

Final Evaluation Report

Your Details	
Full Name	Hira Fatima
Project Title	Assessing the status and distribution of wolves in Pakistan using genetics
Application ID	25854-1
Date of this Report	7-10-2019

1. Indicate the level of achievement of the project's original objectives and include any relevant comments on factors affecting this.

Objective	Not achieved	Partially achieved	Fully achieved	Comments
Conduct a genetic survey of wolves across major regions of Pakistan				Out of the sampling locations in northern Pakistan, we were able to collect and sequence ~200 scats. We confirmed wolf presence in all the major regions sampled.
Provides estimates of genetic diversity and potential gene flow to adjacent wolf populations within Pakistan				With sequencing the Cytochrome B and D loop region of the mitochondria, we were able to only study the genetic diversity (in terms of maternal lineage diversity) of wolves from each region. Assessing potential gene flow to other wolf population would involve using microsatellites on our known wolf extracted DNA samples.
Provide a clearer picture of the distribution of the Indian and Himalayan maternal wolf lineages				With the D-loop region sequenced from our wolf samples, we were able to further distinguish the extent of the Himalayan and Indian wolf maternal lineages.

2. Describe the three most important outcomes of your project.

a). Updates on wolf lineage distribution in Pakistan – we were able to genetically confirm over 60 scat samples as wolves from all the regions we sampled (South Waziristan, Chitral, various regions in Gilgit-Baltistan, Swat Valley). We did not find any Indian or Tibetan maternal lineage haplotypes in these samples. This may suggest that the region along the Indian-Pakistan border (where it transitions from the Karakoram to the Tibetan plateau) might represent the extent of the Tibetan wolf maternal lineage.

b). Baseline data on the carnivore communities – some rare and endangered species were confirmed to be present in different national parks in Pakistan. We genetically confirmed snow leopard in different valleys of Gilgit-Baltistan and lynx in Chitral National Park in Khyber-Pakhtunkhwa.

c). Collaboration – this project led to many collaborations with other researchers studying related fauna. We are collaborating with a PhD student from Bioresource Research Center (BRC) in Islamabad to combine efforts on a longer termed project on wolf genetics in Pakistan. This Rufford project's results have been incorporated into two manuscripts so far – one on wolves in South Waziristan (lead by a master's

student studying wolf-human conflict) and one on the phylogenetic status, diet and distribution of the Kashmir Hill red fox (currently in review). The Kashmir hill fox paper utilised our scat samples that were identified as red fox (n=37) and we conducted additionally analyses on these red fox samples to provide more insight into red foxes in Pakistan.

The tissue and blood samples that we collected will be added to a large-scale study on evolutionary history of wolves in South and Central Asia using whole genome sequencing, in collaboration with PhD candidate Lauren Hennelly from University of California, Davis. Lastly, we are collaborating with WWF-Pakistan on an outreach film on wolves in Gilgit Baltistan.

The two manuscript titles are below:

Hamid A, T Mahmood, H Fatima, LM Hennelly, R Akrim, A Hussain, M Waseem. 2019 (in press). Origin, ecology, and human conflict of grey wolf (*Canis lupus*) in Suleiman Range, South Waziristan, Pakistan. *Mammalia*

Mahmood T, I Rafaqat, LM Hennelly, H Fatima, F Akrim, MR Khan, A Hamid, S Andleeb, IU Zaman. In review. Phylogenetic status, diet composition, and distribution of the Kashmir red fox inhabiting Ayubia National Park, Pakistan. *Animal Biology*

3. Explain any unforeseen difficulties that arose during the project and how these were tackled.

Delay in the project update was observed mainly due to collection delays. Additionally, shipment delays (due to distance in samples collection [field sites] and processing unit [UC Davis]) was the reason. Beside these difficulties we were all good.

4. Describe the involvement of local communities and how they have benefited from the project.

We primarily worked with the forest department staff, professors from Gilgit Baltistan, and PhD/Master students from PMAS Arid Agriculture University. Therefore, we did not directly work with local communities on this project yet worked with a large community of conservationists from NGOs, universities, and forest department staff.

5. Are there any plans to continue this work?

We are currently collaborating with a PhD student from the Bioresource Research Center to work together on a joint project to improve our understanding of wolf genetics in Pakistan. This will involve combining our wolf samples with this PhD student's efforts to obtain a broader understanding of wolf maternal lineage diversity across Pakistan. Additionally, the blood and tissue samples will be shipped to UC Davis Veterinary Genetics Laboratory to obtain genomic data, as part of a PhD project of a Rufford project collaborator (Lauren Hennelly). Therefore, the samples collected will be further used for scientific research.

6. How do you plan to share the results of your work with others?

Our results will be published in peer-reviewed scientific journals and will be presented at conferences (such as the Pakistan Congress of Zoology). In addition, we are collaborating with WWF-Pakistan on creating an awareness video on wolves in Gilgit Baltistan.

