MOLECULAR MARKERS AND FEATHER MICROSTRUCTURES OF SELECT TRADED AVIAN SPECIES IN ASSAM

Ph D Thesis submitted to the

BHARATHIAR UNIVERSITY, COIMBATORE

For the award of degree of

DOCTOR OF PHILOSOPHY IN ZOOLOGY

By

SWAPNA DEVI RAY

(Reg. No: 2018R0206)

Under the Guidance of

Dr. RAM PRATAP SINGH

Ex-Senior Scientist (SACON)

Associate Professor and HoD, Department of Life Science

Central University of South Bihar, Gaya, Bihar-824236

Under the Co-guidance of

Dr. P. PRAMOD

Senior Principal Scientist, SACON

SÁLIM ALI CENTRE FOR ORNITHOLOGY AND NATURAL HISTORY (SACON)

ANAIKATTY, COIMBATORE - 641 108

TAMIL NADU, INDIA

JANUARY 2023

Certificate

CERTIFICATE

This is to certify that the thesis entitled "**MOLECULAR MARKERS AND FEATHER MICROSTRUCTURES OF SELECT TRADED AVIAN SPECIES IN ASSAM**" submitted to the Bharathiar University, in partial fulfillment of the requirements for the award of the degree of Doctor of Philosophy in **Zoology** is a record of original research work done by **Ms. SWAPNA DEVI RAY** during the period **October, 2018 – January, 2023** of her research in the Avian Physiology and Genetics Division at **Sálim Ali Centre for Ornithology and Natural History Anaikatty, Coimbatore, Tamil Nadu - 641108** under my supervision and guidance, and the thesis has not formed the basis for the award of any Degree/Diploma/Associateship/Fellowship or other similar title of any candidate of any university.

Countersigned

Director SACON

निदेशक प्रभारी / Director in-charge
सालिम अली पक्षिविज्ञान एवं प्रकृति विज्ञान केन्द्र
Sálim Ali Centre for Ornithology and Natural History
आनैकट्टी, कोयम्बत्तूर / Anaikatty (Post), Coimbatore - 641 108

Signature of the Research Supervisor

(Dr. Ram Pratap Singh)

Dr. Ram Pratap Singh
Associate Professor & Head
Department of Life Science
Central University of South Bihar

Signature of the Research Co-Supervisor

(Dr. P. Pramod)

डॉ. पी प्रमोद / Dr. P. Pramod
वरिष्ठ प्रधान वैज्ञानिक/Senior Principal Scientist
सालिम अली पक्षिविज्ञान एवं प्रकृति विज्ञान केन्द्र
Sálim Ali Centre for Ornithology and Natural History
आनैकट्टी, कोयम्बत्तूर/Anaikatty, Coimbatore – 641 108

Declaration

DECLARATION

I, **Ms. SWAPNA DEVI RAY** hereby declare that the thesis, entitled **"MOLECULAR MARKERS AND FEATHER MICROSTRUCTURES OF SELECT TRADED AVIAN SPECIES IN ASSAM"**, submitted to the Bharathiar University for the award of the degree of **DOCTOR OF PHILOSOPHY IN ZOOLOGY** is a record of original and independent research work done by me during **October 2018 –January 2023** under the supervision and guidance of Dr. Ram Pratap Singh, ex-senior Scientist, SACON, Associate Professor & Head, Department of Life Science, School of Earth, Biological and Environmental Sciences, Central University of South Bihar, Gaya - 824236, Bihar and under the co-supervision of Dr. P. Pramod, Senior Principal Scientist, SACON, Anaikatty, Coimbatore, Tamil Nadu-641108, and it has not previously formed the basis for the award of any Degree/ Diploma/ Associateship/ Fellowship or other similar title of any candidate of any university.

Signature of the candidate

(Swapna Devi Ray)

Certificate of Genuineness of the Publication

CERTIFICATE OF GENUINENESS OF THE PUBLICATION

This is to certify that the Ph.D. candidate **Ms. SWAPNA DEVI RAY** working under my supervision has published a research article in the refereed journal named **Journal of Threatened Taxa** with Vol. No. **14,** Issue No. **6**, Page Nos. **21161–21169** and year of publication **2022** published by **Wildlife Information Liaison Development Society, Coimbatore**. The contents of the publication incorporate part of the results presented in her thesis.

Countersigned

Director SACON

डॉ. एस. मुरलीधरन / Dr. S. Muralidharan
निदेशक प्रभारी / Director in-charge
सालिम अली पक्षिविज्ञान एवं प्रकृति विज्ञान केन्द्र
Sálim Ali Centre for Ornithology and Natural History
आनैकट्टी, कोयम्बत्तूर / Anaikatty (Post), Coimbatore - 641 108

Signature of the Research Supervisor

Dr. Ram Pratap Singh
Associate Professor & Head
Department of Life Science

(Dr. Ram Pratap Singh)

Signature of the Research Co-Supervisor

(Dr. P. Pramod)

डॉ. पी प्रमोद / Dr. P. Pramod
वरिष्ठ प्रधान वैज्ञानिक/Senior Principal Scientist
सालिम अली पक्षिविज्ञान एवं प्रकृति विज्ञान केन्द्र
Sálim Ali Centre for Ornithology and Natural History
आनैकट्टी, कोयम्बत्तूर/Anaikatty, Coimbatore - 641 108

CERTIFICATE OF GENUINENESS OF THE PUBLICATION

This is to certify that the Ph.D. candidate **Ms. SWAPNA DEVI RAY** working under my supervision has published a research article in the refereed journal named **Biologia** with Vol. No. , Issue No. , Page Nos. and year of publication **2023** published by **Springer Nature**. The contents of the publication incorporate part of the results presented in her thesis.

Countersigned

Director SACON

निदेशक प्रभारी / Director in-charge
सालिम अली पक्षिविज्ञान एवं प्रकृति विज्ञान केन्द्र
Sálim Ali Centre for Ornithology and Natural History
आनैकट्टी, कोयम्बत्तूर / Anaikatty (Post), Coimbatore – 641 108

Signature of the Research Supervisor

(Dr. Ram Pratap Singh)

Dr. Ram Pratap Singh
Associate Professor & Head
Department of Life Science
Central University of South Bihar

Signature of the Research Co-Supervisor

(Dr. P. Pramod)

डॉ. पी प्रमोद / Dr. P. Pramod
वरिष्ठ प्रधान वैज्ञानिक/Senior Principal Scientist
सालिम अली पक्षिविज्ञान एवं प्रकृति विज्ञान केन्द्र
Sálim Ali Centre for Ornithology and Natural History
आनैकट्टी, कोयम्बत्तूर/Anaikatty, Coimbatore – 641 108

Certificate of Plagiarism Check

பாரதியார் பல்கலைக்கழகம்
BHARATHIAR UNIVERSITY
COIMBATORE – 641 046, TAMILNADU, INDIA

State University | Re-Accredited with "A" Grade by NAAC | Ranked 15th among Indian Universities by MHRD-NIRF

CERTIFICATE OF PLAGIARISM CHECK

1	Name of the Research Scholar	SWAPNA DEVI RAY
2	Course of study	M.Phil., / Ph.D.,
3	Title of the Thesis / Dissertation	MOLECULAR MARKERS AND FEATHER MICROSTRUCTURES OF SELECT TRADED AVIAN SPECIES IN ASSAM.
4	Name of the Supervisor	Dr. RAM PRATAP SINGH
5	Department / Institution/ Research Centre	Zoology Department, Bharathiar University. Avian Physiology and Genetics Division, Sálim Ali Centre for Ornithology and Natural History Anaikatty, Coimbatore, Tamil Nadu-641108, India
6	% of Similarity of content Identified	03 %
7	Acceptable Maximum Limit	10 %
8	Software Used	URKUND
9	Date of verification	26/12/2022

Report on plagiarism check, items with % of similarity is attached

Signature of the Supervisor
(Seal)

Dr. Ram Pratap Singh
Associate Professor
Department of Zoology
Bharathiar University
Coimbatore - 641 046.

Signature of the Researcher

Head of the Department
(Seal)

डॉ. पी. आर. अरुन / Dr. P. R. Arun
वरिष्ठ प्रधान वैज्ञानिक / Senior Principal Scientist
सालिम अली पक्षिविज्ञान एवं प्रकृति विज्ञान केन्द्र
Salim Ali Centre for Ornithology and Natural History
Anaikatty (P.O.), Coimbatore – 641 108

University Librarian (BU)
(Seal)

University Librarian
Arignar Anna Central Library
Bharathiar University
Coimbatore - 641 046.

Director i/c
Center for Research & Evaluation (BU)
(Seal)

Ouriginal

Document Information

Analyzed document	Swapna Devi Ray.pdf (D154532511)
Submitted	12/26/2022 8:31:00 AM
Submitted by	
Submitter email	buaacl.urkund@gmail.com
Similarity	3%
Analysis address	bhauni.urkund.buaacl bhauni@analysis.urkund.com

Sources included in the report

SA **Swapna_PhD - Thesis_Introduction_18112022.RPS.docx**
Document Swapna_PhD - Thesis_Introduction_18112022.RPS.docx (D154312780)

SA **Swapna_PhD - Study Area_19112022.RPS.docx**
Document Swapna_PhD - Study Area_19112022.RPS.docx (D154313062)

SA **Swapna_PhD - Chapter 3_02122022.RPS.docx**
Document Swapna_PhD - Chapter 3_02122022.RPS.docx (D154312959)

SA **Swapna_PhD - Chapter-4_11122022.RPS.docx**
Document Swapna_PhD - Chapter-4_11122022.RPS.docx (D154312985)

SA **SWAPNA_Chapter-5_20122022.docx**
Document SWAPNA_Chapter-5_20122022.docx (D154312992)

Entire Document

Page 1 of 68

Acknowledgement

ACKNOWLEDGEMENT

This present doctoral research work is the outcome of my last four years, which gives me immense pleasure to those who were associated with me and encouraged, motivated and also inspired me during these times. This work is a part of a larger project titled "*Establishment of National Avian Forensic Laboratory at SACON for National Certification to Prevent Illegal Trafficking of Birds*" which was funded by Ministry of Environment, Forest and Climate Change (MoEFCC).

First of all, I would like to acknowledge my parents Mr. Suddha Nath Ray and Mrs. Anima Rajbonshi; who recklessly planted the seed of dream inside me to earn a doctorate degree in the field of my interest. During these whole times they become the "*Backbone*" of who actually I am today. Without their support, I could not have able to complete my work.

Without a supervisor no work can be done in the right track. I would like to sincerely acknowledge my supervisor Dr. Ram Pratap Singh and co-supervisor Dr. P. Pramod for giving me such an emerging platform of research and for their immense support and guidance during this whole time. I am sincerely thankful for the work freedom they provided to me. Thank you for being the "*Guiding Light*" for me.

I would like to convey my sincere thanks to Dr. Goldin Quadros for always being there in the time of need during my research time exactly like the "*Philosopher's Stone*". Without his support and guidance, I could not have completed my work.

I would also like to thanks Dr. Sanjeev Kumar Sharma, Prateek Dey and Indrani Sarkar with whom I have worked for three years in the same project. Thank you for helping me out in the time of need.

I am deeply grateful to Assam Forest Department and Assam State Biodiversity Board for providing necessary permissions. I am also thankful to Aaranyak and their family for their support; specially Dr. Jyoti Das and Mr. Arif Hussain for their immense help during my field work. I am also grateful to my field assistance Mr. Niren Singh, Mr. Bijay Panika, Mr. Pravat Kalita, Mr. Nabojyoti Das, Mr. Randeep Barman, Mr. Anupam Ray for their cooperation in field.

My sincere thanks to Jeevith, Andrew, Jaydeep, Priyanka, Gautam, Rajesh, Nitheen, Siddhesh, Prathamesh, Durgesh, Ashique and all SACON's fellow researchers for encouraging me and always being there for any help. Brother Anoop, Shiba, Suresh, Baburao was there exactly like the elders to guide me and to discuss regarding any confusions related to my research work. I would also like to thanks whole SACON family for their help and support.

I am also deeply grateful to Mohammad Ibrahim, Julfia Begum and Devika i.e., to the whole SACON Envis team for their immense help and support during my Ph.D. Work. Especially to Mohammad Ibrahim brother, who helped me preparing the maps and with other advance software. Vighneswaran, I don't know how much thanks I owe you; without all of your technical supports I couldn't have able to finish my PhD on time.

Last but not least, I am deeply grateful to Mr. Gaurab Nandi Das for everything he did so that I will not give up my research work in the halfway. I am really thankful to him helping me to keep my mental sanity with his immense efforts.

"Which is more important?" asked the pupil,
"The journey or the destination?"
"THE COMPANY", said the teacher- by Unknown

Thank you all!!

Table of Contents

TABLE OF CONTENTS

List of Tables

LIST OF TABLES

List of Figures

LIST OF FIGURES

List of Images

LIST OF IMAGES

List of Appendices

LIST OF APPENDICES

Chapter 1

Chapter 1

Introduction

1.1. Wildlife trade

The sale and exchange of wild animal and plant resources by people and the effect of these acts on the conservation of biodiversity and its sustainable development is commonly known as "wildlife trade" (Oldfield *et al.*, 2003). The fifth pillar edict of King Ashoka in third century BC chronicled the protection and conservation of India's wildlife (Sharma, 2014). During the reign of king Ashoka, hunting and the indiscriminate slaughter of animals were banned. But, during the time of Mughal empire and the British colonial rule, the increased passion among the royal families for the recreational hunting (Shikar), unchecked poaching and wildlife trade of birds, primates, ivory, reptile skin and wild products were directly affected on the species population declining (Sharma, 2014). At the critical moments, the Wildlife (Protection) Act, 1972 comes to the action which mandates the need to protect all the forms of wildlife of the country. But yet, both terrestrial and marine species are illegally traded in the India whilst the overharvesting results the critical population declination of the species (Lewis *et al.*, 2022).

The history of wildlife trade in East and Southeast Asia is age-old. In the 20th Centuries, trade in wildlife was a major source of foreign exchange in Cambodia (Martin & Phipps, 1996). Until the 10th Century, the Vietnamese Kings collaborated with their Chinese leaders to sell and exchange live animals and wildlife products (Nash, 1997; Grieser-Johns & Thomson, 2005). The biodiversity of a region is largely the outcome of the long-thriving interactions between people and nature. However, as the human population expanded in due course of time and as trade flourished, especially in the twentieth century, biodiversity came to the edge due to serious threats from several socio-economic as well as cultural dimensions, among which one of the top reasons was the strong demand for wild products (Grieser-Johns & Thomson, 2005). In the 1860s, in the Lao People's Democratic Republic, the French explorer Garnier observed a flourishing trade of wildlife products, elephant ivory, rhinoceros' horns, peafowl feathers, and animal bone by the Chinese and Thai nationals (Garnier, 1996). By the 1960s, Lao's wildlife was being trafficked into Thailand by foreign businesses (Mills & Servheen, 1990; Mills &

Servheen, 1994), and by the late 1970s, $25 million worth of wild animal parts were traded to the Chinese by Cambodia's Khmer Rouge for weapons and supplies (Nooren & Claridge, 2001). Thus, so far, the trade of wildlife and wildlife products is becoming a potential driving force for species eradication from these regions (Grieser-Johns & Thomson, 2005). In the last two decades, the high demands of animal body parts in East Asia affected the wildlife population genetically by the drastic depletion in the wild (Niraj, 2009). While there is documentation on the history of wildlife trade from East and Southeast Asia, such records are very scarce from India.

The porous international boundaries of India with China, Myanmar, and other Southeast Asian countries, the growing aviation market with fast-expanding airport sectors, and the use of social media as an online marketplace makes India one of the top 20 wildlife trafficking countries of the world (Krishnan, 2022).

1.2. Illegal wildlife trade

The term "illegal" trade of species refers to the trade of specimens (or any easily recognizable part or derivate of the species, included in CITES Appendices I, II and III) except in accordance with the provisions of the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES)' (Source: CITES, Art. 1 and 2). Under international environmental treaties, i.e., most noticeably CITES, and under national legislations, those species categorized as endangered when traded are considered as illegally traded (Grieser-Johns & Thomson, 2005). Illegal trade in wildlife is a major form of international environmental crime (Brack, 2004). Studies on the illegal trade in wildlife have estimated values ranging from US $10 billion to US $20 billion globally per year (Brack, 2004; Wyler & Sheikh, 2008; Wilson-Wilde *et al.*, 2010). In East and Southeast Asia, the illegal wildlife trade is known as a "multi-million-dollar business", which comes in a most intensive nature (Compton *et al.*, 1999; Nooren & Claridge, 2001).

1.3. Characteristics of illegal wildlife trade

The local, national and international level illegal wildlife trade is also a commodity business that runs through different socio-cultural and socio-economic motives (Grieser-Johns & Thomson, 2005). The patterns and trends of illegal wildlife trade depend on a few commercial factors: improved transport infrastructure and development, market access and

acceleration of national and regional economic developments (Grieser-Johns & Thomson, 2005). Structurally, the well-framed illegal wildlife trade involves an extremely complex relationship between collectors, intermediaries, traders and wholesalers, where the characters of these relationships rotate based on their time and locations (Broad *et al.*, 2003). Along with changing circumstances, traders adapt their ways of contacting customers and carrying out transactions to maintain their substantial income (Broad *et al.*, 2003). When the traders sense the risk of being exposed or the supplies depleted, they adapt by changing target areas, sources, and species or groups to a new one, targeting new species within the commodity group. This is channelized by selecting and developing new methods and routes for the trade while picking out areas with weak wildlife law enforcement for their exploitations (Grieser-Johns & Thomson, 2005). Whenever wildlife law enforcement strengthens the implementation of rules in an illegal trade-affected area, the illegal trade machinery immediately goes underground, where it becomes difficult to monitor the activity, or determine the quantities, value, or number of species involved (Grieser-Johns & Thomson, 2005).

In recent times, with updated digitization and easily accessible modern technologies, illegal wildlife trade has become an integral part of cybercrime. By taking the benefits of different online platforms such as social media viz., Facebook, Instagram, Twitter, as well as online business sites like E-bay, Amazon and some self-created online sites, the traders provide information about their stocks to the customers (Woolloff *et al.*, 2022). The traders verify the customer identity first and, only after the confirmation, carry out the transactions by developing personal contact (Woolloff *et al.*, 2022). These types of cybercrimes are hard to detect, and identifying the source of the supplier can be a real task due to the use of fake ID's or multiple IP addresses. In addition, sales are now facilitated through cryptocurrencies such as Bitcoin (Grieser-Johns & Thomson, 2005). Hence, the constantly progressing, illusive and illicit nature of the illegal wildlife trade grows mysterious and threatens the wildlife population.

1.4. Quantum of illegal wildlife trade

The illegal wildlife trade always threatens peace, security, livelihood and biodiversity (World Bank Report, 2018). After narcotics, counterfeiting and human

trafficking, the "illegal wildlife trade" is reported as the fourth largest global illegal trade (Sollund & Maher, 2015). This is despite the different actions to curb wildlife trade that were taken since the early to mid-1990s, while most of the work done by the nongovernmental organizations (NGOs) and the trade and advocating accession to CITES were monitored by the region's non-party nations (Grieser-Johns & Thomson, 2005; World Bank report, 2018). In 1975, CITES came into force, providing a broad systematic framework to manage legal and illegal trade in wildlife and plants. CITES in operation is also supplemented by domestic legislation providing the highest degree of protection to more than 30,000 wild animal and plant species (both dead and alive) that are traded. CITES also provides strict protection to the species not categorized as endangered, permitting only their limited harvest to reduce the stresses upon wildlife and their derivatives (Grieser-Johns & Thomson, 2005).

1.5. Indian scenario of illegal avian trade in a global context

Based on the abundance, diversity and nature of avian species, the illegal bird trade is widespread in India as they provide good commercial values (Ahmed *et al.*, 2002). Approximately 3300 bird species are globally traded (WWF TRAFFIC India report, 2016). India has about 900 bird markets where about 450 avian species are sold (WWF TRAFFIC India report, 2016). Globally, India stands as the third highest in illegal/legal bird trade, which becomes the main reason for at least 176 bird species in India being categorized as 'threatened' (WWF TRAFFIC India report, 2016). In rural India, various parts of the birds and the bird itself are being used in traditional medicines and sorcery practices due to superstition (Ahmed, 1997; Ahmed, 1999; Ahmed, 2010). Most wild-caught birds are either destined for sale as pets or are hunted for their meat (Thomsen *et al.*, 1992; Ahmed, 2002). Birds hunted for their meat later on being sold/traded in the local markets based on their demands. The owl trade in India is well documented by Ahmed (2010) where he specifically mentioned about the live bird trade where 15 owl species from India are being traded; with the description of hotspot areas of owl trade, reasons behind the trade and the people who are involved in this trade. Bhupathy *et al.*, (2013) reported about annually trade of 13,067 birds in the Tuensang market of Nagaland from which the local people generate a revenue of about 18.5 lakhs/year by trading different kinds of bushmeat. Hunting/ poaching for bushmeat and for illegally trading birds for traditional medicine or sell them

for ancient cultural activities are also well reported where locally abundant to migrant bird species continued to be victim in the North-East region of India (Aiyadurai, 2012; Bhupathy *et al.*, 2013; Naro *et al.*, 2015). The song bird trade in India is also reported as a major threat to the population of wild bird species; as among the 18 species of mynas and starlings (Sturnidae) from India, nine of them have reported to present in illegal bird trade for pets, food and merit release (Ahmed, 1997; Ahmed, 2004). The Hill Mynas (*Gracula religiosa*) were reported to be smuggled out of India to various international markets especially to the Middle East via the illegal trade route of Pakistan and Bangladesh (Ahmed *et al.*, 2013). Although only a few reports are available from Indian context yet there is a serious dearth of systematic study to pull out the actual ongoing scenario.

