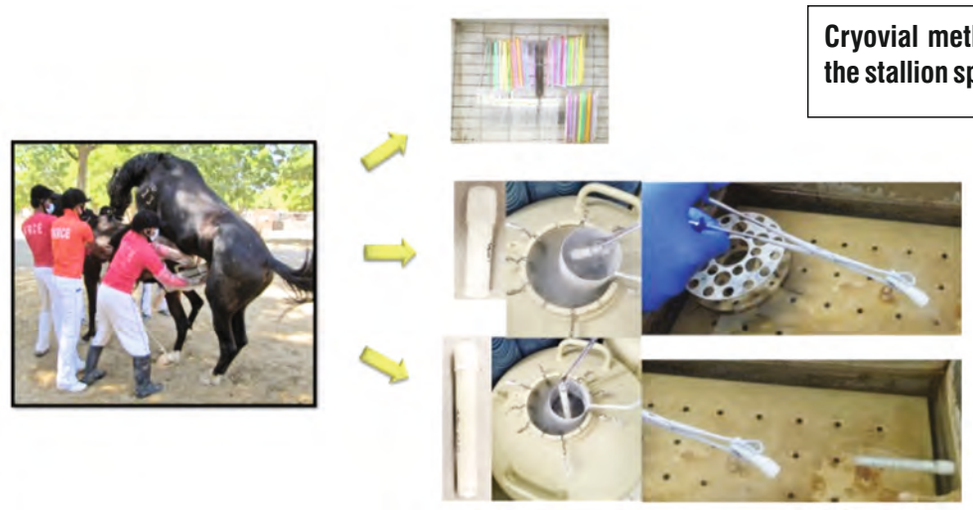
Note:- Mean values with different superscripts in a column differ significantly (P<0.05).

(T.R. Talluri, R.K. Dedar, S.C. Mehta & T.K. Bhattacharya)

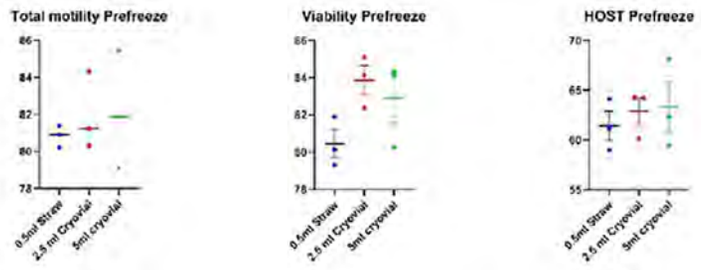
Cryovial methods of cryopreservation of stallion spermatozoa

Current methods of cryopreservation of stallion semen is storing in 0.5 ml straws.

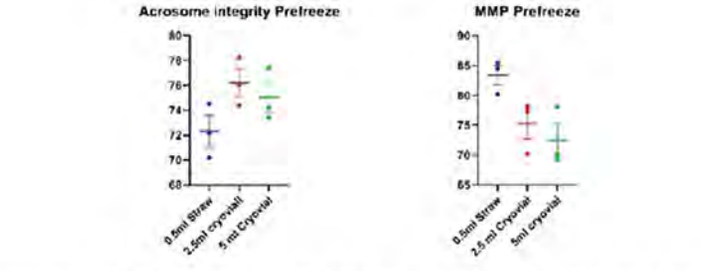
Therefore, we need to collect 10 straws from the LN2 container and thaw them at same time and cut them uniformly in the same time to get more live and motile spermatozoa for insemination. To circumvent these process alternative methods of storage of equine semen were explored and successfully stored the equine semen with appreciable post thaw semen quality using 2.5ml and 5 ml cryovials.

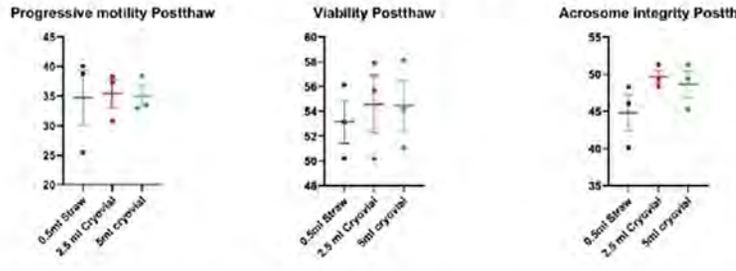


Cryovial method of cryopreserving the stallion spermatozoa

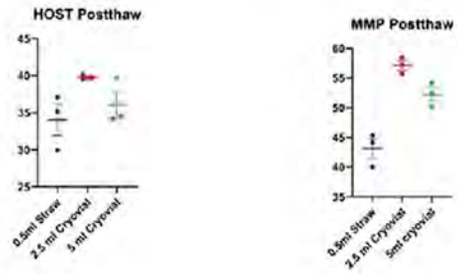


Seminal parameters at prefreeze stage of cryopreservation (0.5 ml, 2.5 ml and 5 ml cryovials)





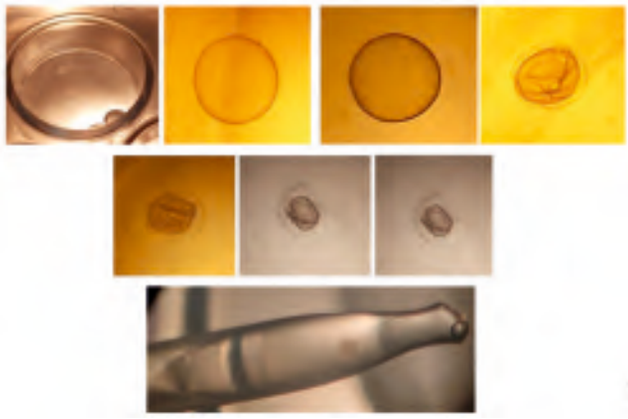
Seminal parameters at postthaw stage of cryopreservation (0.5 ml, 2.5 ml and 5 ml cryovials)



(T.R. Talluri, R.K. Dedar, S.C. Mehta & T.K. Bhattacharya)

Vitrification of horse embryos

Vitrification is a fast freezing process. It is highly difficult to vitrify the horse embryos and especially if the size of the embryos reaches above 300μm. The horse embryos flushed on day 7 and 8 would ideally be ranging between 300-400μm. This year we attempted 50 flushings out of which 35 embryos were recovered. Out of the 35 embryos, we attempted 20 embryos for vitrification and 18 embryos were successfully vitrified using hemi-straw, cryoloop and Cryotop devices. The embryos survival rate after thawing was observed to be 95.6%.



Vitrification of horse embryos (day 8.5)

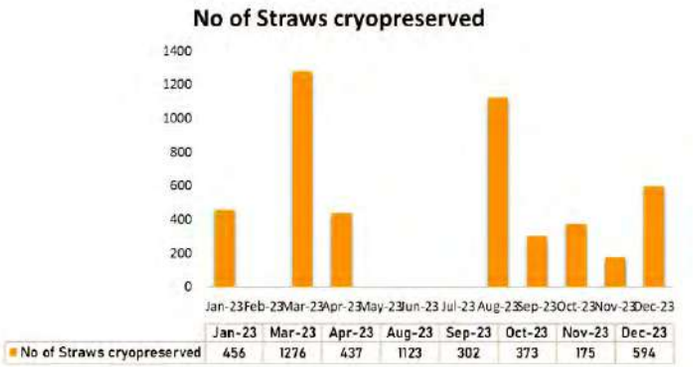
Summary of vitrification experiments of horse embryos

Day of Flushing	Total Flushings	Embryos recovered	Recovery rate	No. of Embryos Vitrified
Flushings on Day 7.5 post - AI	13	12	92.30	10
Flushings on Day 8.5 post - AI	11	8	72.72	8
	24	20	**83.33**	**18**

(T.R. Talluri, Yash Pal, R.A. Legha, R.K. Dedar & T.K. Bhattacharya)

Cryopreservation of semen from indigenous horses

In the current year, we have cryopreserved a total of 3967, 594 and 175 straws from Marwari Manipuri and Zanskari breed of stallions available at Equine Production Campus, ICAR-NRC on Equines. During the current year, the team also visited the Dundlod stud farm for stallion semen cryopreservation and successfully cryopreserved 901 semen straws from six Marwari stallions.



Total number of semen straws cryopreserved during the year 2023

Semen collection and cryopreservation at farmer's doorstep

Artificial insemination at the farer's doorstep using frozen semen

Detection of novel SNPs of the MSTN gene among Indigenous donkey breeds

The whole blood samples were collected from the jugular veins of 36 donkeys of Halari, Spiti breeds and also donkeys from the Leh region and Genomic DNA was extracted. For the amplification of the *MSTN* gene, PCR reactions were conducted. The 36 PCR products of *MSTN* gene were purified and purified PCR products were sequenced through AgriGenome Pvt Ltd, Kochi, Kerala. The single nucleotide polymorphisms (SNPs) have been analysed MSTN gene through UGENE software. The obtained gene sequences were aligned with the reference sequence of one international horse (AY840554.2) and 3 international donkey (MZ169554.1, MW970078.1, MW970079.1) through Clustal-W by using MEGA-X. Total three SNPs has been detected in exon 2 of chromosome 18 after analyzed with the international horse reference sequence (Mongolian horse-AY840554.2) in 36 samples of four Indian donkey breeds. Out of three SNPs, two novel SNPs (T>C, transition) have been detected at nucleotide position 2396 (codon12) and 2398 (codon 13) respectively and One SNP (G>A, transition) has been found at nucleotide position 2422 (codon 21) as shown in figure 4 and 5. The Phenylalanine (T**T**T) is converted into Serine (T**C**T) at codon 12 and Alanine (**G**CT) is converted into Threonine (**A**CT) at codon 21 so it is a non-synonymous mutation, while Leucine (**T**TG) is converted into Leucine (**C**TG) at codon 13 so it is a synonymous mutation. Based on the analysis of all *MSTN* gene sequences with reference sequences, we found only mutant type mutations in all studied Indian donkey breeds. We also compared all 36 samples of four Indian donkey breeds with the international donkey sequence (Guangling donkey-MZ169554.1, Turkey donkey-MW970078.1, MW970079.1). In this analysis, we found that the studied Indian donkey breeds are 100% similar to the reference sequence of Guangling and Turkey donkey breeds. In our study, novel SNPs of the *MSTN* gene have been discovered for the first time and partial DNA fragments of the *MSTN* gene have been obtained from Indian donkeys for the first time. The sequences of *MSTN* gene were submitted to the NCBI GenBank with the accession number: OQ436746- OQ436755, OQ447192-OQ447217.

Novel SNPs in *MSTN* Gene in Indian Donkey

Sr. No.	SNPs present in codon	Nucleolid e change	Codon Position	Mutation Type	Mutation position (nucleotide)	Coden No.
1	T**T**T**12**T**C**T	T > C	Second	Transition	2396	12
2	**T**TG**13C**TG	T > C	First	Transition	2398	13
3	**G**CT**21A**CT	G > A	First	Transition	2422	21

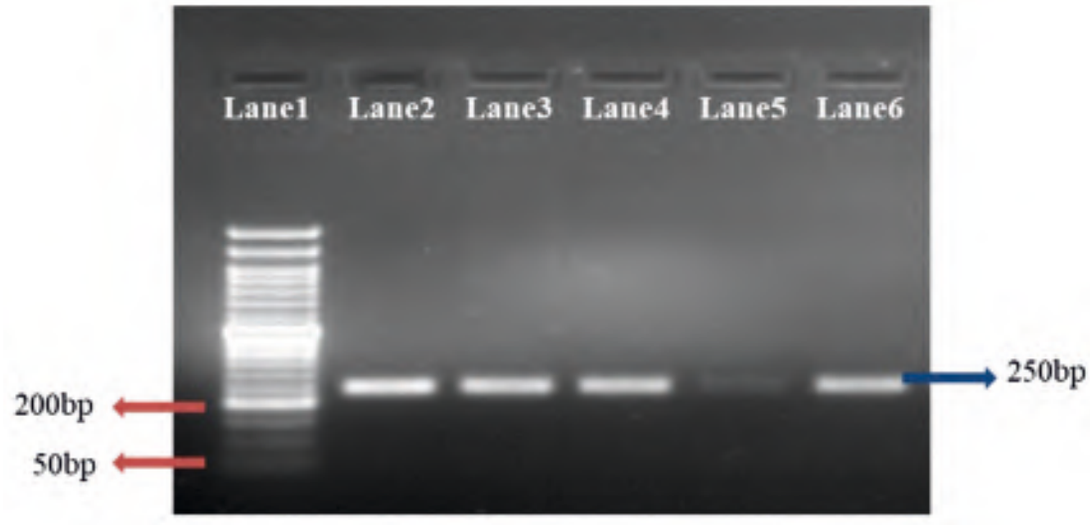


Fig. Lane-1 is showing 50bp DNA ladder, **Lane-2** Halari, **Lane-3** Spiti, **Lane-4** Leh, **Lane-5** Blank and **Lane-6** French (Poitu) donkey are showing 250 bp PCR product of *MSTN* gene of Indian Donkey breeds.

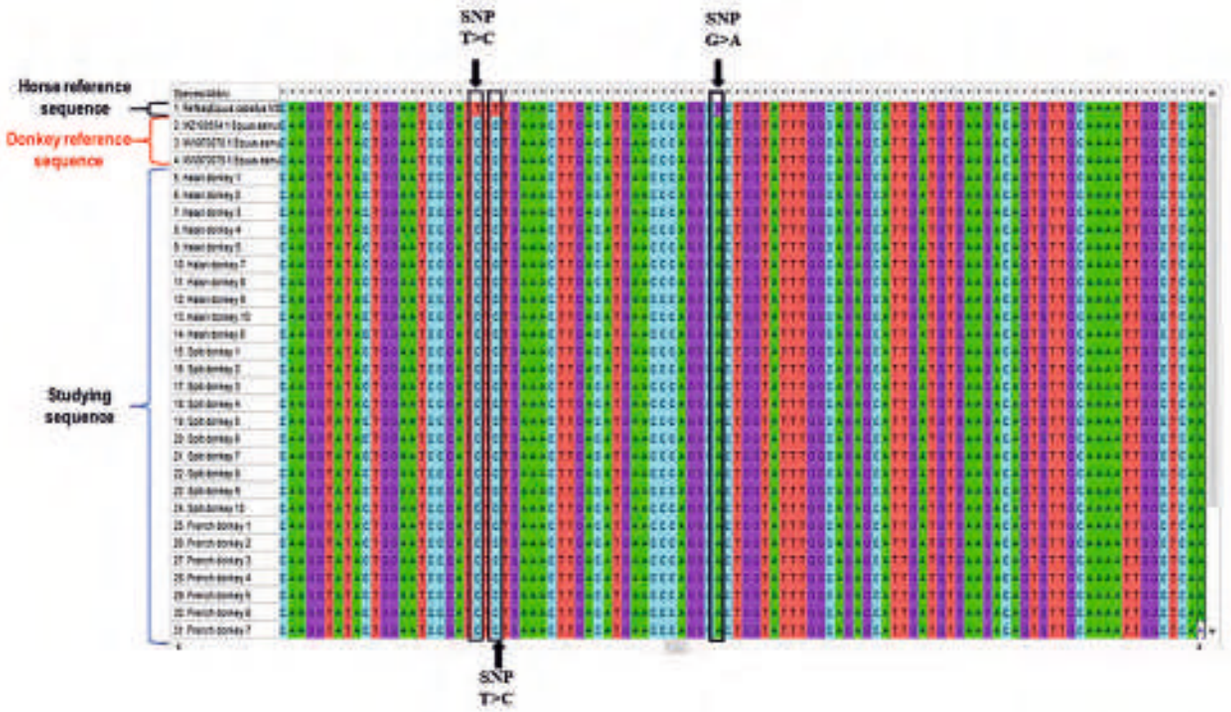
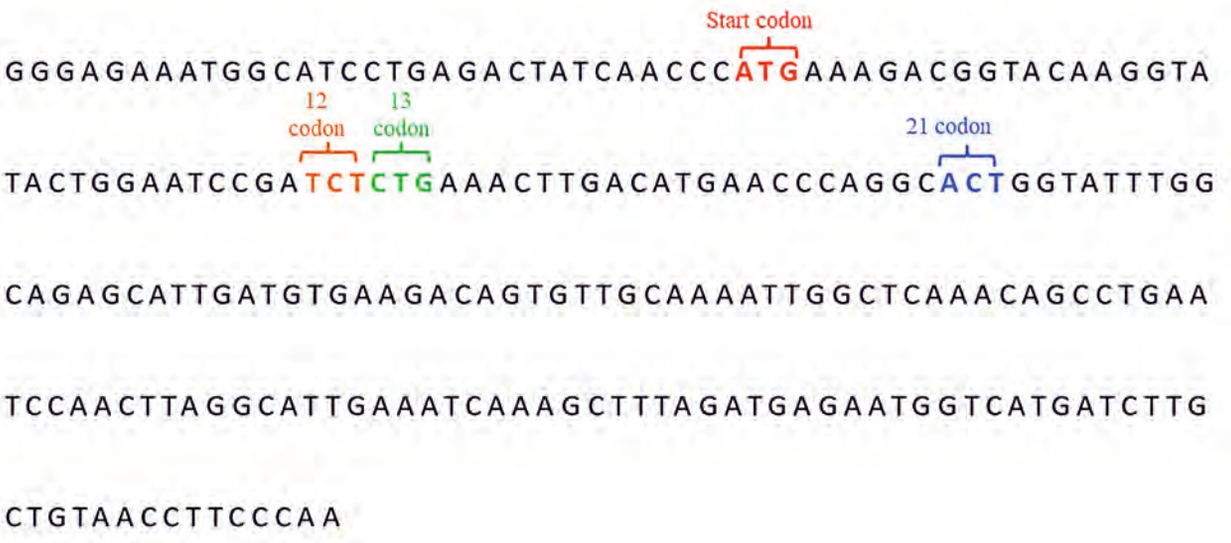


Fig. SNP's of T>C (nucleotide positions 2396 and 2398) and G>A (nucleotide position 2422) have been detected in MEGA software in the Donkey breeds



T>C SNP (12 and 13 codon) and G>A SNP (21 codon) has been detected.

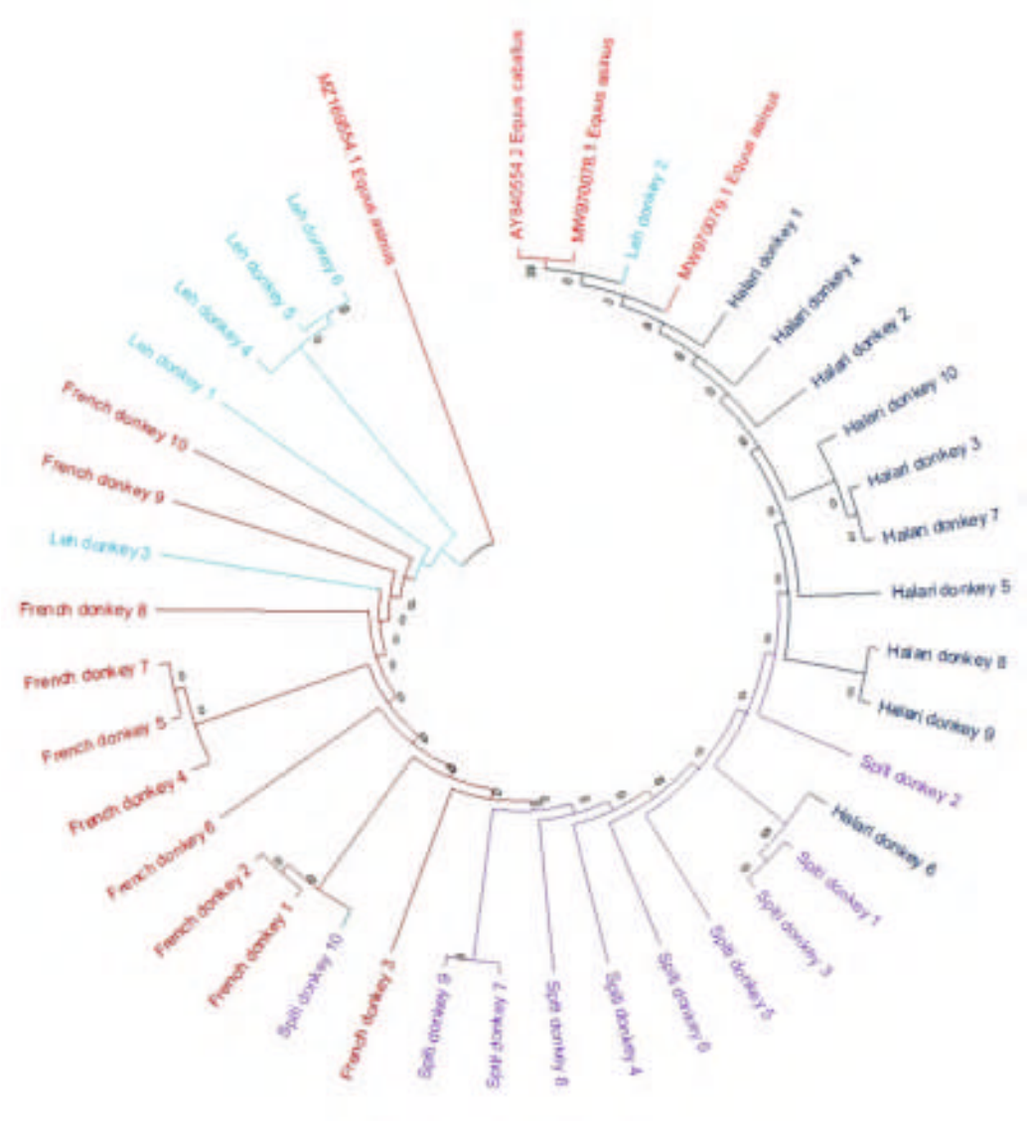
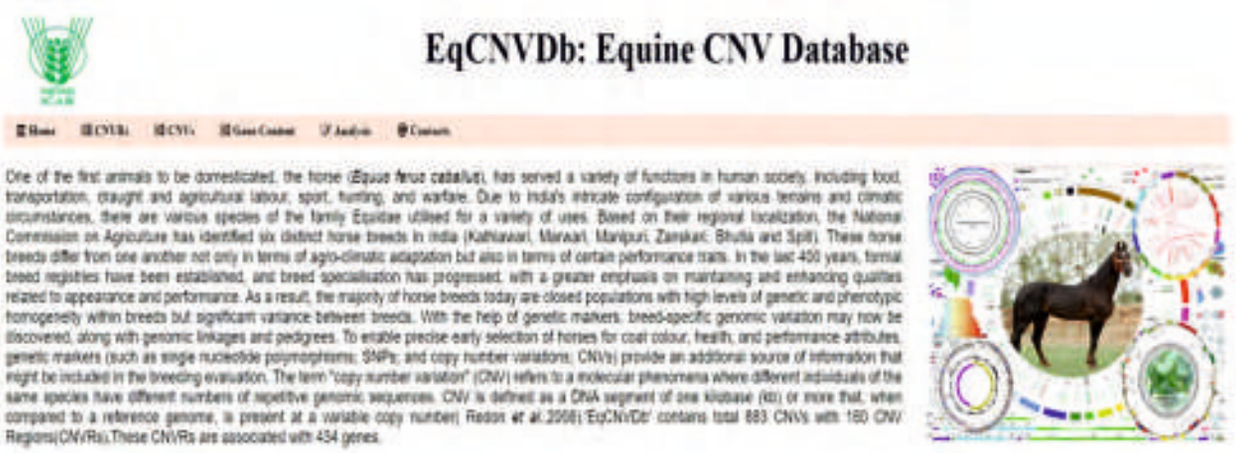


Fig. Maximum likelihood tree with 1000 bootstraps using Kimura 2 distance parameter model is showing the phylogeny of 36 sequences of Indian Donkey breeds {Halari, Spiti and Leh donkey and French donkey (Poitu)} respectively

(Sonali, Anuradha Bhardwaj, Shiv Kumar Giri, Yash Pal, Varij Nayan, Mir Asif Iquebal, Sarika, Dinesh Kumar & B. N. Tripathi)

Creation of Equine CNV Database

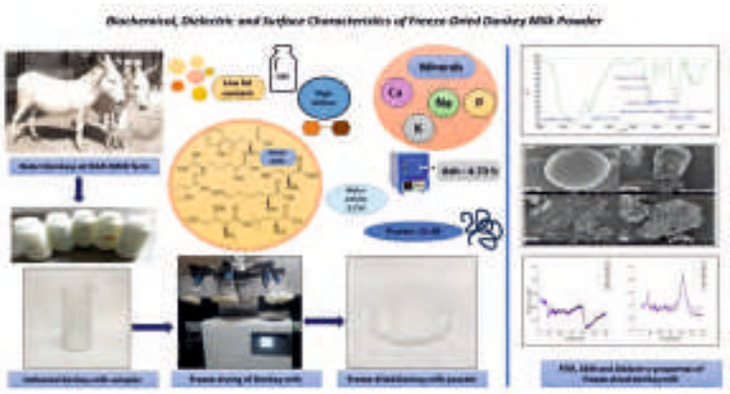
These horse breeds differ from one another not only in terms of agro-climatic adaptation but also in terms of certain performance traits. With the help of genetic markers, breed-specific genomic variation may now be discovered, along with genomic linkages and pedigrees. To enable precise early selection of horses for coat colour, health, and performance attributes, genetic markers (such as single nucleotide polymorphisms; SNPs; and copy number variations; CNVs) provide an additional source of information that might be included in the breeding evaluation. The term "copy number variation" (CNV) refers to a molecular phenomena where different individuals of the same species have different numbers of repetitive genomic sequences. CNV is defined as a DNA segment of one kilobase (kb) or more that, when compared to a reference genome, is present at a variable copy number (Redon et al.,2006).'EqCNVDb' contains total 883 CNVs with 180 CNV Regions (CNVRs). These CNVRs are associated with 434 genes.



(Nitesh Kumar sharma, Prashant Singh, Bibek Saha, Anuradha Bhardwaj, Sarika,Yash Pal, M. A. Iquebal, Varij Nayan,U B Angadi, Shiv Kumar Giri, Anil Rai, Ram Avatar Legha, Dinesh Kumar & T.K. Bhattacharya)

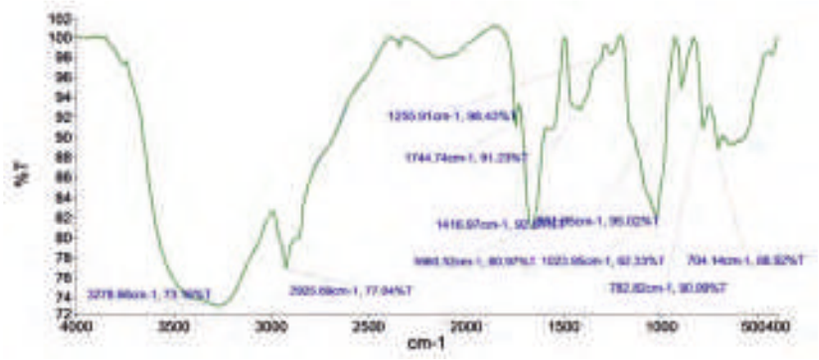
Biochemical, dielectric and surface characteristics of freeze-dried donkey milk powder

Freeze-dried donkey milk powder (DMP) was analysed for its chemical composition, amino acids, mineral composition, fatty acid profile, surface morphology using SEMEDS, FTIR, dielectric properties, as well as functional and flow characteristics. Samples of donkey milk obtained from nine distinct animals were collected throughout the 21st to 30th week of lactation, during the months of August to October. The findings of this study revealed substantial amount of various nutritional parameters in DMP. Notably, high lactose content (62.09 %) was obtained, protein content was determined to be 21.49 %, indicating a significant presence of this essential macronutrient. Fat content was relatively low (2.19 %). Minerals, amino acid, and fatty acid profiles were found to be adequate, further highlighting its nutritional value of DMP. SEM-EDS revealed slightly spherical shape particles with a smooth surface. The FTIR spectra analysis validated the existence of distinct functional groups indicative of carbohydrates and proteins in DMP.



The Fourier Transform Infrared (FTIR) spectra of DMP are shown in above figure

The spectrum band below 900 is often referred to as the "fingerprint" region in scientific literature. This region is used for identifying conformational changes and crystal regions inside the material being investigated. The infrared (IR) bands within the range of 782.82 – 704.14 cm^{-1} and 1023.95 – 891.95 cm^{-1} have been linked to the presence of carbohydrates in DMP. The spectrum areas ranging from 1660.52 to 1416.97 cm^{-1} and at 1255.91 cm^{-1} are associated with the amide I band, which primarily represents the stretching of the carbonyl (C=O) groups in proteins. The presence of a prominent absorption peak at a wavenumber of 1744.74 cm^{-1} indicates the occurrence of C=O stretching in milk lipids. The observed peak at 3278.66 cm^{-1} may be attributed to the stretching vibrations of the hydroxyl (OH) bond and the NH– bond in proteins present in donkey milk powder.



FTIR spectrum of donkey milk powder

Scanning Electron Microscopy (SEM) Analysis: The surface properties of a powder may be ascertained by the analysis of its scanning electron microscopy (SEM) data. The SEM pictures acquired for the DMP particles in this study indicated the presence of smaller vacuoles, some of which included minute particles that were entrapped on the surface. The major observation indicates a small inclination towards a spherical form with a smooth surface. The elemental content and distribution of DMP were examined using energy-dispersive X-ray spectroscopy (EDS).

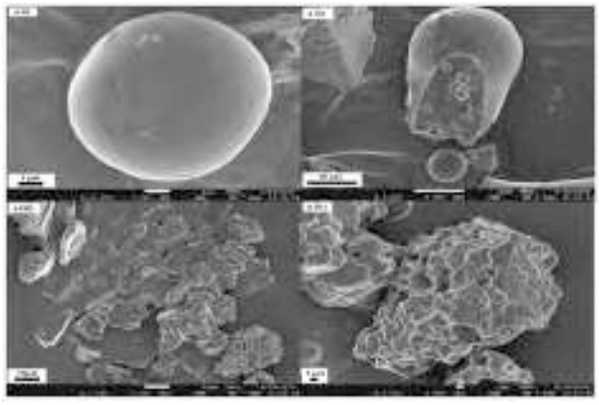


Figure (a) shows the scanning electron microscopy (SEM) micrograph of DMP at different magnifications. The EDS spectrum

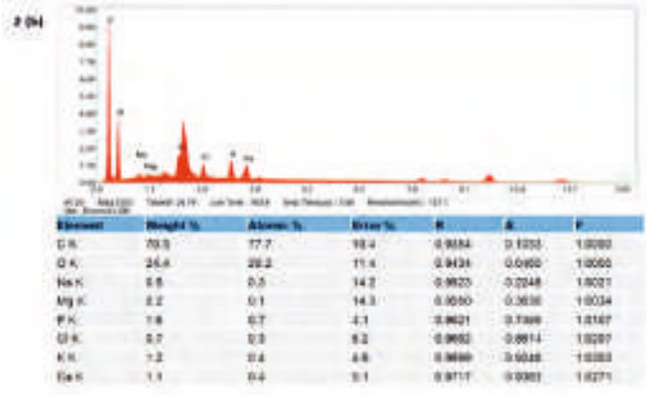


Figure (b) illustrates the existence of sodium, magnesium, phosphorus, chlorides, potassium, and calcium elements in different amounts, namely 0.5, 0.2, 1.6, 0.7, 1.2, and 1.1 (% weight), respectively. Additionally, the surface of DMP exhibits abundant levels of carbon and oxygen as compositional components.

Dielectric Constant and Electrical Conductivity : The dielectric constant of DMP had the greatest value, peaking at around 230 at a temperature of 90 °C. Moreover, it displayed comparable fluctuations with increasing frequency

across all temperatures. The values experienced a substantial decrease, ranging from 0.001 MHz to 1 MHz, and continued to decline as the dielectric constant increased. Eventually, a notable decline was seen in the vicinity of the 4 MHz frequency range. The observed trend persisted and exhibited additional growth till reaching a frequency of 6 MHz, as shown from the data. The frequency-dependent dielectric loss coefficients of WRP are graphically shown within a temperature range spanning from 25 °C to 90 °C. Similar to the dielectric constant, the dielectric loss factor of DMP exhibited an upward trend as the frequency increased during the early phase, followed by consistent oscillations until a significant rise in the loss factor of 0.75 was seen at around 4 MHz frequency range. The data indicates that there is no discernible trend in the dielectric constant of DMP as temperature increases. The loss factor saw a recovery and subsequently decreased, ultimately reaching a frequency of 6 MHz. The correlation between the rise in dielectric loss and elevated temperatures has been substantiated as evidence for energy dissipation, as reported by other resaerchers. The potential correlation between increased electric polarisation at elevated temperatures and the significant escalation of dielectric loss values with temperature may be seen. Numerous researchers have observed alterations in the dielectric characteristics of substances with respect to temperature and frequency. These changes could potentially be associated with the activity of water dipoles, enhanced mobility of bound water, stability of compositional elements, or diminished ionic conductivity.

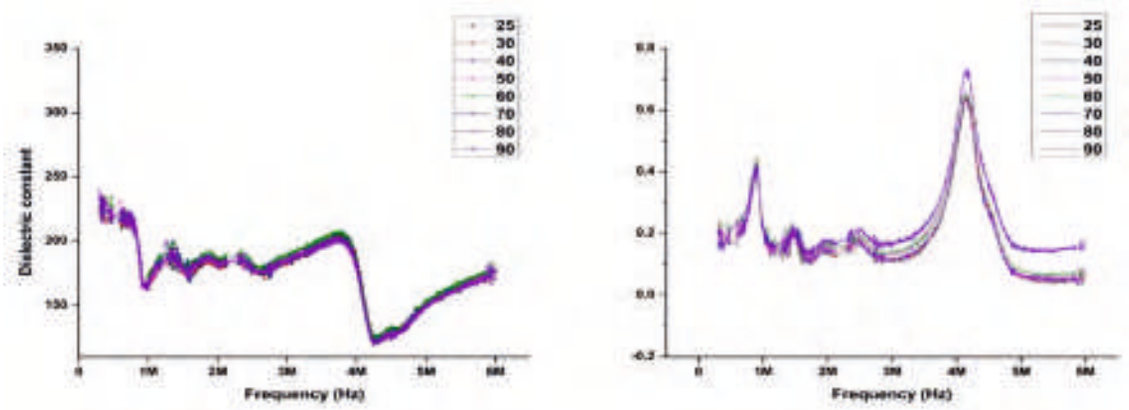


Figure displays the variation of the dielectric constant with frequency throughout the temperature range of 25 to 90 °C.

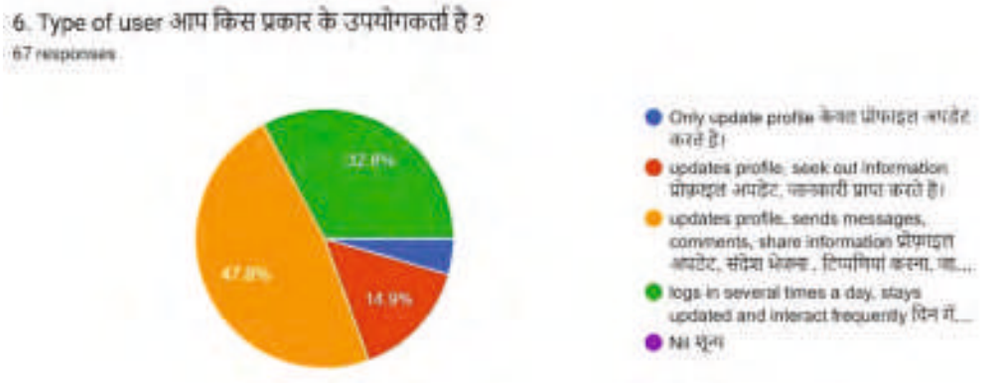
(Renu Garhwal, Anuradha Bhardwaj, Harish Kumar, Karnam Sangwan, Ankur Kumari, Bhavya, Varij Nayan, Yash Pal, Tarun Kumar Bhattacharya & B. N. Tripathi)

Dielectric Constant and Electrical Conductivity : The dielectric constant of DMP had the greatest value, peaking at around 230 at a temperature of 90 °C. Moreover, it displayed comparable fluctuations with increasing frequency across all temperatures. The values experienced a substantial decrease, ranging from 0.001 MHz to 1 MHz, and continued to decline as the dielectric constant increased. Eventually, a notable decline was seen in the vicinity of the 4 MHz frequency range. The observed trend persisted and exhibited additional growth till reaching a frequency of 6 MHz, as shown from the data. The frequency-dependent dielectric loss coefficients of WRP are graphically shown within a temperature range spanning from 25 °C to 90 °C. Similar to the dielectric constant, the dielectric loss factor of DMP exhibited an upward trend as the frequency increased during the early phase, followed by consistent oscillations until a significant rise in the loss factor of 0.75 was seen at around 4 MHz frequency range. The data indicates that there is no discernible trend in the dielectric constant of DMP as temperature increases. The loss factor saw a recovery and subsequently decreased, ultimately reaching a frequency of 6 MHz. The correlation between the rise in dielectric loss and elevated temperatures has been substantiated as evidence for energy dissipation, as reported by other resaerchers. The potential correlation between increased electric polarisation at elevated temperatures and the significant escalation of dielectric loss values with temperature may be seen. Numerous researchers have observed alterations in the dielectric characteristics of substances with respect to temperature and frequency. These changes could potentially be associated with the activity of water dipoles, enhanced mobility of bound water, stability of compositional elements, or diminished ionic conductivity.