7. Looking ahead, what do you feel are the important next steps?

The next steps towards understanding wolf genetics in Pakistan would involve doing additional sequencing of other parts of the genome to investigate the genomic distinctiveness and connectivity among wolf populations, as well as increase sampling effort to other parts of the wolf's range in Pakistan. Furthermore, disseminating these findings to the public in Pakistan and scientific researchers would be an important next step. There are an increasing number of reports of illegal wolf trafficking in Pakistan, in which wolves are caught and sold as pets in major cities. The knowledge of how wolf populations vary across the country can aid in distinguishing what region trafficked wolves are being captured from. More broadly, a focused awareness and educational program on the endangered wolves of Pakistan is needed to conserve the remaining populations.

8. Did you use The Rufford Foundation logo in any materials produced in relation to this project? Did the Foundation receive any publicity during the course of your work?

We acknowledged the Rufford Foundation grant funding in the acknowledgements of the red fox paper. We plan on using the Rufford Foundation logo for conference talks and outreach material associated with findings from our research.

9. Provide a full list of all the members of your team and their role in the project.

Dr. Tariq Mahmood - Assistant Professor at PMAS Arid Agriculture University in Rawalpindi, Pakistan. Advisor of Hira Fatima.

Dr. Muhammad Zafar Khan – Assistant Professor in the Department of Environmental Sciences of Karakoram International University, Gilgit. Dr. Khan assisted with sample collection in Gilgit-Baltistan region.

Yasir Abbas – Ecologist at Central Karakoram National Park. Mr. Abbas assisted with sample collection efforts in Karakoram National Park.

Hameed Wazir - Masters student at PMAS Arid Agriculture University studying distribution and livestock depredation of wolf in South Waziristan. He assisted with sample collection and the genetic results contributed to his master's thesis.

Raza Khan – Masters student studying wolf ecology and diet in Swat Valley, Pakistan. Raza assisted with sample collection, which the results will contribute to his master's thesis on wolf ecology in Swat Valley.

Mehdi Sadpara – Forest ranger in Deosai National Park, Gilgit-Baltistan, Pakistan. He assisted with sample collection of carnivore scats.

Dr. Ben Sacks – Directory of the Mammalian Ecology and Conservation Unit (MECU) of the UC Davis Veterinary Genetics Laboratory. Dr. Sacks assisted with sequencing logistics and the MECU hosted Hira Fatima during 2018.

Lauren Hennelly – Ph. D student in the Mammalian Ecology and Conservation Unit at University of California, Davis. Lauren assisted with sequencing the carnivore scat samples in the MECU genetics lab at UC Davis.

Hussain Ali Sukhera – Director of Lahore Zoo. Sukhera assisted with sample collection.

10. Any other comments?

I would like to thank you Rufford on behalf of my entire team. We had an amazing experience, and your financial assistance made this journey possible. Believe me It wouldn't have been done without you guys, so I am grateful.



UNIVERSITY OF OKARA
WEBINAR ON
Unraveling the History of Wolves in South Asia to Inform Conservation

Under Supervision
Prof. Dr. M. Zakria Zakar
Vice Chancellor, University of Okara

Guest Speaker
Lauren Mae Hennelly
(Ph.D Scholar) Mammalian Ecology and Conservation Unit, University of California, Davis, USA

Dated:
1st Feb. 2022

Time: 10 :00 a.m.
Venue: Auditorium

ORGANIZERS

Mudasser Hassan
Deputy Director Wildlife (Head Quarter)
Punjab Wildlife & Paks Department

Dr. Muddasir H. Abbasi
HOD Dept. Wildlife & Ecology
University of Okara

Prof. Dr. Muhammad Wajid
Director, PAZ & SAB
University of Okara

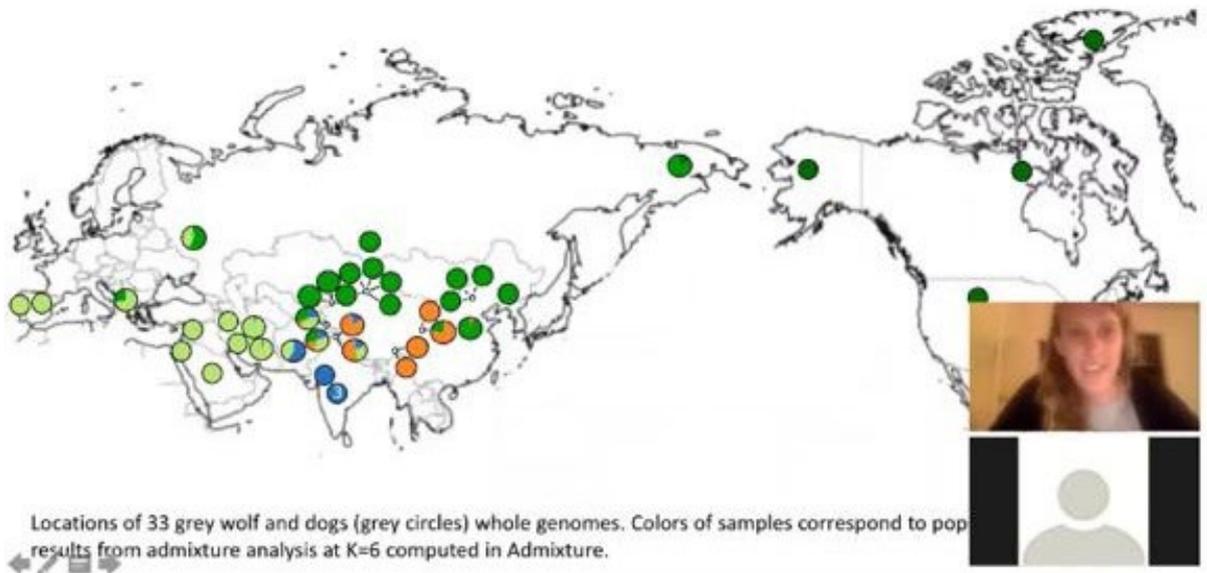
Dr. Muhammad Idnan
Lecturer Wildlife & Ecology
University of Okara

