



**One Health in Wakhan:
A summary of health investigations carried out by WCS
in the Wakhan District, Afghanistan
2006–2014**

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SUMMARY

Between 2006 and 2014 the Wildlife Conservation Society implemented and organized clinical and laboratory investigations on livestock in Wakhan with the intention to detect zoonotic diseases and those infectious agents capable of affecting sympatric wild ungulates. Bovine tuberculosis and glanders were not found. Brucellosis had a very low prevalence among tested livestock indicating that the disease is not endemic in Wakhan and of low exposure risk to people. Toxoplasmosis and Q fever did occur with relatively high prevalence in livestock and should be considered as endemic in the area, both diseases being of public health concern. During the study period we found no evidence of presence of rabies, but locals suggested that the disease might occur occasionally as epidemics affecting wild carnivores. Tick-borne relapsing fever seemed endemic in the area. Among diseases that could pose a risk to wildlife, foot-and-mouth disease was endemic across the district, and bluetongue and Border disease in the Wakhan Valley. Peste des Petit Ruminants occurred as irregular and localized epidemics. Contagious caprine pleuropneumonia an emerging disease in Central Asia was not detected in Wakhan. The relative paucity of infectious agents in livestock in Wakhan is remarkable and may result from a combination of factors, including the extreme remoteness and isolation of the area, the fact that Wakhan is a livestock production area where non-native domestic animals are seldom introduced, and the harsh winter conditions and healthy predator population that may constitute efficient selection factors against the weakest and sick animals. However, any relevant contagious pathogen introduced into such isolated and immunologically naïve livestock population could have disastrous effects. It is therefore of utmost importance to avoid any importation of domestic animals into the district without thorough medical check-up and a quarantine period outside of the district.

The human population in Wakhan suffers significant health problems. We estimated by extrapolation the infant mortality rate (i.e. the number of deaths of infants under one year old per 1,000 live births) in the Wakhi population in 2013-2014 at 106, significantly higher than the average value for Afghanistan in 2010-2015 (ca. 70). In 2013-2014 acute respiratory infections were allegedly responsible of 56.7%, and 55.2% of the deaths in infants less than one year old, and five years old, respectively. Also 83% of households in Wakhan Valley reported that at least one family member suffered an acute respiratory disease syndrome. The infant mortality is reported to be even higher in the Kyrgyz community.

Despite significant efforts in the last decade, human and animal health facilities in Wakhan are still few, unequipped, and staffed by a small number of poorly trained personnel.

Human health

In 2014 the One Health team of WCS carried-out a questionnaire survey on the socio-demography and health of the Wakhi community in the Wakhan Valley between the village of Ftur at the western extremity of the district and the village of Sarhad-e Broghil, the eastern-most settlement in the valley that can be reached by car. The main aim of the survey was to evaluate the health of people, of their livestock and livestock predation cases by wild carnivores. In the course of the survey WCS investigators collected some succinct information on the health of the Wakhi community (ca. 90% of the population in the Wakhan district).

During the 2.5 month-long survey the team visited 1,404 households (allegedly all households) in the Wakhan Valley. The total population residing in the valley (98% Wakhi) was estimated at 14,948 people, including 7,666 males (51.3%) and 7,282 females (48.7%), in perfect accord with the estimated sex ratio at birth for Afghanistan of 1.05 male for 1 female. The average number of people per household was 10.64 ± 6.02 (SD) (min-max: 1-43). The population was young with 10,645 people aged less than 29 years old, or 71.2% of the surveyed population.

During the year prior to the survey (summer 2013 to summer 2014), 205 people died. Twenty-six (12.7%) of them were less than one month old, 41 (20.0%) were between one month old and one year old, and an additional 20 (9.7%) were one to five years old. Overall 87 (42.4% of reported deaths) infants less than five years old died, or ca. 3.5% of this age class. We estimated by extrapolation the infant mortality rate (i.e. the number of deaths of infants under one year old per 1,000 live births) at 106. For comparison the World Health organization reports an estimated average infant mortality rate in Afghanistan during the period 2010-2015 of ca. 70. http://www.who.int/maternal_child_adolescent/epidemiology/profiles/neonatal_child/afg.pdf. A report produced by Orphan Refugee Aid (ORA) International in 2006 suggested that the infant mortality rate in the Kyrgyz community could be even higher and proposed an under 5 mortality of ca. 520 per 1,000 live births (however, this dramatic figure was based on an extrapolation from a very small sample size).

In the Wakhi community acute respiratory infections were responsible for 56.7%, and 55.2% of the deaths in infants less than one year old, and five years old, respectively. In addition 118 people (57.6%) aged 5 to ca. 90 years old died during the year prior to the survey, with 'oldness', digestive, cardiovascular disorders and sudden deaths being responsible of ca. 60% of deaths. Acute respiratory disease was responsible of only 7.6% of deaths in this age class. However, 83% of households reported that at least one family member over the past year showed symptoms of an acute respiratory disease syndrome including one or a combination of the following; sore throat, muco-purulent nasal discharge, cough, chest pain and fever.

