

Risks of disease transmission between wildlife and livestock in the Chang Tang Nature Reserve: Suggestions for baseline data collection in wild ungulates

Background: the Chang Tang Nature Reserve

Covering around 300,000 km² in the north-western part of the Tibetan Autonomous Region of the People's Republic of China, the Chang Tang Nature Reserve (CTNR) is one of the largest protected areas in the world. It was established in 1993 to protect an ecosystem unique in the world. The Reserve especially upholds an exceptional community of large mammals, including six endangered ungulate species: the Tibetan antelope (*Pantholops hodgsoni*), the Tibetan wild ass or kiang (*Equus kiang*), the Tibetan gazelle (*Procapra picticaudata*), the wild yak (*Bos grunniens*), the blue sheep (*Pseudois nayaur*) and the Tibetan argali (*Ovis ammon hodgsoni*). Although remote and difficult to access, the Reserve still faces many threats, which endanger wild species and the rangeland ecosystem that supports them and the local pastoralists. Because of the high elevation of the Reserve (most of it lies between 4000 and 5000 m), the ecosystem there is fragile and difficult to restore once damaged. Overhunting and changes in traditional pastoral production systems especially threaten wild ungulates. For example, increased livestock numbers, sedentarization of pastoralists, and rangeland fencing, pose problems of overgrazing and increased competition for forage between non-domestic and domestic ungulates. Habitat loss may also result in changes of migratory patterns of wild species and affect their survival through the introduction by livestock of infectious agents.

Raising livestock is a major economical resource for local people but wildlife would be a major attraction for any ecotourism project. As a matter of fact preserving a healthy ecosystem today may also contribute to secure an economical resource for the region in the future. One of the main challenges of decision makers is therefore to integrate the needs of local development with the conservation of Tibet's unique wildlife and ecosystem. The project on Biodiversity Conservation and Sustainable Natural Resource Use in the Chang Tang Region of Tibet aims at fulfilling this challenge by developing coordinated planning and improving enforcement of plans and policies through cooperation with and between government departments. In particular the project should facilitate the development of a comprehensive landscape-level conservation management plan for the region. The Wildlife Conservation Society (WCS) will play the role of a cooperating agency, will build capacity and help strategize the different components of the conservation plan.

Until the 1990's, the CTNR's wildlife and ecosystem had been little studied. Recent surveys have provided information on rangelands, wildlife, and pastoral systems in the Reserve. However, the health status of both wild and domestic ungulates in the Reserve, and therefore the risk of disease transmission between them, is still poorly

known. Recent information is apparently nonexistent or not accessible. To enhance the survival of both wildlife and livestock, we will need to gain insight into the presence of infectious agents in wild ungulates, their prevalence and incidence, and the extent to which they may affect the survival of exposed populations. The WCS ecosystem health component — which I represented during this visit — will help Chinese partners test on a small scale a cooperative system on epidemiological surveillance and disease detection. It will bring expertise in the field of wildlife disease, but will also be in charge of identifying partners involved in the project and their level of expertise, outlining the frame of their future cooperation, and launching pilot cooperative projects in the field involving data collection. Eventually it will help outlining a strategy for the health management policy of the protected area. Principal project partners are the Tibet Academy of Agriculture and Animal Sciences (TAAAS), the Tibet Autonomous Region Forestry Bureau (TFB), the Institute of Zoology (IOZ) of the Chinese Academy of Science, and the World Wildlife Fund (WWF) China. TAAAS, TFB and IOZ are the three principal partners involved in the wildlife health component of the project. As a matter of fact TAAAS supervises all health issues related to livestock and poultry in Tibet and TFB those related to wildlife, while IOZ, through its National Research Center for Wildlife Borne Diseases (NRCWBD), has been mandated by TFB and the Chinese Academy of Science to investigate any wildlife disease issue in Tibet.

