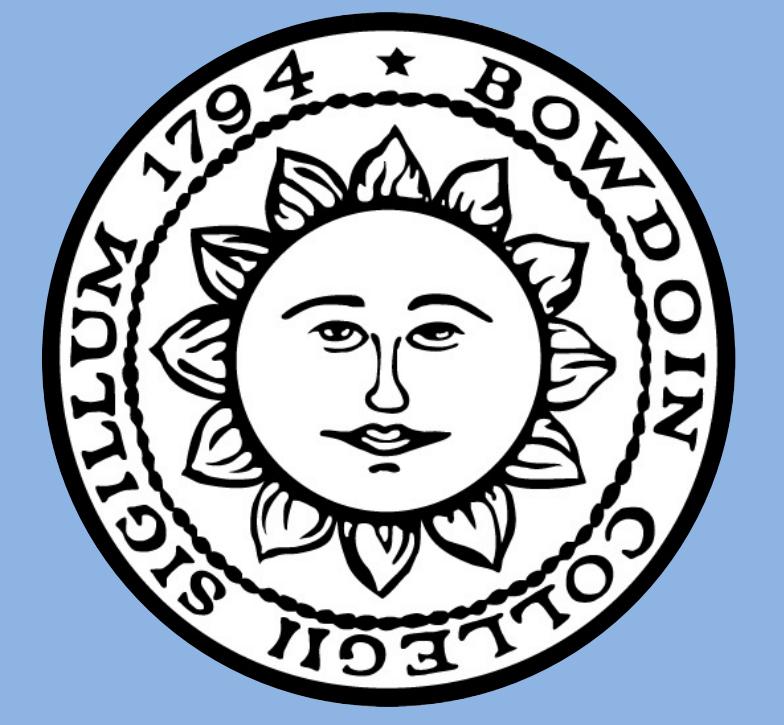


Zoonotic Disease Risk Assessment in Bardia National Park, Nepal

CENTER FOR MOLECULAR DYNAMICS - NEPAL



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Introduction

Emerging infectious diseases (EIDs) pose substantial threats to the health of animals, people and economies globally. Zoonotic pathogens shared with wild or domestic animals account for the majority of EIDs, and viruses comprise 75% of these emerging and re-emerging pathogens. Building on the surveillance activities to date, Center for Molecular Dynamics is focusing on setting up surveillance systems in locations where environments and market systems are changing in ways that are conducive to the spillover of pathogens from animals to people. Despite greater recognition of emerging zoonoses, the exact mechanisms of viral spill over and transmission from animals to humans are poorly understood. This project aims to provide a better understanding of the drivers and host-pathogen dynamics, including which human behaviors and practices increase risk, and under what circumstances these behaviors facilitate spillover of zoonotic pathogens.



Bardia National Park (BNP) stands as the largest national park in Nepal, encompassing a buffer zone that is home to over 130,000 people across multiple municipalities and districts. These communities' livelihoods are closely intertwined with their interactions with local wildlife, and the park's popularity among tourists elevates the global risk of zoonotic transmission. The park's diverse ecosystem, nurtured by the Babai and Karnali Rivers, hosts a rich variety of wildlife, including Bengal tigers, Indian elephants, spotted deer, and numerous bird species, attracting birdwatching enthusiasts.

Bardia was chosen for this research since residents of the buffer zone rely on a diverse range of park resources, including grass, fodder, timber, firewood, thatch, medicinal herbs, and more, shaping their livelihood strategies accordingly. The frequency of wildlife encroaching into villages and causing damage to crops, infrastructure, and, regrettably, even human casualties has been on the rise. Consequently, these incidents have led to an escalation in human-wildlife interactions. These interactions, coupled with the influx of national and international tourists, elevate the potential for the transmission of zoonotic diseases between wildlife and humans.



Project Goals & Questions

The project aims to address the significant threats posed by emerging infectious diseases (EIDs) to global health, economies, and ecosystems. These diseases, often zoonotic in nature, with viruses accounting for a majority, have historically been managed reactively. The project seeks to shift towards a proactive approach, building on initiatives like USAID's Pandemic Influenza and the Emerging Threats Program and the PREDICT project. By focusing on pathogen discovery and surveillance in changing environments and market systems, particularly in Nepal, the research aims to shed light on viral spill over, transmission dynamics, and potential reservoirs. The study's general objective is to establish an active zoonotic disease surveillance and mitigation program, beginning with a pilot study in Bardia National Park. Specific objectives include generating baseline data on prevalent zoonotic diseases, engaging stakeholders, collecting non-invasive samples for analysis, understanding and identifying risks of disease spillover, assessing potential reverse spillover, and conducting capacity building activities. This research is crucial for a more proactive and effective approach to preventing future epidemics.