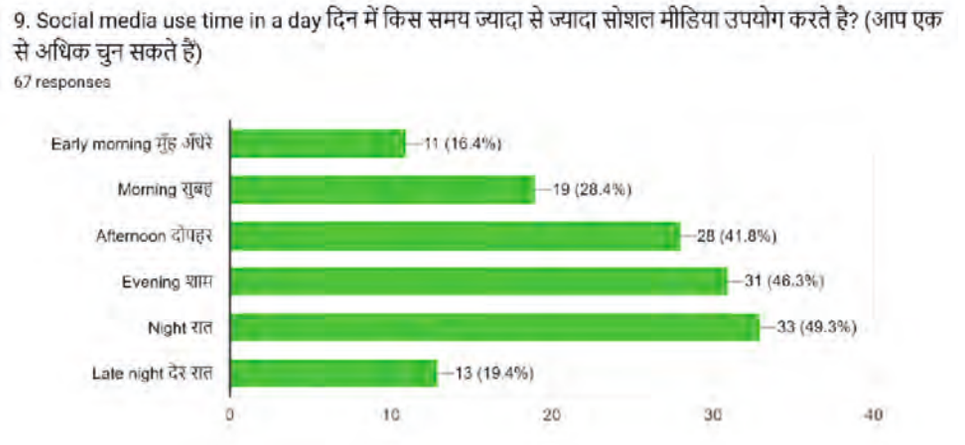
EXTENTION EDUCATION

Social media usage pattern among horse keepers and field veterinarians

The ever-increasing number of smartphone users in India (650 million in Dec, 2022) opened immense opportunities to connect instantly and without any borders. Information-sharing and communication through social media have enhanced relationships and collaborations among different stakeholders. To better exploit this new opportunity, a survey was conducted to study the social media usage patterns among horse keepers and field veterinarians of Haryana state. A bilingual online questionnaire (Hindi & English) was designed to obtain responses from horse keepers and veterinarians with 10 question items. A total of 32 horse keepers and 35 field veterinarians (N = 67) recorded their responses within the given time frame. All the respondents owned smartphones whereas laptops were owned by only 18% (n=12). WhatsApp topped the list (95.50%) followed by Facebook (76.10%) and YouTube (65.70%) in social media participation. Around 71.60% of the respondents reported that they are using social media for finding out news and events whereas 68.70% of the respondents are using social media for exchange of information. It was found that only 30% of the respondents are using social media for sharing their personal events. Another interesting finding of the study is that majority of the respondents are in advanced social media user level – level 3 (47.80%) and level 4 (32.80%) as depicted in Fig. 1. When enquired about the drawbacks in using social media, 35.80% revealed about poor internet connectivity followed by 24% expressing their concerns on lack of reliable information. It was also found that majority of the respondents are interested to use social media platforms during evening (46.30%) and night hours (49.30%) as shown in Fig. 2.



Findings showing the social media nature of respondents



Results of social media use in a day

(Ana Raj J, R. K. Dedar & Gururaj M)

NATIONAL CENTRE FOR VETERINARY TYPE CULTURES

Authentication and accessioning of viruses of animal origin

NCVTC virus repository is being strengthened with the addition of viruses from different geographical locations of the country through the deposition/collection of isolates and clinical samples from different animals and poultry. The primary objectives of the virus repository include the isolation & characterization of viruses of animal origin, accessioning, preservation & development of database of the isolates in the repository and the distribution of authenticated virus isolates / cell lines to stakeholders.

In this regard, thirty-six viral isolates were received as deposits from NCVTC Network units. The deposits included Avian infectious bronchitis (n=2), Fowlpox virus (n=1), Marek's disease virus (n=2), Inclusion body hepatitis virus (aka Fowl adenovirus) (n=1), African swine fever virus (n=1), Bovine Corona virus (n=3), Rous Sarcoma Virus (RAV-1), Duck enteritis virus (n=1), PPR virus (Mutant strain) (n=1), Bluetongue virus (n=15), Sheeppox virus (n=3), Orf virus (n=2) and Lumpy skin disease virus (n=2). The samples/isolates were processed for authentication & accession of the different viruses.

Four viruses were accessioned (VTCC AVA 381- 384), in the NCVTC repository which include Rous Sarcoma Virus (RAV-1), Duck enteritis virus (n=1), PPR virus (Mutant strain) (n=1) and African swine fever virus (n=1). The remaining viruses (received recently) are under processing for confirmation of their viability. Besides, the bulk production and preservation of 30 previously accessioned viruses (10 vials each) including NDV (n=11), fowl adenovirus (n=3), Infectious bronchitis virus (n=4), Bluetongue virus (n=4), Chicken astrovirus (n=2) and Fowlpox virus (n=2), Swinepox virus (n=1) and Infectious bursal disease virus (n=3) was also completed. Furthermore, 21 previously preserved viruses including NDV (8), SWPV (1), LSDV (1), BTV (6), IBDV (3) and IBV (2) were revived and checked for their viability which is an important activity of the virus repository.

(Sanjay Barua, Naveen Kumar & Riyesh, T)

Evaluation of the safety, immunogenicity and efficacy of a new live-attenuated lumpy skin disease vaccine in India

Lumpy skin disease (LSD) was reported for the first time in India in 2019 and since then, it has become endemic. Since a homologous (LSD-virus based) vaccine was not available in the country, goatpox virus (GPV)-based heterologous vaccine was authorized for mass immunization to induce protection against LSD in cattle. This study describes the evaluation of safety, immunogenicity and efficacy of a new live-attenuated LSD vaccine developed using an Indian field strain (LSDV/India/2019/Ranchi). The virus was attenuated by continuous passage (P=50) in Vero cells. The vaccine (50th LSDV passage in Vero cells, named as Lumpi-ProVac^{Ind}) did not induce any local or systemic reaction upon its experimental inoculation in calves (n=10). At day 30 post-vaccination (pv), the vaccinated animals were shown to develop antibody- and cell-mediated immune responses and exhibited complete protection upon virulent LSDV challenge. A minimum Neethling response (0.018% animals; 5 out of 26940 animals) of the vaccine was observed in the field trials conducted in 26940 animals. There was no significant reduction in the milk yield in lactating animals (n=10108), besides there was no abortion or any other reproductive disorder in the pregnant animals (n=2889) (Fig.) Sero-conversion was observed in 85.18% animals in the field by day 30 pv.

(a)

State wise number of animals vaccinated	
Jammu and Kashmir	20600
Rajasthan	4932
Haryana	249
Uttar Pradesh	751
Delhi	63
Punjab	345
Total	**26940**

(b)

Species/type of animal	Number of animals vaccinated
Cattle	26527
Buffaloes	413
Pregnant Cattle/buffaloes	2889
Lactating cattle/buffaloes	10108

(c)

Parameter	Observations (Number of animals examined)
Swelling at site of injection	Rare (5/26940) (0.018%)
Skin nodules at the site of inoculation	Nil (n=26940)
Generalized skin nodules	Nil (n=26940)
Viremia	Nil (n=62)
Fever	Nil (n=53)
Vaccine virus excretion (Nasal/fecal /ocular routes)	Nil (n=62)
Excretion of vaccine virus in milk	Nil (n=24)
Excretion of vaccine virus in semen	Nil (n=8)
Abortion in pregnant animals	Nil (n=2889)
Natural disease after completion of 3 weeks of vaccination	14 Animals including 9 calves & 5 adults (n=26940)

n : Number of animals examined; Nil: No reaction observed or absent

Fig. Safety and efficacy of Lumpi-ProVac^{Ind} in field animals. A total of 26940 animals across six Indian states **(a)** comprising of 26527 cattle, 413 buffaloes (2889 pregnant cattle/buffaloes and 10108 lactating buffaloes) **(b)** were included in the study. All the animals were injected with 1 ml of Lumpi-ProVac^{Ind} (containing $10^{3.5}TCID_{50}$/dose) by subcutaneous route and monitored for swelling/skin nodules at the site of injection, generalized skin nodules, abortions in pregnant animals and efficacy of the vaccine **(c).**

(Naveen Kumar, Sanjay Barua, Ram Kumar, Amit Kumar, Sukdeb Nandi, Karam Pal Singh, Yash Pal, Triveni Dutt, & B. N. Tripathi)

Evaluation of the immune responses in buffaloes vaccinated with a live-attenuated lumpy skin disease vaccine (Lumpi-ProVac^{Ind})

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. However, 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 UVinactivated 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 (Fig.). In conclusion, the buffaloes also mount a potent antibody- and cell-mediated immune response following vaccination with Lumpi-ProVac^{Ind}.

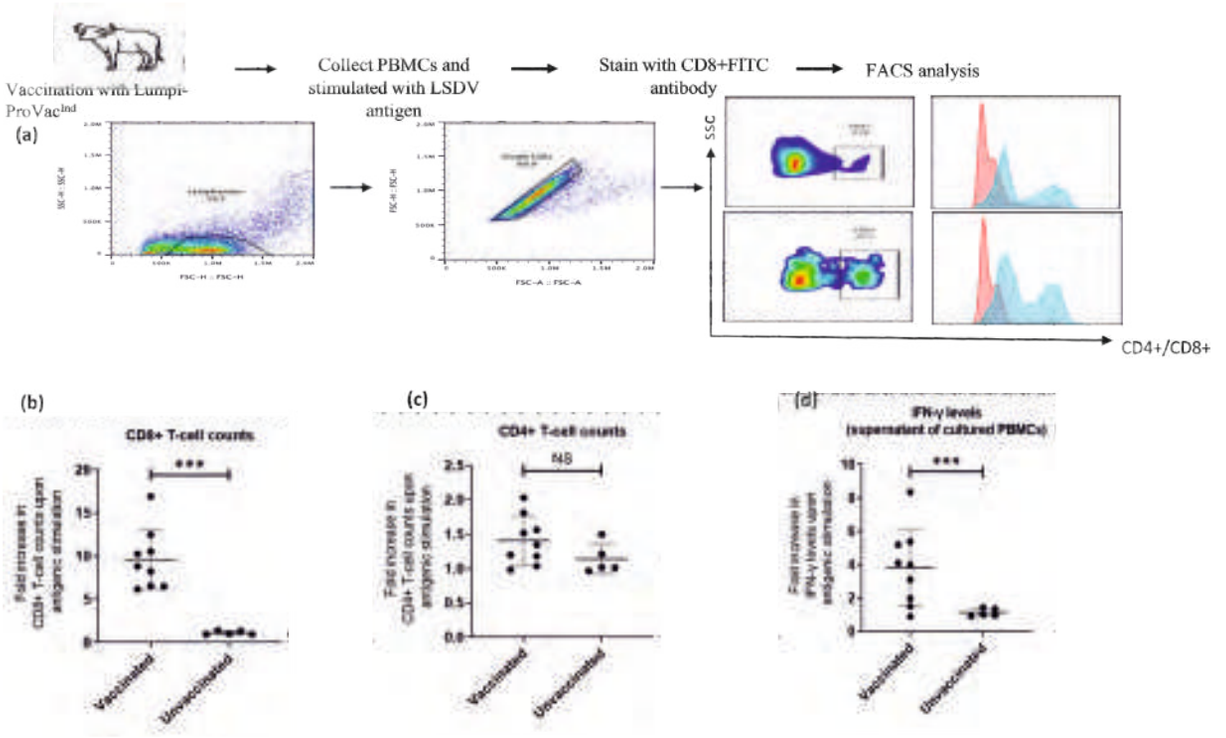


Fig. Cell-mediated immune response in buffaloes vaccinated with Lumpi-ProVac^{Ind}. Buffaloes (n=9) were vaccinated with live-attenuated LSD vaccine. At 4 months following vaccination, equal numbers of PBMCs from vaccinated (n=9) and unvaccinated control (n=5) animals were treated with UV-inactivated LSDV antigen and incubated for 36 h at 37°C. The cells were then stained with the respective antibodies. Flow Cytometry was performed using a CytoFLEX Flow Cytometer, and the results were analyzed using FlowJo™10 software. **(a)** ***The gating strategy for FACS analysis***. Total lymphocytes were first gated in a forward scatter (FSC)/side scatter (SSC) plot, and then a single-cell population was selected in an FSC-A/FSC-H plot. Single cells were subsequently gated for CD4 or CD8 expression. ***(b) CD8+ T cell counts.*** Relative fold-change in CD8+ counts upon antigenic stimulation. ***(c) CD4+ T cell counts.*** Relative fold change in $CD4^+$ counts upon antigenic stimulation. ***(d) IFN-γ*** response. PBMCs from vaccinated and unvaccinated cattle were either unstimulated or stimulated with LSDV antigen for 24 h. The cell culture supernatants were subjected for qunatitation of *IFN-γ*. Values are means ± SD. Error bars indicate SD. Pair-wise statistical comparisons were performed using Student's t test (*** = P<0.001). NS indicates no statistically significant difference.

(Shweta Dhanda, Ram Kumar, Sanjay Barua, Deepak Kumar Sharma & Naveen Kumar)

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

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 **(Fig.).** 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.

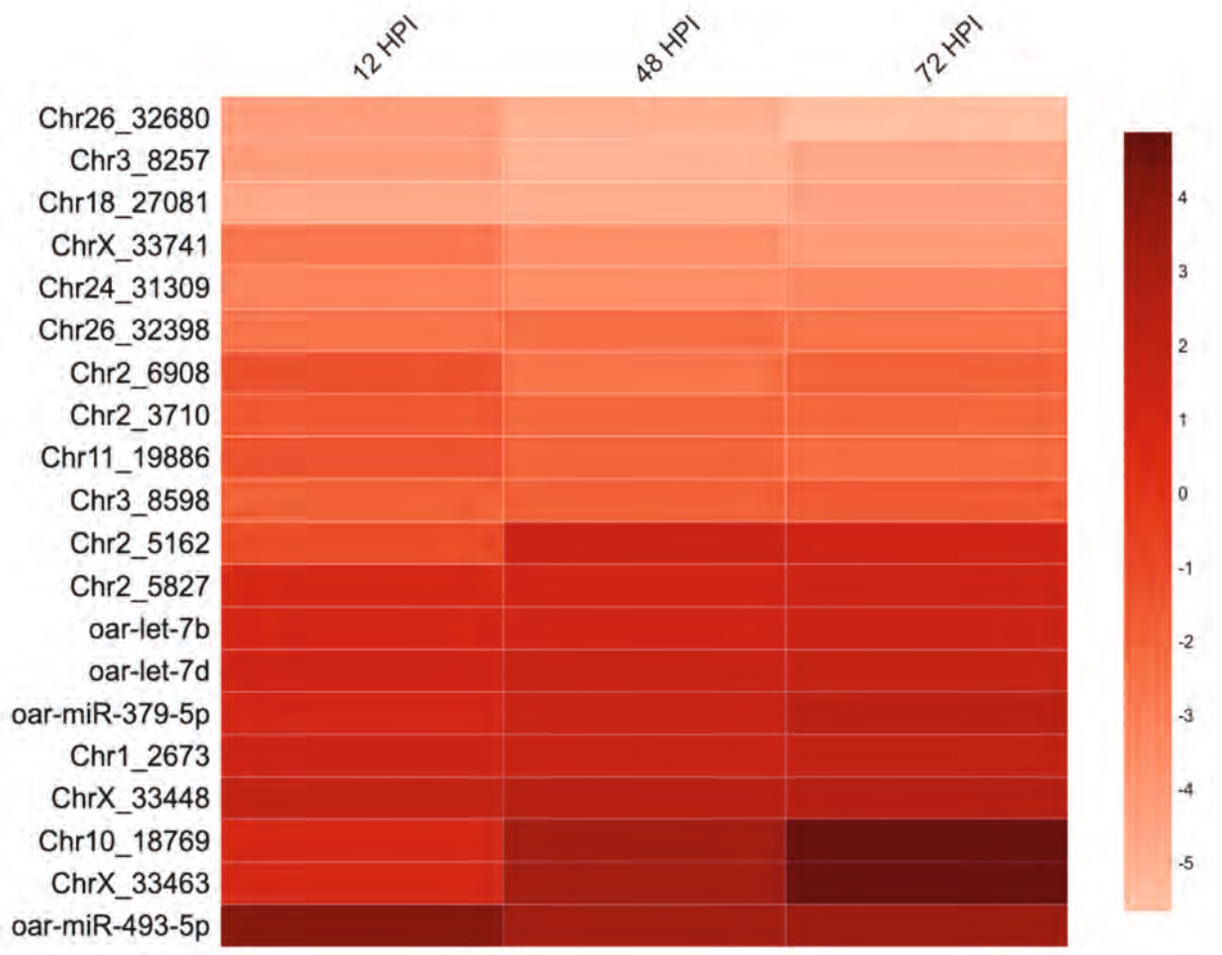


Fig. Heatmap. The hierarchical cluster analysis of 10 top commonly dysregulated miRNAs in mock-infected and LSDV infected LT cells at different times post-infection is shown. The colour code on the heatmap is linear, where dark red and faint red represent upregulated and downregulated miRNA expression levels, respectively. Each row represents a miRNA, and each column represents a sample. Heatmap plots were generated from the made4 R package based on log2 FC.

(Sakshi Pandita, Sanjay Barua & Naveen Kumar)

A novel HRM-based gap-qRT-PCR for identification and quantitation of the vaccine and field strain(s) of lumpy skin disease virus

Lumpy skin disease (LSD) has become the most important animal health problem in India due to high morbidity, mortality and production losses. A homologous live-attenuated LSD vaccine (LumpiProVac^{Ind}) was recently developed by using a local LSD virus (LSDV) strain (LSDV/2019/India/Ranchi) in India which is likely to replace the existing practice of vaccinating cattle with goatpox vaccine. It is essential to differentiate the vaccine and field strains, if a live-attenuated vaccine has been used for control and eradication of the disease, particularly in endemic areas. As compared to the prevailing vaccine and field/virulent strains, the Indian vaccine strain (Lumpi-ProVac^{Ind}) has a unique deletion of 801 nucleotides in its inverted terminal repeat (ITR) region. We exploited this unique feature and developed a novel high resolution melting-based gap quantitative real-time PCR (HRM-gap-qRT-PCR) for rapid identification and quantitation of the vaccine and field strain(s) of LSDV (Fig.).

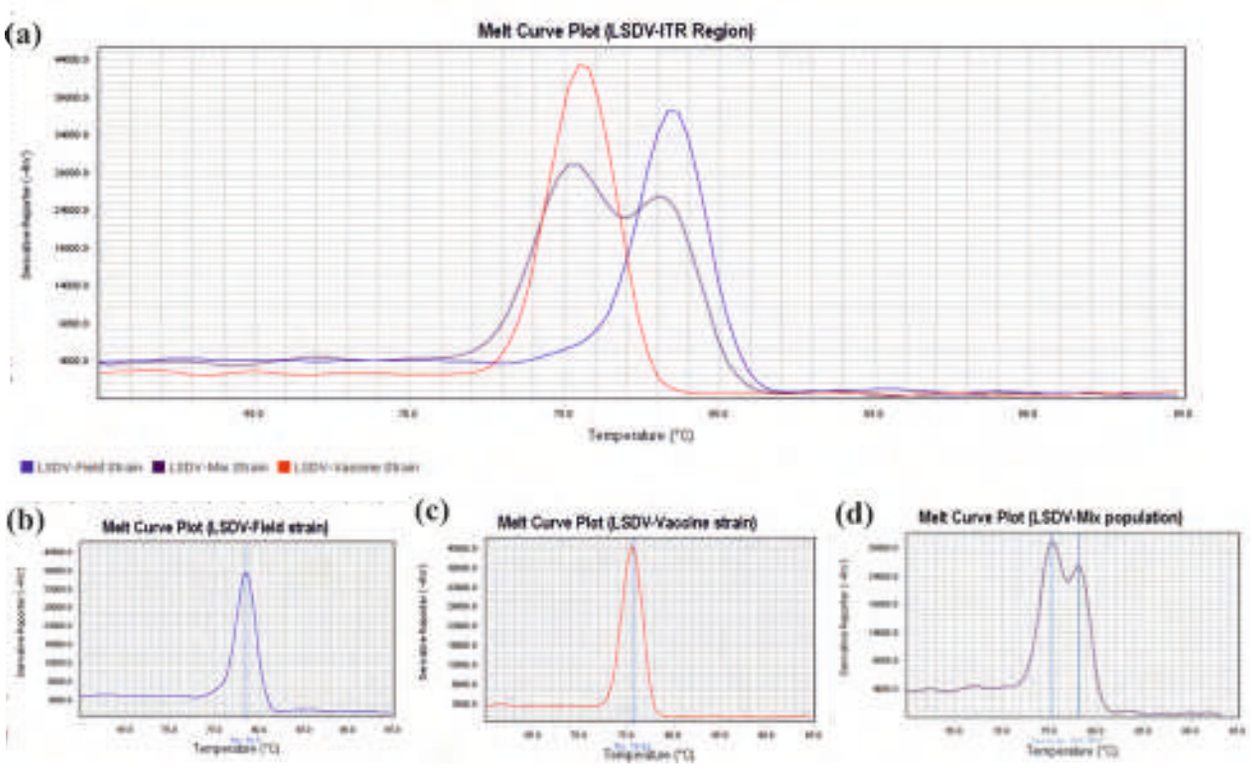


Fig. High resolution melt curve analysis of ITR region of LSDV: The ITR region of LSDV vaccine and field strains were amplified. The Tm of PCR-amplicons were examined by change in their relative fluorescence intensity over a temperature range of 60–95°C in 0.2°C increments by using QuantStudio™ Design and Analysis Software v1.4.2. **(a and b)** Melting curve of LSDV field strain (Blue line; Tm=78.5°C).(**a and c**) Melting curve of LSDV vaccine strain (Red line; Tm=75.62°C). (**a and d)** The mixed (vaccine plus field strain) population of LSDV produces two amplicons with Tm of 75.29°C and 78.12°C.

(Ram Kumar, Sanjay Barua & Naveen Kumar)

Identification of miR-29a as a novel biomarker for lumpy skin disease virus exposure in cattle

miRNAs have been implicated in the regulation of maturation, proliferation, differentiation, and activation of immune cells. In this study, we demonstrated that miR-29a antagonizes IFN-γ production in response to lumpy skin disease virus (LSDV) exposure in cattle (Fig.). Stimulation of sensitized peripheral blood mononuclear cells (PBMCs) exhibited lower levels of miR-29a, concomitant with a potent cell-mediated immune response (CMI), characterized by an increase in LSDV-specific CD8+ T cell counts and enhanced levels of IFN-γ, which eventually facilitated virus clearance. Furthermore, compared to sensitized crossbred cattle, PBMCs from sensitized Rathi (a native Indian breed) animals exhibited lower levels of miR-29a along with an increase in CD8+ T cell counts and enhanced levels of IFN-γ. Finally, we analyzed that a ≥60% decrease in miR-29a expression levels in the PBMCs of sensitized cattle correlated with a potent CMI response. In conclusion, miR-29a serves as a novel biomarker for predicting the functionality of CMI responses in sensitized cattle.

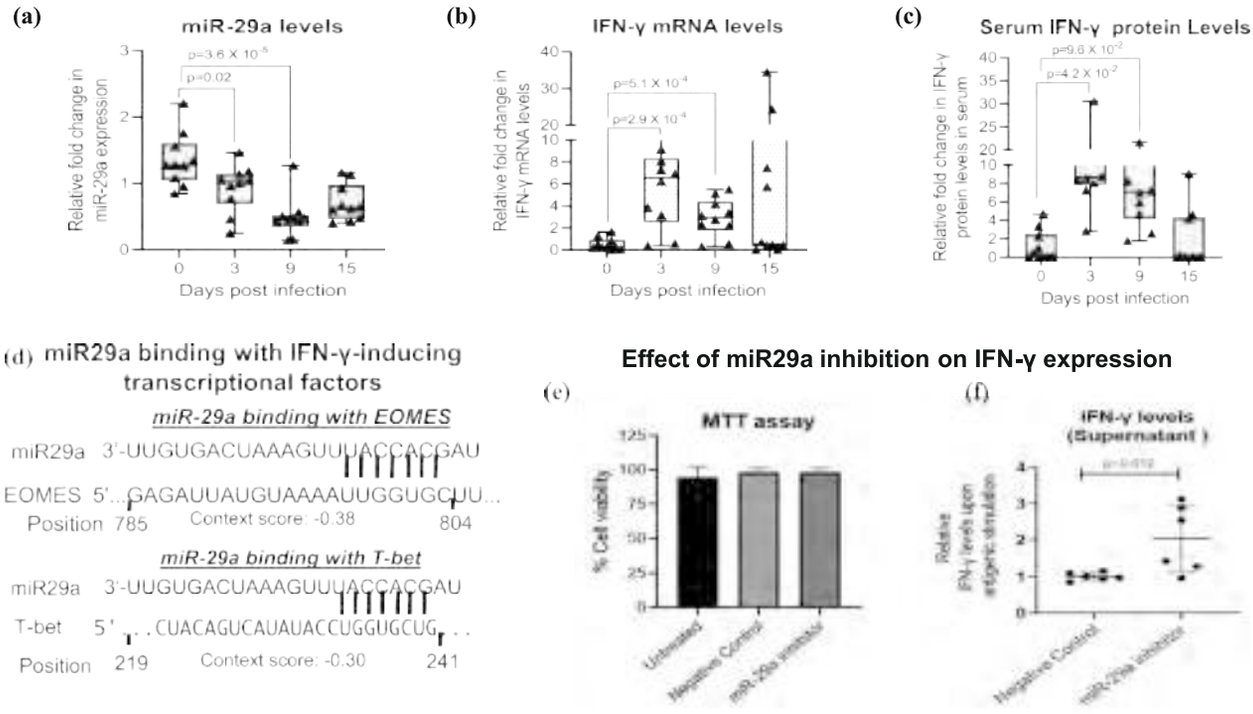


Fig. miR-29a negatively regulates IFN-γ response in LSDV-infected cattle. Blood/serum samples from cattle were collected at the indicated times post-LSDV infection. **(a)** ***miR-29a.*** RNA from PBMCs was subjected to poly(A) tailing, cDNA synthesis, and qRT-PCR using a universal adaptor primer and a miR-29a-specific forward primer **(Table)**. The miRNA expression levels were calculated using the $2^{-\Delta\Delta}$Ct method and normalized to the respective U6 (endogenous control) expression levels. **(b)** ***IFN-γ*** in PBMCs. RNA from PBMCs was subjected to cDNA synthesis, and *IFN-γ* levels were determined by qRT-PCR. **(c)** ***IFN-γ*** in serum. The IFN-γ levels in serum were determined by the Bovine IFN-γ-ELISA kit. Differences in the levels of miR-29a/IFN-γ at various days post-infection were compared using Student's t-test. Values are means ± SD. The p value indicates the level of statistically significant difference. ***(d) miR29a binds to IFN-γ***-inducing transcriptional factors EOMES and T-bet. The miR29a binding sites on IFN-γ-inducing transcriptional factors EOMES and T-bet were identified by pairing the miRNA seed region and complementary sites within target mRNAs using Target Scan. The context score and position of miR29a binding to EOMES and T-bet mRNA are shown. **(e)** ***Evaluation the cytotocity of miR-92a inhibitor.*** The PBMCs, in triplicates, were transfected with bovine miR-29a inhibitor or negative control and cultured for 72 h. The cytotoxicity was determined by the MTT assay. The % cell viability was measured by comparing it with the untransfected control. ***(f) Effect of miR29a inhibition on IFN-γ*** expression. The PBMCs from sensitized cattle were transfected with bovine miR-29a inhibitor or a negative control. After incubation for 24 h, cells were either unstimulated or stimulated with UV-inactivated LSDV antigen. The level of IFN-γ released in the supernatant at 36 h post-stimulation was measured by the Bovine IFN-γ-ELISA kit. The IFN-γ levels were normalized with the respective unstimulated controls. The relative levels of IFN-γ secreted in miR-29a inhibitor-treated- and negative control-treated PBMCs are shown. Values are means ± SD. The "p" value indicates the level of a statistically significant difference.

(Ram Kumar, Himanshu Kamboj, R.K. Dedar, Sanjay Barua, Bhupendra N. Tripathi, Shalini Sharma & Naveen Kumar)

miR-29a determines breed susceptibility to lumpy skin disease virus infection in cattle

One interesting observation made during the 2022 outbreak of LSD in India was that indigenous cattle, particularly the Rathi breed of cattle showed lower morbidity (13.75%) and case fatality rate (27.27%) as compared to the crossbred cattle (87.77% morbidity and 69.51% case fatality). As compared to the sensitized crossbred cattle, PBMCs from sensitized Rathi (a native Indian breed) animals exhibited lower levels of miR-29a, along with an increase in CD8+ T cell counts and enhanced levels of IFN-γ (Fig.). This suggests that Rathi cattle mount a more potent CMI response against LSDV as compared to the crossbred cattle.

(a) **Herd strength, morbidity and case fatality rate in Crossbred, Rathi and Sahiwal cattle**

S.N.	Breed	Location	Herd strength	Morbidity rate (%)	Case fatality rate (%)
1	Crossbred (HF)	Shri Radha Krishan Dairy & Durga colony, Nohar	139	87.77	69.51
2	Rathi	Livestock Research Station, Nohar	240	13.75	27.27
3	Sahiwal	Dadhich Cattle farm, Nohar	72	33.33	29.17

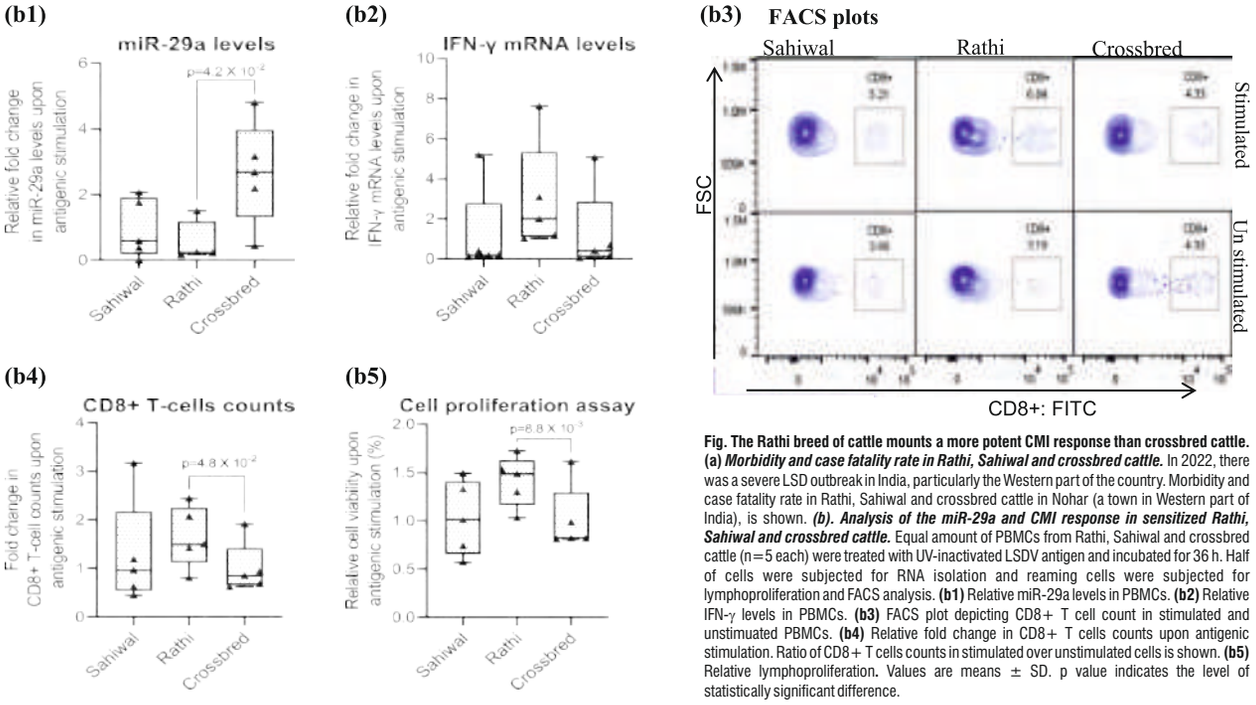


Fig. The Rathi breed of cattle mounts a more potent CMI response than crossbred cattle. **(a)** ***Morbidity and case fatality rate in Rathi, Sahiwal and crossbred cattle.*** In 2022, there was a severe LSD outbreak in India, particularly the Western part of the country. Morbidity and case fatality rate in Rathi, Sahiwal and crossbred cattle in Nohar (a town in Western part of India), is shown. ***(b). Analysis of the miR-29a and CMI response in sensitized Rathi, Sahiwal and crossbred cattle.*** Equal amount of PBMCs from Rathi, Sahiwal and crossbred cattle (n=5 each) were treated with UV-inactivated LSDV antigen and incubated for 36 h. Half of cells were subjected for RNA isolation and reaming cells were subjected for lymphoproliferation and FACS analysis. **(b1)** Relative miR-29a levels in PBMCs. **(b2)** Relative IFN-γ levels in PBMCs. **(b3)** FACS plot depicting CD8+ T cell count in stimulated and unstimuated PBMCs. **(b4)** Relative fold change in CD8+ T cells counts upon antigenic stimulation. Ratio of CD8+ T cells counts in stimulated over unstimulated cells is shown. **(b5)** Relative lymphoproliferation. Values are means ± SD. p value indicates the level of statistically significant difference.

(Ram Kumar, Himanshu Kamboj, R.K. Dedar, Sanjay Barua, Bhupendra N. Tripathi, Shalini Sharma & Naveen Kumar)

Evaluation of the duration of the immunity of Lumpi-ProVac^{Ind}

The average anti-LSDV antibody titres (log2) at 1 month (m)-, 3 m- and at 6 m-post-vaccination were 5.7, 5.8 and 5.9, respectively. Thereafter, it started declining, as the average antibody titres (log2) were 5.2 and 3.85 respectively, at 9 m- and 12 m-post-vaccination. After attaining the peak titres (1-3 m), there were no significant increase in the average antibody titres, suggesting animals were not exposed to natural LSDV infection during the course of experiment and, therefore, the antibody titers truly reflected the end result of vaccination. For estimation of cell mediated immune response, at 12 m following vaccination of Lumpi-ProVac^{Ind}, the blood was collected and PBMCs separated. FACS analysis of PMBCs was performed upon stimulation with UV-inactivated LSDV (antigen). The CD8+ T cell counts were significantly higher (~2.6-fold) in vaccinated cattle as compared to unvaccinated animals (seronegative calves). Further, the IFN-γ level in the supernatant of stimulated PBMCs was also significantly higher in vaccinated as compared to the unvaccinated controls. This suggests that vaccinated animals mount a potent cell-mediated immune response even at 12 m following vaccination. Two of the animals that did not show detectable amount of anti-LSDV antibodies at 12 m, also mounted cell-mediated immune response. Taken together, the data seems to suggest that an effective immune response following Lumpi-ProVac^{Ind} persists at least for a period of 1 year. Blood samples were also collected from the cattle that were vaccinated with goatpox vaccine. At six months after vaccination, there was no significant increase in the CD8+ T cell counts following stimulation of the PBMCs with LSDV antigen (Fig.). This suggests that the LSD-specific cell-mediated immune response persists for less than 6 months in cattle vaccinated with heterologous vaccine.

