

# Annual Report 2006-2007

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राष्ट्रीय अश्व अनुसंधान केन्द्र  
National Research Centre on Equines



***It is the Horse***

***An animal to some, an inspiration to us***

*Throughout history and across geographies, the horse has been considered second to none in the animal kingdom. Its speed, power, elegance and sensitivity has been the inspiration for many, across countless centuries.*

*And so it is with us at NRCE. We think we have learnt a thing or two from this magnificent animal, like speed and precision to address the issues concerning equine breeders and farmers.*

*It is this dedication to excellence and our sensitivity to equine problems that turns a small research centre in to an organization for utilization of equine power for agricultural activities.*



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The achievements and activities of the centre from April 2006 to March 2007 are presented in this report. Mention of trademark, proprietary product, or firm in text or figures does not constitute an endorsement and does not imply approval to the exclusion of other suitable products or firms.

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**NRCE is dedicated to improve equine health & productivity**



## DIRECTOR'S FOREWORD



NRCE is dedicated to undertake researches on equine health related problems in order to understand the subtleties involved in defining the best attributes of host (equines)

and worst armors of the agent. The dramatic emergence of glanders among equines of Maharashtra, Uttar Pradesh, Uttarakhand and Punjab in 2006 has had a tremendous impact on the equine population. In response to this acute crisis, the centre alerted the animal husbandry departments of various state governments. Our nation-wide glanders vigil deciphered that 97 of the 4395 equines tested from various states were affected by the glanders. Our strategic and tactical measures helped to restrict the spread of the disease. We continued our active equine disease serosurveillance in 16 states that helped us unravel that equines in our country have been free from equine infectious anemia, African horse sickness and equine viral arteritis in recent years. Primarily due to our relentless efforts, India has been recently declared free of African horse sickness by World Animal Health Organization (OIE). Japanese encephalitis (JE) being endemic in India, is a possible threat to equines. Therefore, screening of equines for JE has been incorporated in our nation-wide surveillance programme since 2006.

Development of diagnostics and vaccines has been the mainstay of the centre. The inactivated vaccine for the control of equine herpes virus-1 developed at the centre is now on way to field trial stage. It has

been our constant endeavor to improve and refine diagnostics for equine diseases. A monoclonal antibody-based assay developed at the centre for detection of rotavirus infection in young animals is mandated for patenting. Recombinant protein-based immunoassays for equine piroplasmiasis and EHV-4 have been standardized.

The centre has generated excellent information about indigenous breeds, including phenotypic and genotypic characterization of Marwari horse. The Marwari breed characteristics were finalized for registering true-to-breed Marwari horses. Manipuri ponies have been targeted for phenotypic and genotypic characterization.

For the benefit of poor equine owners, NRCE organizes several health camps in different regions and also offers artificial insemination services in field for conservation of indigenous equines. We also provide frozen semen of exotic jack to farmers for superior mule production. This is a small step in our efforts to improve the sustainability of subsistence level livelihood systems.

All our efforts have yielded fruits under the able guidance and support of Indian Council of Agricultural Research, New Delhi; particularly Dr. Mangala Rai Ji (DG, ICAR and Secretary, DARE), DDG (Animal Sciences) and Dr. Lal Krishna, ADG (Animal Health). I would like to congratulate the members of the publication committee for bringing this publication in this shape. My heartfelt appreciation goes to these dedicated members of the NRCE family.



**(S.K. DWIVEDI)**

## EXECUTIVE SUMMARY

During 2006-07, NRCE focussed on pursuing international competitiveness by developing infrastructure and resources to enable us to meet the global challenges in equine health in post-WTO scenario. The centre developed additional state-of-the-art laboratories at its Hisar and Bikaner campus. In addition, the renovation of the existing laboratories was done to meet the international (OIE) guidelines. The centre has also taken initiatives for the establishment of Veterinary Type Culture facility at NRCE. To promote trade and conservation of the indigenous equines, a one-day seminar was organized by the centre in collaboration with Department of Animal Husbandry, Dairying & Fisheries (Govt. of India) for finalization of breed characteristics of Marwari horses. A short-term training course was conducted for upgrading the skills of equine practitioners from different parts of India for better management of the problem of infertility and abortion in equines. We continued our efforts towards development & refinement of biologicals and diagnostics for improvement in health and production potentials of equines. Nation-wide monitoring of equine diseases, popularizing artificial insemination using quality cryopreserved semen and providing advisory & consultancy services to the equine farmers and breeders have been our priorities during the year. A brief account of achievements of NRCE in year 2006-07 is outlined below:

An equine herpes virus-1 (EHV-1) killed vaccine incorporating indigenous strain of EHV-1 has been developed by the centre. During the year, protective response of this vaccine was evaluated in pregnant indigenous mares. This study indicated that the vaccine is safe and generates good immune response in equines. The level of protection of immunized mares after virus challenge was satisfactory. Before recommending the vaccine for mass scale production and use, its immune response in mares needs to be evaluated in selected organized equine farms.

The centre is continuously working towards development and refinement of diagnostics for important equine diseases. During the year, a study was initiated to develop diagnostics for Japanese

## कार्यकारी सारांश

वर्ष 2006-07 के सत्र में राष्ट्रीय अनुसंधान केन्द्र हिसार ने अपनी अन्तर्राष्ट्रीय प्रतिस्पर्धात्मक क्षमता को बढ़ाने हेतु आधारभूत ढाँचे एवं अन्य संसाधनों का विकास किया जिससे हम अन्तर्राष्ट्रीय व्यापार संगठन नियमों के नए परिपेक्ष में विश्वस्तरीय चुनौतियों का सामना कर सकें। केन्द्र ने अपने हिसार एवं बीकानेर परिसर में बुनियादी सुविधाओं से सुसज्जित उच्चकोटि की प्रयोगशालाओं का निर्माण किया। इसके इलावा अन्तर्राष्ट्रीय मापदण्ड पर खरे उतरने के लिए हमने अपनी वर्तमान प्रयोगशालाओं को नया रूप दिया है। केन्द्र ने "पशु रोग जीवाणु संवर्धन सुविधा" की स्थापना करने की दिशा में कदम उठाए हैं। देश में अश्वों की उन्नत नस्लों के व्यापार को बढ़ावा देने एवं उनके संरक्षण हेतु केन्द्र ने पशु-पालन, डेयरी एवं मात्स्यिकी विभाग (भारत सरकार) के सहयोग से एक दिवसीय गोष्ठी का आयोजन किया। इस गोष्ठी में मारवाड़ी नस्ल के अश्वों की प्रजाति विशेषताओं के मानकीकरण को आधिकारिक स्वीकृति दी गई। केन्द्र में एक अल्प अवधि प्रशिक्षण पाठ्यक्रम का भी आयोजन किया गया, जिसमें भारत के विभिन्न भागों से आए अश्व पशु-चिकित्सकों ने भाग लिया। इस प्रशिक्षण कार्यक्रम में अश्वों में प्रजनन, बॉझपन एवं गर्भपात की समस्याओं के प्रबंधन के बारे में जानकारी दी गई। अश्वों के स्वास्थ्य संवर्धन एवं उत्पादन में सुधार के लिए संशोधित जैविक एवं नैदानिक सामग्रियों के विकास के लिए केन्द्र प्रयत्नशील रहा है। अन्य कार्यक्रमों के अन्तर्गत देशव्यापी अश्व रोग निगरानी, अति हिमीकृत वीर्य से कृत्रिम गर्भाधान एवं अश्व पालकों के परामर्श एवं सलाह आदि के कार्य यथोचित चलते रहे हैं। राष्ट्रीय अश्व अनुसंधान केन्द्र द्वारा वर्ष 2006-07 की उपलब्धियों का एक संक्षिप्त ब्यौरा नीचे दिया जा रहा है:

केन्द्र द्वारा घोड़ियों में गर्भपात से बचाव के लिए एक स्वदेशी निष्क्रिय अश्व हरपीज विषाणु-1 प्रयुक्त टीका तैयार किया गया है। वर्ष के दौरान इस टीके का देशी नस्ल की गर्भित घोड़ियों में रोग प्रतिरक्षण क्षमता का परीक्षण भी किया गया। इस परीक्षण के परिणाम दर्शाते हैं कि टीका पूर्णतयः सुरक्षित है एवं रोग प्रतिरोधक शक्ति का स्तर रोगकारक विषाणु से संक्रमण के पश्चात भी संतोषप्रद है। इस टीके के बड़े स्तर पर उत्पादन एवं प्रयोग से पूर्व इसका अश्व प्रजनन फार्म पर गर्भित घोड़ियों में परीक्षण आवश्यक है, ताकि इसके द्वारा उत्पन्न रोग प्रतिरोधक शक्ति का फार्म स्तर पर मूल्यांकन हो सके।

केन्द्र निरन्तर अश्व रोगों की नैदानिक विधियों के विकास एवं परिष्कृतिकरण की ओर अग्रसर है। वर्ष के दौरान अश्वों में जापानी मस्तिष्क ज्वर के निदान एवं भारत में इस रोग की व्यापकता जानने के लिए अनुसंधान प्रारम्भ किया





encephalitis (JE) and to know its status and geographic distribution among equines in India. Haemagglutination inhibition (HAI) assay was standardized using the HA antigen produced at the centre for serodiagnosis of JE. On testing equine serum samples (n=754) from different states for antibodies to JE serocomplex by HAI, 108 (14.32%) samples were detected positive. In addition, a RT-PCR was also developed for detecting the virus in infected tissues.

During the year, a recombinant protein-based ELISA was standardized for differentiation of EHV-1 and EHV-4 viruses which cause respiratory disease and abortions in equines. On testing 209 serum samples by this assay, antibodies to EHV-4 were found in 171 (81.8%) samples. Further work on the sensitivity and specificity of this assay is under progress.

Equine piroplasmiasis is an important tick-borne haemoprotozoan disease caused by intra-erythrocytic protozoa, *Babesia equi* and /or *Babesia caballi*. An ELISA has been developed employing recombinantly expressed merozoite surface protein EMA-2, for detection of *B. equi* specific antibodies. This assay could detect *B. equi* antibodies in equines from 6 days post-infection till 90 days of observation period. Out of 1172 equine sera tested by this assay, antibodies to *B. equi* were found in 610 (52.04%) samples.

Our unique program of nation-wide active equine disease surveillance facilitated poor equine owners in saving their precious indigenous equine germplasm through timely diagnosis and management of diseases. An outbreak of glanders was reported during July-August 2006 in Maharashtra. Subsequently, cases of glanders were also reported from Punjab, Uttar Pradesh and Uttarakhand. Nation-wide testing on 4395 serum samples revealed 97 samples to be positive for glanders during the year. Active equine disease sero-surveillance was conducted in the states of Gujarat, Rajasthan, Haryana, Punjab, Chandigarh, Delhi, Uttar Pradesh, Madhya Pradesh, Maharashtra, Andhra Pradesh, Tamil Nadu, Karnataka, West Bengal, Manipur, Chattisgarh and Uttarakhand. Antibodies to EHV-1 were detected in 25 (2.2 %) of the 1150 samples, while *Babesia equi* was detected in 610 (52.0 %) of the 1172 sera tested, Japanese encephalitis in 108 (14.3%) of the 754 serum samples tested. None of the serum samples tested was detected positive for equine infectious

gaya। केन्द्र द्वारा उत्पादित हीमएग्लुटिनिन के इस्तेमाल से जापानी-ज्वर के निदान के लिए हीमएग्लुटिनेशन दमन परीक्षण का मानकीकरण किया गया। इस विधि द्वारा विभिन्न राज्यों से प्राप्त कुल 754 अश्व नमूनों की जाँच करने पर 108 (14.3%) अश्वों में जापानी-ज्वर के प्रतिपिण्ड पाए गए। इसके अतिरिक्त संक्रमित पशुओं के ऊतकों से जापानी ज्वर के विषाणुओं की जांच के लिए एक आर. टी. पी. सी. आर. विधि विकसित की गयी।

वर्ष के दौरान अश्व हरपीज विषाणु-1 एवं 4 के प्रतिलिपित एवं अभिव्यक्त प्रोटीन आधारित एलाइज़ा परीक्षण का इन दो विषाणुओं में पृथक्कीकरण के लिए मानकीकरण किया गया। ये हरपीज विषाणु (1 एवं 4) अश्वों में श्वास रोग एवं गर्भपात के कारक हैं। इस विधि द्वारा कुल 209 अश्वों के नमूनों का परीक्षण किया गया। अश्व हरपीज विषाणु-4 की प्रतिपिण्ड (एण्टीबाडी) इनमें से 171 (81.8%) अश्वों में पाई गई। इस परीक्षण की संवेदनशीलता एवं विशिष्टता पर आगे का काम चल रहा है।

अश्व पाइरोप्लासमोसिस एक चीचड़-जनित रक्त प्रोटोजोआ रोग है। बैबीसिया इक्वाई अथवा बैबीसिया कैबेल्ली नामक प्रोटोजोआ लाल-रक्त कोशिकाओं के भीतर जाकर रोग उत्पन्न करते हैं। एक पुनः संयोजित मीरोज्वाइट सतह प्रोटीन पर आधारित एलाइज़ा का विकास बैबीसिया इक्वाई के प्रतिपिण्ड का पता लगाने के लिए किया गया है। इस विधि द्वारा घोंड़ों में रोग संक्रमण होने के 6 दिन पश्चात् से लेकर 90 दिनों तक एण्टीबाडी देखी गयी। कुल 1172 अश्व सीरम नमूनों को एलाइज़ा द्वारा जांचा गया और बैबीसिया इक्वाई प्रतिपिण्ड 52.04 प्रतिशत नमूनों में पाए गए।

हमारे देशव्यापी सक्रिय "अश्व रोग सर्वेक्षण कार्यक्रम" द्वारा स्वदेशी नस्ल के अश्वों को सामयिक रोग निदान प्रबंधन की सुविधा अश्व पालकों के द्वार पर उपलब्ध कराई गई। महाराष्ट्र राज्य के अश्वों में 2006 (जुलाई-अगस्त) में ग्लैण्डर का प्रकोप पाया गया। इसके पश्चात् इसका संक्रमण उत्तरी भारत के पंजाब, उत्तर-प्रदेश एवं उत्तराखण्ड राज्यों में भी पता लगा। इसके फलस्वरूप एक राष्ट्र-व्यापी ग्लैण्डर परीक्षण कार्यक्रम चलाया गया, जिसमें 4395 अश्व सीरम नमूनों की जाँच करने पर 97 अश्व ग्लैण्डर रोग-ग्रसित पाए गए। इस सक्रिय "अश्व रोग निगरानी कार्यक्रम" के अन्तर्गत देश के विभिन्न राज्यों में अश्वों की जांच की गई। ये राज्य हैं: गुजरात, राजस्थान, हरियाणा, पंजाब, चण्डीगढ़, दिल्ली, उत्तर-प्रदेश, मध्य-प्रदेश, महाराष्ट्र, आंध्रप्रदेश, तमिलनाडु, कर्नाटक, पश्चिम-बंगाल, मणिपुर, छत्तीसगढ़ और उत्तराखण्ड। इस जांच में देश भर के अश्वों में अश्व हरपीज विषाणु-1 प्रतिपिण्ड 1150 में से 25 (2.2%) नमूनों में पाए गए, जबकि बैबीसिया इक्वाई 1172 में से 610 (52%) नमूनों में पाए गए। जापानी-ज्वर प्रतिपिण्ड 108 (14.3%) अश्व सीरम नमूनों

anemia, African horse sickness, equine influenza and *Salmonella Abortus equi*.

Development of drug resistance is a major problem in equines. To address this issue, a study was initiated to identify the high-level drug resistant sentinel bacteria in equines that will work as a model to predict the outcome of antibacterial drug use. A total of 198 samples from equids and their environments were collected and 512 bacterial isolates (234 Gram-positive and 278 Gram-negative). Gram-positive bacterial isolates were predominated by *Enterococcus* spp whereas Gram-negative bacteria belonged to *Escherichia*, *Klebsiella*, *Proteus*, *Enterobacter*, *Aeromonas* and *Edwardsiella* genera. Multi-drug resistance was common in Gram-negative bacteria, particularly in *E. coli* and *Klebsiella* spp isolates. However, resistance was not as common as in Gram-positive bacteria. Drug resistance was more common in isolates from equids maintained on organized farms.

Molecular epidemiology of rotavirus isolates causing severe diarrhoea in foals from two organized farms in northern India was studied. Of the 164 diarrhoeic stool samples, 58 (35.36%) were positive for rotavirus and their RNA exhibited at least 5 different electropherotypes (E1-E5). The serotyping of the representative equine rotavirus isolates was done by ELISA and nucleotide sequencing. It was revealed that E1, E2, E4 and E5 strains exhibited G10, G3, G6 and G1 type specificities that accounted for 19.0, 42.9, 14.3 and 9.5% of the isolates, respectively.

*Rhodococcus equi*, a pathogen causing bronchopneumonia in foals was isolated from 4 of the 48 samples from equine foals and their environment. They were found to be sensitive to chloramphenicol, erythromycin, oxytetracycline, ciprofloxacin, neomycin and rifampicin.

Major histocompatibility (MHC) genes provide a major genetic component of resistance/susceptibility to infectious or autoimmune diseases. To study the polymorphism in the MHC class II genes in Marwari horses, PCR conditions for amplification of polymorphic regions of MHC class-II (DRB-2a and 2b) genes were standardized. The fragments of 276 bp and 229 bp were amplified using Be1 and Be2 primers, respectively from DNA samples of Marwari horses. Further work is under progress to study heterozygosity at these loci at population level.