Because most people living in Wakhan are highly dependent on livestock meat and dairy products for their food security we were particularly interested at searching for the presence of zoonotic diseases that could be transmitted from livestock to man. Also, over the years the number of livestock has increased and as a result there is a competition between livestock and wild ungulate over grazing space. The contact between the two has increased such as the possibility of transmission of infectious agents from wildlife to livestock and vice versa. Recently disease transmission from livestock to wildlife has had devastating consequences on mountain ungulates in Central Asia. This interdependency between animal, human and ecosystem calls for a multidisciplinary health response that is ecologically sustainable. Here we report on zoonosis present in Wakhan and on livestock diseases potentially dangerous to wildlife.

Zoonosis

Toxoplasmosis

The agent of this infection is *Toxoplasma gondii*, a coccidium (protozoa) belonging to the phylum Apicomplexa. This protozoon has a worldwide distribution and is responsible of one of the most widespread of all zoonosis. We tested serologically 472 shoats (354 sheep and 118 goats) sampled in Wakhan Valley, Big Pamir and western Little Pamir in July-August 2008. Positive animals were found in the three landscape and the overall prevalence was 12.7% (CI95% 10.0%-16.0%) suggesting that the parasite is present at a relatively high prevalence in the Wakhan district.

However, the significance of this disease in Wakhan is probably marginal. In man the infection is in general very common but the clinical disease is relatively rare, occurs sporadically and has low level of incidence. A notable exception is congenital toxoplasmosis, which is not frequent but can cause severe disease and sequels. However, this form of the disease is probably very rare in Wakhan, as most pregnant women have been probably exposed on multiple occasions to the parasite before pregnancy through the ingestion of contaminated soil particles or the consumption of infected meat, which confers a lifelong immunity.

Q fever

It is a pneumo-rickettsiosis due to *Coxiella burnetii*. The infection is endemic in many areas and its presence has been confirmed in more than 50 countries including Afghanistan. As a general rule the infection in domestic animals is clinically unapparent. We tested serologically 472 shoats (354 sheep and 118 goats) sampled in Wakhan Valley, Big Pamir and western Little Pamir in July-August 2008. Reactors were found in the Wakhan Valley, western Little Pamir but not in eastern Big Pamir. The overall prevalence was 8.5% (CI95% 6.3%-11.4%). We tested again 309 sheep

sampled across the Wakhan Valley in October 2012 and found a similar prevalence of 10.8% (CI95% 6.6%-16.3%). Because the agent is extremely resistant in the external environment and the pastoral societies living in Wakhan maintain very close contact with livestock, human infection must occur in the area. The human infection however is often asymptomatic and its mild forms can be mistaken for other febrile diseases. However, it can also result in serious febrile infections including pneumonitis (slight cough, mild expectoration and occasionally chest pain). The disease range in severity but is in most cases benign, it rarely attacks children under 10 years old and is in general more severe in adults over 40 in whom it can even take a chronic course, affecting the cardiovascular system. The disease would deserve being thoroughly investigated in Wakhan considering the relatively high prevalence detected in sheep and goats, the mode of life of inhabitants living for the vast majority in very close contact with livestock, and the relatively high prevalence of reported pneumonia and cardiovascular problems in adults.

Brucellosis

The disease is caused by bacteria belonging to the genus *Brucella*, typically *B. melitensis* and *B. abortus* in Afghanistan. It has a worldwide distribution. The epidemiological situation of brucellosis in Afghanistan is increasingly better understood thanks to efforts of the Animal Health department of the Ministry of Agriculture efficiently supported by a modern and well-capacitated laboratory (i.e. Central Veterinary Diagnostic and Research Laboratory). However, the survey effort was undertaken only in several districts of most provinces and the prevalence of infection in livestock has been found to vary considerably from one place to the other. The disease is potentially very debilitating to man with a septicemic phase accompanied by continued, intermittent, and irregular fever, then followed by a suite of organ infections that can occur over a duration varying from several weeks to several years. Because the natural reservoir of *B. abortus* and *B. melitensis* are, respectively goat and sheep, and cattle, the three most common species of livestock in the Wakhan district and transmission by ingestion of animal products (e.g. fresh cheese, raw milk) predominates in areas where small ruminant brucellosis is enzootic, WCS health team invested significant efforts to search for the presence of the disease in livestock of the district. Over the period 2008-2012, we blood-sampled and tested serologically (Rose Bengal Test) 2,341 sheep and goats, 431 cattle and 352 yaks from the Wakhan Valley, Little and Big Pamirs. About half of these samples as well as all doubtful and positive samples with RBT were also tested with cELISA. Positive samples with RBT and cELISA were also tested with Complement Fixation Test. We found no positive case in sheep, goats and yaks, and one seropositive cow (RBT doubtful, cELISA and CFT positive). The prevalence in cattle was 0.2% (CI95% 0.0% – 1.4%) and nil in shoats and yak with an upper limit for CI95% in shoats of 0.15% and 1.1% in yak. Based on

these results the disease seems to have a very low prevalence among livestock in the district.