Summary of first visit to China of WCS ecosystem health component

During my two-week stay (12–27 October 2007) in China I was unfortunately not given the permission to travel to Tibet. The reasons underlying this decision were administrative and contextual; I should therefore be able to visit partners in Tibet during my next visit to China in early 2008. Nevertheless, I could meet people involved in the project at IOZ in Beijing and in particular Prof. Hongxuan He, head of the NRCWBD. In theory any case of wildlife disease in China has to be reported to this laboratory for further investigation. The NRCWBD works in collaboration with sister institutes and research centers in other ministries. The creation of this centre is recent (2003?) and is directly related to the emergence of diseases probably originating from wildlife reservoirs such as severe acute respiratory syndrome (SARS) and avian influenza. NRCWBD was involved in the highly pathogenic avian influenza virus (HPAIV) outbreak in Qinghai Lake in 2005 (6000+ wild birds died of H5N1 infection). NRCWBD is also concerned by diseases affecting endangered species such as the giant panda (*Ailuropoda melanoleuca*) and the Tibetan antelope (*Pantholops hodgsoni*). Those two flagship species of Chinese conservation are under the scrutiny of national authorities. Any diseases affecting them must therefore be investigated in priority. Prof. Hongxuan He has apparently the capacity to run most serological tests for an early detection of a range of diseases, as well as bacterial and virus isolation. However for the handling of dangerous infectious agents, such as HPAIV, the NRCWBD collaborates with P3-

confinement laboratories in China. According to Prof. Hongxuan He, with the exception of emerging diseases such as HPAIV, there are only historical data concerning wildlife disease occurrence in Tibet. I also met Prof. Hanchun Yang at the department of preventive medicine of the College of Veterinary Medicine in Beijing. He was not informed of health issues in livestock in Tibet, and wildlife diseases fell outside his field of expertise. From general discussions with these two interlocutors it appears there is a relative lack of communication between regions in China concerning wildlife and even livestock health issues. In addition, Tibet being a politically sensitive area, communicating about diseases in this region, especially with a foreigner, seems to be a sensitive matter. Prof. Hongxuan He was keen to collaborate to the project within the framework of his mandate and we believe his laboratory has the capacity to run a number of microbiological and serological investigations on samples that will be collected in Tibetan wildlife.

Challenges for ecosystem health management in Chang Tang Natural Reserve

The challenges facing future health managers of the second largest protected area in the world are tremendous. According to my very preliminary analysis of situation, the current lack of knowledge on health issues in the Chang Tang ecosystem seems to be due to five main causes. First, the area is remote and exceptionally vast. Second, there are no easily accessible baseline data. Third, available staff in the field apparently lack capacity and logistical support. Fourth, there seems to be a lack of cooperation and agreement about mandates of animal diseases surveillance between acting agencies (i.e. Ministry of Forestry and Ministry of Agriculture). Finally, there is no regional strategy concerning wildlife disease issues. Remoteness and vastness of the area is an essential factor that will affect every aspect of the strategy and logistical solutions we may propose in the future. Without visiting the area, meeting with the animal health actors in Tibet, and assessing the efforts put into the detection of diseases in wildlife, I can not draw any action plan yet to improve the four last points enumerated above. Therefore the present document will only propose some guidelines concerning baseline data collection. We will focus on wild ungulates, as they are the most susceptible to be infected by, or infect, livestock.

As mentioned earlier, virtually nothing is known about the presence and prevalence of infectious agents in non-domestic herbivore populations of Tibetan Highlands. However, in the context of cohabitation between domestic and non-domestic herbivores such as reported in the Chang Tang Reserve, transmission of diseases from one population to the other through direct or indirect contacts could have a great impact in term of economical and biodiversity losses. Because of that, one important objective of the ecosystem health component of the project will be to promote the acquisition of baseline data on infectious agents circulating in wild ungulates in the Reserve. This could be done through an epidemiological surveillance system. Epidemiological surveillance is the systematic collection, analysis and dissemination of health data for the planning, implementation and

evaluation of health programs. Sensitivity (probability of a positive test among infected individuals) and specificity (probability of a negative test among non infected individuals) are two essential attributes of quality of any epidemiological surveillance operation. Epidemiological surveillance is a very important aspect of disease monitoring in wildlife population. It is based on the ability to detect through a variety of sampling schemes the presence of an infectious agent in a population, either directly via antigen detection, isolation/identification and nucleic acids detection, or indirectly through the measurement of circulating antibodies in the blood. Obviously epidemiological surveillance reaches maximum sensitivity when it is directed towards infectious agents that have a reasonable likelihood of being present in the population. Because clinical diseases are rarely observed in free-ranging animals, wildlife epidemiologists base their detections either on random sampling of live and dead animals, targeted sampling of harvested specimens, or more rarely on animals presenting clinical symptoms of a disease.

