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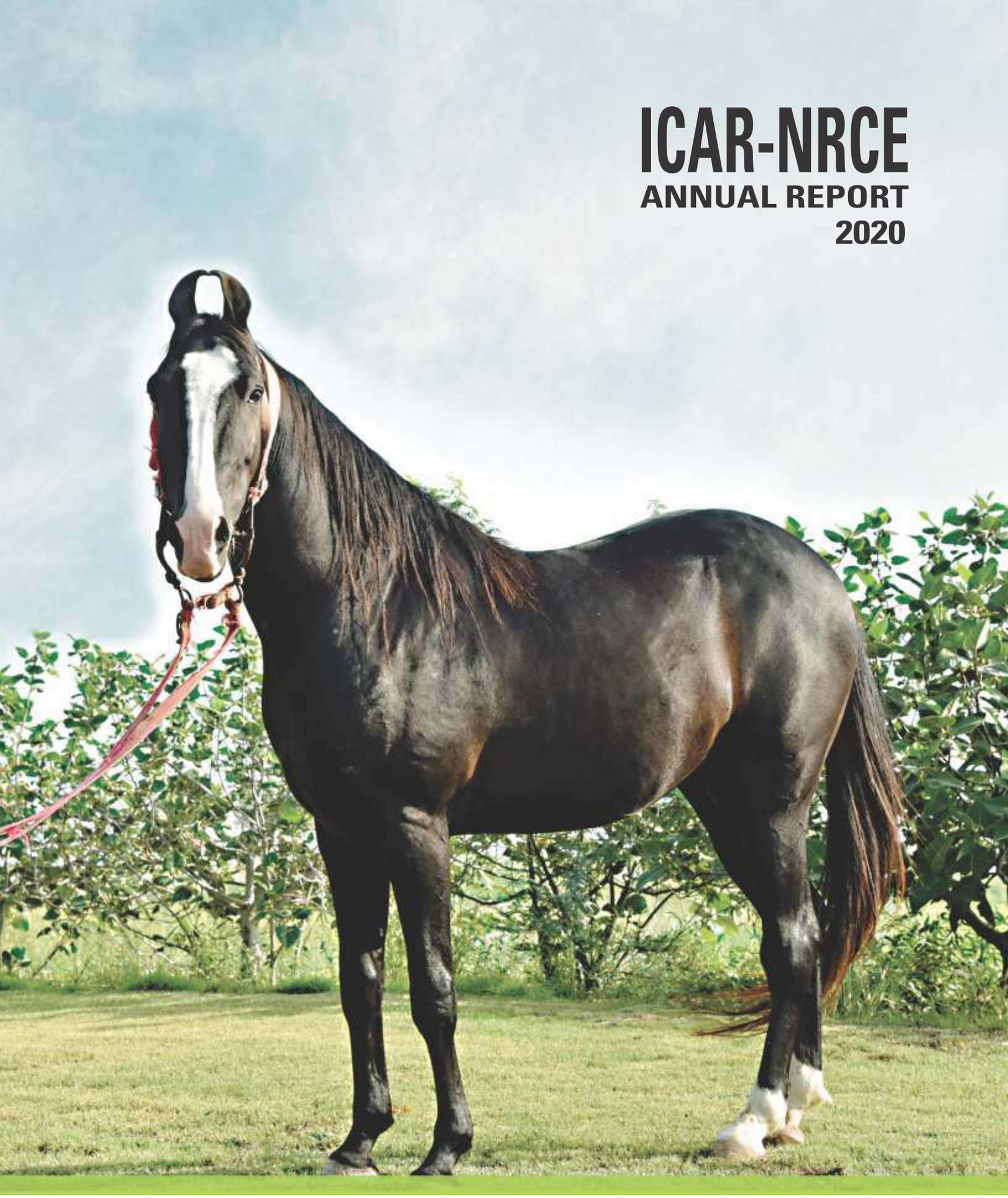
ANNUAL REPORT
2020



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

ICAR-NATIONAL RESEARCH CENTRE ON EQUINES

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The achievements and activities of the Centre from Jan.2020 to Dec. 2020 are presented in this report. Mention of trademark, proprietary product or firm in the report does not constitute an endorsement or rejection of other suitable products or firms.

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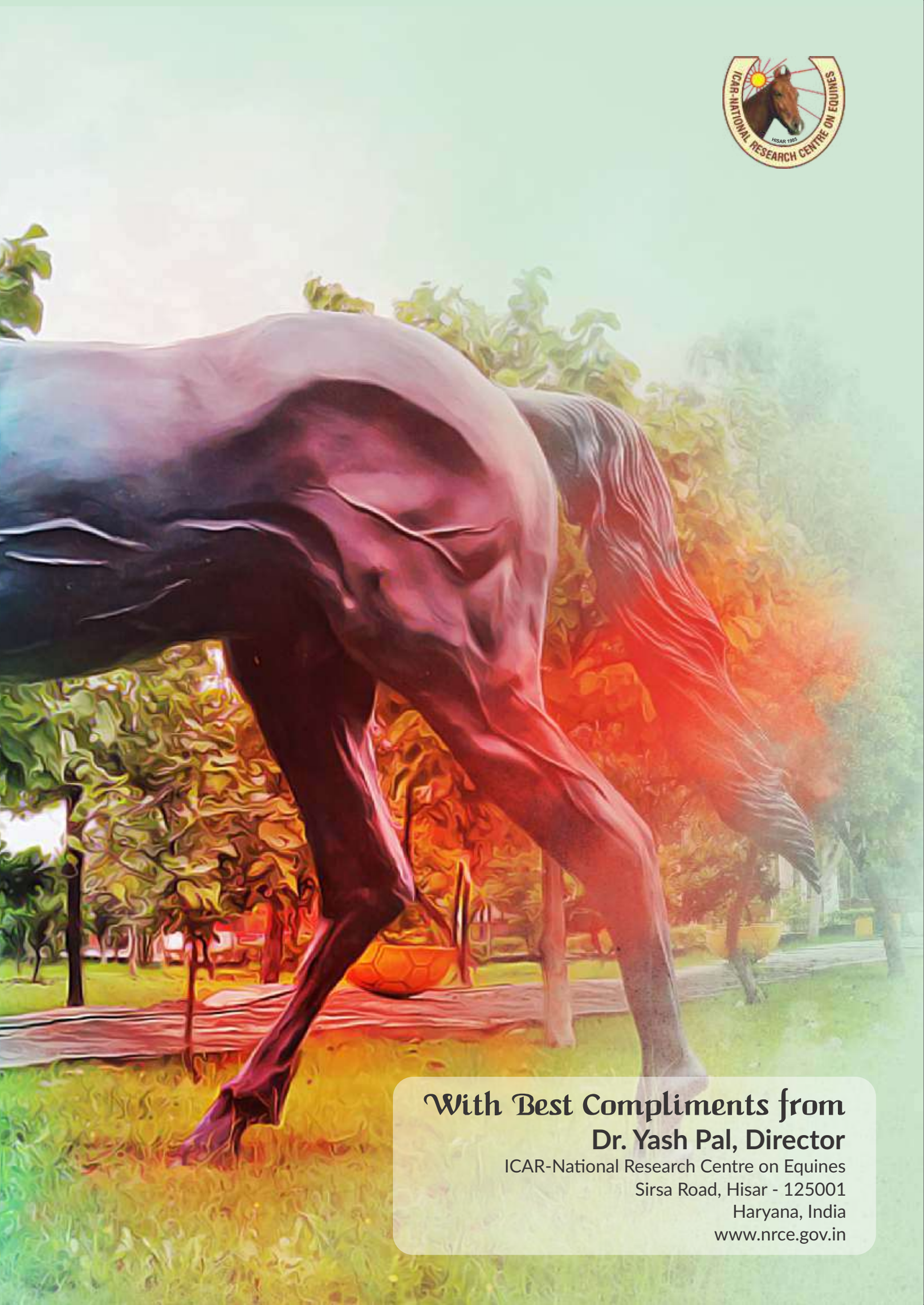

With Best Compliments from

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About the Cover

“The stallion Prince is of Marwari horse breed reared at NRCE-EPC campus Bikaner. Marwari or Malani is a breed of horse from the Marwar region of India. These horses have majestic look with high-carried head, peculiar character of inward curved and touching ears. The breeding tract of this breed is mainly Jodhpur, Jalore, Sirohi, Jaisalmer, Barmer, Rajsamand and Udaipur area of Rajasthan and in some pockets of Gujarat.”

Content

ANNUAL REPORT

Director's FOREWORD

The year 2020, was challenging due to COVID-19 pandemic. However, the Centre extended it's human resources and laboratories for diagnosis of the diseases to make a substantial contribution in the country. ICAR-NRCE being a premier research institute of ICAR was bestowed with an important responsibility of COVID-19 testing which was carried out with full zeal and enthusiasm by the scientists and staff. The Centre is actively working in the areas of equine vaccines, diagnostics, therapeutics, equine management, production and reproduction. It has attained national and international recognition through quality research. The Centre is also gaining appreciation for the research efforts in emerging areas of the new generation vaccines, nanotechnology, host-pathogen interaction studies, ethno-veterinary medicine, repository of novel and emerging microbes including pathogenic viruses and bacteria as well as bacteriophages. The institute strives hard to serve the poorest of the poor farmers by providing timely help through services like artificial insemination, disease diagnosis and active surveillance. The National Action Plan for control and eradication of glanders in India developed by ICAR-NRCE has been launched by the Ministry of Fisheries, Animal Husbandry and Dairying, GOI. For nation-wide invigilation of equine diseases a total of 229 equine serum samples from 10 states were tested for diseases like Equine Infectious Anemia (EIA), Equine Influenza (EI), Equine Herpes Virus (EHV-1), Japanese Encephalitis (JE)/ West Nile, Trypanosomosis, Equine piroplasmosis, Salmonellosis and Brucellosis. Also under disease investigation 1001 samples were tested. A total of 22130 equine sera from 258 districts of 16 states were tested for glanders. Under the contractual diagnostic services, a total of 6775 samples from race industry, turf clubs, riding schools, quarantine animals and other organized sectors were tested for diseases. The revenue generated from sample testing was Rs 49.10 lakh for the year.

During the year 2020, the genetic diversity analysis of 39 *Burkholderia mallei* isolates was performed to reveal the recent circulating isolates in India and MLST typing of 41 isolates of *Streptococcus equi* and *S. zooepidemicus* was carried out which suggested the presence of novel strains in India. To decipher the diverse strains of EHV-1, whole genome sequencing of abortigenic EHV-1 isolates was carried out and genome analysis suggested the diversity of Indian strains in comparison to previously reported. Taqman probes based real time PCR assays were developed to detect the glycoprotein-B gene of EHV-2 & 5 and the assays are useful for the detection of these viruses in the upper respiratory tract of asymptomatic young horses.

In an endeavor to develop the refined modified live virus vaccines, EHV-1 deletion mutants were developed, and their pathology and attenuation studies were carried out in mouse models. The EHV-1 virus constructs provided adequate immune response and are prospective candidates for large animal studies.

In order to develop alternatives to antibiotics, efficacy of novel ZnO nanoparticles against *S. equi* was also carried out. The extracts of *Lawsonia inermis* Linn were analyzed for constituents to develop suitable herbal

formulations with reduced toxicity. Also leaf extracts of *Aerva Javanica* showed efficacy against the melanoma and warts in horses. The research on development of drugs against *Trypanosma evansi* confirmed the anti-trypanosomal activity and apoptotic mechanism of naphthoquinones in axenic cultures of *T. evansi*.

The Centre has undertaken endurance and fertility analyses in indigenous horses using SNP markers and recorded biometry of horses. In order to address farmers demands Nukra male were purchased and semen straws of selected horses were provided to field veterinarians for AI. The biochemical and hormonal profiles of Marwari stallion semen and their kinematic parameters were recorded to perfect the semen cryopreservation technology. Also, looking at the increased demand of donkey milk in-depth analysis of milk parameters has been carried out.

During the year, NCVTC was recognized as the national repository by the Ministry of Environment, Forests and Climate Change under Biological Diversity Act 2002. A total of 204 microbes were accessioned including Lumpy Skin Disease Virus and SARS-CoV2. Studies on apigenin against BPXV and emetine against SARS-CoV2 were carried out. The etiological agents of Pullorum disease and Fowl Typhoid were used for comparative genomics and the analysis of salmonella strains provided basis for underlying pathogenesis and virulence of this microbe. Novel bacteriophages were also isolated and phage therapy was extended in mastitis mouse models.

The scientific contributions under 33 research projects, including 12 externally funded projects, lead to publication of 51 research papers, development of diagnostic kits, generation of value added products and grant of Indian patent along with providing service to nation through recognized COVID-19 diagnostic facility.

I extend heartful thanks to the sincere efforts of scientist and staff for being there and offering services during this difficult year. I congratulate publication committee for bringing out this excellent annual report.

I heartfully acknowledge the support extended by Dr. Trilochan Mohapatra, Secretary DARE and Director General ICAR, Dr. B.N. Tripathi, DDG (Animal Sciences) and many thanks are also due to Dr. Ashok Kumar, ADG (AH), Principal Scientists, Dr. Jyoti Misri, Dr. Rajan Gupta, Dr. Vineet Bhasin at ICAR headquarters for their support to ICAR-NRCE.

(Yash Pal)

Executive Summary | कार्यकारी सारांश

Horses have been domesticated since historic times. They hold a special place in culture and rise of empires as they played a key role in the rise of larger human settlements and great civilizations. Horses, donkeys, ponies and mules collectively referred to as equines have a wide range of utilities including sports, army, police, and as working animals. They have been regarded as most faithful companion animals for the human beings. Mechanisation has taken over the role of equines to a greater extent resulting in decline in their population. However, they still remain preferred means of transport in hilly terrains of India. Mules and donkeys have been a better choice for carrying packs in areas where road networks are still a distant dream.

Considering the importance of the species and limited healthcare facilities available in India the Indian Council of Agricultural Research established National Research Centre on Equines (NRCE) on November 26th, 1985 at Hisar (Haryana). The state-of-the art laboratories and facilities at the main campus of NRCE in Hisar have been undertaking research in the areas of equine virology, bacteriology, parasitology, immunology, pathology, medicine, biochemistry and biotechnology. 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.

A sub campus of NRCE was established in 1989 at Bikaner (Rajasthan) to undertake research on equine production, genetics and breeding, management, reproduction, physiology and nutrition. Equine Production Campus at Bikaner has well maintained nucleus herds of Marwari, Kathiawari, Zanskari and Manipuri horses and Halari and exotic donkeys. The National Centre for Veterinary Type Cultures (NCVTC) was established in the year 2005 at ICAR-NRCE, Hisar main campus for collection, characterization, preservation and distribution of microbes of animal origin having veterinary importance. Presently, the

घोड़ों को ऐतिहासिक काल से पालतू बनाया जाता रहा है। अश्वों का संस्कृतियों और साम्राज्यों के उदय में एक विशेष स्थान रहा हैं क्योंकि उन्होंने मानवता एवं महान सभ्यताओं के विकास में महत्वपूर्ण भूमिका निभाई है। घोड़ों, गर्दभों, टट्टुओं और खच्चरों को सामूहिक रूप से 'अश्व' अभिहित किया जाता है। अश्वों की खेलों, सेना, पुलिस और बोझा ढोने सहित विभिन्न उपयोगिताएं हैं। इन्हें इंसानों के लिए सबसे वफादार जानवर माना गया है। मशीनीकरण ने काफी हद तक अश्वों की भूमिका को सीमित कर दिया है, जिसके परिणामस्वरूप उनकी आबादी में गिरावट आई है। हालांकि, वे अभी भी भारत के पहाड़ी और रेगिस्तानी इलाकों में परिवहन के पसंदीदा साधन बने हुए हैं। खच्चर और गर्दभ उन क्षेत्रों में सामग्री ले जाने के लिए बेहतर विकल्प हैं जहां सड़कें अभी भी एक दूर का सपना है।

भारत में अश्वों के लिए सीमित स्वास्थ्य सुविधाओं को ध्यान में रखते हुए, भारतीय कृषि अनुसंधान परिषद ने 26 नवंबर, 1985 को हिसार (हरियाणा) में राष्ट्रीय अश्व अनुसंधान केंद्र (रा.अ.अनु.के.) की स्थापना की। यह केन्द्र अश्व विषाणु विज्ञान, जीवाणु विज्ञान, पशु औषधि विज्ञान, परजीवी विज्ञान, रोग प्रतिरोध विज्ञान, विकृति विज्ञान, जैव रसायन एव जैव प्रौद्योगिकी के क्षेत्रों में अनुसंधान कर रहा है। अनुसंधान में सहयोग के लिए केन्द्र में पशु व कृषि फार्म, जैव सुरक्षा स्तर –3 की सूक्ष्मजीवी संगरोध प्रयोगशाला, कृषि प्रौद्योगिकी सूचना केन्द्र, कृषि प्रौद्योगिकी प्रबन्धन इकाई, पुस्तकालय् व अश्व ज्ञान सग्रांहलय जैसी केन्द्रीयकृत सुविधाएं स्थापित की गई हैं।

अश्वों के उत्पादन, आनुवंशिकी, प्रजनन, प्रबंधन, शरीर विज्ञान और पोषण पर अनुसंधान करने के लिए 1989 में बीकानेर (राजस्थान) में रा.अ.अनु.के. का एक उप परिसर स्थापित किया गया। अश्व उत्पादन परिसर बीकानेर में मारवाड़ी, काठियावाड़ी, ज़ांस्कारी और मणिपुरी घोड़ों तथा हलारी और विदेशी गर्दभों के समूह स्थापित किए गए। राष्ट्रीय पशु चिकित्सा प्रारूप संवर्धन केन्द्र की स्थापना वर्ष 2005 में भा.कृ.अनु.प.–रा.अ.अनु.के., हिसार के मुख्य परिसर में पशु मूल के रोगाणुओं के संग्रह, लक्षण वर्णन, संरक्षण और वितरण के लिए की गई थी। वर्तमान में, यह केंद्र देश भर में फैली 14 नेटवर्क इकाइयों के माध्यम से काम कर रहा है।

Centre is working through 14 network units spread throughout the country.

ICAR-NRCE has contributed significantly in the development of vaccines and diagnostics for equine infectious diseases and recognized as National Referral Centre for diagnosis of important equine infectious diseases by Department of Animal Husbandry, Dairying & Fisheries, Ministry of Fisheries, Animal Husbandry & Dairying , Government of India. During the COVID-19 pandemic the Centre extended its human resource and laboratories for diagnosis of the diseases and contributed significantly. The centre is providing services to equine breeders, state animal husbandry departments, police and army horses. The continuous surveillance and monitoring of equine infectious diseases has enabled the country to devise control strategies. The other important services include health certification for movement of equines, supply of cryopreserved semen for artificial insemination to augment the production of superior quality horses, mules and donkeys and training of various stakeholders on health and management aspects of equine husbandry. National Action Plan for control and eradication of glanders in India developed by ICAR-NRCE has been launched by the Ministry of Fisheries, Animal Husbandry and Dairying, Government of India. This action plan has been framed for surveillance of the entire equine population of the country reared in different management and animal husbandry practices following the conceptual framework of the OIE Terrestrial Code and the OIE Terrestrial Manual. The Centre is an active partner in the program for capacity building and providing technical inputs including novel diagnostics.

The National Centre for Veterinary type Cultures has now a good collection of accessioned microbes of veterinary use and has started distributing them for teaching, research and development of new technologies in the country.

Sero-surveillance and monitoring of equine infectious diseases in India is the mainstay of the services rendered by the institute. This aims to keep vigilance on the status of economically important equine diseases of viral, bacterial or parasitic origin. During the year, a total of 2299 equine serum samples from 10 states were tested for various diseases like Equine Infectious Anaemia (EIA), Equine Influenza (EI), Equine Herpes Virus-1 (EHV-1), Japanese Encephalitis/ West Nile Virus (JEV/WNV), *Trypanosoma evansi* (Trypanosomosis), Piroplasmosis, *Salmonella* Abortusequi and Brucellosis. The highest sero-prevalence was observed for equine

भा.कृ.अनु.प.–रा.अ.अनु.के. ने अश्व संक्रामक रोगों के टीकों और निदान के विकास में महत्वपूर्ण योगदान दिया है और यह केन्द्र भारत सरकार के मत्स्य पालन, पशुपालन और डेयरी मंत्रालय द्वारा अश्वों के संक्रामक रोगों के निदान के लिए राष्ट्रीय परामर्श केंद्र के रूप में मान्यता प्राप्त है। कोविड–19 महामारी के दौरान केंद्र ने रोग के निदान के लिए अपने मानव संसाधनों और प्रयोगशालाओं को प्रयोग किया और स्वास्थ्य सेवाओं में महत्वपूर्ण योगदान दिया। केंद्र अश्व प्रजनकों, राज्य पशुपालन विभागों, पुलिस और सेना के घोड़ों को सेवाएं प्रदान कर रहा है। केंद्र द्वारा अश्वों के संक्रामक रोगों की निरंतर निगरानी ने देश को नियंत्रण रणनीति तैयार करने में सक्षम बनाया है। केंद्र की अन्य महत्वपूर्ण सेवाओं में अश्वों की आवाजाही के लिए स्वास्थ्य प्रमाणन, बेहतर गुणवत्ता वाले घोड़ों, खच्चरों और गर्दभों के उत्पादन को बढ़ाने के लिए कृत्रिम गर्भाधान के लिए हिमीकृत वीर्य की आपूर्ति और अश्व पालन के स्वास्थ्य् और प्रबंधन पहलुओं पर विभिन्न हितधारकों के प्रशिक्षण शामिल हैं। भारत सरकार के मत्स्य पालन, पशुपालन और डेयरी मंत्रालय ने भा.कृ.अनु.प.–रा.अ.अनु. केन्द्र द्वारा विकसित ग्लैंडर्स के नियंत्रण और उन्मूलन के लिए राष्ट्रीय कार्य योजना शुरू की गई है। यह कार्य योजना विश्व पशु स्वास्थ्य संगठन के वैचारिक ढांचे का पालन करते हुए ग्लैंडर्स निगरानी के लिए तैयार की गई है। यह केंद्र क्षमता निर्माण और नवीनतम तकनीकी विकास में सक्रिय रूप से कार्यरत है।

राष्ट्रीय पशु चिकित्सा प्रारूप संवर्धन केन्द्र के पास अब पशु चिकित्सा उपयोग के परिवर्धित रोगाणुओं का एक अच्छा संग्रह है और केन्द्र ने देश में नई तकनीकों के विकास, शिक्षण व अनुसंधान के लिए उनका वितरण शुरू कर दिया गया है।

भारत में अश्व संक्रामक रोगों की निगरानी के लिए प्रदान की जाने वाली सेवाएं संस्थान का प्रमुख कार्य है। इसका उद्देश्य अश्वों के विषाणुजनित, जीवाणुजनित एवं परजीवी मूल के आर्थिक रूप से महत्वपूर्ण रोगों की स्थिति पर निगरानी रखना है। वर्ष के दौरान, 10 राज्यों के कुल 2299 अश्व सीरम नमूनों का परीक्षण विभिन्न रोगों जैसे अश्व रक्ताल्पता (ईआईए), अश्व फ्लू (ईआई), अश्व हर्पीस विषाणु–1 (ईएचवी–1), जापानी मस्तिष्क शोथ/वेस्ट नाइल बुखार (जेईवी/डब्ल्यूएनवी), सर्रा, पाइरोप्लाज्मोसिस, साल्मोनेला और ब्रुसेलोसिस के लिए किया गया। सीरो–प्रचलन में सर्वाधिक सकारात्मकता मुख्य रूप से इक्वाइन पाइरोप्लाज्मोसिस (51.80%) रोग के लिए पाई गई, इसके बाद जापानी मस्तिष्क शोथ/ वेस्ट नाईल बुखार (9. 13%), अश्व हर्पीस विषाणु–1 (2.87%) और सर्रा (1.43%) थे। अश्व फ्लू, अश्व सक्रांमक रक्ताल्पता, ब्रुसेलोसिस और साल्मोनेला के लिए कोई भी अश्व संक्रमित नहीं पाया गया।

रोग जांच के तहत 10111 नमूनों की जांच की गई। अश्व फ्लू के लिए 192 नमूनों के परीक्षण में एच3एन8 एंटीबॉडी नहीं पाई गई।

piroplasmosis (51.80%) followed by JE/WNV (9.13%), EHV-1(2.87%) and *Trypanosoma evansi* (1.43%). None of the equines were found positive for equine influenza, equine infectious anemia, brucellosis and *Salmonella* Abortusequi.

Under disease investigation 10111 samples were tested. Testing of 192 samples for equine influenza exhibited negative results for H3N8 antibodies. For annual reconfirmation of AHS free status, a total of 185 samples from 5 states were tested and found negative. For EIA, 9397 serum samples obtained from 13 states were found negative by Coggin's test. No EIA positive case has been reported in India for the last 10 years. This surveillance data would be of immense help for obtaining EIA free status of the country. Samples tested for other diseases under disease investigation were found negative.

A total of 22130 equine sera from 258 districts of 16 states were collected and tested for glanders. Out of these, 139 glanders positive cases were reported in 54 districts of 10 states. For rapid and efficient execution of surveillance activities glanders ELISA developed by NRCE has been provided to 12 state diagnostic laboratories/RDDLs. Presently, glanders ELISA kit is commercially available and being used by State laboratories. As per guidelines, ELISA positive samples were retested and confirmed by complement fixation test (CFT) at NRCE. About 50% of the glanders positive cases were from Uttar Pradesh. From zoonotic point of view, 87 sera from occupationally exposed humans (Veterinary Officers, equine handlers, laboratory workers) were tested and local form of glanders was detected in one equine handler.

Under contractual diagnostic services, a total 6775 samples were received from race courses, turf clubs, stud farms, riding schools, animal quarantine & certification services (AQCS) and other organized sectors during the year. These samples were tested for various notifiable and exotic diseases to check ingression of diseases and monitor elite horses in private sectors. A total of 2737 sera samples for EIA and 3316 samples for glanders were found negative. Among exotic diseases, 208 swab samples for contagious equine metritis (CEM), 70 samples for equine viral arteritis (EVA), 83 samples for African horse sickness (AHS) and 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 49.10 lakh was generated through contractual diagnostic services.

अफ्रीकन अश्व रोग मुक्त स्थिति की वार्षिक पुष्टि के लिए, 5 राज्यों के कुल 185 नमूनों का परीक्षण किया गया और वे नकारात्मक पाए गए। अश्व संक्रामक रक्ताल्पता के लिए, 13 राज्यों से प्राप्त 9397 सीरम नमूने कॉगिन परीक्षण से नकारात्मक पाए गए। पिछले 10 वर्षों से भारत में कोई भी अश्व संक्रामक रक्ताल्पता के पॉजिटिव मामला सामने नहीं आए हैं। यह आंकड़े देश को अश्व संक्रामक रक्ताल्पता से मुक्त स्थिति प्राप्त करने के लिए बहुत मददगार होंगे। रोग जांच में अन्य बीमारियों के लिए जांचे गए नमूने नकारात्मक पाए गए।

सोलह राज्यों के 258 जिलों से कुल 22130 अश्व सीरम नमूने एकत्र किए गए और ग्लैंडर्स रोग के लिए उनका परीक्षण किया गया। इनमें से 10 राज्यों के 54 जिलों में 139 ग्लैंडर्स पॉजिटिव मामले सामने आए। तेजी और कुशलता से ग्लैंडर्स की निगरानी के लिए रा.अ.अनु.के. द्वारा विकसित ग्लैंडर्स एलिसा को 12 राज्य–नैदानिक प्रयोगशालाओं को प्रदान किया गया है। वर्तमान में, ग्लैंडर्स एलिसा किट व्यावसायिक रूप से उपलब्ध है और राज्य प्रयोगशालाओं द्वारा उपयोग में लाई जा रही है। दिशानिर्देशों के अनुसार, रा.अ.अनु.के. में एलिसा सकारात्मक नमूनों का पुनः परीक्षण किया गया और पुष्टि की गई। लगभग 50% ग्लैंडर्स पॉजिटिव मामले उत्तर प्रदेश के थे। जूनोटिक दृष्टिकोण से, व्यावसायिक रूप से संवेदनशील मनुष्यों (पशु चिकित्सा अधिकारी, अश्वों की देखभाल करने वाले एवं प्रयोगशाला कार्यकर्ताओं) में 87 नमूनों का परीक्षण किया गया और एक अश्वपालक में ग्लैंडर्स के लक्षण पाए गए।

संविदात्मक निदान सेवाओं के तहत वर्ष के दौरान रेस कोर्स, टर्फ क्लब, स्टड फार्म, राइडिंग स्कूल, पशु संगरोध, प्रमाणन सेवाओं (एक्यूसीएस) और अन्य संगठित क्षेत्र से कुल 6775 नमूने प्राप्त हुए। इन नमूनों का परीक्षण प्रमुख अश्व रोगों और विदेशी बीमारियों के लिए किया गया ताकि बीमारियों के प्रवेश की जांच एवं रोकथाम की जा सके। अश्व संक्रामक रक्ताल्पता के लिए कुल 2737 सीरम नमूने और ग्लैंडर्स के लिए 3316 नमूने नकारात्मक पाए गए। विदेशी बीमारियों में, अश्व संक्रामक गर्भाश्य शोथ (सीईएम) के लिए 208 नमूने, अश्व विषाणु जनित धमनीशोध (ईवीए) के लिए 70 नमूने, अफ्रीकी अश्वों की बीमारी (एएचएस) और डोरीन के लिए 83 नमूने पशु संघरोध केन्द्रों से प्राप्त हुए। इन विदेशी बीमारियों के लिए सभी नमूने नकारात्मक पाए गए। संविदात्मक निदान सेवाओं के माध्यम से केन्द्र को 49. 10 लाख रुपये का राजस्व प्राप्त हुआ।

हरियाणा, उत्तर प्रदेश, हिमाचल प्रदेश और दिल्ली से नासिका स्वैब, ऊतक, फोड़े, गर्भपात भ्रूण जैसे 244 नमूने प्राप्त किये गए जिनमें से 51 विभिन्न जीवाणु प्राप्त हुए जिनका सूक्ष्म विश्लेषण किया गया।

Microbiological analysis was carried out on 244 clinical samples including nasal swabs, tissues, abscesses, aborted fetus etc. originating from Haryana, Uttar Pradesh, Himachal Pradesh and Delhi yielding 51 bacterial isolates.

Burkholderia mallei is the causative agent of glanders, a fatal devastating infectious disease of horses, donkeys and mules. To ascertain the genetic diversity of Indian *B. mallei* strains sequencing of 16S rRNA and ITS genes and phylogenetic analysis of 39 *B. mallei* isolates were performed. For 16S phylogeny, 36 sequences were added from the database thus 75 *B. mallei* sequences were used for tree construction. Phylogeny showed a similar branching pattern and revealed that recent isolates circulating in India are closely related to each other but genetically diverse from older *B. mallei* isolates that were reported from India or elsewhere. The present findings and surveillance data suggests that Uttar Pradesh is the most glanders prone area and equine movement from this place led to rapid spread of glanders to other states.

Streptococcus equi is the causative agent of strangles. The disease is economically important and the most contagious infectious disease of horses, mules and donkeys in India. The organism has been isolated from many diseased animals but not characterized at molecular level to ascertain the diversity of microbial population in India. The MLST scheme for the β-hemolytic, Lancefield group C streptococcal bacteria *Streptococcus equi and S. zooepidemicus* has been developed based on sequencing of seven highly conserved housekeeping genes at the Animal Health Trust. UK. This study analysed the sequences of these seven genes from forty one isolates of the bacteria isolated from field cases. The genomic constellations suggest the presence of novel isolates of *S. equi* in India. Out of the total 41 *S. equi* isolates nine isolates had novel genomic constellations that have not been reported from any part of the world. MLST analysis also revealed that the genomic combination of two isolates of *S. zooepidemicus* and *S. equisimilis* were also novel and not reported earlier throught the World.

Equine herpesvirus 1 (EHV-1) causes abortion, neonatal death, respiratory and neurological disease in equines and is endemic in India. The factors that contribute to the EHV-1 disease severity are not clearly known. To decipher the EHV-1 diversity amongst abortigenic isolates in India, we carried out the whole genomes sequencing of Indian EHV-1 isolates. Phylogenetic analysis of the genome showed

बर्खोल्डेरिया मैलाई ग्लैंडर्स का कारक है जो कि घोड़ों, गर्दभों और खच्चरों में पाया जाने वाला एक घातक विनाशकारी संक्रामक रोग है। भारतीय जीवाणुओं की आनुवंशिक विविधता का पता लगाने के लिए 16एस राइबो न्यूक्लिक एसिड और आई. टी.एस. जीनों का अनुक्रमण और फ़ाइलोजेनेटिक विश्लेषण किया गया। 16एस फ़ाइलोजेनी के लिए 75 अनुक्रमों का उपयोग विश्लेशण के लिए किया गया। फाइलोजेनी से यह खुलासा हुआ कि भारत में हाल के जीवाणु नमूने एक–दूसरे से निकटता से संबंधित हैं, लेकिन आनुवंशिक रूप से पुराने जीवाणु से भिन्न हैं जो प्रायः भारत या अन्य जगहों से एकत्रित किए गए थे। वर्तमान निष्कर्ष और निगरानी से पता चलता है कि उत्तर प्रदेश सबसे अधिक ग्लैंडर्स संक्रमित क्षेत्र है और इस जगह से अश्वों की आवाजाही के कारण अन्य राज्यों में ग्लैंडर्स का तेजी से प्रसार हुआ है।

स्ट्रेप्टोकोकस इक्वाई स्ट्रेन्गल्स का कारक जीवाणु है। यह रोग आर्थिक रूप से महत्वपूर्ण है और भारत में घोड़ों, खच्चरों और गर्दभों का सबसे संक्रामक रोग है। यह जीवाणु कई रोगग्रस्त जानवरों में पाया गया है लेकिन भारत में जीवाणुओं की विविधता का पता लगाने के लिए आणविक स्तर पर कार्य नहीं हुआ है। β–हेमोलिटिक, लांसफील्ड समूह सी स्ट्रेप्टोकोकल जीवाणु स्ट्रेप्टोकोकस इक्वाई और एस. जूएपिडेमिकस के लिए एमएलएसटी योजना को पशु स्वास्थ्य ट्रस्ट, ब्रिटेन में सात अत्यधिक संरक्षित हाउसकीपिंग जीनों के अनुक्रमण के आधार पर विकसित किया गया है। इस अध्ययन में भारत से एकत्रित जीवाणु के इकतालीस नमूनो से इन सात जीनों के अनुक्रमों का विष्लेषण किया। इस विष्लेशण द्वारा भारत में नई प्रकार के जीवाणुओं की उपस्थिति का पता चला। कुल 41 जीवाणु नमूनों में से नौ नमूनों में नए आनुवांशिक संयोजन विश्व के अन्य जीवाणुओं से भिन्न थे जिन्हें दुनिया के किसी भी हिस्से से पहली बार एकत्रित किया गया है। एमएलएसटी विषलेषण से यह भी पता चला है कि एस. जूएपिडेमिकस और एस. इक्विमिलिस के दो आइसोलेट्स का आनुवांशिक संयोजन भी नवीन था और दुनिया भर में पहले इसकी सूचना नहीं थी।

अश्व हर्पीस विषाणु–1 (ई.एच.वी.1) अश्वों में गर्भपात, नवजात मृत्यु, श्वसन और तंत्रिका संबंधी रोगों का कारण है व भारत में स्थानिक है। अश्व हर्पीस विषाणु–1 रोग की गंभीरता में योगदान करने वाले कारक स्पष्ट रूप से ज्ञात नहीं हैं। भारत में गर्भपात करने वाले आइसोलेट्स के बीच अश्व हर्पीस विषाणु–1 की विविधता को समझने के लिए, हमने भारतीय विषाणु के नमूनों के पूरे जीनोम का अनुक्रमण दिया। फ़ाइलोजेनेटिक विश्लेषण से पता चला है कि भारत में 3 अलग–अलग वायरल क्लेड पाए जाते हैं। एसेक्स आइसोलेट को छोड़कर सभी भारतीय आइसोलेट्स विश्वसनीय बूटस्ट्रैप मूल्यों से यूके से प्राप्त

that up to 3 distinct viral clades have been circulating in India. All the Indian isolates clustered away from the UK isolates with reliable bootstrap values except for the Essex isolate. Among the Indian isolates, all the Hisar isolates clustered to a sub-node, away from the H14 and Meerut isolates. Delhi isolate was found to be the farthest from the Meerut isolates. One of the abortigenic isolates had the N752D substitution, whereas all other abortion isolates were non-neuropathic. The bioinformatic analysis suggested that diverse strains of EHV-1 are circulating in India and causing abortions in pregnant mares.

Equid gammaherpesvirus 2 (EHV-2) and 5 (EHV-5) are members of the family *Herpesviridae*, EHV-2 is also associated with keratoconjunctivitis and EHV-5 with equine multi-nodular pulmonary fibrosis syndrome. Infections by EHV-2 and 5 occur in young foals with periodic reactivation of the latent virus during the life of the animal. Although herpesvirus infections (EHV-1& 4) are endemic in horses, herpesvirus infections (EHV-2 & 5) have not been reported from India. Taqman probe-based real-time PCR assays were standardized using primers and probes designed to detect the glycoprotein B gene of EHV-2 & 5. These assays were used to detect the occurrence of EHV-2 and EHV-5 in the upper respiratory tract of asymptomatic young horses.

Comparative pathological evaluation of deletion mutants of EHV-1 was undertaken. Four deletion mutants of EHV-1 developed earlier and coding Δg^{A},Δg^{B}, $\Delta g^{A,B}$ and $\Delta g^{A,B,X,Y}$ were studied in BALB/c mouse model for assessing their pathology and attenuation as compared to the wild parent virus EHV-1- Tohana. Based on the substantial attenuation of virus, broad cell tropism, CD8+ and Th1 mediated immune responses, the deletion mutants of EHV-1 *viz.*$\Delta g^{A,B}$ and $\Delta g^{A,B,X,Y}$ qualified to be good modified live vaccine candidates and were studied further in murine model challenge studies. EHV-1 virus constructs $\Delta g^{A,B}$ and $\Delta g^{A,B,X,Y}$ were selected to assess their pathogenicity and protective efficacy in murine model. Mice immunized with the mutant viruses and further challenged with pathogenic wild virus showed less pathology in terms of clinical signs, body weight loss, gross and histopathological lesions. SNT and ELISA results showed good immune response with attenuated viruses. Flow-cytometric immune-phenotyping revealed Th1 dominated immune response in all groups at all-time points, highest being witnessed in four deletion mutant

आइसोलेट्स से दूर क्लस्टर हुए । भारतीय आइसोलेट्स में, सभी हिसार आइसोलेट्स एच14 और मेरठ आइसोलेट्स से दूर एक सब–नोड में क्लस्टर हुए हैं। दिल्ली के आइसोलेट मेरठ के आइसोलेट्स से दूर पाया गया। गर्भपात करने वाले आइसोलेट्स में से एक में एन.752डी. प्रतिस्थापन था, जबकि अन्य सभी गर्भपात आइसोलेट्स गैर–न्यूरोपैथिक थे। जैव सूचनात्मक विश्लेषण से पता चला अश्व हर्पीस विषाणु–1 के विविध उपभेद भारत में घूम रहे है और गर्भवती मादा अश्व में गर्भपात का कारण बन रहे हैं।

अश्व गामाहर्पीस विषाणु–2 (ई.एच.वी–2) और 5 (ई.एच.वी.–5) हर्पीसविरिडे फैमिली के सदस्य हैं, ई.एच.वी–2 नेत्र श्लेश्मल शोथ और ई.एच.वी.–5 अश्वों में श्वसन तन्त्र में बहु–गांठदार फाईब्रोसिस सिंड्रोम से जुड़ा है। ई.एच.वी.–2 व 5 संक्रमण पशु के जीवन के दौरान अव्यक्त वायरस के आवधिक पुनर्सक्रियन के साथ नवजात अश्वों में होता है। हालांकि –हर्पीसवायरस संक्रमण (ई.एच.वी.–1 व 4) घोड़ों में स्थानिक हैं, गामाहर्पीस विषाणु संक्रमण (ई.एच.वी.–2 व 5) भारत से अभी तक रिपोर्ट नहीं किए गए हैं। पी.सी.आर. द्वारा ई.एच.वी.–2 और 5 के ग्लाइकोप्रोटीन बी. जीन का पता लगाने के लिए डिज़ाइन किए गए प्राइमर और प्रोब का उपयोग करके मानकीकृत किया गया। इन विधियों का उपयोग स्पर्शोन्मुख अश्वों के ऊपरी श्वसन पथ में ई.एच.वी.–2 और ई.एच.वी.–5 का पता लगाने के लिए किया गया।

ई.एच.वी.–1 के विलोपन म्यूटेंट का तुलनात्मक रोग मूल्यांकन किया गया। ई.एच.वी.–1 के पहले विकसित हुए चार विलोपन म्यूटेंट Δजी ^{ए}, Δजी $^{बी}$, Δजी $^{ए, बी}$ और Δजी $^{ए, बी, एक्स, वाई}$ और मूल वाइल्ड पैरेंट ई.एच.वी. 1– (टोहाना) की तुलना में उनकी विकृति और क्षीणन का आकलन करने के लिए बाल्ब/सी. चूहों में अध्ययन किया गया। वायरस के पर्याप्त क्षीणन, ब्रॉड सेल ट्रोपिज्म, सीडी 8+ और टी.एच. 1 द्वारा मध्यस्थता प्रतिरक्षा प्रतिक्रियाओं के आधार पर, ईएचवी 1 के विलोपन म्यूटेंट अर्थात Δजी $^{ए, बी}$ और Δजी $^{ए, बी, एक्स, वाई}$ वैक्सीन बनाने के लिए उपयुक्त थे और उनका चूहों में अध्ययन किया गया। ई.एच.वी.–1 की डेल्टा Δजी $^{एए बी}$ और Δ जी $^{ए, बी, एक्स, वाई}$को चूहों में उनकी रोगजनकता और सुरक्षात्मक प्रभावकारिता का आकलन करने के लिए चुना गया। म्युटेंट वायरस से टीकाकृत चूहों को जब मूल विषाणु के साथ चुनौती दी, तब नैदानिक संकेतों, शरीर का वजन घटने, सकल और ऊतकविकृति विश्लेषण में कम विकृति दिखाई दी गई । एसएनटी और एलिसा परिणामों ने क्षीण विषाणुओं के साथ अच्छी प्रतिरक्षा प्रतिक्रिया दिखाई। फ्लो–साइटोमेट्रिक इम्यून–फेनोटाइपिंग ने सभी समूहों में सभी समय बिंदुओं पर टी. एच. 1 वर्चस्व वाली प्रतिरक्षा प्रतिक्रिया का प्रदर्शन किया, उच्चतम चार विलोपन उत्परिवर्ती में देखा गया।

EHV-1. EHV-1 virus constructs developed are providing adequate immune responses and protective immunity in murine model and are prospective candidates for large animal studies.

Antibiotic resistance is emerging at fast pace against various bacteria including *Streptococcus equi,* the causative agent of strangles, a highly contagious disease of economic importance in horses, mules and donkeys. In order to find an alternative to the antibiotics and counter the antibiotic resistance, we determined the efficacy of our novel ZnO nanoparticles against *S. equi.* Zinc oxide nanoparticles prepared by conventional hydrothermal method and microwave irradiation were found to be highly effective against *S. equi* at various concentrations.

Zinc oxide based polymeric nanoformulations were used in collagen induced arthritis in DBA-1 mice. We evaluated the efficacy of developed nanoformulations (AGZnO NPs/ CsZnO NPs). Remarkable reduction in inflammation and swelling of the inflamed digits/feet were observed in treated and arthritic mice. The gross changes in the affected feet/ digits demonstrated the remarkable efficacy of the nanoformulation. Serum creatinine levels were found higher in untreated arthritic DBA-1 mice as compared to treated DBA-1 arthritic mice. The markers for cartilage degradation determined in treated and untreated DBA-1 experimental arthritic mice revealed the efficacy of the nanoformulations.