1.6. Drivers of wildlife trade

The predominant driving factors of the legal/illegal wildlife trade in Asian countries are the high demand for wild animal products in China, where these are being used as food or medicine (Grieser-Johns & Thomson, 2005; World Bank report, 2018). The improved living standards and fast-growing population of East and Southeast Asia also contribute immensely to the wildlife trade by showing the directly proportional curves for traded species, i.e., as the increase in income directly related to the increase in demand for traded wildlife from these regions (Robinson & Bennett, 2002). The major and complex causes of wildlife trade are rooted in many different structures, such as social, economic, cultural and political (Grieser-Johns & Thomson, 2005). According to a World Bank report (2018), these constraints are compounded by different groups, wildlife hunters, rural communities, government officials, urban consumers, medical professionals, and decision-makers involved in promoting or curbing wildlife trade. Any practical solution applied towards curbing wildlife trade must address all these interest groups and factors from the grassroots levels with a better understanding of the dynamics of the trade. The regulatory mechanism should be set up at various levels, such as state, national and international levels, with adequate scientific knowledge of the species and sustainable management of the same. Public awareness about the trade and conservation of traded species by engaging local people and stakeholders needs to be implemented in policy and action (Grieser-Johns & Thomson, 2005; World Bank report, 2018).

1.7. Combating wildlife trade

Tackling the wildlife trade at the grassroots level needs adequate information and necessary actions covering four areas, i.e., policy, enforcement, supply economics, and consumer demand (Grieser-Johns & Thomson, 2005). The very dearth of systematic study makes illegal wildlife trades ambiguous in nature. Without specific information, any action taken to curb wildlife trade will be in vain. Physical inspection of the trade field is important to understand the gravity and quantum of trade. Once the threats and driving factors of trade are identified, appropriate actions can be taken. The illegal bird trade in India is greatly lucrative, relatively fail-safe and a well-organized business. On average, only 1% of cases of wildlife crime/trade are proven guilty in a court of law in India (WWF TRAFFIC India report, 2016). The low conviction rate is attributed to a lack of evidence, mainly species-level identification that can withstand in the court. One of the solutions to curb illegal trade with such issues is wildlife forensics. Wildlife forensics provides the necessary evidence to present in the court of law to give justice towards the voiceless victims. Forensic studies can include conventional plumology and modern molecular techniques such as genome sequencing and developing nucleotide database for traded species identification.

Along with different uses of plumology, the study of feathers of avian species can be a real asset, an invaluable tool for identification. The study of microscopic characteristics of feathers can assist with identifying avian species in forensic investigations, especially when nothing but feathers remain. The microstructures of different feathers from different locations with selected specific parameters can directly help in species identification aiding forensic investigations. Under the molecular techniques, the study of mitochondrial DNA (mtDNA) signatures for avian species can be crucial in wildlife forensics. However, the unavailability of species-specific mitochondrial DNA markers is also one of the main reasons for the low prosecution of wildlife crime. Furthermore, the study on complete mitogenome also provides a foundation to conduct similar work for other conservation-priority species, including birds.

1.8. Objectives of the study

Taking into consideration the above premise regarding wildlife trade in India, the present study was undertaken with the following objectives;

(1) Assess local markets in Assam for traded avian species.

(2) Identify feather microstructures of select traded avian species.

(3) Generate complete mitogenome sequences of selected traded avian species.

1.9. Organization of the thesis

The thesis is organized into five chapters, as mentioned below. A detailed literature review on each specific topic is given in the respective chapters focusing on each objective.

The first chapter is introductory, providing information on the study's background, need and importance, objectives, and the outline of the thesis.

The second chapter elaborates on the study area with a GIS map and describes its biodiversity, climate, people and conservation issues.

The third chapter explores the study area with respect to the illegally traded avian species and deliberates upon the data on different conservation threats associated with avian species. This chapter also provides the perception of local people from various groups and the role of NGOs in conservation. Thus, the chapter provides systematic information about the illegal avian trade scenario from Assam, where there is a dearth of systematic literature.

The fourth chapter dwells on Plumology and provides feather microstructures of select traded avian species from Assam collected from road-kill samples during the survey detailed in the third Chapter. By adapting plumology as a species identification tool, this chapter provides a feather catalogue for two highly traded avian species from Assam. The microstructures studied for the species can be used by different law enforcement agencies and research organizations for species identification, especially from confiscated samples from crime scenes, even in cases where the sample size is inadequate.

The fifth chapter discusses the complete mitogenome of two highly traded avian species from Assam. The samples collected from the road kill survey was used for

mitogenome study as a tool to be used in wildlife forensics. Using the NextGen Sequencing technique, this chapter provides the complete mitogenomes from tissue samples, valuable baseline data for future research.

The sixth chapter summarizes salient findings with respect to the set three objectives, followed by the conclusion and conservation implications for curbing illegal bird trade in Assam.

Chapter 2

Chapter 2

Study Area

2.1. Selection of the study area

The documentation of illegal trade and trafficking is mainly focused in India's central and northern states, and there is a huge information gap on illegal avian trade, especially from the North-East. It is speculated that the north-eastern states seem more vulnerable to illegal avian trade because of geographical, geopolitical, and socio-cultural reasons (Hilaluddin *et al.*, 2004; Aiyadurai *et al.*, 2010). There is a dearth of systematic literature except for a few newspaper reports on wildlife trafficking in Assam, which confirm that people in Assam collect and sell wild birds during different festivals associated with agriculture and cultural practice. Assam is also known as the "transitory route" for wildlife trafficking in the northeastern region (WCS Workshop Report, 2020). Assam shares its boundary with other states of the North-East while sharing the exit route of all illegal trade through road networks emanating from other states (WCS Workshop Report, 2020). Of India's top 10 traded bird species that contribute nearly 75% of the indigenous bird trade, 8 species are well distributed in the Assam region (Ahmed *et al.*, 2001). Moreover, in Assam, of the 55 Important Bird and Biodiversity Areas (IBA), 50, where poaching, hunting, poisoning and killing of avian species are ongoing, were reportedly under major threats (Rahmani *et al.*, 2016). Therefore, the state of Assam was chosen as the study area. The study was conducted in the districts of Assam, especially those nearby the protected areas, national parks, wildlife sanctuaries, reserve forests, and IBAs.

2.2. A brief history of Assam

Assam (Assamese: অসম) is the easternmost sentinel state among the eight northeastern states of India, formerly known as the Assam Province (British India). The name "Assam" holds numerous decrees for its origin. For the ancient Assam, the names 'Pragjyotisha' and 'Kamrupa' were used in the ancient Sanskrit literature. The Mahabharata, the Ramayana and the Puranas established the antiquity of the mentioned names. Gait (1992, reprint) wrote that the name 'Prajyotisha' or 'Pragjyotishpura' means the 'City of Eastern Astrology' as 'Prag' means 'former' or 'eastern' and 'Jyotisha' means

'a star', astrology, shining. Literature and many epigraphs provide references about the origin of the name "Kamrupa". According to Indian mythology, after Sati (one of the wives of the god Shiva and a daughter of the sage Daksa) deceased, the Gods sent Kamdev (known as the cupid god) to break Shiva's penance. Shiva was enraged about the results of Kamdev's mission and burnt him into ashes by opening his third eye. Later, Kamdev regained his original form here and based on this incident, the country was known as Kamarupa (where Kama regained his Rupa or form). Even though the origin of the name 'Aham' or 'Asom' is ambiguous, it is currently believed that the name was probably given after the Ahoms, who came into Assam in 1228 A.D. and ruled for nearly six hundred years. It is now well believed that the Anglicization of the modern name Assam comes from them.

The history of Assam is very intriguing. With the passing of time and stages of development, Assam's history can be divided into four eras (https://assam.gov.in/about-us/394). According to Samudragupta's inscriptions on the Allahabad pillar, the ancient era began in the 4th century with the mention of Kamarupa and the establishment of the Kamarupa kingdom. After that, according to the "Kanai-boroxiboa rock inscription", the medieval era began in 1206 with the attacks from the Bengal Sultanate, i.e., by Bakhtiyar Khilji, which took place after the breakup of the ancient Kamrupa kingdom with the sprouting of medieval kingdoms. In 1826, after the Treaty of Yandaboo, with the establishment of British control, the colonial era began, and in 1947, after the Independence of India, the post-colonial era began.

2.3. Geology and Physiography

Assam is located in the tropical latitudes 24°N and 28°N; and 90°E and 96°E eastern longitude (https://assam.gov.in/). The total geographical area of Assam is 78,438 sq. km. (https://assam.gov.in/). Assam shares interstate boundaries with Arunachal Pradesh, Nagaland, Manipur, Mizoram, Tripura, Meghalaya and West Bengal and the international boundary with Bhutan and Bangladesh (https://assam.gov.in/). In the northern parts, the state has Brahmaputra valley, in the southern part, it has Barak valley; in between these two, the Karbi Anglong Plateau to the south of the state (https://dgm.assam.gov.in/about-us/history).

Assam consists of three geological structural units, i.e., Karbi Anglong plateau, Tertiary depositional zone and Alluvium depositional plains of Brahmaputra valley and Barak Valley (https://dgm.assam.gov.in/about-us/history; Taher & Ahmed, 2021). The whole of Northeast India, including Assam, falls in seismic zone-V. Along with the Brahmaputra and Barak Valley, three Autonomous Councils are present in Assam; Karbi-Anglong Autonomous Councils (KAAC), Dima Hasao Autonomous Councils (DHAC), and Bodoland Territorial Area Districts (BTAD) (https://assam.gov.in/; Devi, 2019). Assam has 33 districts in it (https://assam.gov.in/about-us/394) (Figure 2.1), and these districts work under 5 different regional divisions (mentioned below) with a single divisional office (https://assam.gov.in/about-us/396).

i) **Barak Valley:** Comprises of Cachar, Hailakandi, and Karimganj districts with the divisional office located at Silchar.

ii) **Central Assam:** Comprises of Dima Hasao, Hojai, East Karbi Anglong, West Karbi Anglong, Morigaon and Nagaon districts, the divisional office located at Nagaon.

iii) **Lower Assam:** Comprises of Baksa, Barpeta, Bongaigaon, Chirang, Dhubri, Goalpara, Nalbari, Kamrup Metropolitan, Kamrup Rural, Kokrajhar and South-Mankachar districts with the divisional office located at Guwahati.

iv) **North Assam:** Comprises of Biswanath, Darrang, Sonitpur and Udalguri districts, the divisional office located at Tezpur.

v) **Upper Assam:** Comprises of Charaideo, Dhemaji, Dibrugarh, Golaghat, Jorhat, Lakhimpur, Majuli, Sivsagar and Tinsukia districts, the divisional office is located at Jorhat.

Recently, two more districts have been carved from the previously existing districts, i.e., Bajali from the Barpeta district and Tamulpur from the Baksa district, making the total number of districts 35 (https://assam.gov.in/about-us/396).

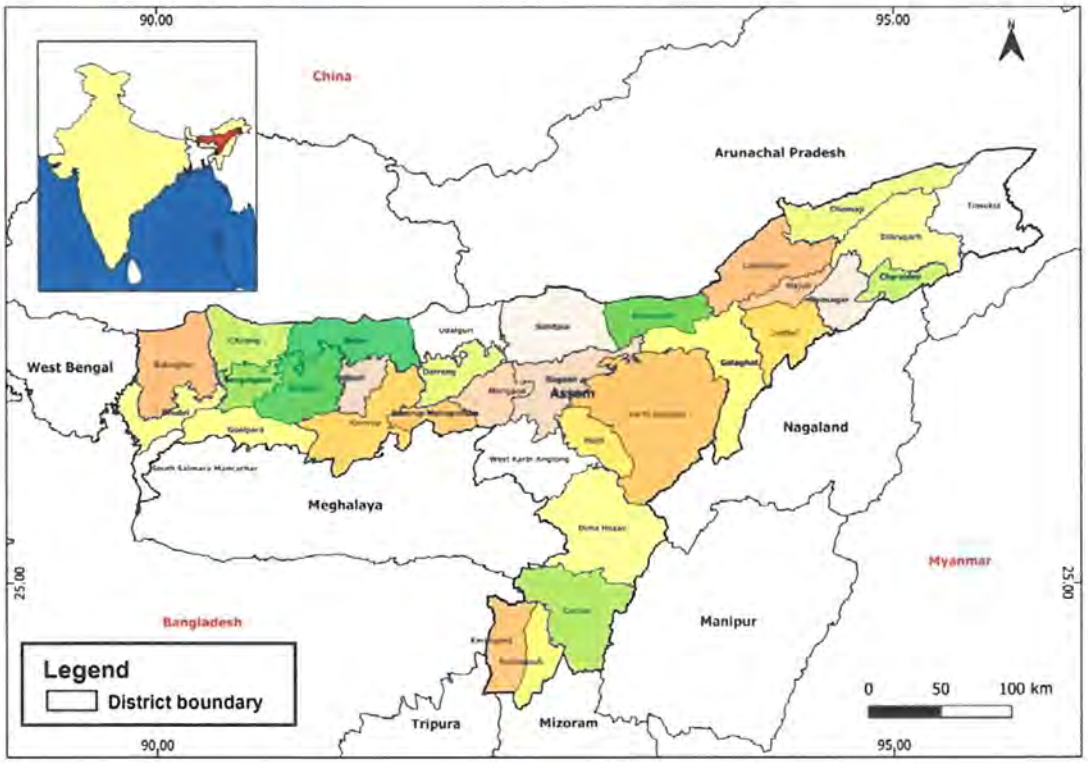

Figure 2.1: Districts of the state Assam.

2.4. Climate

Assam has the 'Tropical Monsoon Rainforest Climate', with high humidity levels and heavy rainfall throughout the years (https://assam.gov.in/about-us/assam-glance). The climate of Assam can be broadly divided into two major seasons, i.e., the winter and the monsoon season, along with two shorter seasons, spring and autumn, representing the transition between winter and monsoon and between monsoon and winter, respectively. The climate of Assam varies from district to district. The major districts present in the plains of Assam experience the maximum temperature of around 90°F (or 32°C) during rainy seasons. During the winter, the plains of Assam have a minimum temperature of about 47°F (or 8°C). A salubrious climate is present in the three hill districts of Assam, whereas the other 32 districts in the plain have comparatively warm summers and cool winters (http://asmenvis.nic.in/Database/Climate_1042.aspx). Because of these patterns, the climate of Assam is characterized by alternate cool and warm periods with high

humidity, especially from May to November. The two seasons, i.e., spring (March–April) and autumn (September–October), are usually pleasant with moderate rainfall and temperature. Assam gets some rainfall between March and May, i.e., in spring. From 1989 through 2018, the state received the highest rainfall (28.7%) during the southwest monsoon in July, followed by 28.6% in June, whereas August and September received 23.8% and 18.9% southwest monsoon rainfall (Guhathakurta *et al.*, 2020).

2.5. Vegetation

According to the Atlas, Forest type of India, 2020, there are 23 different forest types in Assam. The forest types occurring in the state are Assam valley tropical wet evergreen forest (*Dipterocarpus*), *Kayea* forest, *Mesua* forest, Cachar tropical evergreen forest, Assam alluvial plain semi-evergreen forest, Sub-Himalayan light alluvial semi-evergreen forest, *Syzygium* parkland, Pioneer Euphobiaceous scrub, Eastern alluvial secondary semi-evergreen forest, Cachar semi-evergreen forest, Secondary moist bamboo breaks, *Khashi* hill sal, East Himalayan upper *bhabar* sal, Kamrup sal, East Himalayan moist mixed deciduous forest, Northern secondary moist mixed deciduous forest, Low alluvial savanna woodland (*Salmalia-Albizia*), (*Terminalia-Lagerstroemia)*, Creeper swamp forest, Eastern seasonal swamp forest, *Khair-sissu* forest, Assam subtropical hill savannah woodland and Assam subtropical pine forest (Forest Survey of India, 2021). Of these, the major forest types are Tropical Wet Evergreen, Tropical Semi-evergreen, Tropical Moist Deciduous, Subtropical Broadleaf Hill, Subtropical Pine, and Littoral and Swamp Forests (https://assam.gov.in/about-us/397; Rahmani *et al.*, 2016). As per the 2021 assessment reports, Assam has 3,017 sq. km. of very dense forest (VDF), 9,991 sq. km of moderately dense forest (MDF) and 15,304 sq. km of open forest (OP) area, composing a total of 28,311.51 sq. km forest cover area with 0.29% scrub area (Forest Survey of India, 2021).

2.6. Biodiversity and protected areas

Among the 17 Mega bio-diverse countries of the world, India holds an important position with 7-8% of the recorded species (https://asbb.gov.in//biodiversity.html). With the prevalence of good climatic conditions and heterogenic physiography, phyto-geographically Assam is the home of many endemics, rare and centre of the origin of many commercially important plants, such as Banana, Citrus, Mango, *Zizyphus*, and Tea. The

luxuriant vegetation and the floristic richness made Assam worthy of the title of "Biological Gateway" of North-East India (https://asbb.gov.in//biodiversity.html).

Assam holds 193 mammalian species with wide distributional ranges in the region (https://asbb.gov.in//biodiversity.html). It is the home of a few species, such as One-horned rhinoceros (*Rhinoceros unicornis*), Water buffalo (*Bubalus bubalis*), Pigmy hog (*Porcula salvania*), Swamp deer (*Rucervus duvaucelii*), Golden langur (*Trachypithecus geei*), Hoolock gibbon (*Hoolock hoolock*) with their distributions limited to this state, especially in a few isolated pockets and protected areas. Assam also holds a key place in the evolutionary process of mammalian divergence in India with the close relationship of the relict mammalian fauna of Western Ghats and Assam and the Northeast region. Assam is recognized as one of the "endemic bird areas" in the world, being home to 950 numbers of bird species, i.e., 53.5% of bird species found in the Indian Sub-Continent (https://asbb.gov.in//biodiversity.html). There are several bird's species endemic to Indian region recognized from Assam state, including *Paradoxornis flavirostris, Perdicula manipurensis inglisi*. 45 species of birds recorded in the state are mentioned in the Indian Red Data Book. The varied physiography and climatic condition support habitats for a rich reptilian population, including Gangetic gharial (*Gavialis gangeticus*), 19 tortoises, and 77 snakes and lizards (https://asbb.gov.in//biodiversity.html). 70 Amphibian species reported from the Assam and North-East region (https://asbb.gov.in//biodiversity.html). The Brahmaputra and Barak River system, along with their tributaries and floodplain wetlands, are the main water source of the rich aquatic diversity in the state. These river basins provide conducive habitats to the 185 reported fish species, of which 25 are identified as threatened. Of the 1500 butterfly species reported from India, approximately half are reported from Assam and North-East India region (https://asbb.gov.in/ /biodiversity.html). Among 387 moth species of India, most are reported from Assam. The presence of high numbers of these "Environmental Indicator Species" proves the state's suitable habitat conditions and rich biodiversity.

Until December 2021, India had 106 National Parks covering 44,372.42 km^2 areas, 564 Wildlife Sanctuaries covering 1,22,509.33 km^2 areas, 218 numbers of Community Reserves covering 1,445.71 km^2 and 99 numbers of Conservation Reserves covering 4,726.24 km^2 area. The total protected area in the country is 987 covering a total of 1,73,053.69 km^2 area (http://www.wiienvis.nic.in/Database/Protected_Area_854.aspx). The protected areas in Assam include seven national parks (i.e., 2.51% of Assam's area) and 16 wildlife sanctuaries (1.88% of Assam's area), and two proposed wildlife sanctuaries (Government of Assam, 2021a; Rahmani *et al.*, 2016). The seven national parks cover 2,634.05 km^2 areas, and the 16 wildlife sanctuaries cover 1728.9 km^2 areas in Assam (Government of Assam, 2021b). Of the total area of Assam, about 17% is legally protected as forest areas (13590.267 km^2) (Government of Assam, 2021b). As part of the Eastern Himalayan Biodiversity Region, Assam is also part of one of the two biodiversity "Hotspots" of India (https://assam.gov.in/about-us/397). Two "World Heritage Sites" declared by UNESCO exist in Assam, Kaziranga National Park and Manas National Park (Rahmani *et al.*, 2016). Assam also has the world's largest river island, home to many unique species (Rahmani *et al.*, 2016). A total of 55 IBA sites are present in Assam, which is crucial for the threatened species' survival (Rahmani *et al.*, 2016). Deepor Beel is a "Ramsar Site" famous for its enriched biodiversity, especially the migratory birds (https://rsis.ramsar.org/ris/1207). A map of Assam showing the protected areas (national park and wildlife sanctuaries) is given in Figure 2.2.

The present study was conducted in the nearby locales of Kaziranga National Park, Manas National Park, Gibbon Wildlife Sanctuary, Chakrasila Wildlife Sanctuary, Majuli Island and Dehing-Patkai National Park. Each of these PAs is unique in its specific biodiversity. Whereas Kaziranga is famous for One-horned rhinoceros (*Rhinoceros unicornis*), Dehing-Patkai is famous for the endangered state bird of Assam (White-winged wood duck *Asarcornis scutulata*), Manas National Park is known for its population of Royal Bengal tiger (*Panthera tigris*), Gibbon Wildlife Sanctuary is home of six different primate species, and Chakrasila Wildlife Sanctuary is famous for the Golden langur (*Trachypithecus geei*). The Majuli Island is known for its migratory bird species and Ganges River dolphin (*Platanista gangetica gangetica)*. On top of all, these studied protected areas are well known for their rich species number and abundance of birds. Two

different migratory flyways, i.e., Central Asian Flyway (CAF) and East-Asian Australasian Flyway (EAAF) run through the state of Assam, which makes the state significantly important for bird migration (Vaithianathan, 2022). The mighty Brahmaputra, its tributaries, riverine lakes, and beels are hosts to numerous migrating birds, especially during the winter, due to the very suitable habitat conditions. At this point, the IBAs in Assam play a crucial role by providing feeding and breeding ground to the winged visitors on their odyssey.