Guidelines for baseline data collection in wild ungulates

Considering the large variety of potential infectious agents in any wild population, and in order to save time and money, we recommend to prioritize detection efforts according to the following criteria: 1- the recognized susceptibility (RS) of wild herbivores to the infectious agent; 2- the recognized presence (RP) of this infectious agent in livestock in Tibet, 3- the likelihood of transmission (LT) of the infectious agent through direct or indirect contacts between domestic and non-domestic herbivore populations. Diseases that fulfill those criteria should receive priority allocation of resources, logistics and manpower. Also a cooperation agreement should be put in place as soon as possible between the main diagnostic laboratory of the project (IOZ, NRWBD?) and National Reference Laboratories for Foot and Mouth Disease (FMD) and Peste des Petits Ruminants (PPR). The detections should be organized so as to evaluate the presence of these diseases in surveyed populations assess their prevalence, and distinguish between a spillover event of infection from domestic animals, a sustainable infection in wild species or a state of asymptomatic carriage. Recent investigations carried out by WCS in Mongolian gazelles (*Procapra gutturosa*) in the eastern steppes of Mongolia have shown how critically important it is to understand livestock/wildlife interface issues.

Priority diseases

We propose to evaluate in priority the exposure of non-domesticated herbivores in the Chang Tang ecosystem to: Foot and Mouth Disease (FMD), Peste des Petits Ruminants (PPR), Brucellosis, Blue tongue (BT) /Epizootic hemorrhagic disease (EHD), Hemorrhagic Septicemia (HS) and mange. These diseases clinically affect wild ungulates, are present in livestock in Tibet, and are likely to be transmitted from livestock to wild herbivores and possibly vice versa for a number of them.

Foot and Mouth Disease (RS, RP, LT)

- *Agent:* Foot and mouth disease virus (FMDV) is a member of the *Aphthovirus* genus in the Picornaviridae family. Seven distinct serotypes are recognized: A, O, C, SAT-1, SAT-2, SAT-3, Asia-1, the O serotype being the most common.
- *Occurrence:* FMD is widely distributed throughout the world. It is endemic in parts of Asia, Africa, the Middle East and South America. Since 1999, FMD outbreaks (O and Asia-1 serotypes) have been recorded in livestock in several provinces of China including Tibet.
- *Hosts:* FMD is an infectious disease affecting cloven-hoofed animals, both domestic (cattle, zebu, domestic buffalo, yak, sheep, goats, and swine) and wild (ruminants and Suidae). Camelidae (Bactrian camels, dromedaries, llamas, vicunas) have low susceptibility.
- *The disease in wildlife:* In Africa, natural reservoirs of FMDV are domestic cattle and African buffalo (*Syncerus caffer*). Although other domestic and wild species become infected, they seem to be unable to maintain the infection for more than a few months in the absence of cattle or African buffalo. Elsewhere in the world cattle are usually the main reservoir, although in some instances the viruses involved appear to be specifically adapted to domestic pigs or sheep and goats. Wildlife outside Africa has not, so far, been shown to be able to maintain FMD viruses. The evidence indicates that infection of deer in the past was derived from contact, direct or indirect, with infected domestic animals. Circumstantial evidence indicates, particularly in the African buffalo, that carriers are able, on rare occasions, to transmit the infection to susceptible animals with which they come in close contact: the mechanism involved is unknown. The carrier state in cattle usually does not persist for more than 6 months, although in a small proportion it may last up to 3 years. In African buffalo individual animals have been shown to harbor the virus for at least 5 years, but it is probably not a lifelong phenomenon. Within a herd of buffalo, the virus may be maintained for 24 years or longer. There is no information on the duration of the carrier state in another domestic buffalo, the swamp buffalo of East Asia. Domestic buffalo, sheep and goat do not usually carry FMD viruses for more than a few months. Understanding the role of wild ungulates in the epidemiology of FMD in Tibet is critical to developing efficient FMD control strategies.
- *Proposed actions:* Targeted and random detection. Evaluate the level of exposure to FMD in wild ungulates in contact with livestock as well as in animals captured, harvested, culled or found recently dead with no reported contact with livestock. Methods: virus neutralization, competitive solid phase ELISA, or liquid phase blocking ELISA.
- *Carrier stage:* Evaluate the presence of the viral agent in individuals presenting positive antibody prevalence (methods: antigen detection test, virus isolation, nucleic acid detection).

