Diergaarde BLIJDORP rotterdam zoo

HOSTED BY
THE ROTTERDAM ZOO
INTERNATIONAL ELEPHANT FOUNDATION

IEF INTERNATIONAL ELEPHANT FOUNDATION

10 October 2011 – 14 October 2011

INTRODUCTION

Rotterdam Zoo, or **Diergaarde Blijdorp** as it is known, is 152 years old and one of the oldest zoos in the Netherlands. The original zoo was established close to the central station but, as the city expanded, this commercially important site was needed for other purposes and it was decided to move the zoo to its current location. The construction of new zoo was nearly complete by the outbreak of the Second World War, this was fortunate as the old zoo was destroyed in the bombing of the city. Although many animals were killed in the destruction, a number managed to escape; sealions were found swimming in the canals and zebras galloping down the streets. These escapees were soon captured and transferred to the new zoo.

Today Diergaarde Blijdorp is both a zoological and a botanical garden. It is unique in that is was designed by a single architect and, as a result, is now a national monument. The zoo has seen many changes over the last 20 years; it has expanded and today houses its broad collection in modern, naturalistic exhibits. This change in presentation is a reflection of a change in the primary goal of the zoo from education to conservation in the broadest meaning of the term. To this end Diergaarde Blijdorp has recently established a dedicated conservation and research department with a mandate to streamline and develop the zoo's existing conservation and research activities into a coherent and more effective whole.

The zoo has a long history of keeping and breeding elephants. Fifteen Asian elephants were born in the past 25 years, during which period the zoo has managed the European studbook for this species. One black rhino was born at the Rotterdam Zoo, the fifth ever born in captivity, until this species had to make place for the Indian rhinoceros, of which five calves have been born to date.

The **International Elephant Foundation (IEF)** supports conservation, education and research of the world's elephants with a commitment to affect positive change through the facilitation of elephant conservation. Every year IEF facilitates an annual International Elephant Conservation and Research Symposium, a key component of IEF's mission to provide support for in situ and ex situ conservation of elephants.

This symposium, now in its eleventh year, is a multi-disciplinary meeting which offers elephant experts the unique opportunity to learn from others and share information in order to further conservation efforts of elephants in the wild and human care. The symposium deals with issues from veterinary care to management to field conservation and human elephant conflict. During the course of the meeting, the international participants are encouraged to exchange ideas, network and form partnerships that ultimately benefit the elephants with which they work.

As a non-profit organization dedicated to elephant welfare, IEF solicits donations to fund worthy elephant conservation and research projects worldwide. To learn more about the International Elephant Foundation or to contribute to elephant conservation efforts, please visit IEF's website at *www.elephantconservation.org*.

AGENDA

Monday, 10 October 2011

15:00 – 18:00 **Registration**

18:00 – 21:00 Ice-breaker

Tuesday, 11 October 2011

08:00 – 09:00 **Registration**

09:00 – 10:30 Opening Ceremonies

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09:30 – 10:30 *Keynote Speaker* – Cathy Dean, Save the Rhino International
Back to the trenches: The bloody battle against rhino and elephant poaching

10:30 – 11:00 Break

11:00 – 12:00 **Session I** ***In situ*** **Asia and Africa**

11:00 – 11:20 Human-elephant conflicts and elephant conservation in Sri Lanka
Jayantha Jayewardene

11:20 – 11:40 Rhino Translocation in Assam under Medetomidine Anaesthesia
Dr. Bhupen Sarma

11:40 – 12:00 Elephants and Children in Burkina Faso: Developing a conservation ethic through the *My Elephant Neighbor* program, an assessement
Julien Marchais (introduction) & Erica Rogers MA

12:00 – 13:30 **Lunch**

13:30 – 15:10 **Session II** ***In situ*** **Asia and Africa**

13:30 – 13:50 Elephants, land and people striking a balance
Jim Nyamu

13:50 – 14:10 The Sabah Rhino project – Captive Breeding, habitat protection and habitat reforestation
Dr. Petra Kretzschmar

14:10 – 14:30 Ascertaining the causes and proposed mitigatory action for the Human-Elephant Conflict in selected areas
Pubudu Darshana Weerarathana

14:30 – 14:50 One horned Rhinoceros Conservation in Manas Tiger Reserve, India
Dr. Pranjal Bezbarua

14:50 – 15:10 GIS Analysis of Asian elephant Ranging Pattern in Manas National Park, India suggested Landscape Level Conservation Action in Indo-Bhutan Landscape
Bhrigu Prasad Saikia

15:10 – 15:30 Break

15:30 – 17:10 Session III *In situ* Asia and Africa

15:30 – 15:50 Activity Budgeting of the great Indian one horned rhino (*Rhinoceros unicornis*) in the Rajiv Gandhi Orang National Park, India
Dr. Buddhin C. Hazarika

15:50 – 16:10 Characterizing Sleep Behavior of the Wild Black Rhinoceros in Addo Elephant National Park, South Africa
Dr. Rachel Santymire

16:10 – 16:30 Prevalence of Tuberculosis among wild elephants in Sri Lanka
Dr. B. Vijitha Perera

16:30 – 16:50 Mycobacterium tuberculosis in free ranging Asian elephants
Arun Zachariah

16:50 – 17:10 Status of Human-Elephant Conflict in Manas Tiger Reserve, India with Special Reference to Eastern Buffer Zone
Pranjal Bezbarua

Wednesday, 12 October 2011

08:00 – 09:00 Registration

08:00 – 10:00 Session IV Emerging Diseaess – Elephant Endotheliotropic Herpesvirus (EEHV)

08:00 – 08:20 Emerging Viral diseases in wildlife with special emphasis on elephants and rhinos
Prof. Ab Osterhaus

08:20 – 08:40 Three fatal cases of endotheliotropic herpesvirus in Asian elephant (Elephas maximus) calves
Andrew Routh BVSc

08:40 – 9:00 Expanded Genetic Analysis of Five More Elephant Endotheliotropic Herpesvirus Genomes (EEHV2, 2x EEHV5 and 2x EEHV6) from Asian or African Elephants and its Use for Development of New Diagnostic and Serological Reagents
Dr. Gary Hayward

09:00 – 09:20 EEHV-5, a newly recognized elephant herpesvirus associated with clinical and subclinical infections in a herd of captive Asian elephants (*Elephas maximus*).
Dr. Lauren L. Howard

09:20 – 09:40 New ELISA and vaccine development for EEHV
Dr. Byron Martina

09:40 – 10:00 Endotheliotropic Elephant Herpes Virus Hemorrhagic Diseases in Asian Elephants of Southern India
Dr. Arun Zachariah

10:00 – 10:20 Break

10:20 – 11:40 Session V Emerging Diseases

10:20 – 10:40 Clinical research into endotheliotropic elephant herpesvirus at ZSL Whipsnade Zoo
Andrew Routh BVSc

10:40 – 11:00 Herd monitoring for elephant endotheliotropic herpesvirus (EEHV) in captive Asian elephants (*Elephas maximus*).
Dr. Lauren L. Howard

11:00 – 11:20 Pericardiocentesis: Description of a new technique ICU for the management of EEHV
Dr. Jon Cracknell

11:20 – 11:40 Launch of Website www.eehvinfo.com
Dr. Jon Cracknell

11:40 Lunch & VISIT THE ROTTERDAM ZOO

EEHV WORKSHOP (EEHV researchers and guests)

Thursday, 13 October 2011

08:00 – 09:00 Registration

08:00 – 10:00 Session VI Behavior

08:00 – 08:20 From Chitwan to Vienna – How do gait parameters change in a pair of Indian Rhinos (*Rhinoceros unicornis*) coming from semi-wild conditions to a European Zoo?
Regina Pfistermüller MSc

08:20 – 08:40 African elephants change gate when walking downhill
Dr. Robert Dale

08:40 – 09:00 The use of salivary cortisol to assess the welfare of elephants
Dr. Immanuel Birmelin

09:00 – 09:20 Personality assessment and factors influencing personality in captive
Saki Yasui

09:20 – 09:40 Insightful Problem-solving in an Asian elephant
Preston Foerder

09:40 – 10:00 The self-recognition of an African elephant
Dr. Immanuel Birmelin

10:00 – 10:20 **Break**

10:20 – 12:00 **Session VII Reproduction**

10:20 – 10:40 Results from two decades of reproductive steroid monitoring in white rhinoceroses kept in European zoos
Dr. Franz Schwarzenberger

10:40 – 11:00 Elephant-sized hot flashes? The relationship between social status and ovarian cycle activity provides evidence for menopause in female African elephants
Dr. Elizabeth Freeman

11:00 – 11:20 Emergency neonatal care of an Indian Rhinoceros (*Rhinoceros unicornis*) calf
Andrew Routh BVSc

11:20 – 11:40 Fertility and aggression control in elephant bulls through GnRH vaccination
Dr. Imke Lueder

11:40 – 12:00 Establishment of elephant gamete and embryo bank of wild founders for captive propagation programs
Dr. Thomas B. Hildebrandt

12:00 – 13:30 **Lunch**

13:30 – 15:10 **Session VIII Reproduction**

13:30 – 13:50 Using Fecal Hormonal Analysis to Determine the Factors affecting the success of the Black Rhinoceros in Addo Elephant National Park, South Africa
Dr. Rachel Santymire

13:50 – 14:10 Sperm Sorting and Preservation Technologies for Sex Ratio Modification in the Elephant and Rhinoceros: An Update
Dr. Justine O'Brien.

14:10 – 14:30 Ovulation induction for timed artificial insemination in the Sumatran Rhinoceros (*Dicerorhinus sumatrensis*)
Dr. Terri Roth

14:30 – 14:50 The white rhino EEP breeding Programme: Analysis throughout the years
Lars Versteege

14:50 – 15:10 Oestrous synchronization in white rhinoceros
Dr. Robert Hermes

15:10 – 15:30 **Break**

15:30 – 17:00 **Session IX Nutrition**

15:30 – 15:50 Body Condition Scoring of captive white rhinoceros *(Ceratoherium simum)*
Thijs D van den Houten

15:50 – 16:10 Laser assisted photogrammetric technique to measure shoulder height and body length of elephants
S. Wijeyamohan

16:10 – 16:30 Comparison of serum electrolytes and minerals between wild, captive concentrate fed, and captive forage on fed White rhinoceros (*Cerathotherium simum simum*)
Dr. Ray Ball

16:30 – 17:00 Black rhino population performance in relation to Plant Available Nutrient and Plant Available Moisture.
Benson Okita Ouma

Friday 14 October 2011

08:00 – 09:00 Registration

08:30 – 10:10 Session X Veterinary Care

08:30 – 08:50 Preliminary epidemiological findings using serology in an outbreak of Mycobacterium tuberculosis in managed Asian elephants (*Elephas maximus*) at a single facility
Dr. Ray Ball

08:50 – 09:10 Tuberculosis in elephants: the IFNgamma assay added to the diagnostic spectrum
Taweepoke Angkawanish

09:10 – 09:30 Update from the Stakeholders Task Force For The Management And Research Priorities Of Tuberculosis For Elephants In Human Care
Dennis Schmitt

09:30 – 09:50 Hemosiderosis and Clinical Findings Consistent with Black Rhino Syndromes in Greater One Horned Rhinoceros (*Rhinoceros unicornis*)
Dr. Ray Ball

09:50 – 10:10 Romifidine-Ketamine and its Reversal with Atipamezole in elephant paediatrics
Dr. Bhupen Sarma

10:10 – 10:30 Break

10:30 – 12:10 Session XI Veterinary Care

10:30 – 10:50 Health care feeding and adaptation in two Black Rhinoceros (*Diceros bicornis*) imported from Japan to the National Zoological Gardens in Sri Lanka
Jagath Jayasekara

10:50 – 11:10 Chronic perforating foot sole lesions in 2 captive great Indian rhinoceroses (*Rhinoceros unicornis*)
Dr. Willem Schaftenaar

11:10 – 11:30 The Mobile Elephant Clinic, a tool for disease monitoring in Thailand
Taweepoke Angkawanish

11:30 – 11:50 Investigating the hemoglobin of white rhinoceros (*Cerathotherium simum simum*) and possible implications for anesthesia
Dr. Ray Ball
11:50 – 12:10 Prevalence of *Cobboldia elephantis* in free ranging elephants in Sri Lanka
Dr. B. Vijitha Perera

12:10 – 13:30 Lunch

13:30 – 14:30 Session XII Conservation Organizations
13:30 – 13:50 Asian Elephant Support
Linda Reifschneider
13:50 – 14:10 Research and Rhinos: what we need to know to conserve them
International Rhino Foundation, Dr. Terri Roth
14:10 – 14:30 International Elephant Foundation
Deborah Olson

14:30 – 15:00 CLOSING CEREMONY

Saturday 15 October 2011

The post conference tour - Beekse Bergen Safari Park

KEYNOTE ADDRESS

BACK TO THE TRENCHES: THE BLOODY BATTLE AGAINST RHINO AND ELEPHANT POACHING

Cathy Dean
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Rhino and elephant poaching is on a scale not seen since the 1970s and 1980s, when Richard Leakey burned Kenya's ivory stockpile and Zimbabwe fought the rhino wars.

This presentation looks at the factors involved in the poaching crisis: the rise in demand, attributed to the growing Chinese footprint in Africa and the increasing wealth of an expanding middle class in East Asian countries; the patterns of poaching, which vary from country to country; and the impact on the elephant and rhino populations of range countries.

A variety of different approaches are being adopted to attempt to prevent or mitigate the affects of poaching, including increased levels of security and specialised equipment and the use of intelligence and informer networks. Save the Rhino International, with its partners such as the International Rhino Foundation and several European zoos, provides funding and technical support for the ongoing running costs of some 15 field programmes in Africa and Asia.

Attempts to promote awareness in consumer countries of the problems caused by poaching of endangered species are in their infancy, although efforts have been made through the CITES Standing Committee meeting in August 2011 and bilateral talks between the South African and Vietnamese governments. Other approaches are also being considered, such as a move to develop a legal trade in rhino horn; though those opposed to the legalisation of trade point to the continuing elephant poaching despite one-off sales of ivory from national stockpiles. There remains much work to be done in rhino range countries and in the consumer countries to protect wild populations of both taxa.

Cathy Dean read History of Art at university and then worked for specialist art bookshops and publishers, before becoming Campaign Manager for the Tate Modern and Tate Britain capital campaigns, raising over £50 million from the private sector. She then freelanced as fundraising consultant until a wildlife holiday to Madagascar sparked a determination to raise money for endangered species and habitats.

Since joining Save the Rhino International as Director in 2001, Cathy has developed the organisation's fundraising, grant-making and partnership activities. Save the Rhino now raises US $1.5 million per year to help support c. 15 in situ rhino conservation programmes, and has close working partnerships with other conservation bodies such as the IUCN African and Asian Rhino Specialist Groups, the International Rhino Foundation and EAZA headquarters and member zoos.