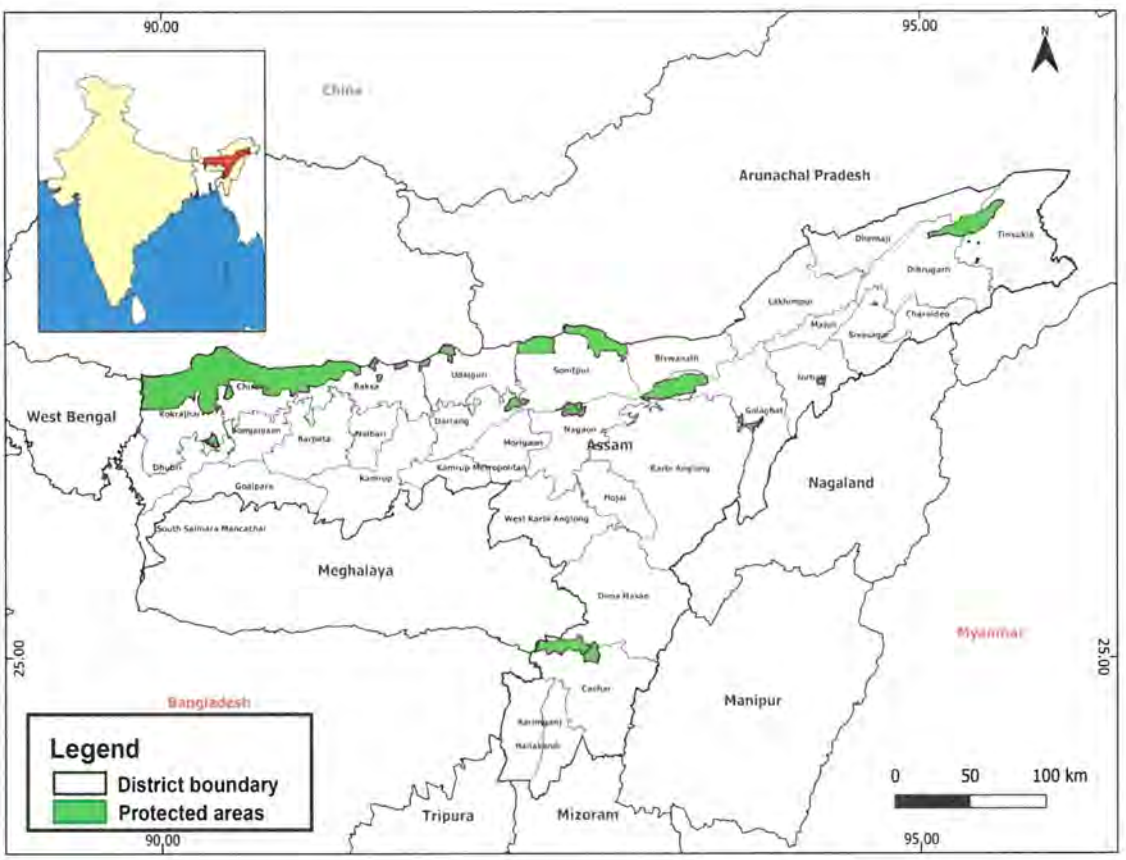

Figure 2.2: Protected areas of Assam.

2.7. Population and tribes

The very state of Assam is credited as the most populous state in entire Northeast India (https://assam.gov.in/about-us/394). The total population of Assam in 2021/2022 is 34,586,234 (34.59 million) as per the Aadhar Statistics (https://www.indiagrowing.com/Assam). According

to the 2011 census, 31,205,576 million people were present, with 15,266,133 million males and 6,406,471 million females (https://www.indiagrowing.com/Assam). Among the total population, 85.90% is rural, and 14.10% is urban, with an average population density of 398 persons per sq. km. (Forest Survey of India, 2021).

The people of Assam are basically an intermixture of various racial stocks such as Mongoloid, Indo-Burmese, Indo-Iranian and Aryan. The native people of the state of Assam are commonly known as "Asomiya" (Assamese), which is also spoken by the people and scheduled as the principal state language of Assam. Many tribes, each unique in tradition, culture, dress and exquisite ways of life, are present in this state. The Brahmaputra valley majorly is inhabited by the Assamese community and by the other major plain tribes such as Boro, Mishing, Rabha, Tiwa, Sonowal-Kachari and Deuri. However, other non-scheduled population groups are also present in the valley as Koch-Rajbongshi, tribal communities like tea garden laborers and Muslims (Devi, 2019). Whereas in the Barak valley, the district's main inhabitants include Bengalis, Manipuri and Assamese, interspersed with several different tribes such as H'mar, Riang, Kuki, Naga, Mizo, Barman (Dimasa Kachari), Vaiphei and Jaintia (Devi, 2019).

2.8. Livelihood

Four major livelihood patterns exist in Assam, i.e., Agriculture-based, livestock-based, non-timber forest products (NTFP)-based, and non-farm-based (https://asrlms.assam.gov.in/portlet-innerpage/assam-livelihoods-%E2%80%93-the-scenario). The fundamental base of the economy in the state is agriculture, as the agriculture sector supports more than 75% of the state economy and 53% of the workforce. Assam's most cultivated crop is rice, respectively, after mustard and tea. The economy of Assam also depends predominantly on an agrarian economic system, as more than 85% of Assam's population lives in rural areas. According to the 19th livestock Census 2012, a significant proportion of landless labourer and small and marginal farmers make use of livestock resources, with a total of 19.08 million livestock population. Of the livestock, the cattle population constitutes the largest, with 54.02%, followed by goats at 32.23% and pigs at 8.5%. Livelihoods based on NTFP, mainly products from bamboo and medicinal plants, are very limited across the state. Handloom weaving has a glorious history in Assam and is inexorably linked with

Assamese Culture and Heritage. The handloom Industry of Assam provides the maximum employment after agriculture, led by the traditional making of handloom and handicraft products representing the non-farm-based livelihood sector. Among these livelihood options, the present study assessed the community's reasons and involvement in avian trade along the select protected areas of Assam.

Chapter 3

Chapter 3

Objective 1: Assessment of local markets of Assam for traded avian species

3.1. Introduction

3.1.1. Illegal Wildlife trade

The illegal wildlife trade, also known as Illegal unsustainable wildlife trade (IUWT), affects all taxonomic groups in numerous ways (Cardoso *et al.*, 2021; Fukushima *et al.*, 2021; Morton *et al.*, 2021). Globally, wildlife trade is considered a major cause for the decline of wild fauna and flora (Niraj, 2009). The "illegal trade, smuggling, poaching, capture or collection of endangered species, protected wildlife (including animals or plants that are subject to harvest quotas and regulated by permits), derivatives or products thereof" (Wyatt, 2009) are all covered under wildlife trafficking or illegal wildlife trade (Utprey *et al.*, 2021). Global illegal trade in wildlife per year is estimated to be between US $10 and US $20 billion (Brack, 2004; Wyler & Sheikh, 2008; Wilson-Wilde, 2010; WWF Traffic India Report, 2016). Illegal wildlife trade is reportedly the fourth largest global illegal trade after narcotics, counterfeiting and human trafficking (Sollund & Maher, 2015). This illegal unsustainable wildlife trade (IUWT) has been identified as the major "Key challenge" for modern conservation, with the number of 100 million of plants and animals being internationally trafficked per year (Morton *et al.*, 2021).

3.2. Review of literature

3.2.1. Illegal trade in South-East Asia

In Asia, wildlife trade is one of the major challenges to species conservation with its dynamic nature and a prominent threat to biodiversity, very challenging to pin down (Menon, 1996; McNeely *et al.*, 2009; Nekaris *et al.*, 2010; Esmail *et al.*, 2020; Utprey *et al.*, 2021). The IUWT generates considerable annual revenue for some of the least economically affluent people in South-East Asia, making the trades very adaptive and lucrative (Nijman, 2010). In South-East Asia, the variety of critical goods and services provided to the rural poor by plant and animal products are undeniable, which includes

their daily source of food, income, energy and medicine (TRAFFIC, 2008; Krishnasamy & Zavagli, 2020).

One of the major driving factors of illegal unsustainable wildlife trade in South-East Asia is the demand for traditional medicine prepared using different body parts of wild fauna and flora (Veríssimo *et al.*, 2012). These demands directly affect the existing population of particular species, taxa and sometimes the entire biodiversity leading to the continuous decline of the same (Veríssimo *et al.*, 2012; Morton *et al.*, 2021). The trade is also directly correlated with the climate change of a particular region, with the quick functional decline of the ecosystem services associated with biodiversity loss (UNODC, 2022). The intensive extraction and high demand for illegal trade together contribute as a major driving factor for species' extinction (McClenachan *et al.*, 2016).

A few countries are identified as the major exporters of wild-caught animals, viz. Malaysia, Vietnam, Indonesia and China, while the European Union and Japan are the most significant importers (Nijman, 2010). Due to the informal chain of trade-network, the actual values and scale of the trade are not quantified, which also connotes the quantum of forest-related income earned by the poor rural people (TRAFFIC, 2008). The illegal unsustainable wildlife trade reflects the weakness in environmental governance and other associated exacerbating factors. It is mostly led by ineffective regulations, failure of effective monitoring and enforcement alongside rule-breaking, corruption and other organized crime (UNDOC, 2020; UNODC, 2022). The upgradation of illegal trade from physical trade to the online platform is right now the topmost issue that needs to be counter-fired by tackling with tech experts as soon as possible (Lavorgna, 2014). The Covid-19 outbreak also provided a strong platform to uprise these online trades, including the pet trade (McNamara *et al.*, 2020), with an outstanding increasing rate of hunting from some parts of the world (Morcatty *et al.*, 2021).

3.2.2. Birds in trade

Birds form the major component of the multi-billion-dollar international illegal wildlife trade industry involving a third of the global avifauna (Butchart, 2008; Marshall *et al.*, 2020a). Wildlife trafficking of avifauna is huge worldwide, with approximately 3300 species being subject to illegal trade (WWF TRAFFIC India report, 2016). According to

the data on international trade in CITES-listed animals, from 1998–2007, of the 35 million traded, 1.0 million birds were illegally exported from South-East Asia (Nijman, 2010). The most commonly traded bird in national and international illegal trade are parrots (including macaws, cockatoos, and lorries), followed by others such as flamingos, eagles, toucans, and several songbirds (Gilardi, 2006). Convention on International Trade in Endangered Species reports that birds from the passerine group were traded internationally as pets in Latin American countries (FAO, 2011). Due to various cultural factors and local biodiversity effects, birds from the passerine group were traded throughout Brazil as pets (Destro *et al.*, 2012; Souto *et al.*, 2017). In the Netherlands, 65% of parrots (Psittaciformes) from South America and 23% of birds of prey from the Middle East were trafficked between 2009 and 2013 (van Uhm & Spapens, 2018). Singapore has been known as the major conduit for bird trade from Africa and Europe to East Asia and Middle East from 2005 to 2014, wherein it imported nearly 225,600 birds in that period (Name, 2016). In South-East Asia, the rapid decline of bird diversity is due to the uptake of the "cage-bird phenomenon", also labelled the "Asian Song Bird Crisis" ((Nijman, 2010; Su *et al.*, 2014; Lee *et al.*, 2016; Harris *et al.*, 2017 Marshall *et al.*, 2020a).

In South-East Asia, more than >1000 species of different wild birds are traded for various reasons, mostly due to the demand to be used as pets (cage-bird, song-bird) along with the use for nutritional and medicinal resources, symbolic or cultural practices and gambling-recreational contest (Jepson, 2010; Harris *et al.*, 2017; Souto *et al.*, 2017; de Oliveira *et al.*, 2018; Rentschlar *et al.*, 2018; Bezerra *et al.*, 2019). Keeping birds as pets, especially the song-birds is a central belief of South-East Asia's people, deeply rooted in different social values and cultural practices (Marshall *et al.*, 2020a; Marshall *et al.*, 2020b; Flink *et al.*, 2021). Indonesia has the world's highest number of threatened native bird species (n=175), resulting from illegal and unsustainable overexploitation for the pet trade (IUCN Red List, 2021). The bird trade research of the various states of Indonesia was focused on very specific taxa, e.g., Straw-headed bulbul (*Pycnonotus zeylanicus*), Black-winged myna (*Acridotheres melanopterus*), Bali myna (*Leucopsar rothschildi*) and Sumatran laughing thrush (*Garrulax bicolor*) (Leupen *et al.*, 2022). On the island of Java, Indonesia, over 70 million cage birds are kept across 12 million households, which signifies the seriousness and demand for songbirds as pets (Marshall *et al.*, 2020b). Interestingly,

the film and television media have also recently influenced the bird trade. Nijman & Nekaris (2017) reported on the effect of fiction series on the illegal owl trade in Indonesia, where the owls were previously called *Burung hantu* ("Ghost birds"); but after releasing the Harry Potter series in the 2000s, owls are referred as *Burung Harry Potter* ("Harry Potter birds") in the bird markets.

3.2.3. Bird trade scenario in India

The first study on India's bird trade was in 1971 on 275 bird species exported to Heathrow airport in London (Ahmed, 2002). In India, all export and domestic trade of wild avifauna has been banned since 1990-1991; yet the illicit trade of birds is ongoing in a huge quantum (Ahmed, 2014; Singh *et al.,* 2011). Among all illegal trades, the bird trade in India is the most organized, where traders and customers are well connected via the internet for transferring information to cash transactions (Singh *et al.,* 2011). The study by TRAFFIC India revealed that annually one million bird species are traded in and around India (Ahmed, 2002; Ahmed, 2004). It has also been estimated that the primary grass-root level domestic wild bird trade is approximately a minimum of Rs. 25 million/year (Ahmed, 2004). WWF TRAFFIC India report (2016) divulges that around 450 bird species are reportedly traded in more than 900 markets in India, making the country globally the 3rd highest in illegal/legal bird trade. This huge unlawful trade is perhaps the main reason for at least 176 bird species coming under the 'threatened' category (WWF TRAFFIC India report, 2016). The illegal bird trade in India is greatly lucrative and relatively fail-safe; hence, it has developed into a well-organized illicit business. On average, only 1% of wildlife crime/trade cases are proven guilty in a court of law in India (WWF TRAFFIC India report, 2016).

According to Ahmed (2004), the major commercial driving factors of the Indian birds in trade mainly depend on the abundance, diversity and nature of avian species the illegal trade (i.e., demand). In several parts of India, dried-up body parts and the bird itself of a majority of wild-caught birds are either destined for sale as pet, food or meat trades, merit-release, black magic, medicinal value, feather trade, lab specimens, zoos and bird sport (Thomsen *et al.,* 1992; Ahmed, 1997; Ahmed, 1999; Ahmed, 2002; Ahmed, 2008; Ahmed, 2010; Ahmed, 2012; Ahmed, 2014). One of the fraudulent practices in the illegal

avian trade is selling small wild birds as exotic species in the markets by disguising them by applying artificial colour (Ahmed, 1999; Ahmed, 2014). Most trappers intentionally target species like weaver birds and munias, which are sold in the markets based on colour distinction (Ahmed, 1997; Bhargava, 2000; Ahmed, 2014; Bhargava, 2017). Ahmed (2004) well reported the trade of the nest of edible-nest swiftlet from Andaman and Nicobar Islands that were traded internationally on a large scale just because of the derivatives. Hunting of Galliformes for bush-meat from wetlands is well-reported in India (Gupta, 2004; Sundar & Kittur, 2013; Ramachandran *et al.*, 2017). In Arunachal Pradesh, most avian species are hunted for their meat, but many small birds, which do not come under the taboos of different tribes, are also being hunted despite their size and commercial values (Aiyadurai, 2012). Earlier, the hunting and poaching in the northeastern parts of India were done for domestic consumption, but nowadays, due to commercial importance, people are getting attracted to this lucrative trade (Kumara & Singh, 2004; Datta *et al.*, 2008; Aiyadurai *et al.*, 2010; Aiyadurai, 2011; Bhupati *et al.*, 2013; Selvan *et al.*, 2013; Velho & Laurance, 2013).

Since time immemorial, avifaunal species have been included for human consumption by many tribes who used to hunt bird species along with other species (Tynsong *et al.*, 2012). In the early 1900s, in India, when trapping bird species was a legal profession, some people specialized in trapping specific bird species (Bhargava, 2017). From the Northern parts of Baheliyas, Mir-Chikaris are the epic example of some tribes known for their expert avifaunal hunting technique (Ahmed, 1999; Ahmed, 2004). In North-Eastern India, War Khasi communities of Meghalaya passed their hunting techniques using traditional methods from generation to generation (Tynsong *et al.*, 2012). In Arunachal Pradesh, several indigenous tribes practice indigenous hunting traditions for food, trade, culture and leisure, directly affecting the state's biodiversity (Aiyadurai *et al.*, 2010). However, hunting in Arunachal Pradesh is mostly done by the tribal groups during the time of jhum cultivation and at the time of crop harvesting, where most of the avian species are hunted for bush-meat (Chutia & Solanki, 2013). Catapults and guns are the most common tools for hunting birds in India. However, different other types of traditional traps include loop bamboo strip traps, canopy traps, stone traps, noose with a coloured food bait, triangular traps and using direct metal wire or bamboo, which are mainly used in

Arunachal Pradesh but in Nagaland nowadays people mainly hunt avian species by using guns along with their traditionally perishable materials (Aiyadurai, 2012; Imchen & Joglekar, 2015; Naro *et al.*, 2015). The most commonly used trap for avian species in India is the net trap, and most of them die by entangling themselves while trying to be free from the net (Ahmed, 2004; Singh *et al.*, 2011).

Ahmed (1997) reported northern India as the centre point of illegal avian trade. The illicit avian markets, particularly for the wild birds in India, connected to Patna city, Lucknow, Meerut, Hyderabad, Bangalore, Mumbai and Chennai, are all interlinked with each other based on public demands (Niraj & Ghosh, 2014). Based on survey reports of TRAFFIC India, Kolkata, Delhi and Sonapur are declared as the testimony-bearing cities for the wild animal pet trade in India (Niraj & Ghosh, 2014).

Most of the hunting reports of avian species are from Arunachal Pradesh and Nagaland due to cultural and social reasons where wild birds are readily available in markets (Aiyadurai, 2011; Bhupati *et al.*, 2013). The report states that 13,067 birds are traded annually in the Tuensang market of Nagaland, generating revenue of 18.5 lakhs/year from birds and other animals (Bhupati *et al.*, 2013). Because of the rich biodiversity and low human population in Arunachal Pradesh, hunting is directly linked with poverty, commerce, and customary and cultural practices, where most avian species like hornbills are being hunted and are becoming threatened (Bhatt & Pandit, 2019).

There are still only scanty records from India on the national and international illegal trades of wildlife, especially birds, whereas the actual ongoing scenario is still unknown due to the elusive nature of illegal trade (UNODC, 2022). Busina *et al.*, (2020) suggested that the direct census-based market surveys to estimate illegally traded bird species never represent the actual data and are unable to represent the true volume of the trade despite continuous efforts and intensity of visits. However, the urgent necessity is to bring to light the actual scenario of these trades, despite any political, socio-cultural or economic issues. The WCS Workshop report (2020) mentions "Guwahati", the capital of Assam, as the known "transitory route" for wildlife trafficking. Still, until now, no conclusive report describing the illegal avian trade in Assam exists. So, with this study, a systematic attempt has been carried out to assess the status and driving factors of illegally traded avian species focusing on an important state of India, i.e., Assam.

3.3. Materials and methods

3.3.1. Selection of the markets for the study

The market survey was undertaken in the vicinity of the Important Bird and Biodiversity Areas (IBA) sites as well as Protected Areas (PA) across 16 districts of Assam. According to Rahmani *et al.*, (2016), avian species in 50 IBAs (of the 55) face serious threats in Assam due to hunting, poisoning and poaching for various human purposes. Secondary data from published and grey literature were used to identify and select "wildlife trade" markets in the state to conduct the survey (Ahmed, 2002; Islam & Rahmani, 2004). Information on the targeted markets for the survey was also collected from personal contacts such as forest personnel, researchers, birding groups, individual birders, bird photographers and local birding guides. In addition, local newspaper articles also helped in the selection of the markets for the survey.

3.3.2. Data collection

The snowball sampling method was opted to assess and understand the quantum of illegal avian trade in different districts of Assam (Johnson, 2005; Atkinson & Flint, 2001; Gamborg *et al.*, 2012; Naderifar *et al.*, 2017; Utprey *et al.*, 2021). The select daily and weekly markets were surveyed, covering two seasons, i.e., winter and monsoon, in 2019. Using a custom-made semi-structured questionnaire, the markets were surveyed physically based on chain referral to obtain maximum information (Gamborg *et al.*, 2012; Utprey *et al.*, 2021). The detailed questionnaire has given in Appendix-1. People present in the markets on a daily/regular basis were mainly targeted as the primary respondent. Regular vendors and butchers in the markets were approached as the first respondents. Spiritual mendicants (locally called Babajis, Sannyasins), fortune tellers, occasional vendors and fishermen selling diverse biological products were also interviewed. The respondents' perception of wildlife trade, wildlife laws and NGOs (Local/ International) associated with various local conservation issues was also documented. The perceptions of local people were measured through these questions, viz. Should avian species be traded for different human use? Can avian species be protected through legislation? Can NGOs be helpful to protect wildlife/avian species? and the frequency of respondents per district to encounters provided the information on the birds being traded.

In each market, 5 to 13 respondents were interviewed. During the survey, respondents were initially sceptical about sharing information about the locally traded avian species. They were apprehensive about sharing the information (along with their names) with forest officials. Hence, they initially denied knowledge about any illegal trades, and they opened up only when they were assured about the interacting person's identity and trust. In the field, the researcher's identity was disguised as a bird/butterfly researcher, photographer, and sometimes as a tourist to extract maximum accurate information.

The survey was conducted in the local languages translating the questions into local/tribal languages. The Assamese language was broadly used to communicate with the forest officials, whereas other languages, viz. Mising, Koch-Ranjbongshi, Bodo, Garo, Rabha, Bengali, Nepali and Hindi were also used to communicate with the local respondents. To remove the language barrier and extract the local people's exact information, field assistants were only selected from the surveyed areas. Moreover, discussions were started by the lead of local people only. With the help of local interpreters (field assistants), the conversant statements were translated for both the respondents and the interviewer (Ogra, 2009).

Photography was not encouraged by the respondents, and we also found that it influenced the responses; hence, it was avoided, except when the respondents were requested photographs of their collecting/trapping gear. After the field interaction, the information/data collected was translated into English and tabulated in excel sheets for further analysis.

3.3.3. Data analysis

At first, a checklist of traded avian species was prepared based on the information sourced from the respondents while surveying markets across districts (May 2002; Utprey *et al.*, 2021). The present status of avian species was obtained from the 'Indian BIRDS' journal's checklist of birds of India (Praveen *et al.*, 2021). The distribution status of the species was adopted from Grimmett *et al.*, (2016). The Assamese vernacular names of available traded bird species were opted from "ENVIS Resource Partner on Avian Ecology" of Bombay Natural History Society (http://bnhsenvis.nic.in/Database/VernacularNames_832.aspx).