Peste des Petits Ruminants (RS, RP, LT)

- *Agent:* Like rinderpest virus, Peste des Petits Ruminants virus (PPRV) is a member of the *Morbillivirus* genus in the Paramyxoviridae family.
- *Occurrence:* Once thought to occur only in Africa, PPR is now present in the Middle East, Pakistan, Afghanistan, Nepal, Bhutan, Bangladesh and India. It has been reported in Tibet in 2007.
- *Hosts:* Goats and to a lesser extent sheep. Cattle and pigs develop unapparent infections.
- *The disease in wildlife:* To date the disease has only been diagnosed in captive dorcas gazelle (*Gazelle dorcas*), Nubian ibex (*Capra [ibex] nubiana*), gemsbok (*Oryx gazella gazella*) and Laristan sheep (*Ovis orientalis laristanica*). Experimentally the white tailed-deer (*Odocoileus virginianus*) is fully susceptible. No case has been reported so far in free-ranging wildlife and the susceptibility of Tibetan ungulates is unknown. To date no wildlife reservoir has been identified but the fact that cattle can carry and disseminate the virus without clinical symptoms suggest that the virus could have co-evolved to a commensal relationship in other hosts.
- *Proposed actions:* Targeted and random detection. Evaluate the level of exposure to PPR in wild ungulates in contact with livestock as well as in animals captured, harvested, culled or found recently dead with no reported contact with livestock (methods: virus neutralization test or competitive ELISA).
- *Carrier stage:* Evaluate the presence of the viral agent in individuals presenting positive antibody prevalence (methods: antigen detection test, virus isolation, RNA detection).

Brucellosis (RS, RP, LT)

- *Agent:* Coccobacilli (short-roded Gram-negative bacteria) of the genus *Brucella*. There are several different species of *Brucella*, with different host specificities.
- *Occurrence:* It has a worldwide distribution although several countries have succeeded to eradicate it. It is present in Tibet.
- *Hosts:* In the Artiodactyla order, *B. abortus*, *B. melitensis*, and *B. ovis* affect cattle, sheep and goats.
- *The disease in wildlife:* *Brucella* infections have been documented worldwide in a great variety of terrestrial and marine wildlife species. *B. abortus* has been isolated from a range of non-domestic artiodactyls including the bison (*Bison bison*), the elk (*Cervus elaphus*), and the Cape eland (*Taurotragus oryx*). Arabian oryx (*Oryx leucoryx*), chamois (*Rupicapra rupicapra*) and alpine ibex (*Capra ibex*) are susceptible to *B. melitensis*, with clinical signs, while reindeer (*Rangifer tarandus*) can be infected by a biovar of *B. suis*. An important consideration with regard to terrestrial brucellosis in wildlife is to distinguish between a

spillover of infection from domestic animals and a sustainable infection in wild species.

- *Proposed actions:* Targeted and random detection. Evaluate the level of exposure to *Brucella* in wild ungulates in contact with livestock as well as in animals captured, harvested, culled or found recently dead, with no reported contact with livestock (methods: at least two serological tests such as Rose Bengal and competitive ELISA).
- *Carrier stage:* Evaluate the presence of the bacterial agent in individuals presenting positive antibody prevalence (methods: antigen detection test, virus isolation, nucleic acid detection).

Blue Tongue (RS, RP, LT?) and Epizootic Hemorrhagic Disease (RS, RP?, LT?)