Lawsonia inermis Linn popularly known as the Henna has played an important role in natural herbal medicines. We assessed the phytochemical profile for presence of phyto-constituents from various extracts of the plant leaves and found alkaloids, steroids, flavonoids, saponins, tannins, proteins/amino acids in different extracts. The extracts were further purified by column chromatography for the isolation of plant constituents and monitored by thin layer chromatography (TLC), analysed by FTIR, HNMR, and GC-MS analysis. All the spectral results (IR, NMR, GC-MS) suggest that the compounds from the extract contain aromatic nucleus and OH group along with methoxy group, allyl as well as vinyl group. Cytotoxicity studies revealed some of the leaf extracts have potential cytotoxic activity on vero cells. Reducing the chloroform concentration during extraction decreases the cytotoxic effect on the cells. Although most of the components are biocompatible, the presence of cytotoxic compounds in some of these extracts warrants research for fabrication of suitable formulations comprising these constituents

विकसित ई.एच.वी.–1 विषाणु चूहों में पर्याप्त प्रतिक्रिया और सुरक्षात्मक प्रतिरक्षा प्रदान कर रहे हैं और बड़े जानवरों के अध्ययन के लिए संभावित उम्मीदवार हैं।

घोड़ों, खच्चरों और गर्दभों में आर्थिक महत्व की अत्यधिक संक्रामक बीमारी स्ट्रेंगलस के खिलाफ एंटीबायोटिक प्रतिरोध तेज गति से उभर रहा है। एंटीबायोटिक दवाओं का विकल्प खोजने और एंटीबायोटिक प्रतिरोध का मुकाबला करने के लिए, हमने अपने नवीन जिंक ऑक्साइड नैनोकणों की स्ट्रैप्टोकोकस इक्वाई के खिलाफ प्रभावकारिता मूल्याकिंत की। पारंपरिक हाइड्रोथर्मल विधि, माइक्रोवेव विकिरण द्वारा तैयार जिंक ऑक्साइड नैनोकणों को विभिन्न सांद्रता में एस. इक्वाई के खिलाफ अत्यधिक प्रभावी पाया गया।

डी.बी.ए.–1 चूहों में कोलेजन प्रेरित गठिया में जिंक ऑक्साइड आधारित पॉलीमेरिक नैनोफॉर्म्यूलेशन का उपयोग व विकसित नैनोफॉर्म्यूलेशन की प्रभावकारिता का मूल्यांकन किया। उपचारित और गठिया रोग से ग्रसित चूहों में सूजन में उल्लेखनीय कमी देखी गई। प्रभावित पैरों में सकल परिवर्तन ने नैनोफॉर्म्यूलेशन की उल्लेखनीय प्रभावकारिता का प्रदर्शन किया। इलाज किए गए डीबीए–1 गठिया रोगी चूहों की तुलना में अनुपचारित गठिया डीबीए–1 चूहों में सीरम क्रिएटिनिन का स्तर अधिक पाया गया। उपास्थि क्षरण के लिए मार्करों ने जैसे सूजन से पूर्व साईटोकाइन्स को उपचारित और अनुपचारित डी.बी.ए.–1 प्रायोगिक चूहों में निर्धारित किया, जिससे नैनोफॉर्मुलेशन की प्रभावकारिता का पता चला।

मेंहदी ने प्राकृतिक औषधी के रूप में महत्वपूर्ण भूमिका निभाई है। हमने पौधों की पत्तियों के अर्क से विभिन्न घटकों की उपस्थिति के लिए रासायनिक आंकलन किया और अर्क में विभिन्न एल्कलॉइड, स्टेरॉयड, फ्लेवोनोइड, सैपोनिन, टैनिन, प्रोटीन/अमीनो एसिड पाए गए। पौधों के घटकों के अलगाव के लिए कॉलम क्रोमैटोग्राफी द्वारा अर्क को और शुद्ध किया गया और एफटीआईआर, एचएनएमआर और जीसी–एमएस द्वारा विश्लेषण किया गया तथा थिन लेयर क्रोमैटोग्राफी (टीएलसी) द्वारा निगरानी की गई थी। सभी वर्णक्रमीय परिणामों से पता चलता है कि अर्क के यौगिकों में मेथॉक्सी समूह, एलाइल व विनाइल समूह के साथ सुगंधित नाभिक और हाईड्रोक्सी समूह होते हैं। विषाक्तता अध्ययनों से पता चला है कि पत्तों के अर्क में कोशिकाओं पर विषाक्तकता थी। निष्कर्षण के दौरान क्लोरोफॉर्म सांद्रता को कम करने से कोशिकाओं पर विषैला प्रभाव कम हो जाता है। यद्यपि अधिकांश घटक जैव–संगत हैं। इस पौधे में उपस्थित विषैले योगिकों को कम खुराक व विषाक्तता निवारण के अनुसंधान के पश्चात् उपयोग की सलाह दी जा सकती है।

घोड़ों सहित अन्य जानवरों में कई प्रकार के ट्यूमर जैसे

to reduce its dose/toxicity for the use of beneficial effects of the plant components.

Many types of tumors such as sarcoids, melanoma, carcinoma and warts are observed in animals including horses. Leaf extract of *Aerva javanica* was used against melanoma and warts in horses. A fraction of methanol soluble fraction of the aqueous extract of leaf of *Aerva javanica* was applied topically against two clinical cases of melanoma and two clinical cases of warts in horses. All the cases of tumor resolved completely in 30 to 50 days of topical application of the extracts. Occurrence of seasonal allergic dermatitis, especially insect bite hypersensitivity and atopic dermatitis are very common in horses. At present there is no satisfactory treatment available for management of these skin allergies. A total of 11 clinical cases were treated successfully using the plant extract.

Trypanosoma evansi is an extracellular flagellate blood protozoan parasite and an etiological agent of animal trypanosomosis. Presently, only a few drugs are registered in India and have been used for the treatment of animal trypanosomosis, but these drugs show severe toxic effects and also have the problem of drug resistance. Naphthoquinones (NTQ) have been reported for their antitrypanosomal potential against other trypanosomes-*T. brucei* and *T. cruzi*. Six naphthoquinones (NTQ1-NTQ6) derivatives were selected and procured for evaluation by demonstrating their growth inhibitory effect against *T. evansi*. All NTQs significantly ($p<0.001$) exhibited activity against parasite growth and multiplication. We confirmed the antitrypanosomal activity and apoptotic-like mechanism of NTQs in an axenic culture of *T. evansi*.

Endurance and fertility analyses in indigenous horses using SNP markers were performed for a total of 12 SNPs loci associated with endurance and fertility in 10 animals each in Marwari, Kathiawari and Kachchi Sindhi and a minimum of 5 animals each in Manipuri and Zanskari breeds. In general, the ponies i.e., Manipuri and Zanskari exhibited lower polymorphism as compared to the horse breeds probably because of small sample size and /or small population base even in the breeding tract. Existence of polymorphism in Indian horses and ponies indicates the possibility of their use in selection programmes.

The inventory database of the Marwari, Kathiawari, Manipuri, Zanskari horses and donkeys maintained at the Campus has been updated for the period 1989 to 2020, *i.e.* from date of inception of the Campus to till

सारकॉइड, मेलेनोमा, कार्सिनोमा देखे जाते हैं। सफेद बूई (एर्वा जावनिका) के पत्तों के अर्क का इस्तेमाल मेलेनोमा के खिलाफ किया गया। ऐर्वा जावनिका की पत्ती के जलीय अर्क के मेथनॉल में घुलनशील अंश मेलेनोमा और घोड़ों में वार्ट के दो नैदानिक मामलों के खिलाफ प्रयोग किया गया। अर्क के सामयिक अनुप्रयोग के 30 से 50 दिनों में ट्यूमर के सभी मामले पूरी तरह से ठीक हो गए। मौसमी एलर्जी, विशेष रूप से कीड़े के काटने से होने वाली अतिसंवेदनशीलता और एटोपिक त्वकशोथ घोड़ों में बहुत आम है। वर्तमान में इनके प्रबंधन के लिए कोई संतोषजनक उपचार उपलब्ध नहीं है। पौधे के अर्क का उपयोग करके कुल 11 अश्वों मामलों का सफलतापूर्वक इलाज किया गया।

ट्रिपैनोसोमा इवान्साई एक बाह्य कोशिकीय, रक्त प्रोटोजोआ परजीवी है और पशुओं में सर्रा का एक कारक है। वर्तमान में, भारत में इसके लिए केवल कुछ दवाएं पंजीकृत हैं और जानवरों के इलाज के लिए उपयोग की जाती हैं, लेकिन ये दवाएं गंभीर विषाक्त प्रभाव दिखाती हैं और इनमें दवा प्रतिरोध की समस्या भी होती है। नेफ्थोक्विनोन (एनटीक्यू) को ट्रिपैनोसोमा ब्रुसाई व ट्रि. क्रुजाई के खिलाफ उनकी परजीवी के विरूद्ध क्षमता के लिए जाना जाता है। । छह नेफ्थोक्विनोन व्युत्पन्न रासायनों का चयन किया गया और ट्रि. इवान्साई के खिलाफ उनके निरोधात्मक प्रभाव के मूल्यांकन के लिए चुना गया। सभी रसायन महत्वपूर्ण रूप से परजीवी वृद्धि के विरूद्ध उपयोगी थे। हमने ट्रि. इवान्साई की एक अक्षीय संवर्धन में एनटीक्यू की परजीवी के विरूद्ध गतिविधि और एपोप्टोटिक जैसी गतिविधियों की पुष्टि की।

मारवाड़ी, काठियावाड़ी और कच्छी–सिंधी नस्ल के 10 जानवरों और मणिपुरी व ज़ांस्कारी नस्लों में से प्रत्येक में न्यूनतम 5 जानवरों में तितिक्षा और प्रजनन क्षमता से जुड़े कुल 12 एसएनपी लोसाईं के लिए एसएनपी मार्करों का उपयोग करके विश्लेषण किया गया। सामान्य तौर पर, टट्टुओ (मणिपुरी और ज़ांस्कारी) ने घोड़ों की तुलना में कम बहुरूपता का प्रदर्शन किया। भारतीय घोड़ों और टट्टुओं में बहुरूपता का अस्तित्व चयन कार्यक्रमों में उनके उपयोग की संभावना को इंगित करता है।

अश्व उत्पादन परिसर में मारवाड़ी, काठियावाड़ी, मणिपुरी, ज़ांस्कारी घोड़ों और गर्दभों के आकंड़ो को 1989 से 2020 की अवधि के लिए पूर्ण किया गया है।

1989 से अब तक मारवाड़ी घोड़ों की बायोमेट्री अर्थात विदर ऊंचाई, शरीर की लंबाई, हृदय की परिधि और शरीर के वजन के आंकड़ों का विश्लेषण वंशावली संबंध, अंतर्गर्भाशयी गुणांक की गणना, फेनोटाइपिक प्रवृत्ति का विश्लेषण, प्रजनन मूल्यों का अनुमान, आनुवंशिकता के विश्लेषण के लिए किया गया। कुल 30 वर्षों की अवधि के दौरान 226 अभिलेखों का विश्लेषण किया गया। अश्व की औसत ऊंचाई 150.15 ± 0.04 सेमी, शरीर की

The biometry, *viz*. wither height, body length, heart girth and body weight data of Marwari horses since 1989 till date were analyzed for pedigree relationship, calculation of inbreeding coefficient, analysis of phenotypic trend, estimation of breeding values, estimation of heritability and analysis of variance. In all 30 years' period was covered and 226 records were analyzed. The average height at withers was 150.15±0.04 cm, body length was 151.44±0.06 cm, heart girth was 170.02±0.19 cm and body weight was 371.34±0.52 kg. The analysis of variance indicated non-significant effect of sex, tier and interaction of sex and tier. The breeding values were quite close to the population means except for the body weight. The effect of sex, tier and sex *tiers was non-significant on EBV of wither height, body length and heart girth. The effect of tiers on EBV of body weight was significant (P<0.05) indicating that body weight received favour in breeding programme. The breeding plan was prepared for Marwari, Kathiawari, Manipuri and Zanskari animals. The average height of selected Marwari stallions was 160.6 cm and the average height of breedable females was 150.30 cm. The average body length and heart girth of selected stallions were 150.60 cm and 173.1 cm, respectively.

Looking at the demand of the farmers, two Nukra males were purchased from the field and cryopreserved about 100 doses of semen for AI of farm/ field mares.

A total of 47 doses of the Marwari semen were sold to the veterinary professional for artificial insemination. Though, the semen doses of Marwari stallions were supplied to field veterinarians free of cost in previous years for artificial insemination in field mares.

The semen straws of Marwari (500), Manipuri (480), Zanskari (500) horses and Halari donkeys (500) have been submitted to the National Semen Bank at NBAGR, Karnal. The NBAGR has also collected the somatic cells from the Centre for cryopreservation.

Various biochemical and hormonal profiles of the Marwari stallion semen were carried out and correlated with that of seminal quality parameters. GOT, GPT, TP and Ca differed significantly in seminal plasma while glucose differed significantly in spermatozoa (10^9 million) among stallions. GOT, GGT, TP, Ca and P were significantly lower in the stallions below 4 years of age. Glucose and Ca levels were found to have significant positive correlation with progressive motility and cholesterol was found to have significant positive correlation with viability and membrane integrity.

लंबाई 151.44 ± 0.06 सेमी, हृदय की परिधि 170.02 ± 0.19 सेमी और शरीर का वजन 371.34 ± 0.52 किलोग्राम था। शरीर के वजन को छोड़कर प्रजनन मूल्य जनसंख्या औसत के काफी करीब थे। ऊँचाई, शरीर की लंबाई और हृदय की परिधि के प्रजनन मूल्यांक पर लिंग, टायर और लिंग– टायर का प्रभाव गैर–महत्वपूर्ण था। शरीर के वजन को प्रजनन मूल्यांक के स्तरों पर प्रभाव महत्वपूर्ण था (पी <0.05), यह दर्शाता है कि प्रजनन कार्यक्रम में शरीर के वजन का चयन किया गया। मारवाड़ी, काठियावाड़ी, मणिपुरी और ज़ांस्कारी जानवरों के लिए प्रजनन योजना तैयार की गई। चयनित मारवाड़ी घोड़ों की औसत ऊंचाई 160.60 सेमी और प्रजनन योग्य मादाओं की औसत ऊंचाई 150. 30 सेमी थी। चयनित घोड़ों के शरीर की औसत लंबाई और हृदय की परिधि क्रमशः 150.60 सेमी और 173.10 सेमी थी।

किसानों की मांग को देखते हुए दो नुकरा घोड़े खरीदे गए और कृत्रिम गर्भाधान के लिए वीर्य की लगभग 100 खुराक हिमीकृत व रक्षित की गई।

मारवाड़ी वीर्य की कुल 47 खुराक कृत्रिम गर्भाधान के लिए पशु चिकित्सको को बेची गईं। मारवाड़ी घोड़े के वीर्य की खुराक गत वर्षों में भी फील्ड में कृत्रिम गर्भाधान के लिए पशु चिकित्सकों को मुफ्त आपूर्ति की गई थी।

मारवाड़ी, मणिपुरी, जांस्कारी घोड़ों और हलारी गर्दभों का वीर्य राष्ट्रीय पशु आनुवंशिक ब्यूरो, करनाल में राष्ट्रीय वीर्य बैंक में जमा किए गए हैं। राष्ट्रीय पशु आनुवंशिक ब्यूरो ने हिमीकरण एवं रक्षण के लिए केंद्र से कायिक कोशिकाओं को भी एकत्र किया है।

मारवाड़ी अश्वों के वीर्य के विभिन्न जैव–रासायनिक और हार्मोनल रूपरेखा तैयार की गई और उन्हें वीर्य गुणवत्ता मानकों के साथ जोड़ा गया। जीओटी, जीपीटी, टीपी और कैलशियम वीर्य प्लाज्मा में काफी भिन्न थे जबकि ग्लूकोज की मात्रा शुक्राणुओं (10^9 मिलियन) में विभिन्न घोड़ों के बीच काफी भिन्न थी। चार वर्ष से कम उम्र के घोड़ों में जीओटी, जीजीटी, टीपी, कैल्शियम और फासफोरस काफी कम थे। ग्लूकोज और कैल्शियम स्तरों में प्रगतिशील गतिशीलता के साथ महत्वपूर्ण सकारात्मक सहसंबंध पाया गया और कोलेस्ट्रॉल का व्यवहार्यता और झिल्ली अखंडता के साथ महत्वपूर्ण सकारात्मक सहसंबंध पाया गया।

शुक्राणु की गतिशीलता और गतिज मापदंडों पर नस्ल के प्रभाव का मूल्यांकन किया गया। मारवाड़ी नस्ल के शुक्राणु की सिर की लंबाई और पूंछ की लंबाई में मणिपुरी और ज़ांस्कारी घोड़ों के मुकाबले महत्वपूर्ण अंतर देखा गया, जबकि मणिपुरी और ज़ांस्कारी घोड़ों के बीच कोई महत्वपूर्ण अंतर नहीं था।

घोड़ों के सेमिनल मैसेंजर राइबोन्यूक्लिक एसिड को अलग किया गया और विभिन्न प्रजनन संबंधी जीन की अभिव्यक्ति के लिए

Evaluation of effect of breed on motility and kinematic parameters of the spermatozoa were studied and recorded. A significant difference was observed in the parameters like head length and tail length of Marwari and that of Manipuri and Zanskari stallions and there were no significant differences observed among the Manipuri and Zanskari stallions.

The stallion seminal mRNA was isolated and subjected to analysis for the expression of various fertility related genes. All the six fertility related marker genes showed differential expression between the seasons in Marwari stallions.

We also standardized the protocols for assessment of seminal quality parameters using flow cytometry. The study showed that cryopreservation process has significant effect on MMP and production of ROS.

Equine Production Campus, ICAR-NRCE has perfected the technology of semen cryopreservation of elite stallions not in lab and farm conditions but also off campus at farmers' doorsteps. In this endeavour, a team collected and cryopreserved semen from village Kankroli, Rajsamand (Rajasthan) from three elite Marwari stallions.

Ten mares were monitored for duration of foal estrus and other parameters for foal heat breeding of mares- a strategy to obtain a foal per year. The conception rates varied from 39.14 to 68.89 % for different breeds.

Studies were performed for development of fatigue cum fitness score card for working equines. For development of improved saddle and harness for working equines, different saddles fabricated during previous year were tested in mules and donkeys during carting trials at our centre. Refinement of saddle for carting equids was done. Testing of these saddles has been done on institute donkeys and mules during carting trials. The donkeys and mules were comfortable and no injury or saddle mark was observed on the back of experimental animals. Adjustable type saddles were fabricated for mules and ponies.

There is a recent renewed interest in the economic utility of donkeys (*Equus asinus*), especially, for the production and consumption of donkey milk. Due to inherent nutritive content, milk provides a rich medium for microbial growth, and thus may constitute a public health risk if consumed raw.

Donkeys are primarily known as pack animals, reared date.

विश्लेषण किया गया। सभी छह उर्वरता संबंधी मार्कर जीनों ने मारवाड़ी घोड़ों में ऋतुओं के बीच विभेदक अभिव्यक्ति दिखाई।

हमने फ्लोसाइटोमेट्री का उपयोग करते हुए मौलिक गुणवत्ता मापदंडों के आकलन के लिए प्रोटोकॉल को भी मानकीकृत किया। अध्ययन से पता चला है कि हिमीकरण प्रक्रिया का एमएमपी और आरओएस के उत्पादन पर महत्वपूर्ण प्रभाव पड़ता है।

अश्व उत्पादन परिसर, भा.कृ.अनु.प.–रा.अ.अनु.के. ने प्रयोगशाला और फार्म की स्थितियों में ही नहीं बल्कि परिसर के बाहर भी विशिष्ठ अश्वों के वीर्य की हिमीकरण की तकनीक को विकसित किया है। हमारी टीम ने कांकरोली, राजसमंद (राजस्थान) के तीन उत्तम किस्म के मारवाड़ी घोड़ों से वीर्य एकत्र किया और हिमीकरण द्वारा संरक्षित किया।

प्रति वर्ष एक बछड़ा प्राप्त करने की रणनीति के अंतर्गत दस मादा अश्वों में फ़ोल ईस्ट्रस अवधि एवं अन्य मापदंडों की निगरानी की गई। विभिन्न नस्लों के लिए गर्भाधान दर 39.14 से 68.89% तक पाए गए ।

काम करने वाले अश्वों के लिए थकान सह फिटनेस स्कोर कार्ड के विकास के लिए अध्ययन किए गए। काम करने वाले घोड़ों के लिए बेहतर काठी और साज़ (हार्नेस) के विकास के लिए, पिछले वर्ष के दौरान निर्मित विभिन्न काठी का परीक्षण हमारे केंद्र में खच्चरों और गर्दभों में किया गया था। सामान ढोने के लिए काठी का संशोधन किया गया। कार्टिंग ट्रायल के दौरान इनका परीक्षण संस्थान के गर्दभों और खच्चरों पर किया गया है। गर्दभ और खच्चर कुशल थे और प्रायोगिक जानवरों की पीठ पर कोई चोट या काठी का निशान नहीं देखा गया था। खच्चरों और टट्टुओं के लिए समायोज्य काठी बनाई गई।

गर्दभों (इक्वस असिनस) के दूध की आर्थिक उपयोगिता हाल ही में अत्यधिक प्रचलित हो रही है। निहित पोषक तत्वों के कारण, दूध माइक्रोबियल विकास के लिए एक समृद्ध माध्यम प्रदान करता है, और इस प्रकार कच्चे दूध के सेवन से स्वास्थ्य को जोखिम हो सकता है।

गर्दभों को मुख्य रूप से बोझा ढोने वाले जानवरों के रूप में जाना जाता है, जिन्हें मुख्य रूप से काम के लिए पाला जाता है। भारत में गर्दभ के दूध की संरचना का अधिक विस्तार से अध्ययन नहीं किया गया है। भा.कृ.अनु.प.–रा.अ.अनु.के. ने हलारी गर्दभों के दूध और दूध पाउडर के भौतिक और जैव–रासायनिक गुणों का अध्ययन किया। दूध के नमूनों में कोई कीटनाशक अवशेष और धातु संदूषक नहीं पाया गया। जैविक परीक्षण मापदंडों के तहत, कुल प्लेट काउंट (टीपीसी) 88 सीएफयू/एमएल और <10 सीएफयू/एमएल यीस्ट और मोल्ड काउंट था। औसत दूध वसा,

mainly for work, and seldom milk. The composition of donkey milk is not studied in much detail in India. ICAR-NRCE studied physical and bio-chemical properties of milk and milk powder of Halari donkeys. No pesticide residues and metal contaminants could be detected in milk samples. Under biological test parameters, the total plate count (TPC) was 88 cfu/ml and <10 cfu/mL of yeast and mould count. Average milk fat, ash and total solids content were found to be 0.2%, 0.39% and 8.2%, respectively. The rheological properties of donkey milk were determined in terms of flow behaviour property and temperature sweep. A Jenny dairy unit was established by purchasing 10 female donkeys and one male foal from Horse and Donkey Breeding Farm, Chanasma, These female donkeys will be used for research on donkey milk characterization.

The molecular markers are the revolutionizing tool that can be used for breed assignment as done in various domestic animals. By genotyping, allelic data of 24 microsatellite loci in 8 horse breed populations viz., Marwari, Kathiawari, Kachchhi-Sindhi, Thoroughbred, Zanskari, Bhutia, Spiti, and Manipuri were generated. A total of 29280 DNA fingerprinting data were generated which were formatted for computational analysis and breed prediction. After data pre-processing, different classifiers like BayesNet, NaiveBayes, Artificial Neural Network (ANN), Support Vector Machine (SVM), and Random Forest (RF) were deployed using scripts.

National Centre for Veterinary Type Cultures (NCVTC) has been recognized as a national repository by the Ministry of Environment, Forest & Climate Change under the Biological Diversity Act, 2002 to keep safe custody of voucher specimens of microorganisms pertaining to veterinary sciences. During the year, a total of 204 microbes were accessioned in the NCVTC repository leading to a cumulative strength of 4191. These include 1539 bacteria and 324 virus isolates from 32 different species. The important virus isolates accessioned notably include Avian nephritis virus, Fowl adenovirus, Bluetongue virus, Infectious bronchitis virus, Lumpy skin disease virus and SARS-CoV-2. The network centres at NIANP Bengaluru and NDRI Karnal were also able to strengthen their repositories of rumen and dairy microbes scaling up their collections to 562 and 677 microbes, respectively.

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

राख और कुल ठोस सामग्री क्रमशः 0.2%, 0.39% और 8.2% पाई गई। गर्दभ के दूध के रियोलॉजिकल गुण प्रवाह व्यवहार और तापमान स्वीप के संदर्भ में निर्धारित किए गए थे। एक जेनी डेयरी इकाई की स्थापना अश्व एवं गर्दभ प्रजनन फार्म, चानस्मा से 10 मादा गर्दभों और एक नर गर्दभ को खरीदकर की गई, इन मादा गर्दभों का उपयोग गर्दभ के दूध के लक्षण वर्णन पर शोध के लिए किया जाएगा।

आणविक मार्कर क्रांतिकारी उपकरण हैं जिनका उपयोग विभिन्न जानवरों की तरह नस्ल जानने के लिए किया जा सकता है। जीनोटाइपिंग द्वारा, 8 अश्व नस्लें जैसे मारवाड़ी, काठियावाड़ी, कच्छी–सिंधी, थोरोब्रेड, ज़ांस्कारी, भूटिया, स्पीति और मणिपुरी में 24 माइक्रोसेटेलाइट लोसाई के एलील डेटा तैयार किए गए थे। कुल 29280 डीएनए फ़िंगरप्रिंटिंग डेटा उत्पन्न किए गए थे जिन्हें कम्प्यूटेशनल विश्लेषण और नस्ल असाइनमेंट के लिए स्वरूपित किया गया था। डेटा प्री–प्रोसेसिंग के बाद, बेयसनेट, नाइवबेयस, आर्टिफिशियल न्यूरल नेटवर्क (एएनएन), सपोर्ट वेक्टर मशीन (एसवीएम), और रैंडम फॉरेस्ट (आरएफ) जैसे विभिन्न क्लासिफायर की स्क्रिप्ट का उपयोग किया गया ।

पशु चिकित्सा विज्ञान से संबंधित सूक्ष्मजीवों के नमूनों को सुरक्षित रखने के लिए जैविक विविधता अधिनियम, 2002 के तहत पर्यावरण, वन और जलवायु परिवर्तन मंत्रालय द्वारा राष्ट्रीय पशु चिकित्सा प्रारूप संवर्धन केन्द्र (एनसीवीटीसी) को राष्ट्रीय भंडार के रूप में मान्यता दी गई है। वर्ष के दौरान, कुल 204 रोगाणुओं को शामिल किया गया जिससे एनसीवीटीसी भंडार में अब कुल 4191 रोगाणु हो गए हैं । इनमें 1539 बैक्टीरिया और 32 विभिन्न प्रजातियों के 324 विषाणु शामिल हैं। विशेष रूप से परिग्रहण किए गए महत्वपूर्ण विषाणुओं में एवियन नेफ्रैटिस वायरस, फाउल एडेनोवायरस, ब्लूटॉन्ग वायरस, संक्रामक ब्रोंकाइटिस वायरस, लम्पीस्किन रोग वायरस और सार्स कोरोना विषाणु–2 शामिल हैं। राष्ट्रीय पशु पोषण एवं शरीर क्रिया संस्थान, बेंगलुरु और राष्ट्रीय डेयरी अनुसंधान संस्थान, करनाल के नेटवर्क केंद्र भी रुमेन और डेयरी रोगाणुओं के अपने संग्रह को क्रमशः 562 और 677 रोगाणुओं तक बढ़ाने में सक्षम रहे।

एनसीवीटीसी विषाणु भंडार को देश के विभिन्न भौगोलिक स्थानों से विषाणुओं को जोड़कर और विभिन्न जानवरों और कुक्कुट और नैदानिक नमूनों के संग्रह के माध्यम से मजबूत किया जा रहा है। वर्ष के दौरान, विभिन्न विषाणुओं के लिए कुल 100 विभिन्न जैविक नमूनों को संसाधित किया गया। कुल 27 नए वायरस शामिल किए गए।वर्ष के दौरान, 22 परिग्रहित विषाणुओं को संरक्षित किया गया। इसके अलावा, पहले से संरक्षित 34 विषाणुओं को पुनर्जीवित किया गया और उनकी व्यवहार्यता के लिए जाँच की गई व विषाणु व्यवहार्य पाए गए।

different animals and poultry. During the year, a total of 100 different biological samples were processed for isolation of different viruses. A total of 27 new viruses were accessioned.

During the year, 22 accessioned viruses were preserved through bulk production. Furthermore, 34 previously preserved viruses were revived and checked for their viability. The viruses were found viable.

Lumpy skin disease (LSD) has devastating economic impact. During the last decade, LSD had spread to climatically new and previously disease-free countries, which also includes its recent emergence in the Indian subcontinent. We have described the first successful isolation of LSDV in India, besides providing insights into the life cycle of Vero cell-adapted LSDV.

Apigenin, which is a dietary flavonoid, exerts a strong *in vitro* and *in ovo* antiviral efficacy against buffalopox virus (BPXV). Apigenin treatment was shown to inhibit synthesis of viral DNA, mRNA and proteins, without affecting other steps of viral life cycle such as attachment, entry and budding. The study also provides mechanistic insights on the antiviral activity of Apigenin and selection of potential Apigenin-resistant mutants upon long-term culture.

Emetine is a FDA-approved drug for the treatment of amebiasis. In the recent times we had also demonstrated the antiviral efficacy of emetine against some RNA and DNA viruses. Following emergence of the COVID-19., we further evaluated the *in vitro* antiviral activity of emetine against severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). Emetine treatment was shown to decrease viral RNA and protein synthesis without affecting other steps of viral life cycle such as attachment, entry and budding. Emetine targets SARS-CoV-2 protein synthesis which is mediated via inhibiting the interaction of SARS-CoV-2 RNA with eIF4E. This is a novel mechanistic insight on the antiviral efficacy of emetine. *In vitro* antiviral efficacy against SARS-CoV-2 and its ability to protect chicken embryos against IBV suggests that emetine could be repurposed to treat COVID-19.

CRISPR/Cas9-mediated genome editing applied to generate knockout (KO) cells with disrupted Phosphoribosyl Formyl Glycinamidine Synthase (FPPS) gene. PFAS knockout BHK21 cells were found to produce about 50-fold fewer virus particles then wild type BHK21 cells suggesting a proviral role of PFAS in FMDV life cycle. PFAS plays an antagonistic role in the

लम्पीस्किन रोग (एलएसडी) का विनाशकारी आर्थिक प्रभाव पड़ता है। पिछले दशक के दौरान, एलएसडी पहले से रोग मुक्त देशों में फैल गया, जिसमें भारतीय उपमहाद्वीप में इसका हालिया उद्भव भी शामिल है। हमने वेरो कोशिका–अनुकूलित एलएसडीवी के जीवन चक्र में अंतर्दृष्टि प्रदान करने के अलावा, भारत में एलएसडीवी के पहले सफल अलगाव का वर्णन किया है।

एपिजेनिन, जो एक आहार फ्लेवोनोइड है, भैंस के विषाणु (बीपीएक्सवी) के खिलाफ इन विट्रो और इन ओवो में एक मजबूत एंटीवायरल प्रभाव दिखाता है। एपिजेनिन विषाणु डीएनए, एमआरएनए और प्रोटीन के संश्लेषण को जीवन चक्र के अन्य चरणों जैसे जुड़ाव, प्रवेश और नवोदय को प्रभावित किए बिना बाधित करने में सक्षम पाया गया था। एपिजेनिन की एंटीवायरल गतिविधि और दीर्घकालिक संवर्धन पर संभावित एपिजेनिन–प्रतिरोधी म्यूटेंट के चयन पर यंत्रवत अंतर्दृष्टि भी शोधन से प्राप्त हुई ।

एमेटाइन अमीबीया के इलाज के लिए एफडीए द्वारा अनुमोदित दवा है। हाल के दिनों में हमने कुछ आरएनए और डीएनए विषाणुओं के खिलाफ एमेटाइन की एंटीवायरल प्रभावकारिता का भी अध्ययन किया। काविड–19 के बाद, हमने गंभीर तीव्र श्वसन सिंड्रोम कोरोनावायरस–2 (सार्स–कोरोना विषाणु–2) के खिलाफ एमेटीन की इन विट्रो एंटीवायरल गतिविधि का और मूल्यांकन किया। वायरल जीवन चक्र के अन्य चरणों जैसे जुड़ाव, प्रवेश और नवोदय को प्रभावित किए बिना वायरल आरएनए और प्रोटीन संश्लेषण को कम करने के लिए एमेटाइन को उपयुक्त पाया गया। एमेटाइन सार्स कोरोना विषाणु–2 के प्रोटीन संश्लेषण को लक्षित करता है जिसकी मध्यस्थता सार्स कोरोना विषाणु–2 के आर.एन.ए. की eIF4E के साथ इंटरेक्शन को रोककर की जाती है। यह एमेटाइन की एंटीवायरल प्रभावकारिता पर एक नवीन यंत्रवत अंतर्दृष्टि है। सार्स कोरोना विषाणु–2 के खिलाफ इन विट्रो एंटीवायरल प्रभावकारिता और इन्फेक्शियस ब्रोंकाईटिस विषाणु के खिलाफ चिकन भ्रूण की रक्षा करने की इसकी क्षमता से पता चलता है कि एमेटाइन को कोरोना–19 के इलाज के लिए पुर्नउद्देशित किया जा सकता है।

क्रिस्पर/केस–9 मध्यस्थ जीनोम एडिटिंग के द्वारा एफ.पी.पी.एस. जीन नॉकआउट कोशिकाओं को उत्पन्न किया गया। पीएफएएस नॉक आउट बीएचके 21 कोशिकाओं को लगभग 50 गुना कम वायरस कणों का उत्पादन करने के लिए उपयुक्त पाया गया था, फिर वाइल्ड प्रकार की बीएचके 21 कोशिकाएं एफएमडीवी जीवन चक्र में पीएफएएस की भूमिका का सुझाव देती हैं। पीएफएएस विभिन्न पिकोर्नाविरिडे परिवार के सदस्यों के जीवन चक्र में एक विरोधी भूमिका निभाता है। इसलिए

life cycle of different *Picornaviridae* family members. PFAS may therefore, serve as a potent host target for anti-FMDV drug development. The rise in incidence of drug resistance has prompted a shift towards development of antiviral drugs. We screened a library of small molecule chemical inhibitors and identified antiviral efficacy of sarco/endoplasmic reticulum calcium-ATPase (SERCA) inhibitor (Thapsigargin) and MNK1 (MAPK-interacting kinase). Calcium has been identified as an essential component in the different stages of the lifecycle of viruses. We tried to explore the role of calcium in the growth of NDV.

During the year, 50 cultures were accessioned in the bacterial repository. Besides this, 57 pathological/ other samples submitted/collected at NCVTC bacteriology laboratory lead to isolation of nearly 100 bacterial cultures, which are preserved in general preservation. Isolation and biochemical identification of bacterial isolates from pathological and environmental samples obtained from horse, donkey, buffalo, goat, sheep and fisheries pond water was performed. Bacterial cultures were biochemically investigated for identification.

Salmonella enterica serovar Gallinarum biovars Pullorum (S. Pullorum) and Gallinarum (S. Gallinarum) are the etiological agents of pullorum disease (PD) and fowl typhoid (FT) respectively, which cause huge economic loss to poultry industry in developing countries. Comparative genomics of 9 strains of *Salmonella* species elucidated the average genome size to be 46,57,781 bp, with average GC% of 52.19. This comparative genomic analysis of S. Gallinarum strains has provided a basis for future experimental studies to be carried out to decipher the underlying mechanisms driving the pathogenesis and virulence of this important pathogen.

The antimicrobial resistance surveillance work was extended outside Hisar city in the village area where rectal swabs from Beetel Goats, Nali and Munjal breeds of sheep (24 nos.) were collected from Village Thaska, Hisar. From 24 samples, 43 *Escherichia coli* isolates were isolated, confirmed biochemically and by duplex PCR (*lacy* & *pho*A positive). Disc diffusion test results showed that 13 (68.5%) isolates were resistant to one or more class of antimicrobials.

Widespread antibiotic use in medicine and livestock industry has contributed to the global spread of multidrug-resistant (MDR) bacterial pathogens, including *Acinetobacter baumannii*. Bacteriophages against this pathogen offer an alternative to

पीएफएएस एफएमडीवी विरोधी दवा के विकास के लिए एक शक्तिशाली मेजबान हो सकता है।

दवा प्रतिरोध की घटनाओं में वृद्धि ने एंटीवायरल दवाओं के विकास की ओर एक बदलाव को प्रेरित किया है। हमने छोटे अणु रासायनिक अवरोधकों के एक लाइब्रेरी की जांच की और पैरामाइक्सोवायरस (न्यूकैसल रोग विषाणु) के खिलाफ सार्को/एंडोप्लाज़मिक रेटिकुलम कैल्शियम–एटीपीस (एसईआरसीए) अवरोधक (टेस्पिगैरगिन) और एमएनके 1 (एमएपीके–इंटरेक्टिंग किनेज) अवरोधक की एंटीवायरल प्रभावकारिता की पहचान की। विषाणु के जीवनचक्र के विभिन्न चरणों में कैल्शियम को एक आवश्यक घटक के रूप में पहचाना गया है। हमने न्यू कैसल विषाणु के विकास में कैल्शियम की भूमिका का पता लगाने की कोशिश की।

वर्ष के दौरान, 50 जीवाणुओं को जीवाणु भंडार में शामिल किया गया। इसके अलावा, एनसीवीटीसी बैक्टीरियोलॉजी प्रयोगशाला में जमा/एकत्र किए गए 57 रोगजनक/अन्य नमूने से लगभग 100 जीवाणुकल्चर प्राप्त किये गए, जिन्हें सामान्य संरक्षण में संरक्षित किया जाता है। अश्वों, गर्दभ, भैंस, बकरी, भेड़ और मत्स्य पालन, तालाब के पानी से प्राप्त रोगजनक और पर्यावरणीय नमूनों से बैक्टीरिया प्राप्त किये गए और जैव रासायनिक जांच की गई।

साल्मोनेला एंटरिका सेरोवर गेलिनेरम बायावार्स पुलोरम (एस. पुलोरम) और गैलिनेरम (एस. गैलिनेरम) क्रमशः पुलोरम रोग और फाउल टाइफाइड के कारक हैं, जिससे विकासशील देशों में पोल्ट्री उद्योग को भारी आर्थिक नुकसान होता है। साल्मोनेला प्रजातियों के 9 उपभेदों के तुलनात्मक जीनोमिक्स ने औसत जीनोम आकार को 46,57,781 बीपी, 52.19 के औसत जीसी % के साथ स्पष्ट किया। इस तुलनात्मक जीनोमिक विश्लेषण ने इस महत्वपूर्ण रोगज़नक़ के रोगजनन और विषाणु के अंतर्निहित तंत्र को समझने के लिए भविष्य के प्रयोगात्मक अध्ययनों के लिए एक आधार प्रदान किया है।

रोगाणुरोधी प्रतिरोध निगरानी कार्य को हिसार शहर से बाहर गावों के क्षेत्र में विस्तारित किया गया था। गाँव ठस्का, (हिसार) से बीटेल बकरियों, नाली और मुंजाल नस्ल की भेड़ों से रेक्टल स्वैब एकत्र किए गए थे। 24 नमूनों में से 43 एस्चेरिचिया कोलाई आइसोलेट्स को जैव रासायनिक रूप से और डुप्लेक्स पीसीआर द्वारा अलग किया गया। डिस्क प्रसार परीक्षण के परिणामों से पता चला है कि 13 (68.5%) आइसोलेट्स एक या एक से अधिक वर्ग के रोगाणुरोधी प्रतिरोधी थे।

दवा और पशुधन उद्योग में व्यापक एंटीबायोटिक उपयोग ने एसिनेटोबैक्टर बॉमनी सहित मल्टीड्रग–रेसिस्टेंट (एमडीआर) बैक्टीरियल रोगजनकों के वैश्विक प्रसार में योगदान दिया है।

antibiotics. A highly lytic phage - VTCCBPA145 against *Acinetobacter baumannii* was isolated from sewage by enrichment technique. The phage produced clear plaques with a zone of halo.

Phage therapy using a characterized phage was carried out in Swiss albino mice after 10-12 days of parturition with 4-5 litter size. The host bacteria used for the infection of mammary glands was *S. aureus (*MTCC96). Beneficial effects for curing mastitis were observed in mice model and need to be validated further in large animal model.

The complete genome analysis of bacteriophage VTCCBPA139 against *Proteus mirabilis* was carried out. This phage was isolated from poultry litter against a drug resistant *P. mirabilis*.

ICAR-NRCE executed a total of 33 research projects out of which 12 research projects were externally funded. A total of 51 papers were published in national and internal journals of high repute. ICAR-NRCE also developed a diagnostic kit entitled *"Japanese Encephalitis Virus Antibody Test kit, iELISA for equids and pigs"*. The kits were released by Hon'ble Union Minister of Agriculture & Farmers' Welfare during the 91st Annual General Meeting of ICAR. A technology entitled, *"Donkey Milk Based Products (Bathing Soap, Body Butter and Lip Balm)"* was transferred for commercial use. During the year one Indian patent was granted for the assay developed for diagnosis of glanders. ICAR NRCE inked four MoUs with universities and research organizations in India.

The centre organized four training programmes on use of equine operated agricultural implements, one short course, and four webinars for the stakeholders. Nearly 300 stakeholders were trained in these programs. During the period 18 students were guided for their Masters, PhD and Post-doctoral programs.

इस रोगज़नक़ के खिलाफ जीवाणुभोजी एंटीबायोटिक दवाओं के विकल्प की तरह चुना जा सकता है । एसिनेटोबैक्टर बॉमनी के खिलाफ एक अत्यधिक लाइटिक जीवाणुभोजी–वीटीसीसीबीपीए 145 को संवर्धन तकनीक द्वारा सीवेज से निकाला गया। जीवाणुभोजी ने प्रभामंडल के साथ स्पष्ट प्लाक्स बनाईं।

प्रोटीयस मिराबिलिस के विरुद्ध जीवाणुभोजी का संपूर्ण जीनोम विश्लेषण किया गया। इस जीवाणुभोजी को एक दवा प्रतिरोधी पी. मिराबिलिस के खिलाफ पक्षियों से निकाला गया था।

भा.कृ.अनु.प.– रा.अ.अनु.के. ने कुल 33 अनुसंधान परियोजनाओं को क्रियान्वित किया, जिनमें से 12 अनुसंधान परियोजनाएं बाह्य वित्त पोषित थी। उच्च प्रतिष्ठित राष्ट्रीय और आंतरिक पत्रिकाओं में कुल 51 शोध पत्र प्रकाशित हुए। भा.कृ.अनु.प.– रा.अ.अनु.के. ने जापानी एन्सेफलाइटिस वायरस एंटीबॉडी टेस्ट किट, आइ–इलिसा फॉर इक्विड्स एंड पिग्स नामक एक नैदानिक किट भी विकसित की है। किटों को भा.कृ.अनु.प. की 91वीं वार्षिक आम बैठक के दौरान माननीय केंद्रीय कृषि और किसान कल्याण मंत्री द्वारा जारी किया गया था। गर्दभ दूध पर आधारित उत्पाद (साबुन, बॉडी बटर और लिप बाम) नामक तकनीक को व्यावसायिक उपयोग के लिए स्थानांतरित किया गया। वर्ष के दौरान ग्लैंडर्स के निदान के लिए एक भारतीय पेटेंट प्रदान किया गया। भा.कृ.अनु.प.–रा.अ.अनु.के. ने भारत में विश्वविद्यालयों और अनुसंधान संगठनों के साथ चार समझौता ज्ञापनों पर हस्ताक्षर किए।

केंद्र ने हितधारकों के लिए अश्वों से संचालित कृषि उपकरणों के उपयोग पर चार प्रशिक्षण कार्यक्रम, एक लघु पाठ्यक्रम और चार वेबिनारों का आयोजन किया। इन कार्यक्रमों में लगभग 300 हितधारकों को प्रशिक्षित किया गया। इस अवधि के दौरान 18 छात्रों को उनके परास्नातक, पीएचडी और पोस्ट डॉक्टरेट कार्यक्रमों के लिए निर्देशित किया गया।

Introduction

Horse symbolizes power, bravery, wisdom and beauty. They have been domesticated by humans since historic times. Horses hold a special place in culture and rise of empires as they served unsung hero of wars and played a key role in the rise of larger human settlements and great civilizations. In India, the Aryans started domestication of horses and since then many wars were won by cavalry of this species. Human association has been thousands of years old with horse, however with automobiles it is only a few hundreds of years. But with the advent of modern means of transportation, utility of equines is decreasing resulting in decline in their population. Horses still remain preferred means of transport in hilly and desert terrains for the rural poor, nomadic tribes in the north, north-west and north- eastern parts of India. Mules and donkeys have been a better choice for carrying packs in the mountains and hence are more popularly known as tractor and truck of the hilly tract of the mountains.

