

Snow Leopard Conservation Grants, Snow Leopard Network

FINAL REPORTS FOR 2014 PROJECTS

DUE: 15 FEBRUARY 2015

Please submit your final report by the due date above. We would appreciate it if you could follow the suggested format below. Additionally, please send us copies of any detailed technical report(s), papers, and other output arising from this work. Please refer to your original proposal for items such as objectives, methods, etc. unless those were substantially altered during the course of the work. If so, please explain why.

1. **Executive Summary:** No more than 750 words. Please describe the original goals and the final results of your project. This may be used in press releases and other publicity material about the Grants Program, so please write it for the general public who may not have scientific background.

Maximising One Health Approaches to Enable Detection of Emerging Disease Threats in a Changing World

This project investigated the health status and disease threats faced by Snow Leopards (*Panthera unica*) in the South Gobi desert of Mongolia. Disease threats to endangered species are not an isolated issue, but a series of complex and interconnected threats, which in combination can manifest in disease from a number of sources. This includes interactions with domestic species, leading to direct spread of disease, subsequent conflict with humans, reduction in populations of prey and the difficult-to-predict, complex impacts of climate change on predators, prey and vectors. For this project it was important to identify potential sources of disease, reservoirs and the disease linkages in the system. Due to the broad scale of diseases possible only zoonotic diseases were selected to test for.

Originally this project was to address 4 of the priority needs identified in the Snow Leopard Network's 2013 Threat Analysis. It primarily addresses disease threats;

- 1) Diseases of Snow Leopards, and secondarily through links to disease;
- 2) Prey reduction due to disease
- 3) Growing Livestock Populations & Intensifying Human-Wildlife Conflict
- 4) Impacts of Climate change.

A "One Health Approach," which reaches across the traditional boundaries of conservation, human health and production animal diseases to understand the complex interactions and relationships among and between snow leopards, humans, livestock and their prey was applied to this project. As human populations grow and economies develop, expectations and needs for livelihoods for indigenous peoples also grow. The primary source of livelihood for the indigenous people of the southern Gobi desert is their goats. In other areas, although leopards do kill livestock, losses due to disease outnumber leopard kills. Understanding this helps to mitigate the conflict between herders and carnivores and provides other ways to build human well-being through herd health and productivity, without increasing conflict (Dar *et al.* 2004). There is

a strong interactive effect between intensification of herding, increasing pressure on fragile rangeland productivity, reduction of vegetation standing crop and the population of wild herbivores that snow leopards depend on. Under these conditions snow leopards become more likely to prey on the goats as wild herbivores decline, initially causing both human/wildlife conflict, but also increasing the frequency and intensity of interactions that can transfer diseases between all the elements of the system- leopards, natural prey, environmental reservoirs, livestock and people.

Understanding the prevalence of zoonotic diseases and parasites in this system is critical to predicting future potential for diseases to endanger vulnerable populations of leopards as well as human health and livelihood.

This project is still continuing due to logistical delays concerning the laboratory analyses. However preliminary results are exciting and encouraging with a number of the pathogens tested for to date being present in several of the species. So far we have had positive results for Toxoplasmosis- present in snow leopards, rodents and goats. Results for ibex and dogs are pending and we are waiting to hear from the human health authorities if cases have been reported from within the study area. Leptospirosis has also been confirmed in several of the rodent species and two of the Snow Leopards tested. It has yet to be tested for in the dogs and again we are waiting for human reports.

However the collection of data on climate change was proving too difficult and valid information could not be obtained for the short time-frame of the project. Therefore the data collected will provide a baseline for future climate change work with respect to the identification of pathogens and vectors likely to transmit disease being present.

2. Objectives: What was the purpose of the project? How was it expected to contribute to the knowledge or conservation of snow leopards, their prey, or habitat?

The project was designed to directly build knowledge and understanding of the disease threats present in the south Gobi desert of Mongolia and the disease linkage interactions between Snow Leopards, their prey (both domestic and wildlife) and humans. As this type of study has not been performed previously, it will provide a basis for future conservation strategies of snow leopards with respect to disease prevalence in a changing world. Management and action plans will be developed so that the necessary knowledge, tools and infrastructure can be in place or easily accessible in the event of a major disease outbreak.