में पाए। इस जॉच में देश भर से कोई भी नमूना अश्व संकामक एनीमिया, अफीकी अश्व रोग, अश्व इन्फ्लूएंजा और साल्मोनैल्ला एबारटस इक्वाई से ग्रसित नहीं पाया गया।

एण्टीबायोटिक के विरुद्ध जीवाणुओं की बढ़ती प्रतिरोधक क्षमता एक बड़ी समस्या है। इस समस्या के समाधान के लिए अश्वों में एण्टीबायोटिक प्रतिरोधी सन्तरी जीवाणुओं को पहचानने की एक शोध परियोजना पर कार्य शुरू किया गया। इस अध्ययन से अश्वों में एण्टीबायोटिक दवाओं के इस्तेमाल से होने वाले परिणाम का पता लगाया जा सकेगा। कुल 198 अश्वों और उनके वातावरण के नमूनों में से 512 जीवाणुओं को पृथक कर निकाला गया (234 ग्राम-सकारात्मक एवं 278 ग्राम-नकारात्मक)। ग्राम-सकारात्मक जीवाणुओं में मुख्यतः एन्टेरोकॉकस पाए गए और ग्राम-नकारात्मक में एस्चरीशिया और क्लैब्सिएल्ला जाति के जीवाणु पाए गए। ग्राम-नकारात्मक जीवाणुओं में कई एण्टीबायोटिक्स के विरुद्ध प्रतिरोधी शक्ति पाई गई। यह प्रतिरोधी शक्ति ग्राम-सकारात्मक जीवाणुओं में अपेक्षाकृत कम थी। एण्टीबायोटिक्स प्रतिरोधक क्षमता संगठित क्षेत्र के अश्व फार्म के अश्वों से प्राप्त जीवाणुओं में अधिक थी।

अतिसारित अश्व शावकों में पाये गये विभिन्न रोटा वायरस विषाणुओं में विविधता जानने के लिये उन पर आण्विक स्तर पर महामारी-वैज्ञानिकी अध्ययन किया गया। ये विषाणु उत्तर-भारत के दो संगठित फार्मों के 164 अतिसारित अश्व-शिशुओं में से 58 (35.36%) में मिले। इन विषाणुओं को उनके आर. एन. ए. की गतिशीलता के अनुसार 5 विभिन्न प्रकार के विद्युतजातिवर्गीकरण समूह (इलैक्ट्रोफोरोटाइप) ई.1 से ई. 5 में वर्गीकृत किया। अश्वों के इन रोटा विषाणुओं के सीरम वर्गीकरण (सीरोटाइपिंग) एवं न्यूक्लीयोटाईड पंक्तिकरण ने इंगित किया ई.1, ई.2, ई.4 एवं ई.5 समूह के विषाणु क्रमशः जी.10, जी.3, जी.6 एवं जी.1, सीरोटाइप से सम्बंधित थे तथा उनकी निश्चितता क्रमशः 19, 42.9, 14.3 और 9.5 प्रतिशत विषाणुओं में पाई गई।

अश्व शावकों और उनके वातावरण से लिए गए 48 नमूनों में से 4 में रहोडोकाकस इक्वाई नामक निमोनिया करने वाले रोगाणु पाए गए। ये रोगाणु क्लोरमफैनीकाल, एरिथ्रोमाइसिन, आक्सीटैट्रासाईक्लिन, सिपरोफ्लोक्सासिन, नियोमाइसिन और रिफैम्पीसिन एण्टीबायोटिक दवाइयों से निष्क्रिय होता पाया गया।

प्रधान कोशिका-अनुरुपता जीन समूह (मेजर हिस्टोक्मपैटीबिलिटी काम्प्लैक्स) प्राणियों को संक्रामक एवं स्वप्रतिरक्षारोग (आटोइम्यूनिटी) से प्रतिरोध एवं संवेदनशीलता के गुण देता है। इन एम.एच.सी.-2 वर्ग के जीनों की बहुरूपता का मारवाड़ी अश्वों में अध्ययन किया गया। ये परीक्षण जीन बी. ई.1 एवं बी.ई.2 पर पी. सी. आर. विधि द्वारा किए गए। दो प्राइमरों द्वारा 276 एवं 229 बेसयुग्मों का विस्तार 49 मारवाड़ी अश्वों के डी. एन. ए. से किया गया। इन जीनों के डी. एन. ए.



The study was envisaged to evaluate the role of fertility related proteins from equine seminal plasma. The effect of the heparin and gelatin binding proteins (HBP and GBP) from stallion seminal plasma on *in vitro* capacitation by induction of acrosome reaction (%) and on osmotic swelling by HOST test was evaluated. Significant difference was observed in the percentage of acrosome reacted spermatozoa at 4 hr incubation between HBP treated (40 µg) and control groups. The percentage of hypo-osmotic swollen positive sperms was higher in HBP and GBP protein treated samples than in untreated control.

In an inter-institutional research project on characterization of indigenous breeds of equines, biometric indices of fifty Manipuri ponies were evaluated. The study indicated that height at wither in both the sexes of Manipuri ponies ranged from 119-134 cm. In addition, heart girth (143.6 cm), hind leg length (82.3 cm), foreleg length (78.4 cm) and height at knee (39.4 cm) were also recorded. DNA of representative animals of this breed is being analyzed for molecular characterization of the breed.

The semen of Marwari stallions (n=42) and exotic donkey stallions (n=50) was compared by physical and biochemical parameters. Progressive motility (%) in gel free semen was recorded as  $73.33 \pm 0.94$  and  $77.80 \pm 1.00$  per cent in horse and donkey stallions, respectively. Activity of GOT, GPT and LDH was 174.3, 26.08 and 1552 IU/l in seminal plasma of Marwari horses and 387.7, 28.2 and 3913 IU/l in seminal plasma of donkey stallions, respectively.

During the period, semen from selected Marwari stallions at farmers' door was cryopreserved using mobile laboratory. This semen was used for artificial insemination (AI) of 10 Marwari mares at the centre to produce superior offspring. In addition, AI of 14 Marwari mares at farmers' door was done, of which 7 were confirmed pregnant. The semen of Poitu jacks was also cryopreserved for doing AI in the farm as well as in field animals. A total of 14 mares in the field were inseminated with frozen semen of exotic jacks in Haryana, U.P. and Uttarakhand for mule production. Survey of mule producers of these regions was also done to know about their mule management and production practices and necessary interventions to educate farmers for better mule production.

में स्थित स्थानों की विभिन्नरूपता का अध्ययन प्रगति पर हैं।

अश्व वीर्य पीत-तरल में पाए जाने वाली प्रोटीनों का घोड़ियों में गर्भाधान क्षमता पर प्रभाव का अध्ययन किया गया। नर अश्वों से प्राप्त हिपैरिन और जिलेटिन बंधित प्रोटीनों का प्रजनन प्रभाव शुक्राणु-शीश प्रतिक्रिया विधि द्वारा देखा गया और उनके फूलने का असर होस्ट परीक्षण द्वारा देखा गया। यह ज्ञात हुआ कि हिपैरिन बंधित प्रोटीन (40 माइक्रोग्राम) के प्रयोग से शुक्राणुओं की सामर्थ्यता में महत्वपूर्ण सुधार होता है। नियन्त्रण समूह से तुलनात्मक रूप में हिपैरिन और जिलेटिन बाईंडिंग प्रोटीन से संश्लेषित शुक्राणुओं में फूलने की मात्रा अधिक थी।

एक अंतर-संस्थानीय अनुसंधान शोध परियोजना के अन्तर्गत विभिन्न भारतीय नस्लों के अश्वों के जातिगुणों का विश्लेषण किया जा रहा है। इस वर्ष 50 मणिपुरी अश्वों के शारीरिक सांख्यिकी (लम्बाई-चौड़ाई) गुणों का अध्ययन किया गया। अध्ययन से ज्ञात हुआ है कि मणिपुरी अश्वों के नर और मादाओं के डिल्ले की ऊंचाई 119-134 सें.मी. है। इसके अतिरिक्त छाती का घेरा (143.6 सें.मी.), पीछे की टांगों की लम्बाई (82.3 सें.मी.), आगे की टांगों की लम्बाई (78.4 सें.मी.) और घुटनों तक की ऊंचाई (39.4 सें.मी.) का लेखा लिया गया। इस नस्ल का आण्विक विश्लेषण के लिए इनके डी. एन. ए. के नमूनों का अध्ययन भी किया जा रहा है। इसके अतिरिक्त मारवाड़ी प्रजाति के अश्वों पर भी गहन अध्ययन किए गए।

मारवाड़ी अश्वों और विदेशी गदर्भों के वीर्य का भौतिक एवं जैवरासायनिक तुलनात्मक परीक्षण किया गया। वीर्य की उत्तरातर गतिशीलता मारवाड़ी अश्व और गदर्भ नरों में क्रमशः  $73.33 \pm 0.94$  एवं  $77.80 \pm 1.00$  प्रतिशत पाई गई।

इस अवधि में अश्व पालकों के चुनिंदा मारवाड़ी नर अश्वों का वीर्य अश्व पालकों के द्वार पर जाकर हिमीकृत किया गया। इस वीर्य से मारवाड़ी गुण-संपन्न अश्वों के उत्पादन के लिए हमारे परिसर स्थित 10 एवं ग्राम स्तर पर 14 मारवाड़ी घोड़ियों का कृत्रिम गर्भाधान कराया गया। हमारे परिसर में सात घोड़ियां इससे गर्भित हो गईं। पोइटू जाति के नर गदर्भों का वीर्य भी संगठित फार्म और ग्राम-क्षेत्र की घोड़ियों में कृत्रिम गर्भाधान करने के लिए शीत-परिरक्षित किया गया। इस नस्ल के उत्तम नर-गर्भों के वीर्य से हरियाणा, उत्तर-प्रदेश एवं उत्तराखण्ड राज्यों में टट्टू उत्पादन के लिए वहां जाकर घोड़ियों का कृत्रिम गर्भाधान किया गया। इस क्षेत्र के टट्टू उत्पादकों का सामाजिक एवं आर्थिक स्तर जानने एवं उनके द्वारा अपनाई गई टट्टू-प्रबंधन और उत्पादन कार्यप्रणाली जानने के लिए सर्वेक्षण किया गया, जिससे इन अश्व पालकों को बेहतर टट्टू-प्रबंधन तकनीकी जानकारी दी जा सके।

वर्ष 2006-07 में केन्द्र ने भारतीय कृषि अनुसंधान परिषद् द्वारा प्रायोजित "अश्वों में बांझपन एवं गर्भपात" विषय पर एक अल्प-अवधि (4-13 जुलाई, 2006) प्रशिक्षण कार्यक्रम का आयोजन किया। इसमें अश्व-चिकित्सकों, वैज्ञानिकों और

During 2006-07, the centre organized an ICAR sponsored short course on “Infertility and abortion syndrome in equines” from July 4-13, 2006. Participants in the course included equine veterinary practitioners and scientists working in the field of equine breeding from different parts of the country. The scientists of the centre published 20 original research articles in international and national journals and their 10 research articles have been accepted for publication. In addition, 11 popular articles and 12 abstracts were published by the scientists. Expert lectures (22) were delivered by the scientists in various advanced training/refresher courses organized by various state universities and national institutes. Scientists participated in 14 different national and international conferences/symposia. Seven scientists participated in different training or refresher courses for upgradation of their skills. Dr. R. V. S. Pawaiya, Scientist (Veterinary Pathology) has been awarded 'Jawaharlal Nehru Award for P.G. Agricultural Research 2005' by ICAR for his Ph.D. research. A research paper published in the Animal Reproduction Sciences by Dr. A. Arangasamy (Scientist) has been selected for G.B. Singh Memorial Award.

The centre extended equine welfare activities in different parts of the country by organizing equine health camps and farmer meets (*Ashwa Palak Goshthis*) to enlighten the equine owners on various aspects of disease control and management. In addition to the treatment of major equine ailments in these camps, deworming and tetanus vaccination was done in equines. Feedback from farmers was obtained for further research and development in equine health and production. Foundation Day of the centre was celebrated with great zeal and enthusiasm on 26th November 2006. On this occasion, a horse show was also organized in which indigenous horses from various states participated in different equestrian events.

The centre also offered consultancy and diagnostic services for important infectious diseases of equines. Under this programme, 5032 equine serum samples were tested for equine infectious anemia, 4395 for glanders in addition to other diseases. The centre generated revenue of Rs. 51.61 lakh from its internal sources, mainly through the contractual diagnostic services.

अश्व-प्रजनन विशेषज्ञों ने भाग लिया।

इस केन्द्र के वैज्ञानिकों ने 20 शोध-पत्र राष्ट्रीय एवं अंतरराष्ट्रीय शोध-पत्रिकाओं में प्रकाशित किए। इसके अतिरिक्त 10 शोध-पत्र प्रकाशन के लिए स्वीकृत किए जा चुके हैं। इसके अतिरिक्त 11 लोकप्रिय-लोकप्रयोगी वैज्ञानिक लेख और 12 संक्षिप्त शोध-पत्र भी वैज्ञानिकों द्वारा प्रकाशित किए गए। वैज्ञानिकों द्वारा 22 विषय-विशेषज्ञ व्याख्यान कृषि विश्वविद्यालयों एवं राष्ट्रीय संस्थानों में आयोजित प्रशिक्षण कार्यक्रमों में प्रस्तुत किए गए। वैज्ञानिकों ने 14 राष्ट्रीय एवं अंतर्राष्ट्रीय सम्मेलनों एवं परिचर्चा-गोष्ठियों में भी भाग लिया। सात वैज्ञानिकों ने विभिन्न प्रकार के प्रशिक्षण एवं पुनश्चर्या पाठ्यक्रमों में अपनी कार्य-कुशलता बढ़ाने के लिए भाग लिया। डा० राजवीर सिंह पवैया, वैज्ञानिक (पशु-विकृति विज्ञान) को भारतीय कृषि अनुसंधान परिषद्, नई दिल्ली द्वारा डाक्टरेट अनुसंधान के लिए जवाहरलाल नेहरू पुरस्कार प्रदान किया गया। डा० ए० अरंगासामी (वैज्ञानिक) के पशु-प्रजनन विज्ञान से संबंधित एक उत्कृष्ट शोधपत्र को लिए जी०बी० सिंह स्मारक पुरस्कार के लिए चयनित किया गया।

केन्द्र ने कई अश्व कल्याणकारी कार्यक्रमों का देश के विभिन्न भागों में आयोजन किया। इसके अन्तर्गत अश्व स्वास्थ्य शिविर एवं किसान गोष्ठियां आदि आयोजित की गई, जिससे अश्व-पालक काशतकारों को अश्वों के रोगों और उनसे बचाव और रोकथाम के उपायों से अवगत कराया गया। इसके अतिरिक्त इन शिविरों में अश्वों की बिमारियों का इलाज, पेट के कीड़ों की औषधि वितरण एवं टिटैनस टीकाकरण आदि कार्य भी किए गए। अश्व स्वास्थ्य एवं उत्पादन कार्यक्रमों के माध्यम से अश्व पालकों को प्रबंधन संबंधी जानकारी दी गई और उनकी अश्व समस्याओं की प्रतिसूचना भी प्राप्त की गई, जिससे इनका केन्द्र में शोध द्वारा समाधान किया जा सके। केन्द्र का स्थापना दिवस 26 नवम्बर, 2006 को बड़ी धूमधाम से मनाया गया। इस उपलक्ष पर विभिन्न राज्यों से लाए गए भारतीय नस्लों के घोड़ों का प्रदर्शन, अश्व खेल एवं घुड़सवारी करतबों का आयोजन भी किया गया।

केन्द्र अश्वों के संक्रामक रोगों के विषय में परामर्श एवं निदान सेवा भी प्रदान करता है। इस कार्यक्रम के अन्तर्गत 5032 अश्व सीरम नमूनों की अश्व संक्रामक एनीमिया और 4395 सीरम नमूनों की ग्लैण्डर रोग के लिए जाँच की गई। केन्द्र ने अपने आन्तरिक स्रोतों से 51.61 लाख रूपयों का राजस्व, जो कि मुख्यतः सलाह एवं निदान सेवाओं द्वारा अर्जित किया।



# INTRODUCTION

The endurance of an organization is based on the foundation of its values. It is strength of this foundation that determines how far it can grow and how long it will endure. For National Research Centre on Equines, this strength emanates from our commitment to improve health and production of equines. They are the basis of our growth and inspire us along every path. India has 1.77 million equines comprising 0.70 million horses and ponies, 0.29 million mules and 0.78 million donkeys. In spite of mechanization of civilization, equines have great relevance, especially for poor farmers and for people living in hilly and difficult terrains, where other means of transport are inaccessible. These animals provide livelihood to the landless, small and marginal farmers and other section of our rural and semi-urban society through draught and transport. Our efforts have been concerted to understand infectious diseases confronting equines and thereby increasing the efficiency and sustainability of equine farming so as to improve health and diminish diseases of equines in India. The vision of the centre is enhanced utilization of equines for agricultural and transport purposes through *in situ* equine development programmes to elevate socio-economic status of under privileged. The approved mandates of the centre are :

The main campus of NRCE is located at

- To undertake research on health and production management in equines;
- To develop diagnostics/biologicals for major equine diseases;
- To act as national referral facility for diagnosis, surveillance and monitoring of equine diseases;
- To provide diagnostic, advisory and consultancy services.

Hisar (Haryana). It has state-of-the-art laboratories for undertaking research in areas of equine virology, bacteriology, parasitology, immunology, pathology, medicine, biochemistry and biotechnology. In addition, NRCE has a sub-campus at Bikaner (Rajasthan) where a new building has come up this year with research laboratories for genetics and breeding, reproduction, physiology and nutrition to undertake research on equine production. National Centre for Veterinary Type Cultures has also been established in the year 2005 at NRCE with a mandate for collection and typing of microbes of veterinary importance. Research activities are carried out by a team of 19 dedicated scientists under the dynamic leadership of Dr. S.K. Dwivedi. The research activities are supported by centralized services like animal and agriculture farms, experimental animal facility, a library and internet facility. The centre has well-maintained herd of Marwari & Kathiawari horses and exotic donkeys at Bikaner. In addition, the centre is in the process of development of BSL-III laboratory, agricultural technology information centre and veterinary type cultures facility.