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 (NRCE) on November 26, 1985 at Hisar (Haryana). The state- of-the art laboratories and facilities at the main campus of NRCE in Hisar have been undertaking research in areas of equine virology, bacteriology, parasitology, immunology, pathology, medicine, biochemistry and biotechnology. 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.

Equine Production Campus (EPC) was established in 1989 at Bikaner (Rajasthan) to undertake research on equine production, genetics and breeding, management, reproduction, physiology and nutrition. Bikaner campus has well maintained herds of Marwari, Kathiawari, Zanskari and Manipuri horses and Halari and exotic donkeys. The National Centre for Veterinary Type Cultures (NCVTC) was established in the year 2005 at ICAR-NRCE, Hisar main campus for collection and preservation of microbes of animal origin having veterinary importance. Presently, the Centre is working through 14 network units spread throughout the country.

MANDATE OF NRCE

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

OBJECTIVES OF 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 & socio-economic scenario.

SALIENT ACHIEVEMENTS

During past 35 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. The Centre is striving hard for conservation and characterization of Indian breeds of equines in the country and has even established nucleus herds of representative breeds of equines at its Bikaner campus. Some of the major achievements and accolades of the Centre are enlisted below:

Development of diagnostics for equine diseases

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

- HERP kit for field diagnosis of equine herpesvirus 1 (EHV1) infection.
- COFEB kit for diagnosis of *Theileria equi*.
- membrane potential of the stallion spermatozoa.

- A neutralizing monoclonal antibody-based diagnostic kit 'Equiherpes B-ELISA' for EHV1 antibody detection.
- A type-specific ELISA and real-time PCR for differentiation of EHV1 and 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 of *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 serodiagnosis of glanders and equine infectious anemia.
- Lateral flow assay based rapid diagnostic for *Theileria equi* infection.
- LFA kit for glanders.
- LFA kit for EIA.
- Nested (gB-nPCR) and real-time PCR (gB- qPCR) targeting gB were standardized for detection of EHV1 latency.
- Indirect ELISA was developed using recombinant protein of 444 bp for detection of JEV specific antibodies in horse and pig. The assay has been transferred to NE region labs and is used for validation on pig serum samples.
- Multiplex PCR has been standardized to differentiate *Streptococcus equi subsp. equi and zooepidemicus.*
- Lateral flow assay was validated for rapid diagnosis of trypanosomosis using different *T. evansi* antigens.
- ELISA has been developed to detect *T. evansi* antibodies in multiple sps.
- Peptide ELISA for serodiagnosis of BHV-1.
- Multiplex PCR for simultaneous diagnosis of *Theilleria equi and Babesia caballi* infection.
- Recombinant protien (BC-48 gene) based ELISA for detection of *B. caballi*.

Development of vaccines and immuno-biologicals

- Inactivated EHV1 vaccine "Equiherpabort" using indigenous virus for prevention of abortions in mares.
- Updated equine influenza vaccine using indigenous isolate (A/equi-2/Ludhiana/87). The vaccine was updated in 2008-09 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*.
- EHV-1 vaccine was formulated with inactivated EHV-1 vaccine using montanide adjuvant and tested in murine model for generation of immune response.

Surveillance and monitoring of equine diseases in India

ICAR-NRCE is involved in nation-wide monitoring and sero-surveillance of important equine infectious diseases with a view to manage, control and eradicate diseases. Some of the salient achievements under sero-monitoring include:

- India has gained OIE disease-free status for African horse sickness (AHS) in 2014 based on sero-monitoring data generated by NRCE.
- Clinical cases of equine infectious anemia (EIA) have not been reported since 1997. Only two sero-positive cases (one mule from Uttarakhand in 2009 and one horse from Haryana in 2011) were detected and culled. Control of EIA in India was possible due to timely diagnosis and implementing package of practices formulated by NRCE.
- Outbreaks of glanders in equines have been detected since 2006-07 from different states and control measures are being adopted for preventing their further spread.
- Effective control of equine influenza outbreak of 1987 (involving 83000 equines) was done by implementing

biosecurity and development of effective vaccine. 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.

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, A/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 equines.
- The *in-vitro* cultivation of *Trypanosoma evansi* and *Theileria equi* was successfully established.
- Experimental mouse models for equine influenza and equine herpesvirus-1 infections were developed.
- Complete genome sequencing of two EHV1 isolates was carried out using NGS. The primary NGS data obtained covered up to 90% of 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 with the reference isolates V592.

Phenotypic and genotypic characterization of Indian equine breeds

- 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.
- High genetic diversity observed between Spiti and Thoroughbred, followed by Spiti and Kathiawari while Zanskari and Manipuri are the least differentiated.
- Indian breeds form three distinctive clusters based on Bayesian analysis: (a) Kathiawari; (b) Zanskari, Spiti & Manipuri ponies and (c) Bhutia.

Establishment of nucleus herd

- 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:
- Marwari horses from Rajasthan; Kathiawari horses from Gujarat; Zanskari ponies from Zanskar valley (Jammu & Kashmir) and Manipuri ponies from Imphal (Manipur) and herds of indigenous and exotic donkeys are being maintained:
- Large white (Halari) donkeys for conservation and improvement of donkeys are being maintained.
- Poitou donkey herd for production of superior mules are also maintained.

Improvement in production potential of equines

- In order to conserve the germplasm of indigenous equine breeds, cryopreservation of semen of Marwari, Kathiawari, Zanskari and Manipur stallions and Halari & Poitou donkeys has been done.
- 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 donkey and horse 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 have been studied and established their correlation with DNA integrity and mitochondrial

- **Utilization of equine energy in agricultural activities**
- Single animal drawn matching plough, seed drill (two furrow) and harness have been designed and developed for donkeys and mules for agricultural operations like ploughing and sowing.
- The mules have been used for chaff cutting operation with average output capacity of 660 kg/hour of chopped bajra straw in rotary mode chaff cutter.
- Draughtability studies conducted on adult donkeys using conventional pneumatic two wheel cart showed that Halari donkeys may pull draft of 33Kg for 3 hour, 44, 55, 66 Kg for 2 hour, 77, 88 Kg for 1 hour without much stress to donkeys.
- The technique of vermicomposting of equine dung has been optimized for use in agricultural fields.

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 diagnostic kit for forecasting equine herpesvirus-1 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.

Patents filed

- A highly sensitive kit for detection of antibodies against *Theileria equi* in serum of equids. Application No. 2763/DEL/2012 dated 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/2010 and PCT/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.
- Polymeric metal nanocomposites and methods of synthesis thereof Application No. 201911009696, Dated 13.03.2019.

Services

- ICAR-NRCE provides following services to the farmers and equine breeders:
- Disease diagnostic services for various infectious and non-infectious diseases to equine owners, breeders, state animal husbandry departments, police and army horses.
- Surveillance, monitoring and control of equine infectious diseases in India.
- 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.
- Ar tificial 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.
- Onsite and online consultancy in equine health and production, including toll-free telephonic advisory at Hisar and Bikaner campuses for farmers and stakeholders.

- Trainings and supply of educational materials for equine management, production and health.
- Organization of health camps, awareness campaigns and farmers meets in different areas of the country.
- During pandemic of Corona virus ICAR-NRCE served as a COVID-19 testing facility amongst one of the 4 labs of ICAR.

National Centre for Veterinary Type Cultures

National Centre for Veterinary Type Cultures (NCVTC) initiated its activities in the year 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 14 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 in conservation of animal microbial diversity in three specialized areas: veterinary microbes at 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.

During the year 2020, a total of 204 microbes were accessioned in the repository leading to a cumulative strength of

Year	2009 -15	2015 -16	2016 -17	2017 -18	2018 -19	2019 -20	Till Dec 31, 2020	Current Strength (31 December, 2020)
Veterinary Microbes								
Bacteria	927	110	164	70	123	95	50	1539
Virus	156	14	28	27	31	44	24	324
Bacteriophage	32	44	29	24	8	8	48	193
Recombinant clone	466	45	10	36	16	8	0	581
Phage library	27	0	0	0	0	0	0	27
Genomic DNA	223	57	0	8	0	0	0	288
Total	**1831**	**270**	**231**	**165**	**178**	**155**	**122**	**2952**
Rumen microbes								
Anaerobic bacteria	142	74	37	37	49	46	62	447
Fungi/Yeast	107	0	0	0	0	0	0	107
Meth. Archae	8	0	0	0	0	0	0	8
Total	**257**	**74**	**37**	**37**	**49**	**46**	**62**	**562**
Dairy microbes								
Bacteria	468	39	40	30	36	44	20	677
Total	**468**	**39**	**40**	**30**	**36**	**44**	**20**	**677**
Grand Total	**2556**	**383**	**308**	**232**	**263**	**245**	**204**	**4191**

4191. In the bacterial repository at NCVTC, 50 new bacteria were accessioned during the year, making cumulative culture collection of 1539 bacteria of veterinary importance. In the virus repository, a total of 24 virus isolates were processed, of which all were accessioned in the repository that increased the strength of the virus repository to 324 virus isolates from 32 different species. The important virus isolates accessioned include, Avian nephritis virus (04), Fowl adenovirus (11), Bluetongue virus (4), Infectious bronchitis virus (4) and Lumpy skin disease virus (2) and SARSCoV-2 (2). During the current year, emphasis was on the isolation and characterization of bacteriophages against pathogens of mastitis: *Staphylococcus* sp., and *Streptococci* sp. A total of 48 bacteriophages were isolated and preserved. In rumen microbial repository at NIANP Bengaluru, with the accessioning of 62 rumen bacteria, the total strength of the rumen microbe's repository has reached to 562. Furthermore, with the deposition of 20 bacteria, the dairy microbe's repository at NDRI, Karnal has also increased its strength to 677 dairy microbes.

The distribution of microbes for teaching, research and development of new technologies is another mandated activity of NCVTC. In this regard, bacterial cultures (*Truperella pyogenes*, *Pseudomonas aeruginosa*, *Enterobacter spp*, *Streptococcus pyogenes*, *E. coli*, *Streptococcus agalactiae*, *Staphylococcus aureus*, *ESBL producing Klebsiella*, *Klebsiella pneumoniae*, *Actinomycetes spp*, *Actinobacillus spp*, *Clostridium perfringens*, *Bacillus subtillis*, *Corynebacterium spp.*), Bacteriophages against *E. coli*, *Bacillus sp. and Staphylococcus aureus* as well as cell lines (RK13, Porcine Stable, PK15, Hela & Vero) were distributed to different Institutes/Universities in India for research and teaching purposes.

Some of the salient achievements of NCVTC are listed below.

Veterinary Microbes

- First laboratory confirmed camel pox virus zoonosis.
- First report on isolation and genetic characterization of swinepox virus from India.
- Accessioning of vaccine strains of viruses *viz*., Peste des petits ruminants virus, Sheeppox (Srinagar strain), Goatpox virus (Uttarkashi strain), Orf virus (Mukteswar strain), NDV (R2B strain) and NDV (F strain).
- Complete genome sequencing of Classical swine fever virus (2), chicken astro virus(2) & porcine circo virus (4) .
- First isolation and characterization of *Bordetella bronchiseptica*, *Actinobacillus equilli*, *Staphylococcus hyicus*, *Trueperella pyogenes*.
- Whole genome sequencing of *Pasteurella multocida* sub spp. *multocida* B:2 serotype.
- First isolation and identification of *Moraxella* (*Branhamella) ovis* from ovine keratoconjunctivitis in sheep and methicillin-resistant coagulase negative *Staphylococcus sciuri* from goats.
- Whole genome sequencing of *Trueperella pyogenes*, *Bordetella bronchiseptica*, *Pasteurella multocida*, *Actinobacillus equuli* and *Salmonella* Gallinarum.
- Accessioning of rare strains of bacteria: *Campylobacter spp*., *Bacillus megaterium*, *Enterococcus casseliflavus*, *E. cecorum*, *Barrientosiimonas humi*, *Corynebacterium amycolatum*, *Enterococcus devriesei*, *E. hirae*, *E. faecium*, *Nocariopsis alba*, *Ignatzschineria larvae* and *Escherichia hermanii*.
- Isolation of bacteriophages against a variety of pathogenic bacteria was added to NCVTC repository, including a novel thermotolerant bacteriophage isolated from Ganga river water.
- The whole genome sequencing of *Proteus mirabilis* phage VTCCBPA139 against MDR host.
- Phage therapy against MDR *K. pneumoniae* in mouse model.

Rumen Microbes

- Isolation and characterization of seven tannin degrading bacteria *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 - *Anaeromyces* sp., *Orpinomyces intercalaris* and *Orpinomyces joyonii* from buffaloes; *Piromyces* sp. and *Neocallimastix sp*. from goats.

Dairy Microbes

- Preservation of dairy microbes, viz, *Lactobacillus* spp., *Lactococcus* spp., *Lactococcus lactis* ssp. lactis, *Lactococcus lactis*. *ssp*. Cremoris, *Lactococcus lactis ssp*. lactis bv. Diacetylactis, *Streptococcus thermophilus*, *Leuconostoc* ssp., *Bifidobacterium* sp. *Bifidobacterium dentium*, *Bifidobacterium longum*, *Micrococcus* sp., *Kluyveromyces lactis* and *Saccharomyces bisporus*. Combination of *L. lactis* ssp lactis-C12 and *Leuconostoc mesenteroides* ssp. mesenteroides is very suitable for curd and buttermilk 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 milkcereal fermented products.

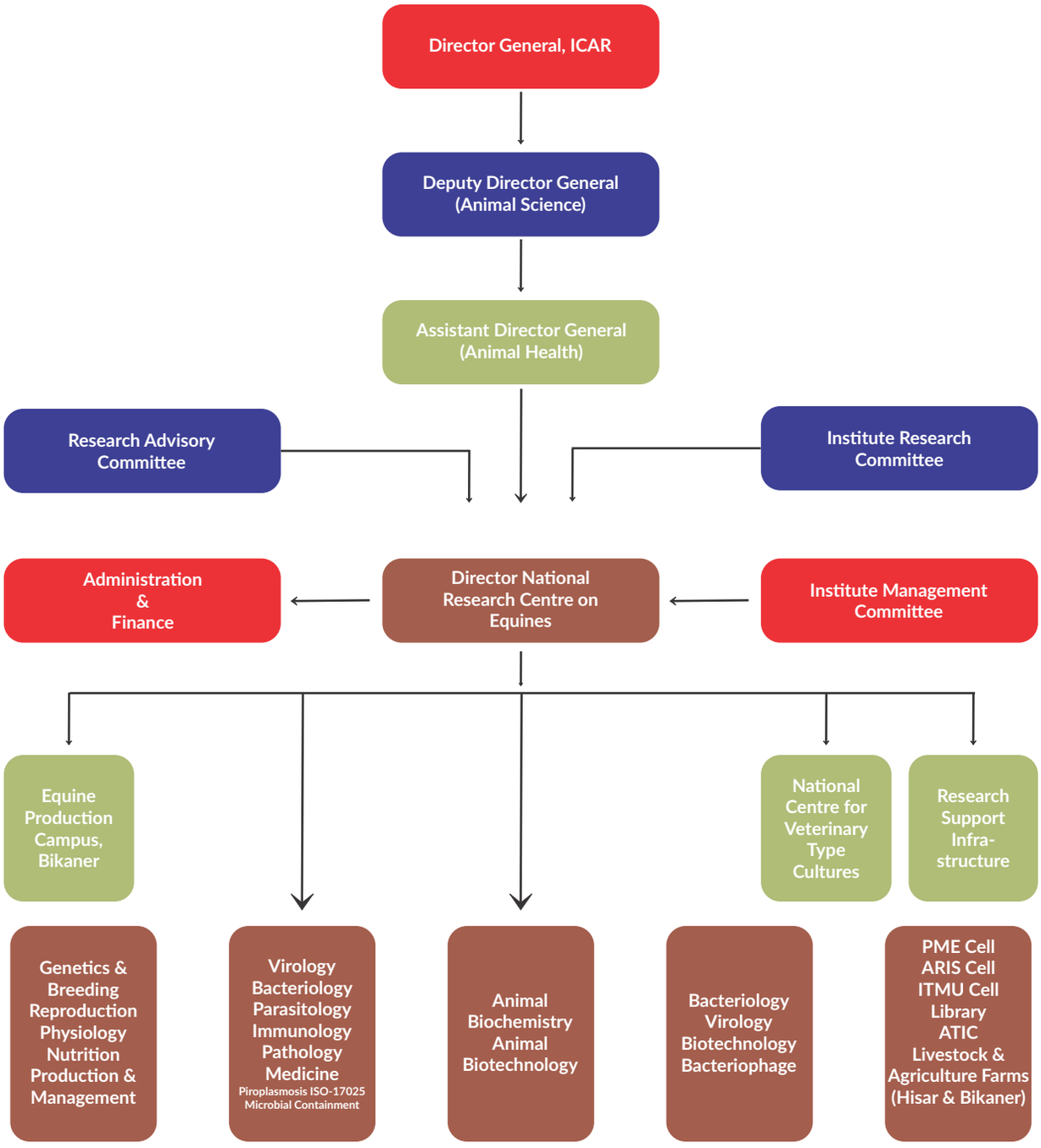
SUMMARY OF EXPENDITURE & REVENUE GENERATION

Summary of Expenditure	Year 2020 (In Lakh)		
	NRCE	NCVTC	Total
Establishment charges including LSP/PF, wages, OTA	1168.85	0	1168.85
Travelling allowances & HRD	14.14	0.37	14.51
Others charges including equipments & recurring charges	637.34	214.13	851.47
Works	41.90	0	41.90
Loans and Advances	0	0	0
Total Plan Expenditure	1862.23	214.50	2076.73
Summary of Revenue Generation			
Sale of farm produce	17.80	0	17.80
Sale of livestock	1.57	0	1.57
Sale of publications and advertisements	0	0	0
License fee	1.60	0	1.60
Interest on loans and advances	6.10	0	6.10
Interest on short term deposits	17.06	0	17.06
Contractual diagnostic services	47.83	0	47.83
Receipt from services	0.98	0	0.98
Other miscellaneous receipts	30.34	0	30.34
Eco-tourism	3.52	0	3.52
Total	**126.80**		**126.80**

STAFF POSITION AT NRCE & NCVTC

Name of the Post	NRCE			NCVTC		
	Sanctioned	Filled	Vacant	Sanctioned	Filled	Vacant
Director	1	0	1	-	-	-
Scientific	26	14	12	10	8	2
Technical	24	21	3	-	-	-
Administrative	14	11	3	-	-	-
Supporting	20	20	0	-	-	-

ORGANISATIONAL STRUCTURE OF NRCE



RESEARCH ACHIEVEMENTS

Sero-surveillance and monitoring of equine infectious diseases in India

Surveillance and monitoring of equine infectious diseases is one of the continuous service projects of the institute. The project aims to keep vigilance on the status of economically important equine diseases of viral, bacterial or parasitic origin. During the year 2020, a total of 2299 equine serum samples from 10 states were tested for various diseases like Equine Infectious Anaemia (EIA), Equine Influenza (EI), Equine Herpes Virus-1 (EHV-1), Japanese Encephalitis/West Nile Virus (JEV/WNV), *Trypanosoma evansi* (Trypanosomosis), Piroplasmosis, *Salmonella* Abortus equi and Brucellosis. Total number of positive cases and sero-positive percentage are indicated in Table 1. The highest sero-prevalence was observed for equine *piroplasmosis* (51.80%) followed by JE/WNV (9.13%), EHV-1 (2.87%) and *Trypanosoma evansi* (1.43%). None of the equines were found positive for equine influenza, equine infectious anemia, brucellosis and *Salmonella* Abortus equi.

Table 1: Sero-prevalence of important equine diseases among indigenous equines, numbers in parenthesis indicates sero-positive samples.

State	EIA	EI	Piroplasmosis	EHV-1	*T. evansi*	JE/WNV	*Sal.* Ab.equi	Brucellosis
New Delhi	53	53	53(14)	53(4)	53	53(1)	53	53
Haryana	526	526	526(317)	526(10)	526(1)	526(36)	526	526
Uttarakhand	249	249	249(148)	249(25)	249(2)	249(34)	249	249
Uttar Pradesh	1206	1206	1206(565)	1206(20)	1206(17)	1206(123)	1206	1206
Rajasthan	15	15	15(7)	15	15	15	15	15
Chhattisgarh	11	11	11(4)	11(6)	11	11(4)	11	11
Jharkhand	33	33	33	33	33	33(2)	33	33
Madhya Pradesh	46	46	46(30)	46	46(4)	46(3)	46	46
Himachal Pradesh	110	110	110(64)	110	110(1)	110	110	110
Karnataka	50	50	50(42)	50(1)	50(8)	50(7)	50	50
Total	**2299**	**2299**	**2299(1191)**	**2299(66)**	**2299(33)**	**2299(210)**	**2299**	**2299**
Sero-prevalence (%)	**-**	**-**	**51.8**	**2.87**	**1.43**	**9.13**	**-**	**-**

The samples tested under disease investigation are shown in Table 2. Testing of 192 samples for equine influenza exhibited negative results for H3N8 antibodies. For annual reconfirmation of AHS free status, a total of 185 samples from 5 states were tested and found negative. For EIA, 9397 serum samples obtained from 13 states were found negative by Coggin's test. State wise distribution of samples tested for EIA surveillance is shown in Table 3. No EIA positive case has been reported in India for the last 10 years. This surveillance data would be of immense help for obtaining EIA free status of the country. Samples tested for other diseases under disease investigation were found negative.

Microbiological analysis was carried out on 244 clinical samples including nasal swabs, tissues, abscesses, aborted fetus etc. originating from Haryana, Uttar Pradesh, Himachal Pradesh and Delhi yielding 51 bacterial isolates including *Klebsiella*

Table 2: Number of samples tested under disease investigation

Disease	No. of samples tested
Equine Influenza	192
Equine Infectious Anaemia (EIA)	9397
African Horse Sickness (AHS)	185
Piroplasmosis	6
T. evansi	14
EHV-1	144
JE/WNV	155
Histopathology	6
Covid-19	7
Total	**10111**

Table 3: State wise distribution of equine samples tested for EIA under disease investigation

Sr No	State	Sample no.
1	Uttar Pradesh	8401
2	Uttarakhand	28
3	Delhi	541
4	J& K	7
5	Haryana	43
6	Maharastra	36
7	Rajasthan	2
8	Himachal Pradesh	62
9	Karanatka	14
10	Chandigarh	4
11	Chhattisgarh	20
12	Madhya Pradesh	29
13	Gujarat	210
	Total	**9397**

pnuemoniae (6), *E. coli* (11), *Rhodococcus equi* (9), *Streptococcus zooepedemicus* (22) and *Burkholderia mallei* (3) (Table 4).

Revenue generation through contractual diagnostic services and consultancy

Under contractual diagnostic services, a total 6775 samples were received from race courses, turf club, stud farm,

Table 4: Bacteria isolated from 244 bio-samples yields from different states

Organism	No.	Sample type/ swab site	Place
Klebsiella pneumoniae	6	Heart (3), Stomach (1), Spleen Rajasthan (2)	
E. coli	11	Liver (1), Kidney (1), Small & Large Intestine (4), Tissue (1), Heart (1), Spleen (1), Lung (1), Rectal Swab (1)	Rajasthan (8), Haryana (1)
Streptococcus zooepidemicus	22	Spleen (1), Small & Large Intestine (1), Wound Swab (6), Skin Sample (3), Pus (9), Nodule Swab (1), Nasal Swab (1)	Rajasthan (2), Haryana (20)
Rhodococcus equi	9	Soil sample (2), Nasal Swab (1), Fecal Sample (2), Pus (3), Hand Sample (1)	Rajasthan (1), Haryana (8)
Burkholderia mallei	3	Pus swab (2), Feed (1)	Haryana (3)

riding schools, animal quarantine, certification services (AQCS) and other organized sector during the year. These samples were tested for various notifiable and exotic diseases to check ingression of diseases and monitor elite horses in private sectors. A total of 2737 sera samples for EIA and 3316 samples for glanders were found negative. Among exotic diseases, 208 swab samples from for contagious equine metritis (CEM), 70 samples for equine viral arteritis (EVA), 83 samples for African horse sickness (AHS) and dourine were received from AQCS, Govt. of India, collected from imported equines. All the samples were found negative for these exotic diseases. Revenue of Rs 49.10 lakhs was generated through contractual diagnostic services (Table 5).

Table 5: Number of samples tested and revenue generation through contractual diagnostic services

	EIA	Glanders	CEM	Dourine	AHS	EVA	*T. equi*	*B. caballi*	WNV	*T. evansi*	Culture isolation	Total
Sample No.	2737	3316	260	139	139	130	20	20	9	2	3	6775
Revenue (Rs.)	1505350	2321200	416000	152900	152900	260000	40000	40000	18000	1100	3000	4910450

(H. Singha, K. Shanmugasundaram, B.R. Gulati, Nitin Virmani, Rajender Kumar, Sanjay Kumar, Sanjay Barua, R.K. Vaid, Ramesh Dedar, Anju Manuja, Balvinder Kumar and Yash Pal)

Glanders Surveillance

National Action Plan for control and eradication of glanders in India has been launched by the Ministry of Fisheries, Animal Husbandry and Dairying, Government of India in 2019 (http://dadf.gov.in). The overall objective is surveillance, control and eradication of Glanders in equines from India. This action plan has been framed for surveillance of the entire equine population of the country reared in different management and animal husbandry practices following the conceptual framework of the OIE Terrestrial Code and the OIE Terrestrial Manual. Annual glanders surveillance report is presented below.

In 2020, a total of 22130 equine sera from 258 districts of 16 states were collected and tested for glanders. Zone wise surveyed states belong to Northern India (Jammu & Kashmir, Himachal Pradesh, Punjab, Haryana, Uttarakhand, Delhi and Uttar Pradesh), Western India (Rajasthan, Gujarat and Maharashtra), Central India (Chhattisgarh and Madhya Pradesh), Southern India (Karnataka and Andhra Pradesh) and Eastern India (Bihar and West Bengal). State wise glanders surveillance data is shown in Table 6. Out of these, 139 glanders positive cases were reported in 54 districts of 10 states. Glanders affected states (Table 7) included Uttar Pradesh (n=76), Uttarakhand (n=10), Haryana (n=8), Delhi (n=5), Jammu & Kashmir (n=7), Himachal Pradesh (n=7), Punjab (n= 1), Gujarat (n=8), Maharashtra (n=9) and Madhya Pradesh (n=8). It was found that 50% of the samples and glanders positive cases were from Uttar Pradesh. From zoonotic point of view, 87 sera from occupationally exposed humans (Veterinary Officers, equine handlers, laboratory workers) were tested and local form of glanders was found in one equine handler.

For rapid and efficient execution of surveillance activities, glanders ELISA developed by NRCE has been provided to 12 state diagnostic laboratories/RDDLs namely Gujarat, Haryana, Himachal Pradesh, Punjab, Rajasthan, Maharashtra, Karnataka, Jammu & Kashmir, Bihar, Chhattisgarh and Madhya Pradesh. Presently, glanders ELISA kit is commercially available and being used by State laboratories. Out of the total samples, 7775 equine samples were screened by ELISA at 10 State Lab/RDDLS (Gujarat, Haryana, Himachal Pradesh, M.P., Punjab, Rajasthan, Maharashtra, Jammu & Kashmir, Bihar, and Karnataka) during the year. As per guidelines, ELISA positive samples were retested and confirmed by complement fixation test (CFT) at NRCE.

Taking account of past four year surveillance data, it was observed that only 10-11 states regularly participated in the glanders surveillance. On the other hand, negligible, irregular or no surveillance was done in North-East and South

Table 6: Glanders surveillance data

Sr No	State	No of samples tested at NRCE	No of samples tested at State Lab/RDDLs	No of districts surveyed	Positive cases
1	Uttar Pradesh		-	75	76
2	Haryana	919	1198	22	8
3	Punjab	45	745	20	1
4	Himachal Pradesh	162	640	6	7
5	Uttarakhand	390	-	8	10

6	Delhi	613	-	3	5
7	Jammu & Kashmir	7	965	6	7
8	Madhya Pradesh	84	816	16	8
9	Gujarat	30	1314	21	8
10	Maharashtra	35	764	24	9
11	Rajasthan	15	1183	34	0
12	Chhattisgarh	111	-	9	0
13	Chandigarh	4		-	0
14	Andhra Pradesh	36		1	0
15	Karnataka	64	34	3	0
16	West Bengal	22	-	1	0
17	Bihar	114	116	9	0
Total		**14355**	**7775**	**258**	**139**

Table 7: Glanders affected districts in India

State	Positive cases	Dist./ Place
Uttar Pradesh	76	Agra, Aligarh, Amorha, Bahraich, Barabanki, Bhadoi, Bulandshahr, Deoria, Fatehapur, Ghazipur, Gonda, Jaunpur, Kanpur Dehat, Kasganj, Kaushambi, LakhimpurKheri, Lucknow, Meerut, Moradabad, Muzaffarnagar, Prayagaraj, Rampur, Saharanpur, Sambhal, SantKabir Nagar, Sharvasti, Sultanpur, Varanasi **(28 Districts)**
Maharashtra	9	Aurangabad, Jintur, Parbhani, Hingoli, Buldhana and Akola
Haryana	8	Rohtak, Hisar, Ambala and Faridabad
Madhya Pradesh	8	Ujjain, Vidisha and Indore
Jammu & Kashmir	7	Udhampur, Rajouri, Ramban and Reasi
Himachal Pradesh	7	Mandi
Uttarakhand	10	Dehradun, Nanital, Almora and Haridwar
Gujarat	8	Sabarkantha and Mahisagar
Delhi	5	Jwala Nagar, Delhi
Punjab	1	Shaheed Bhagat Singh Nagar
Total	**139**	**54 Districts**

India. Therefore, pro-active participation of all State Animal Husbandry Departments in the surveillance programme is necessary to assess state wise sero-prevalence, epidemiology and risk factors to devise future strategies for control and eradication of glanders in India.

(Harisankar Singha, K. Shanmugasundaram and Yash Pal)

Analysis of phylogenic relationship of Indian *Burkholderia mallei* strains based on 16S rRNA and ITS gene sequences

Burkholderia mallei is a Gram-negative bacterium and causative agent of glanders- a fatal devastating infectious disease of horses, donkeys and mules (Fig 1). *B. mallei* is believed to be evolved from a clone of *B. pseudomallei*- a saprophyte and causative agent of melioidosis by genome reduction and rearrangement events. In India, equine glanders cases were regularly reported since 2006. However, genetic diversity of Indian *B. mallei* strains isolated from glanders affected equines at different geographical space and time has not been investigated. Here, we report phylogenetic analysis of 39 *B. mallei* isolates recovered between 2013 to 2019 by 16S rRNA and ITS gene sequences analysis.

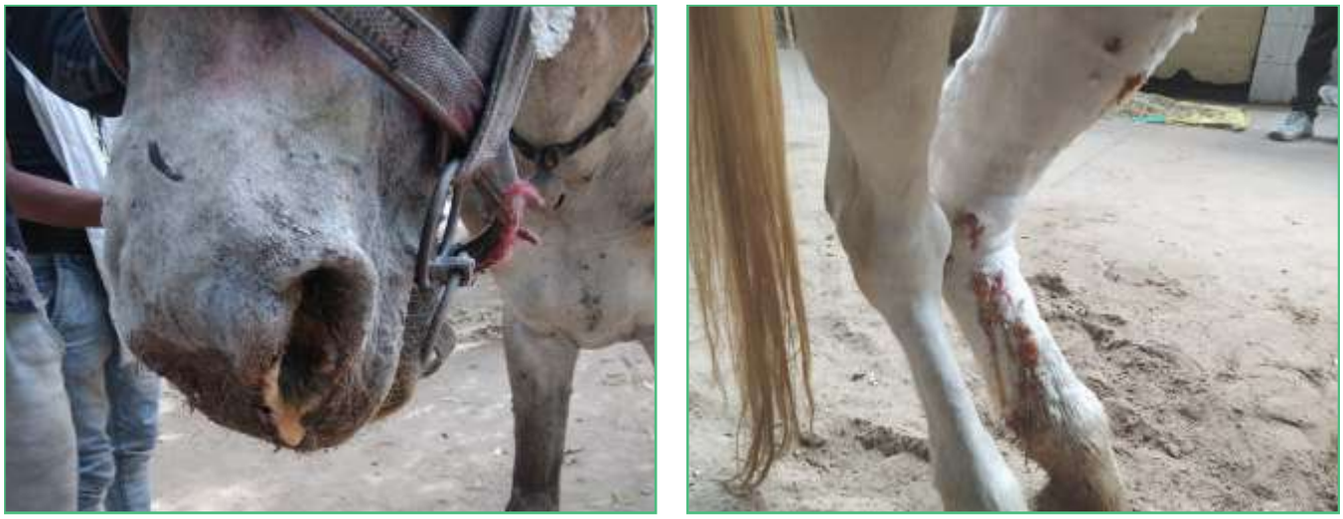


Fig.1 Representative photographs of glanders affected equines.

We aligned and compared 460 bp and 620 bp of 16S rRNA and ITS gene sequences for construction of phylogenetic tree (Fig 1 (a,b) & Fig 2 (a,b). Furthermore, corresponding gene sequences of global *B. mallei* isolates available in the GenBank were also included for the analysis. For 16S phylogeny, 36 sequences were added from the database thus 75 *B. mallei* sequences were used for tree construction. *B. mallei* originated from Iran, Bahrain, Hungary and the USA (2002734299, KC_1092, Bahrain1, FMH and JHU) were grouped in the first branch. The second branch contains three older Indian isolates (2000031066, BMQ and NCTC3709) and four new Indian Isolates (3701, 4629, 4844 and 3880). The third branch is the largest branch and it accommodates most of the recent Indian (n=33) and global (n=31) *B. mallei* isolates (Fig. 2a). For ITS phylogeny, 18 sequences were added from the database thus 57 *B. mallei* ITS sequences were used for tree construction. A similar branching pattern was observed in ITS phylogenic tree. The first branch consists of six isolates such as 2002734299, strain 11, KC_1092, Bahrain1, FMH and JHU. The second branch contains twelve isolates, which include three older Indian isolates (SAVP1, BMQ and India 86-567-2) and nine global isolates. The third branch consists of all 39 recent Indian *B. mallei* isolates (Fig. 2b).

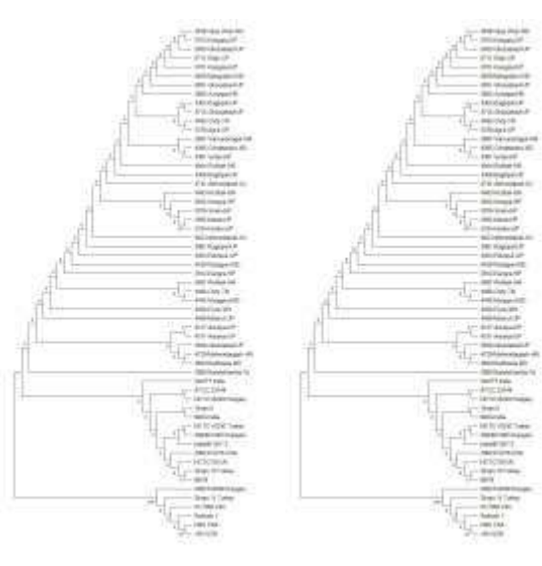


Fig 1(a) & (b) : Phylogeny for ITS genes

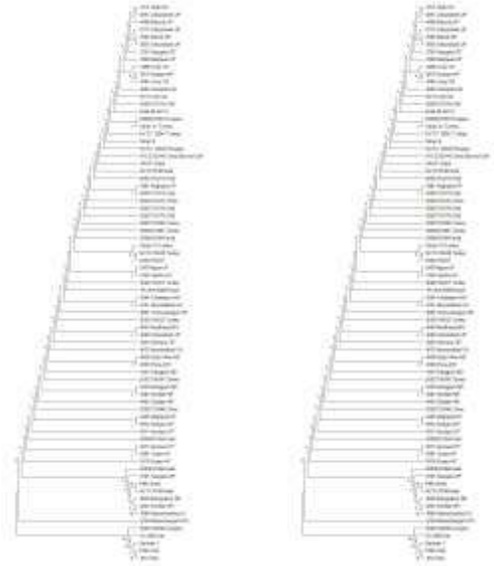


Fig 2(a) & (b) : Phylogeny for ITS genes

Phylogeny showed a similar branching pattern and revealed that recent isolates circulating in India are closely related to each other but genetically diverse from older *B. mallei* isolates that were reported from India or elsewhere. We have observed that *B. mallei* originating from Uttar Pradesh was closely related to isolates obtained from Himachal Pradesh (Ghaziabad/Mandi- 3595/3855, Baghpat/Kangra- 4365/3932, Auraiya/Solan-4517/3081), Maharashtra (Ghaziabad/Buldhana- 4508/4600) and New Delhi (Kasganj/Mongalpuri-3701/4629, Kasganj/Vijay Vihar-3703/4638). The present findings and surveillance data suggests that Uttar Pradesh is the most glanders prone area and equine movement from this place led to rapid spread of glanders to other states.

(Harisankar Singha, Shanmugasundaram K. and B. N. Tripathi)

Comparative pathological evaluation of deletion mutants of EHV-1

Four deletion mutants of EHV-1 developed earlier and coded Δg^{A}, Δg^{B}, $\Delta g^{A,B}$ and $\Delta g^{A, B, X,Y}$ were studied in BALB/c mouse model for assessing their pathology and attenuation as compared to the wild parent virus EHV-1- Tohana. For this BALB/c mice (n=252) were inoculated with respective viruses @ 10^{5}pfu/25ul and observed for clinical signs, body weight reduction, gross and histopathology, immune-histochemistry and immune responses for 56 days. This was done for selection of most appropriate modified live attenuated virus for challenge studies and as a potential candidate for developing live EHV-1 vaccine.

The maximum percentage of body weight reduction observed in virus followed by Δg^{B}, Δg^{A}, $\Delta g^{A, B}$ and $\Delta g^{A, B, X, Y}$ which were 5%, 4%, 3.5%, 2% and 2%, respectively. The clinical signs appeared from 1 dpi onwards in all experimental groups and the signs resolved by 6 dpi in wild virus and gB whereas, in ΔgA, clinical signs lasted up to 4 dpi. On the other hand, gA and gA, B, X, Y group mice showed clinical signs up to 2 to 3 dpi only.

In wild EHV-1 infected mice the lesions were mainly characterized by congestion and grey hepatization at 3 and 5 dpi respectively. The gross lesions were significantly less in Δg^{A} and $\Delta g^{A, B, X, Y}$ group mice than rest other groups at 1 dpi to 5 dpi. The histological lesions in wild ToH-BAC were characterized by diffuse moderate congestion of the pulmonary vasculature with moderate interstitial infiltration, diffuse severe thickening of the interalveolar septae and severe necrosis of the bronchiolar epithelium. Though, the gene deleted mutant recombinant viruses were showing similar type of lesions, the severity of the lesions was reduced at various time intervals. At 3 dpi lesions peaked in all the groups and EHV-1 immunopositivity in bronchiolar epithelium and macrophages was observed in the lungs of mice. At 7 dpi, mild diffuse infiltration in lungs was observed in ToH-BAC and ToH-BAC/IR6 groups whereas no lesions were appreciated in ΔgE deleted mutants. The overall lung lesion scores were significantly high in wild virus group followed by ΔgB at 1 dpi to 7 dpi. Among all groups, the overall lung lesions were significantly lower in $\Delta g^{A,B}$ and $\Delta g^{A, B, X, Y}$ groups. Shedding of EHV-1 in nasal washings and residual virus in lungs as estimated by TaqMan qPCR and immunohistochemistry staining in mice infected with the mutant viruses showed a similar pattern.

The immune responses were assessed through flow cytometry analysis of various cells. The antibody response was detected at 7 dpi followed by a peak at 14 dpi and there was no significant difference between groups. The higher CD8+ T-cell and CD4+ TH1 cell populations observed in four deletion mutant groups followed by double deletion mutant.

Based on the substantial attenuation of virus, broad cell tropism, CD8+ and Th1 mediated immune responses, the deletion mutants of EHV-1 *viz.* $\Delta g^{A, B}$ and $\Delta g^{A, B, X, Y}$ qualified to be good modified live vaccine candidates and were studied further in murine model challenge studies.

Challenge studies in murine model to assess the potential of mutant EHV-1 as vaccine candidates

On the basis of comparative pathology studies EHV-1 virus constructs code named ΔgA, B and Δg A, B, X, Y were selected to assess their pathogenicity and protective efficacy in murine model.

In vitro studies after GFP removal: Plaque size and growth kinetics of EHV-1 deletion mutants when compared to wild type vTOH revealed a significant reduction in the plaque size area. The reduction was greater in group I (85.86%) than group II (76.54%). Moreover, the mutants showed similar time course of replication when compared to the wild type regardless of low cell infectivity. Despite forming smaller plaques, mutants in RK-13 cells replicated to titers similar to that of the parental virus.

In vivo studies: For the *in vivo* studies, firstly a pilot trial was conducted for dose determination of gene deleted EHV1 mutants *vis à vis* wild type vRaj so as to optimize the best dose of inoculum which exhibits maximum immunological responses with minimal pathological lesions. From the pilot study, it was concluded that in EHV-1 mutants, 10^{7} PFU dose group generated maximum immune response while the virulence as assessed through reduction in clinical signs, body weight loss and gross and histopathological lesions was less as compared to the wild type virus.

For challenge studies BALB/c mice were divided into 5 groups. Mice from group I, II and III were respectively immunized intranasally with 20 µl inoculum (10^{7} PFU) of four deletion, two deletion and wild virus under mild anaesthesia followed

by a booster at 14 days. Group IV and V mice were mock immunized with 20 µl of sterile PBS. On 35th day, mice from group I to IV were challenged intranasally with 20 µl of 1.5×10^7 PFU of EHV-1 strain vRaj, while group V animals were mock challenged with PBS. Following challenge, blood collection and necropsy were done on 38, 40, 44 and 49 dpc.

Mice immunized with the mutant viruses and further challenged with pathogenic wild virus showed less pathology in terms of clinical signs, body weight loss, gross and histopathological lesions. Following challenge, considerable amount of nasal shedding of virus as estimated through qPCR was seen in all groups. On 3 and 5 dpc, group IV showed significantly higher nasal shedding than the mutant groups and by 5 dpc, the viral load in the immunized groups had decreased significantly as compared to the positive control animals. The EHV-1 copy numbers from the lung tissue showed similar trend.

The SNT and ELISA results showed good immune response with attenuated viruses and the SNT titres hovered around 128-256 on 35 days post immunization. The isotype ELISA showed IgG2b as the most prevalent type pre challenge, however, post challenge IgG1 was most prevalent followed by IgG2a and IgG2b.

Flow-cytometric immunophenotyping revealed Th1 dominated immune response in all groups at all-time points, highest being witnessed in four deletion mutant EHV-1. The T cell response of this group increased from 14 dpi (52.52±3.22) while attaining its peak on 35 dpi (63.56±6.45). This is contrary to what was observed in mice infected with wild virus (30.83±3.16 on 14 dpi & 39.27±3.49 on 35 dpi).

In nutshell, the EHV-1 virus constructs developed by our group are providing adequate immune responses and protective immunity in murine model and are prospective candidates for large animal studies.