Bovine tuberculosis

Tuberculosis (TB) in man is mainly due to *Mycobacterium tuberculosis* that is a most specific bacterium rarely producing progressive disease in the lower animals other than nonhuman primates and occasionally in a handful of other species. In contrast *Mycobacterium bovis* the agent of bovine tuberculosis can cause progressive disease in most warm-blooded vertebrates, including man. As for *M. tuberculosis*, not all *M. bovis* infections in man progress to TB disease. When progressing to a disease form, the symptoms of *M. bovis* infection are similar to the symptoms of TB caused by *M. tuberculosis*, this can include fever, night sweats, weight loss and other symptoms depending on the part of the body affected by the disease (e.g. cough and chest pain for lung disease, or abdominal pain and diarrhea for the gastrointestinal disease). If untreated a person can die of the disease. Cattle are the main reservoir of *M. bovis*, and eating or drinking contaminated unpasteurized dairy products most commonly infects people. We have therefore tested 128, 153, and 143 dairy cows across villages of the Wakhan Valley in October 2010, 2011, and 2012, respectively. We have used the intradermal tuberculin test, which is the single most important diagnostic test for TB. We have found no reactor and therefore concluded that the prevalence of bovine TB in milking cows in the surveyed area was less than 0.9% (upper limit of the 95% Confidence Interval of the 0.0% prevalence). We have so far found no evidence that the disease exists in dairy cows in the Wakhan district.

Glanders

Glanders is caused by *Burkholderia mallei* bacteria and is primarily a disease of solipeds. Once distributed worldwide the disease still persists in some African and Asian countries. It is present in Pakistan and Iran and has been recently confirmed from Afghanistan too. The disease course is predominantly chronic in horses and is almost always acute in asses and mules. The acute form results in high fever, depression, dyspnea, diarrhea and usually death in a few weeks. The chronic form may last years, some animals recover whereas others die. Three forms characterize clinically apparent, chronic glanders, occurring alone or simultaneously: pulmonary glanders, upper respiratory tract disease, and cutaneous glanders. The disease in man is exceptional, but with high mortality when expressed clinically. Man is usually contaminated through close contact with sick solipeds, especially those kept in crowded conditions. Because solipeds in Wakhan have been observed to suffer often upper respiratory tract disease sometimes associated to cutaneous abscesses the WCS team investigated healthy and a few sick horses and donkeys in 2012 and 2013. We used serological investigation

(ELISA) on 115 solipeds sampled in 2012 in the Wakhan Valley, and intrapalpebral mallein test on 122 horses and 109 donkeys in the Wakhan Valley and Big Pamir and. We found no reactors. In addition we also sampled the muco-purulent discharge of three donkeys and one horse showing upper respiratory tract disease compatible with glanders. The laboratory confirmed the absence of *B. mallei* and found instead *Streptococcus equi* ssp *equi* which is the causative agent of strangles, a disease sometimes mistakenly taken for glanders, with similar symptoms but not zoonotic. We have so far found no evidence that the disease exists in horses and donkeys in the Wakhan district.

Other zoonosis

Other zoonotic diseases may be present in the Wakhan district but we have not found clinical evidence of their occurrence in livestock or wildlife and did not do other laboratory investigations. Several of these diseases may even be clinically unapparent in livestock. They include anthrax, bubonic plague, Crimean-Congo Hemorrhagic Fever, cryptosporidiosis, giardiasis, and hydatidosis. These diseases have been documented to occur in the past or in recent times in adjacent districts of Badakhshan (e.g. anthrax, CCHF, cryptosporidiosis, giardiasis), or in the autonomous oblast of Gorno-Badakhshan in Tajikistan (plague, cryptosporidiosis, anthrax) or in Gilgit-Baltistan and Khyber Pakhtunkwa provinces of Pakistan (anthrax, hydatidosis).

Three zoonotic diseases merit further attention. Rabies is enzootic in foxes, wolves, and jackals in the region, although dogs are considered the primary source of human exposure in Afghanistan. This infectious disease causes hundreds of human deaths annually in Afghanistan but we did not hear of any suspicious human mortality following a contact with a rabid-like animal and we have observed and heard of no clinical cases suggestive of rabies in domestic or wild animals in the Wakhan district for the past 10 years. However, we were told by locals of a disease with symptoms evocative of rabies that was killing foxes in Little Pamir in the early 2000's, shortly before we started working in the area. Rabies could occur in Wakhan in the form of irregular epidemics but the disease does not appear enzootic in wild carnivores in the area.

The second disease is the endemic relapsing fever also named tick-borne relapsing fever, which is caused by spirochetes of the genus *Borrelia* and more specifically in the region *B. persica*. The infectious agent is transmitted through bites or fluid of coxal glands of soft ticks of the genus *Ornithodoros* spp. The disease is endemic in the northern third of Pakistan and in western Pamirs of Tajikistan where human cases tend to be sporadic but are increasing in frequency. *Ornithodoros* ticks live hidden in walls, cracks, animal stables where they can fast in infected state for months or years. Ticks transmit the *Borrelia* bacterium from a wild reservoir (typically rodents) to livestock

and humans. The disease in man is characterized by an initial fever that lasts 3-4 days and begins and disappears suddenly. After several days without fever the attacks of fever recur several times. Complications consist of meningitis and some other neurological disorders. We have found *Ornithodoros lahorensis* infesting livestock in several places in the upper Wakhan Valley in December 2006 and *Ornithodoros* spp. was also found in Kharich village (upper Wakhan) by Jens Soelberg, a botanist visiting Wakhan, in August 2010. The disease is known from herders who associate it to the soft tick, which they avoid touching or even removing from their animals when found.