## OUR MISSION

- Achieving freedom from dreaded equine diseases through development of modern diagnostics and vaccines
- Technology transfer for superior mule and true to breed indigenous horse production in their home tracts using AI & embryo transfer technology with an aim to establish embryo bank of Marwari and Kathiawari horses to enhance export
- Enhancing performance of working equids especially in arid, semi-arid and mountainous regions
- Income generation through market intelligence activities

## Thrust Areas

- ❑ Surveillance and monitoring of important equine diseases including emerging and existing diseases giving special emphasis on indigenous breeds.
- ❑ Development of effective and preferably field based diagnostics and potent immunoprophylactics against major equine diseases threatening equine population in India especially those affecting reproduction, work performance, foal mortalities and morbidities.
- ❑ Development of traditional herbal products for enhancement of performance in equids and management of arthropod borne equine diseases.
- ❑ Identification and demarcation of disease-free zones with reference to some of the important equine diseases endemic in India.
- ❑ To provide diagnostic, consultancy services and training to equine farmers, veterinarians and breeders of India and SAARC countries.
- ❑ Transfer of AI technology for superior quality mule production to end users.
- ❑ *In situ* conservation of true to breed horses/ponies of Indian origin through AI using frozen semen of proven stallions.
- ❑ Improvement and updation of the techniques involving artificial insemination and cryopreservation of the semen and embryos of true to breed indigenous horses.
- ❑ Breed characterization of indigenous breeds of horses.
- ❑ To conduct explorative research for using by-products of equines, namely blood, serum, dung, urine, milk, placenta and hair.
- ❑ To develop balanced ration for equines of different regions based on the local resources for the benefit of underprivileged equine

owners.

- ❑ Studies on utilization of equine energy (donkey, mule & ponies) for agricultural operations and transport purposes using suitable agricultural implements.
- ❑ Promotion of equine sports-related activities at the centre.
- ❑ Achieving the status of 'OIE International referral laboratory' for diagnosis of equine infectious diseases.
- ❑ Commercialization of technologies developed by NRCE with the help of pharmaceutical and drug houses.

## Major achievements of the centre

The centre has made following salient achievements in a short span since its inception:

- ❑ Vaccines for the control of equine diseases: The centre has developed equine influenza vaccine using indigenous isolate (A/equi-2/Ludhiana/87). Improved bacterin and outer membrane protein-based vaccines have been developed for *Salmonella Abortus equi*. Experimental trials of equine herpes virus-1 have been completed and vaccine is on the way to field trials.
- ❑ Disease diagnosis: 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 Agriculture (Government of India). The centre has developed diagnostic kits for equine herpes virus-1 (HERP kit) and *Babesia equi* (COFEB kit) infections. In addition, the centre has developed various tests for diagnosis of equine diseases including equine influenza, equine rhinopneumonitis, equine rotavirus diarrhoea, equine infectious anaemia, equine



piroplasmosis, trypanosomosis, equine viral arteritis, Japanese encephalitis, leptospirosis, mycoplasmosis, streptococci, *etc.*

- ❑ Equine disease surveillance: NRCE is involved in nation-wide monitoring and sero-surveillance of important equine infectious diseases. The database generated on prevalence of equine diseases from different geographical areas is helping in their management. For instance, the centre contributed significantly in the control of equine influenza outbreak of 1987 involving 83000 equines. Effective influenza vaccine was developed subsequent to this outbreak. The equine babesiosis and equine herpes virus infections are endemic in our country in 16 different states of the country. Therefore, development of control strategies against these diseases is one of the priorities of the centre. Control of EIA in India was possible due to timely diagnosis and adopting package of practices formulated by NRCE. The disease has not been reported from India since 1997. Outbreaks of equine glanders in country in 2006 were timely detected and its control measures were taken.
- ❑ Immunobiologicals: Monoclonal antibodies have been developed for diagnosis and characterization of equine herpes, equine influenza and equine rotaviruses. Monoclonals have also been developed against equine chorionic gonadotropin hormone.
- ❑ Molecular characterization of pathogens: DNA finger-printing of EHV-1 virus, sequencing of antigenically important genes of equine influenza virus was done to identify different strains prevalent in equines of India. Sequencing of outer surface proteins (VP4 and VP7) of equine rotaviruses for their genotyping

and molecular epidemiology was done.

- ❑ Repository of veterinary pathogens: The centre has initiated efforts for establishment of a culture repository of veterinary pathogens under Veterinary Type Culture facility.
- ❑ Artificial insemination: The technique of artificial insemination using frozen semen for production of superior quality mules and donkeys has been developed. The true-to-breed germplasm of endangered indigenous horses is being conserved using this technology. Seminal plasma proteins have been isolated, characterized and their role in equine fertility is being evaluated.
- ❑ Indigenous breed characterization: Phenotypic and molecular characterization of indigenous breeds of horses has indicated the existence of genetic variability within Marwari breed and molecular markers for breed identification have been established. Characterization of Manipuri ponies is in progress.
- ❑ Baseline data has been generated on some of the important haematological, physiological and biochemical indices of Marwari and Kathiawari horses, Manipuri ponies as well as local donkeys.
- ❑ Early pregnancy diagnosis: Pregnancy diagnosis between days 14 and 18 post-insemination has been achieved using ultrasonography in donkey and horse mares. An ELISA for pregnancy diagnosis in mares using serum samples has been developed.
- ❑ Donkey fibre has been used to produce carpets by mixing with sheep fibres in the ratio of 40:60.

### Patents

- ❑ Patent has been granted by the Patent Office, Government of India entitled "A method for preparation of a diagnostic kit useful for

forecasting Equine Herpes Virus-1 disease".

- ❑ A patent has been filed for “COFEB-Kit for diagnosis of *Babesia equi* infection in equines”.
- ❑ A patent has been filed for “A method for preparing complement fixation test based (COFEB) kit for the diagnosis of *Babesia equi* infection in equines”.
- ❑ The centre has filed a patent for “A kit for detection of pregnancy in equines and assay thereof”.

### Services

NRCE provides following services to the farmers and equine breeders:

- ❑ Disease diagnostic services for various infectious and non-infectious equine diseases to equine owners, breeders, state animal husbandry departments, police and army organizations.
- ❑ Artificial insemination to augment the production of true-to-breed Marwari horses and superior quality mules and donkeys.

- ❑ Quality jacks and jennies are supplied to various states, breeding societies and farmers, for production of mules and donkeys.
- ❑ Health certification for movement of equines within and outside the country. This facility has helped in promotion of export of horses.
- ❑ Assessment and transfer of technology using the latest know-how of information technology is also given due importance to extend the technologies to the end-users. The scientific and technical staff provides clinical and diagnostic (including pregnancy diagnosis) services and consultancy to the farmers on demand in the areas of equine health and production. Farmers are imparted trainings and supplied education materials for equine management, production and health.
- ❑ Extension activities: To receive feedback from the equine owners, various activities like health camp, awareness and farmers meets are organized on regular basis in different areas of the country.

## STAFF POSITION

Name of the post	Number of posts		
	Sanctioned	Filled	Vacant
Director	1	1	-
Scientific (NRCE)	25	17	8
Scientific (VTC)	10	2	8
Technical	23	22	1
Administrative	11	11	-
Supporting	22	21	1
<b>Total</b>	<b>92</b>	<b>74</b>	<b>18</b>





# EXPENDITURE & REVENUE

Rs. in Lakh

Summary of Expenditure (un-audited figures)		2005-06	2006-07
<b>NON-PLAN</b>			
1.	Establishment charges including LSP/PF, wages, OTA	166.78	165.80
2.	Travelling allowances	2.91	2.90
3.	Others including equipments & recurring charges	69.87	90.79
4.	Works	21.05	44.81
<b>Total</b>		<b>260.61</b>	<b>304.37</b>
<b>PLAN</b>			
1.	Establishment charges including LSP/PF, wages, OTA	0.62	9.36
2.	Travelling allowances & HRD	3.00	3.62
3.	Others including equipments & recurring charges	76.28	193.13
4.	Works	192.19	126.23
<b>Total (NRCE)</b>		<b>271.92</b>	<b>332.47</b>
<b>Total (VTC)</b>		<b>68.79</b>	<b>-</b>
<b>Total Expenditure (Plan and Non-Plan)</b>		<b>601.32</b>	<b>636.84</b>

In Rupees

Summary of Revenue Generation		2005-06	2006-07
1.	Sale of farm produce & auction of dry trees	-	101262
2.	Sale of livestock	-	132500
3.	Sale of publication and advertisements	1560	2300
4.	License fee	58059	67885
5.	Interest on loans and advances	156829	115606
6.	Interest on short term deposits	216914	77593
7.	Leave salary & pension contribution	-	-
8.	Income from internal resource generation	1601350	2359050
9.	Receipt from services	3300	350
10.	Recoveries of loans and advances	-	1467261
11.	Other miscellaneous receipts	953847	837385
<b>Total Revenue</b>		<b>2991859</b>	<b>5161192</b>

## RESEARCH ACHIEVEMENTS

### Evaluation of protective efficacy of indigenously developed EHV-1 vaccine in pregnant mares

An equine herpes virus-1 (EHV-1) killed vaccine incorporating indigenous strain of EHV-1 has been developed by the centre. During the year, protective response of this oil-adjuvanted and mannide monooleate emulsified vaccine prepared from inactivated EHV-1 isolate (Hisar-90-7) was evaluated in pregnant indigenous mares. For testing the potency of the candidate vaccine, 4 mares (Group-I) were inoculated 2 ml of vaccine by intramuscular route. Booster vaccination was done on 10<sup>th</sup> week after primary vaccination. Another 4 pregnant mares (Group-II) were kept as unvaccinated control. Neither hyperthermia nor any untoward reaction was noticed in vaccinated ponies.

Serum samples from experimental mares were collected prior to vaccination and

subsequently at weekly intervals till 15 weeks. Virus neutralizing antibody appeared after first week of the primary vaccination and peak antibody titre (1:32) was observed 3<sup>rd</sup> week post-vaccination. Booster effect of the vaccination was noticed and antibody titre (1:64) was recorded on 14<sup>th</sup> day of the vaccination in Group-I mares (Fig. 1). After virus challenge in experimental mares, the antibody titre increased up to 1:256 on 14<sup>th</sup> day of challenge in vaccinated groups.

For studying the protective efficacy and virus clearance, mares of both groups were challenged with EHV-1 (Raj-98 strain) containing 10<sup>8.5</sup> TCID<sub>50</sub> by intranasal route after 14 days of the booster. Nasal swabs and heparinized blood were collected from all mares for virus isolation every 2<sup>nd</sup> day from day 2 onwards. EHV-1 virus could be isolated from nasal swab of vaccinated mares (Group-I) on 4<sup>th</sup> day from all 4 ponies while on 5<sup>th</sup> day from 2 ponies only. However, EHV-1 could be isolated up to day 7 post-challenge from all the non-vaccinated ponies (Table-1).

After virus challenge, hyperthermia was observed in non-vaccinated control ponies on day 2, while no fever or other reaction was observed in vaccinated mares. On challenge of unvaccinated mares (Group-II), one mare aborted due to EHV-1 (Fig. 2) while one gave birth to a weak foal that died within 24 hrs of birth. EHV-1 virus could be isolated

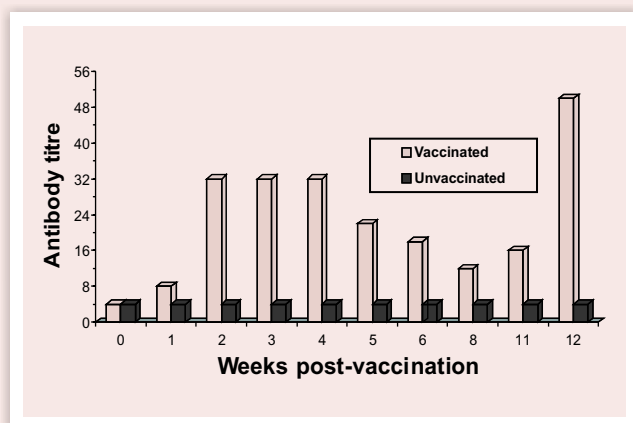


Fig. 1. Virus neutralization antibody response of mares on EHV-1 vaccination

Table 1. Virus isolation from nasal swab of vaccinated and unvaccinated mares on EHV-1 challenge

Group of ponies	Number of mares positive for virus isolation post-challenge					
	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8
Vaccinated (n=4)	0	3	2	0	0	0
Unvaccinated (n=4)	0	2	3	4	4	1





**Fig. 2. Control unvaccinated mare aborted as a result of EHV-1 challenge**

from the lungs and liver tissues of both the foals. Immunoperoxidase staining (IPT) staining of sections of liver and lung tissues of the aborted fetuses and foal (that died within 24 hours from

Group-II) showed presence of EHV-1 antigen. Other two ponies of this group gave birth to healthy foal. On the other hand, out of four EHV-1 vaccinated mares (Group-I), three gave birth to healthy foals even after challenge and only one mare aborted due to EHV-1 infection.

This study indicated that inactivated EHV-1 vaccine developed by this centre is safe and the level of protection of immunized mares after virus challenge was satisfactory, accompanied with good primary and booster humoral antibody response. Before recommending the vaccine for mass scale production and use in field, its immune response in mares needs to be observed in some organized farms in pregnant mares.

**(B.K. Singh, N. Virmani and B.R. Gulati)**

## Development of diagnostics for Japanese encephalitis in equines

Japanese encephalitis (JE) is an endemic disease in several Asian countries including India. Several outbreaks every year are reported from human population in different regions. A study was initiated to develop diagnostics for JE and to know its status & geographic distribution among equines in India.

During the year, haemagglutination inhibition (HAI) assay was standardized using the locally produced HA antigen. Serum samples (n=754) from different states were tested for antibodies to JE sero-complex by HAI and 108 (14.32%) were positive for JE antibodies. Maximum prevalence of JE was observed in equines of Manipur (44 positive out of 48 tested) during the study.

RT-PCR for JEV diagnosis: Primers against different regions of JEV genome including genes for non-structural protein (NS1), envelope (E) and non-translated region (3'-NTR) were designed for standardization of RT-PCR. The 3' NTR-region

primers amplified a *Flavivirus*-specific product of 146 bp of 2 different Flaviviruses tested, while primers directed against E-gene amplified a 291-bp fragment specifically from JEV genome. To determine the sensitivity, serially diluted RNA from



**Amplification of E-gene from serially diluted JEV RNA by RT-PCR**

the infected mice brain tissue ( $10^{6.3}$  LD<sub>50</sub>/20  $\mu$ L) was amplified by RT-PCR. The assay could amplify E-gene amplicon from  $10^{2.3}$  mice LD<sub>50</sub> of the JEV (Fig.). Further work on developing the sensitive and specific assays for JEV is under progress.

**(B.R. Gulati, B.K. Singh and N. Virmani)**

## Recombinant protein based enzyme immunoassay for diagnosis and differentiation of EHV-1 and EHV-4 infections

The GST fusion recombinant protein from fragments of glycoprotein G gene from EHV-1 and EHV-4 were used for standardization of type-specific ELISA for differentiation of EHV-1 and EHV-4. For this, the plates were coated with the known concentration of recombinant proteins. The antibodies from the equine test and control serum were captured on the coated plates followed by detection using anti-horse HRPO conjugate. A total of 209 samples including 27 samples from an organized farm and 182 serum samples from the field were tested for antibodies against both types.

EHV-4 antibodies were found in 9 of 27 (33.33%) samples from organized farm and 162 of 182 (89.01%) samples from the field. Antibodies for both EHV-1 and EHV-4 could be seen in 28 out of 182 samples (15.38%) samples. Eight out of 182 samples had a very high titre of EHV-1 antibodies.

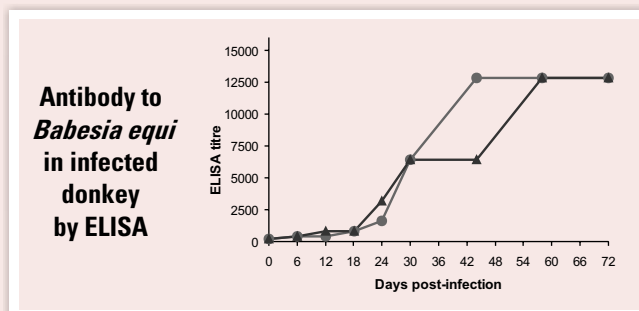
Further work on the sensitivity and specificity of this assay is under progress. This assay will be safe as it does not require handling of any infectious agents.

(N. Virmani, B.K. Singh and B.R. Gulati)

## Development of molecular diagnostics for *Babesia equi*

Equine piroplasmiasis is an important tick-borne haemoprotozoan disease of the equids caused by intra-erythrocytic protozoa, *Babesia equi* and/or *Babesia caballi*. Clinical form of the disease condition is easy to diagnose on the basis of the discrete symptoms and demonstration of the circulating pathogenic parasite in blood smears. Diagnosis of sub-clinical infection is of more relevance and important as these animals aid in the spread of the parasite to the naïve animals and which remain carrier to the *B. equi* parasite throughout their life span. We expressed truncated EMA-2 gene in the *EcoR1* cloning site of the pGEX-4T-1 expression vector and developed ELISA using these purified protein as antigen for detection of specific antibodies against *B. equi* in equine serum. The assay is able to detect *B. equi* specific antibodies and no cross-reaction was observed with *B. caballi*, *Trypanosoma evansi* known positive serum. The assay could detect the infection as early as 6 days post-infection till 90 days of observation period in experimentally infected donkeys (Fig.). The shelf-life of antigen coated plates was tested at 4°C till 6 months and results were satisfactory. Out of 1172 equine sera

tested by ELISA, antibodies to *B. equi* were detected in 610 (52.04%) samples. The important finding of this sero-surveillance was identification of two highly endemic regions i.e. Punjab and



Gujarat where sero-prevalence was more than 80.0%.

We also designed the PCR primers of *B. equi* and *T. evansi* so that PCR amplification can be carried out in one reaction at single annealing temperature. We succeeded in standardization of multiplex PCR and amplified 758 bp (specific to *T. evansi*) and 213 bp (specific to *B. equi*). Further work on sensitivity and specificity of these assays is under progress.

(Sanjay Kumar, Rajender Kumar, A.K. Gupta, S.C. Yadav and S.K. Dwivedi)



## Nation-wide seromonitoring of important equine diseases

During 2006-07, equine disease sero-survey was conducted in various states, namely Delhi, Haryana, Rajasthan, Maharashtra, Andhra Pradesh, Tamil Nadu, West Bengal, Uttar Pradesh,

### Seroprevalence of various diseases among indigenous equines

State	No. of serum samples (positive)		
	EHV-1	<i>B. equi</i>	JE
Haryana	111 (3)	109 (26)	100 (3)
Rajasthan	231 (10)	231 (68)	180 (11)
Uttarakhand	126 (3)	126 (86)	-
Punjab	217	217 (182)	-
MP	142 (3)	166 (51)	153 (11)
Chattisgarh	105 (1)	105 (46)	105 (8)
Manipur	50	50 (9)	48 (44)
Gujarat	168 (5)	168 (142)	168 (31)
<b>Total</b>	<b>1150 (25)</b>	<b>1172 (610)</b>	<b>754 (108)</b>

Karnataka, Chandigarh, Madhya Pradesh, Punjab, Chattisgarh, Gujarat, Manipur and Uttarakhand.