The data collected was categorized into

i) major reasons for avian trade,

ii) different trapping gears used to collect avian species,

iii) demand for specific species in trade and

iv) perception of local peoples towards illegal trade.

Market Prize: Based on the local trade price and the demand, the bird species were divided into three categories, namely high (Rs. 300–Rs 500), moderate (Rs. 200–Rs. 300), and low (Rs. 1–Rs. 200).

Statistical analysis: SPSS (version 23) software was used for statistical analysis; Chi-square tests on the numbers of traded species according to the respondents and Pearson correlation coefficient to see the relation between traded avian species and their market price that reflects the demand and supply. Local people's perception of illegal avian trade was also measured using Likert scale (Talukdar & Gupta, 2016). We have used the five-point Likert scale (1= Strongly agree; 2= Agree; 3= Neither agree nor disagree; 4= Disagree; 5= Strongly disagree) to score local people's perceptions.

3.4. Results

The surveys across 16 districts of Assam were conducted by visiting 130 local markets and interacting with 1003 respondents (Figure 3.1, Image 3.1). The districts surveyed for this study included Kamrup, Morigaon, Nagaon, Tinsukia, Dibrugarh, Sivsagar, Jorhat, Majuli, Golaghat, Dhubri, Kokrajhar, Bongaigaon, Chirang, Barpeta, Cachar and Hailakandi (Table 3.1). The number of respondents varied, ranging from 6 to 175 (Minimum–Maximum) individuals per district based on the market focus and people willing to talk about the trade. Markets were surveyed in the vicinity of 15 protected areas, viz. Deepor Beel, Pobitora Wildlife Sanctuary, Laokhowa and Burhachapori Wildlife Sanctuary, Dehing-Patkai National Park, Pani Dihing Wildlife Sanctuary, Gibbon Wildlife Sanctuary, Majuli World Heritage Site, Kaziranga National Park, Nambor-Doigurung Wildlife Sanctuary, Chakrashila Wildlife Sanctuary, Kaikoijana Reserve Forest, Manas National Park, Borail Wildlife Sanctuary and Katakhal Reserve Forest of Assam.

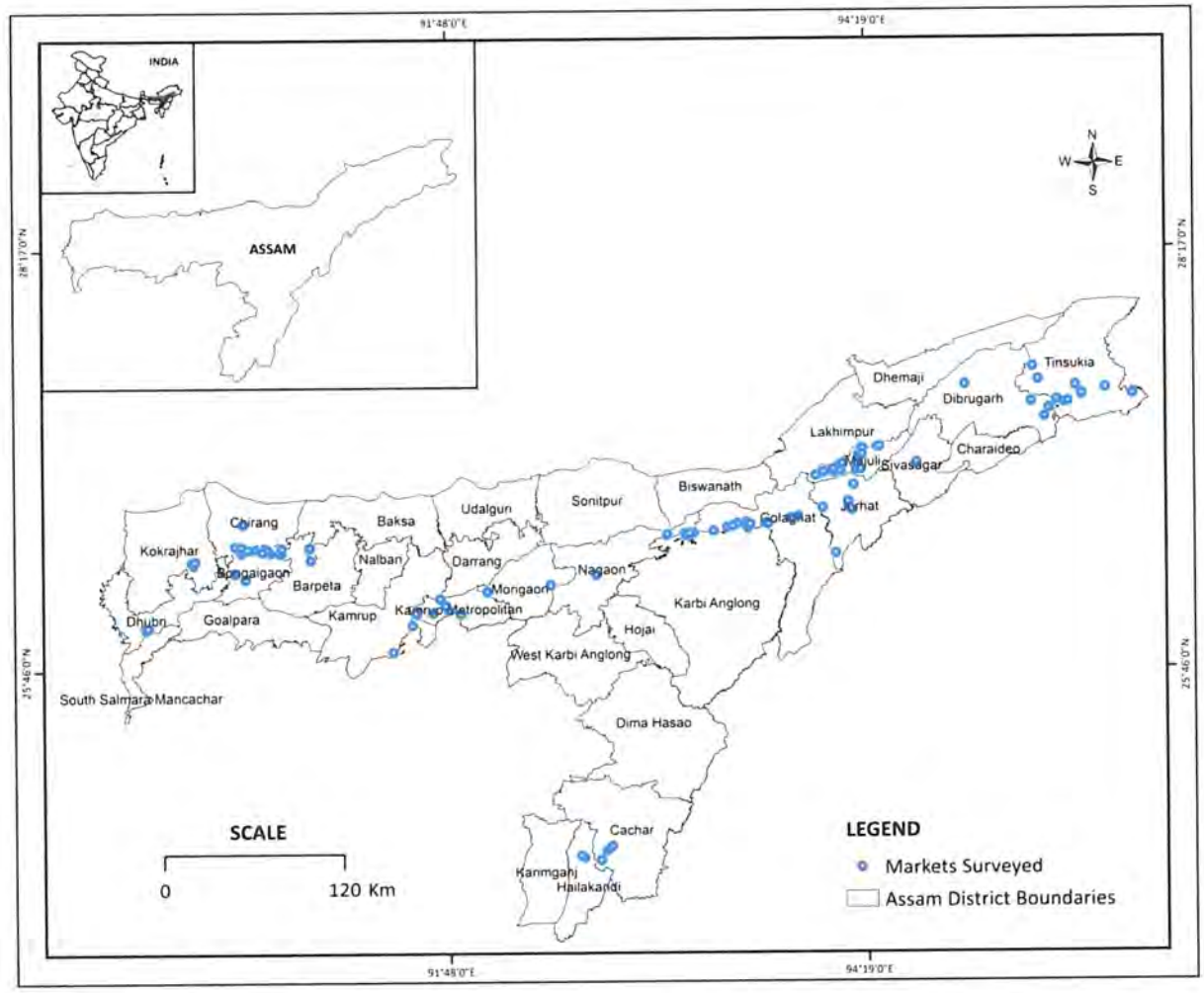

Figure 3.1: Markets surveyed in the districts of Assam.

Image 3.1: Local markets Surveyed in Assam and interaction with the local people. A-Tingrai weekly market, Tinsukia district; B- Diffolu weekly market, Golaghat district; C-Behora weekly market, Golaghat district; D-Nagaon weekly Market, Nagaon; E & F-Interactions with the local people.

3.4.1. Avian species illegally traded from Assam state

In the present study, I documented 82 avian species belonging to 33 families representing 12 orders as illegally traded (Table 3.2) from the state of Assam. The checklist of these 82 species, including their WPA, IUCN status, distributional status and vernacular names, are listed in Table 3.1. Among the species, the Passeriformes (35%) is the highest traded order of birds, while Columbiformes (4%) is the least traded order. Species belonging to the family Anatidae (13%) are highly traded among the families, followed by those under Ardeidae (9%). There are also a few species that we were not able to identify from poor quality pictures and local names; also, the respondents refused to give any detailed specific information regarding the species forcing us to broadly categorize these as ducks (28%), pigeons (24%), water birds (20%), pipits (15%) and raptors (13%).

3.4.2. The demand for the traded species

Based on the respondents, the number of species traded varied significantly (χ^2= 765.798, df= 81, p=0.001). Our result showed a strong positive correlation between the number of traded avian species and their market price (Pearson correlation coefficient: r=0.60, p<0.001), the price largely reflecting the demand in the illegal trade. The percentage of highly demanded bird species is 58%, whereas 26% are in moderate demand and 16% are low in demand. The list of the top 15 bird species with high demand is prepared from their average market price (Table 3.3). Of these 15 species, 14 are resident, except for the Grey-leg goose (*Anser anser*), a winter visitor to Assam.

3.4.3. Reasons behind illegal avian trade

The reasons for the trade of avian species are the demand for bush-meat (48%, N=59), superstition and black magic (14%, N=17), plume trade (13%, N=16), pet trade (13%, N=16), and cultural/traditional uses (12%, N=14) (N= Numbers of traded species) (Figure 3.2).

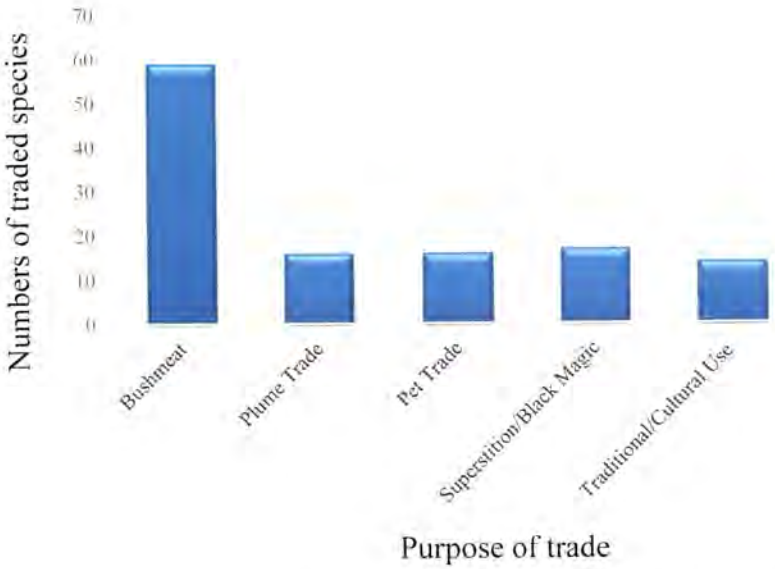

Figure 3.2: Purposes of avian species traded in Assam.

3.4.3.1. Bush-meat

The highest numbers of species (i.e., duck, other water birds, paddy field birds) were traded for bush-meat throughout the year, and the trade peaked during winter, the major bird migration season. Winter is also the season for harvest festivals, and on other auspicious days in Assam, for the people gathering for the celebrations, bush-meat is reportedly an exceptional opulent dish. As the water birds, including the migratory species, become abundant in the IBAs, PAs, wetlands and other water bodies, capturing them and trading in the daily/weekly local markets would be easy. The second peak season in the state for bird trade is during the monsoon (i.e., June-September) when most of the rivers are flooded, and birds are hunted along with some of the mammals for bush-meat. Other locally abundant species like junglefowl, mynas, pigeons and quails are being hunted throughout the year for bush-meat.

3.4.3.2. Plume Trade

Species with intricately attractive feathers with vivid colours are in demand in the plume trade. Feathers of Indian Roller (*Coracias benghalensis)* (n=55), Scarlet Minivet (*Pericrocotus flammeus*) (n=35) and Coppersmith Barbet (*Psilopogon haemacephalus*)

(n=19) fetch high prices in the plume trade as they are used in ornaments like earrings, wind chimes, toys and decorative products (n=number of respondents for the species). Some plain-coloured feathers of birds such as Cattle Egret (*Bubulcus ibis*) (n=42) and ducks are traded as they are easy to colour, and buyers use them for many purposes. Feathers of different colours and sizes are also traded for making dusting brooms commonly used in cars. Feathers of Indian peafowl (*Pavo cristatus*) are traded in weekly markets but reportedly sourced mostly from South India.

3.4.3.3. Pet trade

Birds with excellent voice mimicking characteristics and attractive colours, such as Rose-ringed Parakeet (*Psittacula krameri*) (n=75), Common Hill Myna (*Gracula religiosa*) (n=58), and Common Myna (*Acridotheres tristis*) (n=59) are sold as pets (n=number of respondents for the species). Respondents mentioned that the songbird species encountered mostly in the trade were species like Scaly-breasted Munia (*Lonchura punctulata*) (n=44) and Red-vented Bulbul (*Pycnonotus cafer*) (n=47), are also traded as pets. Generally, all the abundant species of parrots, mynas, weavers and bulbuls are present in the illegal per trade of the state.

3.4.3.4. Superstition and Black Magic

Different owls and raptor species are frequently used in the state's superstitious rituals and black magic. Live birds (mostly owls, four species from the order Strigiformes; n=221) (n=number of respondents for the species) are sacrificed for superstition purposes, and different body parts of these species are available in the illegal trades for use in black magic. Oriental Magpie Robin (*Copsychus saularis*) (n= 20) is sacrificed in some parts of Assam in front of the deities to fulfil promises. Trade in birds is also related to using their body parts in traditional medicines.

3.4.3.5. Traditional/Cultural use

Bird species are also traded for use in different traditional beliefs/rituals. Indigenous tribes in Assam use species such as Spotted Owlet (*Athene brama*) (n=60), White Wagtail (*Motacilla alba*) (n=23) and Lesser Racket-tailed Drongo *(Dicrurus remifer*) (n=20) for performing traditional rituals (n=number of respondents for the species).

3.4.4. Trapping gears used in collecting avian species for trade

Nine trapping methods are commonly used to collect birds from their natural habitats (Figure 3.3, Image 3.2). These are fishing nets, bamboo traps, food poisoning, catapult, pitfall trap, bait, bow and arrow, hand collection and other nets. Of these, catapult (19%) was the highest, while pitfall trap (2%) was used only for a few species. Catapult (19%) is used by adults and children depending on their skill to hunt down locally abundant species (Image 3.2C). Species collected using catapults mostly are for bush-meat. Food bait (18%) is the second-highest trapping method used to collect the birds alive from their natural habitats (Image 3.2D). Poisoning, using pesticides or locally available poisons (16%), is also frequently used to collect birds. Different bamboo traps are the second preferred gear (15%) to collect live birds (Image 3.2B). The type of bamboo traps used varies from tribe to tribe based on their indigenous knowledge and skills. Fishing nets (13%) are also used to capture birds alive, usually during migratory, fruiting, and monsoon seasons (Image 3.2A). Fishing nets are set near wetlands, ponds, water bodies or fruiting trees so that birds are trapped while crossing by or foraging. Other nets (8%), such as modified mosquito nets, are also used to capture birds. Hand collection (5%) of fledglings from their nest is done mostly for the bird species used in the pet trade since that reduces the injury to the bird and ensures a better price. Bow and arrow (4%) are used, mostly by children, often to capture locally abundant species. Pitfall (2%) traps are used to catch elusive species, either using another individual of the same species as bait or setting up the trap in the regular habitats of species. Most trapping gears are locally made with materials like bamboo, jute, wood and fine fishing nets. Capturing live birds from their nest always needs a person with expert climbing skills.

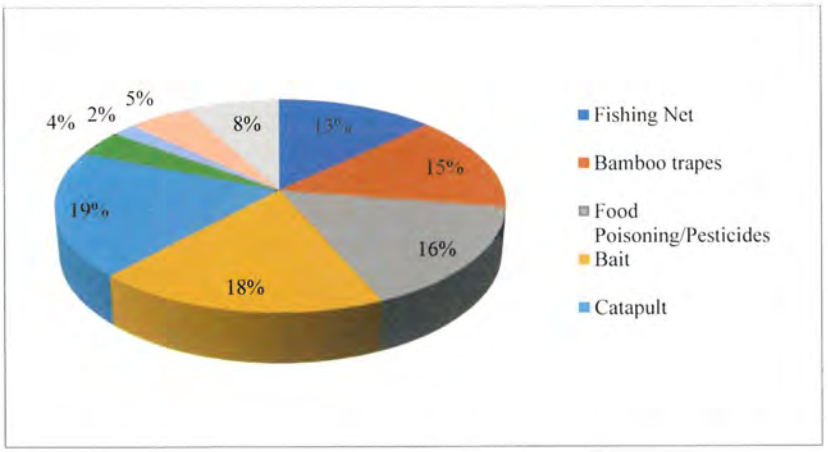

Figure 3.3: Percentage of trapping gears used in the collection of avian species.

Image 3.2: Common techniques/traps used to capture birds in Assam. **A.** Fishing net; **B.** Bow-shaped bamboo trap; **C.** Catapult; **D.** Bamboo basket to trap birds using bait.

3.4.5. Perception of local people towards illegal avian trade

Perceptions of local people regarding different their conservation varied in different districts of Assam (Table 3.4). The perceptions of local people for three different conservation aspects were scored using Likert scales are as-

3.4.5.1. Should avian species be traded for different human use?

Local people from the surveyed districts strongly disagreed about the trade of avian species for different human uses, such as bush meat or as pets (Figure 3.4). Interestingly, all the respondents from Nagaon districts emphatically disagreed in high numbers among all the surveyed districts of Assam. Whereas, in the Dhubri district, alongside the perception of strongly disagree, people also responded as agreed and strongly agreed to trade avian species for different human use purposes.

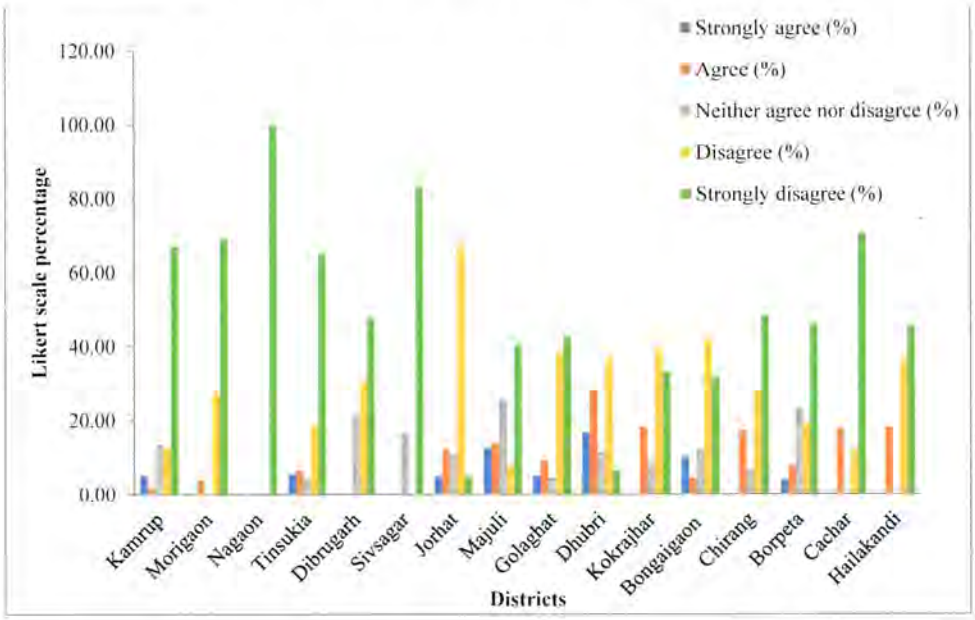

Figure 3.4: Perception of local people towards illegal avian trade in Assam.

3.4.5.2. Can avian species be protected through legislation?

The perception of local people towards conservation was appalling, and people believed that avian species could be protected by strict application of law and legislation (Figure 3.5). In all the surveyed districts, people strongly agreed to protect the avian species through law enforcement. The highest number of respondents from the Sivsagar district strongly agreed with the proper and strict execution of legislation. Most respondents also

expressed the fear of enforcement agencies that can partially stop the open avian trade in the markets. They have also mentioned the fear of common people being put in jail or punished by forest authorities for wildlife crimes, especially trading/hunting/poaching avian species for different human uses. The respondents were categorically positive in selecting the strongly agreed option wherein the legislation is critical for protecting the avian species of Assam.

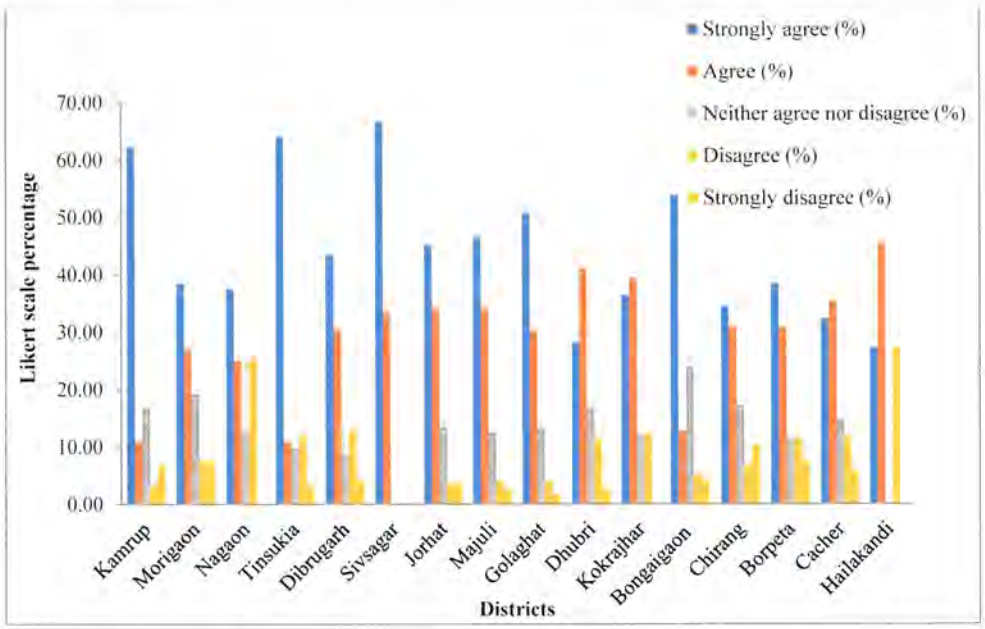

Figure 3.5: Perception of local people for conservation of avian species using strict legislation.

3.4.5.3. Can NGOs help protect wildlife/avian species?

Most of the respondents were aware of the NGOs and their work. They mentioned the awareness programs conducted by the different active NGOs in Assam, namely Aaranyak, Nature's Beckon, Nature's Foster, Korbi, Karpu etc. The respondents strongly agreed with the positive impact of NGOs' work on conserving wildlife/avian species (Figure 3.6). The impact of local NGOs on developing awareness among the local people towards wildlife in certain districts was seen during our surveys. Respondents were well aware of the local conservation heroes involved in different conservation and awareness programs associated with these NGOs. They are also of the opinion that when the local people get involved in conservation action through these NGOs, the wildlife crime rate

reduces as no one wants to harm their property, either their personal belongings or wild animals. The local people were also informed about different conservation issues and their importance by the researchers and scientists working on different aspects of wildlife conservation in their locality. Our results also showed that the upper, central and lower part of Assam is well aware of conservation and related other issues, and people do believe in the conservation-related works done by different NGOs. However, surprisingly respondents from Hailakandi districts responded as neither agree (50%) nor disagree (50%) on the fruitfulness of NGO's activities.