HUMAN-ELEPHANT CONFLICTS AND ELEPHANT CONSERVATION IN SRI LANKA

Jayantha Jayewardene

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Sri Lanka is an island lying south of India with a land area of 6.56 million hectares or 16.21 million acres. It presently has a population of over 20 million people. In 1881 the forest cover in the island was 84% of the land area. At the turn of that century it was down to 70% of the land area. Due to the increasing population, large tracts of forests have been cleared, mainly for settlement and agriculture. The forest area continued to decline and in 1956 it was 44%, in 1981 it was 27% and now it is 20%. This shows a drop of 64% in just 116 years.

Elephants and the people of Sri Lanka have a long history of coexistence. The religious and socio cultural traditions in the country that advocate a benevolent attitude towards wildlife are, however, eroding. Land clearing for agriculture, settlements, plantations and other development in areas occupied by elephants, with no forethought, has raised the frequency of contact –and conflicts – between man and elephant.

Casualties and fatalities of both elephants and humans as a result of these conflicts are high. On an average 120 elephants and 65 humans are killed each year. Sri Lanka has only an estimated 4,000- 4,500 elephants in the wild. Most of the deaths of elephants are due to retaliation for crop depredations, damage to houses and loss of human lives.

Unless immediate action is taken to reduce these conflicts, more importantly, the attitudes of the people, casualties on both sides will rise. Even at present, because of the significance of elephants in Sri Lankan history and culture and the general favourable attitude by most towards conservation, these conflicts have become significant socio-economic and political issues.

Many strategies have been adopted to minimize the HEC. Strategies have also been adopted to conserve the Sri Lankan elephant in the wild.

The Biodiversity and Elephant Conservation Trust (BECT) has a programme that is designed to create an awareness among the younger generation of all aspects of an elephant's life – biology, physiology, religious and cultural significance etc. It also focuses on the urgent need to conserve Sri Lanka's elephant in the wild.

RHINO TRANSLOCATION IN ASSAM UNDER MEDETOMIDINE ANAESTHESIA

Dr. Bhupen Sarma, B.Dutta; A. Talukder; B. Choudhury

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Translocation means transportation of an animal from one place to another. This is commonly applicable in case of wild life with a purpose to reduce the increased population of particular forest or to revive the lost population in another park. It is generally targeted with conservation of a particular species. Therefore, few female and a male animal are generally translocated to a jungle for propagation of the species. In this task it is always kept in view that the female and the male animals are selected from different jungles to avoid inbreeding affect of the offspring. If special care is initiated to protect the translocated animals in their new homeland, they will adapt well, reproduce their own and this wild species will be saved from extinction. As such rhinos from Kaziranga National Park were successfully translocated to Manas National Park.

Five one horn Asian rhinos were initially darted by using mixture of Medetomidine(20µg/kg) and ketamine(200mg/Rhino) anaesthesia of which four were translocated. These rhinos were rescued at a very young age (100kg body weight aprox) by Wild Life Trust of India and reared in their rescue centre until they grew up to 1000 kg body weight approximately. Induction time was recorded at 8.6±2.24 minutes which thereafter became unstable and came to the sternal recumbency. Eyes were closed and total relaxation of muscle with flaccidity of tail was observed in all the cases. Heart rate recorded at 15 and 30 minutes of anaesthesia were 39.25±1.60 and 33.82±1.33 per minutes. The respiration recorded at 15 min was 27.20±2.12 which reduced to 25.60±2.42 at 30 min. Slight drop of temperature from 37.33±1.20 to 37.16±1.85 ºC was recorded at 15 and 30 minutes respectively. After induction, the heart and the lungs of the rhinos were examined and Dexamethasone was injected to attain cardiac stability. Radio collars were fitted to the neck of the rhinos to investigate their movement after release in the forest. Each of the rhino was placed on a sleigh, tied with rope and pulled with a tractor near to the cage and put inside the cage. The rhinos were under anaesthesia for 39.25±3.12 minutes. Inside the cage, the rhinos were reversed from anaesthesia by intravenous injection of Atipamezole (double amount of medetomidine). The reversal time recorded was 8.5±1.89 minutes, until the rhinos were able to stand. Following reversal from anaesthesia, the rhinos urinated profusely. Mild tranquilizer Azaperon (140mg total dose/rhino) was injected to avoid aggressive behavior of the rhino and to make them anxiety free. The cage was lifted by a crane onto a truck and water was sprinkled on the body of the rhino to make it cool. Importance was given for full comfort of the rhino inside the cage and the speed limit of the truck was fixed at 40km/hr to avoid jerking.

Then the rhinos were transported to Manas National Park in presence of a team of veterinary Doctors. At certain interval, the rhinos were examined inside the cage by the expert team of veterinary, which was accompanying just at the back of the rhino bearing truck and thus reached destination. The vet team was accompanied by forest officials and Director IFAW from Boston,USA and the officials of Wildlife Trust of India(WTI). A big team of photography from Discovery Channel, Mexico and national news papers also accompanied the vet team. A pre-release centre was prepared inside Manas National Park by covering an area of 4 kilometers with electric fence. A trench was prepared to adjust the height of the truck and opened the cage by facing towards the release site. An injection of steroid was given to the rhinos as anti stress medication before release to the new homeland. The rhinos walked out slowly from the cage and enter into the jungle covered by electric fence. The site for wallowing and drinking water was accommodated inside the fence area. Radio collars were activated immediately after its release.

Since 1987, medetomidine was used in non domestic animals either alone or in combination with ketamine (Jalanka, 1987) but no record has been made of its use in one horn

rhino. Medetomidine was used for the first time for translocation of one horn rhinos in Assam (India), which is a global record and found to be very smooth in action. Thus it has proved superiority over the commonly used opioid narcotics which are detrimental to the normal functions of the animal. Threat for opioid induced abortion is not at all present with medetomidine. Moreover, the respiratory distress and cardiac arrest leading to 2.9 % mortality recorded by Alford et.al.(1974) following use of the opioids in wild animals are not recorded with this particular anaesthetic. Optimum duration and depth of anaesthesia achieved by this drug was sufficient for performing the operations. Thus zero mortality and absence of deleterious effect of medetomidine acclaimed its use in rhino translocation. Ketamine always balanced the action of its partner anaesthetic and alleviates pain. Atipamezole a specific alpha-2 antagonist effectively reversed the medetomidine induced sedation.

ELEPHANTS AND CHILDREN IN BURKINA FASO: DEVELOPING CONSERVATION ETHIC THROUGH THE *MY ELEPHANT NEIGHBOR* PROGRAM, AN ASSESSEMENT

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The *My Elephant Neighbour* program was implemented in Burkina Faso with the hope that, through its field-based and school-based education program, children will retain knowledge about elephants and develop an attitude of conservation. This research study was implemented in order to evaluate the program's objectives by looking at behavioral, attitudinal, emotional, and knowledge-based changes in the students who participated in 2011. Specifically, a major aim of this research was to compare the impact of direct exposure versus indirect exposure to elephants on the development of a conservation ethic, in order to try to understand which interventions were effective and in what ways.

The sample was made up of 106 students in the CM1 level class (4^{th}-5^{th} grade equivalent) from six different schools in the Deux-Balé forest region, where there is a population of 200-400 elephants. Participants ranged in age from 9-15 years; there were 56 female and 50 male participants; and maternal and paternal ethnicity was distributed across 18 different cultural groups. The sample was split into four groups. The direct exposure group was made up of those who participated in the nature class in the Deux-Balé reserve and did get to observe the elephants. The indirect exposure group was made up of those who were given in-classroom education through the *My Elephant Neighbour* booklet. The booklet, in its first edition, included a culturally-appropriate educational story about the human-elephant conflict, as well as a section about elephants in the surrounding region, with several color photos of local elephants. Because knowledge about elephants is learned not only via this program but also through community, media and other modes of communication in daily life, we used a group of students of the same age, culture and level of education that did not participate in the *My Elephant Neighbour*

program as a control group. This year, because of improved park management and more space for the herds to safely inhabit, the consistency of elephant sightings on the nature class excursions was decreased. This led to the creation of a fourth group, those who went to the park, participated in the nature class, but had no direct exposure to elephants. Study participants were interviewed before and after interventions using a modified version of the Children's Environmental Attitudes and Knowledge Scale (CHEAKS; Leeming, O'Dwyer & Bracken, 1995) as the primary outcome measure. Open-ended questions and drawings across a variety of areas regarding elephant attitudes and knowledge were used.

The International Elephant Foundation proposed that, "personal experiences with elephants can create life-long conservationists and cannot be duplicated in other mediums," (www.elephantconservation.org). This central hypothesis was tested in Burkina Faso. Specifically, it was predicted that direct exposure to elephants will show the highest increase in environmental attitudes and knowledge specifically related to conservation of elephants. Analyses suggested that personal experiences with elephants have a greater impact than any other form of education in the development of a conservation ethic, supporting the hypothesis. Results showed a smaller, yet positive change in the indirect exposure group as well.

ELEPHANTS, LAND AND PEOPLE: STRIKING A BALANCE

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Kenya's elephant population dropped from an estimated 167,000 in 1973 to 19,000 in 1989. This decline was largely due to poaching fueled by demand for ivory. During this period, elephants took refuge in protected areas. The last 20 years population in parks has grown; security boosted, and elephants have started to re-establish their old dispersal areas.

This paper examines re-establishment of elephants in South Rift of Kenya, a key dispersal area sandwiched between Amboseli National Park and Maasai Mara Game reserve. ACC's (elephant project) 7 year research and monitoring in the area has revealed that, sizeable groups of elephants have re-appeared after a 20-year absence and have found haven in the area. The average group size is 2 but herds of 44, 60 and 120 have been sighted.

Movement patterns show that elephants cross both the eastern and western escarpment of the Rift Valley and a possible connection between the Amboseli and Magadi elephants exists. Previously undocumented cross-border movements have also been reported. Land use, as well as livelihood changes witnessed in the area that was predominantly pastoral also pose a major challenge and zoning should be the next inevitable option. As would be expected, human-elephant conflict is on the increase.

The biggest threat to elephant dispersal is land subdivision and opening up of new land for agricultural production. With the changing Land use system from GR to individual ownership breakdown of the group ranch tenure system to individual ownership, negotiating with landowners to maintain open system in a subdivided landscape is one of the few remaining

options. The project has initiated negotiations to win more space for the growing elephant population in the South Rift area.

THE SABAH RHINO PROJECT – CAPTIVE BREEDING, HABITAT PROTECTION AND HABITAT REFORESTATION

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The Sabah rhino (*Dicerorhinus sumatrensis harrissonii*) is a subspecies of the Sumatra rhino (Fig.1). It once occurred all over Borneo but habitat destruction and poaching led to a drastic reduction of the population within the last 15 years. Currently less than 50 individuals of the subspecies still exist. These few individuals occur in the northern tip of Borneo, in the East Malaysian state Sabah. Sabah is characterised by a high diversity in flora and fauna. Its rainforests are among the oldest rainforest of the world. But the majority of the forest areas in Sabah, especially the lowland forests, have been selectively logged in the past. The increasing demand for palm oil on the international market, has led to a high pressure on the remaining forest areas. Forest areas with a very low protection status, such as secondary forest, are therefore at a high risk to be converted into agricultural land. These areas are however very important for the animals such as the rhino and the elephant. They are buffer zones between agricultural land and primary forest and they are corridors for large animals connecting the fragmented landscapes.

The remaining Sabah rhinos are isolated from each other in small pockets of rainforest surrounded by plantations; here they are facing a high risk of inbreeding. Therefore breeding management of this highly endangered species becomes essential. In July 2007 the government of Sabah together with local and international Non Governmental Organisations (NGO) decided to start a rescue and breeding project for isolated individuals.

In the past, the captive breeding of the species has not been a story of success. Basic information from free ranging animals was lacking due to its elusive character, its rarity and the inhospitable nature of its habitat. This resulted in management problems as its basic requirements in terms of food, health and breeding were unknown. In recent years more data has been collected and captive breeding methods have fast developed. Non-invasive hormone analysis, ultrasound techniques and assisted reproductive techniques have been successfully applied for the reproductive assessment in a number of species, including the Sabah rhino. The Leibniz Institute for Zoo and Wildlife Research (IZW) and the Zoo Leipzig support the local government and NGOs with its proven scientific and captive breeding expertise ensuring the preservation of the Sabah rhino. An eventual viable population will largely depend on an intact forest. Afforestation and reforestation efforts are therefore needed to increase the carrying capacity of

the potential habitat. The Rhino and Forest Fund, a German NGO was founded to rise funding for saving the Sabah rhino and its vanishing habitat.

ASCERTAINING THE CAUSES AND PROPOSED MITIGATORY ACTION FOR THE HUMAN - ELEPHANT CONFLICT IN SELECTED AREAS

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Even though Sri Lanka is a small island of 65,000 km² land and a large human population of 22 million, the second largest mammal of the world is still living here without extinction. But the human-elephant conflict in Sri Lanka has grown rapidly in the past three decades.

When analyzing the data of Department of Wildlife Conservation (DWC) for the past 5 years (2006-2010); 353 lives, 1031 elephants and a large area of cultivated land has been destroyed and lost. If this trend persists, there will be loss of a large number of people, and pose a higher risk of extinction for the elephants. A sustainable solution is needed and for to initiate this process data was collected on the more affected people, the nature of the conflict and the period in which elephants mostly come to villages, location and the number of elephants in 10 Districts and 36 Divisional Secretariats.

Furthermore, mapping and the studying of these particular maps has been done with the data collected from the DWC zonal offices, field visits to locations that are extremely vulnerable from the sub offices of these areas.

The main task and the research was done to map the traditional elephant corridors ,locations under DWC, where electric fencing has not been done or fences that are inactive and the isolated forests areas with wild elephants.

Findings from this research showed that a lot of traditional elephant corridors have been blocked by illegal establishment of villages and developments and cultivations, and blocked by electric fencing. This has led the elephants to take alternative routes through villages and has been a reason for the escalation of the human-elephant conflict. Further, establishing of electric fences inside the forest and not on the outskirts of the forest and the difficulty in maintaining them, plus the villagers sending their cattle to graze on the grass and vegetation through the fences has immensely contributed to the scarcity of availability of food sources for the elephants.

Large areas of land have been cleared and extensively planted with crops that are palatable for elephants such as sugar cane and plantain.