The herd and rodent-sentinel monitoring will provide the potential for rapid detection of such an outbreak, so that economical and environmental impacts could therefore be minimized. It will determine the magnitude of livestock loss to disease and the economic losses associated, in comparison with snow leopard related losses. This will allow evaluation of the costs and benefits of potential herd health intervention scenarios against other snow leopard conservation actions planned for the south Gobi region. The herd health surveys provide the basis to develop health management plans for the herders to improve the productivity of their herds, including routine worming and vaccination. Improving domestic herbivore health links to health of the wild herbivores, such as ibex, due to decreases in environmental contamination by pathogens such as intestinal parasites shed onto common feeding grounds. Improving herd health and resilience has the potential to reduce human/wildlife conflict. Awareness and education regarding avoidance of zoonoses provides a direct benefit to the herders themselves.

The project will provide baseline data to aid in predicting or detecting future effects of climate change on the ecosystem in relation to presence/absence of zoonotic disease and the species they affect. Without baseline data to act as a reference point it is impossible to determine what changes, if any, are occurring. As this only becomes apparent over longer time frames it would be beneficial if the study leads to a long-term monitoring program.

Research

- 2. Methods:** Describe the methods you used in detail, so that someone else could repeat the work, or, avoid the problems that you encountered.

DATA COLLECTION – Field trips to the Mongolian study area were performed once a year. During these field trips samples for pathogen identification were collected in similar ways for each species. These included blood, faeces and external parasites. Survey questionnaires on human health and herd/wildlife health, will be distributed to the herders during the 2015 field trip. **Snow Leopards**-The Snow Leopard Trust south Gobi project has been running since 2009 and there is a bank of samples that have already been collected from 19 individual snow leopards (see Johansson et al. 2011 for details on captures). Once captured and sedated, snow leopards are examined for general health. This includes taking heart rate and body temperature, assessing mucous membrane colour, weight, sex, reproductive status, body condition score, lymph node size, tooth wear as an estimate of age and for any signs of wounds or ill health. Rectal swabs are collected prior to temperature monitoring and stored in a charcoal transport medium for future antibiotic resistant microbial gene analyses. Twenty ml of blood is collected from the cephalic vein and placed in serum separating and EDTA tubes. Blood samples are also placed on Waltham FTA cards for DNA and bacterial analysis and Nobuto strips for further serology. The advantages of using these two methods for storing blood samples in remote areas are: 1) requires small blood volumes and 2) can be stored at room temperature. The serum tubes are centrifuged on a solar powered centrifuge, serum is decanted and placed in cryovials for freezing back at the base camp where there is a refrigerator. 1.0ml of whole blood is also collected in Lithium heparin tubes. These samples are then analysed in the field using an Abaxis hand held I-Stat machine for haematology and biochemistry values, thus obtaining baseline values-this has not been performed on Snow Leopards in the wild before. Blood smears are also made in the field using whole fresh blood, dried, fixed and latter stained for identification of haemoparasites and cell morphology. Faecal samples are collected where possible. Half the sample is dried and stored with a dessicant and analysed for parasites, bacteria and viruses. The other half is placed in RNA later for virus and bacteria identification

Rodents are live trapped using (Elliot or Sherman Style traps). Trap lines cover different habitat types to survey as much of the overlap area with Snow Leopards, native herbivores and domestic goats as possible. These areas include the plains grazing areas where the goats and Ibex feed, rocky gorges where snow leopards hunt and around waterholes where all of the target species drink. Traps are also placed, with owners' permission, inside Gers (local houses). Rodents are transferred from the traps into calico bags for handling and sampling. If necessary they are sedated with Zoletil 100 at a dose rate of 15mg/kg to allow handling for body measurements and blood collection. Rodents are ear notched to allow for identification in case of recapture. The ear notch sample is stored in ethanol for DNA analyses of species identification. Blood samples, depending on the size of the rodent, are taken and stored as per those of the snow leopards. If the blood volume is too small for serum separation due to the small size of some of the rodent species, only blood smears and Nobuto strips are used for serum storage. Faecal samples are collected from the traps, calico holding bags or while sampling. Rodents are also weighed,

photographed, morphological features noted and body measurements taken for future identification purposes due to the lack of literature available on these species.

Goats - Approximately 90 herding families live within the study area. Goat herd size ranges from 200-400 animals. Twenty families across the study area have been selected for sampling of goats. Approximately 10% of each herd were sampled. Blood and faeces have already been and will be collected and stored in the same manner as for the other study species.

Dogs- Each herding family also owns dogs, so where possible, blood samples will be taken from those dogs and faecal samples when possible.

ibex -Due to the difficulty of catching ibex, faecal samples only will be collected.

Human Health Data- Local people will be asked to fill in a questionnaire regarding human health and herd health in their lifetime. These will be distributed with the help of local Snow Leopard Trust employees.