Finally, the clinical examination by WCS team of several thousand livestock during the study period suggests that orf, a disease caused by a parapox virus and affecting in priority sheep and goats is common in Wakhan. We have also seen on rare occasions cutaneous lesions on hands of shepherds that could have been caused by this virus. It is in general a benign self-limiting disease in humans.

Livestock health

Livestock diseases investigated serologically

Horizontal inter-species transmission is a central mechanism in the emergence of diseases in wild-living populations. The probability for a pathogen to cross the species barrier from a 'source' to a 'receptor' species depends on the type of pathogen, on the susceptibility of the receptor and on the rate of efficient direct (from animal to animal) or indirect (via environmental contamination or vector transmission) contacts between the species. The contact rate between the source and the receptor is intimately linked to the relationship between these species and the likelihood of sharing the same habitat.

In mountainous areas, the abundance of domestic animals leads to forced cohabitation between them and their wild counterparts. The spillover of contagious diseases from domestic to wild-living ungulates has been largely reported during the last 25 years with sometimes detrimental effects at population level in rare wild ungulates. Domestic and wild-living ungulates are competitors for food, which results in pasture sharing and, thus, in the transmission of infectious agents, especially indirectly transmitted ones.

Afghan Pamirs and the Hindu Kush still host populations of Marco Polo sheep or argali (*Ovis ammon polii*), Siberian ibex (*Capra sibirica*), and urial (*Ovis orientalis*) yet they are under threat of disappearance due to uncontrolled hunting and presumed competition with livestock for suitable habitat. Although in theory cross-species transmission of diseases between livestock and wild ungulates could operate in both ways, in Afghan Pamirs the risk of population collapse for wild ungulates seems to far exceed the anecdotic likelihood of livestock being impacted by a pathogen indigenous

to wild ruminants. Indeed livestock in Afghan Pamirs are a renewable human resource, quickly replaced in the event of massive mortality such as affecting them during harsh winters, whereas wildlife currently suffers overutilization and competition for food resources, allowing only limited productivity. Any relevant contagious pathogen introduced into such pressurized population could have disastrous effects on the short term. Therefore, within the proposed plan to protect remnant populations of wild ungulates in Wakhan, one may legitimately dispute that wild ungulates pose a significant risk of disease transmission to livestock. Here we report on livestock diseases potentially dangerous to wildlife.

Foot-and-mouth disease

This is an extremely contagious viral (family Picornaviridae) disease of cloven-hoofed domestic and wild animals. It is endemic in most of Asia (including the Middle East), Africa, and South America. There are 7 immunologically distinct serotypes and over 60 subtypes. The disease is endemic in Afghanistan and occurs as an annual epizootic. It has a direct effect on food security as it drastically reduces milk production in cows, reduce their fertility rate and incapacitate breeding bulls and oxen. A new serotype (Asia 1) has been identified in Afghanistan in March 2001, bringing the total of known serotypes to three for the country (i.e. A, O, Asia 1). The virus is very stable at low temperatures and can survive in frozen tissues. It may persist for days to weeks in organic matter under moist and cool temperatures. It is however inactivated on dry surfaces and by UV radiation (sunlight). Transmission primarily occurs by respiratory aerosols and direct or indirect contact with infected animals. Sheep and goats are considered maintenance hosts, and sometimes present very mild signs. Cattle are generally the first species to manifest signs of foot-and-mouth disease (FMD) and are therefore considered 'indicators' of this disease. Recovered or vaccinated cattle exposed to diseased animals can be healthy carriers for 6 to 24 months; sheep can be carriers for 4 to 6 months. Vaccination of livestock was not practiced in the Wakhan district before WCS started vaccinating yak and cattle in 2009. In Big Pamir in September 2008, 51.3% and 75% of the sheep and yak respectively had antibodies against FMD, the prevalence was similar in the upper Wakhan Valley in March-April and summer 2009 with prevalence of 54.0% (95%CI 50.0%-57.9%), 76.2% (95%CI 69.2%-82.0%), and 57.9% (95%CI 46.7%-68.3%) in sheep, cattle and yak, respectively. In September 2008 results of virus neutralization tests performed on samples of two sub-adult domestic yaks displaying clinical foot-and-mouth infection confirmed the presence of serotype Asia 1. We also found reactive neutralizing antibodies on five sheep and four cattle positive with the competitive NS ELISA test in 2009. We confirmed the presence of reactive antibodies against Asia 1 serotype in the four cattle and one sheep, against both serotypes Asia 1 and O in three sheep and against serotype O alone in one sheep. According to Wakhi livestock owners we interviewed in 2006, FMD is a relatively new disease in the Wakhan Valley and