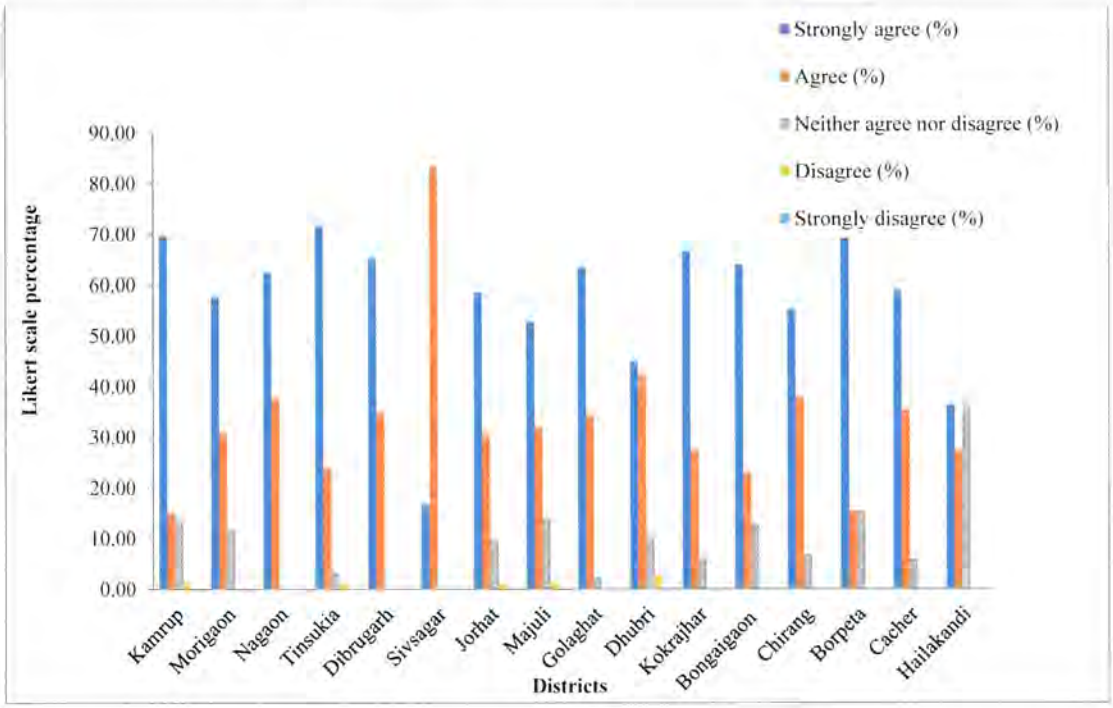

Figure 3.6: Perception of local people on NGO's work for curbing illegal avian trade.

3.4.5.4. Frequency of respondents aware of birds being traded

The local people graded the encounter frequencies of traded bird species as Very frequently, Frequently, Occasionally, Rarely, and Never (Figure 3.7). Respondents from all districts were informed about the encounters or were aware of the birds being traded "Very frequently" and "Frequently". Respondents from the Tinsukia district responded to having encountered avian species traded Very frequently. Respondents from the Nagaon district responded to having encountered these illegally traded avian species highest frequently, and respondents from the Kokrajhar district encountered the highest

Occasionally. Respondents from the Chirang district responded as they encountered avian species trade rarely. A few respondents from the four districts, i.e., Kamrup, Morigaon, Jorhat and Bongaigaon, responded that they had never heard of or encountered the birds being traded.

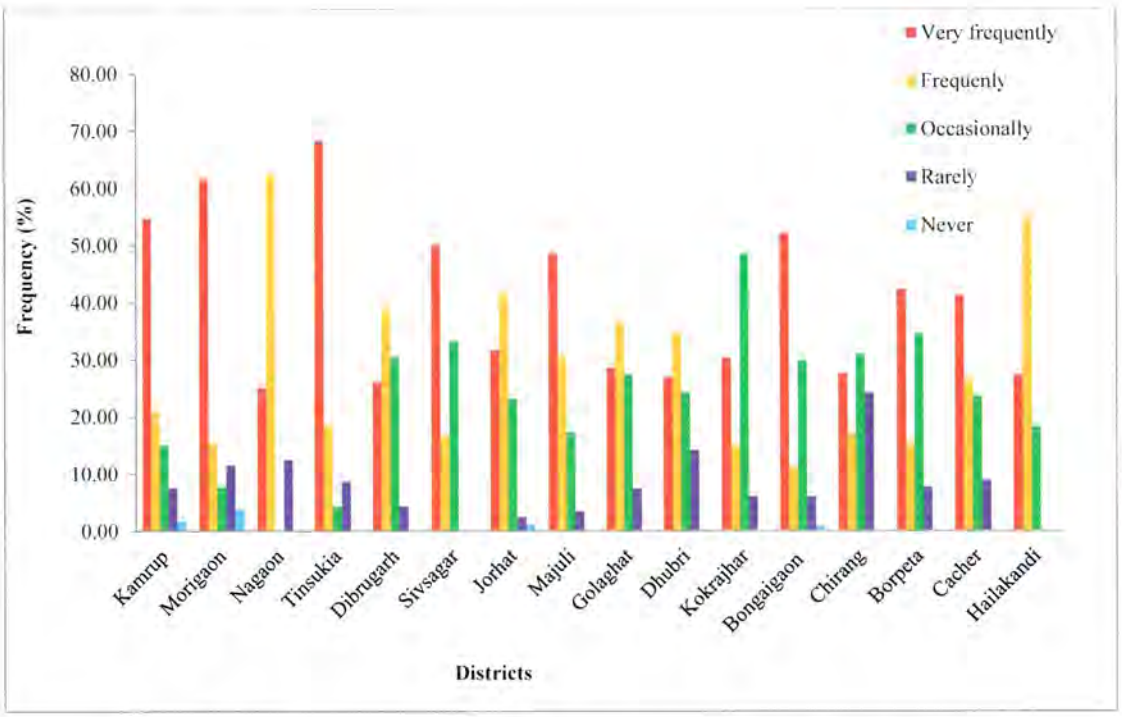

Figure 3.7: Frequency of respondents/districts aware of bird trade.

3.5. Discussion

Illegal trade is one of the important reasons for the ongoing sixth mass extinction with its direct contribution towards population losses (Ceballose *et al.*, 2017), and it is a major conservation threat to biodiversity (Utprey *et al.*, 2021). Nevertheless, on hunting and poaching, only a few works are reported from North East India that showed the illegal avian trade as a serious threat to biodiversity and conservation in the region (Hilaludin *et al.*, 2005; Aiyadurai *et al.*, 2010; Aiyadurai 2011; Mazumdar *et al.*, 2014). Our study is the first systematic attempt to bring to light the ongoing ground-level scenario of illegally traded avian species from Assam. The high numbers of traded bird species also directly reflect the failure concerning avian species conservation in the north-east region of India.

Continuous loss of any species' population adversely impacts that particular region's biodiversity and seriously threatens conservation (Aiyadurai *et al.*, 2010; Aiyadurai, 2011).

Species from the family Anatidae are traded highest in number as most of the birds from this family are sought after for bush-meat. Affluent people prefer to demonstrate their social status by adding bush-meat cuisines during festivities. It also signifies the need for further conservation mitigation measures towards migratory birds in the north-east, where hunting has been reported to be the major threat to the migratory species (Aiyadurai, 2012, Aiyadurai *et al.*, 2010). Bird species such as pigeons (Columbiformes) are also traded as "table bird" (Ahmed, 1997); however, they are less traded probably because it is more locally available. Species demand in trade for plumes is an old trend frequently reported in India (Watt, 1908). A few images presented in this chapter were captured in different parts of India to show the quantum of plume-use for different purposes (Image 3.3). Brightly coloured birds with specific patterns are under threat for this reason. Even feathers of ducks and other waterbird species were traded to be used in quilts, ornaments and other decorative products. Small birds like munias, buntings, and mynas are mostly traded due to their demand as pets (Ahmed, 1997). This high demand also threatens other locally abundant species as sometimes traders suitably colour these birds as required and sell them at high prices (Ahmed, 2002). Superstition, black magic and other traditional uses are also fuelling factors for illegal trade. Dried biological materials of many species are traded for use in traditional medicines (Utprey *et al.*, 2021). In traditional societies with strong belief systems, people usually observe their customary rituals/practices without much explicit or scientific reasoning. However, there will be definite folklore or myths associated with such practices. Keeping Barn owl (*Tyto alba*) at home during Laxmi puja, sacrificing Spotted owlet (*Athene brama*) for rain, and sacrificing Magpie Robin (*Copsychus saularis*) for Durga puja rituals are some of the common local beliefs that directly help illegal trade of avian species (Ahmed, 1999).

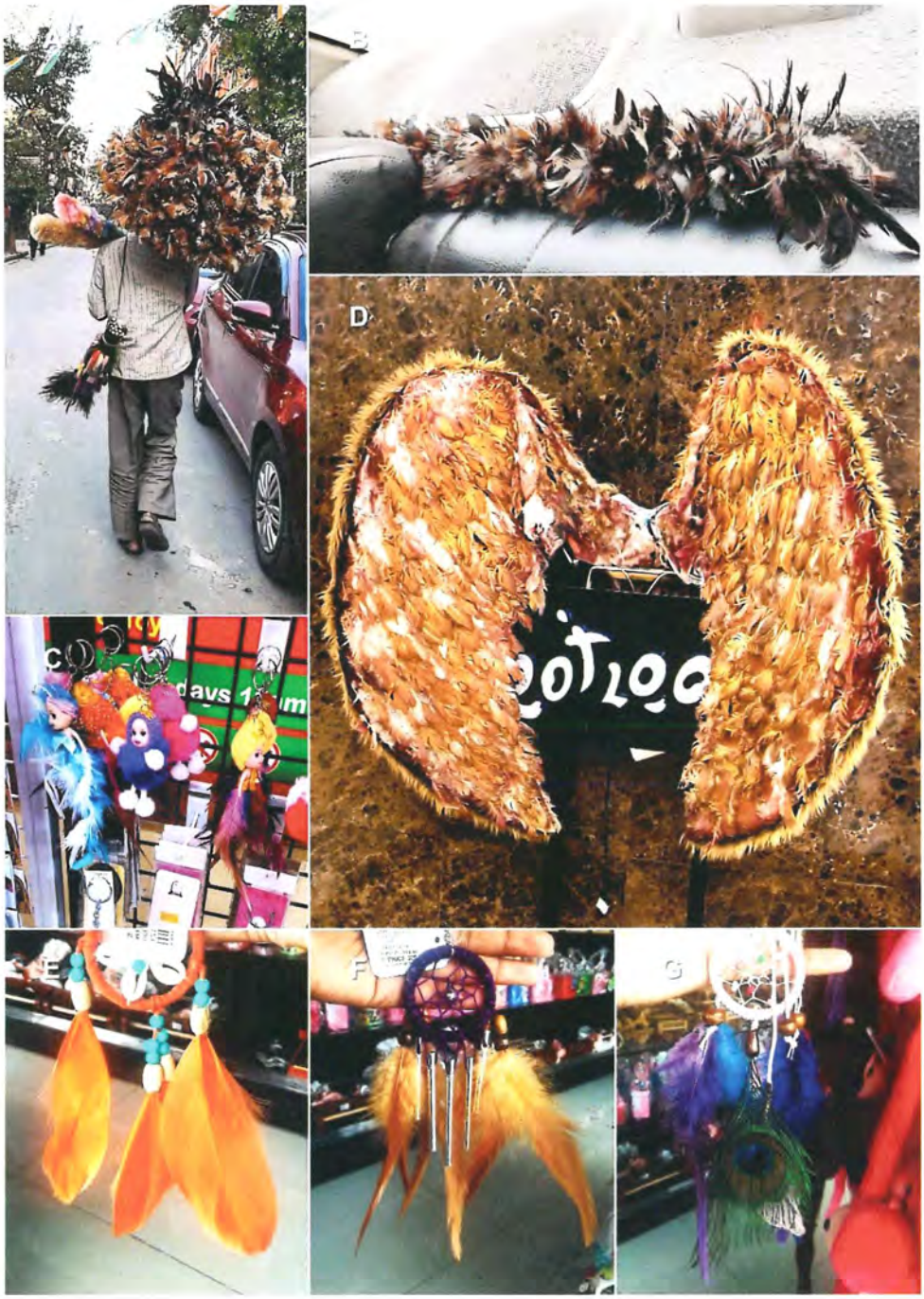

Image 3.3: Different uses of bird plumage all across India. A-Sold as a duster on the road-side (In Kolkata, West Bengal); B- Plumage-duster inside an Ola Cab (In Kolkata, West Bengal); C-Coloured feathers used to make key-chains (In Guwahati, Assam); D-Plumage used to prepare huge wings to showcase in front of cinema hall; E, F, G- Feathers used to prepare dream-catcher (In Coimbatore, Tamil Nadu).

Illegal trade is of transboundary nature and is firmly linked with efforts from other countries to tackle wildlife crimes (Lin, 2005). Assam shares its state boundaries with Arunachal, Meghalaya, Nagaland, Mizoram, Manipur, Tripura and West Bengal; and international boundaries with Bhutan and Bangladesh. Our study shows that the intensity of traded bird species increased in numbers in markets closer to Assam's state and international borders (Figure 3.8). Porous boundaries made it easy for traders and smugglers to organize transportation routes (Lin, 2005; Nijman *et al.*, 2016; Utprey *et al.*, 2021). Establishing frequent checking points by the concerned law enforcement agencies can reduce such transboundary issues. Keeping all these points in mind, sustainable conservation approaches and awareness programs need to be appropriately designed and implemented (Roe *et al.*, 2020; Utprey *et al.*, 2021) to control the menace of illegal trade.

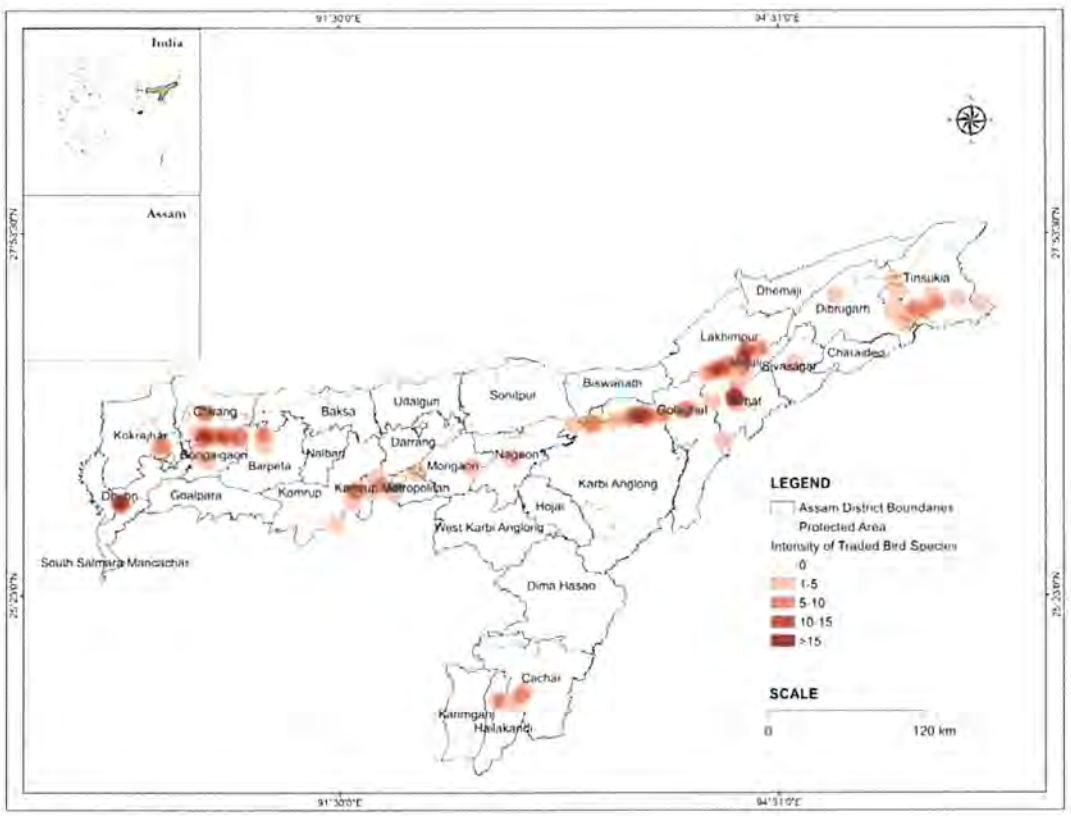

Figure 3.8: The intensity of traded avian species in Assam.

During our survey, we could gauge the well-organized functioning of the illegal trade, no matter how big or small the agencies involved are. The respondents revealed their clear understanding of wildlife laws, the consequences of illegal trade, and ways to dodge

the reach of the enforcement agencies. Such spread of information could also be an undesirable outcome of the active works by local NGOs and forest departments in the surveyed areas to promote wildlife conservation. Even though our results showed that local people's perception directly depends on local awareness, as seen in the border-line district like Hailakandi, where not much awareness work had been done, and no NGOs were active. It was observed that local people from those areas not exposed to the work of NGOs have no fear of any punishments for wildlife crimes. With a clear understanding of the law and wildlife crime among those involved, it is obvious that the wildlife trade is critically based on confirmed personal contacts. Hence, local communities can be crucial in curbing this well-organized illegal trade (Utprey *et al.*, 2021; Vasquaez, 2014). Further, education, intensive awareness programs, and prompt implementation of laws and regulations taking into confidence the local communities could only effectively curb the illegal trade in wild species.

3.6. Conclusion

A laxity in law enforcement allows traders to continue the avian trade even in daily markets. The high demand for species as bush-meat and pets is the major driving force. Our study provides critical information and cues to the forest departments, other law enforcement agencies, and researchers to identify and execute appropriate action plans to curb the illegal wildlife trade. Though some checklists and published knowledge cannot directly stop such unlawful trades (Ahmed, 1997), such details are essential to find better solutions to curtail wildlife crimes. Besides, strong conservation-oriented programs with the participation of the local communities are much required. The active participation of local communities has an effective and superior potentiality to curtail illegal unsustainable wildlife trade with their honest intention and determination (Esmail *et al.*, 2020; Fukushima *et al.*, 2021). Hence, conservation measures and enforcement of related laws must take into confidence the local indigenous communities, ensuring that they are also beneficiaries of the same, is greatly required.

Table 3.1: Surveyed markets in Assam along with GPS location in districts of Assam.

Sl. No.	Name of the Markets	Districts	Latitude	Longitude
1	Beltola Daily Market	Kamrup	26.12280	91.80567
2	Ganeshguri Daily Market	Kamrup	26.15060	91.78488
3	UzaanBozaar Daily Market	Kamrup	26.19280	91.75195
4	Borjhar Daily Market	Kamrup	26.10695	91.60490
5	Azara Chowk Bazaar Daily Market	Kamrup	26.11677	91.61451
6	Garhchuk Tiniaali Daily Market	Kamrup	26.10672	91.70808
7	Chandubi Chawk Market	Kamrup	25.87426	91.47020
8	Rani-Garhbhanga Daily Market	Kamrup	26.03470	91.58456
9	Jorabaat Market	Kamrup	26.09939	91.87628
10	Rani Gate Market	Kamrup	26.09968	91.61227
11	Kahikuchi Market	Kamrup	26.10632	91.60471
12	Morigaon Weekly Market	Morigaon	26.27128	92.41685
13	Morigaon Daily Market	Morigaon	26.23418	92.03523
14	Nagaon Weekly Market	Nagaon	26.33084	92.69166
15	Digboi Daily Market	Tinsukia	27.39547	95.61707
16	Soraipung Daily Market	Tinsukia	27.35257	95.51300
17	Daily Market (Near Digboi college)	Tinsukia	27.48853	95.35568
18	Maya Bozaar Daily Market	Tinsukia	27.35806	95.53361
19	Ouwguri Weekly Market	Tinsukia	27.36806	95.46778
20	Tingrai weekly Market	Tinsukia	27.45361	95.57972
21	Tingrai Tea State daily market	Tinsukia	27.56778	95.32250
22	Borbil tea State daily market	Tinsukia	27.40449	95.61683

Sl. No.	Name of the Markets	Districts	Latitude	Longitude
23	Bhadou-Pachoni Daily Market	Tinsukia	27.31782	95.41948
24	Jagun Daily Market	Tinsukia	27.40169	95.92251
25	Bozaloni Daily Market	Tinsukia	27.36917	95.46778
26	Lekhapani Daily market	Tinsukia	27.26472	95.39278
27	Milan Nagar Daily Market	Dibrugarh	27.46238	94.91455
28	Pengri Weekly Market	Dibrugarh	27.43871	95.75903
29	Duliajaan Weekly Market	Dibrugargh	27.35785	95.31404
30	Sivsagar Daily Market	Sivsagar	26.99447	94.62247
31	Garali Daily Market	Jorhat	26.75637	94.21923
32	Na Ali daily Market	Jorhat	26.74958	94.21236
33	Lichubari Market	Jorhat	26.72819	94.21333
34	Cinamora Daily Market	Jorhat	26.71119	94.23372
35	Rowriah Weekly Market	Jorhat	26.74958	94.21236
36	Jorhat Chawk Bazar Market	Jorhat	26.76196	94.21076
37	Tarajan Puja Mandir Market	Jorhat	26.76143	94.21060
38	Fancy Ali Market	Jorhat	26.76035	94.21366
39	Sankar Hall Local Market	Jorhat	26.76175	94.20933
40	Borhulla Daily Market	Jorhat	26.45379	94.13511
41	Nimati Ghat Daily Market	Jorhat	26.86149	94.24196
42	Kamalabari Daily Chawk Market	Majuli	26.94611	94.16528
43	Garhmur Daily Market	Majuli	26.97916	94.15783
44	Aauniati Chawk market	Majuli	26.93548	94.12281
45	Balichapori Daily market	Majuli	26.94917	94.08861