In this research the main objective was to identify natural corridors, corridors that have been blocked and to open these pathways by reducing the activities of people that contribute to the blocking of the natural trails. Identifying correct locations for development projects that doesn't block the natural trails, doesn't harm the natural food sources of the elephants were other tasks of the research. Cultivating of crops, changing the possibility of the periods and make chena cultivation more methodical so that an extra food source will be there for the elephants. Through this was looked into to reduce the number of deaths to both humans and elephants. It is

intended with the research to come up with a sustainable development and conservation activities for elephants

ONE-HORNED RHINOCEROS CONSERVATION IN MANAS TIGER RESERVE, INDIA

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The Indian one horned rhinoceros, having a global population of 2400 in the year 2005 has been facing increasing threat from organized poaching and habitat loss. In Assam the threat ranges from local elimination from different protected areas including Manas Tiger Reserve, stochastic risk in Kaziranga National Park and Pabitora Wildlife Sanctuary, increasing man-rhino conflict; continuing poaching in the all three rhino exiting areas having a population about 2000 animal. Therefore, forest department, Assam with support of all stakeholders designed a programme Indian Rhino Vision-2020 that focused overall increasing security and rhino translocation to increase the population from 2000 to 3000 by 2020 which will be distributed in six protected areas. Being a partner to this mega programme, we started a specific programme in Manas tiger reserve, a former rhino habitat selected for first translocation.

The programme aims to study the habitat suitability of the one horned rhinoceros in Manas tiger reserve to facilitate translocation, to rehabilitate ex-poachers turned conservation workers and forest dependent of the area and conservation education and capacity building in fringe villages of the reserve. Remote sensing technique and GIS was used to generate maps considering all ground information like former rhino habitat in grassland and forests communities of Manas, aquatic bodies, topography, different anthropogenic factors namely past and present poaching threats, encroachment, weed invasion, grazing pressure from domestic cattle, burning intensity; awareness level in fringe villages, departmental infrastructure etc. The information helped in preparation the final map on rhino habitat suitability indicating most suitable (122 sq. km.), suitable (240.80 sq. km.), less suitable (125.57 sq. km.) and unsuitable zone (31.29 sq. km.) in Manas tiger reserve. The most suitable area can be increased to 220 sq. km. by minimizing the poaching threat and law and order problem near zero level. Currently the suitable area without poaching threat is covering 201.25 sq km. followed by less suitable 69.38 sq. km. and unsuitable as 28.95 sq. km. Secondly, the rehabilitation programme for families of conservation volunteers (that included hardcore surrendered ex-poachers) and forest dependents were started with introduction of capacity building and in kind support on alternative livelihood options on agro and veterinary products.

The result was gradual behavior change in terms of growing interest and confidence building which helped in achieving 50-70 percent success amongst different groups in next phases and replication. We voluntarily helped local NGOs in their ecotourism and 1370 pound was raised to supply ration for 80 conservation volunteers patrolling the Manas tiger reserve during critical period. We also promoted school education for the children (vulnerable to become future poachers) of poor conservation volunteers and forest dependents. Increase of awareness and capacity building on rhino

conservation was initiated by preparing a conservation leaflet in local language which was used in all conservation meetings and distributed amongst all local stakeholders.

GIS ANALYSIS OF ASIAN ELEPHANT RANGING PATTERN IN MANAS NATIONAL PARK, INDIA SUGGESTED LANDSCAPE LEVEL CONSERVATION ACTION IN INDO-BHUTAN LANDSCAPE

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The endangered(IUCN) and schedule-I (WPA,1972) Asian elephant ranging pattern in the Manas National Park(MNP) , India is not known which ranges in the foot hills region of the eastern Himalaya between Bhutan and India also to the fringe villages in the southern boundary of MNP creating conflict with human. Hence this was found necessary to evaluate the ranging pattern in MNP and adjoining areas of the Indo-Bhutan landscape.

Data on Asian Elephant ranging pattern in Manas National Park were collected using Individual identification, recording GPS location of sighting during field work from 2006-2009. Home ranges estimation were done by 100% Minimum Convex Polygon method using GPS Locations of identified elephant sighting in GIS (Arc View) for creating home-range polygons.

Asian elephant seasonal home-ranges along with annual home range, annual core home range were evaluated. All the home range polygons of tracked elephant falls both side of India and Bhutan political boundary. Some of the tracked Asian elephants (both males and females) were seen maintain small or small to medium-sized home ranges throughout the year. Asian elephant home ranges in Manas National Park were oriented in east-west, and more compatible with the idea of habitat existence and availability of grassland in the Park and mineral licking sites inside the Bhutan. This was found that tracked elephant spent time regularly visiting the natural salt licking sites in the Bhutan. All the tracked elephant home ranges covers several movement tracks covering area both in India and Bhutan. But large breeding bulls frequently switched between the southern boundary of Manas National Park. There is distinct difference between the size of home range of Male and Female Asian elephant.

The home range estimation results clearly shows difference between the home range size of male and female and this difference is for the reason that breeding bulls ranges larger area in search of estrous female and takes more risk in entering fringe village area. Annual home range is lesser than exact habitat area in the Indo Bhutan landscape showing, they are not devoid of quality habitat. Again size of core home range area is very small. Most of the elephants were found to be residential inside MNP but visited mineral licking sites, water source, habitats regularly inside the Bhutan. Home range of Bulls falls into the fringe village areas of MNP as their prime target is to raid crops. However those elephants whose home range polygon falls more in Bhutan habitat compared to other were found to contribute less towards human elephant

conflict in fringe village area of MNP. Hence there is a necessity of trans-boundary management plan for the conservation of Asian elephant targeting habitat management and management of the movement tracks on Landscape level between India and Bhutan. This will help in achieving reduced level of human-elephant conflict and more successful action towards the conservation of this endangered Asian elephant in the Indo-Bhutan landscape.

ACTIVITY BUDGETING OF THE GREAT INDIAN ONE HORNED RHINO *(RHINOCEROS UNICORNIS)* IN THE RAJIV GANDHI ORANG NATIONAL PARK, INDIA

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Activity budgeting of an animal denotes the allocation of time in various diurnal (or nocturnal for certain animals) activities in a specific time period. Study of activity budgeting is very essential for a species to understand its life style characteristics and is a foundation stone for interrelating the ecology and behaviour of a animal species. The main objectives of the present study were (a) to investigate the activity pattern of Indian Rhino in RG Orang National Park in different seasons of the year and (b) to identify the major behavioural activities that play a vital role in time allocation in different activities of Indian Rhino. This will help to lay a site-specific conservation strategy for the Indian Rhino, especially in RG Orang National Park or other similar Rhino habitats of the Brahmaputra floodplain area. Activities are the behavioural output of an individual or group of animals of a species in response to ecological and biological factors, such as body size, diet availability, distribution, abundance of food resources, climate, competition for resource, mate availability etc. Time is a hidden constraint that affects all other behaviours. The activities of the animals are also an important indicator of the health of a habitat, which reflects the status and distribution pattern of the resources. Very little attempt was made to study the activity budgeting of Indian rhino in the Brahmaputra floodplain which harbours more than 71 % of th Indian Rhino Population.

Field surveys for activity budgeting of Indian Rhinos were done during day light hours in Orang National Park**.** *Scan Animal Sampling* (Altman, 1974) was found to be suitable for sampling the activity budgeting of Indian Rhino in Orang National Park. The *Ad. Libitum Sampling* method (Altmann, 1974) was also used to record the important activities between two scans. The activity patterns such as feeding, locomotion, comfort, wallowing, vigilance, non-breeding play, breeding play, agnostic and all other behaviours related to its breeding and non-breeding purposes etc. were recorded in the data sheet. During data collection, the uniformity was maintained to represent all age and sex compositions of Rhino. The time allocation for various behavioural activities by an animal may be determined either by availability of time or habitat condition, as well as other ecological factors. To find out this selectivity, the seasonal variation of time spent in different behaviors were compared with the overall time allocation in different activities.

The present study revealed that, the Indian Rhino showed distinct variation of activity pattern in different seasons of the year. The Indian Rhino in Orang National Park spent a maximum of 46.2% time on feeding activities, followed by wallowing 18.4%, vigilance 15.1%, locomotion 9.1%, comfort 8.01% and minimum of 5.6% in other miscellaneous activities. The results indicated that, feeding was the guiding factor, which effect on time allocation in various activities possessed by Indian rhino.

CHARACTERIZING SLEEP BEHAVIOR OF THE WILD BLACK RHINOCEROS IN ADDO ELEPHANT NATIONAL PARK, SOUTH AFRICA

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Sleep is a universal behavior in the daily cycle of mammals; however, little is known about sleep patterns of animals under natural conditions. The present study was aimed at collecting normative sleep data from wild black rhinoceros (*Diceros bicornis bicornis*) managed in two different sections of Addo Elephant National Park, South Africa. Specifically, we investigated the influence of sex, age and the wet/dry season on sleep patterns and sleep duration. We compared two sections (Addo and Nyathi), which differ greatly in biotic/abiotic factors. Digital, infrared cameras were erected on poles at known bedding sites. Three photos were taken when the passive motion detector was activated with 15 second intervals. Data were collected from September 2009 to March 2010 from eight individually identifiable adult rhino that slept at the two bedding sites. A total of 2417 photos (Addo, n =1626; Nyathi, n=914) captured rhino behavior during 40 separate sleeping bouts (Addo, n=15; Nyathi, n=25). Mean age of the individuals was 10.3 ± 2.4 years for females and 8.4 ± 3.4 years for males in the study. Results demonstrated that age and season did not affect (P>0.05) the sleep behavior; however, sex did influence the duration of sleeping bouts with males (n= 27; mean, 105. 6± 11.3 min; range, 14.0 – 202.0 min) sleeping longer ($F_{1,48}$=6.93,P=0.01) than female rhinos (n=13; mean, 58.6 ± 10.4 min; range, 11.0 – 132.0 min). The park sections (Addo vs. Nyathi) did not influence the duration of sleep, but did affect (rw_{40}=0.88; P<0.025) the time at which the rhinos slept (Addo, 2000 - 2400 hr; Nyathi, 2000 – 0400). Through quantitative studies on mammalian sleep patterns in their natural ecosystems, we have a greater understanding of the biological factors that affect sleeping patterns in wild black rhino, providing information to assist the management and conservation of this highly endangered species.

PREVALENCE OF TUBERCULOSIS AMONG WILD ELEPHANTS IN SRI LANKA

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Tuberculosis (TB) is an infectious, granulomatous disease caused by acid-fast bacilli of the genus *Mycobacterium*. The disease affects practically all species of vertebrates. Limited studies have been conducted for TB surveillance in free ranging wild animals, due to difficulties in applying modern surveillance techniques under such conditions. In the case of elephants ante-mortem diagnosis has been limited by challenges associated with sample collection, test administration and interpretation. TB in captive elephants has been recognized as a re-emerging zoonotic disease since at least the 1996. This study was conducted to determine the prevalence of antibodies to TB among free ranging wild elephants and a semi-captive group in Sri Lanka.

The study was performed using 89 serum samples collected from free ranging elephants from different parts of the country (n=42) and a group of young orphaned elephants being rehabilitated for re-introduction to the wild at the Elephant Transit Home (ETH, n=47). The blood samples of free ranging elephants (ages 15-50 years) were collected after immobilization for treatment or for translocation. The blood samples of young orphaned elephants (ages 1-5 years) were collected while they were sleeping or while being subjected to treatments or transportation under sedation. The serum were separated and stored at -20^{o} C. Samples were assayed with Elephant TB Stat-Pak® (Chembio Diagnostic Systems, Inc., Medford, NY) for antibody detection. The presence of antibody to TB was detected in 3 out of 42 (7%) wild elephants, and in 15 out of 47 (31.9%) orphaned elephants at the ETH. This is the first report of the presence of antibody to TB among wild elephants in Sri Lanka. The high prevalence of TB among orphaned baby elephants may be due to chronic stress that leads to reduction of immunity or to transmission of the disease from elephant keepers.

The dilemma of what should be done with a wild animal that tests positive for TB is a major problem in wildlife conservation. Although previous studies have shown that the Elephant TB Stat-Pak serologic assay demonstrated 100% sensitivity and 95-100% specificity, isolation of the causative bacteria through culture is required for the confirmation of presence of disease. Even after confirming that TB is present in a wild species, eradication of the disease is a huge challenge. TB is considered a disastrous disease for wild animals from the conservation point of view and it can be evaluated in three different ways, based on its impact on the economy, zoonotic importance and the harm done by the infection itself. Firstly, wild animal TB represents a permanent reservoir of infection and poses a serious threat to control and elimination programs. Secondly, as a zoonotic disease, it affects their *in-situ* and *ex-situ* conservation programs as well as tourism industries. Thirdly, a TB epidemic in wildlife has a number of implications on the fecundity, morbidity and mortality of the population. Further studies are warranted on the prevalence of TB in wild and captive elephants in Sri Lanka, and on potential strategies for control of the disease.

STATUS OF HUMAN-ELEPHANT CONFLICT IN MANAS TIGER RESERVE, INDIA WITH SPECIAL REFERENCE TO EASTERN BUFFER ZONE

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The human-elephant conflict in the Manas tiger reserve remains complicated even after significant improvement of security after 2003 resulting significant threat to Asian elephant (*Elephus maxima*) population. Therefore, this study deals to collect base line data on the problem and started experimental action to design future strategies. We selected a major part of neglected eastern buffer zone of the reserve including fringes (365 sq. km.) where no work was carried out earlier. With support of remote sensing techniques, the study identified significant encroachments in all Reserve Forests (RF) especially 60% encroachment in Subankhata RF. An elephant corridor was identified in the area that was along the riparian zone. According to villagers, it is a traditional migration route of elephant from northern Bhutan hills to southern forest patches, now heavily degraded. The survey in two zones indicated 10 human deaths by elephant attack, 20-60% crop damage incidents for 71% of families sampled, property and household damage of 53 families and 2 elephant deaths (2010-11). Of course, outside our study site, death of 5 elephants and 7 human were reported in 2011 in eastern buffer. The population of elephant in the studied sites is around fifty (during last year) having herd size of 10-14 and 2- 5 individuals including lone rogue elephants. Crop raiding and household damage can be strongly linked to presence of palatable rice (grass species) in encroached habitat and corridor. Absence of proper grassland in buffer areas may be a reason of longer elephant raid season if compared with core zone that has sufficient grassland. The conflict zone covers about 0-8 km south from reserve boundary, where nearby fringes, corridor and encroached villages were badly affected. Secondly our socioeconomic survey helped in identification of alternative livelihood options like piggery, weaving, bee keeping, horticultural crops including items not preferred by elephant. Two local NGOs that converted local poachers and wood cutters to conservation volunteers are patrolling in the area since 2006. They already restored about 2 sq. km. of forests having 4-5 years old trees and have traditional knowledge to deter elephants. We are working with them and helping 40 self help groups to promote piggery and weaving options besides bee keeping technique. This is a part of our long term community conservation programme for rehabilitation of families of conservation volunteers (that included hardcore surrendered ex-poachers) and forest dependents affected by elephant attack. These activities will be outside the encroached area to make villagers busy in those alternative options. This way we are trying to recover and restore some of the sensitive encroached area falls on the corridor. On the other hand, we are providing anti-poaching kits, dragon lights and solar torch lights and lamps having mobile phone charging facility to strengthen current traditional method of the experienced volunteers to deter elephant including conservation awareness meetings to minimize the poaching threats. The recommendation suggested more detail study covering all areas, continue community

conservation programme and develop innovative techniques to mitigate the human-elephant conflict.