- *Agent:* Blue Tongue virus (BTV) and Epizootic Hemorrhagic Disease virus (EHDV) are members of the *Orbivirus* genus in Reoviridae family. A total of 24 serotypes have been identified worldwide for BTV and 10 for EHDV.
- *Occurrence:* BT and EHD are transmitted by midges of the genus *Culicoides*, which are biological vectors. The central role of the insect vector in BT and EHD transmission makes distribution of those two diseases strongly related to the distribution of *Culicoides*. Hence BT and EHD distribution is governed by ecological factors, such as rainfall, temperature, humidity and soil characteristics. BTV exists in North, Central, and South America; Africa; and parts of Asia; Europe; the Middle East; and the South Pacific; EHDV is probably similarly distributed. BT has been diagnosed in China for the first time in 1979. BTV seems to have recently reached Tibet (*Culicoides* have been found in Tibet at altitudes up to 4,200 m). It is unclear if EHDV is currently present in Tibet.
- *Hosts:* BT is an infectious, non contagious disease of sheep and other domestic and wild ruminants, such as goats, cattle, deer, bighorn sheep, most species of African antelope and other Artiodactyla.
- *The disease in wildlife:* The outcome of BTV infection ranges from unapparent in the vast majority of infected animals to fatal in a proportion of infected sheep, deer and some other wild ruminants. In non domestic ruminants, clinical symptoms can vary from an acute hemorrhagic syndrome with high mortality, as observed in white-tailed deer (*Odocoileus virginianus*), to non existent as seen in the North American elk (*Cervus canadensis*). EHDV can produce a disease in wild ruminants with clinical manifestations identical to those observed in BT. BTV and EHDV have been relatively little investigated in wildlife outside North America.
- *Proposed actions:* Targeted and random detection: Evaluate the level of exposure to BTV and EHDV in wild ungulates in contact with livestock, as well as in animals captured, harvested, culled or found recently dead, with no reported

contact with livestock (methods: agar gel immunodiffusion and competitive ELISA)

- *Carrier stage*: Evaluate the presence of the viral agent in individuals presenting a positive antibody prevalence (methods: virus isolation and serotyping, nucleic acid detection).

Haemorrhagic Septicaemia (RS, RP, LT?)

- *Agent*: *Pasteurella multocida*, a Gram-negative coccobacillus bacterium. Two serotypes, the Asian B:2 and the African E:2 (Carter-Heddleston system), are mainly responsible for the disease. They correspond to serotypes 6:B and 6:E (Namioka-Carter system). In wild animals, serotype B:2,5 is predominant. The association of other serotypes, namely A:1 and A:3 with a HS-like condition in cattle and buffaloes in India has been recorded.
- *Occurrence*: Worldwide occurrence. Present in Tibet.
- *Hosts*: Cattle, water buffaloes and bisons are natural reservoirs of the disease, buffaloes being the more susceptible. *Pasteurella multocida* is a commensal agent in the upper respiratory tract of many animals.
- *The disease in wildlife*: Recent HS cases have been reported in Asia and Europe in wild mammal species, including water buffaloes, deer, elephants and yaks. In Asian countries, disease outbreaks mostly occur under the climatic conditions typical of monsoon (high humidity and high temperatures).
- *Proposed actions and carrier stage*: Targeted, random detection. Evaluate the level of exposure to *Pasteurella* in wild ungulates in contact with livestock as well as in animals captured, harvested, culled or found recently dead, with no reported contact with livestock (methods: tonsillar biopsy or swabbing culture, isolation and typing).

Mange (RS, RP, LT)

- *Agent*: Mange results from infestation by either astigmatid mites belonging to different genus including *Chorioptes*, *Psoroptes*, *Sarcoptes*, *Otodectes*, *Knemidokoptes*, and *Notoedres*, or prostigmatid mites such as *Cheyletiella*, *Demodex* and *Psorobia*.
- *Occurrence*: Several of these mites have been recorded in Tibetan livestock.
- *Hosts*: A very large variety of hosts. Livestock are infested worldwide.
- *The disease in wildlife*: Reported in a large variety of non domestic species. Noteworthy there was a recent occurrence of sarcoptic mange infesting blue sheep (*Pseudois nayaur*) in northern Pakistan that may have reached Tibet.
- *Proposed actions and carrier stage*: Targeted, random detection: Evaluate the level of exposure to mange ectoparasites based on the recovery and identification of the mite from the affected hosts (hair clipping, skin scraping, ear canal swabbing).