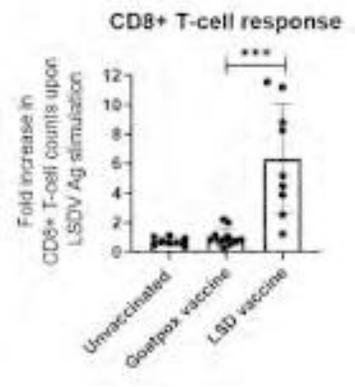


Fig. Cell-mediated immune response following exposure to Capripoxviruses. Cattle were either unvaccinated or vaccinated with the live-attenuated LSD vaccine (Lumpi-ProVac^{Ind}) or commercially available goatpox vaccine At 6 months following vaccination, PBMCs were stimulated with UV-inactivated LSDV antigen. The cells were then stained with anti-CD8 antibody. Flow Cytometry was performed using a CytoFLEX Flow Cytometer, and the results were analyzed using FlowJo™10 software. Relative fold-change in CD8+ counts upon antigenic stimulation is shown. Values are means ± SD. Error bars indicate SD. Pair-wise statistical comparisons were performed using Student's t test (*** = P<0.001).

(Ram Kumar, Himanshu Kamboj, R.K. Dedar, Sanjay Barua, Bhupendra N. Tripathi, Shalini Sharma & Naveen Kumar)

Hesperetin blocks poxvirus replication by competitively inhibiting binding of the 5' cap of viral mRNA with eIF4E

In this study, we demonstrated the antiviral efficacy of hesperetin against multiple poxviruses, including buffalopox virus (BPXV), vaccinia virus (VACV), and lumpy skin disease virus (LSDV). The time-of-addition and virus step-specific assays indicated that hesperetin reduces the levels of viral DNA, mRNA and proteins in the target cells. Further, by immunoprecipitation (IP) of the viral RNA from BPXV infected Vero cells and a cell-free RNA-IP assay, we demonstrated that hesperetininduced reduction in BPXV protein synthesis is also consistent with diminished interaction between eukaryotic translation initiation factor eIF4E and the 5' cap of viral mRNA (Fig.). Molecular docking and MD simulation studies were also consistent with the binding of hesperetin to the cap-binding pocket of eIF4E, adopting a conformation similar to m7GTP binding. Furthermore, in a BPXV egg infection model, hesperetin was shown to suppress the development of pock lesions on the chorioallantoic membrane and associated mortality in the chicken embryos. Most importantly, longterm culture of BPXV in the presence of hesperetin did not induce the generation of drug-resistant viral mutants. In conclusion, we for the first time demonstrated the antiviral activity of hesperetin against poxviruses, besides providing novel mechanistic insights into its antiviral action.

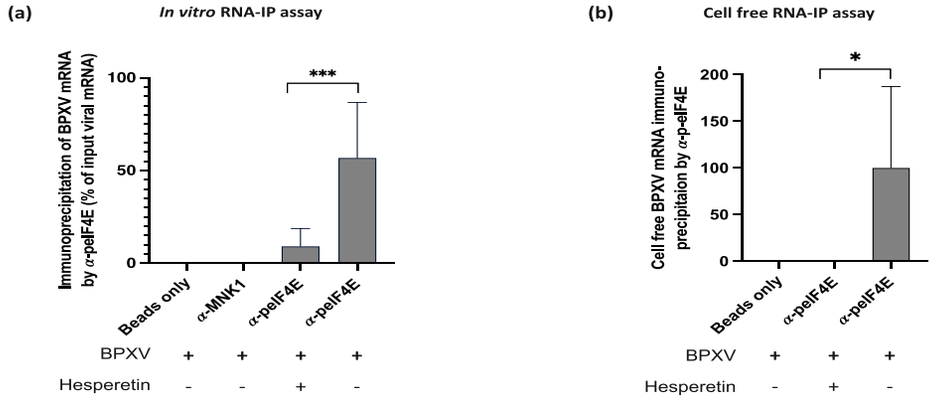


Fig. RNA immunoprecipitation (RNA-IP) assay. ***(a) RNA-IP assay.*** Vero cells, in triplicates, were infected with BPXV at an MOI of 5, followed by washing with PBS and addition of serum-free DMEM supplemented with hesperetin or vehicle control. At 16 hpi, cell lysates were prepared as described in method section and incubated with α-p-eIF4E (reactive antibody), α-MNK1 (non reactive antibody) or equivalent volume of IP buffer (Beads control). This was followed by incubation with a Protein A Sepharose® slurry. The beads were washed with IP buffer, and cross-linking was reversed using Proteinase K. The reaction mixture was centrifuged and the resulting supernatant was subjected to RNA isolation, cDNA preparation, and quantitation of BPXV "*M*" gene by qRT-PCR. The % interaction of BPXV mRNA with α-p- eIF4E was calculated by normalizing with input RNA. **(b) *Cell free RNA-IP assay.*** Uninfected Vero cells were lysed using IP buffer and incubated with a Protein A Sepharose® beads bound with p-eIF4E antibody. The p-eIF4E/Sepharose® beads/α-p-eIF4E complex was then separate by centrifugation. In parallel, Vero cells were infected with BPXV and, at 12 hpi RNA was isolated and incubated with immunoprecipitated p-eIF4E/Sepharose® beads/α-p-eIF4E complex in the presence and absence of hesperetin. The beads were washed with IP buffer and mRNA associated with p-eIF4E/Sepharose® beads/α-p-eIF4E complex was isolated by Triazol and quantified for BPXV *"M"* gene by qRT-PCR. Values are representative of three independent separate experiments. Error bars indicate SD. Pair-wise statistical comparisons were performed using the Student's t test (*=P<0.1, *** = P<0.001).

(Assim Verma, Sanjay Barua & Naveen Kumar)

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

Emerging evidence highlights 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 the 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 (Fig.). The transcriptome analysis revealed a complex interplay among various epigenetic players (methylase and 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 subgenomic 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.

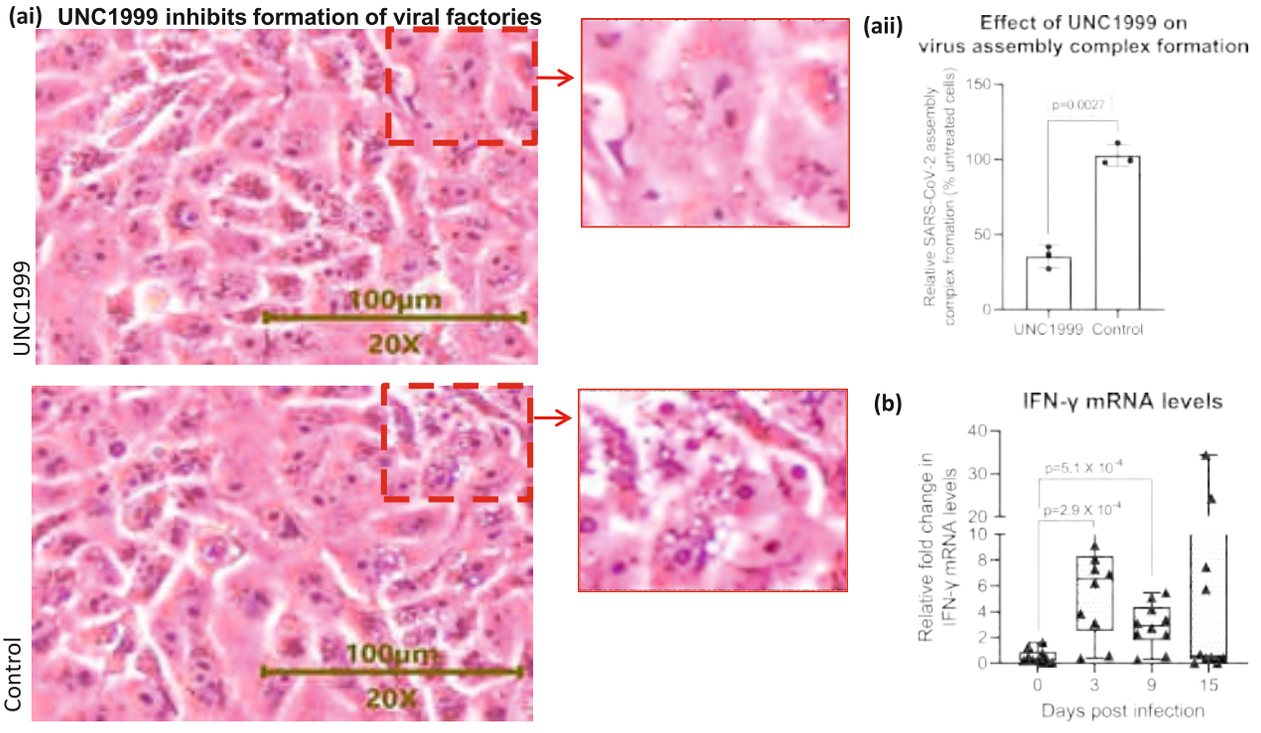


Fig. Effect of UNC1999 on the formation of viral factories. ***(a) UNC1999 inhibits the formation of viral factories.*** Vero cells were grown in chamber slides and infected with SARS-CoV-2 at an MOI of 5 for 1 h in the presence of either 1 µg/ml of UNC1999 or 0.05% DMSO. The cells were subjected to live imaging at 8 hpi. The white colorless dots in the virus-infected cell indicated the viral assembly complex/factories ***(a-i)***. The histogram shows the relative reduction in the number of viral assembly complexes after UNC1999 treatment ***(a-ii)***. ***(b) Effect of UNC1999 on RNP complex assembly.*** Vero cells were infected with SARS-CoV-2 at an MOI of 5, followed by washing with PBS and the addition of fresh MEM. UNC1999 (1 µg/ml) or equivalent volume of vehicle control were applied at 4 hpi. At 10 hpi, cell lysates were prepared and incubated with α- SARS-CoV-2 "N" to immunoprecipitate the RNA. The amount of SARS-CoV-2 RNA in the immunoprecipitate was quantified by qRT-PCR and expressed as % of the input viral RNA. Values are means ± SD and are representative of the results of at least three independent experiments. The "*p*" values indicate the level of a statistically significant difference.

(Ram Kumar, Sanjay Barua & Naveen Kumar)

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

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 (SARSCoV-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 (Fig.) 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-CoV2 replication by p38 MAPK.

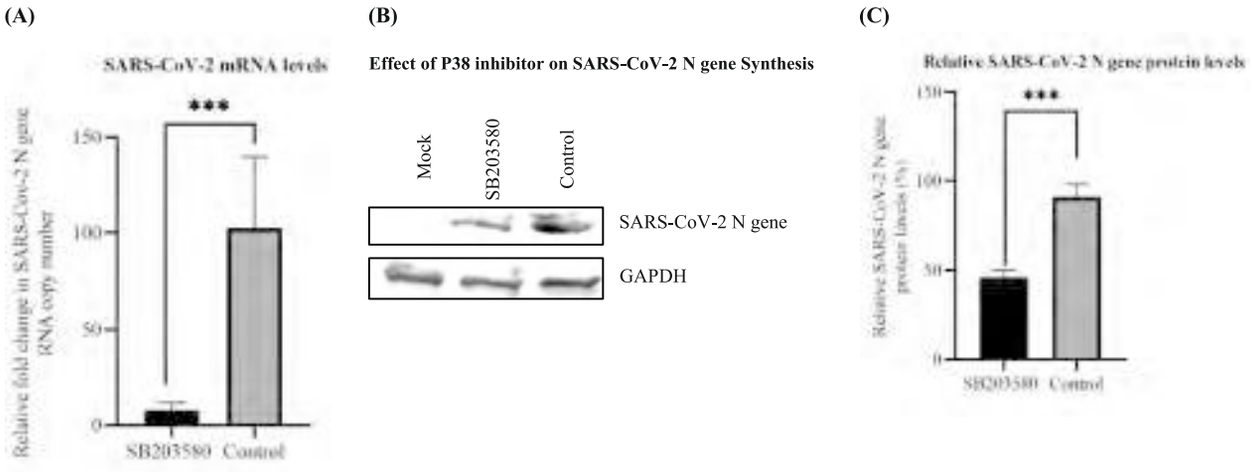


Fig. Effect of SB203580 on levels of viral RNA and protein. ***(A) RNA level.*** Confluent monolayers of Vero cells, in triplicates, were infected with SARSCoV-2 for 1 h at MOI 5. SB203580 was added at 3 hpi and cells were harvested at 12 hpi to determine the levels of SARS-CoV-2 RNA by qRT-PCR. Threshold cycle (Ct) values were analysed to determine relative fold-change in copy numbers of total RNA and mRNA. Values are means ± SD and representative of the result of at least 3 independent experiments. Pair-wise statistical comparisons were performed using Student's t test (***= P <0.001). ***(B) Protein levels.*** Confluent monolayers of Vero cells were infected with SARS-CoV-2 at an MOI of 5. The inhibitor or DMSO was applied at 3 hpi and the cells were scrapped at 24 hpi to examine the levels of viral proteins by immunoblotting. ***(C) Quantitation of protein levels.*** The blots were quantified by densitometry (ImageJ) and the data are presented as mean with SD. The levels of viral proteins (upper panel), along with housekeeping GAPDH protein (lower panels) is shown. Pair-wise statistical comparisons were performed using Student's t-test. ***=P<0.001. Values are means ±SD and representative of the result of at least tree-independent experiments.

(Priyasi Mittal, Sanjay Barua, B.R. Gulati & Naveen Kumar)

Surveillance of SARS-CoV-2 in waste water in Hisar (India) during 2022-2023

The waste-water surveillance of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) may provide information on the potential transmission of COVID-19 infection when mass clinical testing declines. In the present study, a total of 1110 wastewater samples were collected from Hisar, Haryana (India), from August 2022 to June 2023. The SARS-CoV-2 detection in the waste water correlated with clinical cases of COVID-19 in Hisar, besides facilitating the tracking of the ongoing mutations in the viral genome. The study suggests that waste water based epidemiology can potentially complement classical testing of COVID-19 (Fig.) and may serve as an early warning system to prevent potential disease transmission and consequential outbreaks.

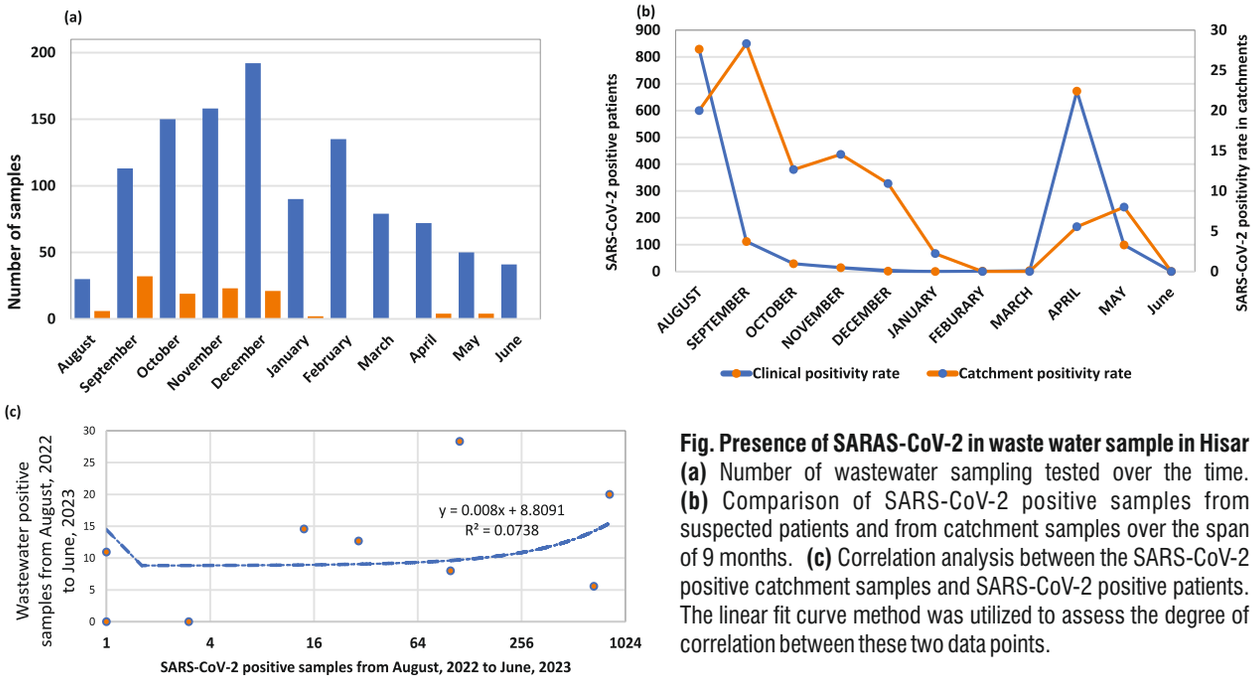


Fig. Presence of SARAS-CoV-2 in waste water sample in Hisar
(a) Number of wastewater sampling tested over the time. **(b)** Comparison of SARS-CoV-2 positive samples from suspected patients and from catchment samples over the span of 9 months. **(c)** Correlation analysis between the SARS-CoV-2 positive catchment samples and SARS-CoV-2 positive patients. The linear fit curve method was utilized to assess the degree of correlation between these two data points.

(Assim Verma, Geetanjali Yadav, Nikita Bishnoi, Ram Kumar, Sanjay Barua & Naveen Kumar)

Assessment of rabies prevalence and identification of a novel sub-genotype AL1b in domestic animals across Haryana, India

Rabies, a deadly zoonotic virus, presents substantial challenges in India, particularly in resourcelimited regions. Limited testing facilities in Haryana restrict rabies information on the prevalence of rabies in the state. Hence, surveillance commenced post-establishment of a diagnostic facility in Hisar, aided by the National Centre for Disease Control, New Delhi. The objectives of this one health study were to assess the prevalence of rabies among domestic animals in Haryana and to characterize the virus. This study will help to fill crucial knowledge gap and to bolster rabies control programme in the state. We collected biological samples included saliva and brain tissue from 50 animals (buffaloes, cows, and dogs) in Haryana, India, between January 2023 and March 2024. The samples were tested using TaqMan RT-qPCR, conventional PCR, and DFAT. Sequencing and phylogenetic analysis were performed for virus characterization. Out of 50 animals, 16 were positive for the rabies virus, which included cattle (n=2), buffaloes (n=4), and dogs (n=10). Sequencing of nine PCR products revealed nucleotide identities ranging from 95.09% to 99.39% and amino acid identities ranging from 95.71% to 99.39%. Phylogenetic analysis classified RABV into two distinct subclades of AL1, namely AL1a and AL1b. Our study showed a comprehensive update on RABV prevalence and unveiled two distinct sub-lineages circulating in Haryana. The discovery of a new AL1b subclade highlights rabies diversity. Interestingly, we have found that forty percent of positive samples were of non-canine origin, of which over 50% were from animals without a dog bite history. Thus, it emphasizes the testing of all animals with encephalitis to rule out rabies (May be taken from conclusion). Underreporting suggests a need for vigilance to achieve zero human rabies deaths by 2030. Furthermore, our study exemplifies the principles of the One Health approach, emphasizing the importance of cooperation between veterinary and medical agencies in combating zoonotic diseases.

(Riyesh T, Shanmugasundaram K, H. Singha, Naveen Kumar, Sanjay Barua & T.K. Bhattacharya)

Identification and isolation of bovine coronavirus from respiratory and enteric infections in cattle

Respiratory infection in bovine is a complex disease caused by spectrum of pathogens. Among them, several viruses cause severe respiratory infections leading to huge economic losses in dairy industry. The bovine coronaviruses (BCoVs) are of the important respiratory pathogens which causes severe respiratory and enteric infections in calf and adult animals. During investigation of respiratory illness and diarrhea cases in cattle in Haryana and Rajasthan, the biological samples (nasal swabs and faecal samples) were collected from infected animals and processed for molecular detection of viral pathogens. The total RNA was isolated from the biological samples and subjected to amplification of conserved region of N gene of bovine coronavirus by RT-PCR. A total of 15 samples showed positive amplification for BCoV (Fig.). Subsequently, the positive samples were processed for isolation of BCoV in NLBK cell line. The BCoV-specific CPE was observed in cell culture at 48hrs after infection. The BCoVs were further passaged upto four times in NLBK cells and identity of the virus was confirmed by RT-PCR.

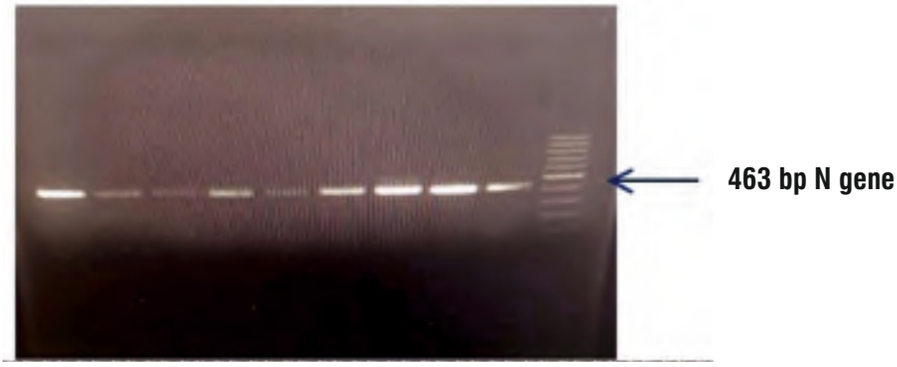


Fig. Detection of BCoV in clinical samples by RT-PCR.

(B.C. Bera, Taruna Anand & Nitin Virmani)

Isothermal "Recombinase Polymerase Amplification" (RPA) coupled with CRISPR Technology and LFA based assay for point-of-care detection of Porcine circovirus 3 (PCV3) in pigs

The Porcine circovirus 3 (PCV3) is a recently identified emerging virus causing reproductive failure in pigs leading to huge economic losses in swine industry. To overcome such important disease conditions, there is need to develop new generation diagnostics for early, rapid, and specific diagnosis of the disease. In the present study, the isothermal recombinase polymerase amplification (RPA) coupled with CRISPR based LFA assay has been developed for point-of-care detection of PCV3 nucleic acids. Various sets of primers and gRNAs were designed targeting conserved regions of the PCV3 genome and standardized RPA assay using plasmids of cloned PCV3 genome. The RPA amplicons were detected in gel electrophoresis. Subsequently, CRISPR reaction was implemented for specific detection of amplicons using designed and in-vitro synthesized gRNAs. The CRISP/CAS based cleaved fluorescence probe in positive RPA reaction, was detected in LFA strips. In positive reaction, test line showed strong band in test line and very light to no band in control line. The specific designed primers and guide RNAs of the assay doesn't cross-react with other associated respiratory viruses of pigs. The assay has been validated with qPCR assay and showed comparative results. The developed assay has been found to be highly specific and detection limit is comparable to qPCR. The standardized RPA-CRISPR-LFA assay was tested for detection of PCV3 in previously collected clinical samples. The developed assay successfully detected the positive field cases (Fig.). In conclusion, the developed assay could serve as versatile POC platform for rapid detection of PCV3 nucleic acids in pigs. PCV3 virus is associated with reproductive failure and piglet mortality in pigs which incurs huge economic losses in swine industry.

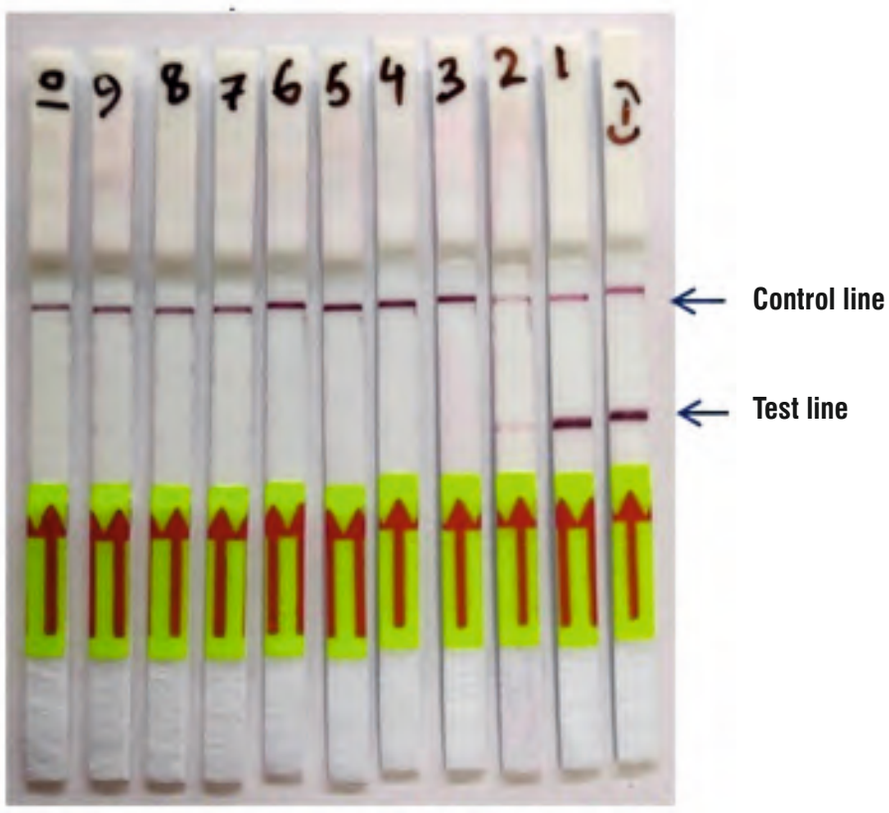


Fig. RPA-CRISPR-LFA detection of PCV3 in clinical samples.

(B.C. Bera, Nitin Virmani & Taruna Anand)

Development of point-of–care diagnostic for detection of SARS-CoV-2 nucleic acids employing RPA CRISPR-LFA assay

The isothermal RT-RPA-CRISPR-LFA assay has been developed for detection of SARS-CoV-2 nucleic acid targeting highly conserved genes of the virus. The RT-RPA reactions were set up first for amplification of the target regions followed by setting up of CRISPR/Cas reaction based cleavage of probe in positive reaction which is detected in LFA strips. The strong positive result indicates strong band in control line and very light to no band in test line. The sensitivity of the developed assay was evaluated in comparison to qRT-PCR. The qRT-PCR was standardized targeting conserved genes using TaqMan probes. The in-vitro transcribed RNA copies of the targeted regions were used as standards.

The serially diluted copies of standards were used as template for qRT-PCR as well as developed RTRPA-CRISPR-LFA assay. The result showed detection limit of developed LFA assay was comparable to qRT-PCR (Fig.). The specificity of the developed assay was evaluated against other respiratory virus viz., bovine coronavirus (BoCV) employing isothermal amplification of the purified RNA of BoCV followed by detection of amplicon using CRISPR-LFA reaction. The developed assay didn't react with BoCV which indicate that the assay is specific for detection of SARS-CoV-2.

The developed assay has been tested using previously purified and stored RNA samples from human clinical samples from SARS-Cov-2 cases. A total of 100 RNA samples purified from human clinical samples were tested using developed RPA assay and result corroborated with the gold standard qRTPCR test. The developed kit has been released during celebration of ICAR foundation and technology day.

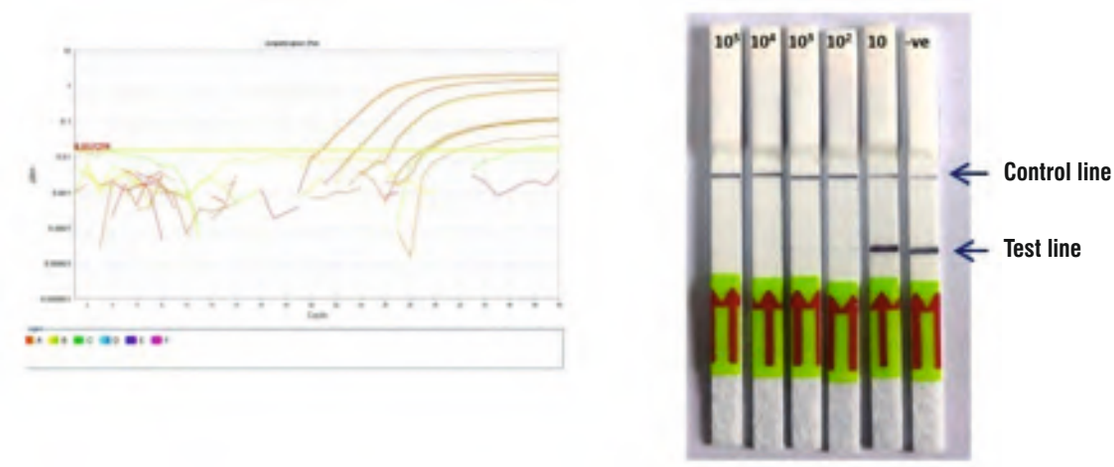


Fig. Evaluation of sensitivity of RT-RPA-CRISPR-LFA assay in comparison to qRT-PC

(B.C. Bera, Nitin Virmani & Taruna Anand)

Evaluation of immunogenicity of mRNA vaccine candidates for SARS-CoV-2 in BALB/c mice

The self-replicating mRNA constructs using immunogenic genes of SARS-CoV-2 were formulated as liposome by encapsulation (LNP) of the in vitro transcribed mRNA in lipid mixture. Subsequently, the immunogenicity of LNP formulations of the mRNA vaccine candidates has been evaluated in BALB/c mice. Different groups of mice were inoculated intra-muscularly with different dosages of sr-mRNA vaccine preparations according to a prime-boost schedule with a 21-days interval. A group of mice was injected with only vector mRNA construct as vector control and one group inoculated with PBS as negative control. To measure immune responses following vaccination, blood samples are being collected at 3 weeks post-prime (day 21) and 2 weeks post-boost (day 35). Immune responses were evaluated by ELISA and flow cytometry. Evaluation of antibody response of the mRNA constructs revealed that overall higher level of antibody production was observed after booster immunization with combined mRNA constructs in comparison to individual construct. The booster immunization increased humoral immune response in vaccine formulations. Evaluation of humoral response of different dosage of individual mRNA constructs revealed that higher antibody production in higher dosage inoculam on 21 and 35 days. The cellular immune response evaluation of immunization of different dosage of mRNA constructs revealed that stimulation of CD8 cells was higher in combined vaccine formulation on 35 days. In conclusion, overall higher level of antibody and cellular immune responses were observed in mice inoculated with higher concentration of mRNA constructs of combined immunogens on 1 week after booster immunization. Hence, the combined mRNA constructs of both immunogenic proteins may be suitable for evaluation of protective immune response of mRNA vaccine formulation in animal model.

(B.C. Bera, Nitin Virmani & Taruna Anand)

Development of RPA-LFA assay for detection of EHV1 & 4 nucleic acids

The isothermal recombinase polymerase amplification (RPA) assays were standardized for isothermal amplification of targeted conserved regions of EHV1 & EHV4 viruses using designed labelled primers, nfo probes and specific reagents using thermal block. The results indicated that all reagents including specific concentration of primers, nfo probes and amplification reagents successfully amplified the targeted region of both EHV1 & EHV4 viruses (Fig.). For detection of RPA amplicons in LFA strip, the set of primers and nfo probes were labelled with different tags for generation of tagged amplicons of EHV1 & EHV4 nucleic acids separately. Upon RPA amplification of target regions of both EHV1 & 4 viruses, the amplicons were mixed with LFA buffer and mixture applied into LFA cassettes. The LFA strip contains implanted bands with both anti-tag antibodies. In positive reaction, two bands –one for control and

one for specific-amplicons were detected. The RPA-LFA assay was successfully standardized using purified DNA of EHV1 & 4. For evaluation of sensitivity of RPA assay for detection of EHV1 virus, the cloned plasmid of target regions of EHV1 was serially diluted to different copies and subjected to standardized RPA reaction as well as qPCR reactions. The detection limit of RPA amplicons was comparable to qPCR assay. Further work on comparative detection limit of RPA-LFA assay with qPCR is underway.

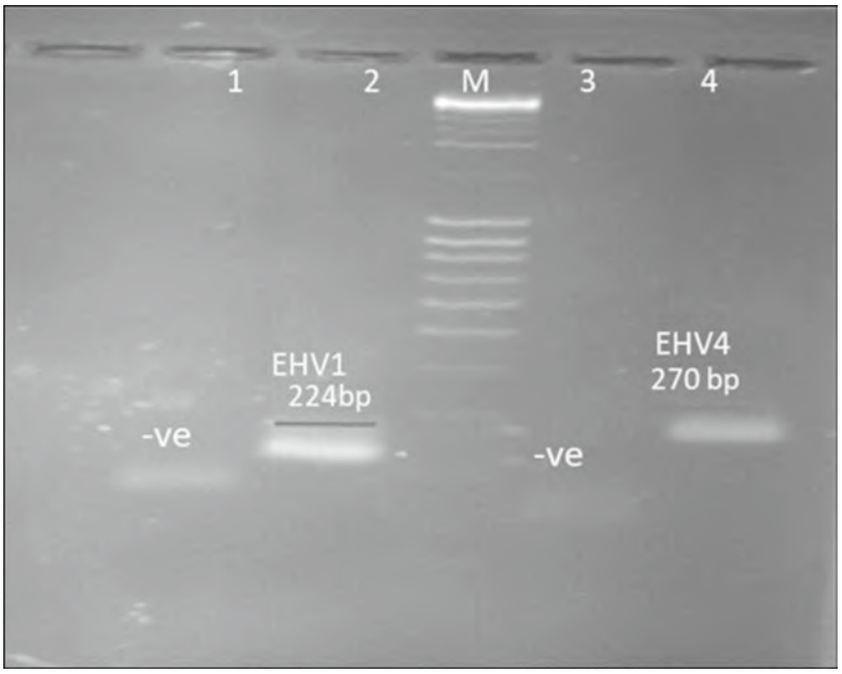


Fig. Isothermal RPA amplification of EHV1 & 4. **L1 :** -ve control, **L2 :** Amplification of EHV1 (224 bp), M: 100 bp DNA ladder, **L3 :** -ve control, **L4 :** Amplification of EHV4 (270 bp)

Molecular detection of Bovine rotaviruses (BRVs)

The bovine rotavirus infection is one of the leading causes of calf mortality due to severe enteric infections. Analysis of circulating strains of BRVs is important to decipher the genetic changes and development of vaccines. We have investigated diarrhea cases in calves in various dairy farms and gaushalas. A total of one hundred and twenty-four (124) fecal samples from Bovine calves were collected from well-organized livestock farms, gaushalas from Haryana and Rajasthan. The viral RNA was extracted from each sample and subjected to detection of rotavirus employing TaqMan based qRT-PCR assay. Out 120 samples, 10 samples were found positive for BRVs. The samples were further confirmed by analyzing purified RNAs from rotavirus positive sample on Polyacrylamide gel by identification of genomic segments of the rotavirus (Fig.).