EMERGING VIRAL DISEASES IN WILDLIFE WITH SPECIAL EMPHASIS ON ELEPHANTS AND RHINOS

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Although many infectious diseases that have scourged humans and animals over the centuries have largely been brought under control in the industrialized world over the past decades – the most striking examples being the global eradication of smallpox and rinderpest - paradoxically we are currently being confronted with an ever increasing number of new infectious disease outbreaks. The origin of most of these outbreaks can be found in wild animal reservoirs from which these agents escape due to a complex mix of predisposing factors in our globalizing world. In turn it also affects wild animal populations by the spill- over or spill-back of the agents involved from humans or domestic animals into wildlife.

Over the past decades, several bacterial and viral infections have been reported in elephants worldwide. Tuberculosis and anthrax have been diagnosed in elephants. A new member of the Mycobacterium spp, *Mycobacterium elephantis*, is now recognized as an emerging pathogen among elephants. In addition, there is serological evidence for leptospirosis in Asian elephants. Cowpox virus, herpesvirus, and coxackievirus infections have been reported in both Asian and African elephants. Since the identification of the elephant endotheliotropic herpes virus (EEHV) in 1995 as a causative agent of fatal hemorrhagic disease of Asian elephants, the virus has caused significant mortality among newborn and young Asian and African elephants both in captivity and in the wild. In 1995, an outbreak of encephalomyocarditis (EMC) virus infection was reported in free-ranging African elephants in South Africa. Surveillance based studies indicated that this outbreak was probably caused by a significant increase of the population of infected myomorph rodents.

In captivity, black rhinoceroses (Diceros bicornis) have been plagued by many disease entities that have not been described in this species in the wild. Recent efforts towards the conservation of endangered rhinoceroses in several areas including re-introduction of the animals into regions where they have lived previously have been been hampered by mortalities among translocated animals. Sero-surveillance studies performed in free-ranging black and white (Ceratotherium simum) rhinoceros in the Republic of South Africa,, Namibia, and Kenya indicated the presence of antibodies specific to viruses causing Rift valley fever(12%), Akabane disease(59.8%), bluetongue (55%), African horse sickness (27.9%), epizootic haemorrhagic disease of deer (19.4%), parainfluenza type 3 (25.3%), bovine herpes 1 (3.1%), equine herpes 1 (8.8%) and bovine viral diarrhea (1.2%) as well as four serovars of Leptospira interrogans, (ranging 1.2 to 8.8%). In addition, infections with cowpox virus, several parasites such as *T. congolense*, *T. simiae spp*, and T. *godfreyi*, Theileria spp., Neospora caninum and microfilaria, as well as fungi (Coccidioidomycosis) have been shown to cause disease of varying severity in black and/or white rhino's.

Laboratory based diagnostics and ongoing epidemiological as well as virus discovery programmes that will also be targeting elephants and rhino`s will benefit greatly from the development of novel serological and molecular techniques. This will yield a better insight into the infectious agents that should be considered in the management of captive and semi-wild elephants and rhino`s and in future conservation efforts for these animals.

THREE FATAL CASES OF ENDOTHELIOTROPIC ELEPHANT HERPES VIRUS IN ASIAN ELEPHANT (*ELEPHAS MAXIMUS*) CALVES.

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ZSL Whipsnade holds a successful breeding group of Asian elephants (*Elephas maximus*). Currently there is one breeding bull and five cows. Two cows have a dependant calf and one cow is in her last trimester of pregnancy. Between 2006 and 2009, three calves (2.1) developed clinical signs of endotheliotropic elephant herpes virus (EEHV). Signs progressed quickly. They started with minor lethargy and depression in all cases. Cyanosis of the tongue appeared in early stages of disease and progressed throughout the disease process. Oedema was present, mainly on the head and trunk and all three calves died within 48 to 53 hours after onset of clinical signs despite intensive treatment with antiviral drugs. One calf also received elephant plasma during treatment.

Post-mortem was highly suggestive for EEHV. Pericardial effusion, extensive petechial to ecchymotic haemorrhages involving the epi- and endocardial heart surfaces and throughout the myocardium, diffusely scattered petechiae within the viscera and parietal peritoneal serous membranes and intracranial haemorrhages have all been reported in the Whipsnade Zoo elephants. No inclusion bodies could be observed during histo-pathological examination in any of the cases. PCR confirmed EEHV1b in the first case and identical EEHV1a in the other two cases.

This presentation will discuss the clinical signs, the treatment protocols and the post-mortem findings of all cases.

EXPANDED GENETIC ANALYSIS OF FIVE MORE ELEPHANT ENDOTHELIOTROPIC HERPESVIRUS GENOMES (EEHV2, 2X EEHV5 AND 2X EEHV6) FROM ASIAN OR AFRICAN ELEPHANTS AND ITS USE FOR DEVELOPMENT OF NEW DIAGNOSTIC AND SEROLOGICAL REAGENTS

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Since their discovery in 1999, novel herpesviruses named Elephant Endotheliotropic Herpesvirus (or EEHV) within the genus Proboscivirus have been identified as the cause of more than 75 cases worldwide of fatal hemorrhagic disease in elephants. These have occurred predominantly in Asian calves (Elephas maximus) between 1 and 8 years of age, both in North American and European zoos, as well as in wild calves and orphans in several Asian countries. A total of eight distinct species or sub- species of EEHV have now been identified within elephants housed in North America, with the predominant culprits being a wide range of strains of EEHV1A. However, a few cases have instead involved EEHV1B, EEHV3 or EEHV4 in Asians and EEHV2 in Africans, and EEHV5 and EEHV6 have caused non- fatal viremia in Asian adults and an African calf respectively. In addition, EEHV2, EEHV3, EEHV6 and EEHV7 have been found in benign lymphoid lung nodules in culled or euthanized adult African elephants. Most captive Asian elephants at several closely monitored breeding facilities also sporadically shed either EEHV1A or its chimeric variant EEHV1B in trunk washes. Many facilities have also had viremic disease cases associated with strains of both EEHV1A and EEHV1B, and five closely monitored Asian elephants have now been observed to have had sequential non- lethal infections by both sub- species. Diagnostic standard or real- time PCR assays for identifying all of these many EEHV species, as well as for epidemiological tracking of strains, have or are being developed. In addition, to better understand the genetic relationships of some of the apparently less pathogenic EEHVs to the most commonly encountered EEHV1s, we have now generated 20- kb of PCR DNA sequence data for a second strain of EEHV2, as well as for both of the only two known strains each of EEHV5 and EEHV6. This data encompasses multiple genomic loci from DNA obtained from viremic blood (2x EEHV5, 1x EEHV6) or from lung nodule samples (1x EEHV2, 1x EEHV6). The genetic differences between all four species EEHV1, EEHV6, EEHV2 and EEHV5 within the AT- rich branch proved to average between 15 and 25% at all loci examined, representing about 10 to 20 million years of evolutionary divergence. All encode at least 15 characteristic core Proboscivirus genes including the TK, gB, MCP, POL, HEL, OBP, gN, gO, gH, gM and MyrTEG proteins. Except for in certain hypervariable genes, intra- species strain nucleotide variability is usually less than 0.5%. The existence of all of these different EEHV species and evidence that many healthy herdmates are likely infected with multiple species of EEHV greatly complicates the interpretation of current serology assays in both Asian and African elephants. Because the intact MyrTEG genes are now known for all seven EEHV types and are small, as well as highly diverged and are expected to be antigenic, we have been cloning and purifying them as GST- fusion proteins in yeast for potential use in next generation Elisa and proteomic chip serology assays to attempt to discriminate between antibodies to some or all of these viruses.

EEHV-5, A NEWLY RECOGNIZED ELEPHANT HERPESVIRUS ASSOCIATED WITH CLINICAL AND SUBCLINICAL INFECTIONS IN A HERD OF CAPTIVE ASIAN ELEPHANTS (*ELEPHAS MAXIMUS*).

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Elephant endotheliotropic herpesviruses (EEHVs) cause acute hemorrhagic disease with high mortality rates in Asian elephants (*Elephas maximus*). Recently, EEHV5 has been described, but its prevalence and clinical significance remains unknown. In February 2011, a 42 year old wild-born female Asian elephant presented with bilaterally swollen temporal glands, oral mucosal hyperemia, vesicles on the tongue, and generalized depression. The elephant had a leukopenia and thrombocytopenia. She was treated with intramuscular flunixin meglumine, and rectal famciclovir and fluids. Signs resolved gradually over 3 weeks, and the white blood cell count and platelets rebounded to elevated values. EEHV5 viremia was detectable starting one week before presentation and peaked at the onset of clinical illness. EEHV5 shedding in trunk secretions peaked after viremia resolved and continued for 2 months. EEHV5 trunk shedding from a female herd mate was detected prior to onset of clinical disease in the 42-year old elephant, suggesting that this animal was the source of the infection. Subsequent EEHV5 viremia and trunk shedding was documented in the rest of the herd, who remained asymptoma tic, except for 1 day of temporal gland swelling in an otherwise healthy 1 year old calf. These findings suggest that sporadic reactivation from a single animal was sufficient to spread infection to all susceptible herd mates and that EEHV5 can cause illness in some animals.

This captive herd had been closed for 28 months prior to detection of EEHV5, illustrating that transmission of EEHVs is unpredictable and may occur years after introduction of animals into a herd.

DEVELOPMENT OF AN ELISA FOR DETECTION OF ANTIBODIES AGAINST ENDOTHELIOTROPIC HERPESVIRUS

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Since the identification of elephant endotheliotropic herpes virus (EEHV) in 1995 as a causative agent of fatal hemorrhagic disease of Asian elephants, the virus has claimed more than 50 deaths of newborn and young Asian as well as African elephants both in captivity and in the wild. Despite the devastating disease that it may cause and the great risk it poses to elephant conservation programs, little is known about the epidemiology, prevalence and occurrence of EEHV. The department of virology of the Erasmus University in Rotterdam, The Netherlands, is involved in a project to develop a serological assay for detecting EEHV specific antibodies. To this end, the glycoprotein B of EEHV was cloned into the baculovirus expression system. Cell lysates were subsequently used as the source of antigen. Preliminary results of the sero-prevalence in sera of elephants from several European zoos will be presented.

ENDOTHELIOTROPIC ELEPHANT HERPES VIRUS HEMORRHAGIC DISEASES IN ASIAN ELEPHANTS OF SOUTHERN INDIA

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We are reporting highly fatal Endotheliotropic Elephant Herpes Virus in 15 cases of young elephants in South India. These represent both captive and free ranging Asian elephants. Etiology is confirmed by gross pathological lesions, detection of intra-nuclear inclusion bodies and direct PCR amplifications. In seven cases so far, data was obtained at multiple gene loci, including the Polymerase, Terminase, U71/gM and vGPCR, revealing five distinct strains of EEHV1A and one of EEHV1B. Two cases from calves that died within a few days of each at the same facility in Dec 2007, gave identical results at all four loci in multiple tissues of both animals showing that their infections were directly epidemiologically connected. The other cases examined that came from different locations and at different times were all readily distinguishable and showed a wide range of variability especially in vGPCR (including subtypes E*, E, E/A, D2, A2, B1). Nevertheless, these closely resembled and fell well within the range of variability seen amongst EEHV1A or EEHV1B strains characterized previously in pathological samples from Asian elephant cases in North America and Europe. Therefore, we conclude that the hemorrhagic disease occurring within young captive and wild Asian elephants in Southern India are exactly the same disease and are caused by closely related EEHV1A and EEHV1B strains as in captive ex situ elephants in North America and Europe. This is the first report of EEHV infections in free ranging elephants highlighting significant emergence of EEHV diseases in the world largest population of Asian elephants.

(CLINICAL) RESEARCH INTO ENDOTHELIOTROPIC ELEPHANT HERPES VIRUS AT ZSL WHIPSNADE ZOO.

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ZSL Whipsnade Zoo has had three fatal cases of endotheliotropic elephant herpes virus (EEHV) between 2006 and 2009 in their Asian elephant (*Elephas maximus*) herd. This has triggered many management changes. Furthermore, (clinical) research projects and collaborations have been set up to minimize further fatal cases of EEHV in our herd. This presentatio n will discuss all changes implemented in the past five years as well as future research goals. Breeding management, daily oral temperature monitoring, haematology and biochemistry monitoring, lysine supplementation and lauric acid supplementation will all be discussed.

In cooperation with the Veterinary Laboratory Agencies in Weybridge, UK, a real-time Taqman PCR was developed to diagnose carrier elephants and possible routes of virus transmission and determine herd prevalence of EEHV. ZSL Whipsnade Zoo also closely cooperates with Erasmus University on the development of an EEHV ELISA and the development of a recombinant vaccine. A future project is planned to improve the sensitivity of the real-time Taqman PCR by including an additional amplification step.

HERD MONITORING FOR ELEPHANT ENDOTHELIOTROPIC HERPESVIRUS (EEHV) IN CAPTIVE ASIAN ELEPHANTS (*ELEPHAS MAXIMUS*)

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Elephant endotheliotrophic herpesviruses (EEHVs) cause acute, often lethal hemorrhagic disease in a juvenile Asian elephants (*Elephas maximus*) and have a devastating impact on many breeding facilities throughout North America and Europe. Pro-active planning and vigilance are an institution's best defense against losing an elephant to EEHV. The Houston Zoo has developed an intensive EEHV monitoring program to optimize detection of illness before it is too late. Daily to weekly herd monitoring includes mouth exams, body weights, trunk wash collection for EEHV PCR, blood collection for CBC, serum chemistries, and EEHV PCR, and measurement of body temperature, heart rate and blood pressure. Any elephants that are "off' are considered EEHV suspects unless proven otherwise.

Thankfully, the landscape of EEHV is changing. In the U.S., there has been a negative stigma associated with EEHV infection, and an institution that lost an animal was a lightening rod for negative publicity. However, more recently, several institutions have shared their EEHV experiences (both positive and negative) publicly and are helping to educate zoo visitors and the general public about this disease and what is needed to fight it. This more open-minded atmosphere allows for information sharing, advancement of research, improved survivability of calves ill from EEHV, and increased fund-raising opportunities.

PERICARDIOCENTESIS: DESCRIPTION OF A NEW TECHNIQUE ICU FOR THE M ANAGEMENT OF EEHV

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A common finding in terminal EEHV cases is severe pericardial effusion leading to cardiac tamponade which can hasten death of certain cases. This paper describes the possible methods of assessing pericardial effusion and management techniques that will support intensive care management of clinical EEHV cases.

LAUNCH OF WEBSITE www.eehvinfo.com

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At the Seventh Annual EEHV workshop hosted by the Houston Zoo, Elephant Managers Association and the International Elephant Foundation it was determined that an EEHV information resource was needed. This resource should meet the needs of all facilities, be a place where people can share experiences in a safe, secure environment, and where information of value to the elephant community would be disseminated quickly and updated frequently. This resource would also be designed to educate and raise awareness.