For equine infectious anemia (EIA), 5032 serum samples from thoroughbred as well as indigenous equines were examined by Coggins test, however, none of the samples tested was found positive. None of the samples have been reported positive for EIA since 1998-99. None of the 1141 serum samples from indigenous equines was positive for equine influenza (EI) by haemagglutination-inhibition (HAI) assay. Similarly, none of the 446 serum samples tested was found positive for African horse sickness (AHS). Similarly, *Salmonella Abortusequi* (H-antigen) was not detected in any of the 1174 equine serum samples tested (Table).

Antibodies to Japanese encephalitis (JE) were detected in 108 out of 754 equine sera by HAI. Out of 1150 equine sera, 25 (2.2%) samples were positive for EHV-1. Out of 1172 serum samples tested for babesiosis, 610 samples were positive by recombinant protein based ELISA giving a point prevalence of 52%.

(S.K. Dwivedi, S.K. Khurana and others)

## Occurrence of glanders in Maharashtra, Punjab, Uttarakhand and Uttar Pradesh

Serum samples and swabs were received in last week of July, 2006 from Regional Disease



A horse affected with glanders showing skin nodules

Diagnostic Laboratory (RDDDL), Pune, Maharashtra. On laboratory testing, one pus sample was positive for *Burkholderia mallei*. Two serum samples were also positive for glanders by complement fixation test (CFT). Following this, a team was sent for investigation to Maharashtra. Investigation by NRCE team was done in Pune and Panchgani area where diseased cases were present (Fig). Serum, nasal swab and pus swab samples were collected at the spot. Samples were also received from RDDDL, Pune. In first lot, of the 13 samples tested, 10 were positive for glanders by CFT; one showed equivocal result later confirmed as positive; one showed negative result but was found positive in subsequent testing. Three nasal swabs also showed

presence of the pathogen. In all, 23 out of 357 samples were positive for glanders from Maharashtra. Since the outbreak of July-August 2006, no new cases have been detected from Maharashtra, indicating the control of the disease in

the states due to timely efforts of NRCE in coordination with state animal husbandry in departments. Besides Maharashtra, 70 cases of equine glanders have been reported from Uttar Pradesh, one from Uttarakhand and three from Punjab during this period (Table). During the year, a total of 4395 equines have been tested for glanders from the entire country and 97 were detected positive. The causative agent, *Burkholderia mallei* was isolated from eight of these cases. NRCE is continuously monitoring the situation in the entire country, with particular emphasis on the equines of Uttar Pradesh and Uttarakhand.

**Glanders cases in equines reported during 2006-07**

State	Number of samples	
	Tested	Positive
Maharashtra	357	23
Uttar Pradesh	548	70
Punjab	723	3
Uttarakhand	164	1
<b>Total</b>	<b>1792</b>	<b>97</b>

(P. Malik, S.K. Khurana and S.K. Dwivedi)

## GLANDERS RESURGENCE IN INDIA

### WHAT IS GLANDERS

A contagious bacterial disease of horses, mules and donkeys, characterized by serial ulcerating nodules in the upper respiratory tract, lungs and skin. Humans can get the infection through cut in skin or mucous membranes and by inhalations.

### CAUSE

Gram-negative bacteria *Burkholderia mallei*

### SYMPTOMS

In equines, high fever (up to 106°F), thick mucopurulent nasal discharge, respiratory signs, nodules and ulcers in nasal septum, lungs and/or skin along lymph vessels. Animal gets debilitated and death may occur.

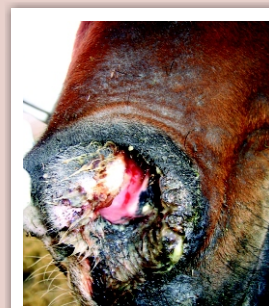
Humans develop fever, nodules and ulcers in skin, nose and lungs.

### STATUS IN INDIA

Sporadic occurrence among equines in Maharashtra, Uttar Pradesh, Uttarakhand and Punjab during 2006-07.

### TREATMENT AND CONTROL

No treatment done in horses. Early detection and elimination as per Glanders and Farcy Act, 1899. No vaccine available.



**Mucopurulent exudate and ulcer on nasal septum**

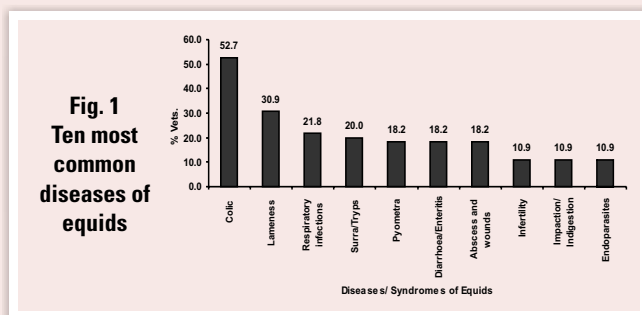


## Anitmicrobial drug use in equids and emergence of multiple drug resistant bacteria

This study was initiated to select the drug resistant sentinel bacteria in equines to determine the evolution of the antimicrobial drug resistance and to predict the outcome of antibacterial drug use in equids, on the basis of antimicrobial drug resistance (AMDR) profile of the selected sentinel bacteria.

Information on equine problems and antimicrobial drug (AMD) usage in equine patients was collected through a questionnaire sent to pan-India veterinarians. The salient conclusions from the survey of 359 respondents are:

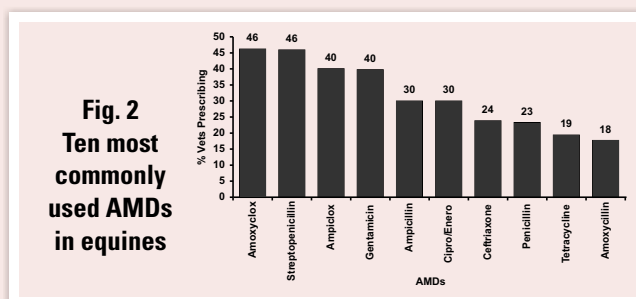
- The most common diseases reported by veterinarians treating equines are: Dystokia, infertility, abortions, thrush, fractures, canker, foal diarrhoea, strangles, lameness and babesiosis (Fig 1).
- The most common AMDs and their combinations prescribed by equine



veterinarians are penicillins, tetracyclines, gentamicin, cotrimoxazole, cefotaxime, ampicillin+gentamicin and metronidazole (Fig 2). Intramuscular route of AMD administration remained the most preferred one (>83%). Adverse effects of sulphadimidine, gentamicin and metronidazoles were more often reported. For prescribing AMDs, 78-89% veterinarians gave weightage to diagnosis, 9-12% to prognosis and 18-26% to clients' economy. More than 75% veterinarians desired that AMD sensitivity of the bacterial isolates is needed for effective treatment.

During the year, a total of 198 samples from

equids and their environments were collected and 512 bacterial isolates were picked up and attempts were made for their preliminary characterization. Of these, Gram-positive bacterial isolates (234) were predominated by *Enterococcus* spp. AMD resistance assay revealed that emipenem and chloramphanicol were the most effective drugs inhibiting nearly 75% and 66% isolates,



respectively. It was observed that addition of antibiotics in media may reduce the recovery rate of even drug resistant isolates. It was revealed that resistance to one drug could be correlated with resistance to other drugs.

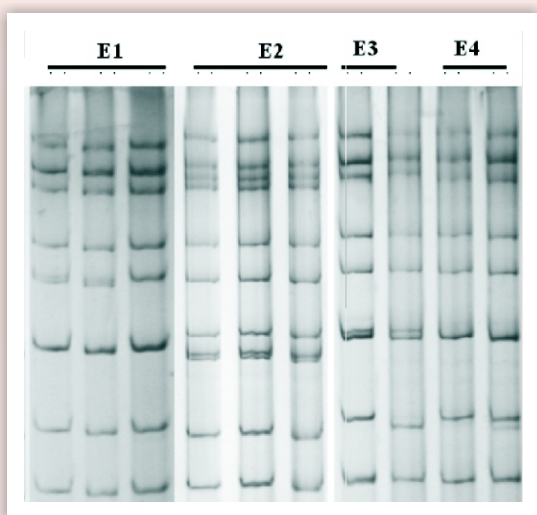
Resistance to any one member of a group of drugs was associated with resistance to similar drugs of the group. For example, in Gram-negative bacteria resistance to ampicillin was strongly associated with carbenicillin, cotrimoxazole, cotrimazine and ciprofloxacin resistance. This indicates that genes for resistance for these drugs might be moving together or might be closely located and are transferred in islandic form.

From analysis of 198 samples, 278 isolates of Gram-negative bacteria belonging to six different genera (*Escherichia*, *Klebsiella*, *Proteus*, *Enterobacter*, *Aeromonas* and *Edwardsiella*) could be isolated. MDR was more common in Gram-negative bacteria (particularly in *E. coli* and *Klebsiella* spp isolates) than in Gram-positive bacteria. Isolates were resistant to 1-13 drugs of the total 14 AMDs tested. Drug resistance was more common in isolates from equids maintained on organized farms.

(B.R. Singh, S.K. Khurana, B.R. Gulati, Mamta and N. Lal)

## Characterization of equine rotaviruses isolated from diarrhoeic foals

Molecular epidemiology of rotaviruses causing diarrhea in foals in two organized farms in northern India during the period 2003-06, was studied. The equine rotavirus isolates were characterized on the basis of their viral RNA electrophoretic mobility in polyacrylamide gel electrophoresis (PAGE). Of the 164 diarrhoeic stool samples, 58 (35.36%) were positive for rotavirus and their RNA exhibited at least 5 different electropherotypes (E1-E5) (Fig).



Electropherotypes of equine rotaviruses

The serotyping of the equine rotavirus isolates was done by serotyping ELISA employing monoclonal antibodies (MAbs) and sequence analysis of the genes encoding the outer capsid proteins (VP7 & VP4) in collaboration with Indian Institute of Sciences, Bangalore. Strains belonging to different electropherotypes exhibited either different serotype/genotype specificity or a lack of reactivity to typing MAbs used in this study. Characterization of twenty one isolates revealed that E1, E2, E4 and E5 strains exhibited G10, G3, G6 and G1 type specificities that accounted for 19.0, 42.9, 14.3 and 9.5% of the isolates (Table). Although the E1 strains possessed genes encoding G10 and P6 [1] type outer capsid proteins, unlike the G10, P8 [11] type strain I321, yet they exhibited high cross-reactivity with the G6-specific MAb suggesting that the uncommon combination altered the specificity of the conformation-dependent antigenic epitopes on the surface proteins. Strains belonging to electropherotypes E3, accounting for 14.3% of the isolates, could not be typed.

(B.R. Gulati and B.K. Singh)

Electropherotypes and serotypes/genotypes of the equine rotavirus isolates

Electropherotype	Serotype/ Genotype	No. of ERV isolates	G Serotype determined by	
			ELISA	Sequencing
E1	G10, P6[1]	4	+*	+
E2	G3, P[?]	9	+	+
E3	UT	3	-	ND
E4	G6	3	-	+
E5	G1, P[?]	2	+	ND

\*Reactivity with G6 MAb, UT-un-typed, ND-not done





## A survey of management practices adapted by mule producers in northern India

The mules are widely used for carrying men and luggage in the hilly terrains and for pulling carts in the plains. Some people living on the banks of Yamuna and *Ganga* river rear mares for mule production. They rear the foals from birth till 4-6 month of age and then before selling these foals. The buyers rear them till adult stage and then sell them out to the actual users, who use these mules for load carrying activities. The study was planned to know socio-economic status as well as the practices followed by the mule producers in Haryana, Uttar Pradesh and Uttarakhand state (n=50). People engaged in mule production generally belong to Muslim community and are locally called as *Seikhs* or *Kalandars*. Literacy rate is poor (6%). Major occupation of these people is mule production. Mean family size was observed as  $8.48 \pm 0.40$  (range 4 to 15). The average age of mule producers was  $41.55 \pm 1.86$  (range 25-65 year). Twenty six percent of mule producers rear the mares on sharing basis and they equally share the amount received from the sale of mule foal with the owner of the mare. The average cost of the pony was reported as Rs.  $21,406 \pm 1596$  (range Rs 2500-82000). The mule producers sell their mule foals in local animal fairs and selling price varies from 10,000/- to 40,000/- depending upon the body size, vigor and colour of the mule foal. Most of the mule breeders were of the opinion that mule produced through AI with the frozen semen of superior jacks provided by the NRCE fetches high price in the market as compared to the mules produced from the local jacks available with the equine owners.

During raining season, 24% mule producers migrate to the areas where they can have plenty of green fodder for their equines. One mule producer generally rears on an average  $9.26 \pm 0.98$  (range 1 to

30) non-descript mares/ponies mares. Eighty two percent farmers rear their mares absolutely on grazing, while 18% mares are stall fed. These mares are exercised 1 to 2 hour daily. They provide *doob* and *nari* as green fodder during rainy season and their hay during winter season and other lean months when there is scarcity of green fodder. No concentrate is being offered to the mares in most of cases. Donkey stallions are provided 3 to 4 kg concentrate/day for a period of 4 months in the winter. Four to five families collectively bear the expenses of concentrate fed to the donkey stallion. Four to five families collectively have one teaser to detect the mares in estrus.

Average age at covering of mares used for mule production was 2.5 years (range 2 to 4). Average age at foaling was observed as 3 years (range 33 to 48 months). The mares in heat were covered by the donkey stallion in morning and evening on day 3<sup>rd</sup> and the in the morning of 4<sup>th</sup> day. Mean age at which donkey stallions were used for breeding was 2.5 years (range 2 to 3). The loss of libido was another problem reported by the equine owners. The reason is excessive use of stallion as well as the use of donkey stallion at an early date before attaining maturity. The donkey stallion becomes adult at an age of 3-3.5 year.

Most of the mule producers do not allow the foals to suckle the first milk i.e. colostrum as they are of the view that colostrum feeding creates problems of diarrhea and death in foals. They have reportedly observed deaths in mule foals due to neonatal isoerythrolysis i.e. incompatibility between blood factors of foal antibodies in mare colostrum. Most owners do not use their mares for work, except for bringing the fodder from the field.

**(R.A. Legha and Yash Pal)**

## Effect of stallion seminal plasma binding proteins on *in vitro* fertilizing ability of spermatozoa

Diagnosis of fertility potential of a semen sample is essential for consistently high reproductive efficiency. The study was envisaged to isolate and purify fertility related (heparin and gelatin binding) proteins from equine seminal plasma and to assess them by *in vitro* fertilization test. The effect of the isolated heparin (HBP) and gelatin (GBP) binding stallion seminal plasma proteins on induction of acrosome reaction (%)

**Table 1. Effect on induction of acrosome reaction during *in vitro* capacitation**

Protein concentration	Acrosome reaction (In % at 4 h.)	
	HBP	GBP
20 µg	20.50 ± 1.28*	19.79 ± 1.63
40 µg	22.13 ± 1.27**	18.68 ± 1.59
60 µg	21.55 ± 1.66*	20.54 ± 1.63
Control	16.31 ± 1.02	16.31 ± 1.02

Means difference between treated and controls (P<0.05)\*, (P<0.01)\*\*

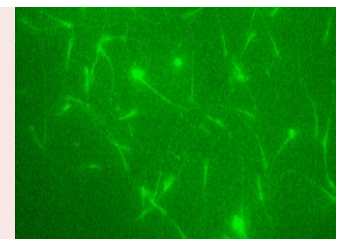
during the *in vitro* capacitation (4 hr) was assessed. Significant difference was observed in the percentage of acrosome reacted spermatozoa at 4 hr incubation between HBP treated and control groups (Table 1). The GBP treated group showed non-significant differences.

The effects of the isolated HBP and GBP (20

µg, 40 µg and 60 µg) on hypo-osmotic swelling test were evaluated. The percentage of hypo-osmotic swollen positive sperms was higher in HBP and GBP protein treated samples than control. The 40 µg HBP treated groups showed significant differences (P<0.01) with control groups in the percentage of hypo-osmotic swollen sperms at 50 and 100 mosmol of osmotic pressure. In the GBP treated group, 40 µg showed significant difference with control group at 50 mosmol (P<0.01) (Table 2).

A random sampling of six samples each from Marwari stallion and donkey stallions were used for the acrosomal integrity test. Acrosomal status was assessed with fluorescein isothiocyanate-conjugated *Pisum sativum* agglutinin lectin (FITC-

**Stallion spermatozoa stained by FITC-PSA Lectin**



PSA lectin) and observed by fluorescence microscopy (Fig). The results revealed that overall intact acrosome percentage was 78.15 ± 3.02 and 84.22 ± 0.99 in Marwari and donkey stallions, respectively.

(A. Arangasamy)

**Table 2. Effect of Heparin (HBP) and gelatin (GBP) binding seminal plasma proteins on hypo-osmotic swelling of equine spermatozoa**

Protein (µg/ml.)	Hypo-osmotic swollen sperms (%) at		
	50 mosmol.	100 mosmol.	150 mosmol.
HBP (20 µg)	21.31 ± 1.38	20.56 ± 1.58	21.48 ± 1.52
HBP (40 µg)	22.92 ± 0.52 **	22.49 ± 1.20	23.04 ± 0.50 **
HBP (60 µg)	20.15 ± 0.62	19.08 ± 0.72	20.37 ± 0.72 **
GBP (20 µg)	22.43 ± 1.88	21.67 ± 1.82	19.26 ± 0.90 *
GBP (40 µg)	22.68 ± 0.57 **	22.71 ± 1.47	21.12 ± 1.66 *
GBP (60 µg)	20.45 ± 0.69 *	19.72 ± 0.85	20.69 ± 1.04
Control	18.24 ± 1.18	20.85 ± 0.75	17.31 ± 0.48

Means bearing different superscripts in a column differ between protein treated and controls (P<0.05)\*, (P<0.01)\*\*



## Physical and biochemical comparison of semen of horse and donkey stallions

To compare the physical and biochemical parameters of seminal plasma of Marwari stallions (n=42) and exotic donkey stallions (n=50), the semen samples were collected using artificial vagina. Total semen volume was recorded as  $76.90 \pm 5.52$  and  $70.6 \pm 4.07$  ml in horse and donkey stallions, respectively. Average gel free semen volume was  $53.69 \pm 2.64$  and  $51.30 \pm 3.56$  ml in horse and donkey stallions, respectively. Progressive motility (%) in gel free semen was recorded as  $73.33 \pm 0.94$  and  $77.80 \pm 1.00$  in horse and donkey stallions, respectively. The details of comparison of physical semen

parameters are given in Table 1.