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

Antimcrobial efficacy of ZnO based nanoformulations against *Streptococcus equi*

Antibiotic resistance is emerging at fast pace against various bacteria including *Streptococcus equi*, the causative agent of strangles, a highly contagious disease of economic importance in horses, mules and donkeys. The disease is characterized by pyrexia, anorexia, frequent nasal discharge, sub mandibular lymphadenopathy and abscesses of lymph nodes. Equines have started to show resistance towards the antibiotics. Novel approaches are necessary to develop the next generation drugs in order to get the bacterial infections under control. Polymeric ZnO NPs have also been suggested for antibacterial activity (Fig. 1, 2) and reduced toxicity against infectious diseases. In order to find an alternative to the antibiotics and counter the antibiotic resistance, we determined the efficacy of our novel ZnO nanoparticles against *S. equi.* Zinc oxide nanoparticles prepared by conventional hydrothermal method, microwave irradiation, AGZnO NPs (Polymeric ZnO NPs-1), CsZnO NPs (Polymeric ZnO NPs-2) were found to be highly effective against *S. equi at* various concentrations as determined by zone of inhibition and microbial inhibitory concentrations tests.

(Prashant, Dharvi, Balvinder Kumar and Anju Manuja)

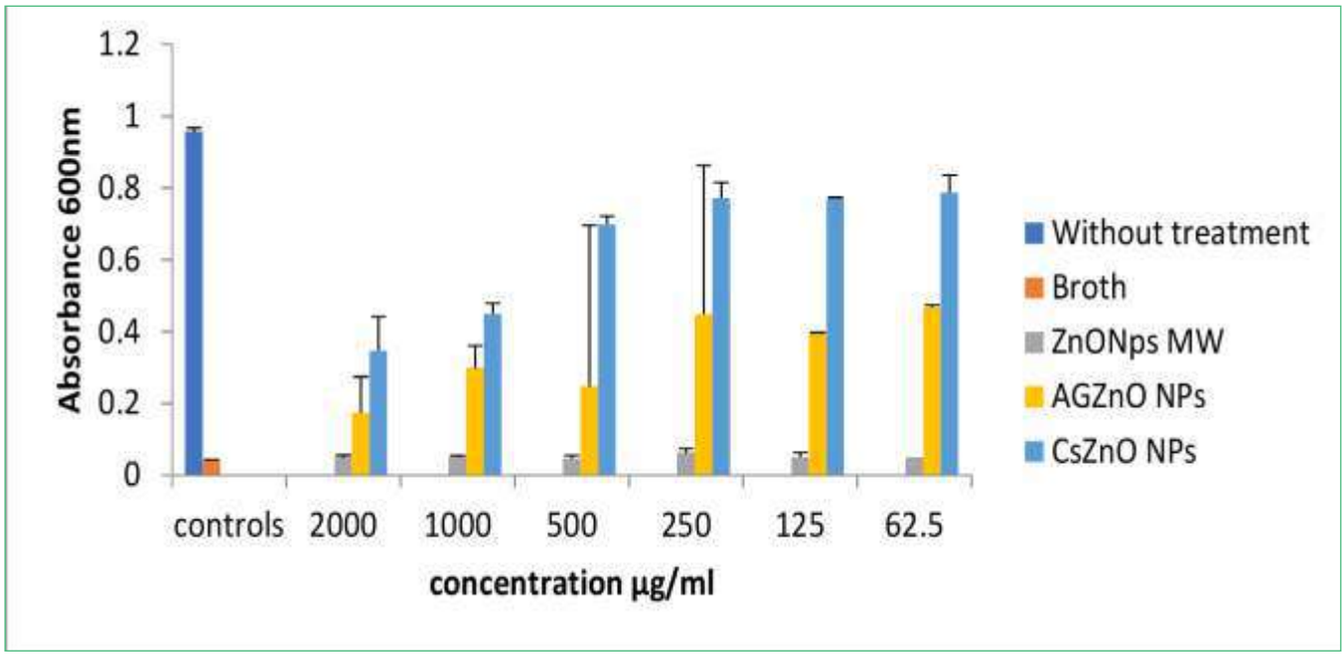


Fig. 1: Concentration dependent antibacterial activity of ZnONPs, AGZnO NPs and CsZnO NPs and controls against *S. equi.* Controls include culture without treatment and only broth (no treatment and no culture)

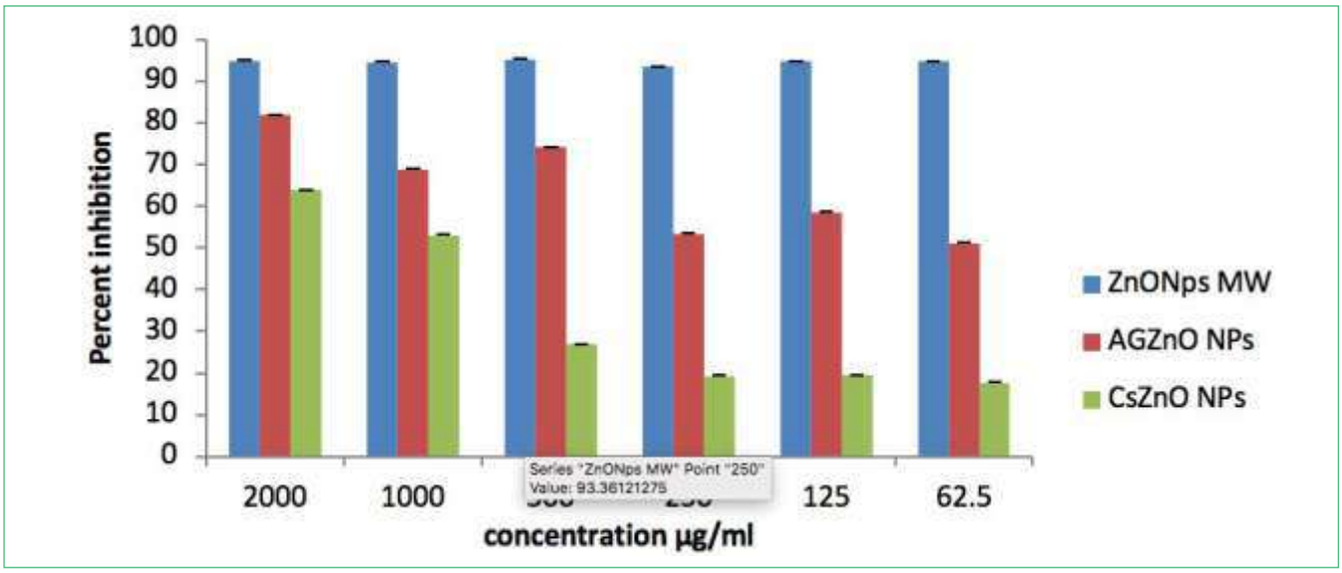


Fig. 2: Concentration dependent percent inhibition of antibacterial activity of ZnONPs, AGZnO NPs and CsZnO NPs against *S. equi*

Efficacy of zinc oxide based polymeric nanoformulations in collagen induced arthritis

Nanoformulation aided delivery using biodegradable scaffolds mimicking extracellular matrix are the novel substitute for tissue repair with minimal side effects and accelerated healing. We have developed novel and safe zinc based polymeric nanoformulations having antibacterial/ anti-inflammatory/re-epithelization properties giving encouraging results in *in-vitro* studies and lab animal models. We developed collagen induced arthritis in DBA-1 mice (susceptible strain for development of osteoarthritis) and Collagen/LPS induced inflammatory arthritis in albino mice and rats. Arthritic changes were confirmed by X-rays, biochemical tests, and markers of cartilage degradation (CTXII, hydroxyproline etc) with individual variations. The structural changes were only observed in DBA-1 mice. However, gross changes, markers of arthritis and histopathological changes confirmed the arthritis in resistant rodent strains (Fig. 1a, 1b, 2a, 2b) We also evaluated the efficacy of developed nanoformulations (AGZnO NPs/ CsZnO NPs). Remarkable reduction in inflammation, swelling of the inflamed digits/feet was observed in treated and arthritic DBA-1 mice. The gross changes in the affected feet/ digits demonstrated the remarkable efficacy of the nanoformulation. Serum creatinine levels were found higher in untreated arthritic DBA-1 mice as compared to treated DBA-1 arthritic mice. The markers for cartilage degradation Cross Linked C-Telopeptide of Type II Collagen, hydroxyproline, proinflammatory cytokines determined in treated and untreated DBA-1 experimental arthritic mice revealed the efficacy of the nanoformulations.

Fig. 1(a) Control rat

Fig. 1(b) Rat after induction of arthritis (collagen +LPS)

Phytochemical profile, cytotoxicity and anti-inflammatory potential of the leaf extracts from *Lawsonia inermis* of Indian origin

Lawsonia inermis Linn popularly known as the Henna has played an important role in Ayurvedic or natural herbal medicines. The presence of phyto-constituents in henna, which may affect adversely the animal or human health,

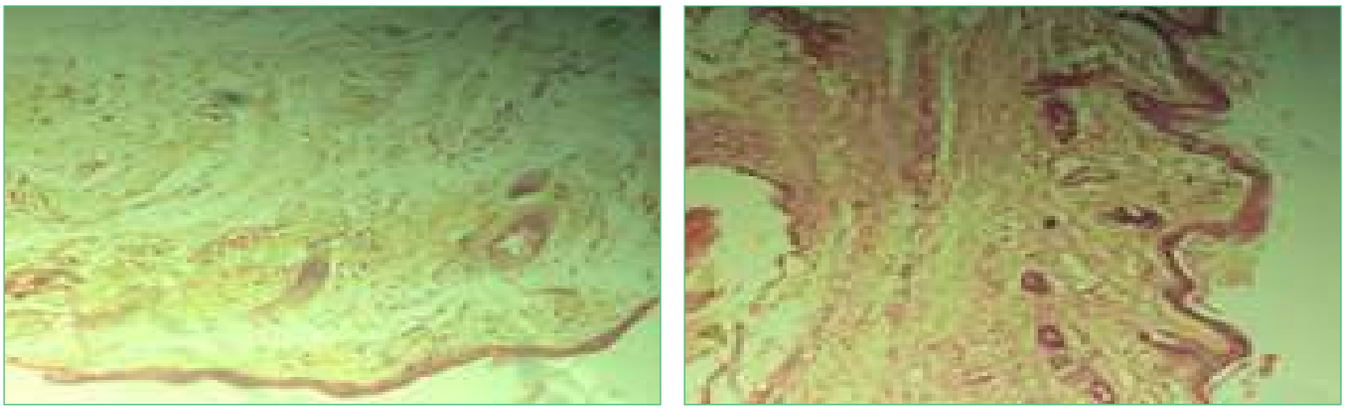


Fig. 2: Histopathological changes in control and arthritic mice. The cartilage is smooth, matrix, chondrocytes are organized into superficial, mid and deep zones in control animals. The surface discontinuity extends through the mid zone in affected animal.

needs to be elucidated for *L. inermis Linn* species especially which is grown in India. We assessed the phytochemical profile for presence of phyto-constituents (alkaloids, carbohydrates, glycosides, steroids, flavonoids, saponins, tannins, proteins/amino acids and gums/mucilage) from various extracts of the plant leaves' and found alkaloids, steroids, flavonoids, saponins, tannins, proteins/amino acids in different extracts.

The extracts were further purified by column chromatography for the isolation of plant constituents and monitored by thin layer chromatography (TLC), analysed by FTIR, HNMR, and GC-MS analysis. All the spectral results (IR, NMR, GC-MS) suggest that the compounds from the extract contain aromatic nucleus and OH group along with methoxy group, allyl as well as vinyl group. Fraxetin 1(3H)-isobenzofuranone structures were confirmed in the fractions of $CHCl_3$ (70%)/ MeOH (30%) extract as observed as the potent constituents. We also assessed the anti-inflammatory activity by Nitric oxide production in various leave's extracts as determined by Griess assay. Overall the highest nitric oxide production by $CHCl_3$ (70%)/ MeOH (30%) was observed amongst various fractions at different concentrations. Cytotoxicity studies revealed some of the leaf extracts have potential cytotoxic activity on vero cells. Reducing the chloroform concentration during extraction decreases the cytotoxic effect on the cells. Although most of the components are biocompatible, however, the presence of cytotoxic compounds in some of these extracts warrants research for fabrication of suitable formulations comprising these constituents to reduce its dose/toxicity for the use of beneficial effects of the plant components.

(Anju Manuja and Balvinder Kumar)

Fig. 1(A): Suppression of malenoma on application of extract

Antiproliferative activity of leaf extract of *Aerva javanica* against malenoma and warts in horses

Many types of tumors such as sarcoids, melanoma, carcinoma and warts are observed in animals including horses. In our earlier study, leaf extract of *Aerva javanica* showed anti proliferative activity against proud flesh and sarcoids. This time this extract was used against melanoma and warts in horses. Melanoma is a cancer of pigment cells of skin. Usually melanoma in horses is benign but cases of malignant melanoma are also reported in horses. Melanoma occurs as black lumps near hairless area usually on tail and around anus. Warts or papillomatosis are the most common tumor of horses

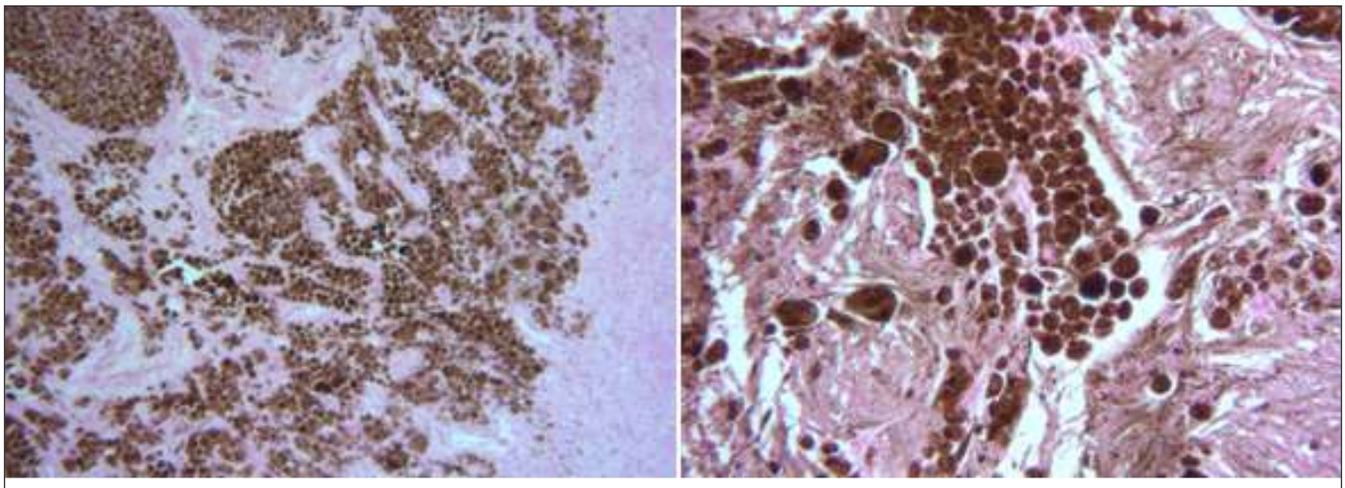


Fig. 1(B): Malignant melanoma with many variable sized melanocytes, few of them are mitotic cell in dermis, necrosed and degenerating cell in epidermis.

Fig. 2: Suppression of warts on topical application of the extract from figure A to D.

caused by papilloma virus. At present no satisfactory treatment is available for warts in horses, however many of the cases show self recovery over the period in horses but malignant form may necessitate culling in some species.

A fraction of methanol soluble fraction of the aqueous extract of leaf of *Aerva javanica* was applied topically against two clinical cases of melanoma and two clinical cases of warts in horses. Biopsy samples were collected for histopathological analysis and differential diagnosis. All the cases of tumor resolved completely in 30 to 50 days of topical application of the extracts Fig. (1A, 1B and 2).

(R.K. Dedar, Naveen Kumar, Shirish D. Narnaware, R.A. Legha and Yash Pal)

Management of seasonal dermatitis in horses by using leaf extract of *Aerva javanica*

Occurrence of seasonal allergic dermatitis, especially insect bite hypersensitivity and atopic dermatitis are very common in horses. At present there is no satisfactory treatment available for management of these skin allergies. Plant flavonoids such as quercetin, kaempferol and their glycoside derivatives have been reported for their anti allergic and anti-inflammatory properties. Clinical signs of the allergic dermatitis in horses are alopecia, thickening of skin and itching. On histopathological examination of skin biopsy samples taken from the clinical cases epidermal hyperplasia, orthokeratotic and parakeratotic hyperkeratosis, spongiosis, occasional trichomalacia, multifocal areas of aggregation of lymphocytes with or without infiltration of eosinophils were observed. Mass spectrometer analysis of more purified extract suggested a glycoside of kaempferol may be an active ingredient of the extract. A total of 11 clinical cases were treated successfully.

(R.K. Dedar, Naveen Kumar, S.D. Narnaware, Jitender Singh, R.A. Legha and Yash Pal)

Identification and evaluation of Naphthoquinone derivatives against *Trypanosoma evansi* infection

Trypanosoma evansi is an extracellular flagellate blood protozoan parasite and an etiological agent of animal trypanosomosis. Presently, only a few drugs are registered in India and have been used for the treatment of animal trypanosomosis, but these drugs show severe toxic effects and also have the problem of drug resistance. Naphthoquinones (NTQ) have been reported for their antitrypanosomal potential against other trypanosomes-*T. brucei* and *T. cruzi*. Six naphthoquinones (NTQ1-NTQ6) derivatives were selected and procured for evaluation by demonstrating their growth inhibitory effect against *T. evansi*. All NTQs significantly ($p<0.001$) exhibited activity

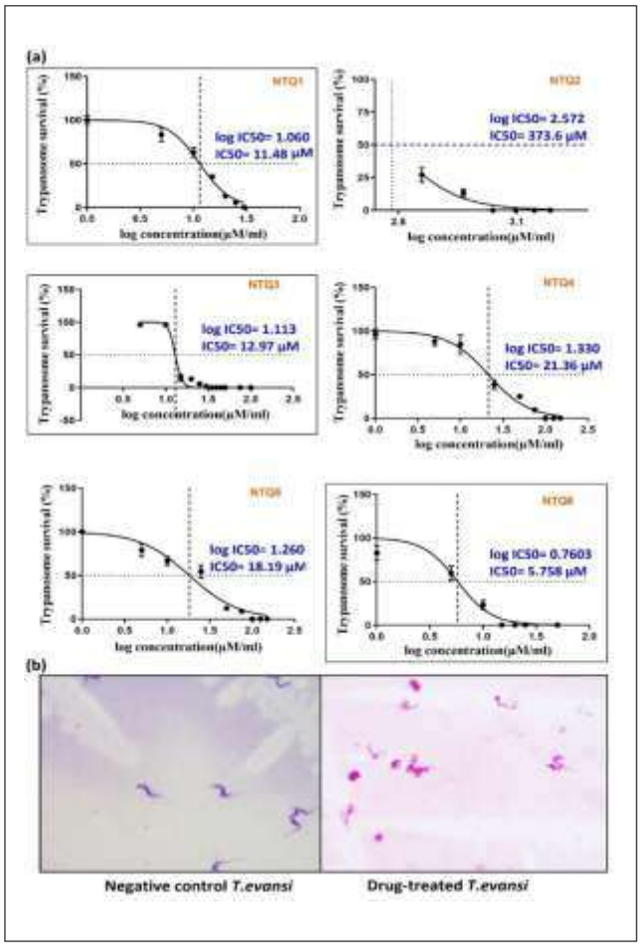


Fig.1 (a): Growth inhibition curves depict log IC_{50} value and IC_{50} of the naphthoquinone derivatives (NTQ1 to NTQ6) against *Trypanosoma evansi* in an axenic culture. (b) Depicts morphological changes in *T. evansi* after drug treatment.

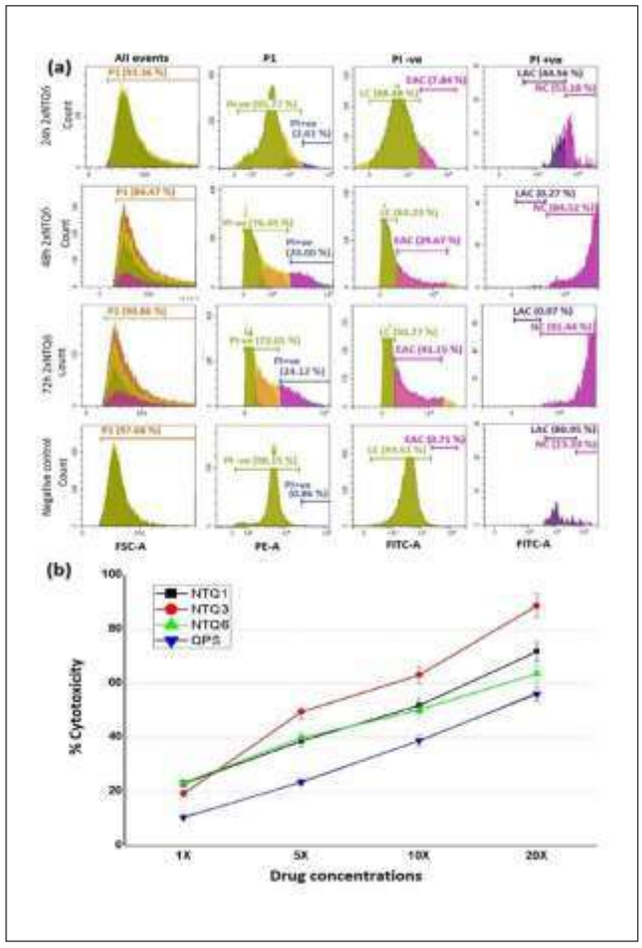


Fig. 2 (a): Flow cytometry apoptosis analysis of 2xNTQ6-treated *Trypanosoma evansi* using Annexin V/propidium iodide staining. LC indicates live cells, EAC indicates early apoptotic cells, LAC indicates late apoptotic cells and NC indicates necrotic cells. (b) *In-vitro* cytotoxicity analysis of different drug concentration on horse PBMCs.

against parasite growth and multiplication with IC_{50} values of 11.48 µM, 373.6 µM, 12.97 µM, 21.97 µM, 18.19 µM and 5.758 µM but NTQ1, NTQ3 and NTQ6 were selected based on their IC_{50} value for further studies (Fig. 1a). The dose-and time-dependent morphological effect (Fig. 1b) on parasite were evaluated including the measurement of reactive oxygen species (ROS) by spectro-fluorometery and measurement of apoptosis using Annexin V-FITC kit by flow cytometry. The selected NTQs exhibited a significant production of ROS and displayed a significant percent early apoptotic cell ($AV^+ PI^-$) and significant percent necrotic cell ($AV^+ PI^+$) in the axenic culture of *T. evansi* than quinapyramine methyl sulphate (QPS), as reference control (Fig. 2a). NTQs also showed more cytotoxic effect on horse peripheral blood mononuclear cells as compared to QPS (Fig. 2b). Therefore, we confirmed the antitrypanosomal activity and apoptotic-like mechanism of NTQs in an axenic culture of *T. evansi*. Further, the nanoformulation of these compounds may be explored to reduce the dose and cytotoxic effect.

(Ruma Rani and Rajender Kumar)

Genetic diversity of equid herpesvirus 1 isolated from abortigenic outbreaks in India

Equine herpesvirus 1 (EHV-1) causes abortion, neonatal death, respiratory and neurological disease in equines and is endemic in India. The factors that contribute to the EHV-1 disease severity are not clearly known. It has been previously reported that the N752D substitution in the viral DNA polymerase (ORF30) is associated with neurological disease (equine herpesvirus myelopathy).

To decipher the EHV-1 diversity amongst abortigenic isolates in India, we carried out the whole genomes sequencing of Indian EHV-1 isolates using NGS under Illumina platform. EHV-1 isolates (n=7) isolated over a period of 15 years included in the study were low passage isolates from abortigenic outbreaks in northern India. On analyzing the 63

Fig. 1: EHV-1 WGS tree

whole genome sequences of EHV-1, Hasegawa, Kishino and Yano (HKY) + G model was found to be the best

substitution model. EHV-1 isolates were analyzed using Jmodellest2 to find out the Randomized Axelerated Maximum Likelihood (RAxML) to construct the phylogenetic tree.

Phylogenetic analysis of regions spanning nearly 90% of the genome showed that up to 3 distinct viral clades have been circulating in India (Fig. 1). All the Indian isolates clustered away from the UK isolates with reliable bootstrap values except for the Essex isolate. Among the Indian isolates, all the Hisar isolates clustered to a sub-node, away from the H14 and Meerut isolates. Delhi isolate was found to be the farthest from the merit isolates. One of the abortigenic isolates had the N752D substitution, whereas all other abortion isolates were non-neuropathic. The bioinformatic analysis suggested that diverse strains of EHV-1 are circulating in India and causing abortions in pregnant mares.

(B.R. Gulati, T. Riyesh, N. Kumar and R.K. Gandham)

Detection of Equine gammaherpesvirus 2 and 5 in nasal swabs of asymptomatic horses in India

Equine gammaherpesvirus 2 (EHV-2) and 5 (EHV-5) are members of the family *Herpesviridae*, subfamily *Gammaherpesvirinae* and genus *Percavirus*. Infections due to EHV are associated with respiratory diseases, pharyngitis, enlarged lymph nodes, nasal discharge, coughing, fever, lack of appetite and poor performance. EHV-2 is also associated with keratoconjunctivitis and EHV-5 with equine multi-nodular pulmonary fibrosis syndrome. Infection by EHV-2 and 5 occur in young foals with periodic reactivation of the latent virus during the life of the animal. Although the sites of latency are not clearly identified, B lymphocytes are considered to be the major site of latency. EHV-2 and EHV-5 are recovered typically from nasal swabs, respiratory fluids and peripheral blood lymphocytes.

Although -herpesvirus infections (EHV-1& 4) are endemic in horses, herpesvirus infections (EHV-2 & 5) have not been reported from India. Taqman probe-based real-time PCR assays were standardized using primers and probes designed to detect the glycoprotein B gene of EHV-2 & 5. These assays were used to detect the occurrence of EHV-2 and EHV-5 in the upper respiratory tract of asymptomatic young horses.

Nasal swabs from 88 apparently healthy young Thoroughbred horses of either sex (between age 3 months and 2 years) were collected in viral transport medium. On testing DNA samples isolated from swabs, 59 (67.04%) were positive for EHV-2, 42 (47.72%) for EHV-5 and 38 (43.18%) were positive for both EHV-2 and EHV-5 (Table 1).

(B.R. Gulati, T. Riyesh, N. Kumar and G. Bishnoi)

Table 1. Detection of EHV-2 and EHV-5

Parameter		EHV-2 Results		
		Positive	Negative	Total
EHV-5 Results	Positive	38	4	42
	Negative	21	25	46
	Total	59	29	88

Novel genomic constellations of Indian isolates of *Streptococcus equi*

Multilocus sequence typing (MLST) is molecular technique devised to study the molecular epidemiology and genetic structure of microorganisms. *Streptococcus equi* is the causative agent of strangles. The disease is economically important and the most contagious infectious disease of horses, mules and donkeys in India. The organism has been isolated from many diseased animals but not characterized at molecular level to ascertain the diversity of microbial population in India. The MLST scheme for the β-hemolytic, Lancefield group C streptococcal bacteria *Streptococcus equi and S. zooepidemicus* has been developed based on sequencing of seven highly conserved housekeeping genes at the Animal Health Trust. UK. This study analysed the sequences of these seven genes from forty one isolates of the bacteria isolated from field cases. The genomic constellations suggest the presence of novel isolates of *S. equi* in India. Allele numbers have been assigned to each unique gene sequence and analyzed to determine sequence types (ST) for each isolate by combination of alleles (Table 1).

Table 1: Allele numbers of seven conserved genes and sequence types of Indian strains of *Streptococcus equi*

Isolate	Organism	arcC	nrdE	proS	spi	tdk	tpi	yqiL	ST
4058	*S. equi*	45	45	45	45	45	45	45	179
4057	*S. equi*	45	47	45	45	45	45	45	283
4399	*S. equi*	45	45	45	45	45	45	45	179
4052	*S. equi*	45	47	45	45	45	45	45	283
4401	*S. equi*	45	47	45	45	45	45	45	283
4403	*S. equi*	45	47	45	45	45	45	45	283
4055	*S. equi*	45	47	45	45	45	45	45	283
2508	*S. equi*	45	47	45	45	45	45	45	283
2759	*S. equi*	45	47	45	45	45	45	45	283
2757	*S. equi*	45	47	45	45	45	45	45	283
3617	*S. equi*	45	45	45	45	45	45	45	179
3615	*S. equi*	45	45	45	45	45	45	45	179
3619	*S. equi*	39	47	45	45	45	45	45	New
2760	*S. equi*	45	47	45	45	45	45	45	283
2756	*S. equi*	45	45	45	45	45	45	12	New
869	*S. equi*	3	3	14	3	36	3	64	New
871	*S. equi*	33	33	40	37	28	1	47	211
873	*S. equi*	13	42	14	21	36	13	41	New
874	*S. equi*	12	42	10	67	44	46	12	New
881	*S. equi*	34	11	45	64	4	14	30	New
905	*S. equi*	45	45	45	45	23	45	45	151
294	*S. equi*	45	47	46	45	45	45	45	New
1245	*S. equi*	2	36	14	42	36	4	2	New
300	*S. equi*	45	47	45	45	45	45	45	283
2576	*S. equi*	45	45	45	45	45	45	45	283
2577	*S. equi*	45	47	45	45	45	45	45	283
2697	*S. equi*	45	47	45	45	45	45	45	283
2839	*S. equi*	45	47	45	45	45	45	45	283
2842	*S. equi*	45	47	45	45	45	45	45	283
2845	*S. equi*	45	45	45	45	45	45	45	179
3565	*S. equi*	45	47	45	45	45	45	45	283
3616	*S. equi*	45	47	45	45	45	45	45	283

3652	*S. equi*	45	47	45	45	45	45	45	283
3658	*S. equi*	45	47	45	45	45	45	45	283
4045	*S. equi*	45	47	45	45	45	45	45	283
4053	*S. equi*	45	47	45	45	45	45	45	283
3566	*S. equi*	6	45	45	45	45	45	45	New
4047	*S. equi*	45	47	45	45	45	45	45	283
4049	*S. equi*	45	47	45	45	45	45	45	283
4051	*S. equi*	45	47	45	45	45	45	45	283
4054	*S. equi*	45	45	45	45	45	45	45	179

Out of the total 41 *S. equi* isolates 6 isolates exactly matched ST 179, 24 isolates to ST 283 and one each to 211 and 151 sequence types already documented in BIGSdb. Nine isolates had novel genomic constellations that have not been reported from any part of the world. MLST analysis also revealed that the genomic combination of two isolates of *S. zooepidemicus* and *S. equisimilis* were also novel and not reported earlier. The nine genetically novel isolates of *S. equi* circulating in India matched partially to ST 3, 17, 96, 179, 180, 245, 250, 283, 328, 358, 362, 420, 423, 425, 457, 461, 487, 509 and 510 isolated from different parts of the World. The two isolates of *S. zooepidemicus* also had novel genomic combination of the conserved gene alleles and partially matched to 17, 69, 151, 179, 281, 282, 283, 325, 358, 379, 395, 396, 402, 422, 423 and 516 sequence types at different number of loci in *S. zooepidemicus* genomic databases.

(Balvinder Kumar, Dharvi Chhabra, R.K. Vaid, Anju Manuja, K. Shanmugasundram and H.S. Singha)

National Centre for Veterinary Type Cultures (NCVTC)

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. During the year 2020, a total of 100 different biological samples *viz*., tissues, swabs and blood were collected / received from Hisar, Ranchi, Sirsa and Jaipur. The samples were processed for isolation of different viruses. The details of virus authentications/isolations and acccessions (27 nos.) are as follows (Table 1).

Authentication of isolates /samples

a. Isolation and authentication of Fowl adenovirus: A total of 10 fowl adenoviruses (FAV) were isolated, authenticated by amplification of DNA polymerase (580 bp) and hexon (900 bp) gene. The isolated viruses were accessioned in the repository upon confirmation of viability in CEL cells.

b. Isolation and authentication of Infectious bronchitis virus: Four Infectious bronchitis virus isolates were also authenticated from poultry intestinal tissues by amplification of spike gene (370 bp). The viability of the virus isolates was confirmed in 10 days old SPF eggs by allantoic route of inoculation.

c. Authentication of Bluetongue virus: One BTV isolates received from CIRB, Hisar was authenticated by amplification

Table 1: Acquisition / Receipt of viral isolates during 2020

Depositor	Virus isolates	Number
NCVTC, Hisar	ANV	4
	LSDV	2
	IBV	4
	SARS-CoV2	2
	FAdV	10
TANUVAS	FAdV	1
VRCVV	Bluetongue virus	3
CIRB, Hisar	Bluetongue virus	1
	Total	**27**

of *NS1* gene (273 bp) and the confirmation of viability in BHK21 cell line is underway.

d. Authentication & characterization of Jaagsiekte sheep retrovirus (JSRV): An outbreak was investigated in Jaipur (India) wherein Sheep were infected. The biological samples (swab) were collected and processed in the lab. The etiological agent was identified as Jaagsiekte sheep retrovirus which has been reported for the first time in India. The samples were subjected to PCR for confirmation by targeting the Protease (313 bp) gene. Upon confirmation the virus sample has been sent for genome sequencing.

e. Isolation and accessioning of SARSCoV-2: Two SARSCoV-2 positive human clinical samples were passaged in Vero cells and their viability was confirmed and accessioned in the repository.

Preservation & revival of preserved viral isolates

The NCVTC repository is currently maintaining more than 300 virus isolates, the isolates are preserved by freeze drying as well as their storage in -80^{0}C deep freezers. During the year, 22 accessioned viruses were preserved through bulk production. The isolates included 10 FAV, 4 ANV, 4 IBV, 2 LSDV and 2 SARS-CoV2. Furthermore, 34 previously preserved viruses including buffalopox virus (5), BTV (6), IBDV (4), NDV (8), CSFV (3), SWPV (2), DPV (3), FPV (2) and Pigeonpox virus were revived and checked for their viability. The viruses were found viable.

First successful isolation of LSDV in India

Lumpy skin disease (LSD) has devastating economic impact. During the last decade, LSD had spread to climatically new and previously disease-free countries, which also includes its recent emergence in the Indian subcontinent (2019). We confirmed the LSD outbreak from cattle in Ranchi (India). Virus was isolated from the scabs (skin lesions) in the primary goat kidney cells. Phylogenetic analysis based on nucleotide sequencing of LSD virus (LSDV) ORF011, ORF012 and ORF036 suggested that the isolated virus (LSDV/Bostaurus-tc/India/2019/Ranchi) is closely related to Kenyan LSDV strains. Further, we adapted the isolated virus in Vero cells. We have described the first successful isolation of LSDV in India, besides providing insights into the life cycle of Vero cell-adapted LSDV.

Host pathogen interaction studies: Virus is an obligatory intracellular parasite which relies on the host machinery upon infection. The virology laboratory at NCVTC has reported some of the important host factors which are critically required for virus replication.

Antiviral activity of Apigenin against buffalopox: Novel mechanistic insights and drug-resistance considerations

We describe herein that Apigenin, which is a dietary flavonoid, exerts a strong *in vitro* and *in ovo* antiviral efficacy against buffalopox virus (BPXV). Apigenin treatment was shown to inhibit synthesis of viral DNA, mRNA and proteins, without affecting other steps of viral life cycle such as attachment, entry and budding. Although the major mode of antiviral action of Apigenin was shown to be mediated via targeting certain cellular factors, a modest inhibitory effect of Apigenin was also observed directly on viral polymerase. We also evaluated the selection of drug-resistant virus variants under long-term selection pressure of Apigenin. Wherein Apigenin-resistant mutants were not observed up to ~ P20 (passage 20), a significant resistance was observed to the antiviral action of Apigenin at ~ P30. However, a high degree resistance could not be observed even up to P60. To the best of our knowledge, this is the first report describing in *vitro* and *in ovo* antiviral efficacy of Apigenin against poxvirus infection. The study also provides mechanistic insights on the antiviral activity of Apigenin and selection of potential Apigenin-resistant mutants upon long-term culture.

(Sanjay Barua, Naveen Kumar and Riyesh T.)

Role of p38 MAP Kinase in buffalo pox virus replication

The study has been planned with an objective to elucidate the mechanisms underlying regulation of BPXV replication by p38 MAPK as well as to study the acquisition of potential antiviral drug resistance against SB239063. During the year, the cytotoxic concentration 50 (CC_{50}) of SB239063 (p38 MAPK) inhibitor was evaluated *in vero* cells. Furthermore the guide RNA's of the four isoforms of p38 MAP kinase were designed. Further work on conducting step specific assays so as to explore the molecular mechanism of p38 MAPK in BPXV replication is underway.

Emetine as an antiviral agent suppresses SARS-CoV-2 replication by inhibiting interaction of viral mRNA with eIF4E:

Emetine is a FDA-approved drug for the treatment of amebiasis. In the recent times we had also demonstrated the antiviral efficacy of emetine against some RNA and DNA viruses. Following emergence of the COVID-19, we further evaluated the *in vitro* antiviral activity of emetine against severe acute respiratory syndrome coronavirus 2 (*SARS-CoV-2*). The therapeutic index of emetine was determined to be 10910.4, at a cytotoxic concentration 50 (CC_{50}) of 1603.8 nM and effective concentration 50 (EC_{50}) of 0.147 nM. Besides, we also demonstrated the protective efficacy of emetine against lethal challenge with infectious bronchitis virus (IBV; a chicken coronavirus) in the embryonated chicken egg infection model. Emetine treatment was shown to decrease viral RNA and protein synthesis without affecting other steps of viral life cycle such as attachment, entry and budding. In a chromatin immunoprecipitation (CHIP) assay, emetine was shown to disrupt the binding of SARSCoV-2 RNA with eIF4E (eukaryotic translation initiation factor 4E, a cellular cap-binding protein required for initiation of protein translation) (Fig. 1).

Further, SARSCoV-2 was shown to exploit ERK/MNK1/eIF4E signalling pathway for its effective replication in the target cells. To conclude, emetine targets SARSCoV-2 protein synthesis which is mediated via inhibiting the interaction of SARS-CoV-2 RNA with eIF4E. This is a novel mechanistic insight on the antiviral efficacy of emetine. *In vitro* antiviral efficacy against SARSCoV-2 and its ability to protect chicken embryos against IBV suggests that emetine could be repurposed to treat COVID-19.

(Naveen Kumar and Riyesh T)

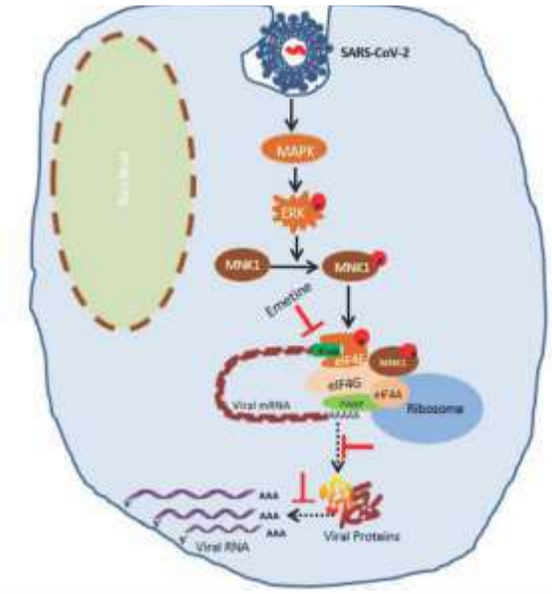


Fig. 1: Emetine blocks viral mRNA and eIF4E interaction in inhibiting SARS-CoV-2 replication. SARS-CoV-2 exploits MAPK/ERK/MNK1/eIF4E cell signalling pathway to effectively replicate in the target cells. Activated eIF4E binds with 5'-cap structure of viral mRNA to initiates translation of viral proteins. Emetine blocks SARS-CoV-2 protein synthesis by inhibiting interaction of viral mRNA with eIF4E.

Isolation and characterization of lumpy skin disease virus from cattle in India

Lumpy skin disease (LSD) has *devastating economic impact*. During the last decade, LSD had spread to climatically new and previously disease-free countries, which also includes its recent emergence in the Indian subcontinent (2019). This study deals with the LSD outbreak(s) from cattle in Ranchi (India). The most prominent clinical findings were skin nodules all over the body surface (Fig. 1) along with fever, oedema of legs, enlarged lymph nodes, lameness, anorexia and abortions with a morbidity rate of ~50% without any significant mortality. Virus was isolated from the scabs (skin lesions) in the primary goat kidney cells (Fig. 2). Phylogenetic analysis based on nucleotide sequencing of LSD virus (LSDV) ORF011, ORF012 and ORF036 suggested that the isolated virus (LSDV/*Bostaurus*-tc/India/2019/Ranchi) is closely related to Kenyan LSDV strains. Further, we adapted the isolated virus in Vero cells. Infection of the isolated LSDV to Vero cells did not produce cytopathic effect (CPE) until the 4th blind passage, but upon adaptation, it produced CPE (Fig 3) and high viral titres in the cultured cells. The kinetics of viral DNA synthesis and one-step growth curve analysis suggested that LSDV initiates synthesizing its genome at ~24 hours post-infection (hpi) with a peak level at ~96 hpi whereas evidence of progeny virus particles was observed at 36-48 hours (h) with a peak titre at ~120 h. To the best of our knowledge, this study describes the first successful isolation of LSDV in India, besides providing insights into the life cycle of LSDV.

Fig. 1: Clinical findings. (a) Nodules all over the body surface in cattle (b) Nodules are circumscribed, round, slightly raised, firm, painful and are 1-3 cm in size. (c) Ruptured nodules that created a deep-seated wound. (d). Wounds invaded by secondary bacterial infection leading to suppuration and sloughing. (e) Extensive lesions in the fetlock region extending up to the underlying subcuitis and muscle. (f) Necrosis of the skin nodule resulting in hard, raised areas "sit-fasts".

Generation of knockout cells using CRISPR/Cas9-mediated genome editing

We exploited CRISPR/Cas9-mediated genome editing to generate knockout (KO) HeLa cells with disrupted Phosphoribosyl formylglycinamidine Synthase (FPPS) gene. The small guide RNAs (sgRNAs) targeting FPPS genes were cloned into pL.CRISPR. EFS.GFP. Upon transfection into BHK21 cells, the recombinant constructs expressed respective target CRISPR RNA (crRNA), trans-activating CRISPR RNA (tracrRNA RNA) and Cas9, besides expressing green fluorescent protein (GFP). All the GFP expressing cells are likely to express sgRNA (crRNA+tracrRNA)/Cas9 complex that cut the genome at the targeted sites. The GFP expressing cells were sorted by fluorescence activated cell sorter (FACS) and cultured in 96-well tissue culture plate to obtain single clones by limiting dilution assay (Fig 1). After culturing for about a week, the wells with a single clone were selected microscopically for further propagation. To evaluate gene editing, all the single clones were subjected to amplify target genes by PCR. For further confirmation of the gene editing, the PCR products were subjected to nucleotide sequencing and Western blotting. Since PFAS is previously known to inhibit enetrovirus replication, we expected that PFAS knockout BHK21

Fig. 2

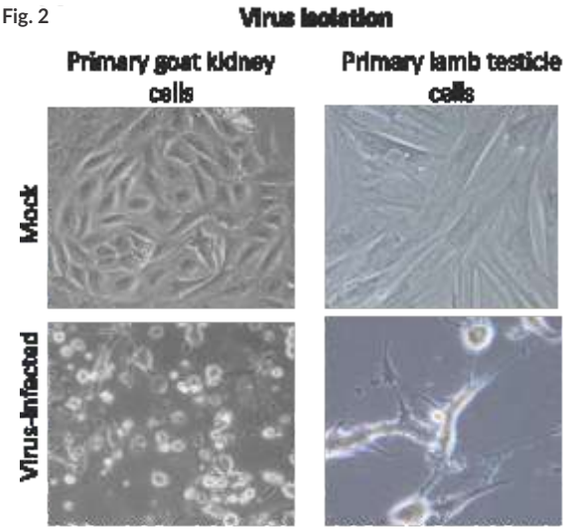
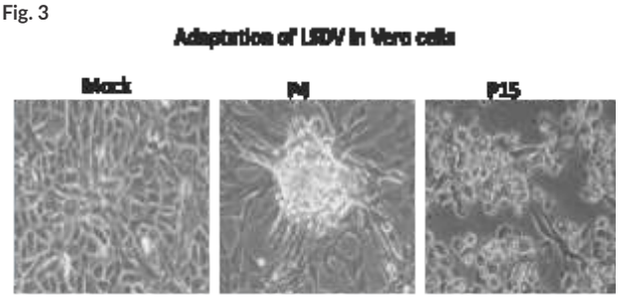


Fig. 3



cells would produce more FMDV. However, in contrary, when tested for FMDV sensitivity, PFAS knockout BHK21 cells were found to produce about 50-fold fewer virus particles then wild type BHK21 cells suggesting a proviral role of PFAS in FMDV life cycle and that PFAS plays an antagonistic role in the life cycle of different *Picornaviridae* family members. PFAS may therefore serve a potent host target for anti-FMDV drug development.