Sl. No.	Name of the Markets	Districts	Latitude	Longitude
46	Goal Gaon Tiniali Daily market	Majuli	26.92960	94.04973
47	Koroti Par High School market	Majuli	26.91750	94.01583
48	Mohotichuk Daily Market	Majuli	26.93889	94.06194
49	Kharjaan Daily Market	Majuli	26.94833	94.10806
50	Borgoya Weekly Market	Majuli	26.95639	94.12250
51	Borguri Chariaali Daily Market	Majuli	27.04168	94.27428
52	Phuloni Weekly Market	Majuli	27.04167	94.27361
53	Jengraimukh Chawk Daily Market	Majuli	27.08500	94.29028
54	Dhakuakhana Road Weekly Market	Majuli	27.08056	94.29667
55	Pohumora Nath Gaon Daily Market	Majuli	26.98750	94.26528
56	Chamguri Tiniaali Market	Majuli	26.95194	94.25806
57	Bongaon Chariaali Chawk Market	Majuli	26.95472	94.28750
58	Kotia Chawk Daily Market	Majuli	26.93889	94.06194
59	Jogi Gaon Daily Market	Majuli	26.99056	94.17722
60	Ratanpur Miri Daily Market	Majuli	27.08889	94.38417
61	Ratanpur Miri Weekly Market	Majuli	27.09278	94.39806
62	Naya Bozar Weekly Market	Majuli	27.08500	94.29056
63	Miri Tiniaali Daily Market	Majuli	27.08056	94.29667
64	Maya Bozar Daily Market	Majuli	27.04600	94.29504
65	Bokakhat Daily Market	Golaghat	26.64062	93.59942
66	Borjuri Daily Market	Golaghat	26.63264	93.54236
67	Subjuri Daily Market	Golaghat	26.63257	93.54957
68	Methoni Tea-State Daily Market	Golaghat	26.62703	93.53505

Sl. No.	Name of the Markets	Districts	Latitude	Longitude
69	Kuthori Daily Market	Golaghat	26.57482	93.22668
70	Jogodomba Tea State Weekly Market	Golaghat	26.56361	93.23444
71	Siljuri Weekly Market	Golaghat	26.61037	93.48249
72	Methoni Weekly Market	Golaghat	26.61796	93.51362
73	Borjuri Weekly Market	Golaghat	26.63225	93.54134
74	Aamtenga Weekly Market	Golaghat	26.62333	93.59361
75	Diffloo Weekly Market	Golaghat	26.60667	93.60806
76	Nahorjaan Daily Market	Golaghat	26.60972	93.60750
77	Nahorjaan Weekly Market	Golaghat	26.62771	93.61901
78	Deusur sang Daily Market	Golaghat	26.56889	93.12000
79	Behora Weekly Market	Golaghat	26.63194	93.71083
80	Kohora Daily Market	Golaghat	26.59000	93.40028
81	Bagori Daily Market	Golaghat	26.57711	93.27910
82	Goshanibar Daily Market	Golaghat	26.61750	93.51361
83	Kuthori Weekly Market	Golaghat	26.57639	93.25333
84	Burha Ali Chawk Daily Market	Golaghat	26.66361	93.86556
85	Baduli Para Daily Market	Golaghat	26.66375	93.86193
86	Sokolatinga Daily Market	Golaghat	26.72278	94.05722
87	A.T. Road Burhalikson Daily Market	Golaghat	26.66361	93.86556
88	Negheri-ting Chawk Market	Golaghat	26.63111	93.72694
89	Ronga Mati Daily Market	Golaghat	26.67406	93.90945
90	Jahaj Ghat Daily Market	Dhubri	26.01323	89.98718
91	Netai Dhubuni Ghat Daily Market	Dhubri	26.02238	89.99523

Sl. No.	Name of the Markets	Districts	Latitude	Longitude
92	Tamranga Beel Daily Market	Dhubri	26.31406	90.58250
93	Pet Shop Gauripur Town Market	Dhubri	26.01882	89.98748
94	Boro Bazar Daily Market	Dhubri	26.02283	89.97905
95	Jugomaya Ghat	Dhubri	26.01518	89.99212
96	Pet Shop Boro Bazar	Dhubri	26.01287	89.98433
97	Ponchu Ghat Daily Market	Dhubri	26.02272	89.99528
98	Bou Bazar Market	Kokrajhar	26.39947	90.26948
99	Daily Local Market	Kokrajhar	26.41600	90.25967
100	Ganjema Bazaar	Kokrajhar	26.41871	90.27770
101	Kanya Mahabidyalaya Daily Market	Bongaigaon	26.47179	90.56095
102	Swaheed Bedi Daily Market	Bongaigaon	26.46904	90.55950
103	Saprakata Daily Market	Bongaigaon	26.48904	90.61335
104	Borpara Daily Market	Bongaigaon	26.46825	90.55837
105	Hapachara Daily Market	Bongaigaon	26.49194	90.64611
106	Goroimari Daily Market	Bongaigaon	26.47795	90.67082
107	Paglasthan flyover Daily Market	Bongaigaon	26.47859	90.55440
108	Manikpur Weekly Market	Bongaigaon	26.47013	90.80152
109	Bhatipara Daily Market	Bongaigaon	26.46693	90.79231
110	Patiladoha Daily Market	Bongaigaon	26.49667	90.79972
111	Patiladoha Weekly Market	Bongaigaon	26.49642	90.79912
112	Chaprakata Daily Market	Bongaigaon	26.48915	90.59727
113	New Bongaigaon IOCL Colony Market	Bongaigaon	26.50839	90.52206
114	Amguri Bazaar	Bongaigaon	26.64106	90.56732

Sl. No.	Name of the Markets	Districts	Latitude	Longitude
115	Amteka Bazaar	Bongaigaon	26.64203	90.56805
116	Chapaguri Bazaar	Bongaigaon	26.50501	90.55432
117	Boitamari Weekly Market	Bongaigaon	26.34856	90.51932
118	Bijni Daily Market	Chirang	26.49695	90.69865
119	Dangaigaon Daily Market	Chirang	26.47056	90.73417
120	Purona-Bijni Daily Market	Chirang	26.48891	90.71452
121	Gas-guri Daily Market	Chirang	26.47547	90.68531
122	Simlaguri Daily Market	Borpeta	26.49574	90.97016
123	Howly Chawk Daily Market	Borpeta	26.42482	90.97415
124	Borpeta Road Daily Market	Borpeta	26.50069	90.96794
125	Irongmara Market	Cachar	24.68821	92.74234
126	Durgakona Market	Cachar	24.69898	92.75699
127	Silcoori Market	Cachar	24.71859	92.77703
128	Dwarbond Market	Cachar	24.63162	92.70946
129	Matijuri Bazaar	Hailakandi	24.64771	92.60900
130	Tempur Bazaar	Hailakandi	24.66078	92.59153

Table 3.2: Avian species traded from Assam along with their conservation status.

Sl. No.	Order	Family	English Name	Scientific Name	IUCN Category	WPA Schedule	Migration Time	Vernacular Name(s)
1	Anseriformes	Anatidae	Fulvous Whistling Duck	*Dendrocygna bicolor*	Least Concern	Schedule-I	Resident	-
2	Anseriformes	Anatidae	Lesser Whistling Duck	*Dendrocygna javanica*	Least Concern	Schedule-IV	Resident	Sorali/Xorali
3	Anseriformes	Anatidae	Bar-headed Goose	*Anser indicus*	Least Concern	Schedule-IV	Winter Visitor	Bornooria hanh, Boga rajhanh
4	Anseriformes	Anatidae	Greylag Goose	*Anser anser*	Least Concern	Schedule-IV	Winter Visitor	Raaj hanh, Dhitraj
5	Anseriformes	Anatidae	Ruddy Shelduck	*Tadorna ferruginea*	Least Concern	Schedule-IV	Winter Visitor	Ramkong, Chakoi-chakoua
6	Anseriformes	Anatidae	Gadwall	*Mareca strepera*	Least Concern	Schedule-IV	Winter Visitor	Saru/Xaru mugi hanh
7	Anseriformes	Anatidae	Indian Spot-billed Duck	*Anas poecilorhyncha*	Least Concern	Schedule-IV	Resident	Bor mugi hanh
8	Anseriformes	Anatidae	Mallard	*Anas platyrhynchos*	Least Concern	Schedule-IV	Winter Visitor	Amrolia hanh, Bonaria pati hanh
9	Anseriformes	Anatidae	Northern Pintail	*Anas acuta*	Least Concern	Schedule-IV	Winter Visitor	Najal hanh, Neji hanh, Dighal

Sl. No.	Order	Family	English Name	Scientific Name	IUCN Category	WPA Schedule	Migration Time	Vernacular Name(s)
10	Anseriformes	Anatidae	Common Teal	*Anas crecca*	Least Concern	Schedule-IV	Winter Visitor	Kalimari, Chila hanh, Patari hanh
11	Anseriformes	Anatidae	Cotton Teal	*Nettapus coromandelianus*	Least Concern	Schedule-IV	Resident	Ghila hanh, Naher, Keeke, Chuwa
12	Galliformes	Phasianidae	Common Quail	*Coturnix coturnix*	Least Concern	Schedule-IV	Winter Visitor	Bota sorai
13	Galliformes	Phasianidae	Blue-breasted Quail	*Synoicus chinensis*	Least Concern	Schedule-IV	Widespread resident	-
14	Galliformes	Phasianidae	Black Francolin	*Francolinus francolinus*	Least Concern	Schedule-IV	Resident	Tetri sorai, Mechentri
15	Galliformes	Phasianidae	Swamp Francolin	*Francolinus gularis*	Vulnerable	Schedule-IV	Resident	Koi, Koi sorai, Koira, Hoi koli
16	Galliformes	Phasianidae	Red Junglefowl	*Gallus gallus*	Least Concern	Schedule-IV	Resident	Ban kukura
17	Columbiformes	Columbidae	Spotted Dove	*Streptopelia chinensis*	Least Concern	Schedule-IV	Resident	Pati kopou
18	Columbiformes	Columbidae	Yellow-legged Green Pigeon	*Treron phoenicopterus*	Least Concern	Schedule-IV	Resident	Haitha, Bor haita
19	Columbiformes	Columbidae	Asian Emerald Dove	*Chalcophaps indica*	Least Concern	Schedule-IV	Resident	Mati kupou, Sil kopou

Sl. No.	Order	Family	English Name	Scientific Name	IUCN Category	WPA Schedule	Migration Time	Vernacular Name(s)
20	Gruiformes	Rallidae	White-breasted Waterhen	*Amaurornis phoenicurus*	Least Concern	Schedule-IV	Resident	Dauk
21	Gruiformes	Rallidae	Purple Swamphen	*Porphyrio porphyrio*	Least Concern	Schedule-IV	Resident	Kaam sorai
22	Gruiformes	Rallidae	Common Moorhen	*Gallinula chloropus*	Least Concern	Schedule-IV	Resident	-
23	Gruiformes	Rallidae	Common Coot	*Fulica atra*	Least Concern	Schedule-IV	Resident	-
24	Pelecaniformes	Ardeidae	Cinnamon Bittern	*Ixobrychus cinnamomeus*	Least Concern	Schedule-IV	Resident	Itaguria
25	Pelecaniformes	Ardeidae	Black-crowned Night Heron	*Nycticorax nycticorax*	Least Concern	Schedule-IV	Resident	Waak chorai
26	Pelecaniformes	Ardeidae	Striated Heron	*Butorides striata*	Least Concern	Schedule-IV	Resident	-
27	Pelecaniformes	Ardeidae	Indian Pond Heron	*Ardeola grayii*	Least Concern	Schedule-IV	Resident	Kona moochuree
28	Pelecaniformes	Ardeidae	Cattle Egret	*Bubulcus ibis*	Least Concern	Schedule-IV	Resident	Go bog
29	Pelecaniformes	Ardeidae	Intermediate Egret	*Ardea intermedia*	Least Concern	Schedule-IV	Resident	Maju bog

Sl. No.	Order	Family	English Name	Scientific Name	IUCN Category	WPA Schedule	Migration Time	Vernacular Name(s)
30	Pelecaniformes	Ardeidae	Little Egret	*Egretta garzetta*	Least Concern	Schedule-IV	Resident	Bamoni bog, Teteri bog
31	Suliformes	Phalacrocoracidae	Little Cormorant	*Microcarbo niger*	Least Concern	Schedule-IV	Resident	Pani kaori
32	Suliformes	Anhingidae	Oriental Darter	*Anhinga melanogaster*	Near Threatened	Schedule-IV	Resident	Maniori, Bejiagir
33	Charadriiformes	Charadriidae	Red-wattled Lapwing	*Vanellus indicus*	Least Concern	Schedule-IV	Resident	Balighora, Tetatua
34	Charadriiformes	Jacanidae	Pheasant-tailed Jacana	*Hydrophasianus chirurgus*	Least Concern	Schedule-IV	Resident	Dol mora
35	Charadriiformes	Jacanidae	Bronze-winged Jacana	*Metopidius indicus*	Least Concern	Schedule-IV	Resident	Dol punga
36	Charadriiformes	Turnicidae	Barred Buttonquail	*Turnix suscitator*	Least Concern	Schedule-IV	Resident	Sansorai
37	Strigiformes	Tytonidae	Common Barn Owl	*Tyto alba*	Least Concern	Schedule-IV	Resident	Lakshmi fecha
38	Strigiformes	Strigidae	Spotted Owlet	*Athene brama*	Least Concern	Schedule-IV	Resident	Fecha
39	Strigiformes	Strigidae	Collared Scops Owl	*Otus lettia*	Least Concern	Schedule-IV	Resident	-

Sl. No.	Order	Family	English Name	Scientific Name	IUCN Category	WPA Schedule	Migration Time	Vernacular Name(s)
40	Strigiformes	Strigidae	Brown Fish Owl	*Ketupa zeylonensis*	Least Concern	Schedule-IV	Resident	Hoodoo
41	Piciformes	Picidae	Grey-headed Woodpecker	*Picus canus*	Least Concern	Schedule-IV	Resident	-
42	Piciformes	Picidae	Greater Golden-backed Woodpecker	*Chrysocolaptes guttacristatus*	Least Concern	Schedule-IV	Resident	Barhoitoka
43	Piciformes	Megalaimidae	Blue-throated Barbet	*Psilopogon asiaticus*	Least Concern	Schedule-IV	Resident	Tuktukra sorai
44	Piciformes	Megalaimidae	Coppersmith Barbet	*Psilopogon haemacephalus*	Least Concern	Schedule-IV	Resident	Basanta sorai, Hetuluka
45	Coraciiformes	Meropidae	Green Bee-eater	*Merops orientalis*	Least Concern	Schedule-IV	Resident	Harial sorai
46	Coraciiformes	Coraciidae	Indian Roller	*Coracias benghalensis*	Least Concern	Schedule-IV	Resident	Nilkantho, Katnas, Konsa
47	Coraciiformes	Alcedinidae	Common Kingfisher	*Alcedo atthis*	Least Concern	Schedule-IV	Resident	-
48	Coraciiformes	Alcedinidae	White-throated Kingfisher	*Halcyon smyrnensis*	Least Concern	Schedule-IV	Resident	Masroka
49	Coraciiformes	Alcedinidae	Pied Kingfisher	*Ceryle rudis*	Least Concern	Schedule-IV	Resident	Pokhora masroka

Sl. No.	Order	Family	English Name	Scientific Name	IUCN Category	WPA Schedule	Migration Time	Vernacular Name(s)
50	Psittaciformes	Psittaculidae	Grey-headed Parakeet	*Psittacula finschii*	Near Threatened	Schedule-IV	Resident	-
51	Psittaciformes	Psittaculidae	Red-breasted Parakeet	*Psittacula alexandri*	Near Threatened	Schedule-IV	Resident	-
52	Psittaciformes	Psittaculidae	Alexandrine Parakeet	*Psittacula eupatria*	Near Threatened	Schedule-IV	Resident	-
53	Psittaciformes	Psittaculidae	Rose-ringed Parakeet	*Psittacula krameri*	Least Concern	Schedule-IV	Resident	Bhatov sorai
54	Passeriformes	Campephagidae	Scarlet Minivet	*Pericrocotus flammeus*	Least Concern	Schedule-IV	Resident	-
55	Passeriformes	Aegithinidae	Common Iora	*Aegithina tiphia*	Least Concern	Schedule-IV	Resident	-
56	Passeriformes	Dicruridae	Bronzed Drongo	*Dicrurus aeneus*	Least Concern	Schedule-IV	Resident	-
57	Passeriformes	Dicruridae	Lesser Racket-tailed Drongo	*Dicrurus remifer*	Least Concern	Schedule-IV	Resident	Bhimraj
58	Passeriformes	Dicruridae	Black Drongo	*Dicrurus macrocercus*	Least Concern	Schedule-IV	Resident	Phenchu, Dhenchu sorai
59	Passeriformes	Dicruridae	Hair-crested Drongo	*Dicrurus hottentottus*	Least Concern	Schedule-IV	Resident	-
60	Passeriformes	Corvidae	Rufous Treepie	*Dendrocitta vagabunda*	Least Concern	Schedule-IV	Resident	Kalo khoa, Chekcheki

Sl. No.	Order	Family	English Name	Scientific Name	IUCN Category	WPA Schedule	Migration Time	Vernacular Name(s)
61	Passeriformes	Corvidae	House Crow	*Corvus splendens*	Least Concern	Schedule-V	Resident	Paati kaori
62	Passeriformes	Nectariniidae	Purple Sunbird	*Cinnyris asiaticus*	Least Concern	Schedule-IV	Resident	Moupiya sorai
63	Passeriformes	Chloropseidae	Golden-fronted Leafbird	*Chloropsis aurifrons*	Least Concern	Schedule-IV	Resident	-
64	Passeriformes	Chloropseidae	Orange-bellied Leafbird	*Chloropsis hardwickii*	Least Concern	Schedule-IV	Resident	-
65	Passeriformes	Ploceidae	Baya Weaver	*Ploceus philippinus*	Least Concern	Schedule-IV	Resident	Tookora sorai
66	Passeriformes	Ploceidae	Finn's Weaver	*Ploceus megarhynchus*	Vulnerable	Schedule-IV	Resident	-
67	Passeriformes	Estrildidae	Scaly-breasted Munia	*Lonchura punctulata*	Least Concern	Schedule-IV	Resident	Tooni sorai
68	Passeriformes	Passeridae	House Sparrow	*Passer domesticus*	Least Concern	Schedule-IV	Resident	Ghon chirika
69	Passeriformes	Motacillidae	Grey Wagtail	*Motacilla cinerea*	Least Concern	Schedule-IV	Winter Visitor	Haldiya balimahi
70	Passeriformes	Motacillidae	Citrine Wagtail	*Motacilla citreola*	Least Concern	Schedule-IV	Winter Visitor	Tooni
71	Passeriformes	Motacillidae	White Wagtail	*Motacilla alba*	Least Concern	Schedule-IV	Winter Visitor	Tipochi

Sl. No.	Order	Family	English Name	Scientific Name	IUCN Category	WPA Schedule	Migration Time	Vernacular Name(s)
72	Passeriformes	Motacillidae	Western Yellow Wagtail	*Motacilla flava*	Least Concern	Schedule-IV	Winter Visitor	-
73	Passeriformes	Cisticolidae	Common Tailorbird	*Orthotomus sutorius*	Least Concern	Schedule-IV	Resident	Paatxla sorai
74	Passeriformes	Pycnonotidae	Red-vented Bulbul	*Pycnonotus cafer*	Least Concern	Schedule-IV	Resident	Bulbuli sorai
75	Passeriformes	Pycnonotidae	Red-whiskered Bulbul	*Pycnonotus jocosus*	Least Concern	Schedule-IV	Resident	-
76	Passeriformes	Leiothrichidae	Jungle Babbler	*Turdoides striata*	Least Concern	Schedule-IV	Resident	Xaatbhani
77	Passeriformes	Sturnidae	Asian Pied Starling	*Gracupica contra*	Least Concern	Schedule-IV	Resident	Kan kuriha
78	Passeriformes	Sturnidae	Common Myna	*Acridotheres tristis*	Least Concern	Schedule-IV	Resident	Salika sorai, Salika, Ghor salika
79	Passeriformes	Sturnidae	Bank Myna	*Acridotheres ginginianus*	Least Concern	Schedule-IV	Resident	-
80	Passeriformes	Sturnidae	Jungle Myna	*Acridotheres fuscus*	Least Concern	Schedule-IV	Resident	-
81	Passeriformes	Sturnidae	Common Hill Myna	*Gracula religiosa*	Least Concern	Schedule-I	Resident	Maina
82	Passeriformes	Muscicapidae	Oriental Magpie Robin	*Copsychus saularis*	Least Concern	Schedule-IV	Resident	Dahikataraa

Table 3.3: Top 15 traded avian species and their price range.

Sl. No	Species Name	Scientific Name	Number of Respondents	Average price (Rs)	Price Range (Rs)
1	Red-breasted Parakeet	*Psittacula alexandri*	67	775	550–1000
2	Rose-ringed Parakeet	*Psittacula krameri*	75	750	500–1000
3	Alexandrine Parakeet	*Psittacula eupatria*	67	700	500–900
4	Common Hill Myna	*Gracula religiosa*	58	675	450–900
5	Common Barn Owl	*Tyto alba*	65	600	500–700
6	Spotted Owlet	*Athene brama*	60	600	550–650
7	Lesser Racket-tailed Drongo	*Dicrurus remifer*	20	550	500–600
8	Grey-headed Parakeet	*Psittacula finschii*	67	525	500–550
9	Fulvous Whistling Duck	*Dendrocygna bicolor*	57	510	450–570
10	Lesser Whistling Duck	*Dendrocygna javanica*	57	510	450–570
11	Common Myna	*Acridotheres tristis*	59	475	450–500
12	Bank Myna	*Acridotheres ginginianus*	33	475	450–500
13	Jungle Myna	*Acridotheres fuscus*	33	475	450–500
14	Black Drongo	*Dicrurus macrocercus*	38	475	450–500
15	Greylag Goose	*Anser anser*	20	475	450–500

Table 3.4: Perception of local people for conservation issues using Likert scale.