Activity of GOT, GPT and LDH was 174.3, 26.08 and 1552 IU/l in seminal plasma of Marwari horses and 387.7, 28.2 and 3913 IU/l in seminal plasma of donkey stallions, respectively. Glucose (mg/dl), total protein (g/dl), cholesterol (mg/dl) and triglycerides (mg/dl) contents were 23.36 and 24.44, 1.25 and 3.63, 64.72 and 74.65, 39.57 and 61.72 in seminal plasma of horse and donkey stallions, respectively. The details of metabolites content and activity of enzymes in marwari and exotic donkey stallions are given in Table 2.

(Yash Pal and R.A. Legha)

**Table 1. Macroscopic and microscopic evaluation of Marwari and exotic donkey stallions**

Parameter	Mean $\pm$ SE (Range) semen values in	
	Marwari Horse	Exotic Donkeys
Total volume (ml)	$76.90 \pm 5.52$ (30-225)	$70.6 \pm 4.07$ (5-150)
Gel volume (ml)	$23.21 \pm 6.48$ (0-200)	$19.0 \pm 3.45$ (0-95)
Gel free volume (ml)	$53.69 \pm 2.64$ (25-90)	$51.30 \pm 3.56$ (15-125)
pH	$07.20 \pm 0.02$ (7.0-7.5)	$07.17 \pm 0.02$ (7.0-7.5)
Initial motility (%)	$79.76 \pm 0.93$ (70-90)	$82.60 \pm 1.02$ (60-90)
Progressive motility (%)	$73.33 \pm 0.94$ (60-90)	$77.80 \pm 1.00$ (60-90)

**Table 2. Seminal plasma evaluation of Marwari horse and exotic donkey stallions (n=24)**

Parameters	Marwari Horse (Mean $\pm$ SE)	Exotic Donkey (Mean $\pm$ SE)
Glucose (mg/dl)	$23.36 \pm 2.05$	$24.44 \pm 1.81$
Cholesterol (mg/dl)	$64.72 \pm 10.23$	$74.65 \pm 9.70$
Total protein (g/dl)	$01.25 \pm 0.19$	$03.63 \pm 0.50$
Triglycerides (mg/dl)	$39.57 \pm 8.35$	$61.72 \pm 7.31$
GOT (IU/L)	$174.3 \pm 6.94$	$387.7 \pm 28.2$
GPT (IU/L)	$26.08 \pm 6.56$	$28.2 \pm 3.96$
LDH (IU/L)	$1552 \pm 114$	$3913 \pm 487$

## Cryopreservation of Marwari stallion semen and fertility trials by artificial insemination

The semen of Marwari stallions was cryopreserved using mobile laboratory at farmers' door. To avoid the use of bio-freezer in field condition, the semen was cryopreserved on LN<sub>2</sub> vapors. For this, the diluted semen filled straws were kept on LN<sub>2</sub> vapors for 5, 8, 10, 12 and 15 min and then plunged in the liquid nitrogen and finally shifted to the LN<sub>2</sub> container for storage. Pre-freezing motility of the semen was 75.0 ± 2.67%. Post-thaw semen motility was observed as 11.25 ± 1.25, 13.75 ± 1.83, 30.62 ± 4.77, 41.25 ± 3.50 and 10.0 ± 1.34% at 5, 8, 10, 12 and 15 min. vapor exposure, respectively. The semen cryopreserved on LN<sub>2</sub> vapors for a period of 12 min gave satisfactory results, without cracking of straws during the process. Semen freezing using bio-freezer and freezing on LN<sub>2</sub> vapors indicated that the post-thaw semen motility 36.25 ± 1.75 and 38.75 ± 2.76%,

respectively. It was observed that results obtained using both techniques are comparable and the technique of freezing on LN<sub>2</sub> vapors can be useful for freezing of semen in field as well as in small laboratories.

Very few superior quality Marwari stallions are available in India. Such stallions were selected by field surveys in Rajasthan and semen was collected and cryopreserved at farmers' door. This cryopreserved semen was used for insemination of Marwari mares to produce superior offspring. On insemination of 10 Marwari mares at our campus, 9 conceptions were reported. Out of these, two have conceived using the semen of an elite Marwari stallion named Rajrattan (height 165 cm). AI of 14 Marwari mares in field at farmers' door resulted in 7 conceptions.

**(Yash Pal, R.A. Legha and A. Arangasamy)**

## Cryopreservation of semen of exotic jacks and artificial insemination in field for superior mule production

During the period under report, the semen of seven exotic jacks was collected, examined and preserved for doing artificial insemination (AI) in the farm as well as field animals. A total of 400 semen doses of exotic jacks were cryopreserved (with assistance from Dr. R.C. Sharma & others). A trial of cryopreservation of exotic jack's semen on LN<sub>2</sub> vapours was conducted. The semen filled straws were put on liquid nitrogen vapours for 8, 10 and 12 min. Post-thaw motility of semen was observed as 22.5 ± 1.21 (15-30%), 40.31 ± 2.16 (20-50%) and 53.73 ± 2.26 (35-65%) for 8,

10 and 12 min, respectively. The results of semen freezing by using liquid nitrogen and bio-freezer were comparable. The post-thaw motility after 12 min on LN<sub>2</sub> vapours and bio-freezer was 53.73 ± 2.26 (35-65%) and 54.38 ± 1.28 (45-60%), respectively.

A total of 14 mares were inseminated for mule production in selected villages of district Panipat (Haryana), Mujaffarnagar (UP) and Udham Singh Nagar (Uttarakhand).

**(R.A. Legha, A. Arangasamy, Yash Pal and J. Singh)**



## Isolation of *Rhodococcus equi* from equines and their antimicrobial sensitivity

Forty eight samples (nasal, faecal, soil) collected from different parts of Haryana were processed for isolation of *Rhodococcus equi*. These included 22 nasal swabs (including 18 from foals with respiratory problem and 4 from in-contact apparently healthy foals), 20 faecal samples (including 18 from foals with respiratory problem and 2 from in-contact apparently healthy foals) and 6 soil samples from contaminated premises. In all 4

isolates of *Rhodococcus equi* were obtained from foals with respiratory problems. These 4 isolates were subjected to *in vitro* antibiotic sensitivity testing with 17 antimicrobial and they were found to be sensitive to chloramphenicol, erythromycin, oxytetracycline, ciprofloxacin, neomycin and rifampicin.

(S.K. Khurana, P. Malik , B.R. Singh and N. Virmani)

## Cloning and expression of surface glycoprotein gene of *Trypanosoma evansi*

For development of sensitive and specific diagnostic for trypanosomiasis in equines caused by *Trypanosoma evansi*, the Ro Tat 1.2, a predominant variable surface glycoprotein (VSG) was selected. A gene fragment (762 bp) of Ro Tat 1.2 gene from the Indian strain of *T evansi* was amplified and sequenced. This gene segment was cloned in expression vector (pGEX 4T-1) and subsequently expressed by transformation into *E. coli* competent cells. The protein of 52 kDa was purified and characterized (Fig).

Standardization of ELISA employing this



recombinantly expressed antigen is in progress.

(Rajender Kumar, Sanjay Kumar, S.C. Yadav and S.K. Dwivedi)

## INTER-INSTITUTIONAL COLLABORATIVE RESEARCH PROJECTS

The centre has developed effective linkages with various national institutes and research organizations. These linkages are aimed at overall improvement in the health and production potential of equines and for meeting the stringent criteria for international movement of horses.

The centre has efficient and effective linkages with various organizations involved in

breeding and production of equines, like RVC of Indian Army, riding and race clubs, turf clubs, equine breeders and breeders' associations. These linkages are also utilized for the validation of research findings and for conducting collaborative researches. Research achievements of three collaborative research projects currently in operation at the centre are presented here.

## Usefulness of recombinant protein for serodiagnosis of glanders

A research project in collaboration with Defence Research & Development Establishment, Gwalior has been started to evaluate the usefulness of recombinant protein for serodiagnosis of glanders. This study will help in development of superior diagnostics for glanders, a resurgent disease of equines.

Glanders is primarily a disease of equines caused by the gram-negative bacillus *Burkholderia mallei*. It is a notifiable disease in India. The disease is of zoonotic significance as it may be transmitted to human beings subsequent to handling of infected animals resulting in a fatal disease. A recombinant protein of *Burkholderia mallei* was expressed from a conserved gene sequence by DRDE and it showed immunogenic potential in preliminary studies at

DRDE/NRCE. This protein appears to have a scope for use in the development of a sensitive and specific diagnostic tool for detection of glanders. Considering this, NRCE was entrusted the responsibility of evaluation of the recombinant protein for its diagnostic application. Initially, a total of 570 serum samples, including both known positive and negative results, were selected and retested by CFT for confirmation of the results. Of the 570 samples selected, 18 were confirmed positive with titres ranging between 1:8 and 1:32. The recombinant protein is being evaluated for use in CFT and for standardization of an indirect ELISA.

(P. Malik and Santosh Kumar)

## Molecular characterization of indigenous breeds of equines

A research project entitled 'Molecular characterization of indigenous breeds of horse for genetic diversity within and between different breeds' was initiated in collaboration with National Bureau of Animal Genetic Resources, Karnal. The objective of the study is to differentiate indigenous breeds of horses at molecular level. During the year,



biometric indices of Manipuri ponies were recorded to established base line data. For this, fifty Manipuri ponies were selected on the basis of their phenotypic characteristic. Data of biometric indices

of these horses revealed that height at wither in both the sexes ranged from 119-134 cm indicating that this breed of horses comes under the categories of ponies as it does not qualify the minimum standards of 150 cm height to be clubbed under the category of horse. Average values of other indices, irrespective of sex, in Manipuri ponies recorded were body length (129.5 cm), heart girth (143.6 cm), hind leg length (82.3 cm), foreleg length (78.4 cm), height at knee (39.4 cm), face length (58.2 cm), face width (20.2 cm), ear length (14.9 cm), ear width (12.8 cm) and gap between poles (14.4 cm).



The sex wise average values with standard deviation of adult animals have been presented in

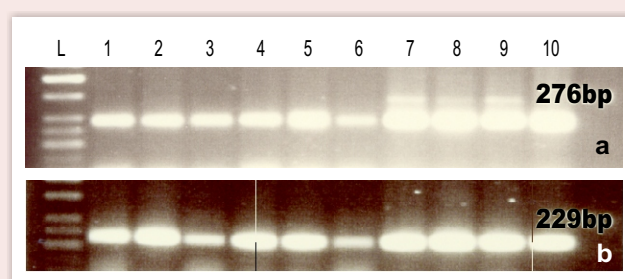
Table 1.

Table 1. Biometrical parameters of Manipuri ponies (cms)				
Biometrical parameters	Mean value (Stallions)	Range	Mean value (Mares)	Range
Body length	129.2 ± 4.229	119-136	129.96 ± 5.062	122-138
Height at wither	129.1 ± 3.917	119-134	129.0 ± 4.286	120-136
Leg length-fore	78.17 ± 4.752	71-95	78.56 ± 2.959	72-85
Height at knee	39.48 ± 2.113	36-45	39.40 ± 1.871	37-43
Leg length- hind	83.05 ± 3.237	77-87	81.64 ± 4.563	72-90
Height at hock	45.12 ± 2.559	39-50	43.80 ± 2.843	39-49
Heart girth	143.70 ± 5.765	132-152	143.40 ± 7.030	127-160
Face length	58.58 ± 1.910	55-63	57.80 ± 2.986	51-65
Face width	20.12 ± 0.900	19-22	20.28 ± 1.100	19-22
Ear length	14.52 ± 1.048	13-17	15.20 ± 0.957	13-17
Ear width	12.56 ± 1.236	8-14.5	13.08 ± 1.115	11.5-16
Gap between poles	14.08 ± 0.917	13-16	14.70 ± 1.307	13-17

(A.K. Gupta, S.C. Gupta, S.N. Tandon, Mamta, Neelam Gupta and P. Malik)

## Genotyping of major histocompatibility complex class-II genes in Marwari horses

During this period, 28 blood samples of Marwari horses (16 from field and 12 from EPC farm) were collected for undertaking the studies on major histocompatibility complex (MHC) class-II genes. The DNA was isolated from the blood samples by the phenol-chloroform extraction method followed by ethanol precipitation. The PCR conditions for amplification of MHC class-II genes (DRB-2a and 2b) were standardized using primers Be1 and Be2. The fragments of 276 bp and 229 bp were successfully amplified using Be1 and Be2 primers, respectively from total of 49 DNA samples



Amplicons of DRB 2a gene using Be1 (a) and Be2 primers (b)

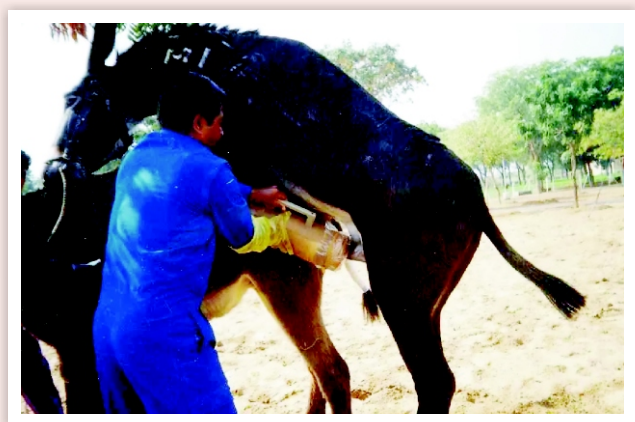
of Marwari horses. Further work is under progress to study heterozygosity at these loci at population level.

(R.C. Sharma and S.C. Mehta)

# TECHNOLOGY ASSESSMENT & TRANSFER

## Field assessment of technology for superior mule production by cryopreserved semen of exotic jacks

Mules are used extensively for pack and transportation by the military and civilians in the hills and inaccessible terrain. However, mules produced by the equine owners in these areas are of low productivity and strength. The paucity of good



**Semen collection from jack**

quality jacks is the major hindrance in producing quality mules in the field. Inseminating the farmer's mares with frozen semen of good quality exotic jacks can solve this problem. With a view to improve the socio-economic status of poor equine owners and landless laborers possessing ponies, donkeys and mules, the centre developed technology for superior mule production. For this purpose, the semen characteristics of jacks including exotic Poitu jacks were evaluated and their semen freezing protocols were standardized. In addition, in experimental trials, the conception rates in artificially inseminated experimental farm mares with cryopreserved semen were estimated. After encouraging results in the farm trials, the technology was disseminated in different areas for superior mule production in field.

**Evaluation of Semen:** The semen quality of exotic

jacks (8) was evaluated by gross and microscopic examination. Average total jack semen volume per ejaculation was  $49.58 \pm 0.84$  ml. The mean gel free semen volume per collection was  $45.04 \pm 0.60$  ml. The means of initial and progressive sperm motility were  $84.31 \pm 0.27\%$  and  $75.43 \pm 0.30\%$ , respectively. Mean sperm concentration was  $293.39 \pm 4.99 \times 10^6$  per ml. The overall live sperm count observed was  $77.47 \pm 0.36\%$ . The quality of the semen collected during breeding season (March to October) was better than that collected in non-breeding season.

**Cryopreservation of jack's Semen:** The semen having more than 70% progressive sperm motility was processed for freezing. Cryopreservation trials were conducted in programmable cryo-freezer with two primary extenders namely (a) Dry skim milk-glucose medium and (b) Citrate EDTA medium and



**AI in field using frozen semen**

secondary extender viz., (a) Lactose-dry skimmed milk-glucose egg yolk media and (b) Glucose-EDTA-egg yolk media. The results of citrate EDTA as primary extender and lactose- glucose-EDTA-egg yolk as secondary extenders were good and





post-thaw sperm motility achieved was as high as 55%. Using these selected extenders, jack semen was cryopreserved for experimental and field trials. Methodology for freezing the jack semen in field over liquid nitrogen vapors has also been standardized. A stock of more than 500 cryopreserved jack semen doses is regularly maintained at the centre.

**Field trials:** In experimental fertility trials using cryopreserved jack semen in the mares at our equine production campus, the conception rate was about 52%. Field trials of this technology were done in selected villages of Haryana, Uttar Pradesh and

Uttarakhand. Mares in these areas were examined by ultrasonography and selected mares in estrus were artificially inseminated twice on 4<sup>th</sup> and 6<sup>th</sup> day of estrous. A total of 254 mares were inseminated. A follow up of 189 mares confirmed pregnancy in 79 of these mares, giving a conception rate of 42% in field trials.

These findings established that using the technique developed at our centre for cryopreservation of jack semen, a conception rate of 42-52% can be achieved and mules so produced are of superior quality.

## External validation of kit for pregnancy diagnosis

The centre has previously developed an eCG-based ELISA for pregnancy diagnosis. This kit can detect the pregnancy between day 35 and 120 of gestation for those mares which have been covered by horse stallion only. Internal validation of this kit has been done that indicated 100% sensitivity of this assay. During the year, the external validation of the assay was initiated and the

results of external validation from one source i.e. Equine Breeding Stud, Hisar confirmed the 100% sensitivity of the assay. Using the kit, the pregnancy diagnosis services was offered to equine owners and results of testing of 181 equine mare serum samples for pregnancy were communicated to the equine owners.