Pamirs. Seemingly the disease appeared in their livestock between 1992 and 1995 when large herds of livestock originating from Panjshir and Badakhshan started using the Wakhan corridor to reach livestock markets of northern Pakistan. Kabul, the traditional marketing outlet for these livestock populations, was no longer accessible, destroyed by the civil war. Arguably contaminated herds moving through the corridor have spread the disease among local livestock. From an historical point of view, the story is believable since Wakhan and Pamirs were renowned for centuries as livestock production areas where non-native domestic animals were seldom introduced. However, from an epidemiological point of view this is a more questionable theory since FMD has been endemic for a long time in neighboring areas of northern Pakistan, north-east Afghanistan and eastern Tajikistan. Nowadays the disease is endemic in livestock in the Wakhan district and because of this and the high stability of the virus in the external environment the FMD virus poses a risk to wild ungulates although their susceptibility remains poorly understood.

Bluetongue

It is an insect-borne viral disease of ruminants, transmitted by *Culicoides* sp. hematophagous midges. Direct transmission between animals is not possible and therefore geographical distribution of the diseases is limited to areas where *Culicoides* vectors occur, roughly in terrestrial areas extending between latitudes N 40° and S 35°. Among domestic animals, clinical disease occurs most often in sheep, resulting in erosions and ulcers of the mucous membranes, dyspnea or lameness from muscle necrosis and inflammation of the coronary band of the foot. Some strains of virus can result in high mortality rates. But bluetongue infection can also be asymptomatic, presumably when it occurs in populations that have been in contact with the virus for long time. The virus belongs to the order Orbivirus and family Reoviridae and 24 serotypes have been identified worldwide. Bluetongue viruses (BTV) are closely related to the viruses of the epizootic hemorrhagic disease (EHD), which is one of the most important diseases of deer in North America. BTV can also cause severe diseases in wild ungulates, including the white-tailed deer (*Odocoileus virginianus*) and the desert bighorn sheep (*Ovis canadensis*). In Big Pamir and the Wakhan Valley 25.0% (95%CI 19.9%-30.9%), 29.1% (95%CI 26.1%-32.4%), 34.0% (95%CI 28.9%-39.4%) of sheep in July-August 2008, July 2011, and October 2012, respectively, tested positive to the VP7 protein of BTV. In the absence of known cross-reactivity with other viruses (e.g. rotavirus), and despite lack of positive results with RT-PCR investigations, it seems that BTV is common in livestock in Wakhan, yet it is debatable whether it actively circulates in Pamirs. Although *Culicoides*, the only known vector of BTV, have colonized a great variety of habitats from sea level to a maximal altitude of 4,100 m in Tibet, their presence in Pamir pastures (between 3,600 and 4,500 m asl on average) is not confirmed. It is also noteworthy that on 31 May 2008, just before leaving to Big Pamir pastures, 56.7% (17/30) of a group of 'sentinel' sheep were

serologically positive to BTV, but only 30% (9/30) were still seropositive on 26 September 2008 at the end of the summer stay in Pamir. Likewise, none of the sero-negative animals prior to spring transhumance seroconverted positive while in altitude pastures, whereas 8 sheep seropositive prior to transhumance converted negative by the end of summer. It is also interesting to note that one animal seroconverted positive between 6 April and 31 May when in the Wakhan Valley (2,800–3,200 m), and before leaving to Pamir. Eventually no circulating viral nucleic acids were detected in tested seropositive sheep (14 prior to and 8 after transhumance). All these observations suggest that BTV might not circulate actively in Pamirs. In such condition it could be the case that wild ungulates living in Pamirs have so far not been exposed to BTV.

Rinderpest

Rinderpest virus (RPV) is classified in the genus *Morbillivirus* in the family *Paramyxoviridae*. The virus is highly contagious and causes an acute to subacute disease of artiodactyls. The disease is characterized by necrosis and erosions in the gastrointestinal tract resulting in severe diarrhea, dehydration, and death. Morbidity and mortality rates often exceed 90%. Recently however, unapparent infections have been more common in cattle. The virus spreads by contact between infected and susceptible animals. Wild ungulates exhibit a wide range of clinical signs, ranging from very severe in kudu (*Tragelaphus strepsiceros*), African buffalo (*Syncerus caffer*), giraffe (*Giraffa camelopardalis*), and eland (*Tragelaphus oryx*) to mild or nonspecific signs in species such as impala (*Aepyceros melampus*). None of the 60 sheep and 20 yaks tested in 2009 showed antibodies against RPV, supporting that the disease is likely absent in the study site. In June 2011, the United Nations FAO confirmed the disease was eradicated globally, making rinderpest only the second disease in history to be fully wiped out, following smallpox.