A Core team of website designers and managers consisting of representatives from each of the critical areas of EEHV research and treatment and husbandry related issues were identified. The group/committee would sign off on everything to be published on the website. The result of this effort is a fully peer reviewed web site dedicated to communicating all aspects of EEHV to both clinicians, animal husbandry staff and any other interested parties.

FROM CHITWAN TO VIENNA – HOW DO GAIT PARAMETERS CHANGE IN A PAIR OF INDIAN RHINOS (*RHINOCEROS UNICORNIS*) COMING FROM SEMI-WILD CONDITIONS TO A EUROPEAN ZOO?

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The Indian rhino (*Rhinoceros unicornis*) is generally regarded as a species with few health problems (Guldenschuh and Von Houwald, 2002). However, one problem is prevalent and has been thoroughly studied by von Houwald (2001), the so-called Chronic foot disease (CFD). CFD is thought to be completely husbandry related. It is characterized by non-healing fissures and ulcers, most prominent between the central toe and the pad, by pad overgrowth, bruising and chronic infection (Von Houwald, 2001). Deviations from a particular gait pattern can be detected through gait analysis. In domestic horses this method serves as a clinical diagnostic tool (Peham, 1999).

The study animals, one female (*2003) and one male (*1997) Indian rhino were wild born in Chitwan National Park, Nepal, and came to the Zoo Vienna, Austria, in March of 2006. Kinetic data were collected during normal gait with a pressure measurement system (Tekscan Walkway 4, Savecomp Megascan GmbH). Spatial resolution was 0.64 mm x 0.64 mm, measuring frequency 39 Hz. Measurements were taken weekly or biweekly between June 2006 and May 2007. Additional sampling aiming at N=10 per foot was performed in fall seasons of 2008-2010, and in spring seasons of 2009-2011.

Twenty-six parameters such as overall and maximum vertical forces, stance duration, foot length and width, peak and maximum contact area, length of the central toe, concavity of the central toe, and Center of Force (COF) were recorded. Changes over time could be observed in the location of the COF and its trajectory, the contact area, and the concavity of the sole of the central toes.

A caudal palmar/plantar shift of the COF (from ~43% to ~51% of the entire hind foot-length in both animals) was probably caused by a bodymass increase during the first year (in the male ~135 kg, in the female ~160 kg). In the following years this trend reversed resulting in a dorsal palmar/plantar shift of the COF (to ~37% of the entire hind foot-length in the male and less than 35% in the female) and a substantial decrease in contact area during stance (from ~325 cm^2 to ~165 cm^2 in the average male hind foot and from ~230 cm^2 to ~130 cm^2 in the average female hind foot). The cause is excessive sole abrasion, especially on the central toes in the hind feet of both animals. Outgrowth of the central toes in the hind feet in both animals could be observed by spring of 2009 by more than 2 cm in the female and ~1 cm in the male. During this time the female was also growing ~12 cm in shoulder height, the male did not. An increased length of the central toe increases the shear forces during breaking over, placing strain on the area of the pad adjacent to the central toe, where CFD usually starts to develop (Von Houwald 2001). First minor cracks of the sole could be detected in winter of 2008/09 in the female (keeper observation). All effects were seen in all four feet but were more severe in the hind feet. This is due to the function hind feet take in during locomotion (Girtler 1987).

Concavity of the sole of the central toe was deemed present when the center of the sole exhibited less than 25% of the force of the rim. Concavity was generally more often visible in the anterior feet. A 'claw-shape' of the central toe – only the anterior rim of the central toe exerting force – was only seen during the first data collection period (2006). Also, concavity of the central toes in the hind feet in both animals could only be detected during the first year.

The trajectory of the COF varied during the first year of data collection in both animals, but due to different causes. In the male animal its poor eyesight (right eye: blind, left eye: visual capabilities an estimated 20%) in combination with the unfamiliar enclosure is a probable cause. In the female the young age might be responsible, since it became similar to the male's trajectory when she reached an age of four years, running from the lateral toe to the pad and the central toe. However, the COF trajectory in the female's right hind foot increasingly differed from the other three feet. To exclude pain as a factor for this change a 3-day therapy with an NSAID (Phenylbutazon) was administered, which acts anti-inflammatory and analgesic. Subsequent data collection did not reveal any difference in COF trajectory. No difference in stance duration or vertical force could be detected.

Data collection in spring of 2011 exhibited an area of increased pressure on the anterior part of the foot pad in the hind right foot of the female animal. Visual control of the sole and pad did not show any signs of CFD. However, since this is the area where CFD usually starts to develop, precautions have to be taken at this time to prevent a future illness.

Apart from the young age, it is still unclear why gait parameters in the female vary more than in the male, especially since CFD is known to occur more often in males (Von Houwald 2001). Both animals share the same habitat, the same keeper routine and nutrition. A behavioural study carried out in 2006 and 2007 (Pertl and Pfistermüller 2011) as well as a chronobiological study carried out in 2008 (Dengg 2009) revealed differences in the animals' activity, but it cannot be excluded that these are individual or due to age. Genetic predisposition, neonate nutrition, hormonal differences as well as overall body structure and conformation might be factors that need to be taken into account.

AFRICAN ELEPHANTS CHANGE GAITS WHEN WALKING DOWNHILL

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Virtually all of the published data indicate that elephants have only one form of locomotion – a lateral sequence gait (with footfalls as follows: left hind-foot → left forefoot → right hind-foot → right forefoot → left hind-foot). Only Hildebrand (1976) has reported that elephants occasionally trot, although several authors have also referred to an "amble" (e.g., Schmitt et al., 2006). Several recent publications have discussed the biomechanics of the lateral sequence gait exhibited by both African and Asian elephants. Hutchinson et al (2003) and Hutchinson et al (2006) indicated that it was unclear whether elephants run: by some measures they do run, according to other measures they don't. Ren & Hutchinson (2008) and Genin et al (2010) demonstrated that Asian elephants are unusual quadrupeds in that their front limbs and

the hind limbs function differently, especially as faster speeds. On the other hand, Ren et al. (2010) provided evidence that, unlike other quadrupeds, elephants use their forelimbs and hindlimbs similarly for propulsion.

Wall et al. (2006) suggested that African elephants tend to avoid steep slopes, perhaps because of the energy cost of moving up and down hills, but all previous studies have examined elephants moving at constant speed on level ground.

Our group has collected information on the gaits exhibited by African elephants moving on level ground, uphill, and downhill. The African elephants we studied at The Indianapolis Zoo rarely walked up or down slopes. However, we were able to collect dozens of samples of the same animals walking uphill, downhill and on level ground while on exhibit. They used the standard lateral sequence gait when walking on level ground and when walking uphill, although the timing of footfalls was different depending on whether the animals were on level ground or walking uphill. The adult elephants frequently changed to a walking trot [left hindlimb and right forelimb move synchronously, right hindlimb and left forelimb move synchronously] when moving downhill, a gait they virtually never used in other circumstances. Juvenile elephants used both the lateral sequence gait and the walking trot on level ground. I will describe how gaits changed as a function of the age of the elephant and the slope of the terrain and suggest why these gait changes occur.

THE USE OF SALIVARY CORTISOL TO ASSESS THE WELFARE OF ELEPHANTS

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All creatures on our planet have a potential of adaption for unpredictable or changed environmental conditions. If the potential of adaption is exhausted, the animal might suffer from stress. A non-invasive assessment of welfare in captive animals can be realized by measuring the salivary cortisol, which indicates stress. Animal rights activists often argue, that circus-elephants suffer from stress under the conditions of the circus. That's why we measured the salivary cortisol of three African circus-elephants in the paddock and during the transport. We did the tests in the paddock always at the same time on four sequently following days to avoid diurnal effects. To measure the salivary cortisol during the transport we took samples before and after the tour from Monte Carlo to Plaschow/Germany. The biochemical analysis of the samples were done by Prof. Sylvia Kaiser of the university of Muenster/Germany. The measured data of the elephants in the paddock was similar to the measured values of elephants in a compound (Menargues et al 2008). There were also no differences between the measured values before and after the transport, which leads us to the conclusion, that the tour didn`t cause stress for the elephants.

PERSONALITY ASSESSMENT AND FACTORS INFLUENCING PERSONALITY IN CAPTIVE ELEPHANTS

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Introduction: Elephants live in a complex society based on matrilineal groups. Management of captive elephants is difficult, partly because each elephant has a unique personality or behavioral traits. Therefore, for a better understanding of elephants' well being in captivity, it would be helpful to systematically assess their personalities. Although personality studies have been conducted in many species, there is little information about individual differences in elephants. Personality can be influenced by many factors such as sex, age and captive settings. Recently the association between personality and gene polymorphism has been reported in some primate species. We aimed to reveal which factors - including sex, age, captivity and genetics - influence personality in captive elephants.

Methods: Subjects for personality assessment were 76 elephants (45 Asian, 30 African savannah and one African forest) kept in 30 zoos. We asked the keepers of subject elephants (n = 95) to answer a questionnaire assessing the elephants' personality traits. The questionnaire consisted of 30 items with adjectives followed by the definition sentence. Five personality factors were extracted by a factor analysis, and individual scores of each factor were calculated. Based on a generalized linear model (GLM), we analyzed which factors (sex, age, the number of elephants kept in the zoo, management procedure) influenced the personality scores of the captive elephants. Additionally we surveyed the polymorphism in genes expressed in the brain using 197 elephant DNA samples. Through genetic analysis, we located the polymorphic regions, and examined the association of elephant genotypes with personality scores as well as other factors.

Results and discussion: Reliability of the five dimensions of elephant personality (Cooperativeness, Dominance, Hyperactivity, Curiosity and Nervousness) was satisfactory. We found that Cooperativeness scores were higher in females and young elephants than those in males and old ones. Hyperactivity and Curiosity scores decreased with age. Nervousness scores were higher in females and elephants managed in free contact than those in males and elephants managed in protected contact. We found polymorphism in three genes; *Androgen receptor* (*AR*), *Fragile X related mental retardation protein interacting protein* (*NUFIP2*) and *Achaete-scute homologs 1* (*ASH1*). The genotype of *ASH1* was related to the Nervousness score, indicating the elephants with short alleles had lower scores than those with long alleles.

These results suggest that the sex differences in personality may reflect differences in lifestyle or the ecological niche of each sex. More detailed analyses about the effects of keeper behavior on elephant personality may help in establishing better relationships between them. This study also showed that *ASH1* is a possible candidate for a personality-related gene. Although we need to increase the sample size to generalize our current findings, the present study provides useful information for the management of captive elephants through a systematic assessment of differences in elephant personalities.

SPONTANEOUS PROBLEM-SOLVING THROUGH TOOL USE IN AN ASIAN ELEPHANT

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Because of their large complex brains, complex social behavior, and facility with tools, elephants are thought to be highly intelligent. Yet, the few cognitive studies conducted with elephants maintain that they perform poorly in spontaneous or "insight" problemsolving tasks. Spontaneous problem-solving without evident trial and error learning has been documented in various animal species. Köhler reported the first evidence for spontaneous problem-solving, calling it "insight," in a non-human species when chimpanzees used objects to reach bananas hung out-of-reach. It has been hypothesized that the elephant's spontaneous problem solving deficit is due to the global cytoarchitecture of the elephant brain. However, in the current research a juvenile male elephant showed spontaneous problem-solving through sudden production of novel adaptive responses to obtain food without evidence of trial and error learning.

Methodology/Principal Findings: We tested three Asian elephants (*Elephas maximus*), two adult females and a juvenile male, at the Smithsonian National Zoological Park, Washington, D.C., USA. Initially, we investigated whether the elephants would use sticks to obtain food out-of-reach on the opposite side of the bars of their stall in two conditions: pulling a tray and knocking the branch of an artificial tree. No elephant attempted to reach the food with the sticks. In an effort to make the experiments more ecologically valid, the experiments were moved outside and food was hung overhead, out-of-reach. Along with the sticks the elephants were given an object that could be moved and stood on to reach the food. Neither of the females attempted to reach the food. However, the 7-year-old male spontaneously moved a large plastic cube, without prior trial and error, and used it to stand on to acquire the food. In further testing he showed behavioral flexibility, using this technique to reach other items within the enclosure, and retrieving the cube from various locations to use as a tool to acquire food. In the absence of the cube, he generalized this tool utilization technique to other objects and, when given smaller objects, the elephant stacked them in an attempt to reach the food.

Conclusions: We have showed that an elephant is capable of spontaneous problemsolving by exhibiting a novel solution without evident trial and error. We posit that previous failures to demonstrate this ability in elephants may not have resulted from a lack of cognitive ability but rather from tasks employing trunk-held sticks as potential tools that prohibited the trunk's use as a sensory organ. Although the trunk is a highly manipulable appendage, in food foraging its sensory function may take precedence. The elephant has an extraordinary sense of smell and the tip of the trunk is as highly enervated as a human fingertip. Holding a stick interferes with these important senses. By adapting our methodology to include an item that can be used as a tool not held in the trunk at the time of food acquisition, we allowed the elephant to show spontaneous problem-solving.

THE SELF-RECOGNITION OF AN AFRICAN ELEPHANT

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It`s generally admitted that apes, whales and dolphins are able to recognize themselves in a mirror. Joshua M. Plotnik, Frans B. M. de Waal and Diana Reiss proved in 2006, that even Asian elephants recognize themselves in mirrors. As far as I know, African elephants were never tested, as they`re not as domesticated as Asian elephants. We tested the elephant cow Mala at an elephant-farm in Platschow/Germany, using a 3mx4m plastic mirror. During the first series of experiments, Mala attacked the mirror and was only held back by her animal-trainer Sonny Francello. In this first series Mala was not able to recognize herself in the mirror. In the second series, three months later, Mala showed all typical stages of behavior and recognized herself in the mirror. To prove, that Mala recognizes herself in the mirror, we performed the marker test; a plastic banana was stuck on the head of the elephant. As Mala didn`t notice the banana for 20 minutes, we were sure, that there was no reason for her to grab for the banana. Then she was confronted with the mirror and immediately picked the banana from her own head instead of grabbing for the banana in the mirror. Her whole behavior shows, that she recognizes the mirror image as herself. The ability to recognize herself reveals that Mala has an idea of her identity, which is supposed to correlate with higher forms of empathy and altruistic behavior.

ESULTS FROM TWO DECADES OF REPRODUCTIVE STEROID MONITORING IN WHITE RHINOCEROSES KEPT IN EUROPEAN ZOOS

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Only 20% of the white rhino EEP population is breeding and as a result the population is not self sustaining. The EEP white rhino Studbook 2009 contains a comprehensive analysis of the situation in Europe. Over the past two decades faecal steroid hormone analysis in female white rhinoceroses was used as a diagnostic tool for investigating reproduction and reproductive problems; these investigations in part were accomplished by rectal ultrasonographic examination. Results of these investigations will be used to analyse and possibly answer the following questions (analysis is currently underway):

- Oestrous cycle length: What is the 'normal' length of the oestrous cycle, 35 or 70 days? Does the oestrous cycle length of 35 or 70 days affect the likelihood of conception? How well do faecal and plasma hormone levels correlate?