# CONSULTANCY, PATENTS AND COMMERCIALIZATION OF TECHNOLOGY

## Institute technology management committee

An institute Technology Management Committee has been constituted under the chairmanship of Dr. S.K. Dwivedi, Director. The committee will be responsible for the management of intellectual property rights portfolio and commercialization of IPR-enabled technologies. Other members of the committee include: Dr. A.K. Gupta (Principal Scientist), Dr. B.K. Singh (Principal Scientist), Dr. R.C. Sharma (Senior Scientist) and Dr. Rajender Kumar (Senior Scientist).

## Consultancy

This centre offers consultancy and diagnostic services for investigation of equine diseases in the country. Under this programme, equine disease investigation is done on-farm by sending teams of experts in different parts of the country. In addition, veterinarians, equine owners and quarantine officers submit samples to the centre for disease investigation. The results of such investigations are conveyed along with advice to the concerned for taking necessary measures for treatment and control of equine diseases.

During 2006-07, a total of 5032 equine serum samples received from quarantine, thoroughbred and indigenous equines were examined for equine infectious anemia (EIA) by Coggins test. None of the samples tested was positive for EIA. In addition, 4395 equines serum samples were received in the centre for Glanders testing. On testing by CFT for glanders, 97 serum samples received from Maharashtra, Punjab, Uttarakhand and Uttar Pradesh were found positive for glanders.

A total of 203 vaginal/preputial swabs received from quarantine stations (177 from Delhi and 26 from

Chennai) were tested for contagious equine metritis (CEM) by agent isolation & identification and all samples were found negative for CEM.

Bacteriological examination of 142 samples including nasal swabs, vaginal swabs, ocular swabs, faecal samples, tissues, exudates, pus samples and aborted foetus yielded 16 isolates (Table).

During this period, necropsies on equines were conducted and the pathological conditions diagnosed were fibrinous pleurisy and pneumonia (1), passive venous congestion (1), pneumonia (1), abortion due to placentitis (1), abortion due to mechanical twisting of the chord (1), acute tubular nephrosis and encephalopathy (1), shock leading to disseminated intravascular coagulation (1), hepatocellular adenoma/ carcinoma and portal cirrhosis of liver accompanied with encephalopathy (1) and eosinophilic enteritis coupled with allergic bronchitis and myocarditis (1).

## Commercialization of technology

The technologies developed at the centre including diagnostic services offered help in generation of revenue for the centre. During the year, the centre generated revenue to the tune of Rs. 23,08,800/- by testing samples for various diseases including equine infectious anaemia (Rs. 12,54,450), contagious equine metritis (Rs. 1,72,000), glanders (Rs. 7,78,700), equine viral arteritis (Rs. 32,000), EHV-1 (Rs. 24,000), equine piroplasmiasis (Rs. 23,800). In addition, the improved germplasm of equines was also provided to the farmers in different parts of the country. Artificial insemination using exotic jack semen was done in field in different states for superior quality mule production in the country.

**Table: Equine bacterial isolates recovered and their origin**

Isolate (Number)	Nature of sample	Place of Origin
<i>Streptococcus equi</i> subsp <i>equi</i> (1)	Lymph node swab (1)	Haryana (1)
<i>Streptococcus equi</i> subsp <i>zooepidemicus</i> (1)	Nasal swab (1)	Haryana (1)
<i>E. coli</i> (2)	Vaginal swab (2)	Rajasthan (2)
Group C Streptococci (3)	Nasal swab (3)	U.P.(2), Haryana (1)
<i>Burkholderia mallei</i> (8)	Pus (1), Nasal swab (7)	Maharashtra (4), U.P. (3), Uttarakhand (1)
<i>Staphylococcus</i> spp. (1)	Pus (1)	Uttarakhand (1)



# AWARDS AND RECOGNITIONS

## A facility for Veterinary Type Cultures established at NRCE

Recognizing the expertise and dedication of this centre in the area of animal health, particularly equine infectious diseases, Indian Council of Agricultural Research (ICAR) sanctioned the establishment of Veterinary Type Cultures (VTC) facility at NRCE during the second half of the 10<sup>th</sup> plan period. According to Dr. S.K. Dwivedi (Director), the VTC facility would serve as a bank for storage, conservation and characterization of veterinary pathogens prevalent in India.

During the year, various research equipments

### Mandate of the VTC

- National repository of microorganisms of animal origin including recombinant cultures and plasmids.
- Identification, characterization and documentation of microorganisms.
- Conservation, maintenance and utilization of microorganisms.
- Surveillance of indigenous/exotic microorganisms.
- Human resource development.

for VTC were procured and functional laboratories for research activities were established by relocation and renovation of the existing space at NRCE. Two scientists have joined VTC during the year.

## Invitation abroad



Dr. Sanjay Kumar, Scientist (Veterinary Medicine) was invited by National Research Centre for Protozoan Diseases, University of Agriculture and Veterinary Medicine, Obihiro, Japan to deliver a talk on “Current status and research activities of protozoan diseases

in India” in the 15<sup>th</sup> Japanese-German Symposium on Protozoan Diseases, from September 16-21, 2006.

## Jawahar Lal Nehru award

Dr. R. V. S. Pawaiya, Scientist (Veterinary Pathology) has been awarded "Jawaharlal Nehru Award for P.G. Agricultural Research 2005" by ICAR for his Ph.D. research on “Pathology of chemically induced neoplasms and evaluation of molecular markers in diagnosis of animal tumours”.



Dr. Pawaiya, during his Ph.D., worked towards application of histochemical studies and molecular markers for differentiation between benign and malignant tumors. He developed markers for early diagnosis of tumor in animals.

## G.B. Singh memorial award



A research paper published in the Animal Reproduction Sciences by Dr. A. Arangasamy (Scientist) entitled “Isolation and characterization of heparin and gelatin binding buffalo seminal plasma proteins and their effect on *in vitro* fertilizing ability (BCMPT and HOST)

with cauda epididymal spermatozoa” has been selected for G.B. Singh Memorial Award. The award was conferred by the Indian Society for the Study of Animal Reproduction in the XXII Annual Convention & National Symposium at Mhow from November, 10-12, 2006.

## EDUCATION AND TRAINING

### NRCE organized a short course on infertility and abortion in equines

The centre organized an ICAR sponsored short course on “Infertility and abortion syndrome in equines” from July 4-13, 2006. Participants in the course included equine veterinary practitioners, scientists and equine breeders from different parts of the country. During the inaugural function on July 4, 2006, Dr. S.K. Dwivedi



**Brig. Singhvi addressing the participants**

(Director) congratulated the Course Co-ordinator, Dr. A.S. Panisup for organizing this need-based training course. He emphasized on the importance of equines in rural economy and role of the veterinarians in equine development programme

### Participation in trainings

- ❑ Dr. Mamta, Scientist (SS) participated in the Summer School on "Sample survey techniques in agricultural research" organized by Indian Agricultural Statistics Research Institute, New Delhi from September 5-25, 2006.
- ❑ Dr. Nitin Virmani (Senior Scientist) participated in the practical training course on "DNA based diagnostics" organized by Department of Animal Biotechnology, College of Veterinary Sciences, CCSHAU, Hisar from November 2-22, 2006.

for agricultural operations. The curriculum of the course included lectures, practicals and hands-on



**Delegates in the valedictory function**

training on ultrasonography for pregnancy diagnosis in addition to rectal examination, diagnosis and management of infectious reproductive problems. Brig. N.M. Singhvi, a renowned equine expert, was the chief guest in the valedictory function on 13<sup>th</sup> July, 2006. Addressing the delegates, Brig. Singhvi underlined necessity of training on infertility in equines that affects up to 40% mares resulting in economic losses to farmers.

- ❑ Dr. Sanjay Barua, Scientist (SS) participated in the practical training course on "DNA based diagnostics" organized by Department of Animal Biotechnology, College of Veterinary Sciences, CCS HAU, Hisar from November 2-22, 2006
- ❑ Dr. R.A. Legha, Scientist (SS) participated in the Winter School on "Sustainable farming systems for arid and semi-arid ecosystems" organized by Central Arid Zone Research



Institute, Regional Research Station, Bikaner from November 4-24, 2006.

- Dr. Rajesh Kumar Vaid, Scientist (SS) participated in the winter school on “Global challenges in vaccinology and control of animal diseases-strategies, approach and blue print” organized by TANUVAS, Chennai from December 1-21, 2006.
- Sh. Mukesh Chand, T-2 (Lab. Tech.) participated in "Laboratory technicians training programme in

electron microscopy" organized by Department of Anatomy, AIIMS, New Delhi from February 12-24, 2007.

- Dr. Niranjana Lal (Scientist) attended "80<sup>th</sup> Foundation course for agricultural research service" organized by National Academy of Agricultural Research Management, Rajendernagar, Hyderabad from January 8-May 7, 2007.

## Lectures, practical trainings and demonstrations for advanced training/refresher courses

1. Dr. B.R. Gulati (Senior Scientist) delivered a lecture on 'Immunological relationship between mare, fetus and foal' in Short Course on Infertility and abortion syndrome in equines, organized by National Research Centre on Equines, Hisar, July 4-13, 2006.
2. Dr. A.K. Gupta (Principal Scientist) delivered a lecture and organized practical on 'Pregnancy diagnosis by equine chorionic gonadotropin (eCG) based sandwich ELISA' in Short Course on Infertility and abortion syndrome in equines, organized by National Research Centre on Equines, Hisar, July 4-13, 2006.
3. Dr. S.K. Khurana (Senior Scientist) delivered a lecture on 'Role of mycoplasmal and mycotic conditions causing infertility in equines' in Short Course on Infertility and abortion syndrome in equines, organized by National Research Centre on Equines, Hisar, July 4-13, 2006.
4. Dr. Rajender Kumar (Senior Scientist) delivered a lecture on 'Parasitic diseases and their role in causation of sub-fertility in equines' in Short Course on Infertility and abortion syndrome in equines, organized by National Research Centre on Equines, Hisar, July 4-13, 2006.
5. Dr. Sanjay Kumar (Senior Scientist) delivered a lecture and organized demonstration on 'Clinical examination of mare and stallion for reproductive soundness' in Short Course on Infertility and abortion syndrome in equines, organized by National Research Centre on Equines, Hisar, July 4-13, 2006.
6. Dr. P Malik (Senior Scientist) delivered a lecture on 'Diagnosis of bacterial agents from cases of infertility among equines' in Short Course on Infertility and abortion syndrome in equines, organized by National Research Centre on Equines, Hisar, July 4-13, 2006.
7. Dr. P Malik (Senior Scientist) delivered a lecture on 'Role of bacteria in causing infertility in equines including diagnosis and control' in Short Course on Infertility and abortion syndrome in equines, organized by National Research Centre on Equines, Hisar, July 4-13, 2006.
8. Dr. Yash Pal (Senior Scientist) delivered a lecture on 'Role of hormones in sustaining pregnancy in mares' in Short Course on Infertility and abortion syndrome in equines, organized by National Research Centre on Equines, Hisar, July 4-13, 2006.
9. Dr. A.S. Panisup (Principal Scientist) delivered a lecture on 'Reproductive disorders in mares leading to abortion and infertility' in Short Course on Infertility and abortion syndrome in equines, organized by National Research Centre on Equines, Hisar, July 4-13, 2006.
10. Dr. B.K. Singh (Principal Scientist) delivered a lecture on 'Viral causes of abortion with emphasis on epizootiology, diagnosis and control of herpes virus infections in mares' in Short Course on Infertility and abortion syndrome in equines, organized by National Research Centre on Equines, Hisar, July 4-13, 2006.

11. Dr. B.R. Singh (Principal Scientist) delivered a lecture on 'Importance of Semen microbiology in the era of artificial insemination in equines' in Short Course on Infertility and abortion syndrome in equines, organized by National Research Centre on Equines, Hisar, July 4-13, 2006.
12. Dr. Nitin Virmani (Senior Scientist) delivered a lecture and organized practical on 'Exfoliative cytology- A tool for diagnosis and prognosis in equines' in Short Course on Infertility and abortion syndrome in equines, organized by National Research Centre on Equines, Hisar, July 4-13, 2006.
13. Dr. B.R. Singh (Principal Scientist) delivered a lecture on 'Use of *Salmonella* in Biotechnology' in College of Biotechnology, SVBP Univ. Agric. And Technology, Modipuram, Meerut (UP), September 26, 2006.
14. Dr. R.C. Sharma (Senior Scientist) delivered a lecture on 'Equine genetic resources and their improvement' in Winter school on Sustainable farming systems for arid and semi arid ecosystems, CAZRI Regional Research Station, Bikaner (Rajasthan), November 4-24, 2006.
15. Dr. B.R. Gulati (Senior Scientist) delivered a lecture and organized practical on 'Laboratory diagnosis of Japanese encephalitis by haemagglutination inhibition assay' in a training course on 'Methods in Virology for Disease Diagnosis', organized by Centre of Advanced Studies, Department of Veterinary Microbiology, CCS Haryana Agricultural University Hisar from December 28, 2006-January 17, 2007.
16. Dr. B.R. Gulati (Senior Scientist) delivered a lecture on 'Screening of equines for viral infections in India' in a training course on 'Methods in virology for disease diagnosis', organized by Centre of Advanced Studies, Department of Veterinary Microbiology, CCS Haryana Agricultural University Hisar. December 28, 2006-January 17, 2007.
17. Dr. B.K. Singh (Principal Scientist) delivered a lecture on 'Viral abortions in animals' in training course on 'Methods in Virology for Disease Diagnosis'. Centre of Advanced Studies, Department of Veterinary Microbiology, CCS Haryana Agricultural University Hisar. December 28, 2006-January 17, 2007.
18. Dr. B.K. Singh (Principal Scientist) delivered a lecture and organized practical on 'Virus neutralization test for equine herpes virus-1 diagnosis' in a training course on 'Methods in Virology for Disease Diagnosis'. Centre of Advanced Studies, Department of Veterinary Microbiology, CCS Haryana Agricultural University Hisar. December 28, 2006-January 17, 2007.
19. Dr. P Malik (Senior Scientist) delivered a lecture on 'Diagnostic techniques in glanders' in National Seminar under ASCAD on 'Emerging and re-emerging animal diseases of public health importance', YASHDA, Pune, January 17-18, 2007.
20. Dr. P Malik (Senior Scientist) delivered a lecture on 'Emerging and existing major equine bacterial diseases in current scenario: Role of State Disease Diagnostic labs' in ASCAD symposium on Strengthening of disease diagnostic facilities for sustainable livestock and poultry development, RDDC, Udaipur, March 10-11, 2007.
21. Dr. Rajender Kumar (Senior Scientist) delivered a lecture on 'Diagnosis of parasitic diseases of equines and their management' in ASCAD symposium on Strengthening of disease diagnostic facilities for sustainable livestock and poultry development, RDDC, Udaipur, March 10-11, 2007.
22. Dr. Nitin Virmani (Senior Scientist) delivered a lecture on 'Collection, preservation and dispatch of biological material for disease diagnosis in equines' in ASCAD symposium on 'Strengthening of disease diagnostic facility for sustainable livestock and poultry development' organized by Regional Disease Diagnostic Centre, Department of Animal Husbandry, Udaipur, March 10-11, 2007.

## Training for post-graduate students

- A short term training course "Production and characterization of monoclonal antibodies" was organized for PhD student of MDU, Rohtak, during June-July, 2006.
- Two MVSc students of Veterinary College, CCS HAU, Hisar are being guided by NRCE scientists as co-advisor.



## RAC, MANAGEMENT COMMITTEE & IRC MEETINGS

### Research Advisory Committee Meeting

The 8<sup>th</sup> RAC meeting was held at our Bikaner campus under the chairmanship of Dr. S.K. Garg, former Vice-Chancellor, *Pashu Chikitsa Vigyan Vishwavidyalaya Evam Go Anusandhan Sansthan*, Deen Dayal Upadhyay Veterinary University, Mathura on April 25, 2006 to discuss various scientific, administrative and policy matters of NRCE. The RAC recommended that additional posts of the technical and supporting staff should be provided to the centre as per the recommended ratio. In view of the achievements made by the



**Members of RAC during their visit to Equine Production Campus, Bikaner**

centre and its services required at national level, the RAC also strongly recommended that the centre may be upgraded to a National Institute on Equines Research, with establishment of more regional stations. The RAC approved three new research project proposals.

### Staff Research Council Meeting

The Annual SRC meeting was held under the chairmanship of Dr. S.K. Dwivedi on May 4-5, 2006 to discuss the progress made in various on-

going research projects. Four new research project proposals were approved by the SRC. The chairman emphasized that training courses for field veterinarians on various techniques in equine practice should be organized regularly at the centre. He also stressed that farmers should be provided training on management and husbandry of horses, mules and donkeys. A training programme on equine farriery should be arranged for technical staff of NRCE and equine farmers.

### New Projects approved

- Development of nutraceutical from medicinal plants for improvement of equine performance.
- Studies on high-level drug resistant bacteria in equines for search of sentinel microbes to use in predictive disease modeling and microbes with vector potential.
- Development of sensitive and specific diagnostics for Japanese encephalitis in Equines.
- To study the existing donkey and mule production system in Haryana, Rajasthan and U.P.

### Institute Management Committee Meeting

Twenty-seventh meeting of the Institute Management Committee was held on May 31, 2006 under the chairmanship of Dr. S.K. Dwivedi, Director. The IMC recommended the purchase of endoscope, remote thermometer, tele-inject system, plasma TV, etc for the centre. In addition, the committee also approved the proposal for purchase of quality imported equipments for Veterinary Type Cultures. The IMC also approved the proposal to write off losses due to animals' mortality at the centre.

### Institute Research Committee Meeting

The half yearly IRC meetings was held under the chairmanship of Dr. S.K. Dwivedi on January 8-9, 2007 at NRCE Hisar to discuss the progress in equine health, production and management. The house reviewed the research work done and made specific recommendations for different ongoing projects. The chairman emphasized that the outcome of the research projects should be in form of development of technologies directly benefiting the end-users and should at the same time have some commercial value.