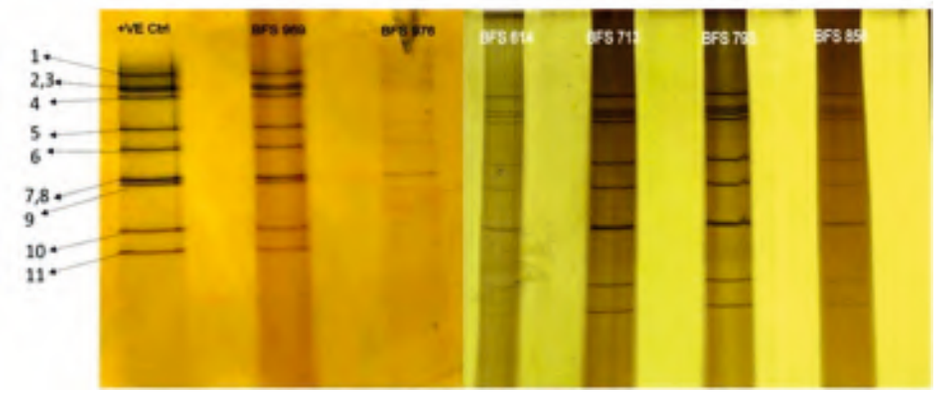


Fig. Electropherogram of group A Rotavirus from diarrheic samples (11 segments of Rotavirus genome)

Isolation of rotaviruses from positive clinical samples

For isolation of BRVs, positive samples were inoculated in MA104 cell culture in Eagle Minimal Essential Medium with 10% fetal bovine serum (FBS) at 37°C in a 5% CO2 atmosphere until CPE has progressed to the point where the cell monolayer is fully disrupted (Fig.). A total of seven rotavirus isolates have been obtained which are confirmed by qRT-PCR and RNA-PAGE analyses. Thirteen clinical positive samples are under process for isolation of BRVs.

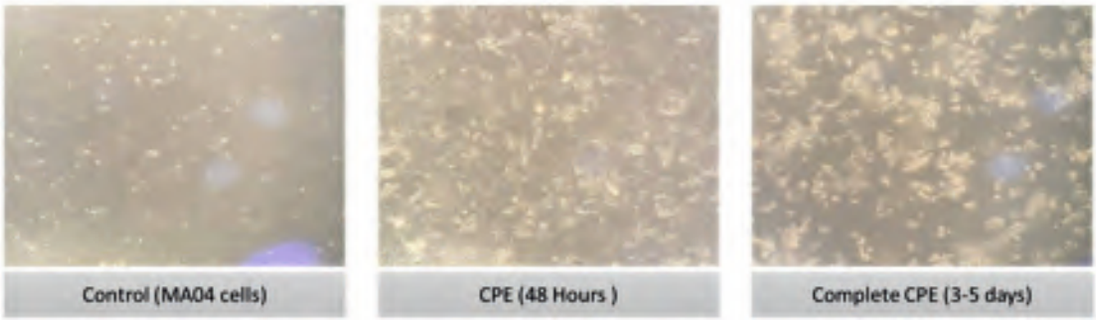


Fig. Isolation of Bovine rota viruses in MA104 cell line

Whole genome sequencing and characterization of rotavirus genomes

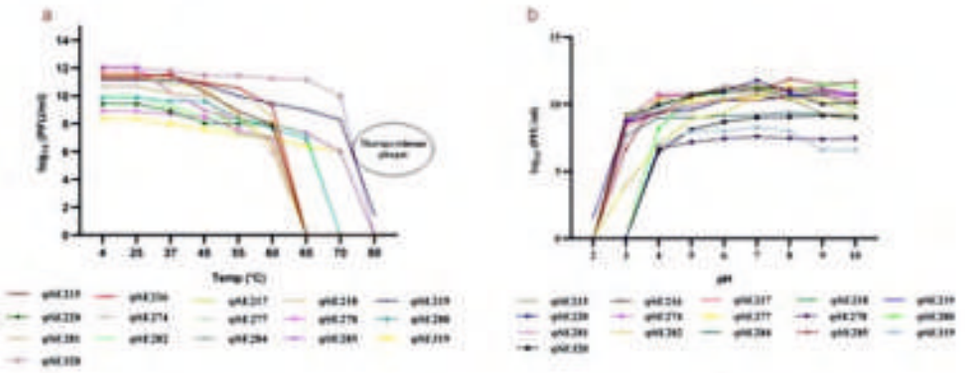
For whole genome sequencing, the two rotavirus isolates were bulk cultured, ultra-purified and extracted viral RNA from the purified viruses. The quality of the RNA was estimated in nanodrop and subjected to whole genome sequencing. The analysis of the genome sequence data revealed that the genotypic constellation of BRVs was G6-P[14]-I2-R2-C2-M2-A11-N2-T6-E2-H3. The genotypic constellation of BRVs is corroborating with the Previously reported in a RVA (Rotavirus A) from stool sample of an Egyptian child in a hospital-based diarrhea surveillance study in the year 2004. BLASTn analysis of each gene with publicly available sequences revealed that the isolates shared a high nucleotide identity with respective segments of human, ruminant and equine origin. The analysis of different structural genes of BRVs revealed that VP1 segment has 97.29% identity with human rotavirus (bovine reassortant) of Thailand; VP2 segment has 96.45% identity with bovine like human rotavirus isolated from Vellore, India; VP3, VP6 and VP4 segments are highly similar to the rotaviruses of African origin; VP6 & VP4 showed 96.27% & 96.70% identity respectively with unusual human rotavirus strain of ruminant origin isolated from a child with severe diarrhea in Malawi and VP7 segment showed 95.50% identity with equine ERV2 isolate from India. Similarly, non- structural genes also showed similarity with other BRVs of different species' origin. We observed that the NSP1 segment has 95.95% identity with a complex reassortant human rotavirus isolated from Kolkata in 2009 with evidence of interspecies transmission; NSP2 segment related to Human strain from Pakistan isolated in 2016 with 98.66% identity; NSP3 & NSP5 segments are highly similar to K-98 Indian caprine rotavirus isolated in 2015 with 97.21% and 98.93% identity respectively and NSP4 segment showed 97.96% identity with an unusual bovine group A rotavirus identified in in Pune, India. The results indicates that BRV isolates shared high nucleotide identity with different genome segments of human, ruminant and equine origin which warrants the thorough investigation of reassortment events in circulating BRVs for implementation of vaccine development and control measures.

(B.C. Bera & Nitin Virmani)

Physiological and genomic characterization of bacteriophages isolated from environmental samples against multidrug-resistant *Salmonella* sp. and *E. coli*

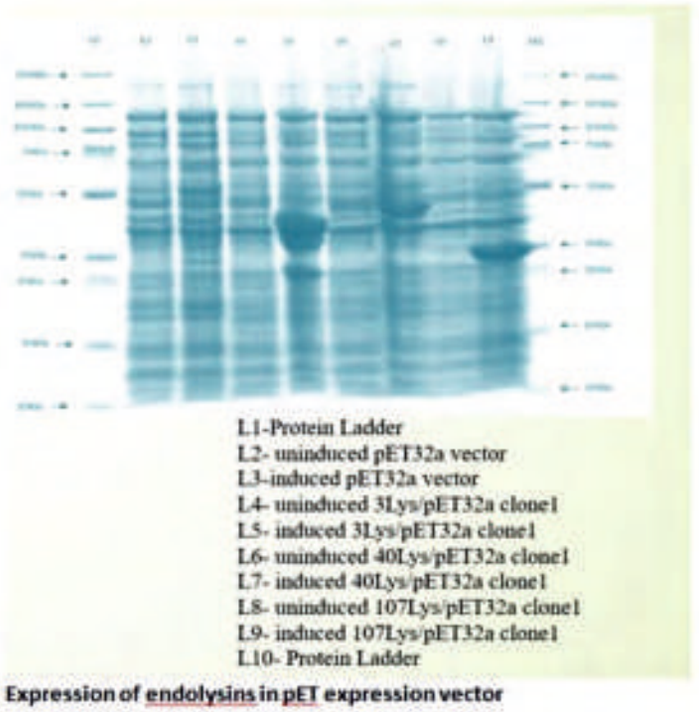
Salmonella enterica and *E. coli* are the leading microbes responsible for foodborne infections, including gastroenteritis, typhoid, and paratyphoid fever, accompanied by diarrhoea, abdominal pain, nausea, and vomiting. Every year, nearly 600 million people get infected, and 4,20,000 lose their lives due to the consumption of contaminated food. This problem is further magnified by the emergence of antimicrobial-resistant strains of these pathogens. The consequences of foodborne disease outbreaks caused by these drug-resistant pathogens extend beyond health impacts, presenting challenges for the attainment of sustainable development goals and thus making

the search for an alternative antimicrobial approach imperative. While targeting foodborne infections, the use of bacteriophages and phage-derived endolysins as antibacterial agents is a promising approach. We have isolated many bacteriophages against *Salmonella* and *E. coli* from various environmental samples like sewage water, pond water, and soil samples. Bacteriophages isolated against *Salmonella* showed a broad host range as compared to *E. coli*. The stability assay of bacteriophages was tested over a wide range of temperature and pH, and we found that some of the bacteriophages remained viable at high temp. (80°C) and also at low pH (pH 3). Further, we selected five bacteriophages for whole genome sequencing based on their screening for lysogenic activities, and we also confirmed their lytic nature by annotating their genome and establishing the absence of genes responsible for the lysogenic nature of the phage. We identified endolysin Lys107 in a *Salmonella* bacteriophage, BPA 319. Globular endolysin of this bacteriophage was cloned in three different expression vectors named pGEX4T, pET32a, and pBAD for assessing their expression level. Results indicated that expression of Lys107 varied in these vectors. Lys107 showed bactericidal activity against many *Salmonella* sp. strains. The results indicated that isolated bacteriophages and endolysin serve as a promising antimicrobial agent against multidrug-resistant strains of *Salmonella* and *E. coli*.



Temp stability assay

pH stability assay



Expression of endolysins in pET expression vector

(Taruna Anand, Nitin Virmani, B.C. Bera & R.K. Vaid)

Authentication and accessioning of bacteria

In 2023, a total of 209 cultures were processed, out of which 115 were accessioned into the bacterial repository, increasing the total collection of veterinary bacteria to 1857. Cultures primarily originated from IVRI, Izatnagar; College of Veterinary Sciences, Palampur; CSWRI, Avikanagar; National Research Centre on Pigs, Gauhati; Assam Agricultural University, Khanapara; NIVEDI, Bengaluru; and MDU, Rohtak. However, the majority of accessions during this period were from IVRI, Izatnagar. Furthermore, the NCVTC Bacteriology laboratory conducted analysis on a wide range of samples obtained from different animal species. These samples included nasal swabs, stomach contents, lung, uterus swabs, fecal samples, and pus swabs from equines, resulting in a total of 113 isolates. Additionally, 20 isolates were obtained from samples collected at the Amarnath Equine Health camp & disease surveillance in Chandanwari and Baltal. Moreover, 26 isolates were derived from horse semen samples, while 31 isolates were obtained from Sheep and Goat pus samples. Overall, a total of 204 bacteria were isolated and cryo-preserved in the repository, falling under the general preservation category.

Some of the significant accessioned cultures are *Pasteurella multocida* Type B, *Aeromonas dhakensis, Ignatzschineria indica, Streptococcus dysgalactiae, Escherichia coli, Moraxella bovoculi, Staphylococcus pseudointermedius, Staphylococcus schleiferi, Streptococcus equi, Brucella melitensis, Truperella pyogenes, Corynebacterium pseudotuberculosis, Pasteurella multocida, Enterococcus gallinarum, Burkholderia cenocepacia, Burkholderia pseudomultivorans, Burkholderia cepacia, Stenotrophomonas maltophilia, Serratia marcescens, Streptococcus porcinus, Staphylococcus lugdunensis, Enterobacter hormaecheii, Enterococcus faecalis, Lactobacillus delbrueckii* ssp. *indicus, Lactobacillus delbrueckii* ssp. *lactis, Solibacillus isroensis, Agrobacterium fabrum, Sphingomonas paucimobilis, Microbacterium zeae, Enterobacter sasburiae, Enterobacter mori, Klebsiella grimoutaii, Enterobacter quasihormaechei, Azospirillum formoseuse, Stutzerimonas stutzeri, Corynebacterium tapiri, Flavobacterium acidificium, Leclercia tamurae, Neisseria canis, Bacillus safensis, Bacillus licheniformis, Staph. warneri, Streptococcus dysgalactiae* ssp. *equissimilis, Staphylococcus caledonicus, Staphylococcus cornubiensis, Pseudomonas songnensis, P. protogenes, P. guariconensis, Streptococcus equi* ssp. *ruminatorum, Streptococcus equi ssp. equi.* Four cultures of STEC, EHEC, EPEC *Escherichia coli* has also been accessioned.

The total of 115 bacterial accessions belonged to 28 taxa of following genera: *Pasteurella* spp., *Aeromonas* spp., *Ignatzschineria* spp., *Streptococcus* spp., *Moraxella* spp., *Staphylococcus* spp., *Brucella* spp., *Truperella* spp., *Corynebacterium* spp., *Enterococcus* spp., *Burkholderia* spp., *Stenotrophomonas* spp., *Serratia* spp., *Enterobacter* spp., *Escherichia* spp., *Lactobacillus* spp., *Solibacillus* spp., *Agrobacterium* spp., *Sphingomonas* spp., *Microbacterium* spp., *Klebsiella* spp., *Azospirillum* spp., *Stutzerimonas* spp., *Flavobacterium* spp., *Leclercia* spp., *Neisseria spp., Bacillus spp, Pseudomonas* spp., have been cryopreserved, which is a significant addition to NCVTC in 2023.

(R. K. Vaid, Taruna Anand, Sumanshu, Shanmugasundaram K, T. Riyesh & B. C. Bera)

DBT network programme on anthrax diagnosis and control in India

DBT network programme on anthrax diagnosis and control in India focuses on combating zoonotic anthrax endemic in Southeastern India, impacting livestock, wildlife, and occasional human outbreaks. Establishing a Bacillus anthracis Culture Collection is pivotal for research. Key activities include protocol development, SOP implementation, biosafety drills, culture processing under BSL3 conditions, and authentication. Emphasis on biosafety included drills with Bacillus cereus cultures. Despite challenges in sourcing UN2814 certified containers in India, they were secured. Culture accessioning and characterization allocated 40 slots in the NCVTC Accession Register. Strains VTCCBAA1760 (BA) and VTCCBAA1761 (SV) from ICAR-IVRI, Izzatnagar were revived and characterized phenotypically in BSL3 labs. Both strains exhibited viability and distinct characteristics on sheep blood agar. Gram staining revealed long bacilli in chains. BA1 showed resistance in penicillin susceptibility tests, while SV1 was sensitive. Conventional PCR detected presence of genes PAG, CAP, and Ba813 in the isolates. SV1 lacked the capsule gene. PCR for rpoB gene confirmed species specificity. Real-time PCR standardized for *CAAX, PL3, cya,* and

capB genes showed PL3 and cya presence in both strains. BA strain was positive for capB, while SV was negative. Both strains were positive for CAAX. These findings refine anthrax diagnostic protocols for effective disease management.

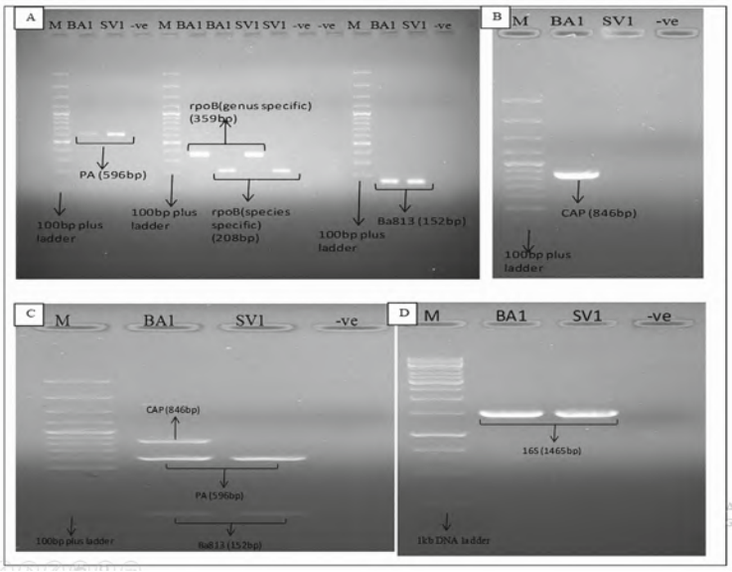


Fig. (A, B) Amplification of PA (596bp), Ba813 (152bp), rpoB(genus specific-359bp) rpoB (species specific 208bp) and CAP (846bp) by Conventional PCR. (C) Multiplex PCR- PA (596bp), CAP (846bp), Ba813 (152bp). (D) Amplification for 16SrRNA (1465bp)

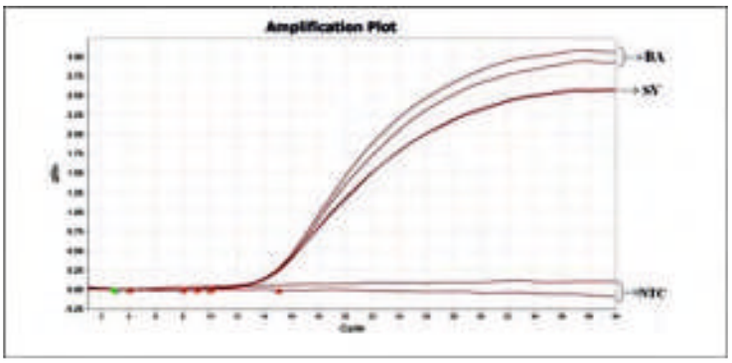


Fig. Amplification plot of cya gene for BA, SV and Negative control (Amplification plot of cya gene for BA, SV and Negative control)

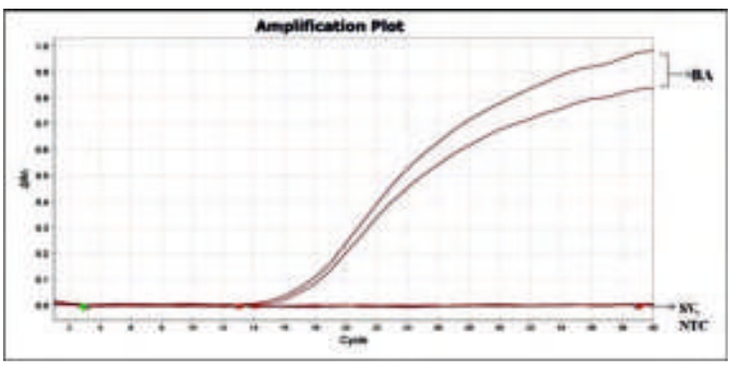


Fig. Amplification plot of capB gene for BA (Ct value: 16.832), SV and Negative control

(R.K. Vaid, B.C. Bera, Shanmugasundaram K, Sudesh Kumar & Ritu)

Antibacterial activity of domestic donkeys' milk against *Escherichia coli* and *Staphylococcus aureus* under various storage conditions

Domestic donkeys have been historically utilized for various purposes, including agricultural and transportation roles. With advancements in technology, their traditional uses have diminished, prompting the exploration of alternative avenues for donkey-related products, such as donkeys' milk. Apart from its nutritional value, recent studies have suggested potential antibacterial properties associated with donkeys' milk. This study aimed to explore its antibacterial effects against *E. coli* and *S. aureus* under varied storage conditions. Donkey milk samples were collected under sterile conditions and frozen at -20°C until analysis. Antibacterial assays were conducted using *Escherichia coli* (ATCC 25922) and *Staphylococcus aureus* (ATCC 29213). Bacterial strains were cultured, suspended in saline, and inoculated into milk samples at 105 cfu/mL. Samples were incubated at temperatures of 4°C, 7°C, and 15°C for 96 hours, with bacterial enumeration at regular intervals using serial dilutions. Results indicated that donkey milk effectively inhibited *E. coli* and *S. aureus* growth across all temperatures tested. Significant differences ($P < 0.05$) were observed between donkey milk and control substances (pasteurized cow milk and nutrient broth) at various time points and temperatures, highlighting its superior antibacterial activity.In conclusion, domestic donkeys' milk shows promise as a natural antibacterial agent, offering potential applications in food and pharmaceutical industries.

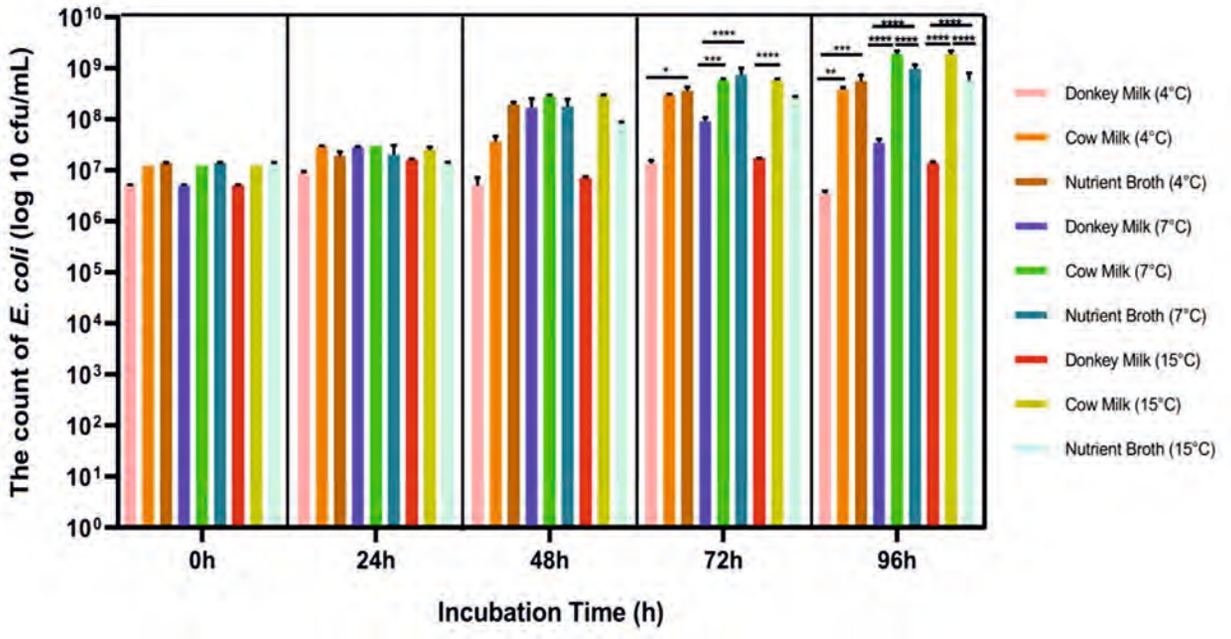


Fig. *E. coli* count changes in raw donkey milk, pasteurized cow milk and nutrient broth at 4ºC, 7ºC, 15ºC temperatures over 96 h

(R.K. Vaid, Sudesh Kumar, Sumanshu Narwal, Ritu, Jyoti Bakshi & A. Bharadwaj)

Indian network for fisheries and animal antimicrobial resistance (INFAAR)

Antimicrobial resistance poses a significant challenge to both animal and public health, necessitating comprehensive management strategies. Surveillance of antimicrobial resistance (AMR) in domestic animals raised for food is crucial. Under the INFAAR surveillance project, 65 milk samples were collected from cattle and buffalo, processed to isolate *Staphylococcus* spp. and *Escherichia coli*. Additionally, *E. coli* was isolated from fecal samples, including rectal swabs from sheep, goats, pigs, and cattle. A total of 274 samples were collected primarily from different blocks of district Sirsa, with some from Hisar. From the milk samples, 69 isolates of *Staphylococcus* spp. and 14 of *Escherichia coli* were obtained. Pig samples yielded 23 *E. coli* isolates, while 138 *E. coli* were isolated from 82 sheep and 63 goat samples. Thirty-two *E. coli* were isolated from 34 poultry cloacal swabs, and 10 *E. coli* were

isolated from cattle rectal swabs. In total, 217 *E. coli* isolates were biochemically confirmed and further validated by multiplex PCR (*lacY, cydA, lacZ, uidA,* and *phoA* genes). Among the 69 morphologically positive *Staphylococcus* spp., genotypic confirmation for *Staphylococcus aureus* was conducted through nuclease gene (*nuc*) PCR, revealing that 55 isolates (80%) were identified as *S.aureus.*

Disc diffusion testing for antimicrobial resistance (AMR) was conducted on 217 *E. coli* isolates. Results showed that 24(11.05%) isolates were resistant to ampicillin; 44(20.27%) to nalidixic acid; 26 (11.9%) to tetracycline; 11(5.06%) to trimethoprim; 10(4.6%) to cefotaxime; 13(5.9%) to ceftraixone; 1(0.46%) to amikacin; 2(0.92%) to aztreonam; 10(4.6%) to norfloxacin; 1(0.46%) to chloramphenicol; 4(1.84%) to cefodoxime; 2(0.92%) to ceftazidime; 6(2.76%) to cefoxitin and 1(0.46%) to amoxiclav. All *E. coli* isolates were sensitive to imipenem. *Escherichia coli* isolates from sheep and goats exhibited low AMR, followed by pigs and poultry.

AMR testing on *Staphylococcus* showed that of 69 isolates, 24(34.7%) isolates were resistant to pencillin. *Staphylococcus* spp. showed comparatively less resistance to rest of the antimicrobials. Three (4.3%) were resistant to cefoxitin, 1(1.5%) to erythromycin, 2(3%) to linezolid. All *Staphylococcus* spp. were sensitive to chloramphenicol, gentamicin, tetracycline and trimethoprim. No isolates were found to be genotypically positive for MRSA. *Escherichia coli* isolates were also tested for ESBL and a total of 13 *E. coli* isolates (6%) were found to be ESBL producers. Out of 8 *E. coli* isolates tested for ACBL producer 6(2.8%) were ACBL producers. The *E. coli* isolates which were ESBL producers were further subjected to PCR analysis to detect the ESBL resistance genes.

(R. K. Vaid, Taruna Anand, Shanmugasundaram K, Jyoti Bakshi & H. S. Singha)

Isolation of *Corynebacterium pseudotuberculosis* from Sheep

Caseous lymphadenitis (CLA) is a contagious bacterial disease caused by *Corynebacterium pseudotuberculosis* in sheep and goats. *C. pseudotuberculosis* is a Gram-positive bacterium that causes CLA, characterized by abscess formation in the lymph nodes, skin and even internal organs of infected animals, such as lungs, kidneys, liver, and spleen. CLA is a significant concern in the livestock industry due to its economic impact on animal productivity and welfare. The isolation of the causative bacteria i.e., *C. pseudotuberculosis* from abscesses is considered to be golden standard for diagnosis.

Sheep and goats were sampled (swabs and pus fluid), and processed for aerobic and anaerobic cultures. The Nasal swabs were processed for aerobic cultures which showed highly mixed growth. All 3 sheep pus samples yielded isolates of *Corynebacterium pseudotuberculosis.* From the nasal swab sample of sheep from Sector II, *Pasteurella multocida*, *Corynebacterium* spp. *Klebsiella* spp., *Proteus, E. coli* were identified. From pus samples of goat 2 isolates of *Gemella haemolysans*, *Micrococcus* spp., and 1 *Corynebacterium* spp., were isolated. From Sirohi goat unit, 7 pus samples were cultured although none of these samples yielded *C. pseudotuberculosis* isolate. 1 *Corynebacterium* spp. isolate from Sirohi goat sample was identified as *Corynebacterium flavescens* (Ch21B). Further, 16S rRNA sequencing has identified many bacterial taxa isolated from pus samples viz., Staphylococcus *aureus* subsp. *anaerobius*; other species of *Staphylococcus*); *Brachybacterium* spp*, Enteractinococcus* spp*.* and (Ch21A); and *Dietzia* spp*.* (Ch21D). It is noteworthy that goat pus samples were positive for *Staphylococcus aureus* ssp*. anaerobius* however these were negative for isolation of *C. pseudotuberculosis.*

(R. K. Vaid, Shanmugasundaram K & Naveen Kumar)

Isolation, characterization and generation of repository of *Mycobacterium* Species

Mycobacteria are gram positive, small, aerobic and acid-fast organisms belonging to the family of *Mycobacteriaceae* and the order *Actinomycetales*. Historically, infections caused by mycobacterium species are well known. In general, isolation, characterization and maintenance of Mycobacterial species are time and resource consuming process and individual laboratories across the country may not be able to do. Therefore, development and optimization of protocols for isolation and characterization of Mycobacterial species from different samples are essential to generate repository of Mycobacterial species and useful in supply of reference strains to the

researchers, academicians and biotechnological exploitations. Samples (n=109) were collected for isolation of mycobacterial species. It includes cattle (n=15) and buffaloes (n=4) fecal samples, sheep fecal (n=47), milk (n=5), sheep blood (n=8), nasal swabs from cattle (n=5), sheep intestine (n=10), cattle lung (n=3), soil samples (n=8), horse dung (n=1), camel lung (n=2) and water sample (n=1) were collected. Acid fast staining of nasal swab samples of cattle (n=5), lung samples of cattle (n=2), and sheep fecal (n=42) samples were performed. Sheep fecal samples (n= 10) found positive for the presence of acid-fast bacilli. Intestinal samples (n=8) and 10 sheep fecal showed positive amplification for *afb* and *hspx* genes. Rest of the samples found negative for *afb* and *hspx* genes. Seven acid-fast bacilli haves been isolated from sheep fecal samples and further characterization is under progress. Four mycobacterial isolates have been submitted for the whole genome sequencing.

(Shanmugasundaram K, RK Vaid, Indu, Soni, Riyesh T & BC Bera)

CRISPR- Cas9 mediated gene editing in *Mycobacterium kansasii*

Mycobacterium kansasii is an acid-fast, slow growing, photo-chromogenic opportunistic pathogenic bacteria that cause range of infections in immunocompromised individuals, including pulmonary tuberculosis. There is a lack of functional genomics studies to understand the role of several genes in bacterial survival in hosts, environments, host-pathogen interactions, and virulence. Several studies have validated the function of MCC genes in survival of many *Mycobacterium* spp. Therefore, in this study we were using CRISPR-Cas9 approach to decipher the role of MCC genes in survival of *M. kansasii*. We designed the Single guide RNA (sgRNA) for MCC genes using CHOPCHOP web interface, and custom PAM sites were used to identify the target sites, designed primers using NCBI primer designing tool. Plasmid pCRISPRx-Sth1Cas9-L5 was digested using BsmBI enzyme, dephosphorylated and purified by gel purification kit. Phosphorylated annealed oligos containing BsmBI overhangs were annealed into the plasmid. After ligation transformed colonies were observed on the LBA plates containing 50ug/ml Kanamycin. Clones with desired amplification were PCR purified and to confirm the insert positive clones were sequenced commercially **(Fig.).** After confirmation, plasmid containing gRNAs was isolated and transformed into electrocompetent *M. kansasii* cells using Eppendorf Eporator. Transformed *M. kansasii* colonies were observed on the Middlebrook 7H10 plates containing Kanamycin and confirmed the insert by colony PCR **(Fig).** For induction, bacterial colonies were picked and grown on Middlebrook 7H10 plates with 50ug/ml Kanamycin and 100 ng/ml ATc (Anhydrotetracycline). After three weeks of incubation, separate colonies were selected, DNA was extracted and *M. kansasii* mutants were confirmed by PCR and sequencing.

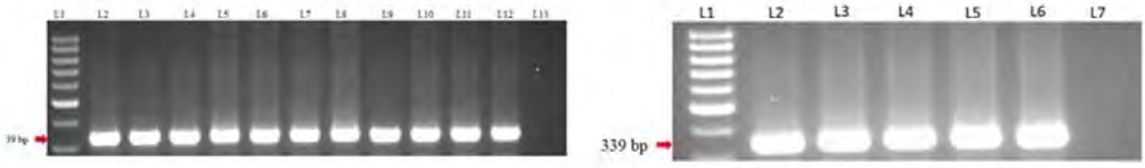


Fig.(A) Fig. (B)

Figures- (A)- Confirmation of transformed E. coli DH5α colonies by amplification of pCRISPRx-Sth1Cas9-L5 plasmid gene sequences using pCRISPRx primer set. (B). Confirmation of pCRISPRx-Sth1Cas9-L5 plasmid containing gRNA in Electrocompetent M. kansasii by PCR.

(Indu Rani, Shanmugasundarm K, Rakesh Kumar, R.K. Vaid & H. Singha)

Technology Development, Transfer and Commercialisation

Since its inception, ICAR-National Research Centre on Equines is continuously striving for the upliftment of equine sector in the country, oriented its research, and focused on the development of farmer friendly technologies for improvement in equine health, production, and utilization in the country. Many diagnostic kits, vaccines and biologicals developed by the scientists of ICAR-NRCE are routinely used in the field. Many of the novel and innovative technologies are under development, transfer, and commercialization.

I. List of Technologies developed by ICAR-NRCE, Hisar

A. Vaccines for Equines and other Livestock species

i. Equiherpabort vaccine.

ii. Updated Equine Influenza vaccine.

iii. A modified vaccine construct for EHV1 and methods of preparing the same.

iv. Corona virus SARS-CoV-2 vaccine for animals.

v. Lumpy Skin Disease vaccine.

vi. Reverse genetics based recombinant equine influenza virus as vaccine candidate.

B. Disease Diagnostics for Equines and other Livestock species

i. Equiherpes B-ELISA Diagnostic Kit.

ii. ELISA kit for differentiation of EHV1 & 4 Infections.

iii. Equine Rotavirus Diagnostic kit. iv. Equine Infectious Anaemia (EIA) ELISA Diagnostic kit.

v. Equine Japanese Encephalitis Virus Antibody Test Kit, iELISA.

vi. Monoclonal antibody based immunoasssay for detection of equine influenza (H3N8) antigen.

vii. Canine SARS-CoV-2 Antibody detection ELISA Kit.

viii. Glanders ELISA Kit.

ix. Duplex Antigen Glanders ELISA Kit.

x. *Theileria equi* ELISA Diagnostic kit.

xi. Lateral flow assay for diagnosis of *Theileria equi* infection.

xii. Surra (*Trypanosoma evansi*) ELISA Diagnostic Kit.

xiii. RPA-LFA kit for point-of-care detection of Porcine circovirus 3.

xiv. LFA kit for SARS-CoV-2 nucleic acid detection.

C. Drug Development and delivery Technologies

i. Polymeric metal nanocomposites and methods of synthesis thereof.

ii. Poly ZnO xeno artificial skin construct.

iii. Yasadbhasma, and hydroxychloroquine formulation as cytoprotective and antiviral effect.

iv. Hydroxychloroquine/ chloroquine zinc oxide nanoparticle formulation.

v. Nanodelivery of quinapyramine sulfate.

vi. *Aerva javanica* extract for the treatment of exuberant granulation tissue and tumors in horses.

vii. *Aerva javanica* extract for the treatment of seasonal dermatitis in horses.

D. Equine Reproduction and Production Technologies

i. A pregnancy diagnostic kit for equine based on detection of eCG by ELISA (Pregmare Kit).

ii. Customised AV (artificial vagina) for semen collection from Stallions.

iii. Semen collection and cryopreservation in Indigenous horses.

iv. Equine DNA Parentage Testing Kit.

v. Development of protocol for embryo transfer in Marwari horses.

vi. Modified vitrification protocols for cryopreservation of 7– 8 day old large sized embryos in mares.

vii. Modified cryoloop and Hemi-straw methods of vitrification of large sized horse embryos.

viii. Improved methods of Cryovial preservation of stallion's semen and thawing protocols.

ix. Development of non-egg yolk based (nano-encapsulated drug delivery based encapsulated systems) semen extender to increase the shelf life and post-thaw motility in stallion semen.

E. Equine Products Technologies

i. Processes for formulation of donkey-milk based cosmetic products – bathing soap, body butter, and lip balm.

F. *Therapeutics*

i. Antiviral activity of Apigenin against buffalopox.

ii. Antiviral activity of Emetine against SARS-CoV-2 virus.

iii. p38-α -a novel drug target against buffalopox virus.

iv. ROCK1/MLC2- a novel drug target against buffalopox virus.

v. Antiviral activity of DZNep against SARS-CoV-2 virus.

G. Methodologies

i. Identification and quantitation of the vaccine and field strain(s) of lumpy skin disease virus.

ii. Development of bacteriophage against drug-resistant (XDR) *Pseudomonas aeruginosa.*

iii. 'Salmoquell" – Encapsulated bacteriophage cocktail for treatment of Salmonella infections in poultry.

iv. Development and treatment regimen using combination of phages and antibiotic to treat multidrug resistant bacterial infections.

v. Construction of Bacterial artificial chromosome (BAC) of Equine herpesvirus 1.

vi. Quadruple genes deletion mutant equine herpesvirus 1 as live vaccine candidate.

vii. Double genes deletion mutant equine herpesvirus 1 as live vaccine candidate.