Local health authorities will also be asked to fill in the human health surveys and provide where possible any extra information about disease in the area.

Water samples will be collected from all waterholes and wells that are accessible in the study area. These will be collected utilizing field filtration methods as described by Goldberg & Strickler (2013) to look for water-borne pathogens including giardia and cryptosporidium.

Diagnostic Lab analyses - The sample analyses were conducted at the Uppsala Biomedical Centre, Uppsala University, Sweden during my visit to the lab for approximately one month in October/November 2014.

These involved molecular techniques- ELISAs and PCRs appropriate to the samples and diseases. The samples will be tested for the following pathogens/diseases: rabies, anthrax, plague, leptospirosis, hantavirus, toxoplasmosis, brucellosis, Q fever, cryptosporidiosis and giardia. These pathogens were selected based on their occurrence in other areas of Mongolia and as they can transfer between wildlife, domestic animals and humans, and cause significant morbidity in all three groups.

Statistical analyses will involve comparisons between the species as to which pathogens are common to each and thus that can be transmitted between species by using Chi squared tests and ANOVA. Multivariate logistic regression analysis will identify relationships between SL and pathogens and the other animal species.

Animal and human health questionnaire surveys - The surveys are mainly qualitative and were kept simple due to the extra issues of having to be translated twice – once into Mongolian and then back to English, which will increase the risk of loss/change of meaning. Comparisons of answers to the the ‘yes/no’ questions will be made using Chi squared tests.

4. Results: Please describe in detail the results of your project. Please illustrate clearly how your stated goals and objectives could be met. You may wish to include tables or graphs in this section if appropriate. This section will be very important to explain the value of these grants to funders of the Snow Leopard Conservation Grant Program. Please be clear, concise, and thorough.

This project is ongoing due to the difficulties in obtaining laboratory space. The results to date are promising and justify the continuance of the project.

To date samples from; 19 Snow Leopards, 200 Goats, 157 Rodents, 4 dogs, have been collected and 100 ibex faecal samples.

As can be seen in Table 1 several of the pathogens that have been tested to date are occurring in more than one species, which verifies our hypothesis that the pathogens we are investigating will cross species.

See Table1 below for a summary of the results to date for pathogen analyses.

	Toxoplasmosis	Leptospirosis	Brucellosis	Hanta Virus	TBE
SPECIES					
Snow Leopard	10% positive out of 20 tested	10% out of 20 tested			
Dog					
Rodent	15.7% out of 70 tested	27.8% Positive out of 90 tested		85 tested all negative	18 tested negative
Goat	94.4% positive out of 90 tested		90 tested all negative		
Ibex					

5. Discussion: Please evaluate your own work. What did you learn that could help others wishing to do similar projects? How do you see the results being applied to conservation? What additional work is now needed based on your findings?

These types of study always take longer than expected. It is critical to be prepared to spend sufficient time getting to know the area and its components to be able to understand how they fit in and work together. For people wanting to do similar projects its important to keep exploring the literature so as to have as much background knowledge as possible and to be flexible. The more you work in the area the more familiar and aware you become of what's required so the study has to be a living entity ready to evolve as more information comes to light. Fleible/adaptive

The results from this study will allow for a greater understanding of disease processes that can occur in this type of desert environment. Vaccination programmes for domestic animals such as distemper and leptospirosis for the dogs and a combined vaccine and worming protocol for the goats should also have advantageous effects on the wild animals by preventing sources of possible environmental contamination. This will also benefit the conservation of the Snow Leopards by reducing the risk of being infected with canine distemper virus and leptospirosis from the dogs. It should also improve the quality and hopefully the quantity of their prey species be reducing parasite burdens through regular drenching of the goats which will lead to less contamination of pastures that the goats share with the Ibex. This should also improve the quality of the meat for the herders and consequently their own health.

Further work needs to look at the effects of climate change on the existence and prevalence of the pathogens we've identified. The study could be further advanced by expanding the number of species to be examined for the presence of disease. This could include animals such as the Pallas cat and some of the larger rodents in the area, which could be a prey source for the snow leopards. A more intense study of the ibex health alone would also be advised and of other carnivores in the area such as wolves. Migratory birds would also be interesting to catch and sample as they are definitely a source for bringing new pathogens into the area.

6. Photographs: If you have good photographic (preferably digital) images of your project that we could use to advertise the Grants Program, please submit them at this time. Please be sure to include a brief description of the photo and provide the credits for the photographer.



Snow Leopard after sampling and radio-collar placement waking from sedation

Final reports and digital images should be emailed to grants@snowleopardnetwork.org.