Scientists deliberating during IRC Meeting

### Members of RAC

Dr. S.K. Garg, Director CAEHS, Meerut	Chairman
Dr. S.K. Dwivedi, Director NRCE, Hisar	Member
Dr. R.P. Mishra, Ex-FAO Expert, Bareilly	Member
Col (Dr.) B. Raut, Consultant Field Research Laboratory, Chandigarh	Member
Dr. M.S. Oberoi, SAARC Regional Coordinator, FAO, New Delhi	Member
Dr. Lal Krishna, ADG (AH), ICAR, New Delhi	Member
Sh. Arvind Yadav, Rewari	Member
Sh. Ram Kripal Bhadoria, Lucknow	Member
Dr. Rajender Kumar, Incharge PME Cell, NRCE, Hisar	Member Secretary

### Members of Institute Management Committee

Dr. S.K. Dwivedi, Director NRCE, Hisar.	Chairman
Dr. Lal Krishna, ADG (AH), ICAR, New Delhi	Member
Director, Animal Husbandry, Govt. of Haryana	Member
Director, Animal Husbandry, Govt. of Punjab	Member
Dean, Veterinary College, CCS HAU, Hisar	Member
Finance & Accounts Officer, NBPGR, New Delhi	Member
Dr. S.N. Tandon, Principal Scientist, NRCE, Bikaner	Member
Dr. A.K. Gupta, Principal Scientist, NRCE, Hisar	Member
Dr. R.K. Sethi, Principal Scientist, CIRB, Hisar	Member
Dr. S. Dey, Senior Scientist, NRCE, Hisar	Member





## WORKSHOPS, SEMINARS & INSTITUTIONAL ACTIVITIES

### Seminar on Marwari horse breed standards organized

National Research Centre on Equines organized a seminar for 'Finalization of Breed Characteristics of Marwari Horse and Procedure for Initiation of Stud Book' on 25<sup>th</sup> November 2006. Various officials from central and state animal husbandry departments, Remount Veterinary Corps (RVC), scientists working in the field of equine breeding & reproduction and representatives of indigenous equine breeders and non-governmental organizations (NGOs) participated in the seminar. The seminar was held under the chairmanship of



**Delegates discussing Marwari breed characteristics in the Seminar**

Dr. S.K. Bandyopadhyay (Commissioner, Animal Husbandry), Department of Animal Husbandry, Dairying & Fisheries (DAHDF), Govt. of India. Sh. Arvind Kaushal, Joint Secretary, DAHDF highlighted the importance of maintenance of stud book for registration of indigenous equines. Addressing the delegates, Dr. S.K. Dwivedi highlighted the role of NRCE in development and conservation of Marwari breed. He invited the comments from the delegates on the 'Marwari Breed Descriptor' developed by NRCE for finalization of Marwari horse characteristics. After in-depth deliberations and inputs from various stake holders, the breed descriptor for Marwari horses was

unanimously adopted. It was also emphasized that horses produced by artificial insemination with pedigreed Marwari stallion should be accepted for entry in the stud book, to be maintained by a registration committee to be constituted by the Animal Husbandry Commissioner in consultation with indigenous horse societies.

### NRCE organizes an indigenous horse show and farmers' meet on its Foundation Day

National Research Centre on Equines, Hisar celebrated its Foundation Day on 26<sup>th</sup> November 2006 with great fanfare. Dr. J.C. Katyal, Vice-Chancellor, CCS Haryana Agricultural University, Hisar was the Chief Guest on this occasion. A horse show was organized on this occasion, in which horse lovers and farmers from various parts of Haryana and Rajasthan exhibited their horses of different indigenous breeds. Different equestrian events like tent pegging, horse jumping, riding skills, etc were also organized on this day. Addressing the equine owners, Dr. S.K. Dwivedi, Director NRCE highlighted the salient achievements in equine research and development in the area of health and production by the centre.



**Horses in action at Foundation Day**

On this occasion, a Kisan Goshti (*Ashwapalak Meet*) was organized in which equine owners from Haryana and neighbouring states deliberated on the problems faced by them in equine production and

health management and were provided sustainable solutions.

### Health camp for donkeys organized at Ratangarh (Rajasthan)

A two-day health camp was organized at Ratangarh Rajasthan during June 2-3, 2006 to provide healthcare to donkey population of this region. A total of 72 donkeys were given treatment for different ailments like lameness, keratitits, debility and weakness. All the equids were dewormed and vaccinated against tetanus. Samples of blood, feed, forage and water were collected for laboratory analysis.

During the camp, farmers were educated on



**NRCE team treating donkeys in the health camp**

equine foot-care, housing and nutritional management of donkeys. They were given training on good donkey management practices to get maximum work with minimum stress to the animals. The farmers were provided literature and know-how for ideal management and formulation of eco-friendly balanced ration for equine from the locally available resources.

### Mule health awareness camp at Gobindghat (Uttarakhand)

A health-cum-awareness camp was conducted from June 7-8, 2006 at Gobindghat (Uttarakhand) for providing healthcare to mules used for transport of pilgrims from Gobind Ghat to Hemkund Sahib and for educating mule owners

about management practices for better health and nutrition of mules. More than 150 animals were



**NRCE expert team at Gobindghat (Uttarakhand)**

examined and 110 samples were collected for laboratory analysis.

### Independence day celebrated

The Independence Day was celebrated at the centre on 15<sup>th</sup> August 2006 with the hoisting of the National Flag by the Director, Dr. S.K. Dwivedi. Addressing employees of the centre on this occasion, he complimented the efforts of scientists and employees of the centre towards betterment of farmers and equine breeders. Dr. Dwivedi



**Director NRCE addressing employees on the Independence Day**

emphasized the need to follow the footsteps of those who sacrificed their lives for the nation and advised to dedicate ourselves whole-heartedly in the service of our motherland. On this occasion, employees and their family members presented a cultural programme.



## हिन्दी सप्ताह का आयोजन

राष्ट्रीय अश्व अनुसंधान केन्द्र में हिन्दी चेतना मास के अर्न्तगत दिनांक 21 से 27 सितम्बर 2006 तक हिन्दी सप्ताह का आयोजन किया गया। हिन्दी सप्ताह समारोह का शुभारंभ आदरणीय राज्य कवि श्री उदय भानु हंस जी मुख्य अतिथि ने 21 सितम्बर 2006 को किया। श्री हंस जी ने कर्मचारियों एवं अधिकारियों को हिन्दी में अधिकाधिक कार्य करने के लिये प्रेरित किया और कहा कि हिन्दी को अपना ही सच्ची राष्ट्रीयता है। इस अवसर पर प्रोफेसर राम कुमार भारद्वाज ने अपने प्रेरणादायी भाषण में कहा कि हमें अपने राष्ट्र के



हिन्दी सप्ताह समारोह में प्रतिभागी भाषण प्रतियोगिता में भाग लेते हुए

चहुँमुखी विकास के लिये मानवीय मूल्यों को संरक्षित करना चाहिये और ये मूल्य हमारे जीवन में झलकने चाहिये। हिन्दी सप्ताह के दौरान केन्द्र सरकार के विभागों के कर्मचारियों एवं विभिन्न विद्यालयों के विद्यार्थियों की भाषण एवं कविता प्रतियोगिता आयोजित की गयी। समापन समारोह के अवसर पर केन्द्र के निदेशक, डा० द्विवेदी जी ने प्रतिभागियों को पुरस्कार वितरित किये और कहा कि वैज्ञानिक उपलब्धियों का सही मायने में फायदा तभी होगा जब हम इन्हें हिन्दी में

भी प्रसारित करें।

## **Kavi Sammelan organized at NRCE**

A *Kavi-Sammelan* was organized on 25<sup>th</sup> November, 2006, on the eve of the foundation day of the centre. In the *Kavi Sammelan*, Sh. Uday Bhanu Hans Ji, first state poet of Haryana was the chief guest and many poets of national repute participated in this *sammelan* and recited their poetry. On this occasion, the centre also dedicated



**Sh. Uday Bhanu Hans and other poets in the *Kavi Sammelan***

its newly constructed laboratory-cum administrative building to the nation.

## **Republic day celebrated at the centre**

The 57<sup>th</sup> Republic Day at the centre was celebrated on 26<sup>th</sup> January 2007 with the hoisting of the National Flag by Dr. S.K. Dwivedi, Director. Addressing employees of the centre on this occasion, Dr. Dwivedi emphasized on the need to follow the footsteps of those who sacrificed their lives for the nation and advised to dedicate wholeheartedly for the development of this country. On this occasion, employees and their family members presented a cultural programme.

## DISTINGUISHED VISITORS

### DG ICAR dedicates new laboratory wing at Bikaner to the nation

Dr. Mangala Rai Ji, Director General, ICAR New Delhi visited Equine Production Campus, Bikaner on 26<sup>th</sup> February 2007. During his visit, Dr. Rai inaugurated the new laboratory-cum-administrative building at the campus. The Director General appreciated the research and development activities at the campus, particularly the quality of the equine germplasm being maintained at the campus.



**Dr. Mangala Rai inaugurating the new building at Bikaner**

### Use molecular tools for equine breed characterization: Bandyopadhyay

Dr. S.K. Bandyopadhyay (Commissioner, Animal Husbandry), Department of Animal Husbandry, Dairying & Fisheries (GOI) visited centre on November 25, 2006 to chair the seminar on Marwari breed characterization. Addressing the participating delegates, Dr. Bandyopadhyay emphasized the importance of development of breed standards and need of characterization of different indigenous equine breeds using molecular tools for their genotyping. Dr. Narain Singh Manaklao, Hon'ble Member of Rajya Sabha congratulated Director NRCE for organizing this need-based seminar. On this occasion, Dr. Arvind Kaushal (Joint Secretary, DAHDF), Col. R.R.



**Dr. Bandyopadhyay and Dr. Manaklao during inauguration of the seminar**

Yadav (Commandant EBS, Hisar) and representatives of various indigenous equines societies expressed their views on breed standards.

### Impact analysis of research benefits to the end users must : Bhargava

Sh. Sudhir Bhargava, Member, ICAR Governing Body visited this centre on October 20, 2006. During his visit, Sh. Bhargava was apprised of different ongoing research and development



**Sh. Bhargava discussing research priorities of the centre during his visit at NRCE**

activities of the centre. On this occasion, Dr. S.K. Dwivedi, Director highlighted the salient achievements of the centre in the area of equine health and production and also discussed about the



future research priorities of the centre. Addressing the scientists, Sh. Bhargava emphasized the need for application of the technologies generated at the centre for the benefits to the farmers and equine owners. He also encouraged the scientists to work on the cost-benefit analysis of the research programmes being pursued at the centre.

### Dr. Katyal inaugurates horse show at NRCE

Dr. J.C. Katyal, Vice-Chancellor, CCS Haryana Agricultural University, Hisar was the chief guest on the occasion on the occasion of Foundation Day of the centre on 26<sup>th</sup> November 2006. During his visit, he inaugurated an indigenous horse show. Addressing the equine owners and scientists of the centre, Dr. Katyal



**Dr. Katyal in indigenous horse show at NRCE**

highlighted the need for the use of equines in agriculture and encouraged farmers for adoption of scientific technologies in the area of equine production for improving the equine germplasm.

### Other visitors to the centre

- Prof. K. Muralidhar of Department of Zoology, University of Delhi visited this centre on September 13, 2006. During his visit, Prof. Muralidhar took keen interest in the research activities of the centre, especially in the field of equine hormones. He sought the collaboration of the centre for characterization and purification of equine chorionic gonadotrophin. On this occasion, Dr. S.K. Dwivedi, Director

highlighted the need for undertaking the inter-institutional projects for commercial utilization



**Dr. Muralidhar being apprised of the research activities at the centre**

of various equine bye-products.

- Sh. Tapesh Pawar, Secretary, Department of Animal Husbandry, Dairying & Fisheries, Govt. of Rajasthan visited Equine Production Campus, Bikaner on October 26, 2006. He appreciated the ongoing research activities in the campus.
- Dr. Khub Singh (Former Director, National Institute of Animal Nutrition & Physiology, Bangalore) and Dr. S.K. Ray (Former Director, Department of Animal Husbandry & Veterinary Sciences, Orissa)



**Dr. Khub Singh during his visit to NRCE**

visited centre on January 16, 2007. They were apprised about the research activities in the area of equine health and production being undertaken at the centre.

# INFRASTRUCTURE DEVELOPMENT

## New administrative-cum-laboratory at EPC, Bikaner dedicated to the nation

The work on new state-of-the-art laboratory-cum-office building was completed at equine production campus, Bikaner during the year. The building was dedicated to the nation by Dr. Mangala Rai Ji, Director General, ICAR on 26<sup>th</sup> February 2007. Inaugurating the building, the Director General appreciated the upkeep of our valuable equine germ plasm, campus plan and research & development programmes being undertaken at the sub-campus, Bikaner. The



**New laboratory building at EPC, Bikaner**

laboratories in the newly constructed laboratory-cum-administrative building have been well furnished, equipped and made fully functional.

In addition, the construction work of open paddocks, stables, foaling lines, internal road, boundary wall and uplifting of main gate of the sub-campus have also been completed.

## New laboratory building constructed at NRCE, Hisar

During the year, the work on construction of additional laboratories at Hisar campus was

completed. The newly-built extension wing has spacious, modern and well furnished laboratories for bacteriology, vaccinology, medicine, parasitology and Veterinary Type Cultures. In addition, the extension wing has well equipped conference room with a sitting capacity of about 120 delegates. The building was dedicated to the nation on November 25, 2006 by Dr. S.K. Dwivedi, Director.

## Agriculture farm production

During the period 2006-07, fodder crops of *Rabi* and *Kharif* season were cultivated in the agricultural farm at Hisar and Bikaner to supply green fodder to animals round the year. The fodder production at Hisar and Bikaner campus is shown in Table 1. The centre produced 2436.6 quintal of fodder during the year, 1397 quintal at Hisar farm and 1039.6 quintals at Bikaner farm.

In addition, 185 dry, dead and diseased trees in the farm area at Hisar were sold to the Haryana Forest Department Corporation at a total cost of Rs.1,01,262/- (Rupees one lakh one thousand two hundred sixty two only).

**Table 1. Fodder production at NRCE, Hisar and Bikaner**

Type of fodder	Production in Quintals		Total
	Hisar	Bikaner	
Lucerne	340.0	484.0	824.0
Oat	203.5	296.2	499.7
Sorghum	419.5	259.4	678.9
Cowpea	128.5	-	128.5
Berseem	184.5	-	184.5
Maize	121.0	-	121.0
<b>Total</b>	<b>1397.0</b>	<b>1039.6</b>	<b>2436.6</b>



## Livestock

The centre has maintained a representative herd of equines at Bikaner campus comprising indigenous horses of Marwari and Kathiawari breed (49), exotic donkeys (40) and mules (5) (Table 2).

The centre is also maintaining some animals

at Hisar campus comprising horses of Marwari breed (11), ponies (12), exotic donkeys (5) and mules (5) of different age groups (Table 3). The centre is also maintaining a frozen stock of the semen which is being used for artificial insemination purpose.

**Table 2. Equine herd strength at Bikaner campus**

Category	Horses		Exotic donkeys		Mules		Total
	Male	Female	Male	Female	Male	Female	
Stock as on 1.4.2006	10	30	18	23	1	1	83
Births during the year	3	3	2	5	2	1	16
Deaths during the year	-	2	-	4	-	-	6
Sold during the year	-	-	2	2	-	-	4
<b>Balance as on 31.3.2007</b>	<b>13</b>	<b>31</b>	<b>18</b>	<b>22</b>	<b>3</b>	<b>2</b>	<b>89</b>

**Table 3. Equine herd strength at Hisar campus**

Category	Horses		Ponies	Mules	Donkeys	G. Total
	Marwari	Others				
Adult Male	1	1	-	2	3	7
Adult female	9	1	11	-	-	21
2-3 yrs	-	4	1	1	-	6
1-2 yrs	-	-	-	-	-	-
0-1 yrs	1	-	-	2	-	3
<b>Balance as on 31.3.2007</b>	<b>11</b>	<b>6</b>	<b>12</b>	<b>5</b>	<b>3</b>	<b>37</b>

# LIST OF PUBLICATIONS

## Research Articles (authored by NRCE scientists)

1. Bansal R. S., Pal Y., Pareek P. K., Purohit G. N., Legha R. A. and Gupta A. K. (2006). Progesterone profile during oestrous cycle, pregnancy and post foaling in donkey mares. *Veterinary Practitioner*, **7**:5-6.
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4. Gulati B. R., Pandey R. and Singh B. K. (2006). Development of monoclonal antibodies against group-A animal rotaviruses. *Indian Journal of Biotechnology*, **5**: 37-41.
5. Gulati B. R., Singh B. K. and Kumar D. (2006). Development of a monoclonal antibody-based sandwich ELISA for detection of equine rotavirus from diarrhoeic foals. *Indian Journal of Microbiology*, **46**: 349-354.
6. Gupta A. K. and Virmani N. (2007). Equine influenza in India. CAB Reviews: Perspectives in Agriculture, Veterinary Science, Nutrition and Natural Resources, (PAVSNNR) 2, No. 016, 9 pp.
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11. Raina O. K., Yadav S. C., Sriveny D., Gupta S. C. (2006). Immuno diagnosis of bubaline fasciolosis with *Fasciola gigantica* cathepsin-L and recombinant L1-D proteases. *Acta Tropica*, **98**: 145-157.
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10. Yadav S. C., Sarvanan B. C. Borkataki S., Baruah K. (2006). A new record of *Parafilaria bovicola* from yak (*Poephagus grunniens*) in India. *Journal of Veterinary Parasitology*: In Press.

### Accepted Research Papers

1. Dey S., Dwivedi S. K., Malik P., Panisup A. S., Tandon S. N. and Singh B. K. (2006). Heat stress associated with mortality in donkey in India. *Veterinary Record*: Accepted.
2. Lal N., Arya H. P. S. and Gupta R. (2007). Farmers' participation on FMD vaccination technology in cattle. *Indian Journal of Extension Sciences*: Accepted.
3. Pal Y., Gupta A. K. and Kumar S. (2006). Draughtability studies in donkey as pack animal. *Annals Arid Zone*: Accepted.
4. Sarvanan B. C, Yadav S. C., Borkataki S. and Pourochottamane R. (2006). Lice infection in Yak. *Indian Veterinary Journal*: In Press.