Acknowledgements

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Methods & Materials

PHASE 1: PROJECT INCEPTION

PHASE 2: TRAINING OF FIELD STAFF IN SAMPLE COLLECTION AND SURVEY ACTIVITIES

Prior to mobilization, field staff from CMDN were trained on several activities as samples could potentially contain highly infectious pathogens. Staff were thoroughly trained on the following topics:

- Biosafety and biosecurity practices:** usage of appropriate and adequate personal protective equipment (PPE)
- Maintaining appropriate cold chain:** proper storage of (stored in liquid nitrogen (LN2) tanks that have a temperature of -190 Celsius)
- Waste management:** waste generated treated, sanitized & discarded with extreme caution.

PHASE 3: LOCAL STAKEHOLDER ENGAGEMENT AND INITIAL SCREENING

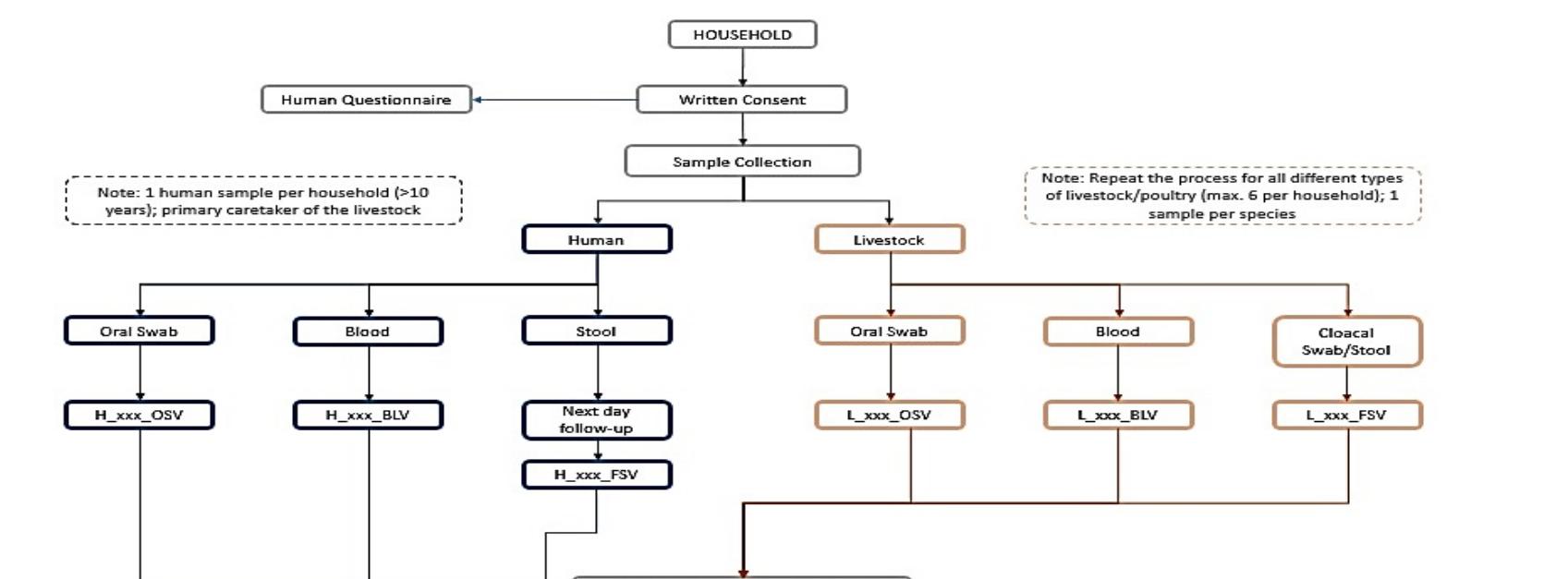


Project coordinators and field leads engaged in active communication with local authorities and community leaders to provide them with comprehensive information about our operations within their respective areas.

PHASE 4: SAMPLING



Thakurdwara's Ward Number 9 was chosen as the study site through a lottery-based selection method. A two-stage cluster sampling approach was employed to gather samples from both the human and livestock populations. Initially, the CMDN team gathered data from the local Ward Offices and conducted a screening survey to distinguish households with and without livestock, with the latter being excluded from the study. Subsequently, a Simple Random Sampling (SRS) technique was employed to randomly select households with livestock to be included in the study. Two teams, comprised of CMDN personnel and local recruits, executed the sampling and survey activities concurrently.