(Naveen Kumar, Sanjay Barua, Riyesh T. and B. Kumar)

Investigating mechanisms underlying acquisition antiviral drug resistance against host-targeting agents

Due to high mutation rates, viruses quickly become resistant at the druggable targets and pre-existing immunity. The rise in incidence of drug resistance has prompted a shift towards development of antiviral drugs. Host factors that are dispensable for host but are critical to virus replication may serve as alternate targets for development of novel antiviral therapeutics. We screened a library of small molecule chemical inhibitors and identified antiviral efficacy of sarco/endoplasmic reticulum calcium-ATPase (SERCA) inhibitor (Thapsigargin) and MNK1 (MAPK-interacting kinase) inhibitor (CGP57380) against paramyxoviruses [(Newcastle disease virus (NDV), peste des petits ruminants virus (PPRV)] and buffalopox virus (BPXV), respectively. Whereas directly acting agents are known to develop a complete

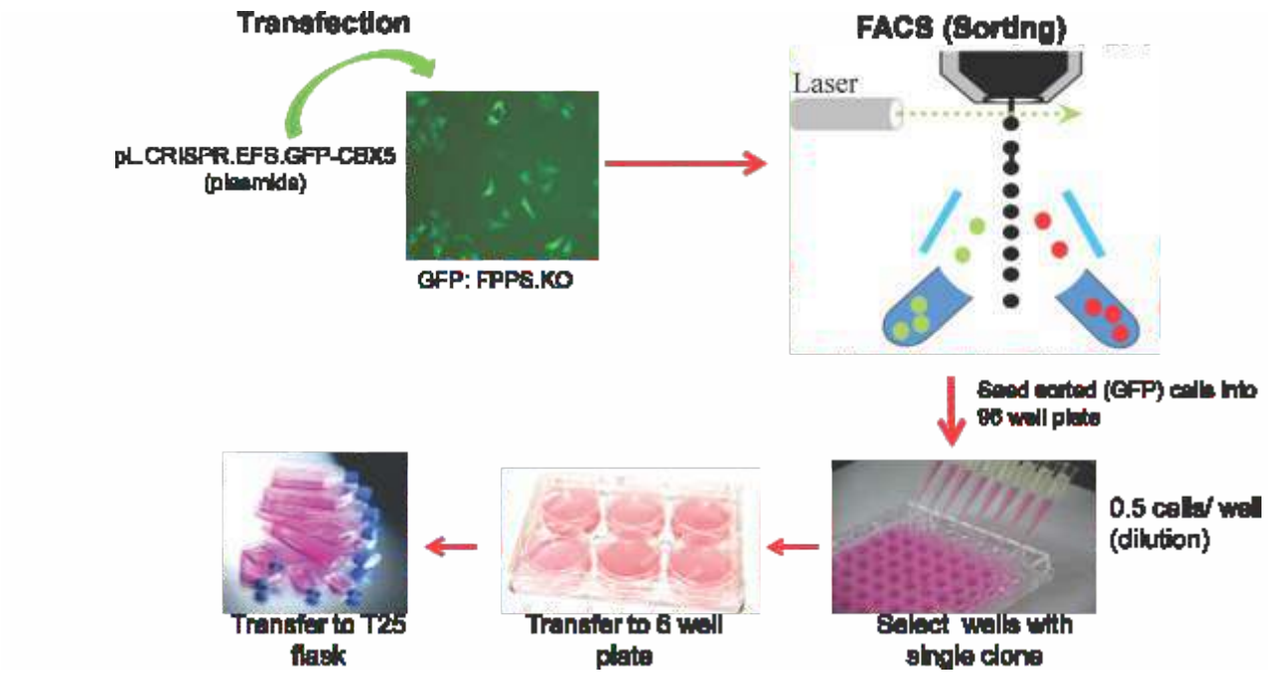
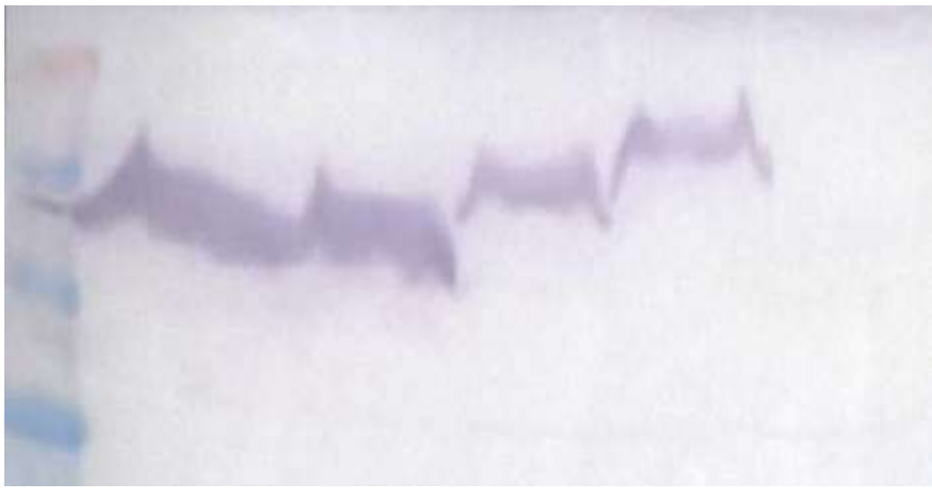


Fig. 1: Generating FPPS knock out cells by CRISPR/cas9-mediated genome editing. BHK21 cells were transfected with pL-CRISPR.EFS.GFP-FPPS constructs (plasmid expressing sgRNA that target FPPS gene). At 48 h post-transfection, cells were trypsinized and subjected to FACS (sorting) analysis. GFP expressing cells were collected aseptically and cultured in 96 well plate at a dilution factor of 0.5 cell/ml so as to get a single cell clone/per well in the 96 well plate. Wells with single clone were identified under microscope and further cultured in- 6-well plate, T25 flasks and finally in T75 flasks. Fourteen clones were evaluated for FPPS dusruption by PCR, nucleotide sequencing and Western blot analysis.

resistance with 6-10 passages (P), Thapsigargin and CGP57380 (host-targeting antiviral agents) did not induce generation of antiviral drug resistant phenotypes against NDV (up to ~P40) and BPXV (upto ~P25), respectively. However, at further higher passage (~P60 in BPXV and ~ P70 in NDV), a significant resistance was observed. Acquisition of antiviral drug resistance against host-targeting agents (SERCA/MNK1 inhibitor) is intriguing and its mechanisms are not well understood. One possibility of acquisition of resistance to host-directed antiviral agents may be to generate defective interfering (DI) particles (upon long-term virus culture) which differently suppress virus yield in drug-resistant and drug-susceptible viruses. In order to test this hypothesis, drug-resistant and drug-susceptible NDV at passage level 70 (P70), plaque were purified (n=10 each) and evaluated for their sensitivity to the drug (host-directed antiviral agent-Thapsigargin). Like virus mixtures (viral quasi species) (P70-Thapsigargin and P70-Control), plaque purified P70-Control viruses showed more sensitivity to the antiviral action of Thapsigargin than plaque purified P70-Thapsigargin viruses. This suggests that acquisition of drug resistance against host-directed antiviral agents is not simply due to the generation of DI particles rather due to the specific events that may involve switch to use alternate host factors or increased affinity of the resistant viruses to its substrate.



Western blot

Fig 1: pCDNA-DEST40_MNK1 and pCDNA DEST40_MNK2 clones were transfected to HEK293/HeLa cells and detected with C-Terminal 6x-His Tag.

In order to further confirm the requirement of MNK1 by BPXV, MNK1 was amplified from Vero cells and cloned in pCDNA3.1 mammalian expression vector by using Gateway cloning technology. The cloning constructs are likely to be expressed as His-tagged fusion protein. The expression of fusion proteins was confirmed by transfection of the plasmid constructs into 293 T cells and subsequently by Western blot analysis using Anti-His antibody (Fig. 1). It is planned to evaluate whether overexpression of MNK1 can rescue the inhibitory effect of MNK1 inhibitor in the target cells.

(Naveen Kumar and Sanjay Barua)

Effect of calcimimetic compound on Newcastle disease virus replication

Calcium has been identified as an essential component in the different stages of the life cycle of viruses. Viruses may use calcium for their attachment, entry, uncoating, genome replication or virus release from cells. The role of calcium has been identified in the growth of different paramyxoviruses such as measles and PPR viruses; however, the same has

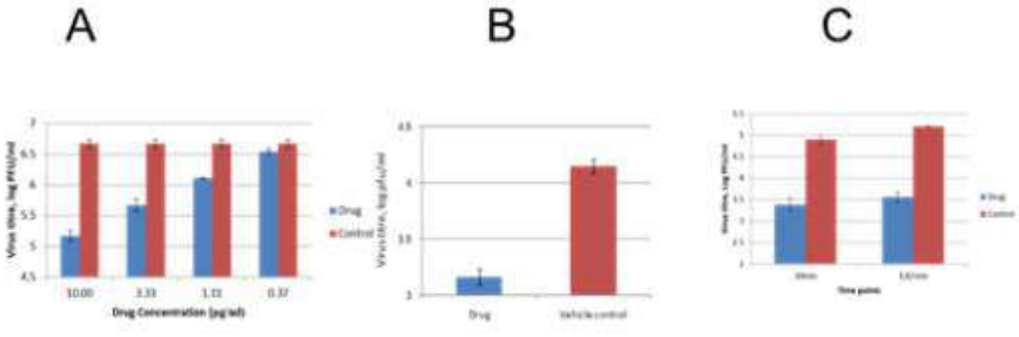


Fig. 1: Preliminary analysis on New Castle disease virus replication.

not been reported for Newcastle disease viruses (NDV). Hence, in this study we tried to explore the role of calcium in the growth of NDV. A plaque purified NDV available at NCVTC, Hisar was used in this study and all assays were performed in Vero cells. We used a calcimimetic compound (AC 265347) that has been found to modulate the entry of calcium into cells by binding to calcium sensing receptor (positive allosteric modulator). Initially, we performed an antiviral assay using noncytotoxic concentration of the compound and a 1.5 log reduction in virus growth was observed. Later, we performed step-specific assays to find out which stage of virus life cycle was affected by the compound. Our preliminary analysis revealed that both virus entry and release stages were affected (Fig. 1a, b, c). Further, analysis is underway to analyze the effect of the compound on virus genome and protein synthesis.

***A. In vitro* antiviral efficacy of calcimimetic compound:** Vero cells, in triplicates, were infected with NDV at MOI of 0.1 in the presence of indicted concentrations of AC 26534or vehicle-control. The virus particles released in the infected cell culture supernatants at 20 hpi were quantified by plaque assay.**B. Virus entry assay:** Vero cell monolayers, in triplicates, were pre-chilled to 4° C and infected with NDV at MOI of 5 in drug-free medium for 1h at 4° C to permit attachment. This was followed by washing with PBS and addition of fresh MEM containing 10 µM AC 265347or vehicle-control. Entry was allowed to proceed at 37° C for 1h after which the cells were washed again with PBS and incubated with cell culture medium without any inhibitor. The progeny virus particles released in the infected cell culture supernatants at 16 h in the treated and untreated cells were titrated by plaque assay.

C. Virus release assay: Confluent monolayers of Vero cells, in triplicates, were infected with NDV, for 2 h at MOI of 5 followed by washing with PBS and addition of fresh MEM. At 10 hpi, cells were washed 5 times with chilled PBS followed by addition of fresh MEM containing 10 µM AC 265347or or vehicle-control. Virus release at indicated time points (post- AC 265347 addition) was quantified by plaque assay.

(Riyesh T, Sanjay Barua and Naveen Kumar)

Authentication and accessioning of bacteria

During the year 2020, among the processed cultures, 75 cultures were accessioned in the bacterial repository which has led to total strength of Bacterial Culture Collection to 1502 veterinary bacteria. Cultures were submitted from Central Institute for Fisheries Education, Mumbai; SKUAST, Jammu; IVRI, Izatnagar; AAU, Khanapara, Post Graduate Institute of Veterinary Education & Research, Jaipur (RAJUVAS); CSWRI, Avikanagar; TANUVAS, Chennai; NRCC Bikaner, apart from accessions from NCVTC Bacteriology laboratory. In addition, many cultures are ready to be accessioned. Besides this, 57 pathological/other samples submitted/collected at NCVTC bacteriology laboratory; viz., samples from goat/sheep (25), buffalo (3), mice (9), equines PM samples (12), dog (1), donkey mare milk (5) including samples from contaminated cell culture used in *Theileria* culturing, lead to isolation of nearly 100 bacterial cultures, which are preserved in general preservation. Some of the significant bacteria identified are *Pseudomonas indoloxydans* (Ad1, Ad4); *Comamona saquatica* ssp. *rana* (Ad6, Ad8); *Faecalibacter macacae* (Ad17); *Pseudomonas hydrolytica* (Ad19, Ad30, Ad21); *Staphylococcus capitis* ssp. *urealyticus* (Ad29); *Arcanobacterium pluranimalium* (Bu91) (Fig 1a, b); *Pasteurella multocida* subsp. *gallicida* strain NCTC 10204 (RR283; RR284); *Streptococcus alactolyticus* (Ss23); *Streptococcus gallinaceus* (As3); *Acinetobacter portensis* (As3A); *Streptomyces cacaoi* subsp. *cacaoi* (As3B); *Luteimonas marina* (As3C); *Escherichia fergusonii* (Eq387, Mm17; Mm18A); *Pseudomonas simiae* (Eq392B); *Streptococcus uberis* (Mm18); *Enterococcus hirae* (Mm17A); *Prolinoborus fasciculus* (Eq414); *Streptococcus suis* (RR216; RR217; RR219; RR221; RR231); *Streptococcus parasuis* (Eq218); *Streptococcus dysgalactiae* subsp. *dysgalactiae* (RR220, RR222, RR224, RR225, RR226, RR227); *Streptococcus dysgalactiae* ssp. *equisimilis* (RR223, RR228); *Streptococcus gallinaceus* (RR232); *Bordetella avium* (RR236); *Aeromonas veronii* bv. *veronii* (Aq1; Aq3; Aq72; BAA1043); *Pseudomonas alcaligenes* (Aq59C); *Aeromonas punctata* (Aq77); *Aeromonas veronii* bv. *sobria* (aq88); *Aeromonas veronii* (Aq88B); *Staphylococcus arlettae* (As14); *Staphylococcus sciuri* (As14A); *Staphylococcus equorum* (As14B); *Corynebacterium terpenotabidum* (As14C); *Leucobacterceler* ssp. *celer* (As14D) (Fig 2a, b); *Brucella tritici* (As14E); *Bacillus zhangzhouensis* (Bu92); *Corynebacterium lipophiloflavum* (Bu92A); *Clostridium tertium* (Bu92B); *Paenibacillus alvei* (Bu96, Bu96A); *Bacillus circulans* (Bu96B); *Lysinibacillus boronitolerans* (Bu96C); *Solibacillus isronensis* (Eq415: Eq415B); *Terribacillus goriensis* (Eq415A); *Enterococcus faecium* (Eq418); *Weissella confusa* (Eq418A). CIFE Mumbai has submitted an important isolate of *Flavobacterium coloumnare*, etiological agent of Columnaris disease, a highly prevalent freshwater fish disease worldwide. SKUAST Jammu submitted 4 cultures out of which 2 significant cultures from dog urine were identified as *Burkholderi acontaminans* and *Achromobacter xylosoxidans*. Anaerobic bacterium *Clostridium butyricum* was isolated from intestinal content of buffalo mortality in NDRI, Karnal. It is a strictly anaerobic, Gram-positive, spore-forming bacillus capable of producing high amounts of butyric acid, and is common human and animal gut commensal bacteria. The bacterium is commonly used as a probiotic in Asia but recently it has been associated with pathological conditions in humans. Apart from this, we have also identified anaerobes *viz.*, *Paraclostridium sordelli*, and *Clostridium vulturis* from buffalo intestinal contents. There are accessions of *Salmonella* Typhimurium and *Listeria monocytogenes* from

RAJUVAS, Bikaner, important milk-borne pathogens. NRCC, Bikaner has submitted cultural isolates *Pseudomonas stutzeri* from camel mastitis, *Streptococcus gallolyticus* and *Staphylococcus hyicus* from camel urogenital tract, and an isolate of *Corynebacterium pseudotuberculosis* from camel pus.

Isolation and biochemical identification of bacterial isolates from pathological and environmental samples obtained from horse, donkey, buffalo, goat, sheep and fisheries pond water was performed. Bacterial cultures were biochemically investigated for identification by API method, which included genera belonging to family *enterobactericeae*, *aeromonadaceae*, *streptococcaceae*, *staphylococcaceae*, *corynebacteriaceae etc*. apart from many glucose non-fermentors. Important isolates were: *Streptococcus dysgalactiae* ssp. *equisimilis* (Eq431); *Staphylococcus saprophyticus* (Ang30); *Staphylococcus xylosus* (Ang31B); *Streptococcus equinus* (As10C); *Aerococcus viridians* (As13); *Brevundimonas vesicularis* (As6, As7); *Streptococcus infantarius* ssp. *infantarius* (Bu94; Bu94B); *Aerococcusurinae* (As4C;

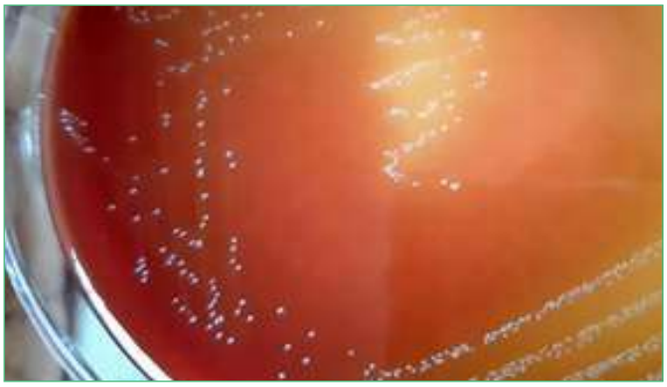


Fig. 1a: Small haemolytic colonies of *Arcanobacterium pluranimalium* on SBA.

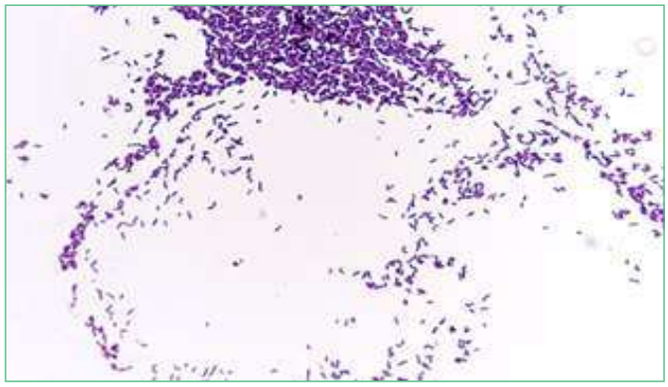


Fig. 1b: Gram positive coryneform rods of *Arcanobacterium pluranimalium* isolate from buffalo.

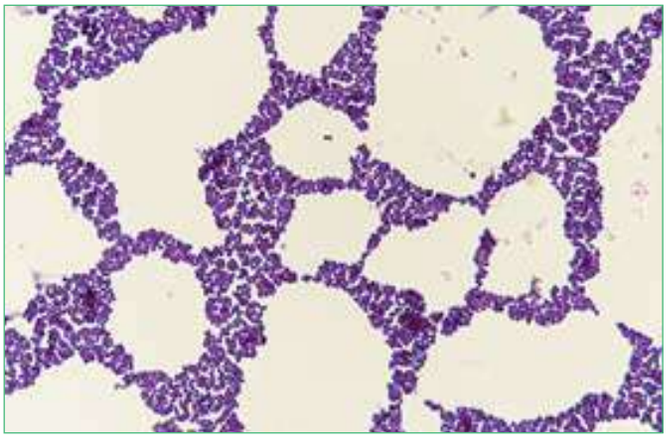


Fig. 2a: Gram-positive, irregular-rod- shaped cells of *Leucobacterceler ssp. celer*

Fig. 2b: Minute non-hemolytic whitish colonies of *Leucobacterceler* ssp. *celer*on SBA

Bu93; Bu94A; Eq414; Fo87A); *Leuconostoc* spp. (Eq418;Eq418A); *Corynebacterium bovis* (Bu92A); *Enterococcus faecium* (E1414A); *Pseudomonas flouroscens* (Bu95); *Pseudomonas aeruginosa* (Bu93A); *Aeromonas sobria* (Aq3); *Aeromonas hydrophila* (Aq1); *Bordetella avium* (RR/2020/236); *Sphingobacterium spiritivorum* (Fo193); *Sphingomonas paucimobilis* (Fo143); *Vibrio alginolyticus* (Oa19A; Fo140; Fo142); *Aeromonas salmonicida* ssp. *salmonicida* (Fo141); *Citrobacter koseri* (Fo55A); *Comamonas testorenii* (Fo54); *Arthrobacter* spp. (Fo53F; Eq408); *Pseudomonas stutzeri*Fo55B); *Corynebacterium argentoratense* (Eq412); *Arcanobacterium haemolyticum* (Eq411B); *Aerococcus viridians* (Eq396; Eq389; Eq391; Eq407; Eq409A, Eq410); *Enterococcus fecalis* (Eq403; Eq403A); *Streptococcus equi* ssp. *zooepidemicus* (Eq388; Eq393; Eq394; Eq395; Eq398; Eq399; Eq400; Eq400A; Eq401; Eq402; Eq404; Eq405; Eq405A; Eq406); *Streptococcus equi* ssp. *equi* (Eq383; Eq383A); *Enterococcus fecalis* (RR/2020/682) *Enterococcus feacium* (RR/2020/683); *Staphylococcus haemolyticus* (RR/2020/679); *Staphylococcus sciuri* (Anf1); *Burkholderia cepacia* (Eq392B); *Corynebacterium propinquum* (Ch19); *Brevundimonas diminuta* (As9A; Eq419A); *Pasteurella* spp. (Oa20); *Escherichia coli* (Eq419); *Enterobacter cloacae* (Eq416).

(R. K. Vaid, Taruna Anand, Jitender Kumar, N. Virmani and Yash Pal)

Distribution of Cultures to researchers all over India

The National Centre for Veterinary Type Cultures (NCVTC) which was established in 2005 with an aim to explore and collect microorganisms of animal origin, their central storage, further characterization, documentation and digitization of microbial database, development of a National Microbial GenBank and broader conservation and utilization of microorganisms. One of the most important mandates of NCVTC is supply of authenticated cultures to users under material transfer agreements (MTA). NCVTC has been working in this direction of providing authenticated cultures to researchers and has taken tentative steps in this direction in February of 2016, when it first supplied cultures of *Salmonella* Typhimurium to Department of VPH at GBPUAT, Pantnagar. Since then, NCVTC has progressed steadily in providing this service to researchers. Initially, in 2016, about 30 cultures of bacteria of genera *Salmonella* spp., *Streptococcus* spp., *Klebsiella* spp., *Mannheimia* spp., *Escherichia* spp., *Proteus* spp., and *Pasteurella* spp. were supplied to researchers in the state universities of Uttarakhand, Karnataka, and Haryana. In the following year 26 cultures of taxa *Staphylococcus* spp, *Streptococcus* spp, *Enterobacter* spp, *Pseudomonas* spp, *Serratia* spp, *Escherichia* spp, *Bacillus* spp., *Proteus* spp, *Enterococcus* spp, *Salmonella* and *Rhodococcus* spp. were distributed. From 2018 to 2020, the demand of cultures was steady as about 60 different cultures were procured by researchers. However, the depth of taxa required by researchers has increased, as researchers are making enquiries of cultures of *Brucella* spp., *Trueperella* spp., *Clostridium* spp, *Shigella* spp, *Nocardia* spp., *Fusobacterium* spp., *Actinobacillus* spp, *Yersinia* spp, *Corynebacterium* spp, *Aeromonas* spp. and *Listeria* spp, from across the length and breadth of country, apart from other cultures. Besides this, researchers also demand antimicrobial resistant strains as reference. NCVTC has now been distributing cultures from all directions of the country with demands coming from Jammu, Kashmir, Himachal Pradesh, Uttarakhand, Uttar Pradesh, Haryana and Punjab in the North; Tamilnadu, Andhra Pradesh, Karnataka and Kerala in the South; Gujarat, Maharashtra, and Rajasthan in the West and Assam, West Bengal, and Meghalaya in the East. Apart from Sate Agricultural Universities, NCVTC has also supplied cultures to CMVL, Meerut; Periyar University, Tamilnadu; MDU, Rohtak, Ella Foundation, Hyderabad; BHU, Varanasi, Central University of Punjab, Bhathinda and Animal Husbandry Department, Mumbai. As the awareness about availability of authenticated cultures will spread across country after opening of webportal ncvtc.org.in, the enquiry and dispatch of cultures will increase.

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

Isolation of *Weissella confusa* (Eq418A) from aborted mare fetus

In July, 2020, a case of abortion in a Zanskari mare was reported. On processing of sample, an isolate of *Weisella confusa* was isolated from fetal liver (Fig 1a,b). The bacterium grew slowly as 1 mm minute α-haemolytic colonies on 5% SBA. The Gram-positive cocci formed short chains and were catalase negative. Tentatively identified as member of *Streptococcus* spp. taxa, the isolate was found to be *Leuconostoc/Lactococcus lactis*; however the isolate was molecularly identified as *Weisella confusa* by 16S rRNA sequence identity. The 16S rRNA which is 98.5% complete showed a 96.88% similarity with *W. confusa* strain JCM1093 (Type) (AB023241). The isolate which is Voges-Proskaeur and esculin positive, needs to be polyphasically analysed to detect any differences from the Type strain to warrant a novel taxa. Phylogenetic analysis also indicate that strain VTC418A is at distance even within the *W. cibaria* clade (Fig 1c). *Weisella confusa*, which was previously classified in taxa *Lactobacillus confusa* has been earlier also reported to be involved in a foal septicaemia case. It has also been reported from human patients with bacteraemia infected solely with *W. confusa*. In this case, an ascending infection of fetus from *W. confusa* is suspected.

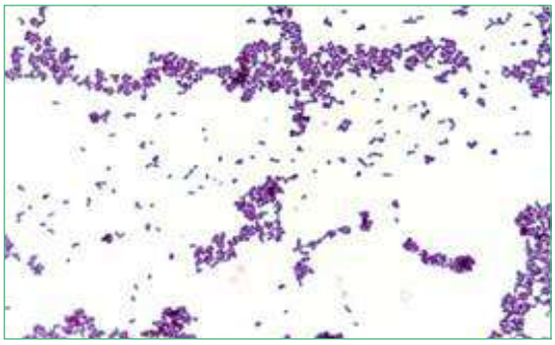


Fig. 1a. Gram positive short coco-bacillary rod like cells of *Weisella confusa*

Fig. 1b Minute dull alpha haemolytic colonies of *Weisella confusa* on SBA

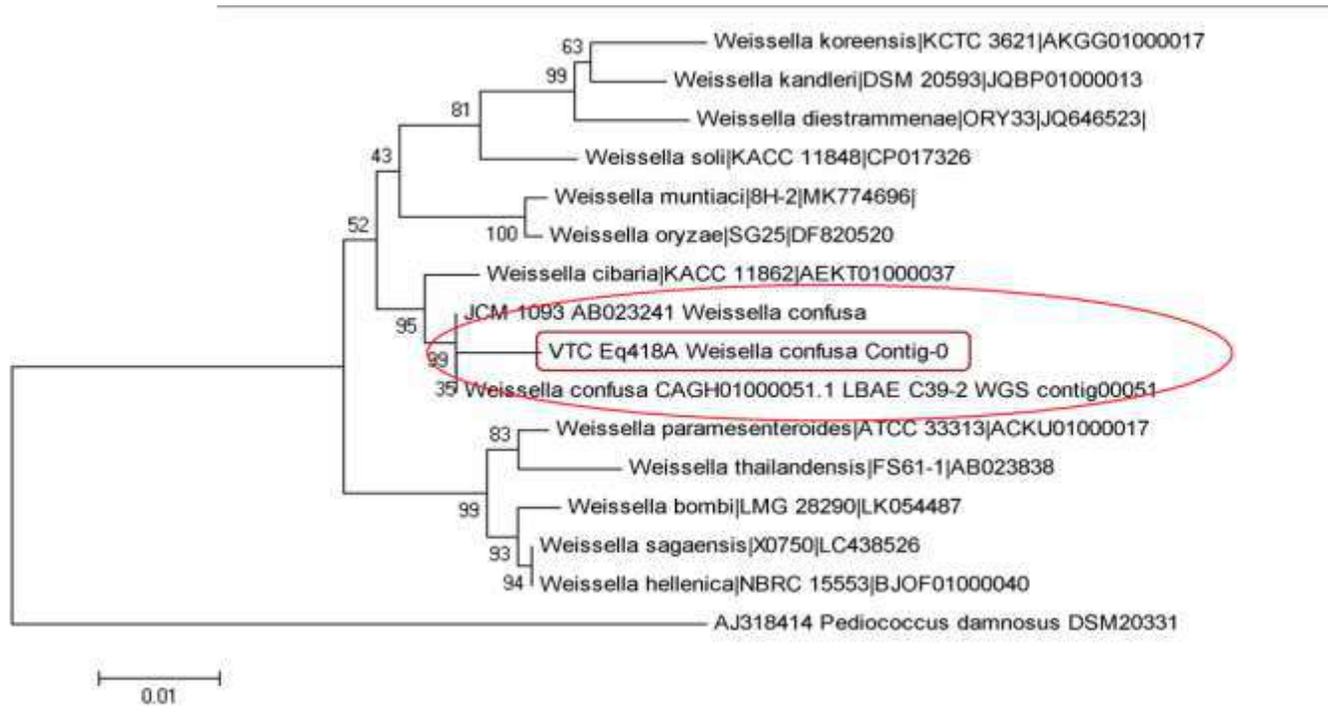


Fig. 1C: Phylogenetic tree based on homologies of 1300 bp sequences in the 16S rDNA of *Weissella* type strains with Eq418A strain

(R. K. Vaid, Taruna Anand and T. Riyesh)

Donkey mare milk microbial quality and identification

There is a recent renewed interest in the economic utility and exploitation of donkeys (*Equus asinus*), especially, for the production and consumption of donkey milk. This is because donkey mare milk is considered to be palatable and a useful source of healthy nutrients, especially for young, old and diseased. However, given the nutritive quality of milk, it may also act as source of bacteria. In India, lately there has been an interest in dairy donkey enterprise. It is therefore important that raw milk to be used as human diet needs to be characterised for an acceptable microbial load. Due to inherent nutritive content, milk provides a rich medium for microbial growth, and thus may constitute a public health risk, if consumed raw. This is borne by the fact that ruminant milk may act as a ready source of bacterial infection such as *Staphylococcus aureus*, *Salmonella* spp., etc. Therefore, under the project, we have undertaken to analyse the microbial load of raw donkey milk by enumeration. In addition, the bacterial colonies growing on media were isolated and characterized by standard phenotypic and molecular level up to species level. The quantitative enumeration of bacterial load in milk and identification of bacteria may give a good idea of the type of microbial hazard raw donkey milk may represent. Total seven samples of donkey milk were received in the bacteriology laboratory. The samples were collected in 50 ml sterile test tubes and transferred to laboratory and were processed immediately. Enumeration was done by plating appropriate decimal dilution on 5% SBA and incubation at 37°C for 24 hours. Colonies were counted and expressed in cfu ml^{-1}. Results indicated that aerobic plate count (APC) of bacteria ranged between minimum 3.6 x 10^2 to 7.4 x 10^3 log CFU ml^{-1}. This APC is lower than values reported (4.15 and 5.63 log CFU. ml^{-1}) in a study in Italy, although our samples are lesser in number. Moreover, the value are lower than internationally acceptable values for raw cattle milk meant for human consumption as raw milk (>3x 10^4 cfu/ml) (New York State regulations for raw milk intended to be consumed as raw milk). A total of 30 bacterial colonies were marked and isolated on the basis of morphology, abundance and hemolysis. Out of this, 22 (73.4%) were Gram-positive and 8 (26.7%) were Gram-negative. Important genera identified from donkey mare milk were *Staphylococcus* spp., *Streptococcus* spp., *Klebsiella* spp., *Acinetobacter* spp., *Streptomyces* spp., *Luteimonas* spp., *Micrococcus* spp., *Bacillus* spp., *Corynebacterium* spp., *Leucobacter* spp., *Mannheimia* spp, *Brucella tritici* (previously, *Ochrobactrum tritici*) (Fig 1a & b) and *Sarcina* spp. Although there was no isolation of *Staphylococcus aureus*, *Staphylococcus* spp. (7 strains-23.4%) were most common taxa followed by *Streptococcus* spp. (4 strains-13.4%), and *Streptomyces* (2 strains-6.7%). Six isolates could not be identified.

Presence of coliforms (*Klebsiella* spp.) in raw milk is not acceptable. The presence of *Staphylococcus* spp., *Streptococcus* spp., *Micrococcus* spp., *Corynebacterium* spp., *Brucella tritici*, *Streptomyces* spp, *Bacillus* spp, may be environmental or soil contaminant. Microbial identification of milk isolates have revealed presence of some unusual taxa *viz*., *Brucella tritici* (As14E); *Leucobacter celer* sub spp. *celer* (As14D); *Streptomyces cacaoi* subsp. *cacaoi* (As3B); *Luteimonas marina* (As3C); and *Corynebacterium terpenotabidum* (As14C).

Comparative Genomics of *Salmonella* Gallinarum

Salmonella enterica serovar Gallinarum biovars Pullorum (*S*. Pullorum) and Gallinarum (*S*. Gallinarum) are the etiological agents of pullorum disease (PD) and fowl typhoid (FT) respectively, which causes huge economic loss to poultry industry in developing countries. The disease caused by the non-motile invasive avian pathogen serovars S. Gallinarum and *S*. Pullorum leads to high morbidity and acute mortality in pullets and adult poultry in India and various countries of Asia, Africa and South America. As limited information is available on the diversity of *S*. Gallinarum strains causing PD and FT therefore, an approach to compare and understand the genetic content and virulence variation among various strains from different countries compared to Indian strain was made using bioinformatics tools.

Comparative genomics of 9 strains of *Salmonella* species elucidated the average genome size to be 46,57,781bp, with average GC% of 52.19. The pan genome of *S.enterica* serovar Gallinarum was found to be in open state with growth exponent value of 0.089 and reached 5091 cds and core genome development plot was limited to 3270 genes. A total of 318 CDS in the range of 3-102 were detected as strain specific genes. The Indian strain, S.Gallinarum Sal40 strain VTCCBAA614, significantly harboured the highest number (102) of singletons among the investigated *Salmonella* strains, most of which are hypothetical genes and need further investigations. The phylogenetic analysis of genomes depicted that all genomes formed two major clades comprising of *S*. Gallinarum and *S*. Pullorum strains. The synteny plot analysis performed by EDGAR of the investigated *Salmonella* genomes in reference to *S*.Enteritidis str. P125109 depicted large scale genomic rearrangements which included relocations, inversions, duplications and deletions. A total of 362 genomic islands (GIs) were detected with an average of 40.22 GIs per strain, a good part of which were related to virulence factors indicating the importance of uptake of foreign DNA in the evolution of virulence in these strains. Significantly, the highest number (44) of GIs were detected in Indian strain. Each of the analyzed *Salmonella* genome was detected to possess at least one candidate prophage region (Fig. 2). In total, 23 prophage regions were identified, with higher quantity in Pullorum strains. TA loci were identified in all the investigated genomes and total of 149 Type II TA loci were detected. The analyzed *Salmonella* genomes harboured acquired aminoglycoside resistance gene.

The pan genome and core genome development plot analysis revealed S. *enterica* serovars Gallinarum to possess an open pan genome state and finding of strain specific genes in all the investigated genomes reiterated the genetically polymorphic status and rapid evolution of *Salmonella* host adapted genomes. The finding of genomic islands, which are the regions of horizontal origin acquired genes that regulate pathogenesis, virulence and multiple drug resistance features needs further research. The finding of highest singleton CDS in Indian strain of *S*. Gallinarum is significant which warrants further investigations. More number of genomes of *S*. Gallinarum needs to be sequenced and compared to understand their pathogenic spectrum. This comparative genomic analysis of *S*.Gallinarum strains has provided a basis for future experimental studies to be carried out to decipher the underlying mechanisms driving the pathogenesis and virulence of this important pathogen.

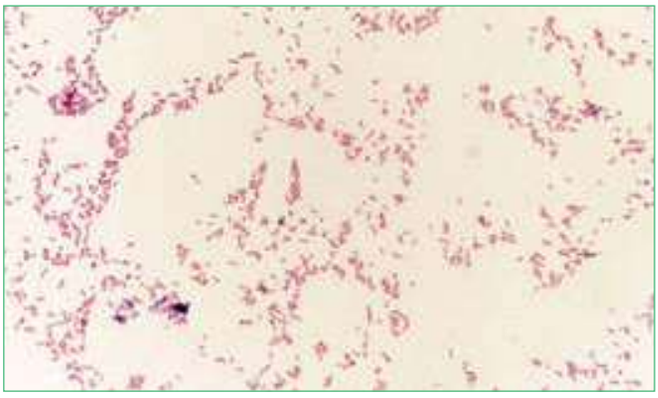


Fig. 1a: Gram negative small rods of *Brucella tritici*

b: Mucoid coalescing colonies of *Brucella tritici* on SBA

(R. K. Vaid, Zoozeal Thakur and Taruna Anand)

Indian network for fisheries and Animal antimicrobial resistance (INFAAR)

The antimicrobial resistance surveillance work was extended outside Hisar city in the village area where rectal swabs from Beetal Goats, Nali and Munjal breed sheep (24 nos.) were collected from Village Thaska, Hisar. From 24 samples, 43 *Escherichia coli* isolates were isolated, confirmed biochemically and by duplex PCR (*lac*Y&*pho*A positive). Disc diffusion test results showed that 13 (68.5%) isolates were resistant to one or more class of antimicrobials. Out of 13 isolates, 10 (77%) isolates were resistant to at least 1 antimicrobial tested, whereas 2 (15%) isolates (Ang19A, Ang2) were MDR strains as these were resistant to 3 classes of antimicrobials. Ang15A was resistant to 2 antimicrobials. Two CoNS were also isolated from goat fecal samples, i.e., *Staphylococcus saprophyticus* and *S. xylosus*. In addition, previously many isolates of *Staphylococcus aureus* were detected to be resistant to erythromycin. In view of the increase in methicillin-resistance in *S. aureus*, the utilization of Macrolide-Lincosamide-Streptogramin B (MLSB) antibiotics to treat *S. aureus* infections, particularly with clindamycin has increased. Clindamycin resistance, which is of 2 types, constitutive or inducible, is difficult to detect in inducible resistant strains, as they appear erythromycin resistant and clindamycin sensitive *in vitro*. This leads to treatment failures as use of clindamycin against strains of *S. aureus* appearing sensitive to clindamycin fails due to inducible resistance. In D test the discs of erythromycin and clindamycin are placed adjacent to each other leading to detection of inducible resistance. The D test was successfully performed for detection of macrolide inducible resistance to clindamycin in *Staphylococcus aureus* isolates which were resistant to erythromycin (Fig). This is an instance of detection of inducible clindamycin resistance in isolates of *S. aureus* from clinical mastitis in buffaloes.

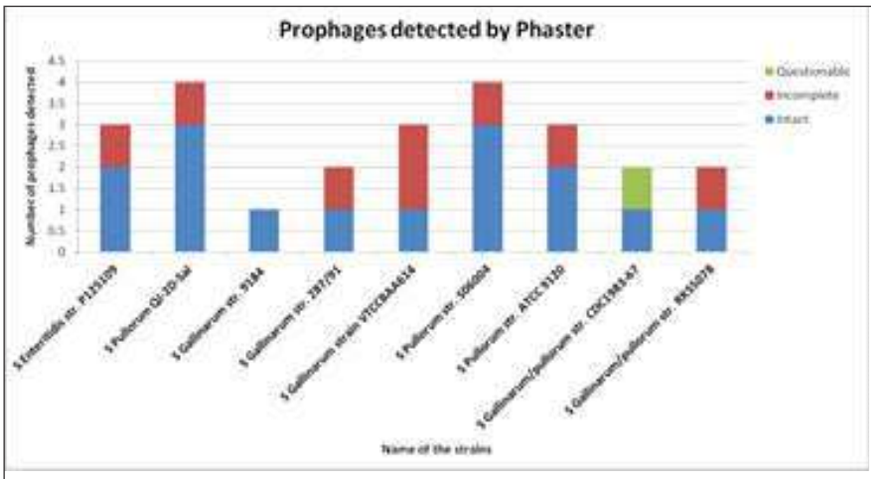


Fig. 2: Prophages detected by Phaster in analyzed *Salmonella* strains.

Fig. 1: 'D test' showing the discs of erythromycin and clindamycin placed adjacent to each other leading to detection of inducible resistance

(R. K. Vaid, Taruna Anand and H.S. Singha)

Strengthening of Bacteriophage repository at NCVTC

Bacteriophages against *Acinetobacter baumannii* added to the phage repository: Widespread antibiotic use in medicine and livestock industry has contributed to the global spread of multidrug-resistant (MDR) bacterial pathogens, including *Acinetobacter baumannii*. *A. baumannii* is one of the ESKAPE pathogens and it has been frequently associated with infections in clinical settings. *A. baumannii* is a Gram-negative bacteria capable of acquiring and maintaining multiple genetic elements encoding antimicrobial resistance determinants. Bacteriophages against this pathogen offer an alternative to antibiotics. Together with their enormous abundance, great diversity, and relative ease of isolation, phages offer an unlimited source of antibacterial agent against *A. baumannii*. A highly lytic phage - VTCCBPA145 against *Acinetobacter baumannii* was isolated from sewage by enrichment technique. The phage produced clear plaques with a zone of halo. The phage belongs to family *Siphoviridae* as observed by transmission electron microscopy (Fig. 1 a, b).

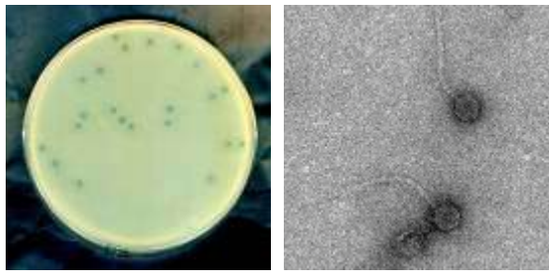


Fig. 1 *A. baumannii* phage –VTCCBPA145 a) Plaques on NA; b) TEM

(Taruna Anand, Medhavi Vashisth, Nitin Virmani, B.C. Bera and R.K. Vaid)

Phage therapy against mastitis in swiss albino mouse model: Phage therapy using a characterized phage was carried out in Swiss albino mice after 10-12 days of parturition with 4-5 litter size. The host bacteria used for the infection of mammary glands was *S. aureus* (MTCC96). Bacterial infection leading to development of mastitis was observed in mammary glands as observed visually and by CFU counts on mannitol salt agar media. Phage therapy using a broad spectrum phage was carried out. Phage particles were recovered back from mammary glands during the period of therapy. Beneficial effects for curing mastitis were observed in mice model (Fig. 2). But the effects need to be validated further with more no. of animals and in large animal model.

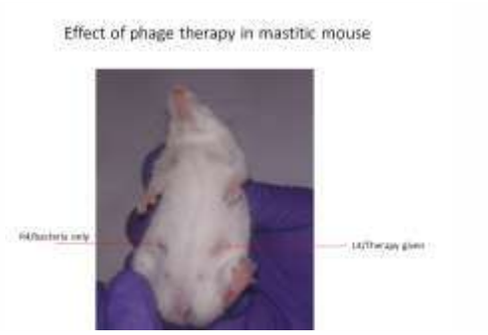


Fig. 2: Effect of phage therapy on mastitis mice

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

Whole genome sequence analysis of VTCCBPA139: The complete genome analysis of bacteriophage VTCCBPA139 against *Proteus mirabilis* was carried out. This phage was isolated from poultry litter against a drug resistant *P. mirabilis*. The phage genome is ~94kb with 95.88% similarity with *Proteus* phage PM135. The genome has 37.69% G+C and carries 153 predicted genes out of which 136 were annotated and 17 tRNAs were found in the genome (Fig. 3).