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2. Gulati B. R. and Singh B. K. (2006). An overview of infectious respiratory diseases of equines. *Centaur*, **22**:32-43.
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### Abstracts in Seminar and Symposia

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2. Chauhan M, Gupta A. K. and Dhillon S. (2007). Evaluation of putative bottleneck in Kathiawari and Spiti horses. *In: 4<sup>th</sup> National Symposium on Role of Animal Genetic resources in rural livelihood security, Ranchi, February 8-9.*



intensively reared goat farm. *In: International Symposium and Annual Conference on new strategies for prevention and control of zoonotic diseases, an integrated Veterinary and Medical approach*, pp 136, IAVPHS, Palampur, October 12-14.

11. Virmani N., Singh B. K., Gulati B. R. and Panisup A. S. (2006). Type specific glutathione S-transferase fusion protein for diagnosis and differentiation of EHV-1 and EHV-4. *In: XXIII Annual Conference of Indian Association of Veterinary Pathologists (IAVP), Department of Veterinary Pathology, Tamilnadu Veterinary and Animal Science University, Chennai, December 27-29.*
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### **Publications as Invited Lectures:**

1. Gulati B. R. (2007). Laboratory diagnosis of Japanese encephalitis by haemagglutination inhibition assay. *In: Laboratory manual on 'Methods in Virology for Disease Diagnosis'. Centre of Advanced Studies, Department of Veterinary Microbiology, CCS Haryana Agricultural University Hisar. December 28, 2006-January 17, 2007.*
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6. Kumar R. and Kumar S. (2007). An update on Equine Babesiosis: Lead paper. *In: National Seminar on Recent Diagnostic trends and control strategies for haemoprotozoan infections in livestock, Department of Parasitology, C.V.Sc. & A.H., S. K. Nagar Dantiwara Agric. Univ., S K Nagar (Gujarat), Feb 9-11.*
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9. Kumar S. (2006). Clinical examination of mare and stallion for reproductive soundness. *In: Compendium of Short Course on Infertility and abortion syndrome in equines.* Gulati B R, Virmani N, Barua S & Vaid R K (eds), National Research Centre on Equines, Hisar.

10. Malik P. (2006). Diagnosis of bacterial agents from cases of infertility among equines. *In: Compendium of Short Course on Infertility and abortion syndrome in equines.* Gulati B R, Virmani N, Barua S & Vaid R K (eds), National Research Centre on Equines, Hisar, pp 48-50.
11. Malik P. (2007). Diagnostic techniques in glanders. *In: National Seminar under ASCAD on "Emerging and re-emerging animal diseases of public health importance",* YASHDA, Pune, January 17-18, pp 37-46.
12. Malik P. (2007). Emerging and existing major equine bacterial diseases in current scenario: Role of State Disease Diagnostic labs. *In: ASCAD symposium on Strengthening of disease diagnostic facilities for sustainable livestock and poultry development,* RDDC, Udaipur, March 10-11, pp 30-37.
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## PARTICIPATION IN CONFERENCES & SYMPOSIA

1. Dr. B.R. Singh (Principal Scientist) participated and presented a lead paper in the 6<sup>th</sup> Indian Veterinary Congress held at OUAT, Bhubaneswar from April 6-7, 2006.
2. Dr. Yash Pal (Senior Scientist) participated in National Seminar on “Artificial insemination: acceptability, impact, constraints and solutions” held at Indian Veterinary Research Institute, Izatnagar from October 12-13, 2006.
3. Dr. S.K. Dwivedi (Director) was invited by FICCI to attend the Agricultural Summit-2006, at Vigyan Bhavan, New Delhi from October 18-19, 2006.
4. Dr. S.K. Dwivedi (Director) chaired a session in the International Conference on Equine Infertility, held at Remount Veterinary Corps Centre, Meerut on November 2, 2006.
5. Dr. R.S. Bansal (T-9) participated in Intermediate Meeting of the World Equine Veterinary Association held at RVC Centre School, Meerut from November 2-3, 2006.
6. Dr. B.R. Gulati (Sr. Scientist) participated in "7th Asia-Pacific Conference on Medical Virology" organized by All India Institute of Medical Sciences, New Delhi from November 13-15, 2006.
7. Dr. S.C. Yadav (Sr. Scientist) participated in XVII National Congress of Veterinary Parasitology and National Symposium held at Puducherry from November 15-17, 2006.
8. Dr. Rajender Kumar (Sr. Scientist) participated and presented a paper in XVII National Congress of Veterinary Parasitology and National Symposium held at Puducherry from November 15-17, 2006.
9. Dr. B.R. Gulati (Sr. Scientist) participated in one-day Workshop on Information and Communication Technology for Agriculture, organized by National Informatics Centre, Rohtak held on December 15, 2006.
10. Dr. Nitin Virmani (Sr. Scientist) participated and presented a paper in the XXIII Annual Conference of Indian Association of Veterinary Pathologists held at Department of Veterinary Pathology, Madras Veterinary College, Tamil Nadu Veterinary and Animal Sciences University, Chennai from December 27-29, 2006.
11. Dr. Niranjan Lal (Scientist) participated in National Seminar on “Information and communication technologies: opportunities and challenges for revitalizing extension system” organized by Indian Society of Extension Education in collaboration with Navsari Agricultural University at Navsari (Gujarat) from December 27-29, 2006.
12. Dr. S.K. Dwivedi (Director) delivered a keynote address on 'Canine Genomics' in Asian Congress on Canine Practice and Symposium on New Dimensions and Challenges to sustain development and competitiveness in canine practice in the new millennium, organized by Indian Society for Advancement of Canine Practice, Veterinary College Nagpur during February 7-9, 2007.
13. Dr. S.K. Khurana (Senior Scientist) participated and presented a paper in VII Indian Veterinary Congress and XIV Annual Conference of Indian Association for the Advancement of Veterinary Research and National Symposium on "Progress and demands in livestock and poultry health and production and new areas like animal welfare and disaster management" held at Mhow during February 8-9, 2007.
14. Dr. B.R. Singh (Principal Scientist) participated and presented a lecture in one day symposium on “Enteric fever and other Enteric infections” at Safdarjang Hospital, New Delhi on March 24, 2007.

# PERSONNEL MILESTONES

## Joining



Dr. Sanjay Barua joined National Centre for Veterinary Type Cultures of this centre as Scientist (Veterinary Microbiology) w.e.f. June 1, 2006 consequent upon his transfer from Central Institute for Research on Goats, Makhdoom (Mathura, Uttar

Pradesh). Dr. Barua joined ICAR services in March 1997 and acquired his Ph.D. in Veterinary Virology from Indian Veterinary Research Institute, Izatnagar (U.P.) in 2002. Before joining NRCE, Dr. Barua was In-charge of Virology and Public Health Laboratory of CIRG, Makhdoom.



Dr. R. K. Vaid joined National Centre for Veterinary Type Cultures at NRCE as Scientist (Veterinary Public Health) w.e.f. June 1, 2006, on his transfer from Central Institute for Research on Goats, Makhdoom, Farah, Mathura (Uttar Pradesh). He

joined ICAR services in December 1997 and completed his Ph.D. in Veterinary Public Health from Govind Ballabh Pant University of Agriculture and Technology, Pantnagar in 2004.

## Selection

- Dr. S. Dey (Senior Scientist) was selected as Principal Scientist (Veterinary Medicine) at Division of Medicine, IVRI, Izatnagar, Bareilly, UP. He was relieved from this centre on August 9, 2006 to take up the new assignment.
- Dr. R.V.S. Pawaiya (Scientist) was relieved from the centre on January 23, 2007 subsequent to his selection as Senior Scientist (Veterinary Pathology) at Division of Pathology, IVRI, Izatnagar, Bareilly, UP.

## Promotions

- Dr. R.V.S. Pawaiya, Scientist (SS) promoted as Senior Scientist w.e.f. July 5, 2005.
- Dr. Nitin Virmani, Scientist (SS) promoted as Senior Scientist w.e.f. October 14, 2005.
- Dr. Sanjay Kumar, Scientist (SS) promoted as Senior Scientist w.e.f. October 31, 2006.

## Dr. Panisup given farewell on his superannuation

Dr. Attar Singh Panisup, Principal Scientist (Veterinary Pathology) retired from active service on July 31, 2006. While working at NRCE, he contributed immensely in the development of



**The Director and scientists presenting memento to Dr. Panisup on his superannuation**

diagnostic techniques for several equine diseases, including COFEB kit for *Babesia equi* diagnosis, for which a patent has been filed. He published more than 100 research papers in journals of national and international repute. Dr. Panisup visited several countries, including Sweden, Canada and USA. He worked as the In-charge of equine health unit at the centre for several years. Dr. Panisup organized several equine health camps and imparted training to veterinarians and equine owners in different areas of equine health. He was the course director of a short course on 'Infertility and abortion syndrome in equines' for veterinarians organized at NRCE in July 2006. We wish Dr. Panisup a happy, healthy and peaceful life after his superannuation.



# Staff at NRCE

## Director

**Dr. S.K. Dwivedi, M.V.Sc., Ph.D.**

### Scientific Staff at NRCE

1.	Dr. A.K. Gupta, M.Sc., Ph.D.	Principal Scientist
2.	Dr. S.N. Tandon, M.V.Sc., Ph.D.	Principal Scientist
3.	Dr. B.K. Singh, M.V.Sc., Ph.D.	Principal Scientist
4.	Dr. B.R. Singh, M.V.Sc., Ph.D.	Principal Scientist
5.	Dr. S.C. Yadav, M.Sc., Ph.D.	Senior Scientist
6.	Dr. S.K. Khurana, M.V.Sc., Ph.D.	Senior Scientist
7.	Dr. Yash Pal, M.Sc., Ph.D.	Senior Scientist
8.	Dr. R.C. Sharma, M.V.Sc., Ph.D.	Senior Scientist
9.	Dr. B.R. Gulati, M.V.Sc., Ph.D.	Senior Scientist
10.	Dr. Rajender Kumar, M.V.Sc., Ph.D.	Senior Scientist
11.	Dr. Praveen Malik, M.V.Sc., Ph.D.	Senior Scientist
12.	Dr. R.A. Legha, M.Sc., Ph.D.	Scientist (SS)
13.	Dr. Sanjay Kumar, M.V.Sc., Ph.D.	Scientist (SS)
14.	Dr. Nitin Virmani, M.V.Sc., Ph.D.	Scientist (SS)
15.	Dr. (Ms.) Mamta, M.Sc., Ph.D.	Scientist (SS)
16.	Dr. A. Arangasamy, M.V.Sc., Ph.D.	Scientist
17.	Dr. Niranjana Lal, M.Sc., Ph.D.	Scientist

### Technical Staff

1.	Dr. R.S. Bansal, T-9	Farm Manager
2.	Sh. R.K. Chaturvedi, T-6	Technical Officer
3.	Sh. K.K. Singh, T-5	Technical Officer
4.	Sh. K.S. Meena, T-5	Farm Manager
5.	Dr. Jitender Singh, T-5	Veterinary Officer
6.	Sh. P.P. Chaudhary, T-4	Lab. Technician
7.	Sh. Ajmer Singh, T-4	Stock Assistant
8.	Sh. Brij Lal, T-4	Stock Assistant
9.	Sh. D.D. Pandey, T-4	Lab. Assistant
10.	Sh. Sita Ram, T-4	Lab. Assistant
11.	Sh. S.K. Chhabra, T-4	Lab. Assistant
12.	Sh. N.K. Chauhan, T-3	Farm Technician
13.	Sh. Joginder Singh, T-3	Lab. Assistant
14.	Sh. Mukesh Chand, T-2	Lab. Assistant
15.	Sh. Sajjan Kumar, T-2	Driver
16.	Sh. Arun Chand, T-2	Driver
17.	Sh. Suresh Kumar, T-2	Driver
18.	Sh. Shankar Lal, T-2	Driver
19.	Sh. S.N. Paswan, T-2	Livestock Assistant
20.	Sh. Om Prakash, T-2	Driver
21.	Sh. Rajendra Singh, T-1	Lab. Technician
22.	Sh. Raghbir Singh, T-1	Driver

### Scientific Staff at VTC

1.	Dr. Sanjay Barua, M.V.Sc., Ph.D.	Scientist (SS)
2.	Dr. Rajesh Vaid, M.V.Sc., Ph.D.	Scientist (SS)

### Administrative Staff

1.	Sh. K.K. Chandna	AAO
2.	Sh. R.A. Parashar	AFAO
3.	Sh. Hawa Singh	Assistant
4.	Sh. Ram Pal	Assistant
5.	Sh. S.P. Kaushik	Assistant
6.	Sh. Ashok Arora	Steno
7.	Sh. Subhash Chander	UDC
8.	Sh. Pratap Singh	LDC
9.	Sh. D.D. Sharma	LDC
10.	Sh. Om Prakash	LDC
11.	Sh. Mahender Singh	LDC

### Supporting Staff

1.	Sh. Ishwar Singh	SSGr. III
2.	Sh. Guru Dutt	SSGr. III
3.	Sh. Jai Singh	SSGr. III
4.	Sh. Mahabir Prasad	SSGr. II
5.	Sh. Ramesh Chander	SSGr. II
6.	Sh. Mardan	SSGr. II
7.	Sh. Balwan Singh	SSGr. II
8.	Sh. Desh Raj	SSGr. II
9.	Sh. Ishwar Chander	SSGr. II
10.	Sh. Om Prakash	SSGr. II
11.	Sh. Deepak Kumar	SSGr. II
12.	Sh. Gopal Nath	SSGr. II
13.	Sh. Satbir Singh	SSGr. I
14.	Sh. Hanuman Singh	SSGr. I
15.	Sh. Subhash Chander	SSGr. I
16.	Sh. Ishwar Singh	SSGr. I
17.	Sh. Ram Singh	SSGr. I
18.	Sh. Raju Ram	SSGr. I
19.	Sh. Mahabir Prasad	SSGr. I
20.	Smt. Ram Kali	SSGr. I
21.	Smt. Santra	SSGr. I

# ONGOING RESEARCH PROJECTS

Research Project	Investigators	Period	
		From	To
<b>EQUINE HEALTH</b>			
Development of vaccine(s) against equine herpes virus-1 infection	B.K. Singh, B.R. Gulati and N. Virmani	June 2005	May 2007
Studies on the improvement of the Diagnostics for differentiation between EHV-1 & 4 infections employing molecular techniques.	N. Virmani, A.S. Panisup, B. K. Singh & B.R. Gulati	May 2004	March 2007
Development of sensitive & specific methods for diagnosis of equine rotavirus from diarrhoeic foals	B.R. Gulati and B.K. Singh	June 2003	May 2007
Development of diagnostic(s) for pathogenic <i>Streptococcus equi</i> in equines	P. Malik, S. K. Khurana, Mamta & B.R. Singh	June 2003	March 2007
Development of diagnostics for <i>Rhodococcus equi</i> infection in foals	S.K. Khurana, B.R. Singh, P. Malik & N. Virmani	May 2004	March 2007
Development of sensitive and specific diagnostics for Japanese encephalitis in Equines	B.R. Gulati, B. K. Singh & N. Virmani	Oct 2006	Sept 2009
Usefulness of recombinant protein for serodiagnosis of glanders	P. Malik	Oct 2006	Sept 2008
Studies on high-level drug resistant bacteria in equines for search of sentinel microbes to use in predictive disease modeling and microbes with vector potential	B.R. Singh, B.R. Gulati, S.K. Khurana, Mamta & N. Lal	Oct 2006	Sept 2009
Epidemiological studies on emerging and existing diseases of equines	S.K. Dwivedi, A.S. Panisup, B.K. Singh, S.C. Yadav, S. Dey, S.K. Khurana, B.R. Gulati, R. Kumar, P. Malik, N. Virmani, Sanjay Kumar, R.V.S. Pawaiya, S. Barua, R.K. Vaid & A. Arangasamy	Continuous Service Project	-
Development of diagnostic tests for equine trypanosomiasis (Surra)	R. Kumar, Sanjay Kumar, S. Dey, S.C. Yadav & S.K. Dwivedi	June 2003	March 2007
Development of sensitive and specific diagnostic tests for detection of equine piroplasmiasis	Sanjay Kumar, R. Kumar, S. Dey, A.K. Gupta, S.C. Yadav & S.K. Dwivedi	May 2004	March 2007
<b>EQUINE PRODUCTION</b>			
Cryopreservation of stallion semen and perfection of AI in Marwari horses.	Yash Pal, R.A. Legha, A. Arangasamy & S.N. Tandon	May 2002	June 2007
Development of equine chorionic gonadotropin (ECG) based ELISA based test for pregnancy diagnosis in equines.	A.K. Gupta, Yash Pal, Sanjay Kumar	May 2002	June 2006
RFLP-based genotyping of major histocompatibility complex class II genes in Marwari horses	R.C. Sharma, S.C. Mehta (NRCC) and R.S. Bansal	Oct 2004	March 2008
Molecular markers based parentage testing in horses of Indian origin	Mamta & A.K. Gupta	Dec 2004	June 2007
Isolation of stallion seminal plasma proteins and their effect on <i>in vitro</i> fertilizing ability of spermatozoa	A. Arangasamy & S.K. Bhure (NRCC)	Oct 2004	Sept 2007
Superior mule production in the field through frozen semen of exotic Jacks	R.A. Legha, R.C. Sharma, Yash Pal, S.N. Tandon, A. Arangasamy & R.S. Bansal	Dec 2004	Nov 2007
Cryo-preservation of embryos for conservation of Marwari Horses	R.K. Chaturvedi and A. Arangasamy	Jan 2007	Dec 2009
Molecular Characterization of indigenous breeds of horse for genetic diversity within and between different breeds.	A.K. Gupta, S.C. Gupta (NBAGR), S.N. Tandon, Mamta, N. Gupta (NBAGR)	Oct 2006	Sept 2009





# Equines in Action



## Contacts

**HISAR CAMPUS :**  
**National Research Centre on Equines**  
Sirsa Road, Hisar - 125 001, Haryana, India  
Phone: 01662-276151, 276748, 275114  
Fax : 01662-276217, Email : nrcequine@nic.in.

**BIKANER CAMPUS :**  
**Equine Production Campus**  
**National Research Centre on Equines**  
Shiv Bari, Jorbeer, Bikaner-334 001 Rajasthan, India  
Phone: 0151-2232541 Fax : 0151-2230114



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