Districts	Likert scale				
	Strongly agree (%)	Agree (%)	Neither agree nor disagree (%)	Disagree (%)	Strongly disagree (%)
Should avian species be traded for different human use?					
Kamrup	5.04	1.68	13.45	12.61	67.23
Morigaon	0.00	3.85	0.00	26.92	69.23
Nagaon	0.00	0.00	0.00	0.00	100.00
Tinsukia	5.43	6.52	4.35	18.48	65.22
Dibrugarh	0.00	0.00	21.74	30.43	47.83
Sivsagar	0.00	0.00	16.67	0.00	83.33
Jorhat	4.88	12.20	10.98	67.07	4.88
Majuli	12.50	13.89	25.69	7.64	40.28
Golaghat	5.14	9.14	4.57	38.29	42.86
Dhubri	16.67	28.21	11.54	37.18	6.41
Kokrajhar	0.00	18.18	9.09	39.39	33.33
Bongaigaon	10.26	4.27	11.97	41.88	31.62
Chirang	0.00	17.24	6.90	27.59	48.28
Borpeta	3.85	7.69	23.08	19.23	46.15
Cachar	0.00	17.65	0.00	11.76	70.59
Hailakandi	0.00	18.18	0.00	36.36	45.45
Can avian species be protected through legislation?					
Kamrup	62.18	10.92	16.81	3.36	6.72
Morigaon	38.46	26.92	19.23	7.69	7.69
Nagaon	37.50	25.00	12.50	25.00	0.00
Tinsukia	64.13	10.87	9.78	11.96	3.26
Dibrugarh	43.48	30.43	8.70	13.04	4.35
Sivsagar	66.67	33.33	0.00	0.00	0.00
Jorhat	45.12	34.15	13.41	3.66	3.66

Districts	Likert scale				
	Strongly agree (%)	Agree (%)	Neither agree nor disagree (%)	Disagree (%)	Strongly disagree (%)
Majuli	46.53	34.03	12.50	4.17	2.78
Golaghat	50.86	30.29	13.14	4.00	1.71
Dhubri	28.21	41.03	16.67	11.54	2.56
Kokrajhar	36.36	39.39	12.12	12.12	0.00
Bongaigaon	53.85	12.82	23.93	5.13	4.27
Chirang	34.48	31.03	17.24	6.90	10.34
Borpeta	38.46	30.77	11.54	11.54	7.69
Cacher	32.35	35.29	14.71	11.76	5.88
Hailakandi	27.27	45.45	0.00	27.27	0.00
Can NGO's be helpful to protect wildlife/avian species?					
Kamrup	69.75	15.13	13.45	1.68	0.00
Morigaon	57.69	30.77	11.54	0.00	0.00
Nagaon	62.50	37.50	0.00	0.00	0.00
Tinsukia	71.74	23.91	3.26	1.09	0.00
Dibrugarh	65.22	34.78	0.00	0.00	0.00
Sivsagar	16.67	83.33	0.00	0.00	0.00
Jorhat	58.54	30.49	9.76	1.22	0.00
Majuli	52.78	31.94	13.89	1.39	0.00
Golaghat	63.43	34.29	2.29	0.00	0.00
Dhubri	44.87	42.31	10.26	2.56	0.00
Kokrajhar	66.67	27.27	6.06	0.00	0.00
Bongaigaon	64.10	23.08	12.82	0.00	0.00
Chirang	55.17	37.93	6.90	0.00	0.00
Borpeta	69.23	15.38	15.38	0.00	0.00
Cacher	58.82	35.29	5.88	0.00	0.00
Hailakandi	36.36	27.27	36.36	0.00	0.00

Districts	Likert scale				
	Strongly agree (%)	Agree (%)	Neither agree nor disagree (%)	Disagree (%)	Strongly disagree (%)
Frequency of respondents/district aware about birds being traded					
Districts	**Very Frequently**	**Frequently**	**Occasionally**	**Rarely**	**Never**
Kamrup	54.62	21.01	15.13	7.56	1.68
Morigaon	61.54	15.38	7.69	11.54	3.85
Nagaon	25.00	62.50	0.00	12.50	0.00
Tinsukia	68.48	18.48	4.35	8.70	0.00
Dibrugarh	26.09	39.13	30.43	4.35	0.00
Sivsagar	50.00	16.67	33.33	0.00	0.00
Jorhat	31.71	41.46	23.17	2.44	1.22
Majuli	48.61	30.56	17.36	3.47	0.00
Golaghat	28.57	36.57	27.43	7.43	0.00
Dhubri	26.92	34.62	24.36	14.10	0.00
Kokrajhar	30.30	15.15	48.48	6.06	0.00
Bongaigaon	52.14	11.11	29.91	5.98	0.85
Chirang	27.59	17.24	31.03	24.14	0.00
Borpeta	42.31	15.38	34.62	7.69	0.00
Cacher	41.18	26.47	23.53	8.82	0.00
Hailakandi	27.27	54.55	18.18	0.00	0.00

Chapter 4

Chapter 4

Objective-2: Identify feather microstructures for select traded avian species

4.1. Introduction

The most eye-catching and morphometrically distinguishing characteristic of avian species is their feathers that cover the entire body (Gill, 1995). Like hair in mammals, different types of feathers are distributed in clearly defined tracts throughout the avian body (Clark & Allen, 1893). Even more, feather morphology varies depending upon the different colours present on the feathers too (Gandow, 1882). Feathers are composed of tubular epidermal beta-keratin structures that are only found in birds, and these structures combine to form a single but complex structure (Terrill & Shultz, 2022). An interesting but seemingly clear statement was made on the functional origin of feathers that, originally, feathers were not evolved for flight (Terrill & Shultz, 2022). Whereas documentation of feathers in fossils (i.e., in avian progenitors) proved that plumulaceous (non-flight) feathers preceded pennaceous (flight) feathers (Harris *et al.*, 2005). The presence of feathers on the fossil specimens of theropods provides "unambiguous" proof that dinosaurs are the ancestors of birds (Qiang *et al.*, 1998; Dove, 2000). By evolving for the performance of the different novel function, feathers achieve these functional structures through basic structural modifications (Terrill & Shultz, 2022). Even more, each different feather type performs specific functions (Rohwer *et al.*, 2021). In another way, feathers perform multiple functions, enabled through diverse evolutionary mechanisms (Stettenheim, 2000; Harris *et al.*, 2005; Terrill & Shultz, 2022).

Feathers of modern birds are now one of the most complicated integumentary derivatives with the hierarchical branches in their formation, i.e., of rachis, barbs, and barbules with the proximal calamus (Lucas & Stettenheim, 1972; Prum & Dyck, 2003). Previous research work stated that the contour (covering) feathers are the most efficient in providing informative characteristics for differentiation among the different taxa of birds (Dove, 2000a; Dove & Peurach, 2010; Lee *et al.*, 2015). Despite its applied importance, Plumology was not explored like other branches of science, and hence pterilography remained "A neglected branch of ornithology" (Clark & Allen, 1893). Now-a-days,

extensive studies on feathers (morphological and microscopic characters) have proven plumology to be useful in various branches of science, including palaeontology, archaeology, feeding habits of different predators and prey-remains, forensics, food contamination, wildlife law enforcement and in recent years to solve bird-strike cases (Dove, 1997). Still, identifying unknown species from feather samples remains "case-specific" based on the amount and types of feather samples and circumstantial evidence (Dove, 1997). Even before microscopic studies, species were identified from feather samples by matching them with known reference samples or museum specimens for a few corresponding macroscopic characters such as colour, pattern, shape, texture, size etc. Yet, for comparing specific microscopic characters between species from the same genus or family and variations in species, it is essential to have a reference collection of micro slides (Dove, 1997; Dove, 2000; Lee *et al.*, 2015). In the case of identification and characterization of avian species samples from wildlife crime spots, the study of feathers' morphological characteristics and examination of microscopic characters can directly help determine the source, assess its status under key national protection categories for illegal hunting, transportation and help resolve trafficking cases of wild birds (Jiang, 2019).

4.2. Review of literature

The study on feathers started long back by Nitzsch in 1876, who described different parts of feathers well with drawings. Following Nitzsch, Wray published significant works on the structure of pennaceous feathers in 1887. The systematic study of feather microstructure and their taxonomic significance were later pursued by Chandler (1916). He was the one who pioneered the systematic works on the microscopic variation of downy feather barbs among large groups of different bird species. The detailed work done by Chandler on the taxonomic significance of microscopic structures of both downy and pennaceous feathers was based on the very first study of Nitzsch (1867) but more systematically.

The study of feather microstructures for species identification gained importance as an applied science in the early 1960s (Dove, 2000). Studies on feathers from inadequate samples (samples from bird-strike, pray remains etc.) were started for species identification in the early sixties (Messinger, 1965; Day, 1966; Gilbert & Nancekivell, 1982). During

that time, Roxie Laybourne, a researcher at the Smithsonian Institution, was called upon to identify bird remains recovered from a Lockheed Electra aircraft crash at Logan International Airport (Lipske, 1982). When the reason for that crash (which killed 62 people) was identified as European Starlings (*Sturnus vulgaris*), the aviation industry began to search for solutions to control the bird strikes in the airfields to reduce damages (Dove, 2000). In the vast field of forensic science, various feather fragments were used for species identification by studying their micromorphology and were considered evidence for conviction by the courts of law (Davies, 1970, Deedrick & Mullery, 1981). Identification of vulture in collision with aircraft at high altitude was reported by Laybourne (1974). Laybourne (1984, 1986) and Brom (1986a) reported on the variations in microstructures of downy barbules of feathers collected at bird-aircraft incidents. The diagnostic and phylogenetic significance of different types of feather structures were well documented by Brom (1991), mainly focusing on feather remains to identify species after the bird's strike. In such case, functional microstructures that present on feather remaining are the keys for diagnosis and also to resolve bird's phylogenetic quarries (Brom, 1990; Brom & Frank, 1992). Laybourne *et al.* (1992) and Laybourne & Dove (1994) documented basic techniques for preparing feathers for species identification, especially from bird-strike remains. Subsequently, systematic feather microstructure works were done for various specific groups of birds by Dove (1997). These microstructures are also useful for museum samples (Dove, 1998). Historic evidence about the use of bird feathers was brought out from feather fragments from archaeological sites in Australia (Robertson, 2002). The use of feather microstructures in species identification and successful case studies demonstrating the importance of plumology in forensic ornithology were reported by Dove (2000b). Identification of feather microstructure alongside with DNA barcode was used to investigate bird-strike accidents in airport (Dove *et al.*, 2009). Feather forensics' practical significance is in bringing reliable proof for courts from the crime scene (Dove & Koch, 2010).

In the 2000s, major works on feather microstructure focused on specific bird groups, particular orders or families. Birds were even identified using feather identifications from ancient feather fragments (Dove *et al.*, 2005). Vinther *et al.* (2009) also studied different colours on various microstructures from fossil feathers. Later works

were mainly on plumulaceous barbs or downy feathers (Dove, 2000a; Lei *et al.*, 2002; Dove & Agreda, 2007; Lee *et al.*, 2016; Jiang, 2019). The feather microstructures were also used to identify birds from historical samples to know what all species were used for different purposes during the Viking age (Dove & Wickler, 2016). Studies were also conducted on down feathers to know about their microstructures that can be of industrial use (Fuller, 2015). Forensic identification of prey species from food particles of predatory birds was a successful attempt at plumology (Dove & Coddington, 2015).

Feather microstructures in Charadriiformes, studied from a functional angle, revealed complex structures of high taxonomic utility (Silaeva, 2019). The functional significances of feathers specifically in the scenario of inter and intra-species variations are yet poorly understood (Pap *et al.*, 2015). Along with the various functions, how these microstructures from downy feathers play a role in the adaptation was studied by Pap *et al.*, (2020). Silaeva *et al.* (2022) recently examined microstructures from the perspective of metric coordination in Anseriformes birds. Also, the current state of plumology in aviation ornithology was recently well describe by Silaeva & Chernova (2022) in Russia. In effect, the field of plumology is emerging from being a "Neglected Branch of Ornithology", as noted by Alen and Clark (1893).

Coming to the Indian scenario of plumology, there is a serious shortage of systematic works and literature. In effect, systematic studies of the feather microstructure of Indian species have not happened so far on a noteworthy scale. Understanding that systemic microstructure studies on bird feathers can effectively be used to explore different taxonomic issues, the present study was initiated. Subsequently, Ray *et al.* (2021) reported on feather microstructures exploring intra-species variations using road-kill bird samples. Dey *et al.* (2021) described the unique feather barbs of an Indian pitta, and Ray *et al.* (2022) reported on the microstructures in different types of feathers, and this thesis presents the details of the work.

4.3. Collection of biological samples

For the collection of biological samples of avian species for this study, the opportunistic road-kill survey method was adopted in select national and state highways of Assam running close to protected areas. Surveys were carried out after obtaining due

permissions from the Assam State Biodiversity Board (Ref. no: ABB/Permission/2012/82) and Office of the Principal Chief Conservator of Forests, Assam Forest Department (Ref. no. WL/FG.31/Pt/Technical Committee/2018) and Office order No. 258, date: 11/01/2019.

4.3.1. Review of literature on road-kill survey

The preparation of the dead list characterized in road-kill science started in the nineteen twenties (Kroll, 2015). Dayton Stoner was among the first naturalists in the nineteenth century to record vehicle-induced wildlife mortality (Kroll, 2015). Roads, acting as human-made barriers for the movements of birds and mammals, were systematically documented by Scott (1938). Effects of roads on various wildlife populations and their natural habitats are well-studied in several countries (Scott, 1938; Hodson, 1960; Hodson, 1962; van der Zande *et al.*, 1980; Seibert & Conover, 1991; Benitez-López *et al.*, 2010). There are also reports on the direct mortality of birds as commonly caused by vehicular collisions due to high-speed vehicular movements and road traffic (Hodson, 1960; Hodson, 1962; Van der Zande *et al.*, 1980; Develey & Stouffer, 2001; Forman *et al.*, 2002; Helldin & Seiler, 2003).

In India, only a few studies report the mortality of different species due to increasing numbers of vehicles and fragmented habitats (Sundar, 2004). Most of the road-kill studies from India are on amphibians, herpetofauna and mammals, occasionally reporting avian road-kill incidents (Kumara *et al.*, 2000; Vijaykumar *et al.*, 2001; Sundar, 2004; Das *et al.*, 2007; Bhupathy *et al.*, 2011; Islam & Saikia, 2014; Mahananda & Jelil, 2017; Jeganathan *et al.*, 2018). A few studies done especially focusing on avian mortality in the central and southern parts of India are by Sharma (1988), Dhindsa *et al.* (1988), Chhangani (2004), Siva & Neelanarayanan (2020) wherein the authors also discussed reasons for avian road causalities such as food availability on the road, high vehicular traffic and trans-passing roads through habitats.

The literature on road-kills shows an overall lack of information from the North East state of Assam, India, with only a couple of studies undertaken in recent years. Such studies include the impacts of roads on butterfly species by Choudhuri & Ghosh (2009) from lower Assam, road-kill of reptiles from Kaziranga National Park by Das *et al.* (2007), and that of Islam & Saikia (2014) on the reptile and amphibian mortality rate from Dehing-

Patkai Wildlife Sanctuary (now Dehing-Patkai National Park). There is a short note on a leopard road-kill from the urban area of Assam by Mahananda & Jelil (2017). Very recently, Sur *et al.* (2022) conducted a systematic study on road-kill vertebrates along National highway 715, the new one passing through Kaziranga National Park, and they reported the road-kill of birds belonging to 15 families.

4.3.2. Materials and methods for sample collection

4.3.2.1. Study area

The survey was conducted on seven road stretches, national and state highways, nearby Deepor Beel, Dehing-Patkai National Park, Gibbon Wildlife Sanctuary, Majuli Island and Kaziranga National Park (Figure 4.1), located in different districts of Assam (Table 4.1).

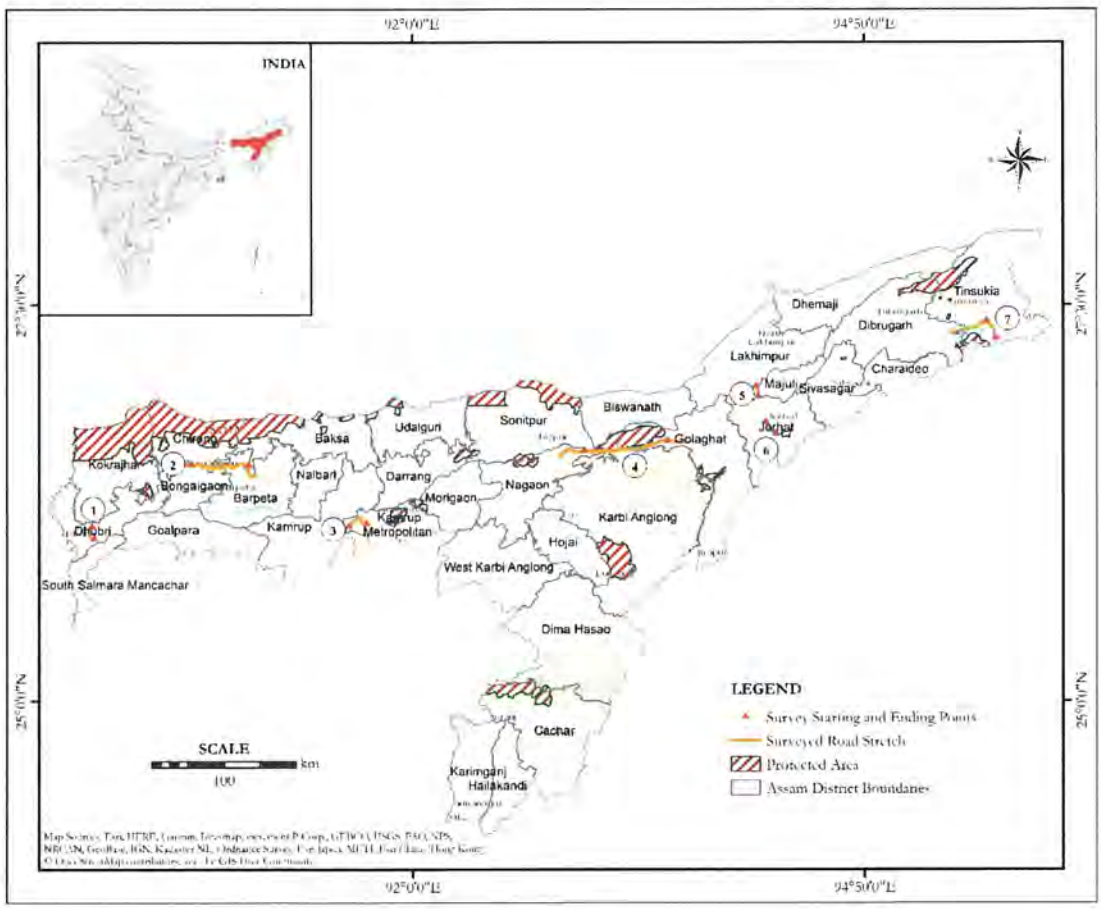

Figure 4.1: The road stretches surveyed for avian road-kill in Assam.

4.3.2.2. Sample collection

The Visual Encounter Survey (VES) method was adopted for the systematic survey of the selected road stretches. These road stretches were surveyed during the "rainy season" (July-October) in the year 2019 when Assam faced critical flood situations due to heavy rainfall (Das *et al.*, 2007). The survey was conducted for 120 days (A minimum of 7 days to a maximum of 15 days per road stretch). Using a motorbike at a uniform speed of 20±5 km/hour, the select road stretches were surveyed twice a day when animal activities are high (i.e., morning and evening). The data was collected by two observers starting at 4.00 am, and the same road stretch was replicated by 15.00 pm. The survey efforts were maintained constant by keeping the daytime increase of vehicular traffic in mind.

The encountered road-kill carcasses of birds were photographed using Canon 80D DSLR camera for identification and documentation while the 'Compas' mobile app was used to geotag the locations. After documenting the road kill, the carcass was removed, following all precautionary safety measures, to the side of the road to avoid duplication.

4.3.2.3. Identification of the species

Taxonomic identification of road-kills was done using the field guides Grimmett *et al.* (2016) and Majumdar *et al.* (2022) for bird identification in the survey using high resolutions photographs. The online database site of "Cornell Lab of Ornithology" (https://www.birds.cornell.edu/home/) was also used for further confirmation of the species and clearing doubts, if any.

4.3.3. Results

During the opportunistic road-kill surveys, 13 individuals from 5 different families of birds were collected from select roads of Assam (Figure 4.2, Image 4.1 and Table 4.2). Among the 7 selected road stretches, we have encountered bird road-kill casualties only on Tinsukia–Dibrugarh stretches (passing through Dehing-Patkai National Park) and Golaghat-Nagaon stretches (passing through Kaziranga National Park). Four birds from three families were collected from Dehing-Patkai National Park, and nine birds belonging to three families were collected from Kaziranga National Park.

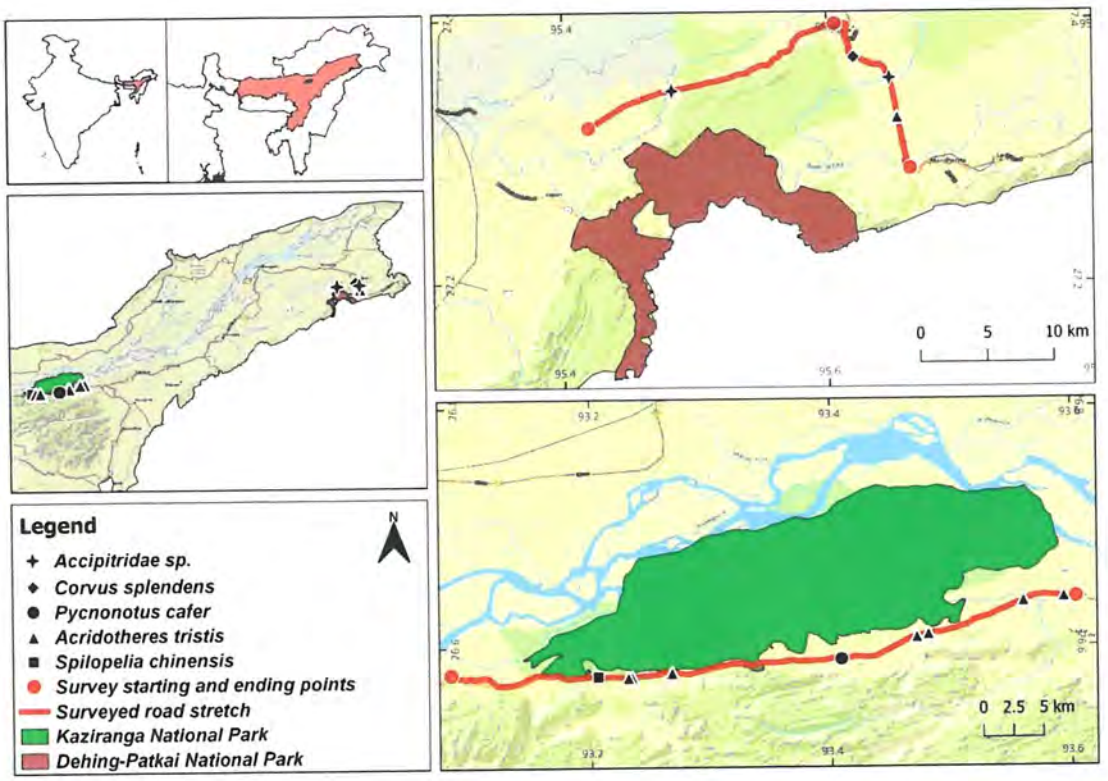

Figure 4.2: Geo-tagged locations of various road-kill avian species obtained from Assam (The legends represent the species obtained during the survey. The Dehing-Patkai National Park and Kaziranga National Park boundaries are marked on the map by two colours, highlighting the two protected areas where opportunistic road-kill samples were obtained).