TECHNOLOGIES CERTIFIED BY ICAR

S. No.	Name	Lead Developer
1.	Hydroxychloroquine/chloroquine zinc oxide-nanoparticle formulation: **Technology**	Dr. Anju Manuja
2.	Polymeric Zinc oxide nanocomposites for wound healing: **Technology**	Dr. Anju Manuja
3.	Bacterial artificial chromosome construct of Indian isolate of Equine herpesvirus 1 (Strain Tohana) : **Technology**	Dr. B. C. Bera
4.	Quadruple genes deletion mutant equine herpesvirus 1 as improved live vaccine candidates for control of EHV1 infection in equines: **Technology**	Dr. B. C. Bera
5.	Double genes deletion mutant equine herpesvirus 1 as live vaccine candidates for control of EHV1 infection in equines:**Technology**	Dr. Nitin Virmani
6.	A novel HRM-based gap-qRT-PCR for identification and quantitation of the vaccine and field strain(s) of lumpy skin disease virus: **Technology**	Dr. Naveen Kumar
7.	Ancovax: Coronavirus (SARS-CoV-2) vaccine for animals: **Technology**	Dr. Naveen Kumar
8.	Development of India's first homologous live-attenuated Lumpy Skin Disease Vaccine: **Technology**	Dr. Naveen Kumar
9.	Monoclonal antibody-based immunoassay for detection of equine influenza (H3N8) antigen: **Technology**	Dr. Nitin Virmani
10.	Recombinant nucleoprotein-based Canine SARSCoV-2Antibody detection ELISA Kit: **Technology**	Dr. Nitin Virmani
11.	Development of Bacteriophage Cocktail to Eradicate Biofilms Formed by Extensively Drug-Resistant (XDR) Pseudomonas aeruginosa: **Technology**	Dr. Taruna Anand
12.	"Salmoquell" – Encapsulated bacteriophage cocktail for oral delivery in poultry for treatment of Salmonella infections: **Technology**	Dr. Taruna Anand
13.	Surra ELISA Kit: **Product**	Dr. Rajender Kumar
14.	Equine Japanese Encephalitis Virus Antibody Test Kit, iELISA: **Product**	Dr. Baldev Raj Gulati

II. Technology Development & Assessment

a. Multiplex taqman qPCR for in vitro detection of *Streptococcus* species and *Streptococcus equi* subspecies *equi* :

A multiplex taqman qPCR assay intended to be used for in vitro detection of *Streptococcus* sps and *Streptococcus equi* subsps. *equi* in respiratory samples (naso-pharyngeal swabs, guttural pouch washes etc) for specific detection of the organisms. The assay has been internally validated by two laboratories at ICAR-NRCE and NCVTC, Hisar.

Inventors: Balvinder Kumar, R.K. Vaid, Anju Manuja, K. Shanmugasundarm & Harisankar Singha

b. LFA kit for SARS-CoV-2 nucleic acid detection

The rapid spread of SARS-CoV-2 virus throughout the world has necessitated quick detection of etiological agents for timely treatment, risk reduction, and prevention of further spread of the virus. Many reports described the incidences of SARS-CoV-2 virus infections in animals including dog, cat, mink, tiger and lion. Thus, there is need to develop highly sensitive point-ofcare test (POCT) for rapid detection of SARS-CoV-2 infections in humans as well as animals. We have developed isothermal RT-RPA coupled with CRISPR/Cas

and LFA assay for detection of SARS-CoV-2 nucleic acids in clinical samples. The isothermal amplification of the SARS-CoV-2 RNAs is carried out in thermal block and amplicons are detected in LFA strip through CRISPR/Cas12a reaction. The assay has been validated with gold test i.e. qRT-PCR assay using human clinical samples and showed comparative results. The assay has been found to be highly specific and detection limit is comparable to qRT-PCR. The salient features of the new generation isothermal RT-RPA-CRIPSR-LFA based point-of-care test includes quick detection of SARS-CoV-2 within 1 1hr; assay can be performed using thermal block at field condition; easy interpretation of result in lateral flow strips; highly sensitive – equivalent to qRT-PCR. The developed kit has been released by Hon'bl Union Minister of Fisheries, Animal Husbandry & Dairying) and Hon'bl Minister of State for Agriculture and Farmers Welfare and DG (ICAR) during celebration ICAR-Foundation and Technology Day.

Inventors : B.C. Bera, Nitin Virmani & Taruna Anand

c. Development of Lysibact antimicrobial nano-spray

A phage endolysin encapsulated antimicrobial formulation for spray application to ameliorate biofilms has been prepared. This antimicrobial formulation carries naturally derived compounds – mainly essential oils in combination with broad activity phage endolysin. The nanoformulation was also assessed for its safety on fibroblast cell lines and in vitro against bacterial spectrum. It was also assessed for reducing bacterial load on meat and in amelioration of biofilms and was found to be effective. The novel nanoformulation named "Lysibact" can be applied on ready to eat foods such as salami and chicken ham.

Inventors : Taruna Anand, Nitin Virmani, B.C. Bera & R.K. Vaid

III. Release of Technologies

a. Release of LFA kit for SARS-CoV-2 nucleic acid detection

Technology entitled "LFA kit for SARS-CoV-2 nucleic acid detection" was released by Hon'ble Union Minister of Fisheries, Animal Husbandry & Dairying) and Hon'ble Minister of State for Agriculture and Farmers Welfare and DG (ICAR) on 16 July, 2023 at Krishi Bhawan, New Delhi.

Release of LFA kit for SARS-CoV-2 nucleic acid detection (16 July 2023 at Krishi Bhawan, New Delhi) by Hon'ble Union Minister of Fisheries, Animal Husbandry & Dairying) and Hon'ble Minister of State for Agriculture and Farmers Welfare and Secretary DARE and Director General ICAR.

IV. List of Technologies developed

a. Development of protocols for embryo transfer in indigenous horses

The 19th and 20th Livestock Census (2012-2019) data reveals that the total horse population in the country is 0.34 million, and they have gone down by 45.58 %. Collectively, the total equine population was reduced by 52.71 % during this period. High demand for genetic preservation of equids has led to developments and improvements in assisted reproduction and a rapidly growing area of equine medicine. Over the decades, NRCE has been working on the genetic conservation and propagation of indigenous horse and donkey breeds. In this endeavour, scientific team at Equine production Campus, Regional Station, ICAR-national Research Centre on equines has standardised the estrus synchronisation protocols, embryo flushing and embryo recovery and to produce foals through embryo transfer technology in indigenous horses. For establishment and standardisation of this technology, donor and recipient mares were screened and selected. Estrus synchronisation protocols using shortening the luteal phase (PGF2α) and extending the luteal phase (CIDR) were tried. Combination of both protocols with injection of HCG as an ovulating agent for timed insemination procedures were also adopted for successful estrus synchronisation of mares. Non-surgical methods of embryo flushing protocols using non-commercial media and without BSA were tried and successfully recovered Grade A Quality embryo. The recovered embryos were successfully transferred into recipient mares and established pregnancies.

Fig. 1. Step by step procedure for producing foals through embryo transfer in horses. Raj –Prathama is the India's First Marwari filly born through embryo transfer. It was born on 19th May 2023 and is healthy

(TR Talluri, Sajjan Kumar, Yash Pal, RA Legha, RK Dedar & TK Bhattacharya)

b. Development of Non-Egg Yolk based Alternative Semen Extender for Stallion Semen

Since the discovery of egg yolk as an ingredient of bull semen extender, it has been extensively used in mammalian semen cryopreservation to protect sperm against initial cold shock. Egg yolk is normally used as a protective agent to freeze semen of equine and other species. However, addition of egg yolk in extenders is not without disadvantages and the demand to find cryoprotective alternatives is strong. Egg yolk has biosecurity risks. Egg yolk is an extremely complex product, whose composition may be extremely variable and may differ between batches. Its lipid composition may change with the hens' diet. Moreover, egg yolk may contain components with beneficial, as well as detrimental, effects on spermatozoa. The presence has been reported of substances in egg yolk that inhibit respiration of spermatozoa or reduce their motility. Considering these disadvantages, there is an urgent need to replace whole egg yolk by its cryoprotective fraction. Keeping the above facts, we at Equine production Campus, ICAR-NRCE have

developed novel liposome-based nano-encapsulated drug delivery systems containing various antioxidants and agents that can protect the spermatozoa from the cold shock and oxidative stress. For the development of novel non- egg yolk semen extender, we used nano-liposomes encapsulated with Vit E, Selenium, Cholesterol and Albumin in varying concentrations. The best combination of these were tested initially and the best combination was selected further for the use as extender. All the preparations of nano-liposomes were prepared at NRCE only in customised methods.

Post-thaw seminal parameters of control and novel extender

	Post thaw seminal parameters	
	Control (Egg Yolk)	Non-Egg Yolk Extender
PTM	38.29 $\pm$ 1.24	45.17 $\pm$ 2.49
Viability	50.17 $\pm$ 2.14	56.17 $\pm$ 1.79
Acrosome Integrity	65.47 $\pm$ 2.29	68.19 $\pm$ 2.19
MMP	52.18 $\pm$ 2.47	56.13 $\pm$ 2.38
HOST	39.14 $\pm$ 3.07	44.37 $\pm$ 2.47

Post-thaw Oxidative parameters

	Pre-free		Post-Thaw	
	C	T	C	T
LPO (MDA)	201.13	180.02	499.02	445.99
ROS	129.8	108.21	213.96	170.94

(TR Talluri, Sajjan Kumar, Pyarelal, RK Dedar & TK Bhattacharya)

c. Methods of cryovial preservation of stallion semen and thawing protocols

Freezing and stock of semen is a safe procedure to preserve reproductive potential of animals with superior genetic heritage. In the horse industry, unlike observed in ruminants, the development of sperm cryopreservation techniques is very slow but despite the technical barriers, the artificial insemination with frozen semen is growing. Ideally, we need 500-600 million of spermatozoa for breeding the mares via artificial insemination either with fresh or frozen semen. The frozen semen is usually stored in 0.5 ml French medium straws. Therefore, we need to collect 10 straws from the LN_2 container and thaw them at same time and cut them uniformly at the same time to get more live and motile spermatozoa for insemination. This whole process is time taking and not economical. To circumvent these process researchers developed alternative strategies like for example, big straws of 4 mL, glass macro tube of 12 mL, and aluminium macro tube of 25 mL and sachet of 15 mL. The cryopreservation of equine semen packaged in these was not found to be optimum for breeding. Hence, we at NRCE have developed a novel, practical and inexpensive method for freezing stallion and donkey semen for use in routine inseminations under farm and filed conditions.

The freshly collected semen from the stallions was assessed for the gross and microscopic properties. The semen with more than 60 % progressive motility was seen further processed for primary dilution and washing of the spermatozoa. The sperm pellet was selected after washing and then further diluted with secondary extender containing the cryoprotectant and egg yolk and the concentration of the sperm were adjusted to 150 million/ml. The semen was either filled in 0.5 ml straws or 2.5 ml or 5 ml cryovials for studying the efficiency of cryopreservation in cryovials in comparison to the 0.5 ml straw methods of cryopreservation. The post thaw parameters did not alter with the methods of cryopreservation in cryovials.

(TR Talluri, Sajjan Kumar, RK Dedar, SC Mehta & TK Bhattacharya)

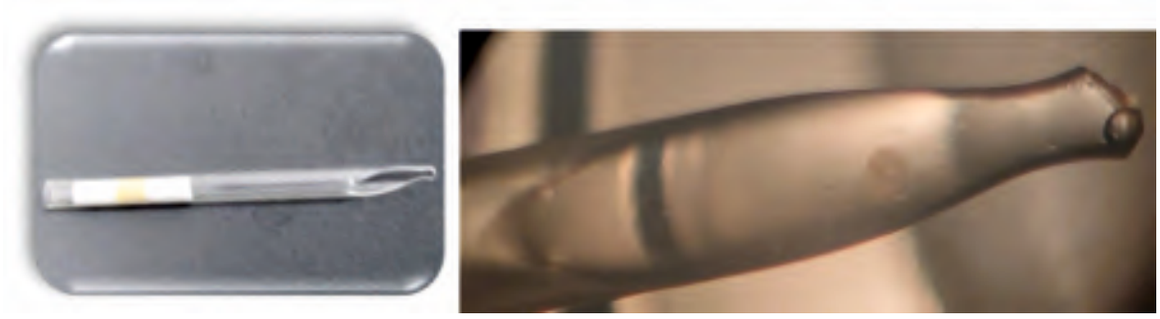
d. Customised novel cryodevices for vitrification of horse embryos

Several vitrification protocols using different cryoprotective agents (CPAs) alone or in combination, different regimes of vitrification and different cryo-devices have been tested for vitrification of small equine embryos recovered at day 6-7. But unfortunately, there were no uniform cryodevices described for vitrification of the large sized equine embryos recovered beyond days 7 or 8. Hence, we at Equine Production Campus, ICAR-NRCE have designed some cryodevices like cryoloop and Cryotop like devices which can hold the large sized embryos and can successfully use for vitrification of the large sized equine embryos. Here at our laboratory, we designed special cryodevices namely cryoloops using fish net over the cryovial, hemi straws using the French medium straws so that they can hold the large size embryos.

Cryoloops were prepared by twisting a loop of fishnet wire (0.5 mm thickness) around a 2.5 ml cryovial. The free end of each loop was fixed to the cap of a 2.5 ml cryovial such that the loop hangs clearly above the bottom of the vial.

Modified cryoloop developed at NRCE

Hemi straws were prepared by cutting the open end of French medium straw at an angle of 45° with a surgical blade.

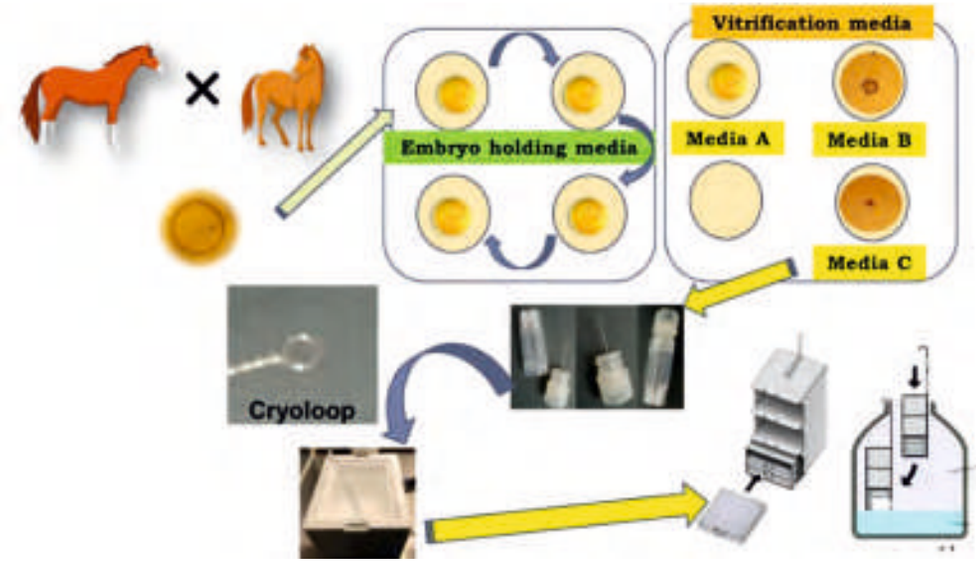


Hemi-straws prepared by using 0.5 ml straws

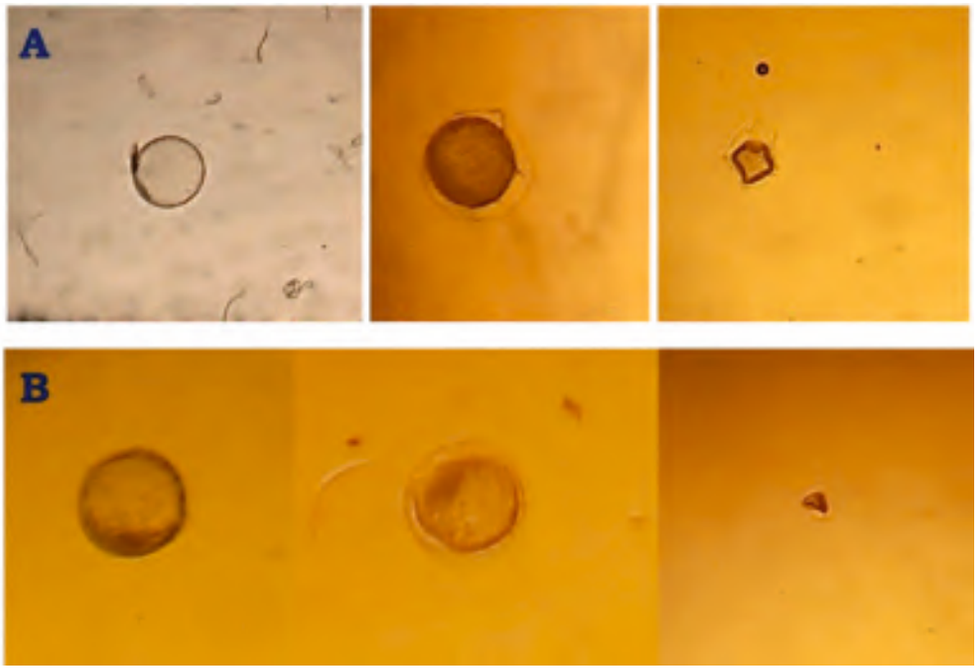
(TR Talluri, Sajjan Kumar, Yash Pal, RA Legha, RK Dedar & TK Bhattacharya)

e. Vitrification of horse embryos

Vitrification is a rapid freezing process. Horse embryo has large volume of fluid and the capsule make it difficult for cryoprotectants to penetrate the inner cell mass in sufficient quantity to prevent chilling injury during cryopreservation. The capsule that is present in larger embryos may impede the intracellular diffusion of cryoprotectants. Vitrification of embryos >300 mm in diameter requires puncture of the glycoprotein capsule, although the size of the hole compatible with embryo survival is unknown. A successful and easy method for embryo vitrification has recently been established at EPC, ICARNRCE. In this technique, embryos were not punctured but modified the vitrification media and timing of exposure to each media before vitrification. This gave better results of vitrification of large sized embryos of 500-700μm.The embryos recovered after the flushing of the mares were initially washed in the embryo holding media containing 2% DMSO for 5 mins. Then the embryo is shifted to the Media A containing the MOPS media for 2 min. Then the embryos are washed in Media B which will be having DMSO and Ethylene Glycol and other antioxidants for 12 mins during which the blastocoel gets shrinkage and the capsule is loosened. Late the embryos are shifted to Media C where the increased concentrations of Sucrose, EG and DMSO are present. The embryo was exposed for brief time of 30-40 sec and then shifted to cryodevices to vitrify in the LN2.In the current year we have successfully vitrified 15 Marwari horse embryos, 2 Zanskari and one Manipuri horse embryos.



Outline of vitrification process of horse embryos



Realtime images of horse embryos undergoing vitrification process

(TR Talluri, Sajjan Kumar, Yash Pal, RA Legha, RK Dedar & TK Bhattacharya)

Breed Recognition

The Bhimthadi (Deccani) breed of horse has been characterized. The animals of the breed were located with the Nomadic Tribe - C of the state and distribution of the breed was found in the area between Bhima and Nira River of the Maharashtra state. The breeding tract encompassed Pune, Ahmednagar, Satara, Sangli, Solapur and Kolhapur districts with an area of about 74329 Km^2 between 15.7469º N to 19.9897º N and 76.414º E to 73.3249º N and the population of the breed has been estimated to be 5134 heads in the year 2023. The breed has now been recognized in the 11th meeting Breed Registration Committee held on December 5, 2023, under the chairmanship of DDG(AS).

(S C Mehta & Sachin D Sorate)

V. Patents/ copy right filed/ granted

a. Patents Filed

S.No.	Title of Patent	Name of the Inventors	Application number details	Institutes
1.	miR-29a serves as a novel immunovirological marker to predict the functionality of immune response to Lumpy Skin Disease virus infection.	Naveen Kumar, Ram Kumar, Sanjay Barua	202311063758, dated 22.09.2023	NRCE, Hisar
2.	Chitosan-Alginate Zinc Artificial Skin Construct	Anju Manuja & Team	202311086503, dated 18.12.2023 (Provisional)	NRCE, Hisar
3.	Development of a Novel test to differentiate the vaccine and field strains of LSDV.	Naveen Kumar, Sanjay Barua, Ram Kumar, Bhupendra N. Tripathi	202211074538, dated 22.12.2022	NRCE, Hisar
4.	Synthetic peptides and antibodies targeted to bovine Mx2 protein	Varij Nayan, Anuradha Bhardwaj *et al*	202311042605, dated 26.06.2023	CIRB, Hisar

VI. MoU for Cooperation in Research and Education

The ICAR-NRCE, Hisar inked the Memorandum of Understanding with various reputed and prestigious academic and research oriented institutes namely Central University of Haryana, Mahendergarh, National Institute of Pharmaceutical Education and Research NIPER, SAS Nagear Mohali, Punjab, Kamdhenu University, Gandhinagar, Gugarat, Kerala Veterinary and Animal Sciences University (KVASU), Wayanad, Kerala, Chaudhary Sarwan Kumar Himachal Pradesh Krishi Vishvavidyalaya, Palampur, Himachal Pradesh, Assam Agrieultural University, Head Quarter Jorhat, Tamil Nadu Veterinary and Animal Sciences University, Chennai. For the cooperation in the areas of Research and Education. Dr. T.K. Bhattacharya, Director, ICAR-NRCE, Hisar and Vice Chancellors/Directors of the Universities/Institutes signed the Memoranda. With the MoU, the NRCE and Universities have agreed for collaborative programmes in the fields of research, education, training and capacity building, extension consultancy and other areas of national interest. Both

the partners have also agreed for mutually recognizing the faculty of both the Institutes for the research and teaching purposes, wherein, the students and faculties can carry out the specific, research and outreach activities at the laboratories of these institutions.

S.No.	Name of the institute under MoU	Date of MoU
1	Central University of Haryana, Mahendergarh.	01.06.2023
2	NIPER, SAS Nagear Mohali, Punjab.	12.10.23
3	Kamdhenu University, Gandhinagar, Gugarat.	26.06.23
4	Kerala Veterinary and Animal Sciences University (KVASU), Wayanad, Kerala.	03.08.23
5	Chaudhary Sarwan Kumar Himachal Pradesh Krishi Vishvavidyalaya, Palampur, Himachal Pradesh.	05.08.23
6	Assam Agrieultural university, Head Quarter Jorhat.	05.12.23
7	TANUVAS, Chennai.	09.12.23

An MoU/MoA was made with Shri Raghavendra Singh Dundlod, Dundlod, Maharashtra on two technologies of Semen collection and cryopreservation in Equines and Artificial Vagina for semen collection in Stallions. These technologies were transferred to Shri Raghavendra Singh during 13th March 2023

(Team: TR Talluri, Yash Pal, SK Ravi & RA Legha)

VIII. Commercialisation and Revenue Generation from the Sale and Service of NRCE Technologies

S.No.	Year	Name of the technology transferred	Name of the Licensee/party	Technology Fee (INR in lakhs)	Date of Licensing
1	2023	Lumpy Skin Disease Vaccine	Hester Biosciences Limited, Ahmedabad	75.0	06.03.2023
2	2023	Semen collection and cryopreservation in Indigenous horses	Shri Raghuvendra Singh Dundlod, Dundlod, Jhunjhunu, Rajasthan	0.5	13.03.2023
3	2023	Customised AV for semen collection from stallions	Shri Raghuvendra Singh Dundlod, Dundlod, Jhunjhunu, Rajasthan	0.177	13.03.2023
4	2023	Customised AV for semen collection from stallions	Dr Balender Goswami, Hanumangarh, Rajasthan	0.177	04.04.2023

Education and
Trainings

Trainings Organized

Training to the field veterinary officer of Rajasthan state under RKVY project

The following trainings were imparted to the Veterinary officers of the Rajasthan State Animal Husbandry Department under the RKVY project titled "*Conservation Marwari (Malani) indigenous breed of horse through I*". A total of 6 training programmes were conducted successfully and 68 veterinary officers were trained in artificial insemination and pregnancy diagnosis in mares.

Name of the Training : Conservation Marwari (Malani) indigenous breed of horse through AI : RKVY

Participants from : Veterinary Officers of Rajasthan State Animal Husbandry Department

Durations : 3 days during the year 2023

Dates/ Duration	Training Details (2023)						Total Participants
	Jan. 23-25	Jan. 30 Feb. 1	Feb. 13-15	Feb. 20-22	Feb. 27 March 1	March 13-15	
No. of Participants	10	9	13	11	13	12	**68**

One week training programme to the field veterinarians

A one-week training programme on "*Equine Husbandry practices and issues involving equine reproduction and their management practices*" was conducted from March 1 - 7, 2023 at Equine Production Campus, ICAR-NRCE, Bikaner. In this training programme, a total of 15 field veterinarians from Rajasthan State Animal Husbandry Department participated. In this training programme, the veterinarians were trained in the areas of follicular examination through Ultrasonography Artificial insemination and Pregnancy diagnosis in Equines.

Training of Field Veterinary Officer's on Glanders awareness

Mass awareness of veterinary officers and equine stake holders on glanders disease is one of the instrumental factors for effective implementation of physical, clinical and serological surveillance, control and containment of glanders outbreak. In 2023, five days training programme were organized at NRCE, Hisar on "Surveillance and Laboratory Diagnosis of Equine Glanders" from 18-22 September 2023. A total of 10 Veterinary Officers from Tamil Nadu state participated in the training programs.

Entrepreneurship development programme on donkey farming

Two training programmes on Entrepreneurship development programme on Donkey farming was organised from April 24th -28th, 2023 and 25-27th August, 2023 at ICARNRC on Equines, Regional Station, Equine Production Campus, Bikaner. For this training programme 23 participants have enrolled and participated. The participation represented from 5 states namely, Andhra Pradesh, Telanga, Karnataka, Maharashtra and West Bengal. Various lectures on Donkey genetics, reproduction, breeding, health and management were taken and practical classes on Artificial insemination, pregnancy diagnosis and milking of donkeys etc were also conducted.

NRCE Scientist and Technician Visited Nepal for conducting training on glanders diagnosis

In continuation of the invitation received from Nepal Veterinary Association and subsequent approval of the DARE, Ministry of Agriculture and Farmers' Welfare vide F. No. 012570-ADHOC Workshop -IC-I dtd 17.02.2023 Dr Harisankar Singha Sr. Scientist and Mr. Sita Ram, ACTO visited Nepal from 20-24 February. They have conducted 5 day's training programme held at Central Veterinary Laboratory, Tripureshwar, Kathmandu. A total of 12 Veterinary Officers participated in the training programme. There was an introductory lecture on glanders and its diagnostics principle followed by hands-on training on glanders diagnosis.

Training programmes conducted under National One Health Programme for Prevention and Control of Zoonoses (NOHP-PCZ)

ICAR-National Research Centre on Equines (NRCE), Hisar is one of the Regional Coordinators under "National One Health Program for Prevention and Control of Zoonotic Diseases (NOHPCZ)" funded by National Centre for Disease Control, Ministry of Health and Family Welfare, Govt. of India. In this context, NRCE is actively involved in capacity building and strengthening of laboratory diagnostic facilities and intersectoral coordination and creating awareness about zoonotic diseases of public health importance by organizing training, workshop and webinar on various zoonotic diseases. In the year 2023, four workshops and two training were conducted as mentioned in the below table.

Sr. No.	Workshops	Date	Participants
1	One day workshop on Zoonotic diseases organized at Kurukshetra, Haryana	20.01.2023	72 participants from medical department, animal husbandry department and framers of Kurukshetra districts participated in the workshop.
2	One day workshop on "Zoonotic diseases:Prevention & Control" atSonipat, Haryana	24.01.2023	A total of 89 participants from Medical & Veterinary (n=49), Farmers (n=40)
3	One-day workshop on Zoonotic diseases organized at Sirsa,Haryana	16.03.2023	84 participants from medical department, animal husbandry department and framers of Kurukshetra districts participated in the workshop.
4	Rabies week is celebrated in collaboration with LUVAS, Civil hospital, Hisar.	27.09.2023 to 06.10.2023	More than 1000 participants including veterinary, medical professional, students, farmers and general public

Sr. No.	Trainings	Date	Participants
1	Diagnosis of Zoonotic Bacterial and Viral Infections was organized at ICAR-NRCE.	31.01.2023 to 03.02. 2023	33 participants from human health sectors and animal husbandry departments and wildlife sectors ofHaryana and Uttar Pradesh
2	Diagnosis of ImportantBacterial, Parasitic and Viral Zoonotic Diseases was organized at ICAR-NRCE.	01.03.2023 to 03.03.2023	20 participants from human health sectors and animal husbandry departments and wildlife sectors of Himachal Pradesh and New Delhi

Training Programme on "Diagnosis of Zoonotic Bacterial and Viral Infections was organized at ICAR-NRCE."

A four days training program on 'Diagnosis of Zoonotic Bacterial and Viral Infections' has been organized at ICAR-NRCE from 31-01-2023 to 03-02-2023. A total of 27 Medical and Veterinary Professionals from Uttar Pradesh and 6 technical staff from ICAR-NRCE, Hisar has attended this training. The goal of this training was to strengthen the knowledge of medical and veterinary professionals about Zoonotic diseases diagnosis and capacity building to carry out surveillance and timely reporting of zoonotic diseases. Hands on training were also imparted on laboratory diagnosis of glanders, rabies, Japanese encephalitis, tuberculosis, paratuberculosis and brucellosis.

Training Programme on "Diagnosis of Important Bacterial, Parasitic and Viral Zoonotic Diseases was organized at ICAR-NRCE."

A three days training program on 'Diagnosis of Important Bacterial, Parasitic and Viral Zoonotic Diseases' has been organized at ICAR-NRCE from 01.03.2023 to 03.03.2023. A total of 19 medical and veterinary professionals from Himachal Pradesh & New Delhi and 1 veterinary staff from ICAR-NRCE, Hisar had attended this training. The lectures and practical on the diagnosis of zoonotic bacterial, parasitic and viral diseases like glanders, leptospirosis, tuberculosis, pox virus, Japanese encephalitis and important zoonotic parasitic infection had been included in the course to update the participants about these important zoonotic diseases.

IEC Leaflet on important Zoonotic diseases on Rabies, Anthrax & Glanders

Trainings/ Workshops/Webinars/Meetings (National and International) attended by the Scientists

S.No.	Name of Staff	Name of training	Organizing institute	Period	Duration
1.	Ana Raj J	Leveraging Extension Strategies for Sustainable development of Allied Agri Sector Enterprises	MANAGE, Hyderabad & ICARNDRI, Karnal	August 16 – 18, 2023	3 days
2.	Dr Anju Manuja	International conference on "Prospects and challenges of environment and biological sciences in food production system for livelihood security of farmers (ICFPLS-2023) Strengthening Veterinary Profession towards One Health through Diversity, Equity and Inclusiveness	Pragati International Scientific Research foundation (PISRF) Merrut& Andaman Science Association (ASA), Port Blair, Andaman & Nicobar, India	18-20 Sept., 2023	3 days
		XVII National Technical Conference of Indian Association of Women Veterinarians on Strengthening Veterinary Profession towards One Health through Diversity, Equity and Inclusiveness	NTR College of Veterinary Science, Gannavaram, Sir Venkateswara Veterinary University, A.P., India	29-30 Nov., 2023.	2 days
		International conference on "Nanotechnology Addressing the convergence of material science, Biotechnology, and Medical Science" (IC NACMBM -2024)	Centre for Interdisciplinary research (CIR), D.Y. Patil Education Society, Kolhapur, Maharashtra, India	12- 14 Feb. 2024	3 days

S.No.	Name of Staff	Name of training	Organizing institute	Period	Duration
3.	Dr. Balvinder Kumar	International conference on "Prospects and challenges of environment and biological sciences in food production system for livelihood security of farmers (ICFPLS-2023)	Pragati International Scientific Research foundation (PISRF) Merrut & Andaman Science Association (ASA), Port Blair, Andaman & Nicobar, India	18-20 Sept., 2023	--
4.	Dr. Balvinder Kumar	International Virtual Conference on Streptococci & Streptococcal Infections	United Scientific Group	27-28 Sept., 2023	2 days
5.	Dr BC Bera	Participated in VIROCON – 2023, on "Advancements In Global Virus Research Towards One Health"	Indian Virological Society at ICAR-NRC on Banana, Tiruchirapally, Tamil Nadu	01.12.2023 to 03.12.2023	3 days
6.	Dr Sanjay Kumar	29th International Conference of the World Association for the Advancement of Veterinary Parasitology	WAAVP	20-24 August, 2023	5 days
7.	Dr HS Singha	XXXV National Conference of Indian Association of Veterinary Microbiologists, Immunologists and Specialists in Infectious Diseases on "Novel Approaches in Animal Health for Realizing One Health Mission"	CSK HPKV, Palampur	7- 8 April, 2023	2 days
		"Uniting for One Health"	NCDC, New Delhi in collaboration with DAHD, MoEFCC, MoAFW, ICAR, and USAID RISE, Jhpiego	6 & 7July, 2023	2 days
		Multi-stake holder Consultation workshop to develop the State Action Plan for the Prevention and Control of Zoonotic Diseases for the state of Rajasthan	NCDC, New Delhi and DMHS,Rajasthan in technical collaboration with U.S. Centers for Disease Control and Prevention (CDC)	12-13 Oct., 2023	2 days
		Laboratory Quality Management System and Internal Audit as per ISO/IEC 17025:2017	Infinity Consultancy, Bhopal.	19-22 July, 2023	4 days
		NABL assessor training programme	IVRI, Izatnagar	21- 25 August, 2023.	5 days
8.	Dr Naveen Kumar	Lead Speaker on "Vaccination against Lumpy Skin Disease". Current state of the art. In, Veterinary Pathology Congress-2023 (IAVP),	IVRI, Izatnagar	20-22 Dec., 2023	3 days

S.No.	Name of Staff	Name of training	Organizing institute	Period	Duration
9.		Lead Speaker on "Vaccination against Lumpy Skin Disease". Current state of the art. In, VII Annual Convention of SVBBI and International Symposium on Multiomics to One Health: Challenges and Way Forward in Biomedical Research. Annual Conference of Society of Veterinary Biochemists and Biotechnologists of India (SVBBI)	IVRI, Izatnagar	14-15 Dec., 2023	2 days
		Lead paper on Lumpi-ProVacInd: Development of LSD vaccine in India. In 35th IAVMI conference on-Novel approaches on Animal Health for reralizing one health mission"	DGCN College of Veterinary and Animal Sciences, CSK HPKV, Palampur (HP)	7-8 April, 2023	2 days
		Invited to deliver a lecture on "Vaccination against Lumpy skin disease in India" in a symposium organized by ICAR-National Institute for Veterinary Epidemiology and Disease Informatics (NIVEDI)	National Institute for Veterinary Epidemiology and Disease Informatics (NIVEDI)	27 Jan., 2022	1 day
10.	Dr Sanjay Barua	Lead paper on "p38 Mitogen-Activated Protein Kinase (MAPKs) as a target for antiviral drug development" In 35th Annual Conference of IAVMI on the theme "Novel Approaches in Animal Health for Realizing One Health Mission"	DGCN College of Veterinary and Animal Sciences, CSK HPKV, Palampur (HP)	7-8 April, 2023	2 days
11.	Dr RK Vaid	Workshop on 'Anthrax outbreak response and sample collection'	Odisha University of Agriculture & technology, Bhubaneshwar, Odisha under DBT Network Programme on Anthrax Diagnosis and Control in India, DBT at Deptt. of Preventive Medicine, College of Veterinary Science and Animal Husbandry, OUAT, Bhubaneshwar	2-4 March, 2023	3 days
		BIMSTEC Workshop 'Epidemiological approaches to prevent and control trans-boundary animal diseases with special focus on zoonotic diseases and FMD".	Nominee of Animal Science Division, ICAR, organized by ICAR-NIVEDI and ICARNIFMD, Bhubaneshwar.	03 Oct. -12 Oct., 2023	10 days