(Taruna Anand, Anubha Pathak, Nitin Virmani, B.C. Bera and R.K. Vaid)

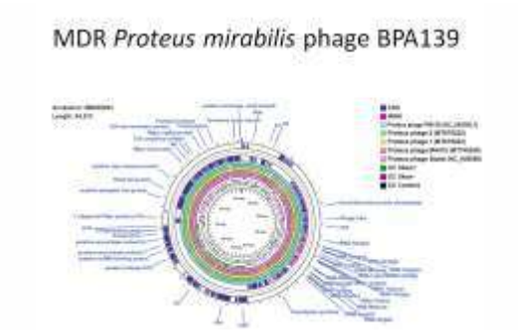


Fig. 3: MDR *Proteus mirabilis* phage BPA139

Development of repository of respiratory viruses of livestock

Viral infections are very frequent in most livestock-rearing systems in the country. This leads to huge annual losses in dairy and meat industries affecting milk and meat production due to mortality and treatment cost. The complicated nature of mixed infections and emergence of new respiratory pathogens, studying responsible pathogens, their variants and interactions are paramount important and are still largely speculative as no detailed investigation of causative agents of respiratory viruses has been thoroughly explored. The repository of respiratory viruses of livestock with long term storage of authenticated viruses will be a ready-made asset for the scientific community to explore these biological resources for future development of diagnostics, immunobiologicals, as reference material, comparative biological studies, etc. Biological samples were collected for identification and isolation of respiratory viruses of livestock species. A total of 55 nasal swabs of cattle were collected from Govt. Livestock Farm, Hisar and Gaushala, Hisar. The samples were collected in viral transport medium maintaining proper cold chain and screened for pathogens like BRSV, BPIV3, BCoV, BVDV and BoHV employing a multiplex RT-PCR assay. None of the samples was positive for any of the targeted viruses.

Standardization of isothermal "Recombinase Polymerase Amplification" (RPA) based assays for detection of Porcine circovirus 2 (PCV2) and 3 (PCV3)

Porcine circoviruses - small non-enveloped circular single-stranded DNA virus under the genus Circovirus within the family *Circoviridae* incur huge economic losses to swine industry globally. The variants of PCV i.e. PCV2 and PCV3 are prevalent in swine population of the country. We have reported the association of PCV3 with reproductive disorder and piglet mortality in different parts of the country. The point-of-care detection based on Recombinase polymerase amplification (RPA) is a new isothermal nucleic acid diagnostic technique that utilizes bacterial genome repair enzymes to rapidly amplify target sequences. Instead of heating and annealing, RPA utilizes a bacterial recombinase and single-stranded DNA binding protein to match primers to their target on the template DNA. One of the advantages of the RPA assay is that the reaction can be conducted in a single tube and produces 'real-time' results. This is made possible by the inclusion of a fluorescent probe. One of the prime advantages cited for RPA is the potentially higher degree of fidelity of the assay, which can result in a test of high specificity. The fact that currently available diagnostics are time-consuming, require expensive laboratory settings and well-trained personnel, hence the aim of development of point-of-care diagnostics employing recently developed RPA technology was rapid and highly sensitive detection of PCV2 and PCV3 viruses at field level. We have designed and synthesized primers targeting the conserved region of ORF2 of PCV2 and cap gene of PCV3. The designed primer sets have been successfully amplified from the PCV2 and PCV3 DNA using

AmplifyRP® Acceler8® kit (Agdia, Life Technologies India Pvt. Ltd) based on isothermal nucleic acid amplification technology which provides all the reagents necessary to amplify DNA at a single operating temperature (39°C). Further, standardization of RPA assays is underway.

(B.C. Bera, Taruna Anand, Nitin Virmani, Riyesh T. & B. R. Gulati)

Isolation, characterization and generation of repository of Mycobacteria

A total of 26 fecal samples collected from sheep were processed for isolation of *Mycobacterium* spp. of these, 9 fecal samples had acid fast bacilli (Table 1). All fecal samples were tested for the presence of acid fast bacilli by PCR targeting *afb* and *hspX* genes. Six fecal samples were positive for the *afb* PCR and four samples were positive for *hspX* genes. All

Table 1: Samples screened to detect *Mycobacterium* species by *afb* and *hspX PCR*

S. No	Sample	Acidfast staining	afb and hspX PCR
1.	Soil samples (n=4)	Negative	Negative
2.	Sheep intestinal samples (n=19)	11 positive	11 positive
3.	Sheep fecal samples (n=26)	9 positive	6 positive for afb and 4 positive for hspX
4.	Nilgai fecal samples (n=6)	3 positive	6 positive for afb and 3 for hspX
5.	Cow fecal samples (n=1)	Negative	Negative
6.	Lung sample (n=1)	Negative	Negative

fecal samples decontaminated and inoculated into in house HYEM and LJ media in duplicates and are under incubation. We were able to isolate two *Mycobacterium* species from a fecal sample and they were identified as *Mycobacterium novocastrense* by sequencing of *afb and hspX* genes (Fig. 1a, b and c). Further, molecular

Fig. 1a and 1b: *Mycobacterium novocastrense* colonies isolated from a sheep fecal sample on HYEM media.(1c) Acid fast bacilli by Ziehl-Neelsen staining.

characterization is under progress. A total of 19 intestinal samples from sheep were triturated and decontaminated, of which, 11 intestinal samples had acid fast bacilli and were positive for *afb* and *hspX* genes by PCR. A total of 6 fecal samples from Nilgai were collected and three fecal samples had presence of acid fast bacilli. All fecal samples yielded specific amplification *afb* gene. However, only three samples showed positive amplification for *hspX* gene. *Mycobacterium abscessus* (Fig. 3 and b) was isolated from Nilgai fecal sample and identified by sequencing of 16S rRNA, *afb* and *hxpX* genes. A total of four soil samples were collected from Hisar, screened for acid fast bacilli by staining as well as PCR and found negative. Dung (n=2) samples were collected and screened for acid fast bacilli by staining and PCR and found negative.

(Shanmugasundaram K, R. K. Vaid, B. N. Tripathi and B. C .Bera)

Equine Production

Endurance and fertility analysis in indigenous horses using SNP markers

PCR amplification and SNP genotyping for a total of 12 SNPs loci associated with endurance and fertility was done for remaining samples of Marwari, Kathiawari, K-Sindhi, Manipuri and Zanskari to have a minimum of 10 animals each in Marwari, Kathiawari and K-Sindhi and a minimum of 5 animals each in Manipuri and Zanskari breeds. The gene and genotype frequency was calculated in each breed at all the loci genotyped. The status of polymorphism and frequency of maior and major alleles has also been worked out. In general, the ponies *i.e.*, Manipuri and Zanskari have exhibited lower polymorphism as compared to the horse breeds probably because of small sample size and /or small population base even in the breeding tract. Existence of polymorphism in Indian horses and ponies indicates the possibility of their use in selection programmes.

(S.C. Mehta and T.R. Talluri)

Assessment and optimization of equine management in an intensive system

Inventory Database: The inventory database of the Marwari, Kathiawari, Manipuri, Zanskari horses and donkeys maintained at the Campus has been updated for the period 1989 to 2020, *i.e.* from date of inception of the Campus to till date.

Biometry Analysis

The biometry, *viz*. wither height, body length, heart girth and body weight data of Marwari horses since 1989 till date were analyzed for pedigree relationship, calculation of inbreeding coefficient, analysis of phenotypic trend, estimation of breeding values, estimation of heritability and analysis of variance. In all 30 years' period was covered and 226 records were analyzed. The average height at withers was 150.15±0.04 cm, body length was 151.44±0.06 cm, heart girth was 170.02±0.19 cm and body weight was 371.34±0.52 kg. The analysis of variance indicated non-significant effect of sex, tier and interaction of sex and tier.

Breeding Value

The breeding values were quite close to the population means except for the body weight. The effect of sex, tier and sex *tiers was non-significant on EBV of wither height, body length and heart girth. The effect of tiers on EBV of body weight was significant ($P<0.05$) indicating that body weight received favour in breeding programme.

Breeding Plan

The pedigree, inventory and biometry databases were updated. The body colour, semen quality parameters and availability of the semen doses was checked. Looking into the demand of the farmers, the horse needs to be bred for body height and body colour. Accordingly, selection differential for body height and independent culling level for body length and heart girth was adopted for selecting the animals. Specific sire was allotted to a dam for breeding keeping in view of above criteria and the pedigree of the animals to be mated. This year the project team visited Dev Stud Farm, Rajsamand and collected semen of three stallions having good height. The breeding plan was prepared for Marwari, Kathiawari, Manipuri and Zanskari animals. The average height of selected Marwari stallions was 160.6 cm and the average height of breedable females was 150.30 cm. The average body length and heart girth of selected stallions were 150.60 cm and 173.1 cm respectively.

Purchase of Nukra Stallions

Looking at the demand of the farmers, two Nukra males were purchased from the field and after cryopreserved about 100 doses of semen for AI of farm/ field mares. (Fig.1)

Fig. 1: Nukra stallion

Sale of Semen

This year after fixing the cost of semen for sale, 47 doses of the Marwari semen were sold to the veterinary professional for artificial insemination. Though, the semen doses of Marwari stallions were supplied to field veterinarians free of cost in previous years for artificial insemination in field mares.

National GenBank

The semen straws of Marwari (500), Manipuri (480), Zanskari (500) horses and Halari donkeys (500) have been submitted in the National Semen Bank at NBAGR, Karnal. NBAGR has also collected the Somatic cells from the Centre for cryopreservation.

(S. C. Mehta, R.A. Legha, R.K. Dedar, T.R. Talluri and Jitendar Singh)

Assessment, evaluation and identification of physical, biochemical and genetic factors affecting stallion fertility.

Biochemical analysis of spermatozoa and seminal parameters

Various biochemical and hormonal profiles of the Marwari stallion semen were carried out and correlated with that of seminal quality parameters (Table 1). GOT, GPT, TP and Ca differed significantly in seminal plasma while glucose differed significantly in spermatozoa (10^9 million) among stallions. GOT, GGT, TP, Ca and P were significantly lower in the stallions below 4 years of age. Glucose and Ca levels were found to have significant positive correlation with progressive motility and Cholesterol was found to have significant positive correlation with viability and membrane integrity.

Table 1: Correlation between biochemical parameters in spermatozoa (10^9) and seminal characteristics

	Total volume	Gel free volume	Progressive motility	Concen-tration	Viability	Total abnormality	Membrane integrity	Acrosome integrity	DNA integrity
Glucose	-0.290*	-0.248	-0.217	0.418**	0.292*	0.167	0.306*	-0.277*	-0.364**
Fructose	0.038	0.086	-0.119	-0.097	-0.171	0.107	-0.064	-0.205	-0.035
TP	-0.191	-0.177	-0.151	-0.113	-0.039	0.111	0.000	-0.118	-0.072
Cholesterol	-0.135	-0.081	-0.127	-0.242	0.109*	0.057	0.116**	0.196*	-0.182
Triglyceride	0.038	-0.030	-0.117	-0.101	-0.080	0.090	0.062	-0.115	-0.065
Ca	0.090	0.040	0.010**	0.045**	0.052	0.004	0.023	0.043	0.089
P	-0.020	0.005	-0.001	-0.011	-0.016	-0.012	-0.100	0.012	0.098

** Correlation is significant at the 0.01 level, * Correlation is significant at the 0.05 level.

Assessment of kinematic and morphological parameters of spermatozoa

Evaluation of effect of breed on motility and kinematic parameters of the spermatozoa were studied and recorded and presented in Table 2. On one way ANOVA effect of breed was found highly significant ($p \leq 0.01$) on some parameters, significant ($p \leq 0.05$) on some parameters and non-significant on some parameters. Sperm morphometry was also studied across different breeds of stallions and the same is depicted in Table. 2. A significant difference was observed in the parameters like head length and tail length of Marwari and that of Manipuri and Zanskari stallions and there were no significant differences observed among the Manipuri and Zanskari stallions.

Fertility genes assessment between seasons

The stallion seminal mRNA was isolated and subjected to analysis for the expression of various fertility related genes like SPATA, PLCz, SP17, PRM1, Ubiquitin and CRISP3 genes. The results of present study showed that all the 6 genes expressed in the Marwari stallions. The results show variation in the expression levels of the same gene between the seasons (Fig. 1). The results also demonstrated that *CRISP3* and *UBQ* genes were expressed more in the breeding season than the non-breeding season while *PLC1*, *PRM1*, *SPATA1* and *SP17* were expressed more in the non-breeding season than the breeding season. All the six fertility related marker genes (*CRISP3*, *UBQ*, *PLC1*, *PRM1*, *SPATA1* and *SP17*) showed differential expression between the seasons in Marwari stallions.

(T.R. Talluri, Yash Pal, S.C. Mehta and Anuradha Bhardwaj)

Table 2: Kinematic and morphometric analysis of seminal parameters between different indigenous breeds and exotic donkeys.

	Marwari (54)	Zanskari (22)	Manipuri (18)	Poitou (36)	Overall (130)	P value	Significance
DSL	20.11±1.22	18.41±0.84	15.78±0.66	19.61±0.99	19.08±0.61	.088	NS
DCL	46.12±2.46	59.01±1.91	60.31±2.60	54.61±3.19	52.62±1.51	.000	**
VAP	63.97±3.57	76.98±3.68	80.57±4.25	90.64±5.29	75.86±2.43	.011	*
VSL	48.415±2.57	51.94±2.61	47.47±2.81	66.79±4.96	53.98±1.95	.633	NS
VCL	108.19±5.29	150.90±6.79	167.44±9.86	170.72±8.82	140.93±4.4	.000	**
STR	75.31±1.20	65.29±1.15	61.08±1.47	72.27±1.80	70.80±0.88	.000	**
LIN	46.93±1.62	34.68±1.33	31.62±1.07	40.36±1.84	40.99±1.02	.000	**
ALH	4.79±0.18	6.92±0.28	8.58±0.29	7.37±0.35	6.39±0.19	.000	**
BCF	35.15±0.55	36.86±0.79	28.55±1.45	31.60±0.93	33.54±0.48	.000	**
WOB	59.66±1.34	50.53±1.17	49.92±1.09	53.50±1.33	55.06±0.79	.000	**
Head length	5.44±0.16	5.41±0.27	4.59±0.14	4.58±0.15	5.081±0.09	.014	*
Head width	3.21±0.10	3.25±0.12	2.86±0.10	2.81±0.08	3.05±0.05	0.119	NS
Head elongation	0.61±0.01	0.63±0.02	0.65±0.01	0.64±0.01	0.63±0.08	0.322	NS
Head perimeter	15.00±0.39	14.99±0.63	12.98±0.41	12.80±0.38	14.11±0.24	0.021	*
Head area	16.57±0.94	16.77±1.16	12.61±0.75	12.52±0.69	14.93±0.51	0.045	*
Tail length	14.28±0.42	13.36±0.62	12.17±0.41	14.18±2.47	13.80±0.71	0.025	*
Tail STR	64.96±1.22	62.62±2.19	67.34±2.10	61.34±2.92	63.89±1.07	0.283	NS
Droplet distance	4.06±0.57	5.82±1.17	3.98±1.01	3.23±0.62	4.11±0.38	0.270	NS
Droplet frame count	4.55±0.66	3.32±1.05	2.22±0.55	2.00±0.44	3.31±0.37	0.147	NS
Bent Tail count	18.63±0.71	18.41±1.17	16.78±1.16	14.08±1.28	17.08±0.55	0.427	NS
Coiled count	6.18±0.45	6.68±0.70	5.00±0.69	5.72±0.94	5.98±0.35	0.250	NS
DMR count	0.24±0.084	0.14±0.07	0.16±0.09	0.26±0.16	0.22±0.05	0.678	NS

Flow cytometric assessment of seminal parameters of stallion spermatozoa

Flow cytometry is a powerful technique which allows the simultaneous analysis of multiple physical characteristics of single particles, usually cells, as they flow in a fluid stream through a beam of light tuned at a particular wavelength. Flow cytometry s used to determine sperm concentration, viability, mitochondrial membrane potential, acrosome integrity, capacitation, DNA content and status etc. In this study, we standardized the protocols for assessment of seminal quality parameters using flow cytometry. Keeping in view of these advantages, the seminal parameters like viability and MMP were assessed in stallion spermatozoa. The reactive oxygen species (ROS) contents were also measured through flow cytometry using MitoSox marker. The study showed that cryopreservation process has significant effect on MMP and production of ROS

JC-1 is widely used in apoptotic studies to monitor mitochondrial health. JC-1 dye can be used as an indicator of mitochondrial membrane potential in a variety of cell types. JC-1 dye exhibits potential-dependent accumulation in mitochondria indicated by two fluorescence emissions green (~529 nm) and red (~590 nm). JC-1 has advantages over other cationic dyes as it can selectively enter into mitochondria and reversibly change colour from red to green as the membrane potential decreases.

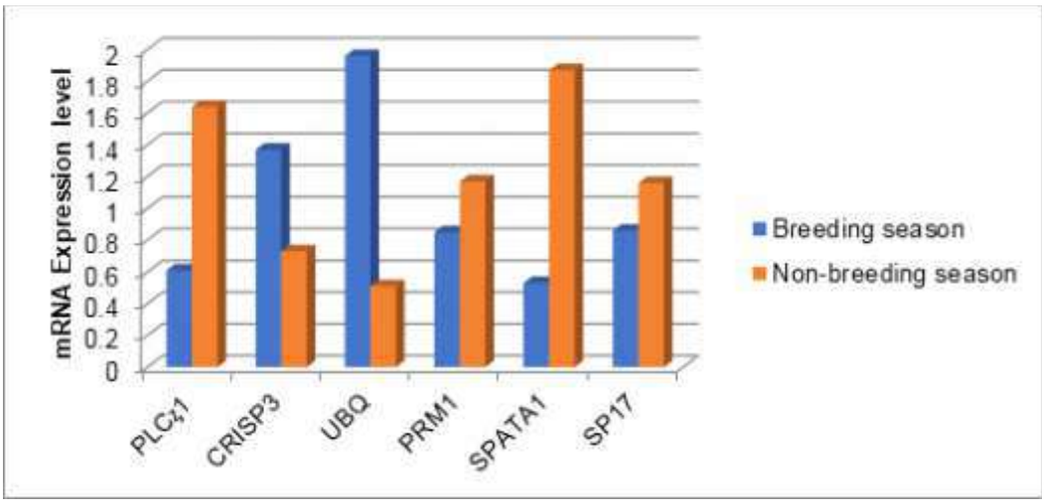
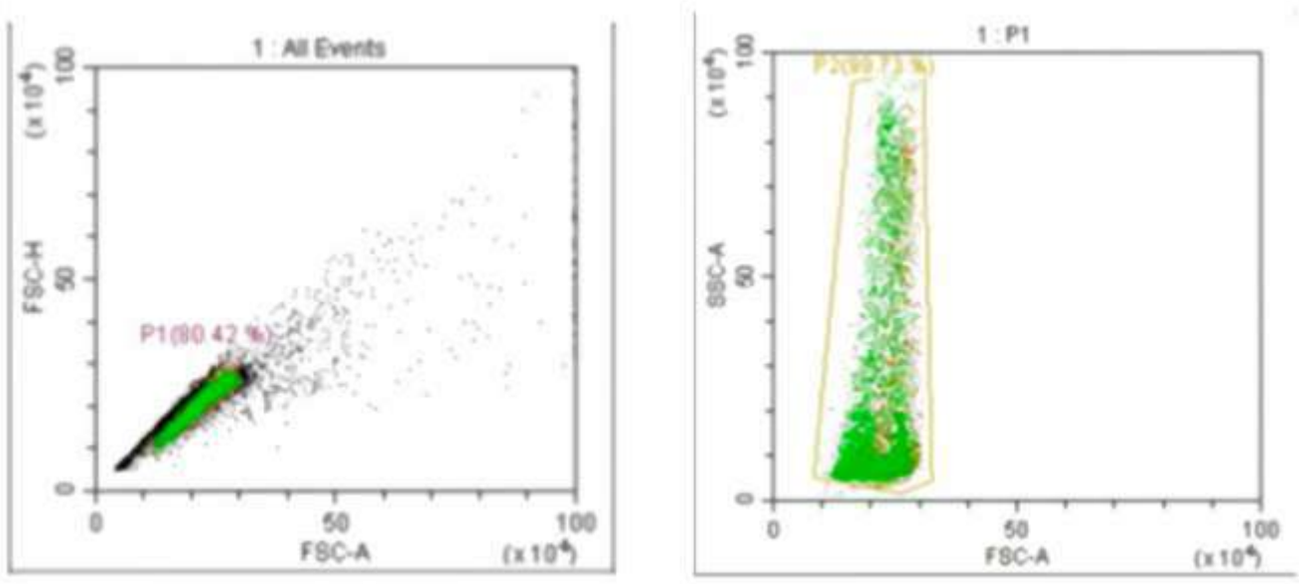
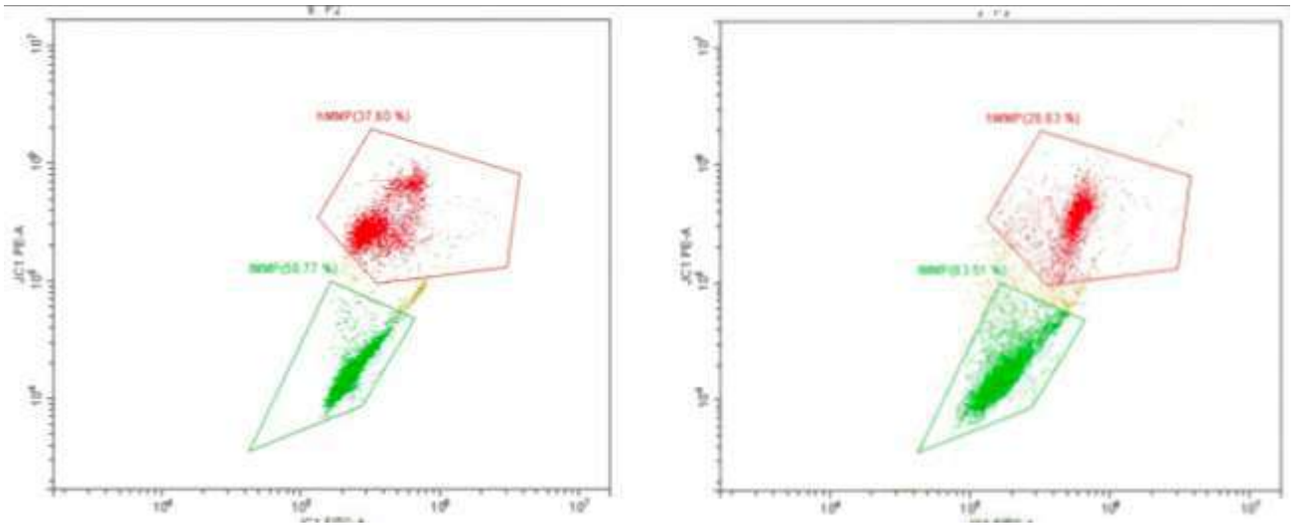


Fig.1: Overall mean C_T values (Mean±SE) of (*PLC1*, *CRISP3*, *UBQ*, *PRM1*, *SPATA1* and *SP17*) in sperm of Marwari stallions during the breeding and non-breeding seasons

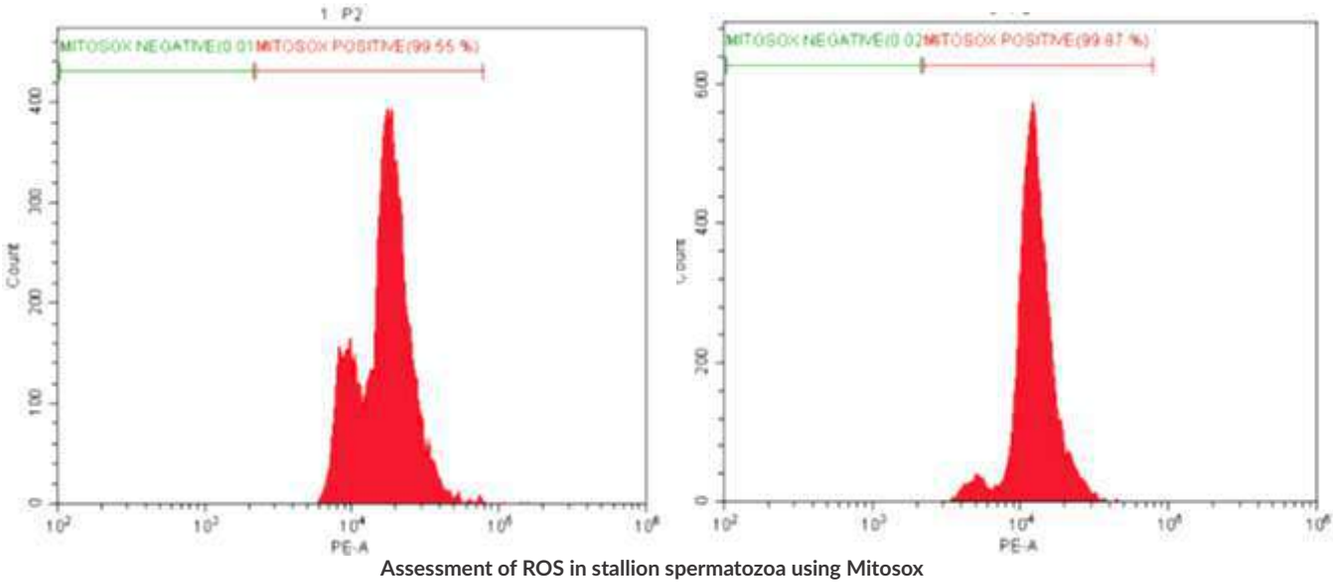


Stallion sperm viability assessment using SYBR-PI



Assessment of MMP in stallion spermatozoa

ROS estimation in spermatozoa: The MitoSOX™ Red is a cationic derivative of dihydroethidium (also known as hydroethidine) designed for highly selective detection of superoxide in the mitochondria. The cationic triphenylphosphonium substituent of MitoSOX™ Red indicator is responsible for the electrophoretically driven uptake of the probe in actively respiring mitochondria of a cell.



Assessment of ROS in stallion spermatozoa using Mitosox

Foal heat breeding of mares- a strategy to obtain a foal per year

To realize the optimum economic return of broodmares, the number of foals produced per dam must be maximized. The horse mare recovers its reproductive cycle soon after delivery, showing foal heat 5 to 12 days after parturition. The resumption of ovarian activity with ovulation after parturition occurs in a physiologically short time frame. A total of ten mares (6 Marwari, 2 of each Kathiawari and Zanskari) were bred during the foaling period and monitored their duration of foal estrus and other parameters. The foaling rate of these animals was also recorded. The pregnancy status was examined at 30, 60 and 90 days post mating and pregnancy rate was calculated as number of mares pregnant at 30, 60 and 90 days post mating out of total mares mated. The foaling rate was calculated as number of mares foaled out of total mare mated. The rates obtained were converted into percentages.

(T.R. Talluri)

AICRP on Increased Utilization of Animal Energy

Development of fatigue cum fitness score card for working equines

Three adult mules were selected for the draughtability trials. The payloads equivalent to 2.0X, 2.5X, 3.0X, 3.5X and 4.0X of their body weights were tested in mules using conventional pneumatic wheel cart on pucca road at normal speed of mules for four hours duration. Physiological responses were studied before, during and after the work and 20 min post work. Rectal temperature, respiration rate and pulse rate levels increased significantly during work at intervals. After 4 hour of work, the physiological responses attained the highest value. There was significant decrease in the values after a rest of 20 min, but these values were significantly higher than control values. Significant decrease in the speed of mules was observed after every hour of work. All the physiological indices increased significantly after work and came to normal range in the next morning. Blood samples were collected before and after work for hematology and estimation of biochemical parameters. Different loads had no significant effect on various hematological indices (Hb, RBC, WBC, HCT, MCV, MCH, MCHC). Fatigue score was estimated based on parameters described by Bhatt *et al.*, (2005). Meteorological data during trial period were also recorded. Significant reduction in body weight was recorded after work but recovered by the next morning. The body weight reduced after work was 9-10 Kg in pay loads equivalent to 2.0X, 2.5X, 3.0X and 3.5X of their body weight. However, it was 14 Kg reduction in 4.0X payload during carting. Body measurements of mules and donkeys in carting and pack load trials were recorded. Average body length, height and heart girth were 142.33, 119.33; 147.33, and 121.66, 168.0,136.33 cms, respectively for mules and donkeys. The trials of different payloads on mules have been completed. The results will be analyzed in consultation with IASRI scientists as advised for development of fatigue score card.

(R.A. Legha and Yash Pal)

Table 1: Details of foal heat occurrence, foal heat duration and conception rate in three indigenous horse breeds (in days).

Breed	First sign of estrus	Estrus duration	Conception
Marwari (6)	9.28±2.47	6.48±1.34	68.89±4.37
Kathiawari (2)	9.19±2.81	5.92±2.19	39.14±2.31
Zanskari (2)	7.04±3.07	7.38±1.20	60.24±4.30

Development of improved saddle and harness for working equines.

Different saddles fabricated during previous year were tested in mules and donkeys during carting trials at our centre. Normally iron base of saddle is fitted with wooden piece to fix the saddle properly on back of the animal. To avoid injury the wooden piece is fitted with locally made padding of rag. But most of the time this rag padding is not able to protect the animal from injury or saddle sore due to hardness, unevenness as well as absorbing of sweat. To refine/improvise the existing saddle, wooden piece was padded with nanda (carpet cloth) in Fig.1 the same was further covered with leather as shown in Fig.2. Refinement of saddle for carting equids was done. Testing of these saddles has been done on institute donkeys and mules during carting trials at our centre. Under saddle, blanket support was provided as shown in picture. The donkeys and mules were comfortable and no injury or saddle mark was observed on the back of experimental animals.

Improved carting saddles for donkeys

Fig 1

Fig 2

Fig 3

Body confirmation of horse, pony and mule is different. Hence, the saddle made for horse can't as such be used for pony or mule. But most the owners use the same saddle for mule and pony. Also, the same type of saddle can't be used for donkeys of different sizes. If saddle is not as per the size and shape of back of animal, it may cause injury to the animal. To tackle the problem, adjustable type saddle was fabricated as shown in Fig.3.

The fabricated saddles and harnesses were distributed to the five farmers of Bikaner using donkeys in the cart. The farmers were contacted and examined the status of animals at fortnightly intervals for two months periods and did not find any saddle sore or any discomfort to the animal. The measurements of equines used in pack load and carting were recorded and correlation will be estimated.

Pack load saddles for mules and donkeys

Mules and donkeys are mainly utilized as carting animals in plains of India except some activities where these are used as pack animals by dhobies, shephereds, brick kilns and in multistoried buildings. However, these animals are mainly used as pack animals in hilly terrains of India for transportation of bag and baggage of people, building material, ration,water, firewood and so many other activities. The condition of saddle used on pack animals is so poor that it causes wounds and injuries on the body of working equids. The saddle available with RVC is quite good and strong but is very heavy. We shall modify the saddle for use of field mules and donkeys as the mules used by army are heavier than field mules. We are in processes of replacing the heavy material with light weight material (composite material). After fabrication of suitable saddles for field mule and donkeys, we will test these at our farm and, if found suitable then we will promote these saddles in field.

(R.A. Legha and Yash Pal)

Determination of physico-chemical, properties of Halari donkey milk

Donkeys are primarily known as pack animals, reared mainly for work, and seldom milk. The composition of donkey milk is not studied in much detail in India due to sampling and other technical issues. Most donkeys in India are non-descript, however, three breeds: Halari, Spiti and Kachchhi were recently registered by ICAR-NBAGR, Karnal. For eons, the donkey milk was used in treating of cow milk protein allergy in infants due to the low casein contents. The research on donkey milk is in infancy in Indian donkey breeds especially. The current study was aimed to determine the physical and bio-chemical properties of milk and milk powder of Halari donkeys. The Halari donkey milk has characteristic taste, odour and appearance and off white in colour. In fresh milk the approximate milk fat is 1.13%, total milk solids 12.75%, and milk solids not fat are 11.62%. No pesticides residues and metal contaminants could be detected in milk samples. Under biological test parameters, the total plate count (TPC) was 88 cfu/ml and <10 cfu/mL of yeast and mould count.

Other important parameters of Halari donkey milk and lyophilised powder are listed in tables as follows.

The quality attributes of donkey milk were determined in terms of milk fat %, milk solids-not-fat (MSNF) %, ash % and total solids %. Average milk fat, ash and total solids content were found to be 0.2%, 0.39% and 8.2%, respectively. The rheological properties of donkey milk were determined in terms of flow behaviour property and temperature sweep. The effect of change in temperature (20-90°C) on the viscosity of donkey milk is shown in Fig1. Attempts were also made on understanding the heat coagulation temperature of donkey milk.

(Anuradha Bhardwaj, Yash Pal, R.A. Legha, R.K. Vaid, Varij Nayan, Narender Raju Panjagari, Ashish Kumar Singh and Rajan Sharma)

DONKEY MILK (HALARI)

CHEMICAL TEST PARAMETERS			
S.No.	**Parameters**	**Unit**	**Results**
1.	Milk Fat	%,m/m	1.13
2.	Milk Solids	%,m/m	12.75
3.	Milk Solids Not Fat	%,m/m	11.62
ORGANOLETPTIC TESTS			
S.No.	**Parameters**	**Unit**	**Results**
1.	Taste	-	characteristic
2.	Odour	-	characteristic
3.	Colour	-	Off white
4.	Appearance	-	liquid
BIOLOGICAL TEST PARAMETERS			
S.No.	**Parameters**	**Unit**	**Results**
5.	Total plate count	cfu/ml	88
CHEMICAL TEST PARAMETERS (NUTRITIONAL ANALYSIS)			
S.No.	**Parameters**	**Unit**	**Results**
1	Energy	Kcal/100g	43.23
2	Carbohydrates	g/100ml	6.89
3	Sugar	g/100ml	4.15
4	Fat	g/100ml	1.03
5	Protein (by Kjeldahl Method)	g/100ml	1.60
6	Vitamin B1	mg/100ml	0.406
7	Vitamin B5	mg/100ml	0.097
8	Lactose	g/100ml	6.84
9	Ash	%	0.31
10	Casein content	%	0.85
11	Titrable acidity	%	0.03

MINERALS			
S.No.	**Parameters**	**Unit**	**Results**
1	Magnesium	mg/100ml	7.65
2	Sodium	mg/100ml	6.33
3	Zink	mg/l	1.75
4	Calcium	mg/100ml	75.50
5	Potassium	mg/100ml	40.50
FATTY ACID PROFILE			
S.No.	**Parameters**	**Unit**	**Results**
1	Saturated fatty acid	g/100g	0.51
2	Polyunsaturated fatty acid	g/100g	0.11
3	Monounsaturated fatty acids	g/100g	0.41
4	Palmitic acid	g/100g	0.32
5	Oleic acid	g/100g	0.35
CHEMICAL TEST PARAMETERS (NUTRITIONAL ANALYSIS)			
S.No.	**Parameters**	**Unit**	**Results**
1	Moisture	%	90.17
2	Water activity	-	0.985

HALARI DONKEY MILK (POWDER)

CHEMICAL TEST PARAMETERS (ORGANOLEPTIC TESTS)			
S.No.	**Parameters**	**Unit**	**Results**
1	Taste	-	characteristic
2	Odour	-	characteristic
3	Colour	-	Off white
4	Appearance	-	Milk powder
BIOLOGICAL TEST PARAMETERS			
S.No.	**Parameters**	**Unit**	**Results**
1	Total plate count	cfu/g	667
CHEMICAL TEST PARAMETERS (FATTY ACID PROFILE)			
S.No.	**Parameters**	**Unit**	**Results**
1	Saturated fatty acid	g/100g	2.36
2	Polyunsaturated fatty acids	g/100g	0.51
3	Monounsaturated fatty acids	g/100g	1.86
4	Capric acid	g/100g	2.76
5	Lauric acid	g/100g	5.69
6	Myristic acid	g/100g	6.68
7	Pentadecanoic acid	g/100g	0.44
CHEMICAL TEST PARAMETERS (ORGANOLEPTIC TESTS)			
S.No.	**Parameters**	**Unit**	**Results**
1	Taste	-	characteristic
2	Odour	-	characteristic
3	Colour	-	Off white
4	Appearance	-	Milk powder

BIOLOGICAL TEST PARAMETERS			
S.No.	**Parameters**	**Unit**	**Results**
1	Total plate count	cfu/g	667
CHEMICAL TEST PARAMETERS (FATTY ACID PROFILE)			
S.No.	**Parameters**	**Unit**	**Results**
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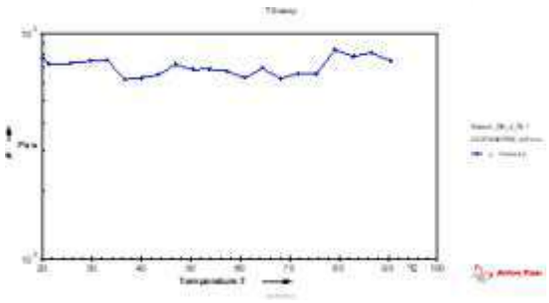
4	Capric acid	g/100g	2.76
5	Lauric acid	g/100g	5.69
6	Myristic acid	g/100g	6.68
7	Pentadecanoic acid	g/100g	0.44
8	Palmitic acid	g/100g	31.21
9	Palmitoleic acid	g/100g	4.97
10	Stearic acid	g/100g	3.21
11	Oleic acid	g/100g	34.26
12	Linoleic acid	g/100g	9.52
13	Gamma linolenic acid	g/100g	1.25



Pattern1. An illustration of the effect of change in temperature on the viscosity of donkey milk

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(Anuradha Bhardwaj, Yash Pal, R.A. Legha, R.K. Vaid, Varij Nayan, Narender Raju Panjagari, Ashish Kumar Singh and Rajan Sharma)

Genomic insights of indigenous equine resources through "*Omics*" approaches.

One of the major problems in sustainability of animals is the breed dilution and warrants the need to identify true to breed type animal. Though this can be done by breed descriptor but only pure breeds can be covered under it, ignoring the admixture in the population. The molecular markers are the revolutionizing tool which can be used for breed assignment as done in various domestic animals. By genotyping, allelic data of 24 microsatellite loci in 8 horse breed populations *viz*., Marwari (282), Kathiawari (69), Kachchhi-Sindhi (50), Thoroughbred (59), Zanskari (50), Bhutia (34), Spiti (16), and Manipuri (50) were generated. We got a total of 29280 DNA fingerprinting data generated which were formatted for computational analysis and breed prediction. After data pre-processing, different classifiers like BayesNet, NaiveBayes, Artificial Neural Network (ANN), Support Vector Machine (SVM) and Random Forest (RF) were deployed using scripts. The performance of these classifiers is represented in Table 1 which shows the classifier "BayesNet" as best having highest precision. The confusion matrix of BayesNet for each horse breed is presented in Table 2. The various evaluation measures, namely, sensitivity, specificity, precision, recall, F-measure and ROC area were computed for each of the eight breeds (Table 3).

Table 1. Performance of different classifiers

	TP Rate/Sen	FP Rate	Precision	Recall	F-Measure	ROC Area
BayesNet	0.889	0.010	0.919	0.889	0.887	0.991
NaiveBayes	0.630	0.035	0.635	0.630	0.604	0.873
MLP	0.741	0.037	0.731	0.741	0.734	0.927
SVM-Poly-2	0.774	0.036	0.778	0.774	0.774	0.932
RBF	0.690	0.038	0.707	0.690	0.679	0.910
RF	0.856	0.021	0.857	0.856	0.854	0.982

Table 2: Confusion matrix to show prediction power of BayesNet for each horse breed

*MG Marwari; KW Kathiawari; KS Kacchi Sindhi; TB Thoroughbred; MP Manipuri; SP Spiti; ZP Zanskari; BP Bhutia.

MG	KW	KS	TB	MP	SP	ZP	BP	Breeds*
281	0	0	1	0	0	0	0	MG
0	37	18	0	13	0	1	0	KW
0	0	50	0	0	0	0	0	KS
0	1	13	36	7	0	2	0	TB
0	0	0	0	45	0	4	1	MP
0	0	0	0	0	15	1	0	SP
0	0	0	0	6	0	44	0	ZP
0	0	0	0	0	0	0	34	BP

Table 3. Prediction accuracies obtained for eight breeds of horse

Breeds	TP Rate/Sen	FP Rate	Precision	Recall	F-Measure	ROC Area
MG	0.996	0.000	1.000	0.996	0.998	1.000
KW	0.536	0.002	0.974	0.536	0.692	0.937
KS	1.000	0.055	0.617	1.000	0.763	0.997
TB	0.610	0.002	0.973	0.610	0.750	0.997
MP	0.900	0.046	0.634	0.900	0.744	0.986
SP	0.938	0.000	1.000	0.938	0.968	1.000
ZP	0.880	0.014	0.846	0.880	0.863	0.994
BP	1.000	0.002	0.971	1.000	0.986	1.000
Weighted Avg.	0.889	0.010	0.919	0.889	0.887	0.991

(Anuradha Bhardwaj, Sarika, M.A. Iqbal, Varij Nayan, Dinesh Kumar, R.A. Legha and Yash Pal)

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Landmark Achievements

Year	Achievement
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	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
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

Technology Development, Transfer and Commercialization

The Centre has made focused efforts for the development of advanced technologies in equine health and production. Also, suitable methodologies were designed to uplift the livelihoods of equine keepers and all stakeholders of equine sector. Many diagnostic kits, vaccines and biologicals developed by the scientists of ICAR-NRCE are being used in the field while several other technologies are in pipeline.

Technologies developed and released

Vaccines

- Inactivated Equine Herpes Virus – 1 vaccine (Equiherpabort)
- Updated Equine Influenza vaccine

Diagnostic kits

- Equiherpes B-ELISA kit for diagnosis of EHV – 1 infection
- Recombinant antigen-based ELISA kit for diagnosis of *Theileria equi*
- LFA for diagnosis of equine piroplasmosis
- LFA for diagnosis of equine trypanosomosis
- Recombinant protein-based ELISA kit for diagnosis of Glanders
- Recombinant protein-based ELISA kit for diagnosis of EIA
- Recombinant gG-based type-specific ELISA for differentiation of EHV 1 and 4 infection
- Monoclonal antibody-based ELISA kit for diagnosis of rotavirus infection
- Japanese Encephalitis Virus Antibody Test Kit, iELISA for equids and pigs

Reproduction technologies

- Pregmare kit for pregnancy diagnosis in mares
- Cryopreservation of equine semen

Dairy technology

- Donkey Milk based products (Bathing soap, Body butter and Lip balm)

Technologies commercialized

- Recombinant protein-based ELISA kit for diagnosis of Glanders
- Recombinant protein-based ELISA kit for diagnosis of EIA
- Donkey Milk based products (Bathing soap, Body butter and Lip balm)

Release of Technology:

ICAR-NRCE has developed a diagnostic kit entitled “Japanese Encephalitis Virus Antibody Test Kit, iELISA for equids and pigs”. Since there is no indigenous kit available for testing equines for JEV, this kit is useful in rapid and specific diagnosis of Japanese Encephalitis in equines. The kit contains sufficient reagents to test 46 samples in duplicate. Rapid test results can be obtained within 3 hours and the shelf life of the kit is six months at 2-8°C.

Release of "JE Virus Antibody Test Kit" by Union minister of Agriculture & Farmers' Welfare

Japanese Encephalitis Virus Antibody Test Kit

The kit was released by Hon'ble Union Minister of Agriculture & Farmers' Welfare Shri Narendra Singh Tomar Ji & other dignitaries on 27th February, 2020 at NASC Complex, New Delhi during the 91st Annual General Meeting of ICAR.