- During the past years several young rhino cows have been imported into the EEP. How do ovarian cycles develop in these animals?
- According to studbook analysis the distribution of birth in the EEP indicates some reproductive seasonality; how does this finding correlate to results from faecal hormone analysis?
- Stress, as indicated by faecal glucocorticoid analysis, is often used as an argument to possibly differentiate more or less stressful keeping conditions. With some results on faecal glucocorticoid metabolites we will try to evaluate this debate.
- The recommendation of the EEP is to keep white rhinos in groups. What is the effect of varying group size on reproductive parameters. What is the influence of a white rhino cow on the reproductive endocrinology of its calf, what is the best timing for natal dispersal?
- The recommendation by the EEP is to move and transfer animals out of non-breeding groups into new groups, or at least to separate males for a few months from their group in case there is no breeding. How does a change in group composition (i.e. arrival of a new male, transfer of females to unknown groups) influences reproductive endocrinology?
- It is a proven fact that non-reproducing white rhinoceroses (males and females) develop reproductive health problems. Can these problems be identified from faecal hormone analysis?

ELEPHANT-SIZED HOT FLASHES? THE RELATIONSHIP BETWEEN SOCIAL STATUS AND OVARIAN CYCLE ACTIVITY PROVIDES EVIDENCE FOR MENOPAUSE IN FEMALE AFRICAN ELEPHANTS

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In large, long-lived, highly social animals such as elephants, cessation of reproductive activity before death may be selected for when energy usually devoted to the care and survival of offspring can instead increase inclusive fitness. African elephants live in a matriarchal, fission-fusion society that relies on the knowledge of the matriarch to access resources, coordinate group defence and facilitate reproductive success. Investigating ovarian activity of free-ranging elephants can enhance knowledge about the physiology of ageing and provide insight into evolutionary phenomena such a menopause. Fecal samples were collected opportunistically (June 2007 to September 2009) from 40 known female African elephants in Addo Elephant National Park, South Africa. Resulting fecal progestagen metabolite (FPM) analyses were used to examine the relationships among age, family social status and ovarian activity in adult elephants. FPM profiles suggested that five females (45-60 years of age) were no longer cycling normally and that pregnant elephants (n = 22) had significantly higher FPM concentrations than non-pregnant individuals. Social status and month of gestation were the strongest predictors of

FPM concentrations in pregnant elephants. The number of years since a female gave birth to her last calf was significantly related to age of the elephant; older females had the longest post-partum duration. Social status was the strongest predictor of FPM concentrations in non-pregnant elephants; grand matriarchs had the lowest values. Combined, our results provide hormonal evidence to support the premise that African elephants experience menopause. The important knowledge that matriarchs impart to their kin may have facilitated the evolution of a post-reproductive lifespan in this long-lived, social species.

EMERGENCY NEONATAL CARE OF AN INDIAN RHINOCEROS (*RHINOCEROS UNICORNIS*) CALF

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An Indian Rhinoceros (*Rhinoceros unicornis*) gave birth after a 491 day gestation. The female calf presented in a breech position. The labour, though ultimately successful, was strenuous. Though the dam had previously successfully reared a calf, in this instance no maternal instincts were seen. The calf was weak and remained entrapped within the placental membranes. After four hours it was decided to separate the mother and calf in order to intervene. On clinical examination the calf was almost comatose. On examination her rectal temperature was 32.8° C, her mucous membranes were pale and she was severely dehydrated. However, she still had a weak suckling reflex. Her heart rate was 64 beats per minute (bpm) but all peripheral veins were collapsed. Over a two day period of intensive, round-the-clock nursing care, the calf was stabilized using a combination of parenteral and oral fluids including, stored Indian rhinoceros plasma, colostrum and milk from the dam, and milk substitutes. In between the interventions, the calf and mother were left together to allow bonding.

The presentation will describe in detail all treatments administered to the calf plus the treatment of *retentio secundinarum* in the dam, standing sedation of the dam to allow milking and milk analysis of the dam's milk, all of which contributed to a successful outcome in this challenging case.

FERTILITY AND AGGRESSION CONTROL IN ELEPHANT BULLS THROUGH GNRH VACCINATION

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Recently, trials on non-surgical castration of elephant bulls using a commercially available GnRH vaccine have been undertaken. Preliminary data indicate that the vaccine is successful in suppressing testosterone production. However, in depth studies are necessary to determine dosages and vaccination intervals, long term effects on the reproductive tract, body condition, social behaviour as well as the effectiveness as a contraceptive method. Therefore, the GnRH vaccine was studied in an Asian elephant and is currently investigated in six African elephant bulls using transrectal ultrasonography of the reproductive tract, semen collection and evaluation, titer and testosterone analysis as well as behavioral observations.

The Asian elephant bull was first vaccined in 2008 at the age of 8 years. He received initially monthly injections of 2ml EquiTe ™ (Pfizer) for 5 months and then was boostered every six months. Transrectal ultrasound of the testicles and the accessory sex glands was performed on a regular base. Semen was evaluated (retrieved via rectal prostate massage). Weekly and around vaccination daily serum testosterone concentrations were determined.

The six African elephant bulls currently studied aged between 8 and 23 years. They were vaccinated with 5 ml of Improvac ® (Pfizer) in March 2011 with a booster following 6 weeks later. Fecal samples were collected weekly and daily a week prior to the vaccinations for testosterone determination. At the day of vaccination, the bulls with shoulder heights between 2.1 and 2.9 m were sedated and tethered (standing sedation using butorphanol [50-100mg] and detomidine [20-40 mg] per animal). One bull (22 yrs, shoulder height 2.8 m) received full immobilization [14 mg M99, 10mg Detomidine]. Anesthetics were reversed after 35-45 minutes. Blood samples were taken and transrectal ultrasound was performed. Subsequently, semen was collected via rectal massage or in the case of full immobilization via electro ejaculation. Ejaculates were evaluated microscopically and via Computer Assisted Sperm Analysis (CASA).

In the Asian elephant, testosterone remained suppressed after the second vaccination. Through the regular boosters, this was maintained up to now (5 years later). Testicles as well as accessory sex glands decreased markedly in size. Testicle diameter declined from initially 9.3 cm (prior vaccination) to 6.4 cm within the first two years. Fluid filling within the ampullae as well as the seminal vesicles disappeared. Sperm count and motility in the ejaculate reduced over the years and as of now it is almost free of sperm.

The examination of the African elephant group had just started. Semen parameter in two bulls that were previously treated, indicate that the GnRH vaccine diminishes ejaculate quality (sperm count and morphology).

Preliminary results suggest that the vaccine may have a great value for ex situ application such as for the handling of mature bulls, musth control as well as the introduction of bachelor groups. In situ, this vaccine may be used as an immuno- contraception measure as well as for the control of wild “problem bulls”.

ESTABLISHMENT OF ELEPHANT GAMETE AND EMBRYO BANK OF WILD FOUNDERS FOR CAPTIVE PROPAGATION PROGRAMS

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Along with improved nutrition and enhanced husbandry standards, assisted reproductive technologies (ART) are very important to the zoo community's attempts to improve the self-sustainability of elephant populations in human care. Establishing genome banks of wild founders available e.g. for artificial insemination (AI) and embryo transfer (ET) programs would help to increase genetic variability without relying on the import of wild animals. High quality reproductive samples banked in liquid nitrogen available year round for AI and ET procedures would dramatically increase the success rate of the application of ART in conservation projects. Besides the advantage of such strategy for the international community of zoological institutions it would also specifically benefit range countries. An international platform for gamete and embryo exchange programs would bring several advantages to the countries of origin. It would support (i) national capacity building by training staff in advanced gamete collection techniques in live and dead animals, (ii) support for establishment of national genome resource banks, (iii) economical compensation for labor and local organization (iv) evidence-based management decisions for definite parks based on reproductive assessments in selected individuals, (v) incorporation of game capture and gamete collection procedures into specific tourism programs.

So far there were two field operations performed in the Republic of South Africa. ZooParc de Beauval and its related conservation society sponsored the first initiative by the Leibniz Institute of Zoo and Wildlife Research (IZW) to collect semen from wild African elephants. The aim of the "Project Frozen Dumbo I" was to establish a semen bank in St. Aignon, France at ZooParc de Beauval, that could be used to artificially inseminate African elephants in Europe. In September 2009, collaborators from ZooParc de Beauval, IZW, University of the Western Cape, Pittsburgh Zoo, and the National Zoological Gardens of South Africa with the assistance of Dr. Douw Grobler of CatchCo conducted the semen collection procedures at the Phinda Game Reserve located close to Richard's Bay in Kwazulu-Natal, RSA. In addition, extensive analysis of the semen samples was conducted by the University of the

Western Cape, and tissue and blood samples were collected and banked by the Wildlife Biological Resource Centre / Bio Bank SA, National Zoological Gardens of Republic of South Africa. The prototype project was extremely successful, with over 1,600 ml of semen from seven African elephant bulls successfully frozen for future use.

The aim of "Project Frozen Dumbo II – North America" was to establish a cryo-bank at the Pittsburgh Zoo and its International Conservation Center, where it will be used to establish a semen bank for artificial insemination in North America.

In context with "Project Frozen Dumbo I and II" the international team of 25 specialists from IZW, Pittsburgh, ZooParc de Beauval, University of the Western Cape, and the National Zoological Gardens of South Africa performed in total 31 immobilizations in 20 adult elephant bulls over a time period of 11 days. In total 5,100 ml of high quality (>50%) cryo-preserved elephant semen were banked. This amount of cryo-preserved semen will be sufficient for more than 120 single inseminations. Parallel there are backup semen samples as well as valuable bio-materials of each bull banked at National Bio-Bank of National Zoological Gardens (NZG), RSA.

USING FECAL HORMONAL ANALYSIS TO DETERMINE THE FACTORS AFFECTING THE SUCCESS OF THE BLACK RHINOCEROS IN ADDO ELEPHANT NATIONAL PARK, SOUTH AFRICA

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Non-invasive fecal hormone analysis is an effective method for monitoring and increasing our knowledge of the health and reproductive success of wild populations. For critically endangered and slow breeding species, it is important to understand the factors that limit population growth. Addo Elephant National Park (AENP) has 70% of South Africa's Southwestern subspecies of black rhinoceros (*Diceros bicornis bicornis*). For the past 3 years, we have been validating non-invasive field techniques for monitoring gonadal (progestin and androgen) and adrenocortical (glucocorticoid) activity in the black rhino in two sections of AENP. Camera traps were strategically placed at rhino latrines to identify individuals and facilitate collection of their fecal samples. Fresh fecal samples were collected, processed and analyzed for pregnancy on site and sent to the United States for androgen (FAM) and glucocorticoid (FGM) metabolites analyses. Approximately, 250 samples were collected from known individuals (21♀; 22♂). Results demonstrate that males and females had similar ($P>0.05$) FGM concentrations; however, individuals in the section of AENP with the highest number of elephants and tourists had higher ($P<0.001$) FGM concentrations compared to the section with fewer elephant and tourist. Concentrations of FGM were lower ($P<0.01$) in the winter for both AENP sections. Pregnancy was accurately diagnosed in 100% (n = 7 females) of

females with fecal progestin concentrations seven-fold higher ($P < 0.001$) in pregnant compared to non-pregnant females. For males, FAM was higher ($P=0.006$) in summer and spring months and lowest in the winter. A similar pattern for progestin was observed in the females ($P=0.012$) with lowest fecal progestin concentrations in the winter. With validated non-invasive monitoring methods, our long-term goal is to establish a multi-year, health monitoring program to investigate the relationship among black rhino hormonal activity, parasitic infection rates and varying abundance of mega-herbivores, predators and humans. Results will be used to facilitate SANParks' management decisions for black rhino conservation.

SPERM SORTING AND PRESERVATION TECHNOLOGIES FOR SEX RATIO MODIFICATION IN THE ELEPHANT AND RHINOCEROS: AN UPDATE

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Management of sustainable *ex situ* populations of elephants and rhinoceroses would be enhanced by the ability to modify the sex ratio of offspring. A bias of the sex ratio towards females, through the insemination of cows with semen samples enriched for X chromosome-bearing spermatozoa, would improve the population's reproductive potential, reduce the number of males, and thereby facilitate the housing of socially cohesive groups. Sperm sexing technology could also address undesirable sex ratio skews and when combined with sperm preservation methodologies and effective genome storage banks, would aid in the cooperative management and long-term conservation of these threatened species. Sperm sorting and preservation research, which has been endorsed by the AZA TAG/SSP Programs of both taxa, has been conducted by our group and collaborators over the past six years. The goal is to adapt flow cytometry technology to sort elephant and rhinoceros spermatozoa and produce fresh and frozen-thawed X-enriched samples of adequate quality for artificial insemination (AI). Standard methods of semen collection in the elephant (manual stimulation of standing males) and rhinoceros (electroejaculation [EEJ] of anaesthetized males) both produce samples exhibiting great variation in quality. For the Asian (*Elephas maximus*) and African (*Loxodonta africana*) elephant, results demonstrated that initial ejaculate quality must meet specific *in vitro* criteria to

allow the subsequent production of high quality sorted spermatozoa. Initial ejaculate criteria necessary for overnight shipment to the sorting facility were determined ("viable samples" >70% progressive motility and plasma membrane integrity, < 15% detached heads, < 0.013 mg creatinine/ml). When viable samples were utilized, *in vitro* quality of sex-sorted, chilled Asian and African elephant spermatozoa was well maintained over a 24 h period, and samples were considered adequate for use in AI trials if inseminated within 24 h after sorting. Despite these promising results, the proportion of ejaculates designated as viable were low for both elephant species. Only 16% (19/118) of manual stimulation collection attempts yielded viable samples from 10 males in the North American and Australasian populations. For the three rhinoceros species examined (white: *Ceratotherium simum*, black: *Diceros bicornis*, Indian: *Rhinoceros unicornis*), the most progress has been made with the former species. White rhinoceros spermatozoa derived from viable electroejaculates displayed *in vitro* characteristics that were considered suitable for use in AI, after sorting and chilled storage for 24 h, and after cryopreservation and thawing using directional freezing technology. Across all three rhinoceros species, the proportion of samples of adequate quality for sorting and preservation after EEJ of 11 males in the North American population was moderate (40%, 8/20), but final numbers of spermatozoa from viable samples available for sorting was highly variable (2059 ± 1932 x 10^6 spermatozoa/male). In summary, methods for sex-sorting and chilled storage of spermatozoa (prior to and after sorting) have been established for both elephant species. Sex-sorting, chilled storage and cryopreservation methods have also been developed for the white rhinoceros. Inconsistent ejaculate quality is the greatest challenge to producing the substantial numbers of sex-sorted spermatozoa necessary for an insemination dose in all of the aforementioned species.