S.No.	Name of Staff	Name of training	Organizing institute	Period	Duration
12.	Dr Shanmugasundaram K	Conference: XXXV Annual Convention and National Conference of Indian Association of Veterinary Microbiologists, Immunologists and Specialists in Infectious Diseases on Novel Approaches in Animal Health for Realizing One Health Mission	Department of Veterinary Microbiology Dr. G.C. Negi College of Veterinary and Animal Sciences, CSKHPKV, Palampur, (HP)-176062, India	April 7-8, 2023	2 days
		NABL Assessor Training	ICAR-IVRI, Izatnagar	21 to 25 Aug., 2023	5 days
		National Conclave "Uniting for One Health	National Centre for Disease Control (NCDC), Delhi	6 & 7 July, 2023	2 days
		Laboratory quality management system and internal audit as per IS/ISO/IEC 17025:2017 (Online)	Infinity Consultancy, Flat No. 401 Sunshine Appartment,	19-22 July, 2023	4 days
13.	Dr SC Mehta	National Conference on Advances in Genetics and Genomics for Sustainable Livestock Transformation & XVII Annual Convention of Indian Society of Animal Genetics & Breeding	ICAR-NBAGR, Karnal	16-17 Nov., 2023	2 days
14.	Dr TR Talluri	XVI AGRICULTURAL SCIENCE CONGRESS & ASC EXPO 2023	ICAR-Central Marine Fisheries Research Institute (CMFRI), Kochi	10-13 Oct., 2023	4 days
		International symposium "Frontiers in Theriogenology: Research and Practice	College of Veterinary and Animal Sciences, Mannuthy, Thrissur, Kerala	6-8 Dec., 2023	3 days
15.	Dr TK Bhattacharjya	EDP on leadership development	NAARM, Hyderabad	21-26 Aug., 2023	6 days

Post Graduate Students' Research and Guidance

Sr. No.	Name of the Student	Name of the Guide	Project/Dissertation Title	Completed/ ongoing
Postdoc Fellows				
1	Dr Shalini Sharma (LUVAS, Hisar)	Dr Naveen Kumar	Prevalence and molecular characterization of *Mycobacterium avium* subspecies Paratuberculosis (MAP) and evaluation of potential role of Th1/Th17 axis in development of Johne's disease in diarrhoeic buffaloes	Ongoing
2	Dr Ruma Rani (DST-SERB NPDF)	Dr Rajender Kumar	Development of recombinant antibody-based nano-diagnostic lateral flow assay for rapid detection of *Trypanosoma evansi* infection at Point-of-Care.	Ongoing

Sr. No.	Name of the Student	Name of the Guide	Project/Dissertation Title	Completed/ ongoing
PhD Students				
1	Raja Kumar (CCS HAU Hisar)	Dr Naveen Kumar	Studies on the epitranscriptomic regulation of Foot-and-mouth disease virus	Ongoing
2	Garvit Kumar (GLA University Mathura)	Dr Naveen Kumar	Studies on the role of role of PPAR-γ in buffalopox virus replication	Ongoing
3	Dr Diksha (LUVAS, Hisar)	Dr Nitin Virmani	Genetic targeting of equine herpes virus 1 as a vector for developing vaccine and to explore its potential as a vaccine vector for targeting infectious bursal disease virus in poultry	Ongoing
4	Yamini (AMITY Univ. NOIDA)	Dr Nitin Virmani	Development of a novel deletion mutant of EHV1 to generate MLV vaccine candidate	Ongoing
5	Swati (GJU&ST, Hisar)	Dr Nitin Virmani	Studies on development of recombinant porcine circovirus vaccine candidate(s) & their immunological response in murine model / piglets	Ongoing
6	Anubala Jaglan (CCS HAU, Hisar)	Dr Taruna Anand	Exploring bacteriophage derived endolysins for targeted delivery into biofilm forming Bacteria	Completed
7	Assim Verma (GJU&ST Hisar)	Dr Sanjay Barua	Studies on the mechanisms underlying antiviral action of Hesperetin against buffalopoxvirus	Ongoing
8	Yogesh Chander (GJU&ST Hisar)	Dr Sanjay Barua	Role of p38 Map Kinase in Buffalopox virus replication	Completed
9	Indu Rani (CCS HAU, Hisar)	Dr Shanmugasundaram K	Targeted genome editing using CRISPR-Cas9 approach to decipher the functional role of predicted genes in survival of *Mycobacterium kansasii*	Ongoing
10	Swati Rani (GJU&ST Hisar)	Dr Anju Manuja	Synthesis, characterization and biological activity of chloroquinine derivatives, their complexes and nanocomposites	Ongoing
11	Dharvi Chhabra (LPU Phagwara)	Dr Balvinder Kumar	Epidemiological and molecular investigations of strangles in equine population of northern India	Ongoing
12	Mamta Tirdia (CBLU, Bhiwani)	Dr Sanjay Kumar	Anti-theilerial and anti-plasmodial activities of herbal based selected lead drug molecules and elucidation of their targets	Ongoing
13	Dr Diksha Sharma (LUVAS, Hisar)	Dr Rajender Kumar	In vitro and in vivo evaluation of chemotherapeutic potential of alkaloids compounds against *Trypanosoma evansi*.	Ongoing
14	Dr Pyarelal (RAJUVAS, Bikaner)	Dr TR Talluri	Study on development of alternative semen extender for stallion semen preservation	Ongoing
15	Sonali (Maharaja Agarsen University, Baddi, Solan)	Dr Anuradha Bhardwaj	Comparative genomic studies on horse and donkey performance genes	Completed
16	Renu Garhwal (Amity University, Jaipur)	Dr Anuradha Bhardwaj	Characterization of physicochemical qualities of donkey milk and its utilization in value added dairy products	Ongoing

Sr. No.	Name of the Student	Name of the Guide	Project/Dissertation Title	Completed/ ongoing
MVSc/MSc Students				
1	Himanshu Kamboj (Amity University Noida)	Dr Naveen Kumar	Studies on the role of Histone deacetylase in lumpy skin disease virus replication	Ongoing
2	Sakshi Pandita (LUVAS Hisar)	Dr Naveen Kumar	Studies on the miRNA response to lumpy skin disease virus infection	Completed
3	Karan	Dr Nitin Virmani	Deletion of Thymidine Kinase gene from mutant *Equine herpesvirus 1* using BAC technology	Completed
4	Dr Vishal Yadav (RAJUVAS, Bikaner)	Dr TR Talluri	Effect of addition of *Spirulina platensis* extract to semen extender on cooled and post-thaw semen quality of Marwari stallion	Completed
5	Naina Paswan (BASU, Patna)	Dr TR Talluri	Effect of melatonin supplementation to the semen extender on cryopreserved stallion semen parameters	Completed
6	Dr Saurabh Daria (RAJUVAS, Bikaner)	Dr TR Talluri	Studies on ultrasonographic, hematological and immunological markers changes during oestrus cycle in mares	Completed
7	Dr Kalpna Godara (RAJUVAS, Bikaner)	Dr RK Dedar	Gene expression studies of clinically important cytokines in summer dermatitis in horses'	Completed

Lead Papers/Invited Papers/Invited Expert Lectures

1. **Dr Anju Manuja** delivered lead lecture in international conference on "Prospects and challenges of environment and biological sciences in food production system for livelihood security of farmers (ICFPLS-2023)" was organized by Pragati International Scientific Research foundation (PISRF) Meerut & Andaman Science Association (ASA), Port Blair, Andaman & Nicobar, India from 18th-20th Sept., 2023.
2. **Dr Anju Manuja** was invited to deliver lecture in a training on "Application of nanotechnology in agriculture: Opportunities and Challenges" under NAHEP-IDP Project at SKUAST Jammu. *(online)*
3. **Dr Anju Manuja** was invited to deliver lecture in XVII National Technical Conference of Indian Association of Women Veterinarians on Strengthening Veterinary Profession towards One Health through Diversity, Equity and Inclusiveness was organized by NTR College of Veterinary Science, Gannavaram, Sir Venkateswara Veterinary University, A.P., India from 29th-30th Nov., 2023.
4. **Dr Anju Manuja** was invited to deliver lecture online "Nanoparticles, their delivery and evaluation for biomedical applications in animals" in Karyashala Workshop on "Advanced Techniques of Preparation, Characterization and Evaluation of Nano formulations for Biomedical Applications" from 16th-25th Jan., 2023 at IVRI.
5. **Dr Balvinder Kumar** was invited to deliver a talk on "Equine Infectious Diseases in India: An Overview", In 2nd International Conference on Prospects and challenges of environment and biological sciences in food production system for livelihood security of farmers (ICFPLS–2023) at ICAR-CIARI, Port Blair, Andaman & Nicobar Islands, India 18th-20th Sep., 2023 Organized by Pragati International Scientific Research Foundation (PISRF), Meerut, India & Andaman Science Association (ASA), Port Blair, A & N Islands, India.
6. **Dr Naveen Kumar** was delivered lead talk on Lumpi-ProVac^{Ind}: Development of LSD vaccine in India. In 35th IAVMI conference on-Novel approaches on Animal Health for realizing one health mission. Held on 7th-8th April, 2023, at DGCN College of Veterinary and Animal Sciences, CSK HPKV, Palampur (HP).
7. **Dr Naveen Kumar** was invited to deliver expert lecture on "Coronavirus (SARS-CoV-2) infections in animals." In 21 Days, Winter School on One Health Approach to Combat AMR, Zoonoses and Food Safety, Organized by Rajasthan University of Veterinary and Animal Sciences, Jaipur, Rajasthan from 1st-21stDec., 2023.

8. **Dr Naveen Kumar** was invited to deliver expert lecture on "CRISPR-cas9-mediated genome editing". In ten days, Training Programme on "Basic Molecular Tools in Animal Biotechnology" from 15th-24th Nov., 2023, at under NAHEP-IDP, SKUAST-Jammu.

9. **Dr Naveen Kumar** was invited to deliver expert lecture on "Detection and management of viral co-infections" In SERB/DST sponsored 10 days training program, organized by IVRI Izatnagar from 17th-26th July, 2023

10. **Dr Naveen Kumar** was invited to deliver expert lecture on "Good laboratory practices." In, Student enterpreneurship training under institutional development plan at GADVASU, Ludhiana, on 4th Dec., 2023.

11. **Dr Naveen Kumar** was invited to deliver expert lecture on "Hands on training on application of modern molecular diagnostic techniques." In, Student enterpreneurship training under institutional development plan at GADVASU, Ludhiana, on 4th Dec., 2023.

12. **Dr Naveen Kumar** was invited to deliver expert lecture on "Hands on RT-PCR molecular technique (LSD) and Production SOPs and testing of lumpy skin disease vaccine in laboratory." In Hands on Training on Good Manufacturing Practices in veterinary vaccines for the veterinary officers of IVBP/DIS, Organized by Training and Education Centre, Pune, in collaboration with Biological Standardization Division, Izatnagar, ICAR Indian Veterinary Research Institute.

13. **Dr Naveen Kumar** was invited to deliver expert lecture on "Pathogenic coronaviruses and their emerging zoonotic potential with emphasis on SARS-CoV-2 infection in animals." In BIMSTEC (The Bay of Bengal Initiative for Multi-Sectoral Technical and Economic Cooperation, an international organisation of seven South Asian and Southeast Asian nations) training on Recent Advances in Diagnosis and Control of Emerging and Re-emerging Zoonoses is being organized virtually by IVRI from 25th-30th Sep., 2023.

14. **Dr Naveen Kumar** was invited to deliver expert lecture on "Sharing experience on development of Lumpy Skin Disease vaccine." In, one day seminar on Sharing experience on development of Lumpy Skin Disease vaccine and role of IPR in enterpreneurship, Organized by LUVAS, Hisar on 14th Nov., 2023.

15. **Dr Naveen Kumar** was invited to deliver expert lecture on "Vaccination against Lumpy Skin Disease in India: SWOT Analysis." In 21 Days, Winter School on One Health Approach to Combat AMR, Zoonoses and Food Safety, Organized by Rajasthan University of Veterinary and Animal Sciences, Jaipur, Rajasthan from 1st-21st Dec., 2023.

16. **Dr Naveen Kumar** was invited to deliver expert lecture on "Vaccination against Lumpy skin disease in India" in a symposium organized by ICAR-National Institute for Veterinary Epidemiology and Disease Informatics (NIVEDI) in collaboration of Indian Virological Society on 27th Jan., 2022.

17. **Dr Naveen Kumar** was invited to deliver expert lecture on "Vaccine manufacturing principles and processes of development." In, Student enterpreneurship training under institutional development plan at GADVASU, Ludhiana, on 5th Dec., 2023.

18. **Dr Naveen Kumar** was invited to deliver lead talk on "Vaccination against Lumpy Skin Disease." Current state of the art. In, Veterinary Pathology Congress-2023 (IAVP), Organized by IVRI, Izatnagar from 20th-22nd Dec., 2023.

19. **Dr Naveen Kumar** was invited to deliver lead talk on "Vaccination against Lumpy Skin Disease." Current state of the art. In, VII Annual Convention of SVBBI and International Symposium on Multiomics to One Health: Challenges and Way Forward in Biomedical Research. Annual Conference of Society of Veterinary Biochemists and Biotechnologists of India (SVBBI), Organized by IVRI Izatnagar from 14th-15th Dec., 2023.

20. **Dr Nitin Virmani** delivered an expert lecture on Development of recombinant vaccine candidate against EHV1 and equine influenza In VIROCON-2023 on "Advancements in Global Virus Research towards One Health" organized by Indian Virology Society and held at ICAR-NRCB, Trichy on 01st-03rd Dec., 2023.

21. **Dr Nitin Virmani** delivered an expert lecture on Development of recombinant virusbased vaccines against respiratory viral diseases of equines in VIBCON-2022 on "Leveraging emerging biotechnologies for one health" organized by Indian Society for Veterinary Immunology and Biotechnlogy and held at SKUAST,

Srinagar, Kashmir on 29th -31st July, 2023.

22. **Dr Nitin Virmani** was invited to deliver lecture on respiratory diseases of equines and modern vaccine approaches. In Veterinary Pathology Congress-2023 on "Advances in Veterinary Pathology for Diagnosis and Control of Emerging Diseases of Livestock and Poultry" held at ICAR-IVRI, Izatnagar, U.P on 20th-22nd, Dec., 2023.
23. **Dr Rajender Kumar** Presented a research paper entitled "multi-recombinant antigensbased ELISA for diagnosis of surra in equines" in 29th International Conference of the World Association for the Advancement of Veterinary Parasitology (WAAVP) held at Chennai during 20th - 24th Aug., 2023.
24. **Dr RK Vaid** presented a lead paper titled Bacteriophages for *Aeromonas veronii* control: isolation, characterization and potential application in aquaculture pathogen management' in the VIROCON 2023 "Advancements in Global Virus research towards One Health" organised by NRC on Banana, Tiruchirapalli and held at Tiruchirapalli, Tamil Nadu from 1st-3rd, Dec., 2023.
25. **Dr RK Vaid** presented annual review progress report of NRCE, Hisar for Surveillance of AMR in Haryana held at 6th Annual Review meet of "Indian Network for fisheries and animals' antimicrobial resistance (INFAAR)" at Bhuwaneshwar, Odisha on 23rd Dec., 2023.
26. **Dr RK Vaid** presented ATR and annual review progress report of NCVTC, XIIth Annual Review Meet as on 9th Jan., 2023 at Committee Room, NRCE, Hisar NRCE, Hisar.
27. **Dr Sanjay Barua** delivered an Invited lecture as a Resource person in BIMSTEC Training (Bay of Bengal Initiative for multi sectoral, Technical and Economic Cooperation) Workshop on "Recent Advances in Diagnosis and Control of Emerging and Reemerging Zoonoses" jointly organized by IVRI, NRCE and NIHSAD from 25th - 30th Sept., 2023 on the topic "Recent advances in the diagnosis and control of poxvirus zoonosis" on 27th Sept., 2023.
28. **Dr Sanjay Barua** was invited to deliver lead lecture at BIMSTEC Training (Bay of Bengal Initiative for multi sectoral, Technical and Economic Cooperation) Workshop on "Laboratory Biosafety and Biosecurity" jointly organized by NIHSAD and NRCE from (10th- 14th July, 2023) on the topic "Waste management of the biorisk materials and biomedical waste management regulations" on 12th July, 2023.
29. **Dr Sanjay Barua** was invited to present a lead paper entitled "p38 Mitogen-Activated Protein Kinase (MAPKs) as a target for antiviral drug development." In Recent advances in the diagnosis and control of poxvirus zoonosis in the 35th IAVMI conference on Novel approaches on Animal Health for realizing one health mission. Held on 7th-8th April, 2023, at DGCN College of Veterinary and Animal Sciences, CSK HPKV, Palampur (HP).
30. **Dr Shanmugasundaram K** delivered an expert lecture on "Bovine Tuberculosis" in the public Awareness Workshop on Zoonotic Diseases organized by ICAR-NRCE in collaboration with Haryana Health Department and Animal Husbandry Department on 20th Jan., 2023 at UPHC, Krishna Nagar Gamri, Kurukshetra; on 24th Jan., 2023 at DCRUST University, Murthal, Sonipat; on 16th Mar., 2023 at Surkhab Complex, Sirsa.
31. **Dr Shanmugasundaram K** delivered an expert lecture on "Diagnosis of glanders: serological and molecular methods" during training programme for Field Veterinary Officers' on Hands on Training Programme on Diagnosis and Control of Equine Glanders organised at ICAR-NRCE, Hisar, 18th-22nd Sept., 2023.
32. **Dr Shanmugasundaram K** delivered an expert lecture on "Sample collection, processing and dispatch for glanders diagnosis" during training programme for Field Veterinary Officers' Hands on Training Programme on Diagnosis and Control of Equine Glanders organised at ICAR-NRCE, Hisar, 18th-22nd Sept., 2023.
33. **Dr Shanmugasundaram K** delivered an expert lecture on Sample collection, packing and dispatch for laboratory diagnosis of important Zoonotic diseases in the training Programme on Approaches for Diagnosis of Zoonotic Bacterial Infections and viral infections organised by ICAR NRCE on 31st Jan., to 03rd Feb., 2023.
34. **Dr Shanmugasundaram** K delivered an expert lecture on Zoonotic Mycobacterial infections of animals, their diagnosis and control in animals in the training Programme on Approaches for Diagnosis of Zoonotic Bacterial and viral Infections organised by ICAR NRCE on 31st Jan., to 03rd Feb., 2023.
35. **Dr Taruna Anand** was invited to deliver expert lecture on "Bacteriophage therapy for mitigating AMR:

Challenges and way forward" during 21 days Winter School on the topic Antimicrobial peptides (AMPs) as an alternative to antibiotics: Hands on training for designing, chemical synthesis, characterization and applications of synthetic AMPs" organized at the Facility for Research & Training on Bioassays and Biosensors, Division of Veterinary Biotechnology, IVRI from 24th Jan., to 13th Feb., 2023.

36. **Dr Taruna Anand** was invited to deliver expert lecture on "Bacteriophage Therapy in veterinary science and phage repository" during Hands on training on Basics of Bacteriophage Biology and Clinical Applications to Combat the Emerging Antimicrobial Resistance from 3rd-7th Oct., 2023, organized by VRDL, Dept of Microbiology, Banaras Hindu University, Varanasi, India.

37. **Dr Taruna Anand** was invited to deliver expert lecture on "Phage therapy in veterinary sector and beyond to tackle emerging antibiotic resistance" for 21 days Winter School on "Molecular Diagnosis of AMR (Anti-Microbial Resistant) pathogen causing Mastitis in Cattle and Buffalo" at Animal Biotechnology Centre, National Dairy Research Institute Karnal from 23rd Feb., to 15th Mar., 2023.

38. **Dr Taruna Anand** was invited to deliver expert lecture on "Phage Threapy to modulate MDR infections in Veterinary science" during 4th International Conference on Bacteriophage Research and antimicrobial resistance from 28th-30th Sept., 2023 organized at Madras University.

39. **Dr Taruna Anand** was invited to deliver lead lecture on "Bacteriophage Therapy in lab animal models to explore the prevention of infections of poultry, aquaculture and veterinary industry", during VIROCON, 2023, Advancements in Global Virus Research Towards One Health organized from 1st-3rd Dec., 2023 in Tiruchirappalli, Tamil Nadu.

40. **Dr TR Talluri** was assigned the duties of teaching and training of handling the equine frozen semen and protocols for deep horn insemination in mares to the officers at Equine Breeding Stud, Babugarh from 04th-6th July, 2023.

41. **Dr TR Talluri** was invited to deliver a lead lecture on "Equine Embryo transfer in Indigenous horse breeds-Challenges and hurdles" at the 38th Annual Convention of ISSAR and International symposium on "Frontiers in Theriogenology: Research and Practice" conducted from 6th-8th Dec., 2023 at College of Veterinary and Animal Sciences, Mannuthy, Thrissur, Kerala

42. **Dr TR Talluri** was invited to deliver an expert lecture on "Follicular Dynamics in Mare" for a short-term training course on "Use of Ultrasonography in Mare Reproduction" organized by Department of Gynaecology & Obstetrics, College of Veterinary Science & A. H., KU., Sardarkrushinagar during 5th-7th July, 2023.

43. **Dr TR Talluri** was invited to deliver an expert talk on "Semen Analysis using flow cytometry" on 7th Feb., 2023 at Kamdhenu University, Anand, Gujarat conducted by Trust for education and Training in Cytometry (TETC) during 24th INDO-US Flow Cytometry Workshop (Vet Applications), organised from 6th-7th Feb., 2023.

44. **Dr TR Talluri** was invited to deliver an expert talk on "Semen Analysis using flow cytometry" on 7th Sept., 2023 at Flow Cytometry workshop (Pakistan's 2nd International Flow Cytometry Workshop) from 7th-9th Sept., 2023 held at Aga Khan University Hospital, Karachi.

45. **Dr Riyesh T** delivered a lecture on "Emerging and Spillover Zoonoses from Bats: Prediction and Monitoring" during the Bay of Bengal Initiative for Multi-Sectoral Technical and Economic Cooperation (BIMSTEC) online training on Recent Advances in Diagnosis and Control of Emerging and Re-emerging Zoonoses, held from 25th-30th Sept., 2023. The event was organized by the ICAR-Indian Veterinary Research Institute, Izatnagar, Bareilly, Uttar Pradesh, in collaboration with the ICAR-National Research Centre on Equines, Hisar, and the ICAR-National Institute for High Security Animal Diseases, Bhopal.

Workshop, Seminar,

Institutional Activities

Workshops

I. A Workshop on QSAR based Computational Drug Design and its Application

A workshop was organized on "Workshop on QSAR based Computational Drug Design and its Application" from 6-8 July 2023 at ICAR-National Research Centre on Equines, Hisar in collaboration with Altem Technologies Pvt Ltd., Bagalore. The workshop was inaugurated by Dr. T. K. Bhattacharya, Director, ICAR-NRCE and workshop manual was released. A total 30 participants attended this hand-on-workshop academic/SAU universities – GJU & ST, Hisar, CBLU, Bhiwani, LPU, Phagwara, LUVAS, Hisar, GADVASU, Ludhiana, CCS HAU, Hisar and ICAR institutes – IVRI, Izatnagar, and NRCE, Hisar. There were seven candidates from ICAR institutes and 23 were from non-ICAR institutes. The workshop covered on various topics viz. Virtual screening of Database, 3D-QSAR Pharmacophore modelling, ADMET and TOPKAT analysis, Molecular docking. The applicants were given live demo on the Discovery software installed in the bioinformatic facility of the ICAR-NRCE and live hands-on-practice were given by the resource person. Dr Dhivya and Dr. Sanjay Kumar were the main resource person for this workshop.

II. Workshop on "Zoonotic Diseases: Prevention and Control

A. An awareness workshop on Zoonotic Diseases has been organized at Urban Public Health Community Centre (UPHC), Krishna Nagar Gamri, Kurukshetra, Haryana, by ICAR-NRCE team on 20 Jan, 2023. A total of 37 Medical and Veterinary Professionals and 35 farmers from Kurukshetra district attended this workshop. The goal of this workshop was to make aware the farmers about common zoonotic diseases and diminish the gap between veterinary and medical profession to control zoonotic diseases.


B. *One day Workshop on "Zoonotic Diseases*: Prevention and Control was organised at at DCRUST University, Murthal, Sonipat, Haryana, by NRCE team on 24 Jan, 2023. A total of 49 Medical and Veterinary Professionals and 40 farmers from Sonipat district marked their presence for this workshop. The goal of this workshop was to make aware the animal husbandry workers and farmers about common zoonotic diseases. Lectures and discussions on various zoonotic diseases such as glanders, bovine tuberculosis, brucellosis, leptospirosis, scrub typhus, rabies, bird flu and Japanese encephalitis were delivered and followed by a discussion with the participants


C. An awareness workshop on Zoonotic Diseases has been organized at Surkhab Complex, Sirsa, Haryana by NRCE team on 16th March 2023. A total of 70 Medical and Veterinary Professionals and 14 farmers from Sirsa district marked their presence for this workshop. The goal of this workshop was to make awareness about common zoonotic diseases and share the knowledge amongst medical officers, and veterinary officers and livestock farmers.


Institutional Activities

a) Foundation Day of ICAR-National Research Centre on Equines

ICAR-National Research Centre on Equines (ICAR-NRCE) proudly celebrated its 39th Foundation Day on November 26th, 2023. Renowned for pioneering advancements in equine health, production, and the conservation of veterinary microbes, ICAR-NRCE has remained dedicated to delivering exceptional services to its stakeholders. Dr SP Kimothi, Member of the agricultural Scientist Recruitment Board, New Delhi graced the occasion along with esteemed Guests of Honour Dr Gaya Prasad Former VC, SVVPUAT, Meerut, Dr Gulshan Narang, Dean CoVSc, LUVAS, Hisar, Director CIRB and Commandant, Equine Breeding Stud, Hisar. Dr Kimothi praised the remarkable work being done at ICAR-NRCE and underscored the necessity of developing cost-effective technologies to advance equine welfare for benefitting the stakeholders. Dr TK Bhattacharya, Director of ICAR-NRCE, provided insights to the guests and stakeholders about the cutting-edge technologies pioneered by ICAR-NRCE and reaffirming its dedication to the industry. Dr Nitin Virmani, Head, Equine Health Division was organizing secretary for the events which highlighted captivating equestrian events showcasing cavalry sports such as tent-pegging, hankey picking, archery on horseback, the mesmerizing dance of horses, and exhilarating show jumping. The event showcased the finest horses of the Marwari breed. The celebrations included a "Brainstorming Session on Protecting Equine Biodiversity in India". The session was attended by industry leaders, stakeholders, experts (scientists, field veterinarians) and policymakers. This session facilitated critical discussions on the declining equine population and issues concerning the preservation and enrichment of India's diverse equine population.

b) 35th Foundation Day Celebration at EPC Bikaner

The 35th Foundation Day of the National Research Centre on Equines, Regional Station-Equine Production Campus, Bikaner was celebrated on September 28, 2023. On this occasion, the chief guest Professor A.K. Gehlot, former Vice-Chancellor, Rajasthan University of Veterinary and Animal Science, Bikaner said that innovation should be done to promote horse rearing. He praised the research being done at the centre. Dr. S.C. Mehta, Head of Regional Station said that this centre has initiated several new aspects of research, they include selection for height at withers of Marwari horses, research on DNA marker for *Rewal Chaal*, characterisation of Bhimthadi breed, work on horse embryo banking and others. Dr Mehta appealed to the horse breeders that they should form a Horse Breeder's Association for the organisation of horse competitions in Bikaner. He assured the horse breeders of every technical support. On this occasion, the guests also released the calendar of horse breeds of India. Special guest of the program, Mr. Anil Rathor, Deputy Director, Tourism Department said that he will try to organize horse races and other competitions in the Camel Festival this year. He personally interacted with the horse breeders and discussed in depth on connecting tourism with horse rearing. On this occasion, a farmer-scientist dialogue program was also organized in which Dr. R. K. Dedar and Dr. T Rao Talluri interacted with the horse breeders.

c) Celebration of Rabies awareness week

Rabies awareness week was celebrated on "World Rabies Day". Starting on 27 September 2023, A team of medical professionals of Civil hospital, Hisar and ICARNRCE organized a health camp at Nagar Nigam office, Hisar. On 28 September 2023, a collaborative event with the NRCE and LUVAS was held at the town park. Interactive workshops and demonstrations on safe animal handling and bite prevention were conducted. On 29 September 2023, an awareness drive was started, banners were pasted on 4 autos to spread the awareness about rabies in whole Hisar district for 10 days. From 2 - 6 October 2023, daily awareness sessions were conducted at Civil hospital to educate visitors about the early signs of rabies infection and the need for immediate medical attention in case of animal bites. On 7 October 2023, rabies awareness lectures were organised at Government school, Neoli Kalan, Hisar. A total of 120 students participated in this and quiz contest was organized, and prizes were distributed to winner students.

The program was successfully conducted under the patronship of Dr Tarun Kumar Bhattacharya, Director, ICAR-NRCE and the chairmanship of Dr Harisankar Singha, Senior Scientist, ICAR-NRCE.

d) ICE activities under National One Health Programme for Prevention and Control of Zoonoses

For information, communication and education, posters (glanders and JE) and leaflets (Rabies, brucellosis, glanders, anthrax, JE) were prepared in Hindi language and distributed to various stakeholders.

e) World Soil Day (5th Dec, 2023)

World soil day was celebrated at the center. Dr. Rajiv Bhatia delivered expert lecture.

f) Activities under SCSP Plan

Livestock health and awareness camps were held under SCSP Plan in six villages during the year 2023 in villages dominated by SC community at Ramayan, Ludas, Tokas, Pattan, Nangthla and Sulakhni villages in the District of Hisar. Around 300 farmers from the community were provided with deworming medicines, mineral mixture etc. Literature with regard to diseases of one health importance and diseases specific to livestock was distributed. Farmers were educated about husbandry practices. Refrigerators were distributed for Infrastruture support to five Veterinary Dispensaries in the villages' viz. Ramayan, Nangthla, Sulakhni, Pattan and Ludas for keeping biologicals and vaccines for maintaining the cold chain.

राष्ट्रीय अश्व अनुसंधान केन्द्र में हिंदी पखवाड़े का आयोजन

भा.कृ.अनु.प. – राष्ट्रीय अश्व अनुसंधान केन्द्र में 01 सितंबर, 2023 से 14 सितंबर, 2023 हिंदी पखवाड़ा आयोजित किया गया। हिंदी पखवाड़े के उद्घाटन समारोह के दौरान दो हिंदी कार्यशालाओं का आयोजन किया गया जिसमें डॉ मोनिका कक्कड़ जी और श्री नरेश शर्मा जी ने व्याख्यान दिया। इस उपलक्ष्य पर डॉ टी.के. भट्टाचार्य, निदेशक भा.कृ.अनु.प. – राष्ट्रीय अश्व अनुसंधान केन्द्र ने बताया कि हिंदी भाषा का अधिक से अधिक प्रयोग करना हमारी कार्यशैली को उत्तम बनाएगा। हिंदी जन जीवन की भाषा और अधिक उपयोगी है। अयोजित की गई कार्यशालाओं में हिंदी का महत्व, उपयोगिता और इस भाषा को जन–जन की भाषा के रूप में प्रोत्साहन दिया गया। संस्थान के सभी कर्मचारियों ने इस कार्यक्रम में भाग लिया और कार्यशाला से लाभान्वित हुए। हिंदी पखवाड़ा के दौरान हिंदी भाषा के प्रोत्साहन हेतु कार्यालय में विभिन्न कार्यक्रम आयोजित किए गए। इनमें प्रमुख रूप से प्रतियोगिताओं में – हिंदी में किया गया सर्वाधिक कार्य, श्रुतलेख प्रतियोगिता, निबंध प्रतियोगिता, हिंदी आशुभाषण, टंकण प्रतियोगिता, वर्ग पहेली प्रतियोगिता और कविता प्रतियोगिता शामिल थे। प्रतिभागियों को इनाम स्वरूप भेंट डॉ. मुकेश जैन, टीटीसी के निदेशक द्वारा प्रदान किया गया। कार्यक्रम में अश्व स्वास्थ्य विभाग प्रमुख डॉ. नितिन विरमानी, एनसीवीटीसी प्रमुख डॉ. नवीन कुमार और हिंदी अधिकारी डॉ. तरुणा आनंद व सभी वैज्ञानिकों का योगदान एवं तकनीकी अधिकारियों व प्रशासनिक विभाग का योगदान सराहनीय रहा। हिंदी के प्रोत्साहन हेतु संस्थान के सभी कर्मचारियों ने पखवाड़े में बढ़–चढ़कर भागीदारी की।

हिंदी सप्ताह एवं राजभाषा कार्यक्रम

केंद्र परिसर की राजभाषा इकाई द्वारा प्रत्येक तिमाही में राजभाषा कार्यान्वयन समिति की बैठक एवं कार्यशाला का सफल आयोजन किया गया द्य राजभाषा, गृह मंत्रालय, भारत सरकार द्वारा पुणे, महाराष्ट्र में 14–15 सितम्बर को हिंदी दिवस एवं तृतीय अखिल भारतीय राजभाषा सम्मेलन का आयोजन हुआ। केंद्र की प्रतिभागिता हेतु श्री कमल कुमार सिंह, राजभाषा अधिकारी ने 11 से 16 सितम्बर के दौरान इस सम्मलेन में भाग लिया गया।

प्रतिवर्ष की भांति इस वर्ष भी हिंदी दिवस एवं हिंदी सप्ताह का आयोजन किया गया। हिंदी सप्ताह आयोजन के दौरान विभिन्न हिंदी प्रतियोगिताएं रखी गई। मुख्यत: इन प्रतियोगिताओं को दो श्रेणी में कराया गया, जिससे हिंदी सप्ताह का आयोजन रोचक बन सके एवं सभी अधिकारी, कर्मचारी (स्थाई व संविदा पर कार्यरत) हिंदी भाषा का प्रयोग आसान तरीकों को अपनाते हुए प्रभावी ढंग से कर सके। प्रतियोगिता हेतु (1) शुद्ध/अशुद्ध शब्द प्रतियोगिता (2) हिंदी पोस्टर प्रतियोगिता एवं (3) नवाचार–विलोम शब्द, पर्यायवाची एवं मुहावरे का आयोजन किया गया। पुरस्कार हेतु प्रथम, द्वितीय, तृतीय एवं सांत्वना दोनों ही श्रेणियों में प्रदान किये गए। परिसर राजभाषा अध्यक्ष एवं केंद्र के प्रभागाध्यक्ष डॉ. एस. सी. मेहता ने राजभाषा के अधिकाधिक प्रयोग पर

बल दिया एवं राजभाषा अनुभाग को बधाई दी एवं आगामी वर्षों में इसे ओर अधिक रोचक एवं प्रभावी बनाने हेतु मार्गदर्शन प्रदान किया। कार्यक्रम में पधारे अतिथियों ने अपने संबोधन में केंद्र की गतिविधियों की भूरी-भूरी प्रशंसा के साथ-साथ, केंद्र द्वारा राजभाषा के क्षेत्र में दिये जा रहे योगदान की सराहना की।

Independence Day celebration at ICAR-NRCE

ICAR-NRCE, Hiasr and EPC Bikaner celebrated 77th Independence Day on August 15, 2023, with great patriotic zeal on 15 August, 2020 with the theme of Meri Mati Mera Desh. The National flag of India was hoisted by Dr T K Bhattacharjya, Honorable Director of the institute at the front yard of ICAR-NRCE; and he inspired the staff and families by memorializing a series of incidents in the history of India for the achievement of independence. To mark the day, children of the employees took pride in glorifying and celebrating the spirit of unity. Besides, Director inspired the staff to accomplish the new horizons in their scientific endeavor. Flag hoisting was done by Dr. S. C. Mehta, Head of the Campus. A quiz on "Indian independence movement" was organised on this occasion and the winners were felicitated. Sporting event were also organised on this occasion at CAR-NRCE, Bikaner Campus.