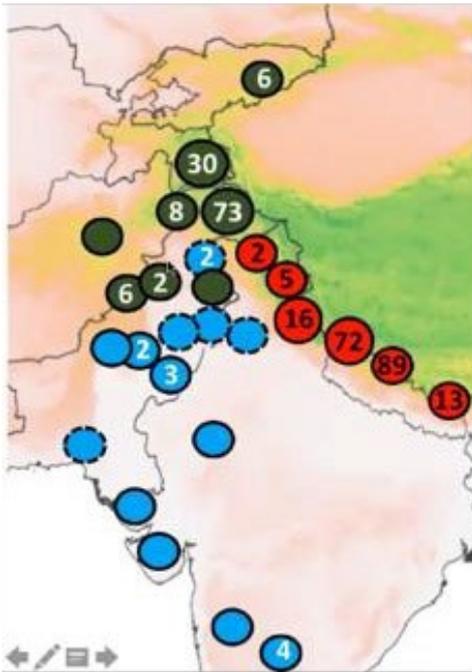
Dr. Hira Fatima
IPF Fellow
University of Okara

Punjab Wildlife & Paks Department | Department of Wildlife & Ecology, UO | Institute Pure and Applied Zoology, UO



Nuclear Genome results





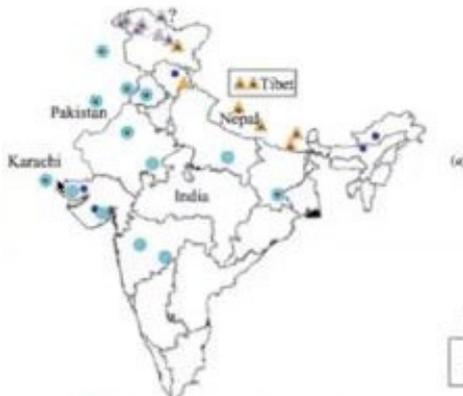
Contact Zones – mtDNA distribution

- Holarctic maternal lineage
- Tibetan maternal lineage
- Indian maternal lineage
- Derived from museum

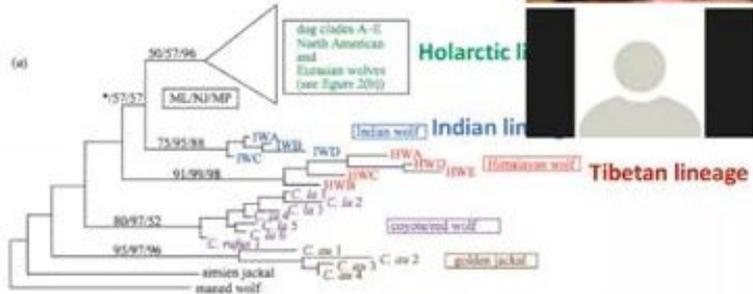


Sharma et al. 2004, Joshi et al. 2020, Werhahn et al. 2020

Tibetan and Indian wolves were actually evolutionarily distinct and old lineages



- *C. l. pallipes*—Indian grey wolf ($n = 45$)
- ▲ *C. l. chanco*—Himalayan haplotype ($n = 16$)
- ▲ *C. l. chanco*—wolf-dog haplotype ($n = 7$)
- *C. familiaris*—feral dogs ($n = 24$)
- museum specimen ($n = 20$)



Sharma et al. 2004



Hira Fatima (University of Okara) , **Ghulam Sarwar** (University of the Punjab), **Tariq Mahmood** (PMAS Arid Agriculture University), **Yasir Rizvi** (Central Karakoram National Park), **Fakhar Abbas** (Bioresource Research Center), **Abdul Majid Khan** (University of the Punjab), **Muhammad Tahir Waseem** (University of the Punjab), **Abdul Hamid** (PMAS Arid Agriculture University), **Muhammad Waseem** (WWF-Pakistan), **Hamera Aisha** (WWF-Pakistan), **Muhammad Ghulam** Baltistan Wildlife Conservation and Development Organization