OVULATION INDUCTION FOR TIMED ARTIFICIAL INSEMINATION IN THE SUMATRAN RHINOCEROS (*DICERORHINUS SUMATRENSIS*)

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The Sumatran rhinoceros is critically endangered with no more than 200 surviving in the wild and just 9 animals in captivity. The captive breeding program was initiated in the 1980s as part of a two-pronged approach consisting of ex situ protection and in situ propagation to ensure the species' survival. To-date, only three calves have been produced from one pair of rhinos at the Cincinnati Zoo. Therefore, four of the nine captive rhinos are closely related. To avoid inbreeding and the international transport of rhinos, the development of artificial insemination for this species has become a priority. Because the Sumatran rhino is an induced ovulator, a proven ovulation induction protocol is essential for successful artificial insemination in this species. The goal of this study was to evaluate the ability of exogenous hormone administration to mimic the physiological responses previously documented in a naturally mating female Sumatran rhino that successfully reproduced. Thrice weekly rectal ultrasound exams were conducted to monitor a young, sexually mature rhino's ovarian activity. When a preovulatory follicle measuring 21 x 22 mm in diameter was observed (Day 0), the rhino was administered

500 µg gonadotropin releasing hormone (GnRH; Cystorelin; i.m.). After treatment, ultrasound exams were conducted every 12 h until ovulation was confirmed by follicular collapse, and then thrice weekly to monitor ovarian activity throughout the luteal phase and subsequent estrus. Urine samples were collected daily prior to GnRH administration, as often as possible during the 36 h period following GnRH, and then every other day through Day 12. Blood samples were collected 41 min and 10 days after GnRH treatment. The follicle was still present 36 h post-GnRH but had ovulated by 48 h post-GnRH (Day 2). Following ovulation, both ovaries remained relatively inactive until Day 22 when a 19 mm pre-ovulatory follicle was observed. No GnRH was administered and the follicle grew to 30 mm without ovulating. Serum LH had increased to 9.97 ng/ml 41 min following GnRH treatment confirming that the pituitary had responded. Baseline serum LH concentrations in this female on Day -11 and Day 10 of the study were 1.97 and 1.37 ng/ml, respectively. Urinary LH concentrations revealed more detail about the pituitary's response to GnRH administration. From an initial baseline value of 962 pg/ng Cr, urinary LH peaked at 2,288 pg/ng Cr 7 h post-GnRH and then dropped rapidly below baseline (161 pg/ng Cr) 11 h post-GnRH. On Day 1, LH had recovered to typical baseline concentrations. Serum progesterone concentrations on Day 0 and Day 10 were 0.32 and 1.25 ng/ml, respectively, confirming the formation of an active corpus luteum. In conclusion, changes in LH, progesterone and ovarian activity following specifically timed GnRH administration mimic those observed following natural mating in the Sumatran rhino. Therefore, this ovulation induction regimen should help pave the way for successful artificial insemination in this species.

THE WHITE RHINO EEP BREEDING PROGRAMME; ANALYSIS THROUGHOUT THE YEARS

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The population of white rhino within the EEP programme has steadily grown to 250 animals at the end of 2010. But is has not been a sustainable population. If we look at the number of births and deaths throughout the years it is apparent that the number of years of negative growth with the number of deaths increasing the number of births exceeds the years of positive growth. Only because of frequent imports from South Africa has the population grown to the current number. However, many institutions have invested in new, larger enclosures and more naturalistic enclosures and group compositions. Already the number of births has increased enormously the last years and only because a large number of old animals (from the original imports from the 1970s) are dying there is not yet a positive growth rate but the anticipation is that this will change in the next years. At this time 20% of the population is breeding, and of the 80% (n=201)which is not (yet) breeding 45% is younger than 10 years and 16% between 10 and 20 years of age. This means that there is a huge potential and much effort is put into getting these animals to breed. Efforts consist of complete reproductive checkups by veterinary experts, hormonal investigations and exchange of non breeding animals to provide them with new surroundings and social group composition. Another initiative is the exchange of young females out of their maternal herd to imitate the natural dispersal of these individuals. A total of 68

institutions participate in the White rhino EEP. Of these 50 have breeding potential, 6 hold a single sex group, 6 hold one last animal and 6 several older animals. Of the 50 zoos with breeding potential, 24 are already breeding with a large majority of their animals, 20 have relatively newly established groups and 6 institutions face breeding difficulties which are addressed.

OESTROUS SYNCHRONIZATION IN WHITE RHINOCEROS

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Ovulation in the white rhinoceros, like in most mammals, is induced by LH surge that occurs at the end of the follicular phase. LH induces the rupture of the Graafian follicle which is followed by the formation of a corpus luteum as the primary source of progesterone synthesis. The length of the oestrous cycle, predominantly measured by pregnane concentrations in the faeces, is 35d or - in extended cycles - 70d. Yet, many of the adult captive white rhinoceros display long periods of anoestrous or erratic cycles despite regular formation of a Graafian follicle. Frequent luteinisation and formation of haemorrhagic follicles instead of follicle rupture and CL formation characterize this commonly occurring anoestrous. Anoestrous in the white rhinoceros is considered as a primary cause for the low reproductive rate in captivity. Transport of females to other facilities or the introduction of new males has shown to initiate regular oestrous cycle activity in some animals. This study attempted to employ an inexpensive and easy to apply hormone treatment to overcome anoestrous periods and to induce oestrous. Thirty-three oestrous synchronizations were attempted in 22 animals. Oral synthetic progesterone was given in combination with either hcG or GnRH analogue to induce ovulation at the end of the oestrous induction protocol. Ovulation was induced in 91% of the treatments. The induced oestrous cycle was in 30 % of the treatments 35d and in 70% 70d long. Forty-one percent of females showed a regular oestrous cycle after the initial oestrous synchronization. In conclusion, oestrous synchronization reliably induced oestrus and ovulation in previously anoestrous white rhinoceros females. In a considerable proportion of treated animals the oestrous synchronization showed a lasting effect as they exhibited a regular oestrous cycle thereafter.

ABSTRACT BODY CONDITION SCORING OF CAPTIVE WHITE RHINOCEROS (*CERATOHERIUM SIMUM*)

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The Body Condition Scoring (BCS) system has been developed by the domestic hoofstock industry as a tool to optimise production, evaluate health and assess nutritional status. The advantages of BCS are that it can be done none invasive, low cost and is not hugely time consuming. Scoring systems are based on body shape and appearance of skeletal features. Body Condition Scoring has been developed for several species in zoos and has been found useful in judging the adequacy of energy supplies. Body Condition Scoring has originally been developed by the domestic hoofstock industry and which requires handling of the animals to assess the relevant body parts. The close handling of animals for body condition scoring is not always possible for wild animals in zoos and therefore body condition scoring of wild animals in zoos is based on visual scoring. A detailed descriptive work on white rhino body condition scoring is not currently available. The goals of this research were to compare the body condition of captive white rhino within European collections and to develop a body condition scoring system that can be used for captive white rhino. A request for pictures has been sent out to all collections within the European breeding programme. 20 Collections responded to the request for pictures, which resulted in pictures for 65 (17.45.3) rhinos. There are 5 scores used within the body condition system: 1=very poor, 2=poor, 3=fair, 4=good and 5=fat. There were no rhinos found that classified within BCS 1. There was 1 animal in BCS 2, 15 animals in BCS 3, 30 animals in BCS 4 and 19 animals in BCS 5. This study was done to assess the BCS of captive white rhino within Europe and to develop a scoring system that can be used for this species.

LASER ASSISTED PHOTOGRAMMETRIC TECHNIQUE TO MEASURE SHOULDER HEIGHT AND BODY LENGTH OF ELEPHANTS

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Elephant weight is been always estimated from linear measurements such as shoulder height, body length, forefoot circumference, chest girth and hind foot length. Here we introduce laser assisted photogrammetric technique (LAPT) to obtain shoulder height and body length directly which could be used to estimate body weight of the wild and captive elephants.

COMPARISON OF SERUM ELECTROLYTES AND MINERALS BETWEEN WILD, CAPTIVE CONCENTARET FED, AND CAPTIVE FORAGE ONLY FED WHITE RHINOCEROS (*CERATHOTHERIUM SIMUM SIMUM)*

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Serum was collected voluntarily from captive Southern white rhino in this study. The first study group consisted of Southern white rhino that are on a strict hay only diet (n=11) from Institution A. The second group, is hay and concentrate fed zoo rhino (n=31from 4 other facilities. All captive rhinos are housed in Florida. The third group consisted of Southern white rhino (n=38) from South African national parks. The wild white rhino are involved in a managed care system, but they are allowed to feed naturally with no supplements. Values for mineral and electrolytes were taken from the literature. Samples were collected after the rhinos were immobilized for other management procedures.

Blood samples from both zoo rhino groups and the wild rhino group were analyzed for mineral; calcium, chloride, cobalt, copper, iron, manganese, magnesium, molybdenum, phosphorus, potassium, selenium, sodium, zinc, and vitamin A and E content. All data from the wild rhino were compiled and summarized from four scientific articles; M.E. Keeper, 1976, van Heerden, 1985 and 1994, Clauss, 2002, and Dierenfeld, 2005, specifically focused on wild Southern white rhino blood parameters. It is noted by the author that all of the wild white rhino in their studies were presumed healthy at the time of blood draws based on blood analysis collected at the time of their study.

The averages for the minerals & vitamins for non-concentrate, concentrate, & wild rhinos.are summarized in the table below. Non-concentrate n=11, concentrate n=31, wild n=17.

Feed	Ca (mg/dL)	Cl (mEq/L)	Cu (ug/mL)	Fe (ug/dL)	Mn (ng/mL)	Mg (mg/dL)	Mo (ng/mL)
Non-concentrate	11.02	93.46	1.38	1.50	6.00	2.35	46.24
Concentrate	11.90	94.79	1.71	2.12	2.95	2.17	31.25
Wild	10.78	94.20	1.62	0.79	2.47	2.06	28.34
	K (mEq/L)	**Se (ng/mL)**	**Na (mEq/L)**	**TP (gm/dL)**	**Vit A (ug/mL)**	**Vit E (ug/mL)**	**Zinc (ug/mL)**
Non-concentrate	4.29	57.80	137.61	8.56	31.19	0.42	2.58
Concentrate	4.45	138.00	139.86	7.97	N/A	2.37	0.93
Wild	5.42	200.80	132.34	9.27	60.00	0.77	1.67

It appears that the means for calcium, iron, potassium selenium, total protein, and zinc for both concentrate and non-concentrate groups of white rhinos varies from wild rhinos. Sodium in concentrate fed rhinos is higher than wild rhinos, copper and magnesium in non-concentrate fed rhinos differs from wild rhinos, while manganese and molybdenum appears equal in all groups.

BLACK RHINO POPULATION PERFORMANCE IN RELATION TO PLANT AVAILABLE NUTRIENT AND PLANT AVAILABLE MOISTURE

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Habitats of low Plant Available Nutrient (PAN) and low Plant Available Moisture (PAM) are expected to sustain low food supply and consequent difficulty for large herbivores to convert the available low nutrients to meet their energy and reproductive requirements. Decreased fecundity and low large herbivore density have been reported to be associated with such situations. Low food, nutrient and moisture may lead to chronic stress and aggravate animal's susceptibility to diseases, immune-suppression, tissue atrophy, and suppress population performance for example by leading to an increase in ages at first calving, increased inter-calving interval, decreased proportion of breeding-age females per year and poor body condition. These relationships are less understood for hind-gut fermenter like the black rhino, also known to feed on over 220 species of plants.

The study attempts to understand the relationships between PAN / PAM and black rhino population performance by comparing and contrasting animal body conditions, ages at first calving, inter-calving intervals and proportions of breeding-age females calving per year in nine black rhino populations of low and high densities in Kenya.

This is a component of a wider PhD research at Wageningen University that investigates the inter-relationships between faecal cortisol, testosterone and oestrogen concentrations on one hand; and population performance indicators of black rhinos at different densities *in situ* on the other hand. The wider study also measures nitrogen, phosphorous and potassium in faeces and common plants to compare between populations in contrasting PAN and PAM. Data for this study are drawn from individually known male and female black rhinos of breeding age from nine well monitored black rhino populations.

PRELIMINARY EPIDEMIOLOGICAL FINDINGS USING SEROLOGY IN AN OUTBREAK OF *MYCOBACTERIUM TUBERCULOSIS* IN MANAGED ASIAN ELEPHANTS (*ELEPHAS MAXIMUS*) AT A SINGLE FACILITY

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Mycobacteria tuberculosis was diagnosed in a 31 yr old female Asian elephant (*Elephas maximus*) at a zoological facility in Florida with a positive culture from a discharge from the trunk in May 2000. Retrospective analysis of serum using the Elephant Stat-Pak™ and multiple antigen immunoassay (MAPIA) for *Mycobacteria tuberculosis* from Chembio™ Inc and

mycobacterial multi-epitope analysis suggested that this elephant had an active infection in March of 1996 with negative results in November 1995. Serum was available on 5 conspecifics at the time of the earliest detection of mycobacterial disease (March 1996) and historically a few months prior. Routine serum chemistry analysis was performed simultaneously on samples frozen at -80C to look for any differences around the time of the re-activation a possible latent infection. Vit D (25-OH) levels around this time period were evaluated.

The most significant trend noted may be that Vitamin D (25-OH) was low and undetectable in some samples on the elephant with tuberculosis. Calculating glomerular filtration rates (GFR) using formulas adapted for marine mammals suggest various degrees of renal insufficiency existed in this herd. Vitamin D deficiency and its role in infectious disease in managed elephants along with possible reason for this possible deficiency is explored. Other environmental factors that may lead to altered immune function including monocytosis, indoor air quality, and hormone status that may parallel disease in humans is also explored.

TUBERCULOSIS IN ELEPHANTS: THE IFNGAMMA ASSAY ADDED TO THE DIAGNOSTIC SPECTRUM

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Mycobacterium tuberculosis is the causative agent of tuberculosis, a chronic respiratory disease in humans, that may be transmitted from infected humans to other species, including elephant, in case of prolonged intensive contact.

M. tuberculosis infection has been diagnosed worldwide in captive elephants, including domesticated elephants in Thailand (Angkawanish et al, 2010). It has an impact on animal welfare and constitutes a risk for human health, hence for the tourist industry and may have economical consequences. Diagnostic procedures available at present are bacterial culture from trunk washes and from affected tissue at necropsy, serological assays like Elephant TB Stat-Pak, MAPIA and DPP VetTB assays (Greenwald et al, 2009) as well as several non-commercial ELISA's employing Mycobacterium Tuberculosis Complex (MTBC) specific antigens. However, assays to assess cell mediated immunity (CMI), that is considered to be elicited early after infection, are not available as yet. The skin test, the CMI assay of choice in humans and

cattle, is not applicable in pachyderms. Like in these species the interferon gamma (IFN?) assay is considered a good alternative for the skin test in elephants. In the present paper we report about our efforts to develop an elephant specific IFN? assay applicable for both Asian and African elephants.