Celebration of 'International Yoga Day'

International Yoga Day was celebrated with great enthusiasm on dated 21st June, 2023 at ICAR-NRCE, Hisar. Yoga practice and awareness camp was organized in front of main office building of the center. Scientists, officers and staff of the center took part is the yoga day. Dr. Naveen Kumar, Head, NCTCC briefed about importance of this day and called upon the gathering to adopt yoga in their daily life.

International Yoga Day-2023

On the occasion of International Yoga Day on 21st June 2023, a program of mass yoga practice was organized under the aegis of Yoga and Naturopathy Centre at Swami Keshavanand Rajasthan Agricultural University. Head of the Campus Dr. SC Mehta and other officers of the Centre participated in the mass yoga practice program. The program was inaugurated by the Chief Guest Dr. Arun Kumar, Vice Chancellor - Swami Keshavanand Rajasthan Agricultural University, by offering havan. On this occasion, Vice Chancellor Dr. Arun Kumar, Dr. Artabandhu Sahu, Dr. SC Mehta, Hoshiyar Singh Meena, Dr. Jagdish Rane, Dr. P.S. Shekhawat, Dr. Vimala Dukaval, Sandeep Khetan etc. expressed their views on the importance of yoga.



Republic Day celebration

Republic Day, January 26, 2023, was enthusiastically observed by ICAR-NRCE and EPC Bikaner. Raising the national flag and remembering a number of significant events in Indian history, the director of ICAR-NRCE Hisar and the officer in charge of EPC, Bikaner, motivated the employees and their families. The employees' children celebrated and honored the spirit of unity on this day with pride.


ICAR-NRCE, Hisar celebrated Kisan Diwas

In the occasion of Kisan Diwas, ICAR-NRCE, Hisar organized a health camp on 23rd December at Pabra Village, Hisar District, Haryana for equine and livestock farmers. A team of Scientists, Technical officers and Students of this Institute observed the Day. NRCE team briefed about various activities and technologies developed by the Institute to benefit the equine and livestock farmers. In the health camp, around twelve equine and livestock owners participated and equines and other livestock were examined to assess their health status. In addition, blood samples and nasal swab samples were collected to test for the presence of equine diseases such as glanders, strangles and other parasitic diseases. In this camp, farmers were briefed about importance of regular screening of animals for important equine diseases. A number of inputs such as mineral mixtures, deworming boluses and ectoparasitic sprays and tincture iodine were distributed to the framers for their animals.

Swachhta Pakhwada

Swachhta Pakhwada activities were observed from16-31 Dec., 2023 at ICAR-NRCE, Hisar and EPC Bikaner. During this time period various activities such as Taking Swachhata pledge, basic maintenance, campaign on cleaning of sewerage and water lines, Campaign on cleaning of sewerage and water lines, awareness on recycling of waste water, water harvesting for agriculture/horticulture application/kitchen gardens in residential colonies/ 1-2 nearby villages, swachhta Awareness at local level (organizing Sanitation Campaigns involving and with the help of the farmers, farm women and village youth in new villages not adopted under any scheme by Institutes/ establishments were conducted/organized by ICAR-NRCE

Health Camps and Kishan Divas and Gosthies-2023

a) 39th State Level Livestock Show, Dadri, Haryana (11-13th March, 2023) : The Centre's stall was put-up at State Level Livestock show at Dadri. Approx. 400 delegates visited the stall and equine related information and literature were provided.

b) Pashu Mahotsav and Pradarshini evam Prashikshan, Muzzaffernagar UP (67th April, 2023) : Stall of the center was put-up at Pashu Mahotsav and Pradarshini evam Prashikshan. Approx. 700 delegates visited the stall and equine related information and literature were provided.

c) National Dairy Mela at ICAR-NDRI, Karnal, Haryana (8-9th April, 2023) : Centre's stall was put-up at National Dairy Mela at ICAR-NDRI, Karnal Haryana. Approx. Approx. 600 delegates visited the stall and equine related information and literature were provided.

d) **Health Camp and Kishan Gosthy at Baltal and Pahalgam Routes, J&K :** Organized health camps on both sides of Amarnath Shrine (Baltal and Pahalgam Routes). The Camps were held on 7th-8th August and 10th August, 2023. Equine stakeholders of around 100 were benefitted by this activity. The animals were checked for different problems like colic, foot sores, and saddle sores. Medications were distributed and treatments were provided. The issues were discussed and information related to problems was provided. Blood samples were also collected for surveillance of equine diseases.

e) **Health Camp and Kishan Gosthy at Hanumanargh (23rd Dec, 2023) :** An equine health camp and kishan gosthy was organized at Bhatner Equine Show, Hanumanargh (Rajasthan).

f) **Tilwara Horse Fair (22nd-23rd March, 2023) :** A team comprising Dr RK Dedar and Dr TR Talluri organised a camp at Balotra, Rajasthan during the Tilwara horse fair from 22-23rd March 2023. A total of 97 farmers have visited our stall and students from 3 schools and one science college also visited our stall and gained the knowledge about our institute and technologies developed by the centre.

Services provide to equine farmers at ATIC-2023

S. No.	Month	AI done in mares brought by farmers at ATIC	PD / Gynecological examination / Consultancy	Consultancy at Kishan call centre on toll free number	Total
1.	January	00	00	00	00
2.	February	02	05	03	10
3.	March	01	03	00	04
4.	April	01	00	01	02
5.	May	02	01	01	03
6.	June	00	00	00	00
7.	July	00	00	10	10
8.	August	01	01	00	02
9.	September	00	00	00	00
10.	October	00	04	01	05
11.	November	00	02	01	03
12.	December	00	00	00	00
Grand Total		**07**	**16**	**17**	**40**

IRC, RAC and
IMC Meetings

26th Research Advisory Committee (RAC) meeting of ICAR- NRCE

The 26th RAC meeting of ICAR- National Research Centre on Equines was held under the Chairmanship of Dr. M C Sharma (Former Director, IVRI) on 20-21st March, 2023 to review the research for the year 2022-23. Dr. T.K. Bhattacharya, Director, ICARNRCE gave a brief presentation on the overall achievement of the Centre for the year 2022-23 which was followed by presentations on equine health, equine production and NCVTC. A total of 40 research projects were discussed in the meeting. The Chairman RAC initiated the proceedings of the meeting with his opening remarks. The Chairman RAC appreciated the efforts of the scientists and emphasized that every scientist should make efforts to develop one product per year to set the pace for developing cutting edge technologies. The scientists should work in mission mode for vaccine development, patenting and commercialization of the technologies. Concerted efforts should be made towards out-reach programs.

Dr. T.K. Bhattacharya, Director, ICAR-NRCE presented the action taken report on the 25th RAC recommendations. During the presentation, Chairman RAC suggested to include veterinary universities, animal farms and academia in the surveillance for sample collection plan. The equine population is decreasing at a faster rate due to mechanization. The Institute should prioritize efforts to improve the utility of animals and work for improving livelihood of equine farmers. The Chairman emphasized the need for transfer of the technologies to equine farmers and initiate extension activities in equine populated area of HP, J&K, Uttrakhand and NEH region etc.

Annual Institute Research Committee (IRC) Meeting

The annual IRC meeting (2022-23) of ICAR-NRCE, Hisar was held in the committee room (ICAR-NRCE) under the chairmanship of Dr. T.K. Battacharya, Director, ICAR-NRCE on 23rd May, 2023 and 07th June 2023 & and 10th July, 2023. A total of 18 institute research projects, 22 externally funded projects and nine concept notes were discussed in the meeting. Dr. T.K. Battacharya, Director, ICAR-NRCE in appreciated the research activities and output from the scientists. He motivated the scientists to do research relevant to the stakeholders' needs. He also motivated the scientists to apply for the external funding in view of budget constraints. He also guided the scientists to work in collaborative mode and to share the manpower, resources and judicious utilization of the institute funds. Two external experts (Prof. M.L. Sangwan, Professor (Retd), LUVAS, Hisar and Dr. Rajesh Kumar Chhabra, Head Department of Microbiology, LUVAS, Hisar) were also invited in the meeting for evaluation of the progress of ongoing research projects and suggestions.

Half Yearly Institute Research Committee (IRC) Meeting

The Half yearly IRC meeting (2023) of ICAR-NRCE, Hisar was held in the committee room under the chairmanship of Dr TK Bhattacharjya, Director on 03-11-2023 and 0611-2023 and 4.01.2023 (online). A total of 22 institute research projects, 24 externally funded projects and two concept note were discussed in the meeting. The meeting was attended by the following scientists. In his opening remarks, the Chairman asked the Scientists to present the salient achievement of the projects in briefly. He also requested that Scientist applying for extenally funded project to submit a copy of the same to PME cell for proper evaluation & records.

The Chairman urged all the scientists to continue working hard in order to achieve further success in the future. Subsequently, Dr Nitin Virmani, the Head of Equine Health Unit was invited to share his comments. Dr Nitin Virmani expressed his opinion that due to the shortage of scientific staff at the institute, there is a need to reorient research programs and he raised concerns about the reduction of equine population in the country. After the Chairman's remarks and Dr Nitin Virmani's comments, the in-charge PME Dr Balvinder Kumar invited each individual scientist to present the progress report.

MoU

The ICAR-NRCE, Hisar inked the Memorandum of Understanding with various reputed and prestigious academic and research oriented institutes namely Central University of Haryana, Mahendergarh, National Institute of Pharmaceutical Education and Research NIPER, SAS Nagear Mohali, Punjab, Kamdhenu University, Gandhinagar, Gugarat, Kerala Veterinary and Animal Sciences University (KVASU), Wayanad, Kerala, Chaudhary Sarwan Ku mar Himachal Pradesh Krishi Vishvavidyalaya, Palampur, Himachal Pradesh, Assam Agrieultural University, Head Quarter Jorhat, Tamil Nadu Veterinary and Animal Sciences University, Chennai. For the cooperation in the areas of Research and Education. Dr. T.K. Bhattacharya, Director, ICAR-NRCE, Hisar and Vice Chancellors/Directors of the Universities/Institutes signed the Memoranda. With the MoU, the NRCE and Universities have agreed for collaborative programmes in the fields of research, education, training and capacity building, extension consultancy and other areas of national interest. Both the partners have also agreed for mutually recognizing the faculty of both the Institutes for the research and teaching purposes, wherein, the students and faculties can carry out the specific, research and outreach activities at the laboratories of these institutions.

S.No.	University Name	Date of signing MoU
1	Central University of Haryana, Mahendergarh	1.06.2023
2	NIPER, SAS Nagear Mohali, Punjab	12.10.23
3	Kamdhenu University, Gandhinagar, Gugarat	26.6.23
4	Kerala Veterinary and Animal Sciences University (KVASU), Wayanad, Kerala	3.8.23
5	Chaudhary Sarwan Ku mar Himachal Pradesh Krishi Vishvavidyalaya Palampur, Himachal Pradesh	5.8.23
6	Assam Agrieultural university, Head Quarter Jorhat	5.12.23
7	TANUVAS, Chennai	9.12.23

Institute Management Committee (IMC) Meeting :

42nd Meeting of Institute Management Committee of ICAR-NRCE, Hisar, held online on 31.07.2023 at 11.00 AM under the Chairmanship of Dr. T.K. Bhattacharjya, Director, ICAR-NRCE, Hisar. Dr. T.K. Bhattacharjya, Director, NRCE and Chairman, IMC briefed the house on the activities and achievements of the Centre. IMC appreciated the efforts and the achievements of the Centre. Shri Raj Kumar, Sr. Administrative Officer & Member Secretary presented the agenda items and the following recommendations were given by the committee:

1. The proposal for write-off of losses on account of mortality of animals at EPC, Bikaner was placed before the IMC for consideration for approval. Dr. Nitin Virmani, Head, Division of Equine Health briefed the IMC of the cause of death of these animals. After discussions, IMC considered and approved the proposal for write off of losses worth Rs. 1,31,873/- (Rupees one lakh thirty one thousand eight hundred seventy three only).
2. IMC was informed that Dr. Sanjay Kumar, Principal Scientist & I/c Medicine Lab. of this Centre reported burnt out incidence in the Medicine Laboratory on 31.3.2023. A committee was constituted to find the reasons of burnt out incidence in the Medicine Laboratory and to suggest further course of action in the present matter and also to suggest preventive measures so as to avoid any such incidence at the Centre. Committee submitted its report to the Director, ICAR-NRCE. Committee recommended appropriate further necessary action on the write-off of these losses as per ICAR guidelines. The committee observed that the loss was not due to the negligence of anyone. Committee also suggested necessary future preventive measures to avoid such incident at ICAR-NRCE.
3. Smt. Shammi Tyagi, Sr. Finance & Accounts Officer briefed the IMC about the financial status of the Centre. She informed that during the financial year 202223, there was 100% utilization of funds under NRCE including SCSP, NEH & TSP as well as NCVTC including SCSP; while 100% funds were utilized under Disaster Emergency fund. SFAO further informed the IMC that against the target of revenue generation of Rs. 137.88 lakhs during 2022-23, the actual revenue receipt generated was Rs 139.94 lakhs.
4. IMC was informed that a tractor (Regn. No. HR20H 2713) was purchased at this Centre in 2002. The tractor is old and not economical to run. This tractor has completed its useful life. IMC was further informed that a committee was constituted for inspection of said tractor. The committee after physical inspection of the condition of tractor recommended its condemnation. The above tractor has completed its useful life in terms of number of hours run as well as number of years in terms of guidelines contained in Council's letter No. FIN/6/1/2016-CDN(A&A) dated 3.9.2019.
5. Extension of term of Part Time Doctor (AMA) at NRCE, Hisar.

Visit of
Dignitaries

A. **Dr S L Goswami, lauds the significant contributions of NRCE :** Dr S. L Goswami, former Director, NAARM visited ICAR-NRCE on 01-02-2023. He interacted with scientists of the Centre on the research activities being undertaken on equine health and production. Furthermore, Dr Goswami stressed the significance of adhering to Good Laboratory Practices (GLP) during the development of new technologies, urging the scientists to uphold rigorous standards in their research endeavours.

B. **Dr Ashok Talyan admired the equine museum at ICAR-NRCE :** Dr Ashok Talyan, Joint Director of Medical Health (Meerut Division, Uttar Pradesh) visited ICAR-NRCE on 02-02-2023. He commended the work done by ICAR-NRCE on equine breed conservation and highly appreciated the educational and informative displays arranged at the equine museum.

C. **Dr Pankaj Kumar appreciated the hard work and dedication of ICARNRCE staff :** Dr Pankaj Kumar, Director (AS), ICAR, visited ICAR-NRCE on 22-07-2023. He engaged with the Centre's scientists regarding their research efforts in equine health production and breed conservation. He appreciated the hard work and dedication of the institute's staff in the field of equine science.

D. **Dr PK Uppal acknowledges the important contributions of NRCE** : Dr P K Uppal, Former Director, ICAR-NRCE visited the institute on 10-09-2023. Dr Uppal commended the ongoing research by the scientists and expressed admiration for the technologies produced by the NRCE team.

E. **Dr SP Kimothi, praises ICAR-NRCE's excellence in equine science and vaccine development :** Dr Shiv Prasad Kimothi, Member ASRB visited ICAR-NRCE on 26-11-2023. During his visit, Dr Kimothi expressed admiration for the institute's exceptional work in equine science research. Dr. Kimothi praised ICAR-NRCE for their exceptional work on a lumpy skin disease vaccine, underlining the institute's role in innovating animal health solutions.

F. **Dr SB Barbuddhe commends ICAR-NRCE's leading role in equine research :** Dr SB Barouddhe, Director, NMRI, Hyderabad visited ICARNRCE on 08-12-2023. During his visit; the Dr Barouddhe praised the institute as one of the leading establishments within ICAR. He commended ICAR-NRCE's significant contributions to research and development in the field of equine science.

G. **Deputy Director General (FS) advocates for fisheries sector collaboration with ICAR-NRCE :** Dr Joykrushna Jena, DDG, Fisheries Sciences visited ICAR-NRCE on 09-12-2023. During his visit, the Deputy Director General (DDG) commended the institute for its outstanding reputation within ICAR. Despite the scientific strength being only half, the DDG acknowledged the institute's remarkable achievements and the valuable contributions it has made in the field of equine science research. Furthermore, The DDG expressed willingness for the fisheries sector to be part of the National Centre for Veterinary Type Cultures (NCVTC) network, indicating a collaborative approach to enhance research capabilities and technological advancements.

Infrastructure, Developmental Activities and Herd Stregth

Facilities creation for vermicomposting by utilizing equine dung

Generally, equine dung (ED) is disposed-off as a waste in nearby barren area and it goes as useless and pollutes the environment. There was neither any scientific document nor any report was available of making vermicompost by utilizing ED. Hence this activity was undertaken. It was a scientific method of making compost by utilizing earthworms in ED. The earthworms were used to enhance the process of conversion of ED into a valuable bio-fertilizer. Earthworms consumed ED and as an outcome produced vermicompost. No chemical element was used in this process, and it is a natural fertilizer.

This activity includes creation of infrastructure facilities of vermicompost unit, collection of material required, starting of the project. Previously, a vermicompost unit was established near equine farm of ICAR-NRCE in rectangular shape (length 40 feet x width 20 feet) consisting of eight vermibeds. Floor of the unit and vermibeds were built up by using bricks and cement. Vermibeds were built up (length 15 feet x width 4 feet x depth 2 feet) little bit tilted from one side to collect vermiwash). Each vermibed was facilitated with a drainage pipe (2-inch size) and a pit (length1 feet x width 10 inch x depth 10 inch) toward tilted side as recommended by Ismail-2005 and along with some modification as per need, space and availability of funds. The roof of the vermicompost unit was made of corrugated galvanized iron sheets fitted on a frame of galvanized iron pipe at height of 10 feet to facilitate proper air flow and to ensure cool environment. Level of the floor of vermicompost unit was kept 6 inch high to ground level to avoid water stagnation. During the year 2023 this infrastructure facilities of vermicomposting has been restored which were not operational during last two years due to water stagnation in farm area. During the year 40 quintals vermicompost was cultivated by utilizing equine dung and supplied to Agricultural Farm for manure in plants and crop.

Inauguration of New gate NCVTC by Dr Jena DDG (AS)

A new gate has been built in the NCVTC campus and the gate has been inaugurated by Dr. Joykrushna Jena DDG (Animal Sciences), ICAR Headquarters, New Delhi on 9th December 2023. Besides, Dr Ashok Kumar ADG (Health), ICAR Headquarters, Dr T K Bhattacharya, Director, ICAR-NRCE and Dr Rajneesh Rana, Principal Scientist, ICAR Headquarters, New Delhi were also facilitated the event.

Equine Museum

An equine museum has been initiated at the Campus for the tourists, farmers and students to get the scientific knowledge about the equines in the country. Comprehensive pictorial information about the world's best horses; breeds of indigenous horses; well-known warriors and their brave horses; research activities and achievements of the institute, and awards and recognition of the institute at various levels has been presented. The equine harness and saddlery items have been displayed. This is how, the Institute is trying to bridge the gap between people and the horses.

DBT Certification of Microbial Containment Laboratory (BSL-3) Facility

The Microbial Containment Laboratory (BSL-3) Facility at ICAR-NRCE, Hisar has undergone significant repairs and upgrades, including the air handling unit, Building Management System (BMS), biosafety cabinets, HEPA filters, and the effluent treatment plant. The laboratory recently received certification compliance issued by DBT, Govt of India on 13.02.2024. Presently, it is part of the National BSL3 Laboratory Network under the National One Health Mission (NOHM) to strengthen nationwide intersectoral joint outbreak investigation by enhancing the capacity of laboratories to collect and test samples from human, animal, and environmental origins.

Department of Biotechnology
Ministry of Science & Technology, Government of India

Certificate of compliance for BSL-3 Facility

Based on the assessment made by the Inter-ministerial Committee for Certification of BSL-3 facility and recommendations of Review Committee on Genetic Manipulation (RCGM), the following facility is certified for BSL-3 compliance:

Indian Council of Agricultural Research-National Research Centre on Equines (ICAR-NRCE),

Sirsa road,
Hisar-125001, Haryana

Certificate No: BT/IBKP/325/2020-BSL3

Issue Date : 13/02/2024 Valid up to: 12/02/2027

N.B. In compliance with IBSC, and other regulatory Biosafety Guidelines of the country applicable from time to time.

Dr. Nitin K Jain,
Scientist- G,
Department of Biotechnology &
Member Secretary,
Review Committee on Genetic Manipulation

Prof. Y. K. Gupta
Chairman,
Review Committee on Genetic Manipulation,
Former Director , IITR, Lucknow and
President, AIIMS, Bhopal

Equine Strength- 2023 (Hisar)

S. No.	Kind of equine	OB 01.01.2023	Addition		Disposal		CB 30.12.2023
			Birth	From EPC	Death	Sale	
Horses							
1.	Mares	05	nil	02	nil	nil	07
2.	Fillies	nil	02	nil	nil	01	01
3.	Colts	01	01	nil	nil	nil	02
4.	Stallion	01	nil	nil	nil	nil	01
	Total	**07**	**03**	**02**	**nil**	**01**	**11**
Ponies							
1.	Mares	04	nil	nil	nil	nil	04
2.	Fillies	01	01	nil	nil	nil	02
3.	Colts	nil	01	nil	nil	nil	01
	Total	**05**	**02**	**nil**	**nil**	**nil**	**07**
Donkeys							
1.	Mares	**12**	**nil**	**nil**	**nil**	**nil**	**12**
2.	Fillies	**05**	**02**	**nil**	**nil**	**nil**	**07**
3.	Colts	**10**	**05**	**nil**	**01**	**09**	**05**
4.	Stallion	**02**	**nil**	**nil**	**nil**	**nil**	**02**
	Total	**29**	**07**	**nil**	**01**	**09**	**26**
	Grand Total	**41**	**12**	**02**	**01**	**10**	**44**

Equine Strength - 2023 (Bikaner)

	MARWARI		KATHIAWARI		ZANSKARI		MANIPURI		POITOU		HALARI		MULE		NUKRA		TOTAL
	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	
Stock as on 01.01.2023	**18**	**27**	**01**	**04**	**03**	**06**	**03**	**01**	**06**	**17**	**03**	**06**	**01**	**00**	**02**	**00**	**98**
Birth	0	3	0	0	0	0	0	1	0	2	2	0	0	0	0	0	**08**
Purchased	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	**00**
Death	0	01	0	0	0	0	0	0	0	0	0	0	0	0	0	0	**01**
Auctioned / Sold	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	**00**
Transferred to NRCE, Hisar	0	02	0	0	0	0	0	0	0	0	0	0	0	0	0	0	**02**
Balance as on 31.12.2023	**18**	**27**	**01**	**04**	**03**	**06**	**03**	**02**	**06**	**19**	**05**	**06**	**01**	**00**	**02**	**00**	**103**
G. Total	**45**		**05**		**09**		**05**		**25**		**11**		**01**		**02**		**103**

Awards, Recognition, Personal Milestones,

New Joinings, Promotions

Awards

1. Dr Naveen Kumar received ICAR Award of excellence for Agriculture research for development of India's first Lumpyi Skin Disease vaccine (Lumpi-ProVacInd).

2. Dr Naveen Kumar received India Animal Health Awards 2023 (Young Scientist) for exemplary contribution to the growth and development of Animal Health in India, In India Animal Health Summit 2023, at New Delhi.

3. Dr Anju Manuja was felicitated as Fellow of National Academy of Agricultural Sciences, by Hon'ble Director General ICAR in January 2023.

4. Dr T. R. Rao received Prof. Nils Lagerlof Award from Indian Society for Study of Animal Reproduction (ISSAR) during the 38th Annual Convention of ISSAR and International symposium "Frontiers in Theriogenology: Research and Practice" organised by College of Veterinary and Animal Sciences, Mannuthy, Thrissur, Kerala from December 6th to 8th, 2023.

5. Dr Anju Manuja awarded Fellow of National Academy of Veterinary Sciences, India.

6. Dr. SC Mehta awarded Fellow of The Indian Society of Animal Genetics and Breeding, New Delhi in 2023.

Best Poster/Oral Presentation Award

1. Dr Naveen Kumar received Best Poster Award for article entitled "*Resistance evolution against host-directed antiviral Agents: buffalopox virus switches to use p38-γ under Long-term selective pressure of an inhibitor targeting P38-α*", In 35th IAVMI conference, held on April 7th -8th, 2023 at DGCN College of Veterinary and Animal Sciences, CSK HPKV, Palampur (HP).
2. Dr Naveen Kumar received Best Poster Award for article entitled *"Emetine suppresses SARS-CoV-2 replication by inhibiting interaction of viral mRNA with eIF4E"*, In XXIX Annual Conference of IAAVR & National Symposium held at Udaipur April 8th -9th, 2022.
3. Dr Naveen Kumar received Best poster Award for article entitled "*Evidence of Lumpy Skin disease virus infection in camels*". In, VII Annual Convention of SVBBI and International Symposium on Multiomics to One Health: Challenges and Way Forward in Biomedical Research. Annual Conference of Society of Veterinary Biochemists and Biotechnologists of India (SVBBI), Organized by IVRI Izatnagar from December 14th -15th, 2023.
4. Dr Naveen Kumar received Best poster Award for article entitled "*Epitranscriptomic regulation of buffalopox virus replication in, VII Annual Convention of SVBBI and International Symposium on Multiomics to One Health: Challenges and Way Forward in Biomedical Research*". Annual Conference of Society of Veterinary Biochemists and Biotechnologists of India (SVBBI), Organized by IVRI Izatnagar from December 14th 15th, 2023.
5. Dr Naveen Kumar received Best poster Award for article entitled "*p38 MAPK regulates SARS-CoV-2 replication*" in, VII Annual Convention of SVBBI and International Symposium on Multiomics to One Health: Challenges and Way Forward in Biomedical Research. Annual Conference of Society of Veterinary Biochemists and Biotechnologists of India (SVBBI), Organized by IVRI Izatnagar from December 14th -15th, 2023.
6. Dr Naveen Kumar received Best poster Award for article entitled "*PPAR-γ regulated buffalpox virus replication*". In, VII Annual Convention of SVBBI and International Symposium on Multiomics to One Health: Challenges and Way Forward in Biomedical Research. Annual Conference of Society of Veterinary Biochemists and Biotechnologists of India (SVBBI), Organized by IVRI Izatnagar from December 14th 15th, 2023.
7. Dr T. R. Rao received the First Best poster award for the paper titled "*Production of live Marwari foals via embryo transfer technology for the first time in the country*" at 38th Annual Convention of ISSAR and International symposium "Frontiers in Theriogenology: Research and Practice" organised by College of Veterinary and Animal Sciences, Mannuthy, Thrissur, Kerala from December 6th to 8th, 2023.
8. Dr H. Singha received Best Poster Presentation Award in a session of XIXth Annual Conference of Indian Association of Veterinary Public Health Specialists (IAVPHS) on "*International Symposium on Promotion of One Health: Opportunities, Challenges and Solutions*" organized at Lala Lajpat Rai University of Veterinary and Animal Sciences, Hisar, December 7th -8th, 2023.
9. Dr A. Bhardwaj received First Prize for the Poster Presentation" Microsatellite markers based genetic characterization of Kachchhi-Sindhi horse" during International Conference on Strategies for Global Food and Nutritional Security, Sustainability and Wellness (NUTRI2023) from December 4th -6th, 2023 organized at Chaudhary Charan Singh Haryana Agricultural University, Hisar, Haryana.

Personal Milestones

1. Dr Naveen Kumar served as Section Editor "Virulence" Journal.
2. Dr Naveen Kumar was designated as Lumpy Skin Disease (LSD) expert by FAO/WOAH of the United Nations.
3. Dr R K Vaid served as expert of Veterinary Medical Science by Central Drugs Standard Control Organisation (veterinary Division) May 22nd, 2023.
4. Dr R K Vaid served as Member, DBT nominee, Central Institute for Research on Buffalo, Hisar Institute Biosafety Committee as on November 9th, 2023.

5. Dr R K Vaid served as Expert Member, Techno Commercial Assessment and Expert Committee meeting in Agrinnovate India Ltd. for technology.

6. Dr H. Singha served as a Guest Editor of Research Topic: Glanders and Melioidosis: One Health Model. Frontiers Research.

7. Dr TR Talluri and Dr RK Dedar were felicitated on the Independence Day for the contribution to the institute in science. The team was felicitated for producing the country's first Marwari filly through embryo transfer technology.

Sr.No.	Name & Designation	Date
New Joining		
1.	Dr. Muhammed Kutty VH, Scientist	Joined on 21.04.2023 at EPC, Bikaner
2.	Sh. Pawan Kumar, AO	Joined on 01.09.2023 at NRCE, Hisar
Transfer		
1.	Dr. Yash Pal, Principal Scientist	Transferred to ICAR-CIRB on 27.10.2023 upon selection as Head of the Division of Animal Physiology and Reproduction, CIRB, Hisar (Haryana)
2.	Dr. RA Legha, Principal Scientist	Transferred to ICAR-CSWRI on 10.10.2023 upon selection of Head of the Division at Regional Station, CSWRI, Bikaner (Rajasthan)
3.	Sh. Raj Kumar, Sr. AO	Transfer on 23.08.023 to ICAR-CIRB, Hisar
Promotion /Appointment		
1.	Dr. S.C. Mehta, Principal Scientist	Appointment to the post of Head, EPC, NRCE, Bikaner on 10.05.2023
2.	Dr. Nitin Virmani, Principal Scientist	Appointment to the post of Head, Division of Equine Health, NRCE, Hisar on 16.06.2023
3.	Dr. Naveen Kumar, Principal Scientist	Appointment to the post of Head, NCVTC, NRCE, Hisar on 19.06.2023
4.	Dr. Jitendar Singh, CTO	Office order dated 18.12.2023
Superannuation		
1.	Sh. Ishwar Singh, SSS	Superannuated on 31.01.2023
2.	Sh. Arun Chand, Sr. Technician	Superannuated on 31.05.2023
3.	Sh. Mahabir Prasad, SSS	Superannuated on 31.08.2023
4.	Sh. Ramesh Chander Sen, LDC	Superannuated on 31.12.2023

Research

Publications

Research Paper

1. Alim S, Laitaoja M, Pawar SS, Talluri TR, Jänis J, Swamy MJ. Primary structure determination and physicochemical characterization of DSP-3, a phosphatidylcholine binding glycoprotein of donkey seminal plasma. International Journal of Biological Macromolecules. 2023 Jun 1;239:124229.
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13. Dhania S, Rani R, Kumar R, Thakur R. Fabricated polyhydroxyalkanoates blend scaffolds enhance cell viability and cell proliferation. Journal of Biotechnology. 2023 Jan 10;361:30-40.

14. Elango K, Karuthadurai T, Kumaresan A, Sinha MK, Ebenezer Samuel King JP, Nag P, Sharma A, Raval K, Paul N, Talluri TR. High-throughput proteomic characterization of seminal plasma from bulls with contrasting semen quality. 3 Biotech. 2023 Feb;13(2):60.

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27. Kumar N, Barua S, Kumar R, Khandelwal N, Kumar A, Verma A, Singh L, Godara B, Chander Y, Kumar G, Riyesh T. Evaluation of the safety, immunogenicity and efficacy of a new live-attenuated lumpy skin disease vaccine in India. Virulence. 2023 Dec 31;14(1):2190647.

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42 Chander, Y., Kumar, R., Verma, A., Khandelwal, N., Nagori, H., Singh, N., Sharma, S., Pal, Y., Puvar, A., Pandit, R., Shukla, N., Chavada, P., Tripathi, B.N., Barua, S. and Kumar, N. Buffalopox virus isolate BPXV/Buffalo/India/2011/SB-P60, partial genome, GenBank: ON974727

Participation, Presentation in Seminars,

Conferences and Symposia

1. Dr Ana Raj J attended training entitled "Leveraging Extension Strategies for Sustainable development of Allied Agri Sector Enterprises" organized by MANAGE, Hyderabad & ICAR-NDRI, Karnal from August 16th – 18th, 2023.
2. Dr Anju Manuja presented a lead lecture in international conference on "Prospects and challenges of environment and biological sciences in food production system for livelihood security of farmers (ICFPLS-2023), organized by Pragati International Scientific Research foundation (PISRF) Meerut & Andaman Science Association (ASA) in Port Blair, Andaman & Nicobar, India from September 18th -20th, 2023.
3. Dr Anju Manuja presented an invited lecture in XVII National Technical Conference of Indian Association of Women Veterinarians on Strengthening Veterinary Profession towards One Health through Diversity, Equity and Inclusiveness, organized by NTR College of Veterinary Science, Gannavaram, Sir Venkateswara Veterinary University, A.P., India from November 29th -30th, 2023.
4. Dr Anju Manuja presented an invited lecture in a training on "Application of nanotechnology in agriculture: Opportunities and Challenges" under NAHEPIDP Project at SKUAST Jammu. (online) 2nd August 2023.
5. Dr Anju Manuja presented an invited lecture online entitled "Nanoparticles, their delivery and evaluation for biomedical applications in animals" in Karyashala Workshop on "Advanced Techniques of Preparation, Characterization and Evaluation of Nanoformulations for Biomedical Applications" from January 16th to 25th at IVRI, Izatnagar, UP.
6. Dr Anju Manuja attended international conference on "Nanotechnology Addressing the convergence of material science, Biotechnology, and Medical Science" (IC - NACMBM -2024) organized by Centre for Interdisciplinary research (CIR), D.Y. Patil Education Society, Kolhapur, Maharashtra, India from February 12th - 14th, 2024.
7. Dr B.C. Bera attended VIROCON, 2023, Advancements In Global Virus Research Towards One Health organized from December 1st -3rd, 2023 in Tiruchirappalli, Tamil Nadu.
8. Dr Balvinder Kumar delivered an invited Talk on "Equine Infectious Diseases in India: An Overview", in 2nd International Conference on Prospects and challenges of environment and biological sciences in food production system for livelihood security of farmers (ICFPLS–2023) at ICAR-CIARI, Port Blair, Andaman& Nicobar Islands, India, September 18th - 20th, 2023, organized by Pragati International Scientific Research Foundation (PISRF), Meerut, India & Andaman Science Association (ASA), Port Blair, A & N Islands, India.
9. Dr Balvinder Kumar attended International Virtual Conference on Streptococci & Streptococcal Infections organized by United Scientific Group from September 27th - 28th, 2023.
10. Dr H. Singha participated in XXXV National Conference of Indian Association of Veterinary Microbiologists, Immunologists and Specialists in Infectious Diseases on "Novel Approaches in Animal Health for Realizing One Health Mission" organized at CSK HPKV, Palampur from April 7th - 8th, 2023.
11. Dr H. Singha participated in a two-days National Conclave "Uniting for One Health" on July 6th & 7th on the occasion of World Zoonoses Day 2023 Organized by NCDC, New Delhi in collaboration with DAHD, MoEFCC, MoAFW, ICAR, and USAID RISE, Jhpiego in New Delhi.
12. Dr H. Singha participated in a Multi-stakeholder Consultation workshop to develop the State Action Plan for the Prevention and Control of Zoonotic Diseases for the state of Rajasthan organized by NCDC, New Delhi and DMHS, Rajasthan in technical collaboration with U.S. Centers for Disease Control and Prevention (CDC) on October 12th -13th, 2023, Jaipur, Rajasthan.
13. Dr H. Singha attended National Control programme for glanders in India: opportunities and challenges

Webinar on Glanders on August 30th, 2023, Tokyo WOAH project on international horse movement in Asia and the Pacific region, Organized by WOAH Regional Representative Asia Pacific.