Lower priority diseases

Other diseases could be investigated, depending on the availability of resources, expertise and timeliness: Infectious Bovine Rhinotracheitis (IBR), Bovine Viral Diarrhea (BVD), Parainfluenza-3 virus (PI-3) infection, Bovine Respiratory Syncytial Virus (BRSV) infection, and Rinderpest (RS, LT). Exposure of wild animals to these agents may have been reported, but seldom with clinical symptoms of infections (IBR, BVD, PI-3 infection, BRSV infection), or the disease is likely to be absent from China (Rinderpest).

Infectious Bovine Rhinotracheitis (RS?, RP?, LT?)

- *Agent:* Family Herpesviridae, genus *Varicellovirus*, 4 recognized subtypes.
- *Occurrence:* Worldwide. Occurrence in Tibet not known.
- *Hosts:* Cattle.
- *The disease in wildlife:* Exposure was detected in a very large variety of wild herbivores but no clinical signs or significant findings associated with the diseases have so far been reported in wild herbivores.
- *Random detection only:* Evaluate opportunistically the level of exposure (for example with competitive ELISA) of any wild ungulate captured, harvested, culled or found recently dead, with or without likely contact with livestock.

Bovine Viral Diarrhea (RS?, RP?, LT?)

- *Agent:* Family Flaviviridae, genus *Pestivirus*, 2 recognized types.
- *Occurrence:* Worldwide. Occurrence in Tibet not known
- *Hosts:* Cattle.
- *The disease in wildlife:* Exposure was detected in a very large variety of wild herbivores but no clinical signs or significant findings associated with the diseases have so far been reported in wild herbivores.
- *Random detection only:* Evaluate opportunistically the level of exposure (for example with competitive ELISA) of any wild ungulate captured, harvested, culled or found recently dead, with or without likely contact with livestock.

Parainfluenza-3 virus infection (RS?, RP?, LT?)

- *Agent:* Family Paramyxoviridae.
- *Occurrence:* Worldwide. Probable occurrence in Tibet.
- *Hosts:* Widespread in cattle.
- *The disease in wildlife:* Exposure was detected in a very large variety of wild herbivores but no clinical signs or significant findings associated with the diseases have so far been reported in wild herbivores.

- *Random detection only*: Evaluate opportunistically the level of exposure (for example with competitive ELISA) of any wild ungulate captured, harvested, culled, or found recently dead, with or without likely contact with livestock.

Bovine Respiratory Syncytial Virus infection (RS?, RP?, LT?)

- *Agent*: Family Paramyxoviridae, genus *Pneumovirus*.
- *Occurrence*: Worldwide. Probable occurrence in Tibet.
- *Hosts*: Widespread in cattle.
- *The disease in wildlife*: Exposure was detected in a very large variety of wild herbivores but no clinical signs or significant findings associated with the diseases have so far been reported in wild herbivores.
- *Random detection only*: Evaluate opportunistically the level of exposure (for example with competitive ELISA) of any wild ungulate captured, harvested, culled or found recently dead, with or without likely contact with livestock.

Rinderpest (RS, LT)

Agent: Virus family Paramyxoviridae, genus *Morbillivirus*, three recognized lineages, lineage 1 and 3 most probably eradicated thanks to vaccination en mass.

Occurrence: In Africa it has been eradicated from several countries and sub-regions, and is normally absent from the northern and southern parts of the continent. Lineage 3 of the virus used to occur in the Middle East, southwestern and central Asia. This lineage may be close to eradication as it has not resurfaced in the region since 2000 (Pakistan). In China, the disease has been eradicated through vaccination since 1955.

Hosts: Cattle, zebu, water buffaloes, sheep, goats are susceptible as well as a very large number of wild animals within the Artiodactyla order, such as the African buffalo (*Syncerus cafer*), the Cape eland (*Taurotragus oryx*), kudu (*Tragelaphus* spp.), wildebeest (*Connochaetes* spp.), various antelopes, etc.