Initially we produced *E. coli* derived recombinant elephant IFN? and immunization of mice and chicken with this bacterial protein gave rise to the monoclonal antibody 5.1 and polyclonal chicken IgY that could be used in IFN? capture assays successfully. More recently, immunization of mice with recombinant Asian elephant IFN? produced by the mammalian cell line HEK293 gave rise to a number of new monoclonal antibodies with high affinity for, mitogen stimulated blood derived, IFN? of both Asian and African elephants that can be used for further improvement of the assay. Detection within concentration ranges to be elicited by MTBC specific antigens is now within reach.

UPDATE FROM THE STAKEHOLDERS TASK FORCE FOR THE MANAGEMENT AND RESEARCH PRIORITIES OF TUBERCULOSIS FOR ELEPHANTS IN HUMAN CARE

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In a spirit of cooperation on August 1-2, 2011, Thirty-three participants – including veterinarians, elephant care experts, human Tb professionals, and public health experts met in Fort Worth, Texas to participate in a workshop entitled **Management and Research Priorities of Tuberculosis for Elephants in Human Care - Stakeholders Task Force.** The American Association of Zoo Veterinarians, Association of Zoos and Aquariums Elephant Taxon Advisory Group, Elephant Managers Association, Fort Worth Zoo, International Elephant Foundation, and the Ringling Bros. Center for Elephant Conservation organized and facilitated the workshop to share current science and research information about tuberculosis in elephants and through facilitated and breakout sessions identified research and management priorities and action plans to further understand the identification, management and treatment of tuberculosis in elephants. The participants discussed concerns, clarified areas of disagreement and identified partnerships to advance our knowledge of tuberculosis in elephants.

There is a critical need for stakeholders to build relationships with and to integrate the efforts of USDA-APHIS Animal Care, USAHA, researchers, veterinarians, and animal managers to ensure the development of best practices for managing TB in elephants. Through this workshop format, participants discussed the current available knowledge, focused on sharing information, established potential areas of collaboration and common goals. They identified actionable items to improve detection, diagnosis, treatment, and ultimately to reduce the impact of this disease so that we may ensure a future for captive and wild elephants.

Key goals include 1) using science-based decision-making to develop practical, feasible and effective guidelines for the management and control of MTb in elephants; 2) defining MTb

exposure; 3) determining the significance of a reactive Stat Pak and MAPIA test result in terms of risk, and how those definitions can best be reflected in the development of new guidelines that are practical, realistic, and effective in controlling MTb in elephants; 4) formulating travel and risk recommendations; and 5) developing short-term and long-range goals in the areas of research, disease management, herd monitoring, herd management, public health, funding, and public relations.

Collaboration, cooperation and communication is essential across the spectrum of elephant care to ensure the well-being of Asian elephants globally. We appreciate the expertise of all involved and the level of commitment we all have for Asian elephants.

HEMOSIDEROSIS AND CLINICAL FINDINGS CONSISTENT WITH BLACK RHINO SYNDROMES IN GREATER ONE HORNED RHINOCEROS (*RHINOCEROS UNICORNIS*)

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A 15 yr-old male Greater one horned rhinoceros (*Rhinoceros unicornis*) was evaluated for chronic weight loss of over 2 years. The diet consistent of over 15kgs of various grain and concentrates offered twice daily for several years. Initial diagnostic work-up was conducted with a combination of butorphanol and detomidine in a chute for blood collection, rectal exam with ultrasound, and upper GI endoscopy. Finding included anemia, suspected renal fibrosis, and Gi inflammation. Serum ferritin from Kansas State University was normal as determined by in house levels established by the laboratory.

The rhinoceros declined in condition and was euthanized several weeks after the exam. Gross findings include severe muscle wasting, fat atrophy, bronzing of the gastro-intestinal viscera, and a firm swollen liver. A complete set of tissues was not available for histopathology but there fibrosis and specialized staining confirmed the presence of hemosiderosis in the liver.

A 10 yr-old female Greater one horned rhinoceros (*Rhinoceros unicornis*) at another facility had a history of recurring ulcerative, bullae cutaneous lesions. Touch prep of these lesions revealed a tendency towards eosinophil accumulation. Empiric therapy with **Diphenhydramine Hydrochloride** (Benadryl™) resulted in some relief of the lesions but only rarely complete resolution and recurrence was common after therapy ceased.

The clinical presentation and findings in both cases closely resemble syndrome seen in captive black rhinos. These findings have not been reported to the best of our knowledge. The possibility of an gastrointestinal hypersensitivity or antigenic source of inflammation is suspected in black rhinos and in these Greater one horned rhinos based on the consistency of eosinophils and the temporary improvement with anti-histamines. The serum assay for ferritin also does not appear to accurately reflect iron stores in Greater one horned rhinos.

Clinical syndromes similar to those seen in black rhinos, anemia, skin disease, chronic wasting are possible in GOH and may actually be under appreciated in captive settings. A nutritional etiology is suspected as is in captive black rhinos.

ROMIFIDINE – KETAMINE AND ITS REVERSAL WITH ATIPAMEZOLE IN ELEPHANT PAEDIATRICS

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Five out of eight elephant calves weighing 600-1000 kg were selected for romifidine-ketamine anaesthesia. Romifidine @ 10µg/kg and ketamine 100 mg/ elephant calf were drawn into a syringe and mixed them thoroughly by shaking and injected intramuscularly to the elephant calves selected for this purpose. The elephant calves were induced at 10,4± 1.20 minutes and the movement were standstill thereafter. The special sign observed during anaesthesia were loud snoring sound, widening of legs, protrusion of penis, relaxed trunk touching ground, dropping of head and ear, closing of eyelids. Ataxia developed at 30± 1.25 and four calves fell down onto the ground at 45±2.15 minutes of anaesthesia. One of them was in sternal recumbency and the rest were in lateral recumbency. Salivation was almost nil. Extremities like ear and tail were not felt cold. The pulse rate reduced from 40.8±1.78 to 36.4±1.12 at 45 minutes of anaesthesa. The respiration continued to reduce from 13.4±0.4 to 11.4±0.03 upto 60 minutes. Slight reduction of body temperature from 96.9±0.52 to 96.6±0.56 was recorded. The elephant calves were under anaesthesia for a duration of 81±9.15 minutes and they recovered at 100±6.14 minutes. However the calves after recovery remained drowsy for several hours. The calves exhibited total analgesia and surgical procedures like wound dressing, injection of drugs into the eye for treating eye disease could be performed satisfactorily

The remaining three elephant calves were also injected with the same dose of anaesthetics allowed them to reach peak of anaesthesia at 48±7.58 minutes. After attaining lateral recumbency, Atipamezole @ 10µg/kg (same dose of Romifidine) was injected into the ear vein. The calves responded immediately and reversed at 1.92± 0.30 minutes. Following reversal each of the elephant calf urinated profusely. The calves walked and started taking grass from the surroundings.

CHRONIC PERFORATING FOOT SOLE LESIONS IN 2 CAPTIVE GREAT INDIAN RHINOCEROSES (*RHINOCEROS UNICORNIS*).

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This report describes the ongoing care of chronic foot problems in a couple of adult captive great Indian rhinoceros. The female developed a bilateral lesion in the hind feet at the age of 6 years, 3 years after her arrival at the zoo. A deep, perforative lesion of the sole cushion was seen approximately 1 cm caudal to the junction of the nail with the sole of D3 (bilateral, right foot worse than left foot). Similar lesions were seen in the male around the same time.

It was hypothesized that the lesions were the result of pressure necrosis: when turning around in the indoor enclosure, the centre point of the turning circle was exactly the site of the lesions. Occasionally the animals showed moderate supporting lameness, which was mostly caused by pressure necrosis and abscess formation due to contact of the lateral nail to the moderately swollen soft tissue of the 3rd digit. The sole around the lesions was thickened. Infected granulating tissue was found deep inside the lesions.

Several attempts to treat these foot problem were made. All of them were based on widening the opening of the lesion to reduce pressure on the deeper lying process and to facilitate drainage of detritus. This presentation demonstrates the different treatment protocols that have been used in these 2 animals, including the use of alginates, supporting blocks attached to the nails, a home-made "rhino-horseshoe", casts and pedicure methods.

To enable treatment, the male needed to be anesthetized 8 times in a period of 9 years before he was transferred to another zoo. The female was anesthetized 26 times in a period of 12 years. In all cases, Immobilon L.A. (etorphine and acetylpromazin) was used under permanent intranasal oxygen insufflation. Since 2010 preventive pedicure without anaesthesia has been performed routinely 1-3 times per week in the female.

Similar sole lesions have been frequently reported anecdotally and written by several veterinarians (Rietschel, 2000, Howald et al. 2001). One published survey indicates that 22.5% of the captive population of great Indian rhinoceroses have lesions at the foot/nail junction (Houwald et al., 1998). Once the sole has a larger perforating lesion, complete cure seems to be an illusion. Training of the affected animal to allow routine pedicure seems to be the best option for treatment.

THE MOBILE ELEPHANT CLINIC, A TOOL FOR DISEASE MONITORING IN THAILAND

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Since 1999 the National Elephant Institute runs a Mobile Elephant Clinic (MEC) to monitor and care for elephant health and welfare. The MEC based in Lampang serves 3,299 elephants that represent almost 100% of the domesticated elephant population in Thailand. Each area of the country is visited twice a year for routine surveys. In addition, the MEC will act in case of emergency, such as gastro-intestinal obstruction, dystocia, severe injury or madness throughout the country. Findings recorded in course of time are believed to be representative for health and welfare conditions among the working elephant population in Thailand, that is mainly threatened by acts of man. Forty percent of the population is injured, in poor condition, has gastro-intestinal problems and suffers from infectious diseases. Tuberculosis, EEHV, FMD and trypanosomiasis are some of the diseases reported as a result of the activities of the MEC around the country.

In conclusion, due to the activities of the MEC concerning a substantial number of domesticated elephants the health and welfare situation in elephant management in Thailand is

well documented. This will help us to solve the present problems and to plan future elephant healthcare and management strategies.

INVESTIGATING THE HEMOGLOBIN OF WHITE RHINOCEROS (*CERATHOTHERIUM SIMUM SIMUM)* AND POSSIBLE IMPLICATIONS FOR ANESTHESIA.

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Current research for white rhinoceros (Ceratotherium simum) anesthetization details protocol for the use of potent anesthetizing agents at varying concentrations. These protocols reportedly cause physiological changes including metabolic acidosis, tachycardia, hypoxemia and capture myopathy. However the oxygen-hemoglobin dissociation curve (ODC) has not been determined, resulting in an information deficit regarding the true physiological effects of anesthetizing agents on blood oxygen saturation. This study was designed to provide valuable information on the white rhinoceros' sensitivity towards anesthetizing agents and to help establish a safety protocol for anesthetized respiration. Although pulse oximetry and blood gas analyses are often used to monitor rhinos under anesthesia, these instruments are calibrated using the human ODC and may result in falsely low readings. To establish a correction factor for measurements from a pulse oximeter blood samples were taken during routine examinations of operantly conditioned white rhinos (n=7) and opportunistically from anesthetized white rhinos (n=2). A pulse oximeter was used to monitor oxygen saturation while blood samples were taken. Measurements from the pulse oximeter were recorded and compared to measurements from an arterial blood gas analysis. Although a steady pulse waveform was achieved, preliminary data has shown significant differences between pulse oximeter and arterial blood gas measurements in standing and anesthetized rhinos. The accuracy of the pulse oximeter was also significantly closer for the yearling rhino when compared to the adult rhinos. The ODC for the white rhinoceros is currently being established using time-resolved optical spectroscopy. It is expected that the ODC will show a higher oxygen affinity of hemoglobin when compared to that of human blood. In addition, current results trend towards an ODC resembling human blood for the yearling rhino probably due to differences in the genetic mechanisms regulating hemoglobin production.

PREVALENCE OF *COBBOLDIA ELEPHANTIS* IN FREE RANGING ELEPHANTS IN SRI LANKA

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The free ranging elephant (*Elephas maximus*) population in Sri Lanka is estimated around 5,000 and it represents 10 % of the endangered Asian elephants. Gastro-intestinal diseases are the major cause leading to mortality and morbidity of elephant calves and adults in Sri Lanka. Among the parasitic diseases of Sri Lankan elephant (*Elephas maximus maximus*) *Cobboldia elephantis* larval (gastric myiasis) infestation is a common parasitic condition and which is not broadly studied. *C. elephantis* is a species of sub family gasterophilinae of family oestridae. It is host specific and obligate parasite. It parasitizes the Asian elephant and is closely related to *Platycobboldia loxodontis* and *Rodhainomyia roverei*, which develop in the African elephant' All of oestridae have reduced mouthparts in the adult stage and cannot feed. Therefore, all of their nutrients for adult activities are ingested in the larval stage and cannot be replenished once the adults emerge. Larvae of *cobboldia* move freely in the stomach and leave the host via its mouthparts.

We have conducted survey to determine prevalence of *Cobboldia elephantis* among free ranging Asian elephants in Sri Lanka in 2007-2011. We conducted 39 post-mortems of elephants those died due to gunshot wounds (n=32), electrocution (n=6), old age (n=2), unknown (01). The post-mortems were conducted within 24 hours of death and gastro intestinal tract was examined to find larvae. The larvae were collected for classification of larval stages and pupate to get adult flies.

The larvae were presented in stomach of 19 elephants and the numbers of larvae range from 12 to 158 Eight instances mature larve/ 3rd instars were kept alive in soil filled jars and four times adult flies emerged after pupating. Currently, examining and drawing of mouth parts and other morphological features of different larval stages and adult flies were going on parallel with the studies on complete life cycle and pathological importance of *C. elephantis*.

ASIAN ELEPHANT SUPPORT

Linda Reifschneider

Asian Elephant Support
www.asianelephantsupport.org

The intent of this presentation is to introduce the International Elephant Foundation membership to Asian Elephant Support (AES). Asian Elephant Support is a U.S. non-profit foundation dedicated to the care and conservation of Asian elephants in their range countries.

Our mission is to:

- Provide financial support for Asian elephant projects in range countries that meet our criteria for care of captive elephants and for conservation of the species.
- Increase awareness of current and future needs of the Asian elephant.
- Increase awareness of humane treatment of elephants living in captivity.
- Provide educational opportunities to those persons who care for captive Asian elephants in range countries.

We would like to share how AES was established, our mission, the projects we support (i.e. CRU units in Sumatra, medical field research in southern India, education funding for those working with elephants, collaborating with other NGOs on efforts we consider worthwhile), our education program, and future plans.

For more information and a list of the current Directors please visit our website at: www.asianelephantsupport.org