Transfer of Technology:

- **"Donkey Milk Based Products (Bathing Soap, Body Butter and Lip Balm)"**

A technology entitled, *"Donkey Milk Based Products (Bathing Soap, Body Butter and Lip Balm)"* was transferred to M/s Dolphin IBA Pvt. Ltd., Ramamangalam P.O., Kochi on 12th February, 2020.

Donkey milk technology transferred to M/s Dolphin IBA Pvt. Ltd

- **Multipurpose iron plough**

The Centre has developed multipurpose iron plough for working equids. Technology was transferred to farmers of Eta district of UP through Brooke India. The farmers are having small land holding and they are facing problem of connection to their field for tractor operated implements. Now they are able to cultivate their fields by equine operated plough, because they can reach their fields with equine and plough on their shoulder along with field bunds of other owners.

Multipurpose iron plough for equines

Technology ready for transfer:

- **Mushroom cultivation with equine dung**

Mushroom cultivation in different combinations of fresh equine dung and wheat straw was undertaken. Encouraging results are being received in both combinations. EPC, Bikaner has produced 35.25 kg button mushroom and sold @ Rs 120/ kg. A total of Rs 4230/- revenue was generated through mushroom sale in 2020.

Mushroom cultivation using equine dung

Patent granted

S. No.	Title	Name of Inventors	Application detail
1.	Recombinant TssA protein for detection of antibodies against *Burkholderia* mallei and uses thereof	Hari Shankar Singha, Praveen Malik, Sachin Kumar Goyal, S.K. Khurana and R.K. Singh	Patent No. 354870, dated 30.12.2020, Application No. 3610/DEL/2015, dated 04.11.2015

MoU for cooperation in Research and Education

ICAR-NRCE, Hisar inked Memorandum of Understanding (MoU) with four universities in 2020.

- Shri Krishna Ayush University, Kurukshetra, Haryana.
- Rajasthan University of Veterinary and Animal Sciences, Bikaner, Rajasthan.
- Maharaja Agrasen University, Solan, Himachal Pradesh.
- D. Y. Patil Education Society (Deemed to be University), Kolhapur, Maharashtra.

MoU with Shri Krishna AYUSH University, Kurukshetra

MoU with Rajasthan University of Veterinary and Animal Sciences, Bikaner, Rajasthan

Revenue generation from samples tested during the year 2020

During the year 2020, diagnostic services were provided to stakeholders from different states of India. A total of 6775 samples received from race courses, turf club, stud farm, riding schools and other organized sector were tested on payment basis. Maximum number of samples was tested for Glanders (3316) and EIA (2737). The revenue generated through contractual diagnostic services was Rs 49.10 lakhs in the year 2020.

Equine samples tested and revenue generated in 2020

Disease	No. of samples tested	Amount (Rs.)
Equine Infectious Anaemia (EIA)	2737	1505350
Glanders	3316	2321200
Contagious Equine Metritis	260	416000
Dourine	139	152900
African Horse Sickness	139	152900
Equine Viral Arteritis	130	260000
Theileria equi	20	40000
Babesia caballi	20	40000
WNV	9	18000
Trypanosoma evansi	2	1100
Culture isolation	3	3000
Total	6775	4910450

Education and Trainings

Trainings attended in 2020

ICAR-National Research Centre on Equines encourages its staff for capacity building in advanced areas of science, administration and skill development. In the year 2020, following trainings were obtained by the staff of ICAR-NRCE.

S. No.	Name of the staff	Designation	Training	Organizing Institute	Duration of the training
1.	Ms. Ana Raj	Scientist	Professional Attachment Training (PAT)	ICAR-CTCRI, Thiruvananthapuram	21st May to 18th August, 2020
2.	Ms. Ana Raj	Scientist	MOOC on "Innovative ideas for Entrepreneurship development in Livestock sector"	MANAGE, Hyderabad	14th to 21st September, 2020
3.	Dr. Anju Manuja	Principal Scientist	ICAR Research Data Repository for Knowledge management	ICAR-CIRB, Hisar	26th to 27th February, 2020
4.	Dr. Anubha Pathak	Scientist	Professional Attachment Training (PAT)	ICAR-NIHSAD, Bhopal	21st May to 18th August, 2020
5.	Dr. Anuradha Bhardwaj	Senior Scientist	Workshop-cum-training on "IPRs in Agricultural Research & Education in India"	NAHEP-IPTM Unit of ICAR	12th to 28th September, 2020
6.	Dr. Balvinder Kumar	Principal Scientist	Online EDP on "ABS Regulations and Nagoya protocol"	ICAR-NAARM, Hyderabad	15th to 17th July, 2020
7.	Dr. H.S Singha	Senior Scientist	Newer approaches in disease diagnosis and vaccines for livestock and poultry	ICAR-NRCE, Hisar	28th January to 6th February, 2020
8.	Dr. H.S Singha	Senior Scientist	ICAR Research Data Repository for Knowledge Management	ICAR-CIRB, Hisar	26th to 27th February, 2020
9.	Dr. Naveen Kumar	Principal Scientist	National Multisectoral One-Health Workshop for Prevention of Zoonotic Diseases	Government of Rajasthan	10th to 12th February, 2020
10.	Dr. Nitin Virmani	Principal Scientist	Online training of Vigilance officers	ICAR-NAARM, Hyderabad	5th to 7th August, 2020
11.	Dr. Nitin Virmani	Principal Scientist	Flow Cytometry Techniques & Applications	Indian Institute of Technology (IIT), Guwahati	21st to 22nd December, 2020

12.	Dr. Yash Pal	Principal Scientist	Virtual workshop-cum-training on "Intellectual Property Rights in Agricultural Research & Education in India"	ICAR Headquarters, New Delhi (NAHEP and IPTM)	12th to 28th September, 2020
13.	Dr. Rajender Kumar	Principal Scientist	Virtual workshop-cum-training on "Intellectual Property Rights in Agricultural Research & Education in India"	ICAR Headquarters, New Delhi (NAHEP and IPTM)	12th to 28th September, 2020
14.	Dr. Shanmugasundaram K.	Senior Scientist	Newer approaches in disease diagnosis and vaccines for livestock and poultry	ICAR-NRCE, Hisar	28th January to 6th February, 2020
15.	Dr. Shanmugasundaram K.	Senior Scientist	Strategies for sustainable control of parasites of livestock, poultry and wildlife and their public health significance	LUVAS, Hisar	21st to 23rd August, 2020
16.	Dr. T.R Talluri	Senior Scientist	Online training cum webinar on "Applications of Flow Cytometry in Semen Analysis"	ICAR-NDRI, Bengaluru and Beckman Coulter Life sciences	21st to 22nd July, 2020
17.	Dr. T.R Talluri	Senior Scientist	Online training on "Advanced Bioinformatics tools and its applications in Agriculture"	ICAR-NAARM, Hyderabad	7th to 11th December, 2020

Trainings imparted in 2020

Training cum exposure visit on use of equine operated agricultural implements

A two days training cum exposure visit on "Use of equine operated agricultural implements" under the "AICRP on Utilization of Animal Energy" has been organized for the farmers at Equine Production Campus, Bikaner from 14th to 15th January 2020. A total of 10 farmers and one blacksmith from Eta District of Uttar Pradesh and 5 officers from Brooke India, took part in this training cum exposure visit programme. Farmers have been imparted with practical training on developing, designing and working with equine operated agricultural implements like different types of plough and seed drillers developed by NRCE under AICRP on UAE project.

Exposure visit of trainees under AICRP on UAE

Practical training on equine operated agricultural implements

ICAR Sponsored Short Course on "Newer Approaches in Disease Diagnosis and Vaccines" for livestock and poultry

A short course on "Newer Approaches in Disease Diagnosis and Vaccines for livestock and poultry" was organized from 28th January to 6th February, 2020. This short course was conducted to provide in-depth knowledge of the concepts and hands-on exposure to participants dealing with diagnostics and vaccine development for livestock and poultry diseases. A total of 14 participants including faculty from various ICAR institutes and State Agricultural and Veterinary universities participated in the course.

Participants of ICAR sponsored short course

The comprehensive list of trainings organized by ICAR – NRCE in 2020 is given below in chronological order.

S. No.	Name of the training	Duration	No. of participants
1.	Training on AI and Ultrasonography in mares to final year BVSc & AH students of College of Veterinary and Animal Science, RAJUVAS, Bikaner	8th to 14th January, 2020	60
2.	Training cum exposure visit of farmers and officials of Brooke India under the "AICRP on Utilisation of Animal Energy"	14th to 15th January, 2020	16
3.	Training on "Sewing, Cutting and Stitching" under SCSP programme in collaboration with PNB Farmer Training Centre, Sacha Khera	20th January to 7th February, 2020	35
4.	ICAR Sponsored Short Course on Newer Approaches in Disease Diagnosis and Vaccines for livestock and poultry	28th January to 6th February, 2020	14
5.	Training on "Fruit Preservation" under SCSP programme in collaboration with PNB Farmer Training Centre, Sacha Khera	3rd to 11th February, 2020	35
6.	Training on "Dairy Farming" under SCSP programme in collaboration with PNB Farmer Training Centre, Sacha Khera	3rd to 9th March, 2020	35
7.	Training on "Tailoring and Cutting" under SCSP programme in collaboration with Saina Nehwal Institute for Agricultural Technology, Training & Education, CCS HAU, Hisar	15th to 19th March, 2020	30
8.	Training on "Baking and Bakery Products for Value Addition of Cereals & Pulses" under SCSP programme	15th to 19th March, 2020	30
9.	One month sponsored laboratory training course for Mr Ankush Dhillon, M Sc student, Dept. of Microbiology, Guru Jambheshwar University of Science and Technology, Hisar	11th September to 10th October, 2020	1
10.	Training on AI and Ultrasonography in mares to internees BVSc & AH students of Arawali Veterinary College, Sikar	9th to 16th December, 2020	40

Skill training under SCSP programme

Training on AI and Ultrasonography in mares

Post Graduate Students' Research and Guidance

S. No.	Name of the Student	Name of the Guide	Title of the Thesis
Ph D Students			
1.	Aashwina Madhwal ICAR –IVRI, Izatnagar, UP	Dr. Nitin Virmani	Development of modified live EHV-1 vectored bivalent vaccine candidate against equine influenza virus and equine herpes virus: Protective efficacy studies in mouse challenge model
2.	Alka Nokhwal CCS HAU, Hisar, Haryana	Dr. R.K Vaid	Isolation and characterization of bacteriophages against aeromonads from fish culture ponds
3.	Anubala CCS HAU, Hisar, Haryana	Dr. Taruna Anand	Exploring bacteriophage derived endolysins for targeted delivery into biofilm forming bacteria
4.	Dinesh Jhamb, RAJUVAS, Bikaner, Rajasthan	Dr. T.R Talluri	Effect of L-arginine and trehalose supplementation to the semen extender on quality and fertility of cryopreserved stallion semen
5.	Medhavi Vashisth CCS HAU, Hisar, Haryana	Dr. Taruna Anand	Characterization of bacteriophages against ESKAPE pathogens and assessment of their synergy with antibiotics
6.	Nitin Khandelwal GLA University, Mathura, UP	Dr. Naveen Kumar	Studies on the antiviral activity of Apigenin against buffalopox virus
7.	Ram Kumar RAJUVAS, Bikaner, Rajasthan	Dr. Naveen Kumar	Studies on the role of ROCK signaling pathway in buffalopox virus replication
8.	Snehil Gupta LUVAS, Hisar, Haryana	Dr. Rajender Kumar	Screening, identification and evaluation of some novel target specific therapeutic compounds against *Tryanosoma evansi*
9.	Stephanie S Pradhan ICAR –IVRI, Izatnagar, UP	Dr. Nitin Virmani	Comparative pathogenicity and immunogenicity of modified live EHV – 1 vaccine candidate(s) in mouse model and development of gE protein based ELISA for differentiation of vaccinated and infected animals
10.	V.K. Pal COVS & AH, Kumarganj, Ayodhya, UP	Dr. Rajender Kumar	Molecular characterization and sero-prevalence studies on equine haemoprotozoan diseases with special reference to *Trypanosoma evansi*
11.	Yogesh Chander GJU S&T, Hisar, Haryana	Dr. Sanjay Barua	Role of p38 MAP kinase in buffalopox virus replication

M V Sc/ M Sc Students			
1.	Diksha Sharma IVRI, Izatnagar, UP	Dr. Rajender Kumar	Epidemiological studies of *Trypanosoma evansi* in cattle, buffaloes and equines of Himachal Pradesh and characterization of immunodominant antigens
2.	Prashant Kadiyan LPU, Jalandhar, Punjab	Dr. Anju Manuja	Antibiotic Resistance and antibacterial activity of zinc oxide nanoparticles against Indian isolates of *Streptococcus equi*
3.	Priyanka GJU S&T, Hisar, Haryana	Dr. Taruna Anand	Isolation and characterization of host specific bacteriophages against *Pseudomonas spp.*
4.	Rajendra Mehra RAJUVAS, Bikaner, Rajasthan	Dr. T.R Talluri	Effect of Addition of Lyophilized heterologous seminal plasma and colostrum to semen extender on cooled and post-thaw Stallion semen quality
5.	Supriya ICAR –IVRI, Izatnagar, UP	Dr. Nitin Virmani	Attenuation of recombinant EHV-1 through deletion of glycoprotein 1 and its pathological and immunological study in mice for modified live vaccine candidate
6.	Tipu Sultan RAJUVAS, Bikaner, Rajasthan	Dr. T.R Talluri	Effect of zinc and gold nanoparticles on cooled and post thaw quality of stallion
Post-doctoral Research Associate			
1.	Dr Ruma Rani Sponsored by CSIR, New Delhi	Dr. Rajender Kumar	Identification and evaluation of target specific novel drug molecules against *Trypanosoma evansi* infection using nanotechnology approach

Workshops, Seminars and Institutional activities

Webinars organized in 2020 at ICAR-NRCE

S. No.	Name of the training	Date
1.	International webinar on "Using phages to combat AMR: Scientific capacity building through laboratory training workshops" in collaboration with Acharya Narendra Dev College, University of Delhi	14th August, 2020
2.	International webinar on "Phage Directory: Making the world's phages more accessible, manageable and shareable" in collaboration with Acharya Narendra Dev College, University of Delhi	3rd September, 2020
3.	International webinar on "Knowing the unknown: Approaches to characterize the uncharacterized proteome of bacteriophages" in collaboration with Acharya Narendra Dev College, University of Delhi	1st October, 2020
4.	Webinar on "Collaboration between Medical and Veterinary officers for the prevention and control of zoonotic diseases"	4th November, 2020

Events and celebrations of the year 2020 at ICAR-NRCE, Hisar and ICAR-NRCE-EPC, Bikaner

Republic Day

At ICAR-NRCE, Republic day was celebrated with pride and enthusiasm. Director, Dr B.N.Tripathi hoisted the national flag and gave an enlightening lecture on patriotism and encouraged members of staff for achieving greater scientific heights for the nation. At EPC Bikaner, the Institutional Awards for best performance were given to Dr. T. Rao, Senior Scientist and Dr.R.A.Pachori, ACTO. On behalf of Rajasthan Tourism Department Dr. Jitendar Singh, Sh. Kamal Singh and the winners of Horse Race, organised on the occasion of Camel Festival, were given mementos. The Campus awards were given to Sh. Narendra Chauhan, Sh. Om Prakash, Sh. S. N. Paswan and Sh. Raju Ram. On this occasion, mementos were also given to the staff working in Reproduction Laboratory, which were sponsored by Dr. T. Rao. Horse Race, Kabbadi Match and Tug-of-war was also organised to mark the occasion.

Farewell to Dr. Bhupendra Nath Tripathi

On February 9th 2020, ICAR-NRCE bid farewell to Director, Dr. B.N. Tripathi who was appointed as DDG, Animal Science at ICAR Head Quarters, Krishi Bhawan, New Delhi. Bidding adieu to one of the most beloved directors of the Centre was an emotional

Republic Day Celebrations ICAR-NRCE, Hisar

moment for the employees and fellow colleagues of NRCE. Principal Scientist (Rtd), Dr. B.K. Singh and Principal Scientist (Retd.) Dr. Attar Singh along with present Director, Dr. Yash Pal, Dr. Sanjay Barua I/C NCVTC, and Principal Scientist, Dr. B.R. Gulati graced the occasion to present a token of love to Dr. B.N. Tripathi on behalf of the NRCE family.

Dr B.N. Tripathi (Centre) being presented with a token of love from NRCE family

Pusa Krishi Vigyan Mela and Health Camps

From March 1-3, 2020, Krishi Vigyan Mela was held at Indian Agricultural Research Institute Campus, IARI, New Delhi, The stall of ICAR NRCE had a total of 700 visitors including prospective and present Equine breeders during the mela. The equine owners were provided the information on major equine diseases like equine Influenza, Equine Herpes Virus, Japanese Encephalitis, Glanders, Strangles, and various parasitic diseases. The farmers were also enlightened on the utilization of donkey milk and milk products.

Director Dr Yash Pal and Principal Scientist, Dr Anju Manuja guiding the equine owners

International Women's day

On March 8th 2020, ICAR-NRCE staff and students gathered to celebrate the progress towards achieving gender equality and women's empowerment. A total of 15 participants, from scientific, administrative and supporting staff participated in the event. The celebration was chaired by Director NRCE, Dr Yash Pal, he discussed the accomplishments of women in the scientific arena focused on striving a greater momentum towards gender equality.

Women Scientists, administrative staff and supporting staff with Director, NRCE on Women's day

Independence day

Centre celebrated Independence Day with gaiety and patriotic passion on 15th August, 2020. In the front yard of ICAR-NRCE, the national flag of India was hoisted with verve by Dr. Yash Pal, Director NRCE. Independence Day Celebration at NRCE inspired the staff and families by commemorating a series of incidents in the history of India for the attainment of independence, making India independent country. To mark the day, children of the employees took pride in glorifying and celebrating the spirit of unity. Director congratulated the staff and inspired the staff to accomplish the new horizons in their scientific endeavour. The Institute reverberated with patriotic fervor and enthusiasm.

केन्द्र में आयोजित हिन्दी सप्ताह

भारत सरकार की राजभाषा की विकासनीति के अंतर्गत गत वर्षों की भांति इस वर्ष भी राजभाषा कार्यन्वयन समिति के तत्वावधान में राजभाषा के प्रचार–प्रसार एवं सरकारी काम–काज में हिन्दी के प्रयोग को अधिकाधिक प्रोत्साहित करने के लिए राष्ट्रीय अश्व अनुसंधान केन्द्र में हिन्दी सप्ताह का आयोजन डॉ. यश पाल, निदेशक की अध्यक्षता में 14 से 21 सितम्बर, 2020 तक किया गया। कार्यक्रम के प्रथम दिन व हिन्दी दिवस के अवसर पर एक हिन्दी कार्यशाला का आयोजन किया गया। डॉ. यश पाल ने हिन्दी कार्याशाला के उदघाटन समारोह में उपस्थित हुए सभी अतिथिगणों और केन्द्र के सभी अधिकारियों एवं कर्मचारियों का संबोधन किया। इस कार्यक्रम में डॉ. हरिदत्त कौशिक, सेवानिवृत प्रोफैसर, चौधरी चरण सिंह कृषि विश्वविद्यालय मुख्य वक्ता के रूप में उपस्थित थे। इस अवसर पर अपने वक्तव्य में केन्द्र के सभी वैज्ञानिकों एवं कर्मचारियों को संबोधित करते हुए मुख्य वक्ता डॉ. हरिदत्त कौशिक ने हिन्दी को एक बृहद भाषा के रूप में परिभाषित करते हुए इसे एक सहज, सरल व आमजन की भाषा बताया जिसमें सभी भाषाओं के

शब्दों के समावेश की अनुपम क्षमता है। उन्होंने हिन्दी भाषा में प्रयुक्त होने वाले विभिन्न शब्दों व उनके सुचित अर्थों के बारे में भी विस्तृत जानकारी प्रदान की। कार्यक्रम के अध्यक्ष व मुख्य अतिथि डॉ. यशपाल ने अपने संबोधन में हिन्दी को एक प्रभावशाली भाषा बताया तथा केवल हिन्दी दिवस या हिन्दी सप्ताह के अवसर पर ही नहीं वरन् पूरे वर्ष हिन्दी को अंगीकर कर अपना अधिक से अधिक कार्य हिन्दी में निश्पादित करने हेतु प्रोत्साहित किया। डॉ. अनुराधा ने हिन्दी सप्ताह के दौरान आयोजित किए जाने वाले विभिन्न कार्यक्रमों की जानकारी दी व साथ ही सभागार में उपस्थित सभी अतिथियों व केन्द्र के वैज्ञानिकों व कर्मचारियों का इस कार्यक्रम में भाग लेने हेतु आभार भी व्यक्त किया। इस दौरान विभिन्न ज्ञानवर्धक एवं रूचिपूर्ण हिन्दी प्रतियोगिताओं क्रमशः हिन्दी परिच्छेद अनुवाद प्रतियोगिता, हिन्दी श्रुत लेख प्रतियोगिता, हिन्दी टंकण, हिन्दी कविता पाठ प्रतियोगिताओं आदि का आयोजन किया गया। दिनांक 21–09–2020 को समापन समारोह में इस केन्द्र के निदेशक महोदय डॉ. यश पाल ने अपने सम्बोधन में सभी कर्मचारियों को अपना अधिक से अधिक कार्य राजभाषा में करने के लिए प्रोत्साहित किया। उन्होंने यह भी कहा कि किसी भी देश की प्रगति में उसकी अपनी भाषा का अहम् योगदान होता है। इसके साथ उन्होंने यह भी कहा कि दूसरी भाषाओं का भी ज्ञान होना देश हित में होता है। निदेशक महोद्य ने हिन्दी सप्ताह में कर्मचारियों की प्रतिभागिता की सराहना की और उन्हें पुरस्कार देकर प्रोत्साहित किया। केन्द्र के निदेशक महोद्य डॉ. यश पाल ने संस्थान के कर्मिकों को अधिकाधिक हिन्दी में कार्य करने के लिए प्रेरित किया। इस समापन समारोह में मुख्य अतिथि के तौर पर हरियाणा विद्यालय शिक्षा बोर्ड के अध्यक्ष डॉ. जगबीर सिंह ने अपने सम्बोधन में कर्मचारियों को सम्बोधित करते हुए हिन्दी को आमजन की भाषा बताया और उन्होंने कहा आजादी मिलने के बाद, देश में अंग्रेजी के बढ़ते उपयोग और हिन्दी के बहिष्कार को देखते हुए सरकार ने हिन्दी दिवस मनाने का निर्णय लिया था। लेकिन अब अधिकतर हिन्दी में ही कार्य किया जा रहा है। डा. अनुराधा भारद्वाज, वरिष्ठ वैज्ञानिक व हिन्दी अधिकारी ने बताया कि इस समारोह में चन्दन साहित्य मंच हिसार के माननीय श्री महेन्द्र जैन ने इस केन्द्र को हिन्दी के उन्नयन के विशेष योगदान हेतू समृति चिन्ह् भेट कर प्रोत्साहित किया। कार्यक्रम के दौरान कोविड–19 के कारण सामाजिक दूरी एवं मास्क का प्रयोग किया गया तथा सब नियमों का पालन करते हुए क्रार्यक्रम को सुचारू रूप से संचालित किया गया।

मुख्य अतिथि डॉ. जगबीर सिंह, चन्दन साहित्य मंच के कवियों के साथ

डॉ. हरिदत्त कौशिक, हिन्दी कार्याशाला के उद्घाटन समारोह के दौरान

अश्व उत्पादन परिसर बीकानेर में हिंदी दिवस का आयोजन

अश्व उत्पादन परिसर बीकानेर में राजभाषा कार्यशाला का आयोजन किया गया जिसमें मुख्य हिंदी वक्ता डॉ. गौरव बिस्सा एसोसिएट प्रोफेसर मैनेजमेंट स्टडीज गवर्नमेंट इंजीनियरिंग कालेज बीकानेर ने "नैतिकता और प्रशासन " पर व्याख्यान प्रस्तुत किया। डॉ. यशपाल निदेशक–भा.कृ.अनु.प. – राष्ट्रीय अश्व अनुसंधान केन्द्र हिसार मुख्य अतिथि के रूप में उपस्थित हुए। उन्होंने वैज्ञानिक व अधिकारियों को अधिकाधिक कार्य हिंदी में करने के लिए प्रेरित किया। उन्होंने कहा की मात्रभाषा में अपने विचार व्यक्त करना आसान है।

Mahatma Gandhi Jayanti Week

Mahatma Gandhi Week was celebrated at ICAR-NRCE from 29th September to 02nd October 2020. Programs based on the life and teachings of Gandhiji were organized during this week. An essay competition and quiz competition on the topic "Mahatma Gandhi - Truth and Non-violence" was conducted. ICAR-NRCE Director, Dr. Yash Pal, gave the message of applying the teachings of Mahatma Gandhi in daily life while Principal scientist Dr. R.K. Vaid gave an informative lecture on the life of Mahatma Gandhi. Dr. Mamta Kakkar, Chairperson, Department of Hindi and Sanskrit, Dayanand, Mahavidyalaya, Hisar, presented a statement on the online platform of Google Meet on the title "Saint of Sabarmati"

Digital Quiz competition held during Gandhi week at ICAR-NRCE

Vigilance Awareness Week

The vigilance awareness week was celebrated on October 27th, 2020. For celebration of the vigilance awareness week, the oath for vigilance awareness was taken by the employees of the institute via online platform. The oath was taken under the leadership of Director ICAR-NRCE, Dr. Yash Pal.

Oath taking ceremony during vigilance awareness week at ICAR-NRCE

Awareness creation about Fundamental Rights and Duties of citizens

In lieu of culminating the "Ambedkar Jayanti", a lecture was arranged on "Fundamental Rights and Duties" to spread awareness about the various rights and duties of the citizen among staff of ICAR-NRCE, Hisar on 6th November, 2020. The lecture was delivered by Dr. Yash Pal, Director, ICAR-NRCE using online zoom platform. All scientific, administrative and contractual staff members of ICAR-NRCE attended the lecture. Dr. Yash Pal has vast experience on the research and legal aspects as he has also pursued LLB (Bachelor of Legislative Law) degree. Dr. Yash Pal has elaborated our "Fundamental rights and duties" as per Constitution of India. He emphasized on the need for every citizen to take his duties towards the nation as a whole. He pointed out the various fundamental rights relating to life, liberty, equality and freedom of expression *etc.* In his closing speech, he urged staff members of the institute to follow the fundamental rights and their duties for the better upliftment of the institute and nation as a whole.

Foundation Day

36th Foundation of ICAR-NRCE was celebrated on Nov 26th 2020, As COVID-19 had firmly gripped the country, the celebrations were conducted on online platform. Dr. B.N. Tripathi, DDG, Animal Science, ICAR, presided over the function as the chief guest. Director ICAR-NRCE, Dr. Yash Pal, Director ICAR-NRCE, Dr. V.P. Singh, Director ICAR-NIHSAD Dr. A.K. Tomar, Director ICAR-CSWRI and Dr. Jyoti Misri from ICAR headquarters graced the occasion with all the staff and members of NRCE family.

Swachhta Pakhwada

Swachhta pakhwada was enthusiastically celebrated at ICAR-NRCE from December 16th to December 21st 2020 with multiple activities promoting cleanliness in and around the campus. Awareness lectures on recycling of wastewater, water harvesting, kitchen gardening in residential colonies was organized. Members from NRCE residential colonies participated in the event. Quiz, essay and drawing competitions for school children, village youth were also organized in NRCE Hisar and EPC Bikaner. In MGMG village Bhojoosar, the public places such as Village Centre, Community Centre, Temple Campus, School Premises, Public Distribution Centre (PDS) were visited and the persons maintaining best cleanliness (without taking any money) were awarded. At ICAR-NRCE Bikaner, programme on swachhata pakhwada and farmers day was organized at selected village under MGMG (Kotari, Rajasthan). A total of twenty farmers attended the meeting in which Dr. R. K. Dedar gave lecture on farmer's day and awareness about different activities of farming. Dr. R. A. Legha delivered lecture on cleanliness and Swachhta of village and animals. Farmers were asked to practise cleaning of village routinely and use of toilets.

Lecture on fundamental rights and duties of citizens at NRCE auditorium

Virtual Foundation day celebration at ICAR-NRCE

Pledge for cleanliness at ICAR-NRCE campus

Activities under "Mera Goan Mera Gaurav" Programme

ICAR-NRCE constituted six teams of scientists for "Mera Gaon Mera Gaurav" related activities in selected villages. A total of six villages (four in Haryana and two in Rajasthan) were adopted by ICAR-NRCE. The various activities were undertaken in Belssar, Bhojoosar, Sinthal villages of Bikaner districts of Rajasthan and Dhandoor Village of Hisar, Haryana. The scientists of the Centre made efforts to coordinate agriculture, animal health related activities and social awareness programme through developing linkages with government officials (Agriculture Development Officers, Veterinary Officers and Anganwari officials & workers) and local village Panchayats. The teams organized various scientific activities *viz*., animal health camps, infertility camps, interface meetings and students programme. Baseline survey of the village Bhojoosar, Bikaner was also done. Distribution of medicines was also carried out. General awareness was created among the villagers about COVID-19 Pandemic, and also on the importance of child and women nutrition. The advisory was provided for Swachchha Bharat. A total of 100 farmers and 27 anganwari workers, 1121 animals including cows, bulls/bullocks, calves, buffaloes, heifers, sheep, goats, camels, horses, mares, etc. were benefitted. Farm management practices were observed and necessary suggestions were given. Scientists also addressed the issues of infertility and malnutrition in the livestock. The effective control of ectoparasites and its importance was deliberated. Good quality mineral mixture, deworming boluses were distributed.

Mahila Kisan Diwas was celebrated at Village Dhandoor, Hisar in the month of October, 2020. The programme was conducted with social distancing and taking other precautionary measures for COVID-19. The 27 anganwadi workers from six villages (Kajla, Mallapur, Jakhod khera, Jhiri, Dhandoor and Bir Babran) tested negative for COVID-19 by state government participated in the event. The focus of the programme was on nutrition of villagers especially women and children with a tagline "Sahi potion and desh rotion". NRCE distributed prizes to the winners of the low cost nutritious recipes of Bajra and anganwadi workers participating in the event.

NRCE staff organized infertility and health camps at Sinthal, Bikaner district, Rajasthan

Medicine distribution to equines owners in Equine Health Camp at Bhojoosar, Bikaner District, Rajasthan

Awareness programme on COVID-19 at Bhojoosar, Bikaner District, Rajasthan

Awareness programme on COVID-19 and Animal health at Belasar, Bikaner district , Rajasthan

NRCE scientist interacting with Anganwadi workers on Mahila Kisan Diwas

Prize distribution by NRCE staff to the winners of low cost nutritious recipes of bajra (pearl millet)

IRC, RAC and Research Review Meetings

Annual Institute Research Committee (IRC) Meeting

The annual IRC meeting of ICAR-NRCE was held on 21-22 & 31 July, 2020 under the Chairmanship of Dr. Yash Pal, Director, ICAR-NRCE. A total of 33 research projects and 3 new concept notes were discussed in the meeting. The Chairman suggested that scientists should come up with new hypotheses based on rigorous brain storming and more emphasis needs to be given on formulation of basic research projects. He expressed satisfaction on the progress made by the institute during the period under report and welcomed suggestions from the scientists for further improvement of the research work in the institute.

The Chairman appreciated the scientists for good research work and active participation in COVID-19 testing at ICAR-NRCE, Hisar. He requested all the scientists to share their research facilities, manpower and reagents in anticipation of a budget cut due to COVID-19 pandemic. He further stressed that efforts should be made to release new technologies this year. The Chairman encouraged all the scientists to apply for externally funded research projects and also to publish research papers in high impact journals.

23rd Research Advisory Committee (RAC) meeting of ICAR- NRCE

The 23rd RAC meeting of ICAR- National Research Centre on Equines was held through video conferencing under the Chairmanship of Dr. M P Yadav (Former Vice Chancellor & Director, IVRI) on 09th July, 2020 to review the research for the year 2019-20. Dr. Yash Pal, Director, ICAR-NRCE gave a brief presentation on the overall achievements of the Centre for the year 2019-20 which was followed by presentations on equine health, equine production and NCVTC. The key recommendations of the committee included (1) Establishment of a donkey farm at the Centre, (2) Evaluation of donkey milk constituents for medicinal, microbiological and nutraceutical properties, (3) Assessment of the impact of disease diagnostics and technologies developed by NRCE at field level, (4) Initiating the processes for achieving disease free status for Equine Infectious Anemia, (5) Giving more emphasis to the basic research and research on bacteriophages, (6) Establishment of nucleus units of Bhutia, Spiti, Kachchhi-Sindhi breeds of horses at ICAR-NRCE for their conservation, (7) Regular meetings with stakeholders for popularization of equine keeping and dissemination of technologies developed by NRCE. In his concluding remarks the Chairman urged the scientists to work hard for the upliftment of equine sector in the country and also to explore value addition, so that equine keeping can be more remunerative and attractive to the farmers.

RAC meeting of ICAR-NRCE in progress

Director ICAR-NRCE also presented the action taken reports on the recommendations of previous RAC meeting. He also apprised about COVID-19 testing and the technologies developed and released by the institute. Action taken report was thoroughly discussed and the Chairman expressed his satisfaction on the initiatives taken by ICAR-NRCE based on the RAC recommendations. The chairman appreciated the contribution of the scientists for achievements of the Centre in the field of Research and Development.

Half yearly Institute Research Committee Meeting

Half yearly IRC meeting of ICAR-NRCE was held on 18th to 19th and 22nd December, 2020 under the chairmanship of Director, ICAR-NRCE, Hisar. In this meeting, 31 research projects from NRCE/NCVTC & EPC including 11 externally funded projects were discussed. Besides these projects, five concept notes were also presented and discussed. The chairman suggested that scientists should limit their role as Co-PI in 2-3 projects. It was suggested to elaborate the results while submitting IRC report to PME Cell. Finally, chairman asked all the scientists to submit research project proposal reports and monthly progress reports on time. It was also requested to submit a copy of the annual progress report of externally funded projects to PME cell for records.

IRC meeting of ICAR-NRCE in progress

Quinquennial Review Team (QRT) meetings

Quinquennial Review Team (QRT) under the Chairmanship of Maj. Gen. (Dr.) Shri Kant Sharma SM, VSM (Retd.), former Vice Chancellor, LUVAS, Hisar reviewed the work done by ICAR-NRC on Equines, Hisar for the period from April 2013 to March, 2018. The QRT constituted by the Council had members from diversified spectra of Veterinary and Animal Science Education and Research, Economics and Management. The Chairman had a vast experience in Equine Health and Production during his distinguished services in Remount Veterinary Corps (RVC). The QRT held five meetings to review the progress of research work at ICAR-NRCE, Hisar, NCVTC and EPC, Bikaner. In addition, a meeting with stakeholders was held at Directorate General RVS, IHQ of MOD (Army) at RK Puram, New Delhi in which DG RVS, Additional DG RVS, Director RVS (Tech), Commissioner Animal Husbandry (GoI), ADG (AH) ICAR, Representatives of Race Club, Brooke India and the Chairman, and members of the QRT participated to discuss the expectations of the stakeholders. The report was discussed in the IMC on 15th July, 2020 and presented to Honourable Director General, ICAR & Secy. DARE on 29th October, 2020.

Quinquennial Review Team (QRT) meeting 2020

Annual Scientific Review meet of NCVTC

The tenth Annual Scientific Review meet of National Centre for Veterinary Type Cultures (NCVTC) was held on 20 November, 2020 online via zoom platform. Dr. B.N. Tripathi (DDG, Animal Sciences), Dr. Ashok Kumar, ADG (Animal Health), Dr. A. Tyagi, ADG (AN&P), Dr. Yash Pal, Director (ICAR-NRCE) Dr. Jyoti Misri, Principal Scientist (Animal Health) at ICAR Headquarters, Dr. Sanjay Barua, In-Charge, NCVTC, PIs, Co-PIs and Scientists from 14 network units attended the meeting. In his opening remarks, Dr. Yash Pal opined that all units have contributed excellently to the NCVTC culture collection despite having manpower and fund constraints. He also urged all the units to deposit only unique cultures to the repository. Dr. Ashok Kumar, ADG (Animal Health) appreciated the progress of NCVTC made in a short span of time including the high impact publications by NCVTC over the years. Dr. A. Tyagi also appreciated the progress of NCVTC. Dr. B.N. Tripathi congratulated the team of NCVTC on being recognized as a National Repository by the MOEFCC, GOI. DDG emphasized on the need for an efficient system for the dispatch of cultures to different parts of the country. He also suggested that cultures being used for different vaccination and challenge studies in the country should be standard and uniform.

Visit of Dignitaries

Dr. Debalina Mitra, Assistant Commissioner, DADF visited NRCE museum and animal shed

Dr. Debalina Mitra, Assistant Commissioner, Department of Animal husbandry, Dairying & Fisheries (DADF), MoA & FW visited Infoequine museum and animal shed of NRCE, Hisar on November 22nd, 2020. She felt amazed with the exhibits in the museum and gathered information on different equine breeds. The eco-friendly nature of NRCE and the well-maintained farm impressed her and the team.

Dr. Priyanka Soni, Deputy Commissioner, Hisar appreciated NRCE

Dr. Priyanka Soni, Deputy Commissioner of Hisar district came for a short visit to NRCE to inspect the COVID-19 testing facility and NRCE Infoequine museum. NRCE scientists explained the equine research activities undertaken by the Centre and the various models exhibited in the museum. She had expressed her gratitude to the NRCE staff for their timely involvement in COVID-19 testing.

ASST. Commissioner, DADF during her visit to NRCE animal shed

Deputy Commissioner, Hisar visiting Infoequine museum

Infrastructure and Development Activities

Installation of fibre horse at EPC, Bikaner

A black fibre horse with a length of 76 inches and height of 90 inches was installed in front of equine museum on March 11, 2020 at EPC, Bikaner.

Fibre horse installed at EPC, Bikaner

Purchase of Nukra Stallions at EPC, Bikaner

Looking at the demand of the farmers, two Nukra males were purchased from the field and cryopreserved about 100 doses of semen for AI in field & farm.

NCVTC got recognized by Ministry of Environment, Forest and Climate Change

National Centre for Veterinary Type Cultures (NCVTC) has been recognized as a National Repository by the Ministry of Environment, Forest & Climate Change under the Biological Diversity Act, 2002 to keep safe custody of voucher specimens of microorganisms pertaining to veterinary sciences. In this regard, the formal communication has been received vide the Ministry's order No. F. No.CS-C12017/138/2020-CS-III dated 25.09.2020.

Establishment of Jenny Dairy unit at main campus, Hisar

A Jenny dairy unit was established by purchasing 10 female donkeys and one male foal from Horse and Donkey Breeding Farm, Chanasma, Dist. Patan, Gujarat. The required infrastructure for the dairy unit was established in the animal shed of NRCE, Hisar in July, 2020. These female donkeys will be used for research on donkey milk characterization.

Establishment of Jenny Dairy unit at main campus, Hisar

Table 1: Equine herd strength at Hisar campus

Particulars	Horse				Pony		Donkey				Total
	Stallion	Mare	Colt	Filly	Mare	Colt	Stallion	Mare	Colt	Filly	
Stock as on 01.01.2020	2	7	3	2	4	0	1	4	1	1	25
Addition	0	0	0	0	0	1	1	10	4	1	17
Disposal	0	1	0	0	0	0	0	2	0	0	3
Balance as on 31.12.2020	2	6	3	2	4	1	2	12	5	2	39
Grand Total	**13**				**5**		**21**				**39**

Table 2: Equine herd strength at EPC, Bikaner campus

Particulars	Horse				Pony				Donkey				Mule		Nukra		Total
	Marwari		Kathiawari		Zanskari		Manipuri		Poitou		Halari						
	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	
Stock as on 01.01.2020	17	29	1	4	3	7	4	3	14	15	4	5	2	1	0	0	109
Birth	5	2	1	0	0	3	0	0	2	4	0	0	0	0	0	0	17
Purchased	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	2
Transferred from main campus, Hisar	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	1	2
Transferred to main campus, Hisar	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	1
Mortality	0	0	0	0	0	2	0	0	1	1	0	0	0	0	0	1	5
Sold	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	1
Balance as on 31.12.2020	22	31	2	4	3	8	4	3	14	18	3	6	2	1	2	0	123
Grand Total	**53**		**6**		**11**		**7**		**32**		**9**		**3**		**2**		**123**

Poitou donkey herd at EPC, Bikaner

Halari donkey herd at EPC, Bikaner

Total agricultural production

In the year 2020, a total of 205 acres of land was cultivated with 100 acres at Hisar and 105 acres in Bikaner. The land was rotationally used for cultivating green fodder, dry fodder and grains for feeding farm equines. Total agricultural production was 5392.90 quintals, including 3200.75 quintals of green fodder, 983.55 quintals of dry fodder and 1208.60 quintals of grains.

Lucerne crop after 60 days at NRCE farm

Wheat crop in flowering stage at NRCE farm

Table 3: Crop-wise production

Type of crop	Production (Quintals)	
	Main campus, Hisar	EPC, Bikaner
Green fodder		
Sorghum	465	896.60
Oats	244	637.50
Lucerne	317	480.60
Sewan grass	-	27.05
Berseem	133	-
Dry fodder		
Oats, Bajra, Guar, Barley, Sewan grass, Wheat straw etc.	611.55	372
Grains		
Oats	199.95	182.95
Wheat	511.25	-
Barley	158.45	-
Paddy	70	-
Rapeseed	-	7.50
Guar	-	78.50
Total	2710.20	2682.70

Awards, Recognitions and Personal Milestones

1. Dr. B.R. Gulati appointed as **Editor of journal - 'Virus Disease'**, an official journal of Indian Virological Society.
2. Dr. B.R. Gulati appointed as **Editor, Special Issue on 'Equine Diseases'** of the journal, Indian Journal of Comparative Microbiology, Immunology and Infectious Diseases.
3. Dr. B.R. Gulati nominated as **DBT representative in the Institute Biosafety Committee** of Lala Lajpat Rai University of Veterinary and Animal Sciences, Hisar.
4. Dr. B.R. Gulati received **Certificate of Appreciation by NRCE** for outstanding contribution to NRCE on Independence Day Function of ICAR-NRCE, 15th August 2020.

Dr. M.R. Dhanda Memorial Oration medal Award to Dr R.K. Vaid

5. Dr. Naveen Kumar served as Member, expert panel for evaluation, monitoring and review of COVID-19 related international research projects funded by International Cooperation Division, Department of Science and Technology (DST), Govt. of India (2020-2023).
6. Dr. R.K. Vaid awarded **Dr. M.R. Dhanda Memorial Oration Medal** at National Symposium on "Public Health Challenge mitigation strategies at the confluence of One Health Approaches" and XIV Biennial National Conference of Association of Public Health Veterinarians (APHV) on January, 24th, 2020 at DUVASU, Mathura.
7. Dr S.C. Mehta conferred with **fellowship of Society for Conservation of Domestic Animal Biodiversity (SOCDAB)** in the XVII Annual Convention of SOCDAB and National Symposium on Enhancement of Farmer income through Management of Animal Genetic Resources held on February 10-11, 2020 at College of Veterinary Sciences and Animal Husbandry, NDVSU Campus, Mhow (MP).

SOCDAB fellowship award conferred on Dr S.C. Mehta

8. Dr. S.C. Mehta received **"Active Camel Research Scientist Award 2020"** by Camel Publishing House, Bikaner, Rajasthan.
9. Dr. Sanjay Barua nominated by ICAR as a **Member of the "National Capital Territory of Delhi Biodiversity Council"** to implement the provisions of the Biological Diversity Act, 2002 on 20 October 2020.
10. Dr. T.R. Talluri awarded with "**Active Camel Research Scientist Award 2020"** for publishing best article in Journal of Camel Practice and Research.
11. Dr. T.R. Talluri awarded with **ICAR-PDF programme** for 1 year (2020-2021) to conduct project based research at ICAR-NDRI, SRS, Bengaluru.
12. Dr. T.R. Talluri felicitated on the occasion of Republic Day, Jan 26th 2020 for the "**Outstanding performance**" to the ICAR-NRCE by the Director ICAR-NRCE. The award was received from the Incharge, EPC Bikaner on 26th Jan 2020.
13. Dr. T.R. Talluri received "**Reviewer excellence award**" from ARCC journal for serving as reviewer for the journal.
14. Ms. Ana Raj received **Best Paper presentation award** for the paper titled "Gender analysis in cassava cultivation" in the NAHEP National workshop on Gender issues and Atmanirbhar Bharat (online) conducted by Central Agricultural University (CAU), Imphal during October 15-16, 2020.