Peste des Petits Ruminants

Peste des petits ruminants (PPR) is an acute contagious disease of small ruminants, particularly goats caused by a paramyxovirus of the genus *Morbillivirus*. The disease is widespread in Afghanistan and occurs also in Tajikistan, Pakistan and western China. Clinical signs are similar to rinderpest in cattle (the two organisms are closely related) and may include fever, necrotic stomatitis, gastroenteritis, and bronchopneumonia. The morbidity and mortality rates can be up to 100% in severe outbreaks. In milder outbreaks, morbidity is still high but the mortality rate may be closer to 50%. Severity depends upon the susceptibility of the population. Goats are generally more susceptible to PPR than sheep. The disease is also known to be transmitted to wild ungulates and may result in significant mortality. We tested serologically sheep and goats sampled in the Wakhan Valley in July 2011, September-October 2011 and October 2012. We

found prevalence of 1.2% (CI95% 0.4%-3.5%), 1.4% (CI95% 0.5%-4.1%), and 20.7% (CI95% 16.5%-25.6%), respectively. The significantly higher prevalence in 2012 may have resulted from an outbreak of PPR in the Wakhan Valley in spring 2012, with high mortality locally. The epidemiology of the disease in Wakhan is poorly known but could occur as epidemic bursts followed by periods of absence. There is no known carrier state (i.e. the infectious agent surviving in a species without affecting it). Because the disease is responsible episodically of high livestock mortality and has been recently shown to spillover to a range of mountain ungulates species, vaccination of sheep and goats should be seriously considered in the future.

Contagious caprine pleuropneumonia

Contagious caprine pleuropneumonia (CCPP) is a pneumopathic disease caused by the bacterium *Mycoplasma capricolum* subsp. *capripneumoniae* (*Mccp*), which can lead to severe losses in domestic goat populations in the developing world. The morbidity and mortality are very high when the disease affects naïve herds, occasionally reaching 100% and 90%, respectively. For the last 10 years the disease has been confirmed in an increasing number of countries. In 2002, *Mccp* was detected only in five countries (Nepal, Oman, Turkey, United Arab Emirates, Yemen) in Asia. Yet in the past 10 years the disease has spread to new territories, including to the European side of Turkey and Tajikistan. The causative agent has also been detected in other countries such as Pakistan as a result of improved diagnostic tests, although the disease was suspected in this country for long time. We sampled 359 and 89 goats in the Wakhan Valley in summer 2011 and spring 2012. Serological tests with cELISA and latex agglutination test failed to detect a reactor. There is therefore no indication that the disease is present in the district and the upper limit of the 95% CI is <1.0%. In the region the disease has not reached the higher elevation districts but is already present in lower elevation mountainous districts in northern Pakistan, southern Tajikistan, and possibly northern Afghanistan. Although *Mccp* was thought to be a pathogen highly specific to domestic goats it has recently been found to affect captive wild goats (*Capra aegagrus*), urial (*Ovis orientalis*) and Nubian ibex (*Capra ibex nubiana*) in Qatar and semi-captive dorcas gazelles (*Gazella dorcas*) in the United Arab Emirates, with mortality rates ranging between 58% and 90%, depending on the species. The introduction of the disease in Wakhan would have significant deleterious effects on both domestic and wild caprids. Because of the extreme lethality of the disease in this group of ungulates, it is of utmost importance not to allow the introduction into Wakhan of any domestic goat, even clinically healthy, from other districts of Badakhshan or other provinces.

Border Disease / BVD-MD

There are four recognized species of pestivirus (Family Flaviridae): the bovine viral diarrhoea virus 1 (BVDV-1) and 2 (BVDV-2) affecting cattle, border disease virus (BDV; seven serotypes recognized) infecting sheep and goat, and classical swine fever virus (CSFV) affecting swine. Ruminant pestiviruses are not strictly host-specific and antibodies against these viruses have been reported in several domestic and wild *Artiodactyla* species. Affected flocks of sheep are recognized first at lambing time by an increase in the number of barren ewes and in the birth of undersized lambs with excessively hairy and sometimes excessively pigmented fleece. Newborn lambs can display skeletal abnormalities, involuntary muscular tremors, particularly of the trunk and hind legs. The tremors are reduced at rest and exacerbated by purposeful movement. In others, skeletal defects such as dropped pasterns and mandibular brachygnathia may predominate. Affected lambs have a poor survival rate. In survivors, nervous signs gradually disappear within 3–4 mo. Even in the absence of typical hairy-shaker lambs, outbreaks of low fertility in ewes and poor viability and ill looking lambs may be associated with border disease virus infection. Serious disease with high mortality has also been reported in the Pyrenean chamois (*Rupicapra pyrenaica*). The CVDRL-Kabul tested sera of 294 sheep and 11 cattle for antibodies against pestivirus, with a blocking enzyme-linked immunosorbent assay (ELISA; BVD/MD/BD P80). Samples were collected in mid Wakhan Valley in October 2014. The prevalence in sheep and cattle were high, respectively at 47.9% (CI95%: 42.1%-53.8%) and 27.3% (CI95%: 6.0%-60.1%). The results underline that pestiviruses are present in Wakhan and circulate actively among sheep and cattle. The significance of this prevalence values in term of risk posed to wildlife is unknown, but should be considered in case of abnormal mortality in wild ungulates coming in contact with domestic sheep and cattle such as the urial (*Ovis orientalis*).