The disease in wildlife: Even though infections with lineage 2 (possibly the last remaining lineage in the world) may pass unnoticed in cattle, the virus is highly infectious in wildlife. Among species commonly regarded as highly susceptible (buffalo, eland, and lesser kudu), it causes fever, a nasal discharge, typical erosive stomatitis, gastroenteritis, and death. Buffaloes infected with lineage 2 showed enlarged peripheral lymph nodes, plaque-like keratinized skin lesions and keratoconjunctivitis. Lesser kudu were similarly affected, but whereas blindness — caused by a severe keratoconjunctivitis— was common, diarrhea was unusual. Eland showed necrosis and erosions of the buccal mucosa together with dehydration and emaciation. Therefore, a diagnosis of rinderpest in any of these species points to the likelihood of a simultaneous transmission of the virus, even at a subclinical level, to neighboring cattle. There are reasonable evidences that the disease may have been eradicated in Asia and as such the relevancy of searching for the presence

of the virus in Tibet may be questionable. However the virus might occur in a mild form and remain unnoticed in remote areas and there are some reasons to look for its presence in wildlife.

Proposed actions: Random detection only: Evaluate opportunistically the level of exposure (with competitive ELISA) of any wild ungulate captured, harvested, culled or found recently dead, with or without likely contact with livestock.

Sampling methods

The agency in charge of animal sampling should obtain as soon as possible the permissions required for the sampling work within a legal and acknowledged framework. It will also need to connect with any project intending to capture wild animals in the CTNR or in its immediate vicinity and get involved in sampling collection during such operation or request participating staff to do so following a standardized sampling method.

Passive sampling

It is the opportunistic sampling from multiple sources, especially related to morbidity and mortality events. Pre-existing programs such as regulated hunting and culling, as well as scientific captures, may also provide occasions to perform sampling. Samples will have to be collected according to a rigorous and standardized protocol during each of such missions. Because these surveillance operations are complicated and require a coordinated planning, an active and careful collaboration between the different partners and biologists involved in these missions will be essential. While capture or harvest operations will provide access to fresh material (blood, fresh tissues), post-mortem examinations, for example in the context of a mass-mortality event, may also provide valuable information about disease ecology such as sex and age ratio of dead animals and probable death causes. In the case of infectious agents resisting post-mortem conditions, sampling of infected tissues can confirm the cause of death (for example bone marrow sampling in suspected *Pasteurella* sp. infection or FMD or scab sampling for detection of parapoxvirus infection).

Active sampling

It is the focused and targeted sampling of a species with predetermined level of accuracy and detection. It is especially designed to collect samples from animals but can also be combined with other objectives (e.g. population productivity assessment). One such mission has been proposed in summer 2006 by Dr Schaller and would consist in sampling blood of newborn calves of Tibetan antelopes to assess the exposure status of their mothers. Ruminants acquire no antibodies from the mother before birth. Because maturation of the immune system is incompletely achieved at birth, antibody production develops slowly in the newborn ruminant. The newborn would rapidly be overwhelmed by invasive organisms if it did not

receive antibodies from the mother. Antibodies acquired in this way come from the colostrums via the neonatal intestine. In most ruminants antibodies contained in the colostrum of the mother are maximally absorbed through the digestive tract within the first 2 hours of life. After 24 hours, a 'closure' phenomenon occurs which prevents absorption of additional antibodies from the milk. However antibodies in the late colostrums and in the milk still contribute at protecting the calf at the level of digestive tract lumen. In the meantime, maternal antibodies that now circulate in the newborn calf are an essential part of its early humoral immunity, a protection capability fully originating from the mother. In theory sampling serum from a less than one-week-old calf would allow the detection of maximal levels of maternal antibodies and provide a qualitative understanding of the diseases the mother has been exposed to. Mother antibody titer will decrease at a rate that varies according to initial antibody level, species, level of antibodies transferred to the calves and type of antibodies. Depending of the initial titer maternal antibodies could be detected for several months after birth. It is therefore not abnormal to find in young calves a high titer of antibodies against a specific pathogen shortly after its birth and no track of such titer a couple of months later. Tibetan antelopes aggregate in specific locations and at specific time of the year to give birth. It would be worth organizing a mission at this time of year to collect samples from newborn calves. In addition freshly dead calves as inevitably reported during calving time and even weak females may also provide the opportunity to better understand the exposure of Tibetan antelopes to the targeted infectious agents.

Conclusion

Acquiring baseline data and understanding whether wild herbivores are exposed to potentially lethal diseases transmitted by livestock or act as reservoir species are important initial steps in the control and management of these diseases. Next step will be to identify quantifiable risk factors, and include them in a landscape-scale management program that will minimize the risk of catastrophic losses in the unique wildlife of the Chang Tang Nature Reserve.