Joining and Superannuation

- Dr. Anubha Prashant Pathak, Scientist (Veterinary Public Health) joined at NRCE, Hisar campus on April 4, 2020 after successful completion of foundation training at ICAR-NAARM, Hyderabad.
- Ms. Ana Raj J., Scientist (Agricultural Extension) joined at NRCE, Hisar campus on April 4, 2020 after successful completion of foundation training at ICAR-NAARM, Hyderabad.
- Shri Ram Pal, Administrative Officer (AO) retired from NRCE, Hisar after successful tenure till January 31, 2020.
- Shri Pratap Singh, Upper Division Clerk (UDC) retired from NRCE, Hisar after successful tenure till January 31, 2020.
- Shri Raj Kumar, Administrative Officer (AO) joined ICAR-NRCE w.e.f. 26.09.2020.

Promotion

- Shri Gopal Nath, Senior Technician has been promoted to Technical Assistant w.e.f. 31st December, 2020.
- Shri Raghbir Singh, Technical Assistant has been promoted to Senior Technical Assistant w.e.f. 5th May, 2020.
- Shri Dinesh Datt Sharma, Upper Division Clerk has been promoted to Assistant w.e.f. 29th June, 2020.
- Shri Om Parkash, Upper Division Clerk has been promoted to Assistant w.e.f. 20th October, 2020.
- Shri Deepak Kumar, Lower Division Clerk has been promoted to Upper Division Clerk w.e.f. 10th November, 2020.
- Shri Mahender Singh, Lower Division Clerk has been promoted to Upper Division Clerk w.e.f. 10th November, 2020.

Dr. T.R. Talluri receiving award for "outstanding performance"

Active Camel Scientist Award 2020 conferred on Dr. T.R. Talluri

Research Publications

Research papers

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11. Chaudhary, A.K., Purohit, G.N., Mehta, J.S., Ravi, S.K. and Talluri, T.R., 2020. 144 serum testosterone profile in Marwari stallions and its relationship with testicular parameters, semen characteristics, reaction time, stallion age, bodyweight, and height. *Reproduction, Fertility and Development*, 32(2),198.
12. Dedar, R.K., Kumar, L., Badsiwal, D., Kumar, S., Legha, R.A., Mehta, S.C. and Tripathi, B.N., 2020. Herbal disinfection against *Rhodococcus equi*. *Indian Journal of Comparative Microbiology, Immunology and Infectious Diseases*, 41(1), 19-30.
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49. Verma, N., Kumar, D., Tiwari, A., Gulati, B.R. and Dogra, S., 2020. Clinical management of equine herpes virus-1 (EHV-1) infected newborn mule foals to prevent neonatal mortality – case studies. *The J Rem Vet Corps*, 59 (1), 80-95.

50. Yadav, J., Goel, P., Mandal, K.D., Yadav, R., Kumar, N., Kumar, R., Tripathi, B.N. and Kumar, S., 2020. Protein kinase inhibitors arrested the in-vitro growth of *Theileria equi*. *Acta Parasitologica*, 65(3), 644-651.

51. Yadav, P., Choudhury, S., Barua, S., Khandelwal, N., Kumar, N., Shukla, A. and Garg, S.K., 2020. *Polyalthia longifolia* leaves methanolic extract targets entry and budding of viruses-An in vitro experimental study against paramyxoviruses. *Journal of Ethnopharmacology*, 248, 112279.

Abstracts published in conferences/ symposia

1. Ana Raj J., Jaganathan D., Prakash P. and Sheela Immanuel. 2020. **Gender analysis in cassava cultivation in Kanyakumari district of Tamil Nadu.** In: National workshop on Gender issues and Atmanirbhar Bharat, CAU, Imphal. October 15-16.

2. Kumar R., Batra K., Sethi K., Rani Ruma, Kumar S. and Tripathi B. N. 2020. **Development of lateral flow assay using recombinant antigens for rapid diagnosis of *Trypanosoma evansi* infection.** Proceedings, 29th National Conference of Veterinary Parasitology and National Symposium on "Challenges and Innovations in Controlling Parasitic Diseases of Livestock and Poultry with Changing Climate", College of Veterinary Science & Animal Husbandry, NDVSU, Jabalpur, M.P. February 5-7.
3. Mandal K.D., Maji C., Tripathi B.N., Kumar R., and Kumar S. 2020. **Anti-*Theileria equi* activity of four Indian medicinal plants in MASP in vitro culture system.** Proceedings, 29th National Conference of Veterinary Parasitology and National Symposium on "Challenges and Innovations in Controlling Parasitic Diseases of Livestock and Poultry with Changing Climate", College of Veterinary Science & A.H., NDVSU, Jabalpur, M.P. February 5-7.
4. Manuja A., Kumar B. 2020. **Nano based bio macromolecules for therapeutic applications in animals.** In: International Online Conference on Macromolecules, Mahatma Gandhi University, Kottayam, Kerala and Gdansk University of Technology, Poland. November 13-15.
5. Vashisth M., Yashveer S., Virmani N., Bera, B.C., Vaid, R.K., Anand T. 2020. **Phage depolymerse mediated destruction of bacterial biofilms of ESKAPE pathogens.** In: International webinar on "Antibiotic Resistance: Renewed Fight". Webinar Jointly organized by All India Institute of Medical Sciences, New Delhi & American Society for Microbiology. October 7-8.

Books, Book chapters, Technical bulletins and Popular articles

1. Anand T., Virmani N., Bera B.C. 2020. **Coronavirus Pandemic - Covid 19.** Published on Slideshare April 29th, 2020.
2. Aswathy C, Ana Raj, Shanthanu Rakshit, Priyojoy Kar, Khrienguno Mepfhuo, HR Ramya, Bharat S Sontakki and K Nagasree. 2020. Institutional Linkages and Community Partnerships for Climate Resilient Agriculture. In: Book **"Climate Change and Indian Agriculture: Challenges and Adaptation Strategies"** published by ICAR-NAARM, Hyderabad. 569 - 583.
3. Kumar, Balvinder and Manuja, Anju. 2020. **Biological functions of Polymers and Metal Composites. In: Book "Current Topics in Medicinal Chemistry"** published by Benthem Science, ISSN: 1873-4294 (Online) and ISSN: 1568-0266 (Print), Vol. 20 (11).
4. Nokhwal, A., Ravikant, Anand. T. and Vaid, R. K. 2020. ***Aeromonas*: an overview.** In: L. T. Duncan (Ed.), Advances in health and research, Nova science publishers, Inc. New York, 1-68.
5. Punetha M., Pathak A., Choudhary S, Sharma M., Yashotha T., Arul S., and Ganesh B. 2020. **Climate Change versus Livestock Health: Impact, Mitigation and Adaptation.** In: Book "Climate Change and Indian Agriculture: Challenges and Adaptation Strategies" published by ICAR-NAARM, Hyderabad. 431 - 448.
6. Singha H, Shanmugasundaram K, Saini, S. 2020. **Diagnosis of glanders through ELISA and CFT.** In: ICAR sponsored short course on 'Newer approaches in disease diagnosis and vaccines for livestock and poultry' compendium organized at ICAR-NRCE, 28 Jan – 6 Feb, 2020, 149-154.
7. Singha H, Shanmugasundaram K, Tripathi B.N. 2020. **Glanders and *Rhodococcus equi* infection in horses and their control.** In: ICAR sponsored short course on 'Newer approaches in disease diagnosis and vaccines for livestock and poultry' compendium organized at ICAR-NRCE, 28 Jan – 6 Feb, 2020, 144-148.

Sequences published (48) in NCBI database

1. Chhabra, D., Manuja, B.K., Manuja, A., Singha, H.S., Vaid, R.K. and Sundaram, S. Diversity analysis of *Streptococcus equi* on the basis of SeM gene. MN974298, NCBI, USA.
2. Chhabra, D., Manuja, B.K., Manuja, A., Singha, H.S., Vaid, R.K. and Sundaram, S. Diversity analysis of *Streptococcus equi* on the basis of SeM gene. MN974297, NCBI, USA.
3. Chhabra, D., Manuja, B.K., Manuja, A., Singha, H.S., Vaid, R.K. and Sundaram, S. Diversity analysis of *Streptococcus equi* on the basis of SeM gene. MN974296, NCBI, USA.
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5. Chhabra, D., Manuja, B.K., Manuja, A., Singha, H.S., Vaid, R.K. and Sundaram, S. Diversity analysis of *Streptococcus equi* on the basis of SeM gene. *Streptococcus equi* strain G3566 M protein (seM) gene, partial cds. MN974294, NCBI, USA.

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7. Chhabra, D., Manuja, B.K., Manuja, A., Singha, H.S., Vaid, R.K. and Sundaram, S. Diversity analysis of *Streptococcus equi* on the basis of SeM gene. MN974291, NCBI, USA.
8. Chhabra, D., Manuja, B.K., Manuja, A., Singha, H.S., Vaid, R.K. and Sundaram, S. Diversity analysis of *Streptococcus equi* on the basis of SeM gene. MN974290, NCBI, USA.
9. Chhabra, D., Manuja, B.K., Manuja, A., Singha, H.S., Vaid, R.K. and Sundaram, S. Diversity analysis of *Streptococcus equi* on the basis of SeM gene. MN974289, NCBI, USA.
10. Chhabra, D., Manuja, B.K., Manuja, A., Singha, H.S., Vaid, R.K. and Sundaram, S. *Streptococcus equi* strain O2845 M protein (seM) gene, partial cds. MN974288, NCBI, USA.
11. Chhabra, D., Manuja, B.K., Manuja, A., Singha, H.S., Vaid, R.K. and Sundaram, S. Diversity analysis of *Streptococcus equi* on the basis of SeM gene. MN974287, NCBI, USA.
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15. Chhabra, D., Manuja, B.K., Manuja, A., Singha, H.S., Vaid, R.K. and Sundaram, S. Diversity analysis of *Streptococcus equi* on the basis of SeM gene. MN974283, NCBI, USA.
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18. Gulati, B.R., Kumar, N., Barua,S., Riyesh, T., Kumar, R., Gupta, S., Manuja, A., Kumar, B., Singha, H.S., Vaid, R.K., Anand, T.,Bera, B.C.,Virmani, N., Bhardwaj, A., Khandelwal, N., Kumar, R. and Pal, Y. Severe acute respiratory syndrome coronavirus 2 isolate SARS-CoV-2/human/IND/Isolate 5844/2020 ORF1ab polyprotein (ORF1ab), ORF1a polyprotein (ORF1ab), surface glycoprotein (S), ORF3a protein(ORF3a), envelope protein (E), membrane glycoprotein (M), ORF6 protein (ORF6), ORF7a protein (ORF7a), ORF7b (ORF7b), ORF8 protein(ORF8) nucleocapsidphosphoprotein (N), and ORF10 protein (ORF10) genes, complete cds. MW555334.NCBI, USA
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31. Manuja, B.K., Chhabra, D., Singha, H.S. and Manuja, A. *Streptococcus equi* strain 3616 heat shock protein 60 (hsp60) gene, partial cds. MT008477, NCBI, USA.
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43. Manuja, B.K., Manuja, A., Chhabra, D., Singha, H.S., Vaid, R.K. and Sundaram,S. Analysis of *Streptococcus equi* on the basis of Multi Locus Typing Sequence. MT154072.NCBI, USA.
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45. Manuja, B.K., Manuja, A., Chhabra, D., Singha, H.S., Vaid, R.K. and Sundaram,S. Analysis of *Streptococcus equi* on the basis of Multi Locus Typing Sequence. MT154070.NCBI, USA.
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47. Manuja, B.K., Manuja, A., Chhabra, D., Singha, H.S., Vaid, R.K. and Sundaram, S. Analysis of *Streptococcus equi* on the basis of Multi Locus Typing Sequence. MT008485. NCBI, USA.
48. Manuja, B.K., Chhabra, D., Singha, H.S. and Manuja, A. Streptococcus equi strain 4045 heat shock protein 60 (hsp60) gene,partial cds. MT008480.NCBI, USA.

Participation and Presentation in Seminars, Conferences and Scientific meetings

- Ms. Ana Raj J. presented a paper on **"Gender analysis in cassava cultivation"** in National workshop on 'Gender

issues and Atmanirbhar Bharat', Central Agricultural University (CAU), Imphal, 15-16 October, 2020.

- Dr. Anju Manuja delivered a presentation on **"Drug Resistance in Trypanosomiasis"** during brain storming meeting at ICAR-NRC on Camel, Bikaner, 6 January, 2020.
- Dr. Anju Manuja delivered an invited lecture on **"Nanobased biomacromolecules for therapeutic applications in animals"** in International Online Conference on 'Macromolecules' organized by Mahatma Gandhi University, Kottayam, Kerala and Gdansk University of Technology, Poland, 13-15 November, 2020.
- Dr. Anuradha Bhardwaj delivered a lead lecture at International e-Conference on Agricultural and Biological Sciences (I-E-CABS 2020), 18-19 December, 2020.
- Dr. B.C. Bera delivered guest lecture on **"Important bacterial and viral diseases of equines in Indian context and their control"** in an online International Seminar on 'Diagnostic Veterinary Pathology' organized by Department of Veterinary Pathology, Madras Veterinary College, TANUVAS, 12 September, 2020.
- Dr. B.R. Gulati delivered an invited lead lecture on **"Distribution of Zoonotic Japanese Encephalitis Virus Infection in India during 2009-2019"** in International Conference of Indian Virological Society (VIROCON 2020) on the theme 'Evolution of Viruses and Viral Diseases', National Agricultural Science Complex, New Delhi, 18-20 February, 2020.
- Dr. H. Singha delivered a lecture on **"Glanders and *Rhodococcus equi* infection in horses and their control"** in ICAR- sponsored short course on 'Newer approaches in disease diagnosis and vaccines for livestock and poultry' organised by ICAR-NRCE, Hisar, 28 January – 6 February, 2020.
- Dr. H. Singha delivered an invited lecture on **"Current status of glanders in India with special reference to Maharashtra"** in a workshop on 'Mapping the issues of Indian donkey and mule population and identify the potential intervention strategies and partners' organized by International Livestock Research Institute (ILRI) in College of Veterinary and Animal Sciences, Parbhani, 4 March, 2020.
- Dr. H. Singha participated in a one day workshop on **'Mapping the issues of Indian donkey and mule population and identify the potential intervention strategies and partners'** organized by International Livestock Research Institute (ILRI) in College of Veterinary and Animal Sciences, Parbhani, 4 March, 2020.
- Dr. H. Singha participated in International Webinar on **'Covid-19 infection: Origin and Transmission'** organized by Association of Microbiologist of India, Hisar Unit, 15 August, 2020.
- Dr. Naveen Kumar delivered a keynote lecture on **"Lumpy Skin Disease: an emerging threat to cattle in India"** in the IAVP zonal conference and national symposium on 'Recent advances in diagnostic pathology for emerging and re-emerging diseases in livestock, poultry under farming conditions and wildlife' held at Ranchi Veterinary college, Ranchi, 22-23 February, 2020.
- Dr. Naveen Kumar delivered a lead lecture on **"CRISPR/Cas9-mediated genome editing: scale up of virus production in vaccine manufacturing"** in the National Symposium on 'Public Health challenge mitigation strategies at the confluence of one health approaches' and XIV National Biennial Conference of Association of Public Health Veterinarians (APHV) at DUVASU, Mathura, 24-25 January, 2020.
- Dr. Naveen Kumar delivered an invited talk on **"Host-directed Antiviral therapies"** in "VAIBHAV"-Vaishwik Bharatiya Vaigyanik Summit, Global Summit of Overseas and Resident Indian Scientist and Academicians, 5-16 October, 2020.
- Dr. Naveen Kumar presented invited distinguished Guest-faculty lecture on **"CRISPR/Cas9-mediated genome editing: scale up of virus production in vaccine manufacturing"** in ICAR sponsored short training course on 'Newer Approaches in Disease Diagnosis and Vaccine for Livestock and Poultry' conducted by ICAR National Research Centre on Equines, Hisar, 28 January – 6 February, 2020.
- Dr. Naveen Kumar presented invited distinguished Guest-faculty lecture on **"Tissue culture techniques in isolation of virus and disease diagnosis"** in ICAR sponsored short training course on 'Newer Approaches in Disease Diagnosis and Vaccine for Livestock and Poultry' conducted by ICAR National Research Centre on Equines, Hisar, 28 January – 6 February, 2020.
- Dr. R.K. Dedar delivered a lecture on **"Equine Dermatology: A Clinical Approach"** in International Online Training on 'Advances in Equine Health Management' organised by College of Veterinary & Animal Sciences, Parbhani, 25-31 October, 2020.
- Dr. R.K. Vaid attended as Country Representative in FAO-RAP 2nd Consultation Meeting on **'Regional AMR Monitoring and Surveillance Guidelines Volume 2 Monitoring and surveillance of antimicrobial resistance in animal pathogens from diseased livestock and poultry',** 21-23 October, 2020.

- Dr. R.K. Vaid participated and presented **NRCE Annual Report for INFAAR Project 2018-19,** 28 August, 2020.
- Dr. R.K. Vaid participated in meeting **"One Health approach to surveillance *of Bacillus anthracis* in India (OHAI)"** Project discussion with VIT Vellore & University of Minnesota, on Anthrax Proficiency Testing Protocol, 26 June, 2020.
- Dr. R.K. Vaid participated in online meeting **"Modalities for ongoing programme of the Indian Network of Fisheries and Animal Antimicrobial Resistance (INFAAR)** 13th SFC plan period (2020-21 to 2024-25)" under the Chairmanship of Dr. J.K. Jena, DDG (Fish. Sci.), ICAR, 27 April, 2020.
- Dr. R.K. Vaid participated in **XIth Annual Scientific Review Meet of NCVTC ICAR - NRCE, Hisar** (2019-20), 20 November, 2020.
- Dr. R.K. Vaid participated in **Xth Annual Scientific Review Meet of NCVTC ICAR - NRCE, Hisar** (2018-19), 11 January, 2020.
- Dr. R.K. Vaid served as Chairman of Poster Evaluation Committee at Kisan Bhawan, College of Veterinary Sciences during National Symposium on **'Public Health Challenge mitigation strategies at the confluence of One Health Approaches'** and XIV Bienneial National Conference of Association of Public Health Veterinarians (APHV) at DUVASU, Mathura, 24 January, 2020.
- Dr. S.C. Mehta attended an online lecture delivered by Dr Cedric Gondro, Professor, Department of Animal Science, Michigan State University, USA on **"Feature Selection for Genomic Prediction"** organized under the distinguished lecture series of the World Bank Funded NAHEP project entitled "CAAST-Advanced Centre for Livestock Health" at ICAR-IVRI, 3 November, 2020.
- Dr. S.C Mehta presented a lead paper on **"Breeding strategies for conservation of indigenous breeds of equines"** in International Webinar on 'Present and future trends in conservation and breeding technologies to enhance production in indigenous animals', Department of AGB, Veterinary College and Research Institute, Tirunelveli, TANUVAS, Chennai, 15 December, 2020.
- Dr. S.C. Mehta presented a lead paper on **"Integration of molecular and traditional breeding tools for enhancing equine productivity"** in the National conference on 'Paradigm shift in livestock management to obtain high quality animal products for enhancing farm economy and entrepreneurship' organised by Rajasthan University of Veterinary and Animal Sciences, Bikaner and Indian Society of Animal Production and Physiology at Post-graduate Institute of Veterinary Education and Research (RAJUVAS), Jaipur, 4-6 February, 2020.
- Dr. S.C. Mehta presented a lead paper on **"Network Approach for Genetic Improvement of Equine Genetic Resources of India"** in the National Symposium on 'Enhancement of farmer's income through management of animal genetic resources' organised by the Society for Conservation of Domestic Animal Biodiversity and College of Veterinary Science and Animal Husbandry (NDVSU), Mhow, 10-11 February, 2020.
- Dr. Sanjay Barua attended a meeting on **"Biological Diversity Act 2002 Revision: Review of Agro biodiversity issues"** through VC as desired by DDG (AS) conducted on 1 August 2020 by ICAR-NBPGR, New Delhi.
- Dr. Sanjay Barua attended a meeting through VC to discuss and provide inputs on the EFC/SFC of ICAR-NIVEDI, Bengaluru under the chairmanship of DDG (AS) – ICAR, 4 June 2020.
- Dr. Sanjay Barua attended a meeting through VC to discuss and provide inputs on the EFC/SFC of ICAR-DFMD and ICAR-NIHSAD, Bhopal under the chairmanship of DDG (AS) – ICAR, 4 June 2020.
- Dr. Sanjay Barua attended a meeting through VC to discuss and provide inputs on the EFC/SFC of ICAR-NRCE, Hisar under the chairmanship of DDG (AS) – ICAR, 5 June 2020.
- Dr. Sanjay Barua attended a meeting to discuss **"Status of bluetongue pentavalent vaccine and about the status of Foot rot as well as Lumpy Skin Disease in the country"** under the chairmanship of DDG (AS) ICAR through VC, 22 October 2020.
- Dr. Sanjay Barua was invited as a panellist by ICAR-Indian Agricultural Research Institute in the Vaibhav Summit-Horizontal 2: Sustainable and Climate Smart Agriculture under the Vertical: Agro-Economy & Food Security, 10 October, 2020.
- Dr. Shanmugasundaram K. participated in a National Webinar on **'Laboratory Animal Management and Techniques in Biomedical Research'** organized by the Department of Livestock Production Management, Veterinary College and Research Institute, Orathanadu, Thanjavur, 24-26 August, 2020.

- Dr. Shanmugasundaram K. participated in a webinar on **'Strategies for sustainable control of parasites of livestock, poultry and wildlife and their public health significance'** organized by Department of Veterinary Parasitology, LUVAS, Hisar, 21-23 August, 2020.
- Dr. Shanmugasundaram K. participated in International Webinar on **'Covid-19 infection: Origin and Transmission**' organized by Association of Microbiologist of India, Hisar Unit, 15 August, 2020.
- Dr. T.R. Talluri acted as rapporteur for a technical session on **"Nutrient utilisation, growth and Lactation physiology"** at XXVIII Annual Conference of Society of Animal Physiologists of India & National Symposium on 'Physiological approaches to address environmental challenges for increasing animal productivity and farmer's income' held at CSWRI, Avikanagar, 18-19 February, 2020.
- Dr. T.R. Talluri delivered an expert lecture on **"Ovarian Follicular Dynamics of four breeds of Indigenous equines"** at XXVIII Annual Conference of Society of Animal Physiologists of India & National Symposium on 'Physiological approaches to address environmental challenges for increasing animal productivity and farmer's income' at CSWRI, Avikanagar, 18-19 February, 2020.
- Dr. T.R. Talluri delivered an invited expert lecture on **"Controlled Breeding programmes in Equine Species"** in National Webinar Series on Controlled Breeding Programmes in Livestock Species jointly organized by Institute Technology Management Unit (ITMU) & Animal Science Division of ICAR-CIARI, Port Blair, Andaman and Nicobar Islands, 16-23 November, 2020.
- Dr. T.R. Talluri delivered an invited expert lecture on the topic **"Semen collection, evaluation and cryopreservation in Equines"** in short term training course on 'Frozen semen technology in domestic animals' organized by Department of Gynaecology & Obstetrics, College of Veterinary Science & A. H., SDAU, Sardar krushinagar, 7-16 July, 2020.
- Dr. T.R. Talluri presented an invited expert lecture on the topic on **"Artificial Insemination and Pregnancy diagnosis in Equines"** in a webinar conducted by ISSAR, Rajasthan chapter, Bikaner, 17 July, 2020.
- Dr. Taruna Anand delivered an invited talk on the topic **"Careful designing of bacteriophage cocktail for biocontrol of MDR poultry pathogens and preliminary results on the encapsulation strategies for safe delivery in poultry gut"** during International conference of virology on Evolution of viruses & viral diseases (VIROCON 2020), 18-20 February, 2020 at NASC Complex, New Delhi.
- Dr. Yash Pal delivered a lecture on **"Equine Milk and its Potential Use"** in "Brain Storming on Potential of Non-bovine Milk" organized by NAAS at Zoom Webinar on June 29, 2020.

On-going Research Projects (2020)

Equine Health

Sr. No.	Title	Team	From	To	PIMS Code/Page
1.	Surveillance, Monitoring and Control of Emerging and Existing Diseases of Equines (Service Project)	H. S. Singha*, S.C. Yadav, B.R. Gulati, Rajender Kumar, Sanjay Kumar, N. Virmani, Sanjay Barua, Rajesh Vaid, Ramesh Dedar, Anju Manuja, B. Kumar, K. Shanmugasundram, and B. N. Tripathi	April, 1997	Contin -uous Service Project	IXX00257
2.	Development of recombinant EHV-1 viruses employing bacterial artificial chromosome mediated mutagenesis and their pathological evaluation in murine model	Nitin Virmani*, B.C.Bera, Taruna Anand and B.N. Tripathi	April 2017	Sept, 2021	IXX14011
3.	Diagnosis and sequence typing of strains *Streptococcus equi*	Balvinder Kumar*, R.K. Vaid, Anju Manuja, K. Shanmugasundram and H. Singha	April 2018	March 2021	IXX14584
4.	Characterization of Equine Herpes Virus isolates in India and documentation of their genetic diversity	B.R. Gulati*, Naveen Kumar, Riyesh T.	Sept. 2018	March 2021	IXX14746
5.	Development of multi-species ELISA for diagnosis of *Trypanosoma evansi* infection in different livestock species.	Rajender Kumar*, Sanjay Kumar and B.N.Tripathi	Nov., 2019	Oct., 2020	IXX15308
6.	Biomacromolecules based nanoscaffolds for wound healing using 3D printing	Anju Manuja*, Balvinder Kumar and Riyesh T.	Oct., 2020	Sept., 2023	IXX15412
7.	Nano-based therapeutic interventions against Osteoarthritis	Anju Manuja*, Balvinder Kumar and Riyesh T.	April, 2016	March, 2020	IXX12559
8.	*In-vitro* growth inhibitory efficacy of different herbal plant extracts against *T. equi* and identification of principal drug molecule(s) thereof	Sanjay Kumar* and Rajender Kumar	Aug., 2019	March, 2020	IXX14012

* Principal Investigator

Equine Production

S. No.	Title	Team	Date of Start	Date of Completion	PIMS Code
1.	Assessment and optimization of equine management in an intensive system (Service Project)	S.C. Mehta*, R. A. Legha, Yash Pal, R K Dedar, P A Bala, T R Talluri, S K Ravi and J Singh.	June, 2016	Service Project	IXX13192
2.	Endurance and fertility analysis in indigenous horses using SNP (Single Nucleotide Polymorphisms) markers	S.C. Mehta*, R.K. Dedar, T.R. Talluri and S.K. Ravi	Oct., 2017	Sept., 2021	IXX13995
3.	Assessment, evaluation and identification of physical, biochemical and genetic factors affecting stallion fertility	T.R. Talluri*, SC Mehta, Yash Pal, Anuradha Bhardwaj	April, 2018	March, 2021	IXX14589
4.	Studies on antitumor and antiviral potential of some plant extracts	Dr. R.K. Dedar*, Dr. Naveen Kumar and Dr. B.N. Tripathi	Nov., 2018	Oct., 2021	IXX14758
5.	Explicating genomic insights of Indigenous equines breed population through "Computational Genomics" and "Artificial Intelligence" based approaches	Anuradha Bhardwaj*, Sarika, Yash Pal, MA Iqbal and Dinesh Kumar	Dec., 2019	Nov., 2022	IXX15401
6.	Elucidation of physico-chemical, metabolomic and functional attributes of indigenous donkey milk	Anuradha Bhardwaj*, Yash Pal, RA Legha, Varij Nayan, AK Singh, PN Raju, Rajan Sharma and RK Vaid	July, 2020	June, 2023	IXX15413
7.	Optimisation of procedures for non-surgical recovery and bio-banking of equine embryos	T.R. Talluri*, Yash Pal and RA Legha	Oct., 2020	Oct. 2023	IXX15417
8.	Area specific mineral mixture for equine of Rajasthan	R. K. Dedar* and R. Nehra (RAJUVAS)	May, 2016	March, 2020	IXX13195
9.	Studies on bioactive components of donkey milk and its application	Anuradha Bhardwaj*	Dec., 2018	March, 2020	IXX14933

* Principal Investigator

NCVTC

Sr. No.	Title	Team	Duration	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.	Development of bacteriophage repository and exploring the therapeutic potential of phages and their encoded endolysin	Taruna Anand*, Nitin Virmani, R.K. Vaid and B.C. Bera	April, 2017	March, 2021	IXX13982

Sr. No.	Title	Team	Duration	To	PIMS Code
4.	Isolation, characterization and reposition of enteric viruses of poultry	NCVTC: Riyesh T.*, Naveen Kumar, Sanjay Barua and LUVAS: Naresh Jindal	June, 2017	March, 2021	IXX13988
5.	Isolation, characterization and generation of repository of Mycobacterium species	Shanmugasundaram K.*, R.K. Vaid, B.C. Bera and B.N. Tripathi	Oct., 2017	Sept., 2020	IXX13994
6.	Development of Knockout cell by CRISPR /Cas9-mediated genome editing	Naveen Kumar*, Sanjay Kumar, Sanjay Barua, Riyesh T, Balvinder Kumar	April, 2018	March, 2021	IXX14586
7.	Development of repository of respiratory viruses of livestock and isothermal based diagnostics for rapid identification.	B.C. Bera*, Nitin Virmani, Taruna Anand, B.R. Gulati and Riyesh T.	Aug., 2020	July, 2023	IXX15338
8.	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
9.	Prevalence studies for porcine respiratory viruses and development of their repository	B.C. Bera*, Sanjay Barua and Taruna Anand	Jan., 2016	March, 2020	IXX12436

* Principal Investigator

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, 2021	OXX00486
2.	All India Network Programme on Neonatal Mortality in Farm Animals	Sanjay Kumar*, Ramesh Dedar and B.R. Gulati (Nitin Virmani was Co-PI upto 8th Aug., 2018)	Jan., 2015	March, 2021	OXX02934
3.	CRP on Vaccines and Diagnostics	B.R. Gulati* Component-I (B.R. Gulati & Nitin Virmani) Component-II (B. R. Gulati & B.C. Bera) Nitin Virmani was Co-PI upto 8th Aug., 2018 Component-III (Sanjay Kumar & Rajender Kumar)	May, 2015	March, 2020	OXX03182
4.	Seroproteome analysis of recombinant secretory proteins of *Burkholderia mallei* towards development of multiple antigen immunoassay for improved diagnosis of glanders	H.S. Singha* & K. Shanmugasundaram	July, 2017	July, 2020	OXX03948
5.	Molecular epidemiology of Japanese Encephalitis Virus in Pigs and Mosquitoes in Assam (DBT Twinning Programme)	PI from Parent Institute: Seema Rani Pegu*, Dilip Kumar Sarma, Swaraj Rajkhowa and PI from Collaborative Institute: B.R. Gulati	Jan., 2017	July, 2020	OXX03737

Sr. No.	Title	Team	From	To	PIMS Code
6.	Elucidating therapeutic role of bacteriophages and encoded endolysins against multidrug resistant enteric pathogens of poultry	Taruna Anand*	June, 2018	May, 2021	OXX04448
7.	Exploration of genomic signatures for indigenous horses using next-generation sequencing approaches (DST-SERB)	Anuradha Bhardwaj*	Dec., 2018	Nov., 2021	OXX04453
8.	Investigating mechanism underlying acquisition of antiviral drug resistance against host targeting agents	Naveen Kumar* and Sanjay Barua	March, 2019	March, 2022	OXX04469
9.	Regional Coordination center under program for Inter-sectoral Coordination for prevention and control of Zoonotic Diseases	B.N. Tripathi* Bacterial Diseases: H.S. Singha, K.Shanmugasundram Viral Diseases: BR Gulati, Naveen Kumar, T. Riyesh	June, 2019	May, 2020	OXX04686
10.	Role of p38 MAP kinase in buffalopox virus replication	Sanjay Barua* and Naveen Kumar	Jan, 2020	Jan, 2023	OXX04792
11.	Development of host-directed anti-coronavirus agents (DST-SERB)	Naveen Kumar* and Riyesh T	June, 2020	June, 2021	OXX04795
12.	Pathogenecity and immunogenecity of recombinant neurogenic and non-neurogenic mutant EHV1 (in tissue explants and murine model) and their potential as vaccine candidate(s)	Nitin Virmani*	April, 2020	March, 2021	OXX03412
13.	Scheduling equines from fatal zoonotic disease – Glanders and Equine Infectious Anaemia (EIA) in India using Point of Care Diagnostics (POCD)	H. S. Singha* and B. N. Tripathi	March, 2018	March, 2020	OXX04100

* Principal Investigator

Staff at ICAR-NRCE
(as on 31.12.2020)

Director (Acting): Dr. Yash Pal

Sr.No.	Scientific staff
Main campus, Hisar	
1.	Dr. B.R. Gulati, Principal Scientist
2.	Dr. Nitin Virmani, Principal Scientist
3.	Dr. Rajender Kumar, Principal Scientist
4.	Dr. Sanjay Kumar, Principal Scientist
5.	Dr. Anju Manuja, Principal Scientist
6.	Dr. Balvinder Kumar, Principal Scientist
7.	Dr. Anuradha Bhardwaj, Senior Scientist
8.	Dr. Harishankar Singha, Senior Scientist
9.	Dr. Anubha Prashant Pathak, Scientist
10.	Ms. Ana Raj J., Scientist
NCVTC, Hisar	
11.	Dr. Praveen Malik, Principal Scientist (on deputation)
12.	Dr. Sanjay Barua, Principal Scientist
13.	Dr. R.K. Vaid, Principal Scientist
14.	Dr. Naveen Kumar, Principal Scientist
15.	Dr. Taruna Anand, Senior Scientist
16.	Dr. B.C. Bera, Senior Scientist
17.	Dr. Riyesh T., Scientist
18.	Dr. K. Shanmugasundaram, Scientist
Equine Production Campus, Bikaner	
19.	Dr. S.C. Mehta, Principal Scientist
20.	Dr R.A. Legha, Principal Scientist
21.	Dr. T.R. Talluri, Senior Scientist
22.	Dr. R.K. Dedar, Senior Scientist
Sr.No.	**Technical staff**
Main Campus, Hisar	
1.	Sh. K.S.Meena, Assistant Chief Technical Officer
2.	Sh. P.P.Chaudhary, Assistant Chief Technical Officer
3.	Sh. Sita Ram, Senior Technical Officer
4.	Sh. Sanjeev Kumar, Senior Technical Officer
5.	Sh. Ajmer Singh, Technical Officer
6.	Sh. Sajjan Kumar, Technical Officer
7.	Sh. Suresh Kumar, Technical Officer
8.	Sh. Joginder Singh, Technical Officer
9.	Sh. Mukesh Chand, Technical Officer
10.	Sh. Raj Kumar Dayal, Technical Officer
11.	Sh. Raghbir Singh, Technical Assistant
12.	Sh. Arun Chand, Senior Technician

Equine Production Campus, Bikaner	
13.	Dr. Jitender Singh, Assistant Chief Technical Officer
14.	Sh. Kamal Kumar Singh, Assistant Chief Technical Officer
15.	Dr. Ram Abtar Pachori, Assistant Chief Technical Officer
16.	Sh. Narender Chauhan, Senior Technical Officer
17.	Sh. Brij Lal, Technical Officer
18.	Sh. Satya Narayan Paswan, Senior Technical Assistant
19.	Sh. Om Parkash, Technical Officer
20.	Sh. Rajender Singh, Technical Assistant
21.	Sh. Gopal Nath, Technical Assistant
Administrative staff	
Main Campus, Hisar	
1.	Sh. Ram Pal, Administrative Officer (Retired on 31.01.2020)
2.	Sh. Raj Kumar, Administrative Officer (w.e.f. 26.09.2020)
3.	Sh. S.P. Kaushik, Assistant Administrative Officer
4.	Sh. Ashok Kumar, Personal Assistant
5.	Sh. Subhash Chander, Assistant Administrative Officer
6.	Sh. Sunil, Assistant
7.	Sh. Dinesh Datt Sharma, Assistant
8.	Sh. Om Parkash, Assistant
9.	Sh. Pratap Singh, Upper Division Clerk (Retired on 31.01.2020)
10.	Sh. Deepak Kumar, Upper Division Clerk
Equine Production Campus, Bikaner	
11.	Sh. Mahender Singh, Upper Division Clerk
Skilled Supporting Staff	
Main Campus, Hisar	
1.	Sh. Ishwar Singh
2.	Sh. Guru Datt Sharma
3.	Sh. Jai Singh
4.	Sh. Mahabir Prasad
5.	Sh. Ramesh Chander
6.	Sh. Mardan
7.	Sh. Desh Raj
8.	Sh. Ishwar Chander
9.	Sh. Om Parkash
10.	Sh. Hanuman Singh
11.	Sh. Subhash Chander
12.	Sh. Ishwar Singh
13.	Sh. Ram Singh
14.	Smt. Santra
15.	Sh. Lilu Ram
16.	Sh. Sant Ram
17.	Smt. Soma Devi
Equine Production Campus, Bikaner	
18.	Sh. Mahabir Prasad Meena
19.	Sh. Raju Ram
20.	Sh. Ashok Kumar

ICAR-NRCE NEWS

राष्ट्रीय अश्व अनुसंधान केंद्र के वैज्ञानिकों ने 100 दिन में कोविड-19 के 20 हजार से अधिक सैंपलों की जांच की

12 अप्रैल को संस्थान को सौंपा गया था टेस्टिंग का जिम्मा

मास्टर न्यूज | हिसार

राष्ट्रीय अश्व अनुसंधान केंद्र के वैज्ञानिक और अन्य ने 100 दिन में कोविड-19 के बीस हजार से अधिक सैंपलों की जांच की है। विभागीय अधिकारीयों ने भी वैज्ञानिकों की खूब सराहना की है।

राष्ट्रीय अश्व अनुसंधान केंद्र का मुख्य कार्य अश्व संबंधी बीमारियों पर अनुसंधान एवं प्रबंधन करना है। मगर कोविड-19 की महामारी को ध्यान में रखते हुए संस्थान ने भारतीय कृषि अनुसंधान परिषद नई दिल्ली से स्वीकृति मिलने के बाद 12 अप्रैल को अपने माइक्रोबियल कंटेनमेंट कॉम्प्लेक्स में एक विशेष कोविड-19 परीक्षण प्रयोगशाला की स्थापना की और कोविड 19 के सैंपलों की जांच का कार्य शुरू किया। यह परीक्षण सुविधा बायोसेफ्टी स्तर -3 प्रयोगशाला है, जहां वायरस रिलीज की रोकथाम के लिए सभी सुविधाएं उपलब्ध हैं। शुरू में यह कार्य धीमी गति से चला और फिर जांच कार्य ने गति पकड़ी- और एनआरसीई ने हिसार के साथ-साथ सिरसा और फतेहाबाद जिला के लोगों के सैंपल जांचने शुरू कर दिए।

राष्ट्रीय अश्व अनुसंधान केंद्र की पीआरओ व सीनियर वैज्ञानिक डॉ. अनुराधा भारद्वाज ने बताया कि 100 दिन में कुल 20305 सैंपलों की जांच की गई, जिनमें से 466 पॉजिटिव जबकि 19839 निगेटिव पाए गए। बताया कि अभी भी संस्थान में सैंपल की जांच जारी है।

निदेशक ने वैज्ञानिकों की सराहना की

एनआरसीई के निदेशक डॉ. यश पाल ने अपनी टीम की प्रशंसा करते हुए बताया कि नोडल ऑफिसर डॉ. बलदेव गुलाटी, डॉ. संजय बरुआ सहित डॉ. राजेंद्र खैयल, डॉ. संजय कुमार, डॉ. नितिन विरमानी, डॉ. राजेश वैद, डॉ. अलविंदर कुमार, डॉ. नवीन कुमार, डॉ. अंजू मनुजा, डॉ. अनुराधा भरद्वाज, डॉ. तरुणा आनंद, डॉ. हरिशंकर सिंरोया, डॉ. बी बेरा, डॉ. शंमुगासुन्दरम, डॉ. रियेश सहित वैज्ञानिकों की टीम द्वारा कोविड-19 के सैंपलों की जांच की जा रही है। परीक्षण में शामिल तकनीकी और सहायक कर्मचारी विजय कुमार, सीताराम, मुकेश चंद, संजीव कुमार, राज कुमार, जोगिंद्र कुमार, पार्थ चौधरी, मुकेश और स्मोटिंग में गुरु दत्त, सुभाष, ओम प्रकाश, महावीर, रमेश चंदर, जय सिंह, नितिन, शीतल, सोनाली, धार्वी, गुरमेश, प्रियशी का भी योगदान है। निदेशक डॉ. यशपाल ने बताया कि जांच में डीडीजी (एएस) डॉ. भीएन त्रिपाठी के साथ संचिव डेयर और डीजी आईसीएआर डॉ. त्रिलोचन महापात्रा ने इसमें प्रमुख भूमिका निभाई है।

आईसीएआर के तहत चारों लैब में हिसार का अश्व अनुसंधान संस्थान कोरोना जांच में आगे

अश्व अनुसंधान केंद्र और राजुवास विवि के बीच समझौता

अश्व अनुसंधान केंद्र के वैज्ञानिक बीकानेर जाकर भी विद्यार्थियों को देंगे जानकारी

अश्व अनुसंधान केंद्र की लैब में 1 लाख से ज्यादा कोरोना सैंपलों की जांच, 6164 निकले पॉजिटिव

अनुसंधान केंद्र ने एक माह में की 4100 ों की जांच, दस आ चुके पॉजिटिव

हिसार

• अश्व अनुसंधान केंद्र के निदेशक समेत 8 वैज्ञानिकों को सौंपी जिम्मेदारी, सैंपलों को भेजे जाएंगे रिपोर्ट

एनआरसीई ने हिसार, भिवानी, सिरसा, फतेहाबाद और जींद के लिए कोविड- 19 के नमूनों का परीक्षण शुरू किया, पहले दिन 21 सैंपल जांचे, सभी की रिपोर्ट निगेटिव

ब्लॉक वाइज मिलेगी मोबाइल ओपीडी की सुविधा, सिविल सर्जन को ड्यूटी रोस्टर व रूट प्लान बनाने के निर्देश दिए

केंद्र लैब में सैंपल जांच की क्षमता हुई चार गुनी

हिसार (संवाद)। अब जिला स्थित केंद्र अश्व अनुसंधान केंद्र की लैब में सैंपल जांचने की क्षमता चार गुना तक हो गई है। इसके लिए विभाग द्वारा लैब में दो शिफ्टों में जांच की जा रही है, जिसमें अब 85 से 90 सैंपल की जांच संभव हो गई है। इससे पहले शुरुआत में केवल 20-21 सैंपल की जांच होती थी, उसके बाद 40 से 45 सैंपल की जांच होनी शुरू हुई।

होस्ट आधारित कोरोना की एंटी वाय दवा को विकसित करेगा एनआरसीः

कोरोना की डायग्नोस्टिक टेस्ट किट व वैक्सीन के लिए आइसीएआर भी उतरा मैदान में

ICAR-NRCE NEWS

अश्व अनुसंधान केंद्र ने किया केरल की डेलिफ्रेन आईसीए के साथ करार

गधी के दूध से बने सौंदर्य उत्पादों का होगा व्यवसाय, साबुन और बॉडी बटर समेत कई प्रोडक्ट हो रहे तैयार

वरिष्ठ वैज्ञानिक डॉ. अनुराग भारद्वाज ने बताया गधी के दूध के गुण

कोरोना सैंपलों की जांच करने वाले घर में रहकर भी दू

एनआरसीई में दो मशीनें आज से करेंगी कोविड-19 टेस्ट

दौर, ऋषिकेश और दिल्ली के पांच घोड़ों में ग्लैंडर्स

हिसार, भोपाल और पूर्णे के बीरोलॉ में जल्द कोविड-19 पर शुरू हो सकती है रिसर्च

हमें वैज्ञानिकों पर नाज, प्रेमवर्क का दिया परिचय

ICAR-NATIONAL RESEARCH CENTRE ON EQUINES

HISAR 1986