Other animal diseases

As previously mentioned orf seems to be endemic in Wakhan, affecting primarily domestic sheep and goats. However, in several places in the world the disease is also known to exist as a natural disease in wild artiodactyls. Currently it is not known whether the orf virus(es) circulating among sheep and goats in Wakhan would be a threat to wild ungulates in the area. Anthrax is another disease allegedly present in Wakhan, but to our knowledge never confirmed through laboratory investigations. The disease could pose a threat to wildlife too.

Also, we were told that an eye disease that causes muco-purulent keratoconjunctivitis affects occasionally livestock, and particularly yaks in summer. Unfortunately we never had the opportunity to sample affected animals and investigate the causative

agent(s). Infectious keratoconjunctivitis (IKC) caused by *Mycoplasma conjunctivae* is a highly contagious ocular infection which in the European Alps is common in domestic sheep and goats and causes inflammation of the cornea and conjunctiva in Alpine chamois (*Rupicapra rupicapra*) and Alpine ibex (*Capra ibex*). Mortality in wild ungulates can occasionally reach 30%. Flies are likely to play a central role in interspecific transmission of *M. conjunctivae* on alpine meadows. Studies are required to evaluate the presence of *M. conjunctivae* in Wakhan and to assess the risk of IKC spillover from domestic animals to wildlife.

We have noticed only few cases of skin diseases in livestock, supporting that scabies, a parasitic disease caused by the mite *Sarcoptes scabiei*, and responsible of mortalities in wild ungulates in Central Asia may be uncommon in Wakhan. However, in winter 2015-2016 Marco Polo sheep affected by a skin disease in Big Pamir were reported by villagers to community rangers who could not confirm these cases when visiting the area two months later.

The infectious agents circulating in domestic dogs and cats have not been investigated, yet several of them such as the viruses responsible of rabies and canine distemper may pose a risk to wild carnivores, including to the endangered snow leopard (*Panthera uncia*).

Health facilities and capacities in Wakhan

Human health facilities in Wakhan are still very few, unequipped, and staffed by a small number of poorly trained personnel. In 2003 the Aga Khan Development Network (AKDN) was contracted by the government to implement a health program in the district (and Badakhshan Province at large) and by 2006 was managing a basic health center with outreach in Khandud and a comprehensive referral health center in Ishkashim in the main city of the district adjacent to the west of Wakhan. In addition Orphans Refugee Aid (ORA) International, a relief and development organization based in Germany has been active in Wakhan since 1998, operating a drug rehabilitation program in the province. Since 2003, a small team started a community health training program in cooperation with the Aga Khan Health Services to implement a 'Basic Package of Health Services. ORA was sponsoring a basic health center in Kipkut village in upper Wakhan with an itinerant British physician and one or two nurses.

Ten years later, in 2016, health facilities were slightly more developed and concomitantly the extent of health coverage had improved. The comprehensive health center in Ishkashim would now qualify as a hospital with some diagnostic capabilities and one physician, a midwife and nurses (no dentist). Serious cases would still require

to be transferred to Faizabad, Kabul or under emergency situation and thanks to a local trans-boundary agreement to hospitals in Ishkoshim or Khorog in Tajikistan. The basic health center in Khandud remained very rudimentary with limited supplies but recently got better staffed with a physician, a midwife and several nurses. There is also a health center in Qila-e Panja but not equipped and not staffed. By 2014 ORA and the small facility in Kipkut ceased functioning. A small basic health center supported by AKDN opened in Ptukh (upper Wakhan) with a pharmacist operating also as a nurse and dentist and prescribing medicines, and a midwife. A very positive achievement is that large-scale child immunization (oral polio vaccine, measles, tetanus) operations have been undertaken in the district every year since 2003, including in Pamirs. Still, these remote high altitude areas remain largely neglected from the point of view of health care, with no permanent health facility or staff in Little Pamir. In 2014 AKDN opened a small health center in Shaitatuk, in the Kyrgyz area of Big Pamir where a new building was built and staffed with a midwife and a health worker (physician or nurse?). This center, the first of its kind in Afghan Pamirs has been approved by the Ministry of Health of Afghanistan and has received in 2016 in-kind donations from the government of Kyrgyzstan. Finally AKDN has trained community health workers in clusters of villages in Wakhan Valley who deliver basic curative services and vaccinations including to Kyrgyz in summer and when necessary refer cases to health centers.