PHASE 5 – LABORATORY PROCESSING AND DATA ANALYSIS

All samples (100 humans, 289 livestock and 14 non-invasive wildlife feces) collected were sorted and processed at CMDN's lab in Kathmandu and screened for the 10 pathogens (6 viruses: Influenza virus, Rhabdovirus, Paramyxovirus, Flavivirus, Coronavirus and Filovirus & 4 bacteria: Salmonella, Brucella, Brucella, Campylobacter, Mycobacteria) using polymerase chain reaction (PCR). Laboratory and survey data was processed and analyzed looking for potential spillover activities and transmission dynamics.

Results & Discussion

All samples collected were screened for 6 viral families—corona, paramyxo-, rhabdo-, influenza, filo- and flavi-viruses and 4 bacterial genera—salmonella, brucella, campylobacter, and mycobacteria—these pathogens were selected based on the known zoonotic disease risks association

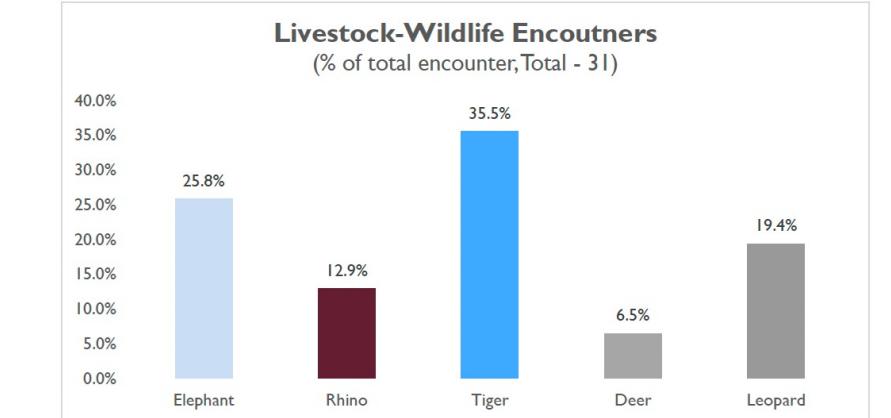
Overall pathogen profile:

Salmonella, mycobacteria, and campylobacter were the only common pathogens among all three (humans, livestock, and wildlife). Brucella was the additional common bacteria detected in both livestock and wildlife. Filovirus, paramyxovirus, and rhabdovirus were detected in humans as well as livestock. Influenza virus was the only pathogen detected in livestock alone.

	Pathogens	Households detected (N)
Humans	Rhabdovirus	13
	Paramyxovirus	2
	Filovirus	5
Livestock	Salmonella	1
	Campylobacter	97
	Mycobacteria	1
Wildlife	Influenza	9
	Rhabdovirus	5
	Paramyxovirus	1
Bacteria	Filovirus	6
	Brucella	3
	Salmonella	2
Virus	Campylobacter	97
	Mycobacteria	15
	Brucella	4
Mycobacteria	Salmonella	2
	Campylobacter	2
	Mycobacteria	4

Given the widespread presence of campylobacter across all three categories and its prevalence, sequencing and phylogenetic analysis were carried out. The results has several potential zoonotic impacts:

- Increased Zoonotic Potential:** Diverse strains in each host group heighten the risk of zoonotic transmission, exposing humans to a wider range of Campylobacter variants.
- Antibiotic Resistance Transfer:** Genetic diversity may lead to varying antibiotic resistance profiles, potentially enabling the transfer of resistance genes between host groups, posing challenges for treatment.
- Transmission Dynamics:** Genetic distances indicate distinct Campylobacter transmission patterns in different hosts, informing targeted control measures for reducing zoonotic risk.
- Emergence of Novel Strains:** Significant genetic diversity may give rise to new Campylobacter strains with altered virulence factors, potentially impacting infection severity.
- Host Adaptation:** Genetic disparities suggest varying degrees of host adaptation, shedding light on cross-species transmission risks.
- One Health Implications:** Campylobacter's presence in diverse hosts underscores the importance of a One Health approach, emphasizing collaborative efforts across human, animal, and environmental health sectors to address zoonotic disease risks.



Risk assessment

- Survey of all 100 households revealed the community lacking several key risks associated with zoonotic disease transmission
- Less than 50% of the houses had permanent structure, 20% had no proper toilets; 92% of the houses used uncovered well as one of their sources of drinking water; people's knowledge or education did not have an impact on mitigating the risk factors
- Campylobacter was the most prevalent pathogen (97% of the household – humans and animals) – is a major food and water-borne pathogen
- Only bacterial pathogens were detected in wildlife samples.

References

USAID BIODIVERSITY (JAL JANGAL) Preliminary Report
Establishing a One Health based active zoonotic disease surveillance in and around Bardia National Park - a pilot study