14. Dr Naveen Kumar was invited as a Lead Speaker on "Vaccination against Lumpy Skin Disease "Current state of the art in, Veterinary Pathology Congress-2023 (IAVP), Organized by IVRI, Izatnagar, UP from December 20th -22nd, 2023.
15. Dr Naveen Kumar was invited as a Lead Speaker on "Vaccination against Lumpy Skin Disease". Current state of the art. In, VII Annual Convention of SVBBI and International Symposium on Multiomics to One Health: Challenges and Way Forward in Biomedical Research in Annual Conference of Society of Veterinary Biochemists and Biotechnologists of India (SVBBI), Organized by IVRI, Izatnagar from December 14th -15th, 2023.
16. Dr Naveen Kumar was invited for a lecture on "Sharing experience on development of Lumpy Skin Disease vaccine" in, One day seminar on sharing experience on development of Lumpy Skin Disease vaccine and role of IPR in entrepreneurship, organized by LUVAS, Hisar on November 14th, 2023.
17. Dr Naveen Kumar presented an invited lecture on "Coronavirus (SARS-CoV-2) infections in animals" in 21 Days Winter School On One Health Approach to Combat AMR, Zoonoses and Food Safety, Organized by Rajasthan University of Veterinary and Animal Sciences, Jaipur, Rajasthan from December 1st – 21st, 2023.
18. Dr Naveen Kumar presented a lead paper on Lumpi-ProVacInd: Development of LSD vaccine in India in 35th IAVMI conference on-Novel approaches on Animal Health for realizing one health mission" held on April 7th - 8th, 2023 at DGCN College of Veterinary and Animal Sciences, CSK HPKV, Palampur (HP).
19. Dr Naveen Kumar presented an invited lecture on "Vaccination against Lumpy skin disease in India" in a symposium organized by ICAR-National Institute for Veterinary Epidemiology and Disease Informatics (NIVEDI) in collaboration of Indian Virological Society on January 27th, 2022.
20. Dr Nitin Virmani presented a lead paper entitled "Development of recombinant vaccine candidate against EHV1 and equine influenza" in VIROCON-2023 on "Advancements in Global Virus Research towards One Health" organized by Indian Virology Society and held at ICAR-NRCB, Trichy on Dec 1st -3rd, 2023.
21. Dr Nitin Virmani presented a lead paper entitled "Development of recombinant virus-based vaccines against respiratory viral diseases of equines" in VIBCON2022 on "Leveraging emerging biotechnologies for one health" organized by Indian Society for Veterinary Immunology and Biotechnology and held at SKUAST, Srinagar, Kashmir on July 29th -31st, 2023.
22. Dr Nitin Virmani presented a lead paper entitled "Respiratory diseases of equines and modern vaccine approaches" In Veterinary Pathology Congress2023 on "Advances in Veterinary Pathology for Diagnosis and Control of Emerging Diseases of Livestock and Poultry held at ICAR-IVRI, Izatnagar, UP on December 20th - 22nd, 2023.
23. Dr Rajender Kumar attended 29th International Conference of the World Association for the Advancement of Veterinary Parasitology (WAAVP) held at Chennai during August 20th – 24th, 2023.
24. Dr R. K. Dedar attended three-days training on 'Instrumentation, technologies and application of sensors in animal husbandry mechanization' from December 18th -20th, 2023 at ICAR-CIAE Bhopal.
25. Dr Riyesh T has participated in the Annual Conference of IAAVR and National Symposium on "Livestock Health and Poultry: Enhancing Productivity for Sustainable Farmers' Livelihood" jointly organized by Lala Lajpat Rai University of Veterinary and Animal Sciences, Hisar-125004 (Haryana) and Indian Association for The Advancement of Veterinary Research (IAAVR) from February 7th- 8th, 2024.
26. Dr Riyesh T has participated in the 5th Biennial Poultry Health Conference and National Symposium on "Poultry Health: Current Challenges and Future Strategies" jointly organized by ICAR - Directorate of Poultry Research, Hyderabad and Association of Avian Health Professionals, Hyderabad during February, 23rd & 24th 2024.
27. Dr. Riyesh T participated in the XVI Agricultural Science Congress & ASC Expo, themed "Transformation of Agri-Food Systems for Achieving Sustainable Development Goals," held in 2023 at Kochi, Kerala from October 10th to 13th.
28. Dr R. K. Vaid presented a lead paper titled 'Bacteriophages for Aeromonas veronii control: isolation,

characterization and potential application in aquaculture pathogen management'' in the VIROCON 2023 'Advancements in Global Virus research towards One Health'' organised by NRC on Banana, Tiruchirapalli, held at Tiruchirapalli, Tamilnadu from December 1st-3rd, 2023.

29. Dr R. K. Vaid presented annual review progress report of NRCE, Hisar for Surveillance of AMR in Haryana'' held at 6th Annual Review meet of Indian Network for fisheries and animals antimicrobial resistance'' (INFAAR) at Bhuwaneshwar, Odisha on December 23rd, 2023.

30. Dr R.K. Vaid attended Workshop on 'Anthrax outbreak response and sample collection' organized by Odisha University of Agriculture & technology, Bhubaneshwar, Odisha under DBT Network Programme on Anthrax Diagnosis and Control in India, DBT at Deptt. of Preventive Medicine, College of Veterinary Science and Animal Husbandry, OUAT, Bhubaneshwar from March 2nd-4th, 2023

44. Dr Shanmugasundaram K delivered an expert lecture on "Sample collection, processing and dispatch for glanders diagnosis" during training programme for Field Veterinary Officers' Hands on Training Programme on Diagnosis and Control of Equine Glanders organised at ICAR-NRCE, Hisar, September 18th -22nd, 2023.

45. Dr Shanmugasundaram K attended Conference: XXXV Annual Convention and National Conference of Indian Association of Veterinary Microbiologists, Immunologists and Specialists in Infectious Diseases on Novel Approaches in Animal Health for Realizing One Health Mission organized by Department of Veterinary Microbiology Dr. G.C. Negi College of Veterinary and Animal Sciences, CSKHPKV, Palampur, (HP)- 176062, India from April 7th -8th, 2023.

46. Dr Shanmugasundaram K attended Anthrax outbreak response and sample collection at OUAT Bhubaneswar Odisha from March 2nd - 4th, 2023.

47. Dr Sanjay Barua presented a Lead paper on "p38 Mitogen-Activated Protein Kinase (MAPKs) as a target for antiviral drug development" In 35th Annual Conference of IAVMI on the theme "Novel Approaches in Animal Health for Realizing One Health Mission" organized by DGCN College of Veterinary and Animal Sciences, CSK HPKV, Palampur (HP) from April 7th -8th, 2023.

48. Dr Sanjay Kumar attended 29th International Conference of the World Association for the Advancement of Veterinary Parasitology (WAAVP) held at Chennai during August 20th - 24th, 2023.

49. Dr Sanjay Kumar organized Hands-on-workshop on *QSAR based Computational Drug Design and its Application* organized in collaboration with Altem Technologies Pvt. Ltd, Bangalore from July 6th to 8th, 2023 at ICAR-NRCE, Hisar.

50. Dr Taruna Anand presented an invited lecture entitled, "Phage Therapy to modulate MDR infections in Veterinary science" during 4th International Conference on Bacteriophage Research and antimicrobial resistance from September 28th -30th, 2023 organized at Madras University.

51. Dr Taruna Anand presented a lead talk entitled "Bacteriophage Therapy in lab animal models to explore the prevention of infections of poultry, aquaculture and veterinary industry", during VIROCON, 2023, Advancements In Global Virus Research Towards One Health organized from December 1st -3rd , 2023 in Tiruchirappalli, Tamil Nadu.

52. Dr TR Rao was invited to deliver a lead lecture on "Equine Embryo transfer in Indigenous horse breeds-Challenges and hurdles" at the 38th Annual Convention of ISSAR and International symposium on "Frontiers in Theriogenology: Research and Practice" conducted from December 6th to 8th, 2023 at College of Veterinary and Animal Sciences, Mannuthy, Thrissur, Kerala.

53. Dr TR Rao was invited to deliver an expert talk on "Semen Analysis using flowcytometry" on February 7th 2023 at Kamdhenu University, Anand, Gujarat conducted by Trust for education and Training in Cytometry (TETC) during 24thINDO-US Flow Cytometry Workshop (Vet Applications), organised from February 6th -7th, 2023.

54. Dr TR Rao was invited to deliver an expert talk on "Semen Analysis using flow cytometry" on September 7th, 2023, at Flow Cytometry workshop (Pakistan's 2nd International Flow Cytometry Workshop) from September 7th to 9th, 2023 held at Aga Khan University Hospital, Karachi.

55. Dr T.R. Rao attended XVI Agricultural Science Congress & ASC Expo 2023 organized by ICAR-Central Marine Fisheries Research Institute (CMFRI), Kochi from October 10th - 13th, 2023.

On-going

Research Projects

A. Equine Health

Sr. No.	Title	Team	From	To	PIMS Code/Page
1.	Surveillance, Monitoring and Control of Emerging and Existing Diseases of Equines	H. Singha*, R. Kumar, S. Kumar, N. Virmani, S. Barua, R.K. Vaid, R. Dedar, A. Manuja, Balvinder Kumar, K. Shanmugasundara m, Anubha Pathak, Ana Raj, Yash Pal and T. K. Bhattacharya	April 1997	Continuous Service Project	IXX00257
2.	Biomacromolecules based nanoscaffolds for wound healing using 3D printing	Anju Manuja*, Balvinder Kumar and Riyesh T.	Oct., 2020	Sept., 2023	IXX15412
3.	Immune responses and host-pathogen interaction analysis in *Burkholderia mallei* infected equines	H. Singha*, K. Shanmugasundara m,	Sept. 2022	Aug. 2025	-
4.	Bio-evaluation of target specific potential drug candidates against *Trypanosoma evansi* and analysis of conventional trypanocides drug-resistance	Rajender Kumar*, Sanjay Kumar and B.C. Bera	May, 2023	April, 2026	-
5.	Preparation and evaluation of zinc based polymeric composites as joint supplements for equines	Anju Manuja*	Oct., 2023	Sept., 2026	-

B. Equine Production

Sr. No.	Title	Team	Date of Start	Date of Completion	PIMS Code
1.	Characterization and recognition of Bhimthadi horse	SC Mehta* and Sachin D. Sorate	July, 2021	June, 2023	IXX15797
2.	Analysis of quantitative traits for genetic improvement of indigenous equines	SC Mehta*, RA Legha and J. Singh	April, 2021	March, 2026	IXX15798
3.	Conservation of Marwari Indigenous breed of Horse through AI	SC Mehta*, T.R Talluri and J. Singh	July, 2022	March, 2024	-

Sr. No.	Title	Team	Date of Start	Date of Completion	PIMS Code
4.	Cryopreservation of semen from indigenous horses and donkeys	T.R. Talluri*, T.K. Bhattacharya, S.C. Mehta, R.K. Vaid, RK Dedar & M Kutty	July, 2023	June, 2026	-
5.	Standardization of protocols for preparation of low dose frozen semen for deep horn intrauterine insemination in mares and jennies	T.R. Talluri*, T.K. Bhattacharya, S.C. Mehta, Dr. RK Dedar, M Kutty & J. Singh	Aug., 2023	Aug., 2026	-
6.	Clinical studies on the colitis and colic in Horses	RK Dedar*, T.R. Talluri, R.K. Vaid	Sept., 2023	Aug., 2026	-
7.	Analysis of fertility associated genes (ProAKAP4, PLCzeta, SPATA1, CRISP3, INHBA, ZAN) in fresh and frozen thawed semen of Equines	Muhammed Kutty*	Dec., 2023	Dec., 2026	-
8.	Diagnostic and therapeutic Studies on incidences of colitis in horses and horses clinically suffering from hind quarter weakness/ataxia leading to recumbency	RK Dedar*	Dec., 2023	Dec., 2026	-
9.	Establishing a model precision Donkey Farming and exploration of therapeutic and cosmetic values of donkey milk to enhance farmers' income	Anuradha Bhardwaj*	Dec., 2023	Dec., 2026	-

C. National Centre for Veterinary Type Culture

Sr. No.	Title	Team	Dura-tion	To	PIMS Code
1.	Authentication and accessioning of viruses of animal origin (Service Project)	Sanjay Barua*, Naveen Kumar, B.C. Bera, Riyesh T. and Taruna Anand	May, 2015	Service Project	IXX11882
2.	Phenotypic and genotypic authentication and preservation of network bacterial isolates	R.K. Vaid*, Taruna Anand, B.C. Bera, Riyesh T. and K. Shanmugasundaram	June, 2015	Service Project	IXX11884
3.	Isolation, characterization and generation of repository of Mycobacterium species	Shanmugasundaram K.*, R.K. Vaid, and B.C. Bera	Oct., 2017	March., 2023	IXX13994
4.	Development of repository of respiratory viruses of livestock and isothermal based diagnostics for rapid identification.	B.C. Bera*, Nitin Virmani, Taruna Anand and Riyesh T.	Aug., 2020	July 2023 extended upto March 2024	IXX15338

Sr. No.	Title	Team	Dura-tion	To	PIMS Code
5.	Indian network for fisheries and animal antimicrobial resistance (INFAAR)	R.K. Vaid*, Taruna Anand, H.S. Singha and Anubha Pathak	June, 2018	March., 2025	IXX15418
6.	Isolation and characterization of bacteriophages against important biofilm forming bacteria (Service Project)	Taruna Anand*, Nitin Virmani, B.C. Bera, and RK Vaid	Dec, 2023	Dec, 2026	IXX15795
7.	A study on bat virome for unravelling the viral diversity in India	Riyesh T*, Naveen Kumar, Shanmugasundaram K, RK Vaid and Sanjay Barua	April, 2021	March, 2024	IXX16007
8.	Adaptation of Lumpy skin disease virus in Vero cells	Naveen Kumar*, Riyesh T and Sanjay Barua	Jan., 2021	March, 2025	IXX16675
9.	Isolation, characterization and development of repository of Infectious Laryngotracheitis virus of Poultry	B.C. Bera*, T.K. Bhattacharya, Nitin Virmani, Taruna Anand & Dr. Riyesh T	Aug., 2023	July, 2025	-
10.	Genetic characterization and evaluation of the immunogenicity of swinepox virus in mice model	Riyesh T.* Shanmugasundaram K, Sanjay Barua and T.K. Bhattacharya	Jan., 2024	Dec., 2026	-
11.	Evaluation of pathogenicity of accessioned bacterial strains by small animal experimental studies-Service Project	R.K. Vaid*	Dec., 2023	-	-

D. External Funded Projects

Sr. No.	Title	Team	From	To	PIMS Code
1.	All India Coordinated Research Project on Utilization of Animal Energy with enhanced system efficiency (AICRP on UAE)	R A Legha* and Yash Pal	July, 2009	March, 2023	OXX004 86
2.	National One Health Program on Prevention and Control of Zoonotic Diseases (NOHPPCZ) Project: Regional Coordination center under program for InterSectoral Coordination for prevention and control of Zoonotic Diseases	Bacterial Diseases: Harisankar Singha, Shanmugasundaram K, Anubha Viral Diseases: Dr Naveen Kumar, Dr Riyesh T.	June, 2019	March, 2024	OXX046 86
3.	Epidemiological studies and development of antiviral therapeutics against coronaviruses	Naveen Kumar, Riyesh T & Shanmugasundar am K	June, 2021	May, 2024	OXX493 5

Sr. No.	Title	Team	From	To	PIMS Code
4.	Validation and translation of the vaccines as well as diagnostic technologies developed in Phase-I of ADMaC".	Nitin Virmani*, Anubha Pathak & Riyesh T.	April, 2021	March, 2024	OXX494 0
5.	Development of ML and ANNbased breed and individual identification system for equine population differentiation	Anuradha Bhardwaj*, T.K. Bhattacharya, Yash Pal, R.A. Legha and T.R. Talluri	July, 2020	June, 2025	OXX501 2
6.	DBT Network Programme on Anthrax Diagnosis and Control in India	R.K. Vaid*, B.C. Bera, K. Shanmugasundar am	Sept., 2021	Sept., 2024	OXX538 3
7.	Development of Diagnostics for Coronavirus infections	Nitin Virmani*, B.C. Bera, & Taruna Anand	June, 2021	May, 2023	OXX511 1
8.	Studies on host pathogen interaction and development of vaccine against zoonotic coronaviruses	B.C. Bera*, Nitin Virmani & Taruna Anand	June, 2021	May, 2024	OXX538 2
9.	Surveillance of Rotavirus a Genotypes in bovine and Equines of India for Identification of Potential vaccine candidates	Anubha Pathak* and B.C. Bera	April, 2022	March, 2025	OXX536 8
10.	Optimisation of procedures for non-surgical recovery and biobanking of Marwari breed horse embryos	T.R. Talluri*, Yash Pal, RA Legha & RK Dedar	April, 2022	March, 2025	OXX536 9
11.	Utilization of desert plants for the treatment of skin diseases of Horses	R.K. Dedar*, RA Legha, Yash Pal, TR Talluri & Naveen Kumar	April, 2022	March, 2025	OXX537 0
12.	Translation of Nano based quinapyramine sulphate formulation into product and its evaluation against Trypanosoma evansi in animals.	Anju Manuja*, Rajender Kumar and Balvinder Kumar	April, 2022	March, 2025	OXX538 8
13.	Development of vaccine against animal's haemoprotozoan parasites for mitigating biotic stress	Sanjay Kumar*, Rajender Kumar & K. Shanmugasundaram	Oct., 2022	May, 2025	OXX544 5
14.	Isolation, identification and characterization of SARS-CoV-2 from sewage and domestic wastewater and COVID-19 patients from Hisar (Haryana)	Naveen Kumar* & Riyesh T.	June, 2022	June, 2023	-
15.	Development and evaluation of genetically engineered vaccine candidates for African swine fever, Equine Herpes virus-1 and Equine Influenza (clade 1 & 2)'	Nitin Virmani* Sanjay Barua, Naveen Kumar, BC Bera & Taruna Anand	June, 2022	May, 2025	-

Sr. No.	Title	Team	From	To	PIMS Code
16.	CRP on Vaccine and Diagnostics: "Development and validation of multiplex assays for laboratory diagnosis of emerging equine herpesviruses (EHV2 & EHV5)"	Nitin Virmani*, BC Bera & Taruna Anand	Jan., 2021	Dec., 2024	-
17.	CRP on Vaccine and Diagnostics "Development of antigen detection point-of-care diagnostics for haemoprotozoan diseases of equines"	Sanjay Kumar* & Rajender Kumar	Jan., 2021	Dec., 2024	-
18.	CRP on Vaccine and Diagnostics: "Development of point of care diagnostics for strangles in equines "	Balvinder Kumar*, RK Vaid, Anju Manuja, K Shanmugasundaram & Harisankar Singha	Jan., 2021	Dec., 2024	-
19.	CRP on vaccine & Diagnostic project: Development of RPA-LFA based point-of-care diagnostic assay for rapid detection and differentiation of equine herpes viruses 1&4	B.C. Bera* & Nitin Virmani	Jan., 2021	Dec., 2024	-
20.	Developing novel therapeutic strategies for mitigating antimicrobial resistance	Taruna Anand*, Nitin Virmani, B.C. Bera, R.K. Vaid	June, 2022	May, 2025	-
21.	National Fellow Project: "Bacteriophage based interventions for therapy and prophylaxis against" Campylobacteriosis, Colibacillosis, Salmonellosis and other diseases as an alternative to antibiotics in poultry.	Taruna Anand*	Oct., 2022	Oct., 2027	-
22.	Development of novel semen extender for the enhancement of post thaw semen quality in equines	T.R. Talluri* Dr. Yash Pal, RA Legha, RK Dedar, Anuradha Bhardwaj and TK Bhattacharya	Feb., 2023	Feb., 2025	-
23.	Integrated analysis for the ultradeep compositional characteristics of donkey colostrum and mature milk	Anuradha Bhardwaj* Yash Pal, parveen Malik, R.A. Legha & T. Rao	Feb., 2023	Jan., 2026	-
24.	Development and evaluation of immunotherapy and vaccine constructs against *Rhodococcus equi* infection to protect foals from pneumonia	H. Singha* R.K. Dedar & Shanmugasundaram K.	Sept., 2023	Aug., 2026	-
25.	Histone acetylation/deacetylation: Potential target for therapeutic intervention and vaccine development	Naveen Kumar* and Sanjay Barua	Sept., 2023	Aug., 2026	-

Staff at
ICAR-NRCE

(as on 31.12.2023)

Director: Dr. Tarun Kumar Bhattacharjya

S.No.	Name	Designation
Scientific Staff		
ICAR-NRCE Main Campus, Hisar		
1.	Dr. Nitin Virmani	Head, Division of Equine Health, NRCE, Hisar
2.	Dr. Rajender Kumar	Principal Scientist
3.	Dr. Sanjay Kumar	Principal Scientist
4.	Dr. Anju Manuja	Principal Scientist
5.	Dr. Balvinder Kumar	Principal Scientist
6.	Dr. Anuradha Bhardwaj	Senior Scientist
7.	Dr. Harishankar Singha	Senior Scientist
8.	Dr. Anubha Prashant Pathak	Scientist
9.	Ms. Ana Raj J.	Scientist
NCVTC, Hisar		
1.	Dr. Naveen Kumar	Head, NCVTC, NRCE, Hisar
2.	Dr. Sanjay Barua	Principal Scientist
3.	Dr. RK Vaid	Principal Scientist
4.	Dr. Taruna Anand	National Fellow
5.	Dr. BC Bera	Senior Scientist
6.	Dr. Riyesh T.	Sr. Scientist
7.	Dr. K. Shanmugasundaram	Sr. Scientist
Equine Production Campus, Bikaner		
1.	Dr. SC Mehta	Head, EPC, NRCE, Bikaner
2.	Dr. RK Dedar	Senior Scientist
3.	Dr. TR Talluri	Senior Scientist
4.	Dr. Muhammed Kutty V.H.	Scientist
Technical Staff		
ICAR-NRCE Main Campus, Hisar		
1.	Sh. Sita Ram	ACTO
2.	Sh. Sanjeev Kumar	ACTO
3.	Sh. Ajmer Singh	ACTO

Sr.No.	Name	Designation
4.	Sh. Joginder Singh	STO
5.	Sh. Mukesh Chand	STO
6.	Sh. Brij Lal	Technical Officer
7.	Sh. Sajjan Kumar	Technical Officer
8.	Sh. Suresh Kumar	Technical Officer
9.	Sh. Raj Kumar Dayal	Technical Officer
10.	Sh. Raghbir Singh	STA
Equine Production Campus, Bikaner		
1.	Dr. Jitendar Singh	CTO
2.	Sh. Kamal Kumar Singh	ACTO
3.	Dr. RA Pachori	ACTO
4.	Sh. Narender Chauhan	ACTO
5.	Sh. Om Parkash	Technical Officer
6.	Sh. Satya Narayan Paswan	Technical Officer
7.	Sh. Gopal Nath	Technical Assistant
8.	Sh. Rajender Singh	Technical Assistant
Administrative Staff		
ICAR-NRCE Main Campus, Hisar		
1.	Sh. Pawan Kumar	AO
2.	Sh. Subhash Chander	AAO
3.	Sh. Sunil	AAO
4.	Sh. Ashok Kumar	Private Secretary
5.	Sh. Dinesh Datt Sharma	Assistant
6.	Sh. Om Parkash	Assistant
7.	Sh. Deepak Kumar	UDC
8.	Sh. Guru Dutta Sharma	LDC
9.	Sh. Ishwar Chander	LDC
10.	Sh. Mahender Singh	UDC (EPC, Bikaner)
Skilled Support Staff		
ICAR-NRCE Main Campus, Hisar		
1.	Sh. Jai Singh	Skilled Support Staff
2.	Sh. Hanuman Singh	Skilled Support Staff
3.	Sh. Subhash Chander	Skilled Support Staff
4.	Sh. Ishwar Singh	Skilled Support Staff
5.	Sh. Ram Singh	Skilled Support Staff

S.No.	Name	Designation
6.	Smt. Santra	Skilled Support Staff
7.	Sh. Lilu Ram	Skilled Support Staff
8.	Sh. Sant Ram	Skilled Support Staff
9.	Smt. Soma Devi	Skilled Support Staff
ICAR-NRCE, Equine Production Campus, Bikaner		
10.	Sh. Mahabir Prasad Meena	Skilled Support Staff
11.	Sh. Raju Ram	Skilled Support Staff
12.	Sh. Ashok Kumar	Skilled Support Staff

भाकृअनुप
ICAR

ICAR-NATIONAL RESEARCH CENTRE ON EQUINES

समाचार पत्रों में एनआरसीई

राष्ट्रीय अश्व अनुसंधान केंद्र के 39वें स्थापना दिवस पर कार्यक्रम आयोजित

जो काम मशीनें नहीं कर पाती वह अश्व कर सकते हैं: कीमोथी

हिसार | एएसआरबी नई दिल्ली के सदस्य डॉ. शिव प्रसाद कीमोथी ने कहा कि जो काम मशीनें नहीं कर पाती वह अश्व कर सकते हैं। नई टेक्नोलॉजी आने से अश्वों का रोल कम हुआ है मगर इनका संरक्षण जरूरी है। एनआरसीई के 39वें स्थापना दिवस पर हॉर्स शो में नवलगढ़ से पहुंचे प्रशिक्षक मातादीन के मारवाड़ी घोड़े अली ने सबका मन मोह लिया। हॉर्स शो में डूंडलोद हनुमानगढ़ के घोड़े समेत कई एक करोड़ से ज्यादा लागत पहुंचे। इस दौरान दौड़ते घोड़ों पर करतब दिखाए।

हिसार, रविवार, 26 नवंबर, 2023 | dainikbhaskar.com

राष्ट्रीय अश्व अनुसंधान केंद्र हिसार का 39वां स्थापना दिवस आज

लम्पी की वैक्सीन बना बचाई लाखों पशुओं की जान, गधों से जुतवा दिए खेत, गधी के दूध से बनाया ब्यूटी प्रॉडक्ट

जानिए... यह पांच उपलब्धि जो एनआरसीई ने देश को दी

शिविर में पशु जन्य रोगों पर विशेषज्ञों को दिया प्रशिक्षण

आईसीएआर-एनआरसीई को डिवीजनों के नए प्रमुख मिले

हिसार| आईसीएआर एनआरसीई के निदेशक डॉ. टीके भट्टाचार्य ने बताया हाल ही में आईसीएआर-नेशनल रिसर्च सेंटर ऑन इक्विन्स, हरियाणा में डिवीजन के नए चयनित प्रमुखों एनिमल हेल्थ यूनिट के प्रमुख के डॉ. नितिन विरमानी ने रूप में काम करना शुरू किया। साथ ही डॉ. नवीन कुमार को आईसीएआर- एनआरसीई, हिसार में एनसीवीटीसी के प्रमुख के रूप में चुना गया। उन्होंने दोनों विभागों के प्रमुखों का नए पदों पर स्वागत भी किया। नव नियुक्त प्रमुख अनुसंधान के अपने क्षेत्रों में प्रसिद्ध वैज्ञानिक हैं। डॉ. नितिन विरमानी एक प्रसिद्ध पैथोलॉजिस्ट हैं जो इक्वाइन इन्फ्लुएंजा, इक्वाइन हर्पीस वायरस और सरस-कोव-2 के लिए काम कर रहे हैं। उन्होंने अपनी टीम के साथ हाल ही में कैनाइन में कोरोना संक्रमण का पता लगाने के लिए कैन कोव की एक किट विकसित की है। डॉ. नवीन कुमार भी एक विश्व स्तरीय वैज्ञानिक हैं जिन्होंने कोरोना वायरस के लिए एनकोवेक्स और गांठदार त्वचा रोग (लंपी स्किन) नाम से वैक्सीन विकसित की है। संभागों के इन गतिशील प्रमुखों के प्रयासों से संस्थान और आगे बढ़ेगा। समस्त स्टाफ इन अधिकारियों का स्वागत किया व शुभकामनाएं दी।

शिविर में पशु जन्य रोगों पर विशेषज्ञों को दिया प्रशिक्षण

लुवास में वैक्सीन बनाने और पेटेंट को लेकर सेमिनार किया आयोजित

राष्ट्रीय उद्यमशीलता दिवस एवं विश्व विज्ञान दिवस के उपलक्ष्य में कार्यक्रम

हिसार | लुवास में राष्ट्रीय उद्यमशीलता दिवस एवं विश्व विज्ञान दिवस के उपलक्ष्य में प्राध्यापकों एवं विद्यार्थियों को वैक्सीन बनाने की संपूर्ण प्रक्रिया एवं पेटेंट व उद्यमशीलता पर जागरूक करने हेतु एक दिवसीय सेमिनार का आयोजन किया गया। अनुसंधान निदेशक डॉ. नरेश जिंदल मुख्य अतिथि रहे। उन्होंने कहा कि अनुसंधान कार्य पशुओं व किसानों के लाभ हेतु लक्षित होना चाहिए।

इस अवसर पर मानव संसाधन प्रबंधन निदेशक डॉ. राजेश खुराना ने बताया कि सेमिनार लुवास के संस्थागत नवप्रवर्तन परिषद द्वारा आयोजित करवाया जा रहा है। आमंत्रित वक्ता, प्रधान वैज्ञानिक एवं विभागाध्यक्ष वीटीसी, राष्ट्रीय अश्व अनुसंधान केंद्र डॉ. नवीन ने लम्पी त्वचा रोग एवं उससे रोकथाम के लिए एनआरसीई में निर्मित वैक्सीन के निर्माण प्रक्रिया के विषय में विस्तार से वर्णन किया। इस अवसर पर उपस्थित दूसरी वक्ता, अधिवक्ता एवं पेटेंट एजेंट, संस्थापक स्वाश सेंगल कंसलटेंट डॉ. भारती ने उद्यमशीलता के विषय में कहा कि विद्यार्थियों को अनुसंधान उद्यमशीलता के लिए प्रशिक्षण एवं आर्थिक सहायता द्वारा प्रोत्साहित किया जाना चाहिए। सेमिनार में तकनीकी सत्र सह अध्यक्ष डॉ. नरेश कक्कड़, संयोजक डॉ. विशाल शर्मा, डॉ. शालिनी शर्मा एवं डॉ. रचना आदि उपस्थित रहे।

भास्कर खास • एनआरसीई में ग्लैंडर्स का सर्विलेंस एवं प्रयोगशाला निदान पर कार्यशाला

ग्लैंडर्स को लेकर सर्विलेंस बढ़ाने पर दिया बल

तामिलनाडु की टीम ने हासिल की पूरी जानकारी

... एंजाइम-लिंक्ड इम्यूनोसॉर्बेंट परख (एलिसा) और पॉलीमरेज चेन रिएक्शन (पीसीआर) जैसी तकनीकों को सीखने और व्यक्तिगत अभ्यास करने का अवसर मिला।

अश्व अनुसंधान केन्द्र बीकानेर ने हासिल की बड़ी उपलब्धि

भ्रूण स्थानांतरण प्रौद्योगिकी से उत्पादित देश की पहली मारवाड़ी बछेड़ी

देश में अश्व प्रजाति पर संकट, 19% से अधिक गिरावट

देश में उपयोगिता के लिए बाजार नहीं मिलने के कारण अश्व प्रजाति की संख्या में 19 प्रतिशत से अधिक गिरावट आई है। इसमें अश्व प्रजाति में घोड़े, खच्चर व गधे शामिल हैं। यह गिरावट साल 2012 से साल 2019 के दौरान की पशु गणना में दर्ज की गई है। ऐसे में अश्व प्रजाति के संरक्षण को लेकर राष्ट्रीय अश्व अनुसंधान केंद्र में कार्यक्रम हुआ। यह आयोजन एनआरसीई के 39वें स्थापना दिवस पर किया गया जिसमें वैज्ञानिक से लेकर अश्व पालक तक एक मंच पर आए। कार्यक्रम में एएसआरबी के सदस्य शिव प्रसाद ने मुख्य अतिथि के रूप में शिरकत की। कार्यक्रम एनआरसीई के डायरेक्टर टीआर भट्टाचार्य के नेतृत्व में हुआ जबकि संचालन विभागाध्यक्ष डा. नीतिन विरमानी ने किया।

सिरसा रोड स्थित राष्ट्रीय अश्व अनुसंधान केंद्र के वार्षिक स्थापना दिवस समारोह के दौरान मंच पर उपस्थित मुख्यातिथि।

अश्व प्रजाति में कमी का ये अहम कारण

- पूर्व में परिवहन के लिए अश्व प्रजाति के पशुओं का कम प्रयोग।
- इनकी उपयोगिता में कमी आना।
- अश्व प्रजाति के खेल देश में अधिक लोकप्रिय न होना।
- अश्व के देखरेख में खर्च भी अहम वजह है
- ग्लैंडर्स की बीमारी इत्यादि भी कारण।

अश्व प्रजाति बचाव में ये लिए फैसले

- जिस जगह ग्लैंडर्स की बीमारी होने के कारण 25 किलोमीटर के दायरे में मेले या अन्य कार्यक्रम नहीं होते थे। अब उस दायरे को घटाया जाएगा। ताकि अश्व मेले अधिक लग सकें।
- ग्लैंडर्स बीमारी मिलने पर दूसरे जिले में भी अश्व कार्यक्रम पर प्रतिबंध हटाने का कदम उठाया जाएगा।
- ग्लैंडर्स या दूसरी बीमारी पाए जाने पर सतर्कता व सर्विलेंस बड़े स्तर पर की जाए।
- उपयोगिता बढ़ाने की दिशा में कार्य किया जाएगा।
- अश्व व पशु पालकों और किसानों अश्व पालन के लिए जागरूक किया जाएगा।

देश में अश्व प्रजाति की संख्या में 19 प्रतिशत कमी आई है। अश्व की प्रजाति को उपयोगिता का बाजार नहीं मिलने से यह कमी बढ़ रही है। इस कमी को बढ़ने से रोकने और अश्व प्रजाति की संख्या बढ़ाने पर मंथन हुआ है।

डा. नीतिन, वैज्ञानिक, एनआरसीई, हिसार।

हिंदी का अधिक प्रयोग करना हमारी कार्यशैली उत्तम बनाएगा : डॉ. भट्टाचार्य

हिसार | राष्ट्रीय अश्व अनुसंधान केंद्र में हिंदी पखवाड़ा मनाया गया। हिन्दी पखवाड़े के उद्घाटन समारोह के दौरान दो हिंदी कार्यशालाओं का आयोजन किया गया। इसमें डॉ. मोनिका कक्कड़ और नरेश शर्मा ने व्याख्यान दिया। इस उपलक्ष्य पर डॉ. टीके भट्टाचार्य, निदेशक राष्ट्रीय अश्व अनुसंधान केन्द्र ने बताया कि हिंदी भाषा का अधिक प्रयोग करना हमारी कार्यशैली को उत्तम बनाएगा। हिंदी जन जीवन की भाषा है और अति उपयोगी है। कार्यक्रम में अश्व स्वास्थ्य विभाग प्रमुख डॉ. नितिन विरमानी, एनसीवीटीसी प्रमुख डॉ. नवीन कुमार, डॉ. तरुणा आनंद मौजूद रहे।

अश्वों के लिए लगाया स्वास्थ्य कैंप

हिसार | राष्ट्रीय अश्व अनुसंधान केंद्र द्वारा गांव पाबड़ा में किसान दिवस मनाया गया। आईसीएआर ने पाबड़ा गांव में अश्व और पशुपालकों के लिए एक स्वास्थ्य शिविर का आयोजन किया। एनआरसीई टीम ने अश्व और पशुपालकों को लाभ पहुंचाने के लिए संस्थान द्वारा विकसित विभिन्न गतिविधियों और प्रौद्योगिकियों के बारे में जानकारी दी। स्वास्थ्य शिविर में लगभग बारह घोड़ों और पशुधन मालिकों ने भाग लिया और उनके स्वास्थ्य की स्थिति का आंकलन करने के लिए घोड़ों और अन्य पशुओं की जांच की गई।

अश्व अनुसंधान केंद्र में ढोलक की थाप पर नाचा राजस्थान का अली

कार्यक्रम में पहुंचे एक-एक करोड़ की कीमत के घोड़े

एनआरसीई के स्थापना दिवस कार्यक्रम में राजस्थान के हनुमानगढ़ और झुंझनू जिले के एक-एक करोड़ के घोड़े पहुंचे।

बगुलों को काटकर इंसानों में जापानी बुखार फैला रहे मच्छर

जागरण विशेष

तीन तरह से किया जा सकता है डायग्नोज

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