In Afghanistan animal health activities are conducted by a range of actors involving state veterinary authorities, private and community paraveterinarians trained by the Dutch Committee for Afghanistan (DCA), a variety of NGOs, and in the recent past foreign military forces. In general, livestock vaccination is one of the main activities of paraveterinarians, a commitment that is achieved with variable efficiency according to the capacity of vaccinators, to the remoteness of targeted areas and the availability of a cold chain and adequate storage facilities. The Aga Khan Foundation promotes veterinary activities in Afghanistan and particularly in the province of Badakhshan. A veterinary health center was established in Ishkashim and staffed with Afghan and Tajik veterinary practitioners. They have been actively vaccinating livestock against a variety of diseases in Ishkashim and Zebak districts of Badakhshan. Yet, until recently little and only sporadic efforts have been devoted to develop animal health programs in the Wakhan district, mainly because of logistical difficulties, the lack of capacity and the chronic difficulty at establishing a reliable cold chain essential to store drugs (and particularly vaccines) before use. A livestock research center supported by AKDN was established in the early 2000's in Khandud but has remained under-equipped and has been active irregularly and according to staff availability. In 2007, WCS has sponsored the training by DCA of the two first paraveterinarians in the area and operationalized two field veterinary units in Abgarch and Kandkhan villages, respectively, in Upper Wakhan. These two paraveterinarians also received training specific to wildlife health. Since 2010 AKDN has sponsored the training of additional paraveterinarians and by the end of 2016, there were eight paraveterinarians and associated field veterinary units in Wakhan in the villages of Qazideh (1), Yamit (1), Khandud (2), Avgarch (1),

Kandkhun (1), Ptukh (1), and Chilkand (1). A veterinary doctor hired from Tajikistan supervises the six paraveterinarians managed by AKDN. Their main duties are to deliver basic curative services, assist livestock parturitions in cases of complication, and undertake vaccinations. Owing to the relative lack of cash in the area and to the reluctance of livestock owners to pay for animal health services, only two of the paraveterinarians can afford to be full-time active at improving animal health; the one in Qazideh, without equipment, is jobless and the others divide their time between animal health work and other livelihood options.

The main livestock vaccinations proposed by paraveterinarians have been sporadically implemented and have targeted clostridial diseases (enterotoxaemia, blackleg, tetanus), and anthrax. Between 2009 and 2014, the WCS One Health team organized with the two paraveterinarians it had mentored the delivery of more than 27,000 vaccinations against foot-and-mouth disease to cattle and yak in upper Wakhan. Finally from 2014 onward, the Ministry of Agriculture in the frame of the National Brucellosis Control Program organized and sponsored with the help of the network of paraveterinarians supervised by AKDN a vaccination program against brucellosis for sheep, goats and cattle (Rev 1 vaccine for sheep and goats and S 19 vaccine for cattle), between the villages of Ftur in Lower Wakhan and Kret in Upper Wakhan. The number of animals vaccinated in 2014 is unknown to us but available at the animal health office of the branch of the Ministry of Agriculture in Faizabad. In 2015, 2,835 sheep and lambs, 2,165 goats and kids, and 1,095 cows and calves were vaccinated. In 2016 vaccinations were continued between Ftur and Kret and extended to the western part of Big Pamir (Wakhi livestock) and included 4,353 sheep and lambs, 3,049 goats and kids, and 1,320 cattle and calves

Appendix 1

Main zoonotic and infectious diseases dangerous to wildlife in the Wakhan National Park, Badakhshan Province, Afghanistan

Disease name	Zoonosis	Serological prevalence in livestock in WNP	Year of evaluation	Risk of clinical disease in wildlife of WNP
Anthrax	Yes	Not investigated		Potentially high but locally
Border disease / BVDV-MD	No	Sheep (>42%); Cattle (>6%)	2014	High if host susceptible
Bovine tuberculosis	Yes	Cattle (<0.9% - not detected)	2010, 2011, 2012	Currently very low
Bluetongue	No	Shoat (>19.9%)	2008, 2011, 2012	High if host susceptible and presence of the midge vector
Brucellosis	Yes	Sheep (<0.15%); Cattle (<1.4%); Yak (<1.1%)	2008 - 2012	Currently very low
Canine distemper	No	Not investigated in domestic carnivores		Potentially high in carnivores
Contagious caprine pleuropneumonia	No	Goat (<1% - not detected)	2011, 2012	Currently very low
Crimean-Congo hemorrhagic fever	Yes	Not investigated in livestock		Unknown
Cryptosporidiosis	Yes	Not investigated in livestock		Unknown
Foot-and-mouth disease	Yes (minor)	Sheep (>50%); Cattle (>69.2%); Yak (>46.7%)	2008, 2009	High if host susceptible
Giardiasis	Yes	Not investigated in livestock		Unknown
Glanders	Yes	Horse/Donkey (<3.5% - not detected)	2012, 2013	Currently very low
Hydatidosis	Yes	Not investigated in domestic carnivores and livestock		Low in rodents and wild herbivores
Infectious keratoconjunctivitis	No	Not investigated in domestic carnivores and livestock		Potentially high if present
Orf	Yes	Presumably high (clinical)	2006 - 2014	Potentially high
Peste des petits ruminants	No	Shoats (>16.5%)	2011, 2012	High if outbreak
Q fever	Yes	Shoats (>6.3%)	2008, 2012	Low in wildlife in general
Rabies	Yes	Very low to inexistent (clinical)	2006 - 2014	High in carnivores if outbreak
Rinderpest	No	Inexistent (eradicated worldwide)	2009	Inexistent
Scabies	Yes (minor)	Unknown	-	Potentially high if present
Tick-borne relapsing fever	Yes	Unknown	-	Unknown
Toxoplasmosis	Yes	Shoats (>10.0%)	2008	Low in wildlife in general