The sampling of feathers was done following the standard National Avian Forensic Laboratory (NAFL) protocol for feather microstructure study (Details in section 4.4). For further genomic study, tissue samples were collected in tissue preservative solution, Dimethyl sulfoxide (DMSO).

Image 4.1: Road-kill birds from Assam. A- *Accipitridae sp.* and B- *Acridotheres tristis* from Dehing-Patkai National Park; C, D, E and F- *Acridotheres tristis*, G- *Spilopelia chinensis* and H- *Pycnonotus cafer* from Kaziranga National Park.

4.3.4. Selection of bird species for microstructure and genomic study

Selection of the bird species for the microstructure and genomic study was done based on two parameters, i.e., the select species are illegally traded in Assam with high demand and have to be locally available species which makes it easy to replicate for the study. Hence, Common Myna (*Acridotheres tristis*) and Spotted Dove (*Spilopelia chinensis*) were selected for further microstructure and genomic study. Both species come under the top 15 highly in demand in illegal avian trade in Assam. Common Myna (*A. tristis*) is traded as a pet, and Spotted Dove (*S. chinensis*) as Table-bird. The sample registration numbers at National Avian Forensic Laboratory (NAFL) at SACON, Coimbatore, Tamil Nadu-641108 for Common Myna (*A. tristis*) are 277, 278 and 280, and for Spotted Dove (*S. chinensis*) it is 286.

4.4. Materials and methods for feather microstructure study

4.4.1. Species details

Common Myna *(A. tristis),* belonging to the family Sturnidae, is widely distributed across the Indian subcontinent. It is a medium-sized (~25 cm) bird with no distinct sexual dimorphisms (Ali & Ripley, 1987; Kannan & James, 2020). It is one of the world's most invasive species, as per IUCN (Lowe *et al.,* 2004), and according to Ahmed (2001), in India, it is among the top five most traded avian species in pet markets. The singing and mimicking abilities of *A. tristis* make it the most frequent species in the illegal pet/avian trades (Ahmed, 1997, Ahmed, 2013). It is sold at a high price in domestic and international illegal pet markets as Hill Myna (*Gracula religiosa*) by disguising it with slight morphological modifications (Ahmed, 1997). Without a forensic examination, it becomes difficult to identify the disguised species in illegal trade (Ahmed, 1997; Lei *et al.,* 2002). The high demand for *G. religiosa* in the pet trade puts continuous pressure on the population of *A. tristis.*

Spotted Dove (*S. chinensis*) is a large-sized bird (30 cm) with a long, square-tipped tail; a black-and-white lacelike pattern of spots present on the hind neck is a diagnostic feature in post-juvenile plumages of *S. chinensis* (Grimmett *et al.,* 2016; Garrett & Walker, 2022). *S. chinensis* is distributed worldwide, including from eastern Afghanistan and India eastward through Indochina and south-eastern China, and south through the Malay

Peninsula, Sumatra, Borneo, the Lesser Sundas, and more recently, its distribution has been reported from the southwest Philippine Islands (Garrett & Walker, 2022). It is closely associated with agricultural areas, human habitats and open forests (Grimmett *et al.*, 2016). In India, *S. chinensis* is used as a table-bird in different regions, which causes its population decline (Ahmed, 1997). From our results presented in Chapter 3, *S. chinensis* comes in the top 15 traded avian species from Assam due to its high demand as bush-meat.

4.4.2. Sampling of feathers

Feathers were plucked from the dead three individuals of Common Myna (*A. tristis*) and a single individual of Spotted Dove (*S. chinensis*) carefully using surgical forceps (number 00). The collected feathers were categorized broadly into two types, contour (wing feathers, tail feathers and body contour) and non-contour (semiplume, down, powder down, bristle and filoplumes) (Gill, 1995; Lovette & Fitzpatrick, 2016). The different types of feathers (i.e., Contour and non-contour feathers) were selected from five different body locations (Dey *et al.*, 2021; Ray *et al.*, 2022), viz. wing, tail, dorsal, ventral and near to the eyes and beak region of the road-kill specimen of *A. tristis* and *S. chinensis* (Image 4.2). Three individuals of *A. tristis* and a single individual (for the unavailability of more samples) of *S. chinensis* were used to develop the feather microstructure atlas.

Image 4.2: Common Myna (*A. tristis)* with locations of feathers sampled (Source Ray *et al.*, 2022).

For the three individuals of *A. tristis*, two numbers of each feather from the remiges (primary and secondary flight feathers) were selected from the right wing. Two numbers of tail contour feathers were sampled from the tail. In duplicates, body contour feathers were taken from the dorsal and ventral sides. For non-contour feathers, two numbers of semiplume, down and powder down feathers were sampled from the tail, dorsal and ventral sides. Five bristle feathers from near the eye and beak and five filoplume feathers from the wings (right and left) were collected as samples. For *A. tristis*, total 38 numbers of feathers from each individual were used for preparing the slides.

For the single individuals of *S. chinensis*, two numbers of each primary and secondary contour feathers were sampled from the right wing. Two numbers of the tail contour feather were sampled from the tail. The body contour, semiplume, down and powder down feathers were sampled each in two numbers from each dorsal and ventral side. From near the eyes and beak, five numbers of bristle feathers were sampled. Filoplume feathers were sampled in five numbers from the right wing. For *S. chinensis*, total 32 numbers of feathers were selected for microstructure study.

The three basic macro-characteristics, viz. colour, pattern and texture, were examined for the sampled feathers from both the species, i.e., *A. tristis* and *S. chinensis* (Dove, 2000).

4.4.3. Feather shaft characteristics

Different characteristics of the feather shafts were studied from the rachis of the flight contour feathers of *A. tristis* and *S. chinensis* following the methods of Trail & Gaffney (2021). The upper, lower and cross sections of the feather's rachis were photographed using a Nikon stereo microscope (Model-SMZ-445) and Redmi Note 10 pro mobile camera for close-up views of their specific characteristics.

4.4.4. Barb sampling

The sampled feathers were cleaned using 70% ethanol solution to remove any contamination/dust/fungal particles (Lee *et al.*, 2016). The feathers were dried and scanned for morphometric measurements using a Samsung SCX-4x21S series printer. Based on lengths, feathers were divided into three equal sections as "proximal", "intermediate" and

"distal" (Ray *et al.*, 2022) (Image 4.3). Five barbs from each section were used for slide preparation. A total of 15 barbs from each feather were used to prepare slides for a feather type. Due to the lack of rachis in powder down feathers, this specific feather was not divided into any sections. In the case of powder down feathers, barbs were sampled directly from the calamus, the single attaching point. Surgical forceps (number 00) were used to sample the barbs carefully for slide preparation.

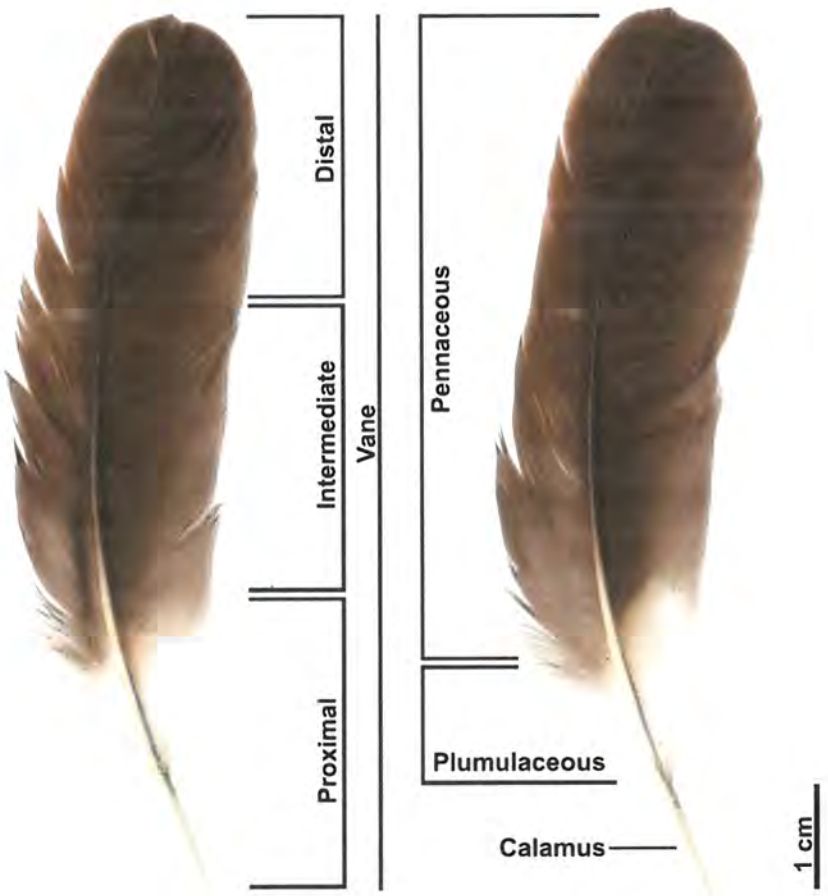

Image 4.3. Divisions of feathers for barb sampling in of *A. tristis* feathers.

4.4.5. Slide preparation

For slide preparation, glass slides and coverslips were cleaned using 70% ethanol. Sampled barbs were placed on a small drop of Xylene upon the pre-cleaned glass slides. The xylene drops were kept to air dry so that it keeps the barbs attached onto the slides (Dove & Koch, 2011). After the barbs dried completely, previously cleaned coverslips were placed on the barbs. Then, using transparent nail varnish, permanent slides were prepared for microstructure study (Robertson, 2002). The permanent slides prepared were labeled with the coding number of NALF (National Avian Forensic Laboratory, SACON) and stored in separate slide boxes at the laboratory.

Total 94 numbers of slides were prepared for a single individual of *A. tristis*. Hence, a total 114 numbers of feathers were used in the preparation of 282 slides for the three individuals of *A. tristis*. For *S. chinensis*, 52 slides were prepared from the sampled feathers to study the microstructures.

4.4.6. Measurements of barb length

Pennaceous and plumulaceous barbs from different types of feathers of *A. tristis* and *S. chinensis* were used to prepare permanent slides for microstructure studies. Barb lengths were measured using ImageJ software from the photographs of prepared slides.

4.4.7. Observation and documentation of feather microstructure

The permanent slides of *A. tristis* and *S. chinensis* were observed for specific microscopic features. The features observed for this study included sub-pennaceous region, villi, nodes and node shape, internode shape, prongs and prong size, hooklets and cilia, ventral teeth and pigmentation. Slides were observed under LaboMed 500X compound light microscope at 100X and 400X resolutions. Different microstructures were documented from barbs using Image aR software. 12 micro-structural observations were made from the barb's proximal, intermediate and distal regions, thus making 36 total observations for a single barb. Thus, from one slide, 180 observations were made on *A. tristis*, i.e., 16,920 observations on a single individual. Likewise, 9,360 observations of different feather microstructures were done on *S. chinensis*.

4.5. Results

4.5.1. Common Myna (*Acridotheres tristis*)

4.5.1.1. Feather macro-characters

As mentioned earlier, to study feathers, macro-characteristics of *A. tristis*, the colour, pattern and texture of nine different types of feathers from three individuals were examined (Table 4.3). The Colour of the feathers from all three individuals were similar but varied from black and white to dark brown to pale white and brown, and even dark brown with a tinge of white (Image 4.4, Table 4.3). Among all the feathers of *A. tristis*, only filoplume feathers showed an additional silvery appearance with pale black-coloured barbs at its tip. No specific patterns were observed on any of the feathers. The texture of feathers varied from being firmly rigid for flight contour feathers, i.e., primary contour, secondary contour and tail contour feathers. Body contour feathers were semi-rigid, whereas, irrespective of their location on the body, the semiplume, down, and powder down feathers were soft and fluffy in texture. In contrast, the modified contour feathers' bristles have a rigid texture.

Image 4.4: Flight contour feather of *A. tristis* from right wing and tail. A- Primary contour feather; B- Secondary contour feathers; C-Tail contour feathers (Scale bar: A, B & C= 1 cm).

4.5.1.2. Feather morphometry

Calamus length, vane length and rachis length of the nine different types of feathers were measured for feather morphometry of the three individuals of *A. tristis* (Table 4.4). Primary contour feathers from the wings were the longest among all feathers in all three individuals. Primary contour feathers possess the longest rachis length

(IND= Individual) (mean±S.E.) viz. in IND-1= 10.8±0.100 cm, IND-2= 11.145±0.000 cm, and IND-3= 10.694±0.119 cm. The bristle feathers were the shortest in rachis length (mean±S.E.) in all the three individuals of *A. tristis* viz. IND-1= 1.26±0.051 cm, IND-2= 0.7288±0.049 cm and IND-3= 0.892±0.052 cm.

Among all studied feathers, barb lengths were longest in primary contour feathers (mean length±S.E.) of *A. tristis* (Table 4.5), accordingly IND-1= 1.875±0.123 cm, IND-2= 1.582±0.070 cm, IND-3= 1.864±0.124 cm; whereas shortest barbs were in filoplume feathers viz. IND-1= 0.288±0.017 cm, IND-2= 0.216±0.009 cm and IND-3= 0.235±0.009 cm.

4.5.1.3. Feather shaft-characters

The feather shaft characters of flight contour feathers observed for feather morphological details (Image 4.5) were as follows.

(a) Feather dorsal surface

The rachis of flight contour feathers (Primary, secondary and tail contour feathers), observed under a microscope, did not show any specific pattern (Image 4.5A, D, G). Instead, only random black/dark brown coloured patchy pigmentation was present on the white-coloured rachis.

(b) Feather ventral surface

The ventral surface of the flight feathers of *A. tristis* does not have any distinct grooves but slightly upward projecting structures only on the proximal region of the rachis, i.e., just right after the end of the calamus (Image 4.5B, E, H).

(c) Cross section of the rachis

Cross-sections were done at the proximal region of feathers, i.e., just right above the end of the calamus in all the primary contour feathers. The cross-section exhibits hollow rachis characteristics in the primary contour feathers (Image 4.5C, F, I). Although the rachis looks like a rod from the outer side (without cross-section), the cross-section confirms its hollowness inside.

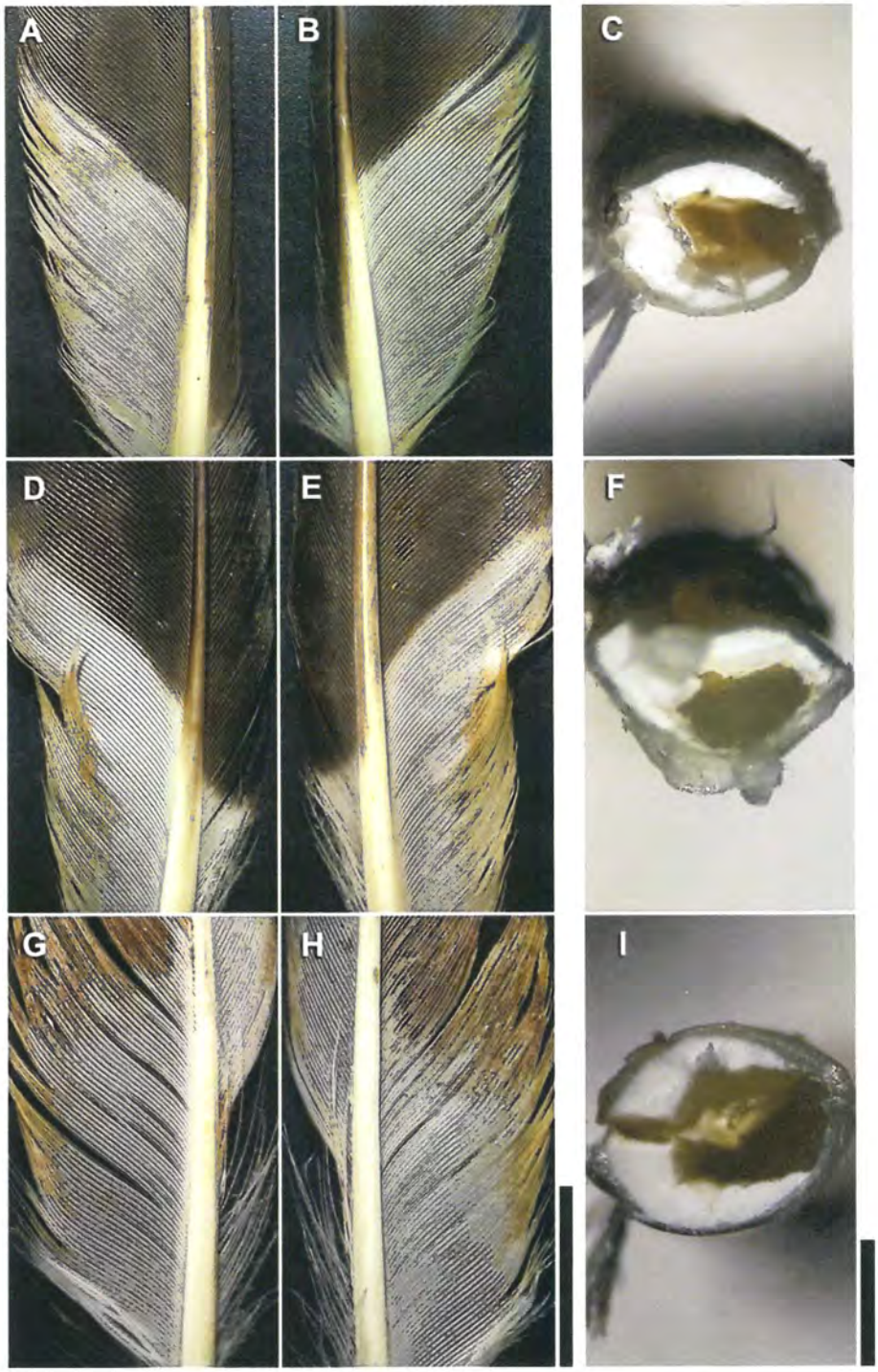

Image 4.5. Feather shaft characteristics of the primary contour feather of *A. tristis*. IND-1: A-Dorsal surface, B-Ventral surface, C-Cross-section; IND-2: D-Dorsal surface, E-Ventral surface, F-Cross-section; IND-3: G-Dorsal surface, H-Ventral surface, I-Cross-section. (Scale Bar: A, B, D, E, G, H= 1 cm; C, F, I= 1 mm)

4.5.1.4. Feather microstructures

The details of the barbs from the nine different feather types of *A. tristis* observed for different feather microstructures are given in Table 4.6, Image 4.6 and Image 4.7. The various microstructures observed are explained below.

(a) **Subpennaceous region:** The barbs of all the feathers showed the absence of subpennaceous region in both pennaceous and plumulaceous barbules in all the feather types.

(b) **Villi:** Villi are the unique diagnostic microstructural characteristic of passerine birds extending from the basal cell of the barbule of the plumulaceous barbs. The shape of villi is either knobbed or pointed, but sometimes both types are present in the basal cells forming finger-like structures (Image 4.6A, B, and C).

(c) **Nodes and their shape:** The barbules of all feathers had swollen nodes bulbous in shape. They are of three different types, viz. plain nodes (Image 4.6D, E), plain pronged nodes (Image 4.6F, G) and quadrilobed nodes (Image 4.6H, I). The plumulaceous barbs have all three types of nodes, while they are absent in pennaceous barbs. The quadrilobed nodes were mainly present in the proximal region of barbules, while the distal region had plain nodes either with prongs or without prongs. These nodes were present in all the different feather types except powder down, bristle and filoplume feathers.

(d) **Internode shape**: The region between two nodes is the internode, straight in shape and present in the barbules of plumulaceous barbs (Image 4.6D, E, F, G, H, I).

(e) **Prongs and prong size:** Prongs are present only on the swollen nodes. Nodes with small prongs were present in the plumulaceous barbs of primary contour, secondary contour, tail contour, body contour, semiplume and down feathers. On the nodes of the bristle (Image 4.7J, K) and filoplume feathers, elongated and large-sized prongs were present. Prongs were absent in powder down feathers.

(f) **Hooklets:** Distinct hooklets were present in pennaceous barbs of primary, secondary, and tail contour feathers. They were present after the basal cells of the barbules (Image 4.7L, M). In *A. tristis*, hooklets were completely absent in all plumulaceous barbs.

(g) **Ventral Teeth:** Pennaceous barbs had ventral teeth at the end of basal cells that were relatively less broad (Image 4.7N, O).

(h) **Pigmentation:** Ramus was present with patchy (Image 4.7P) and dark pigmentation (Image 4.7Q, R). Dark pigmentations were mainly present on the nodes, whereas internodes mostly had patchy pigmentation. In contrast, in semiplume and powder down feathers, nodes had both types of pigmentation (Image 4.7S, T).

4.5.2. Spotted Dove (*Spilopelia chinensis*)

4.5.2.1. Feather macro-characters

Macro-characters of different feathers from *S. chinensis* are summarized in Table 4.7. The colour of *S. chinensis* varied from dark brown to pale brown, creamy with different tinges of rusty, pale brown, and black. There was no distinguishing pattern observed in the different feathers of *S. chinensis*. The texture observed in different feathers of *S. chinensis* varies from firmly rigid to soft and fluffy. In *S. chinensis*, flight contour feathers, i.e., primary contour, secondary contour and tail feathers, were firmly rigid in texture (Image 4.8). Irrespective of their different body locations, body contour feathers were semi-rigid in texture. The semiplume, down, and powder down feathers were soft and fluffy in texture. The very location-specific filoplume feathers are soft in texture.

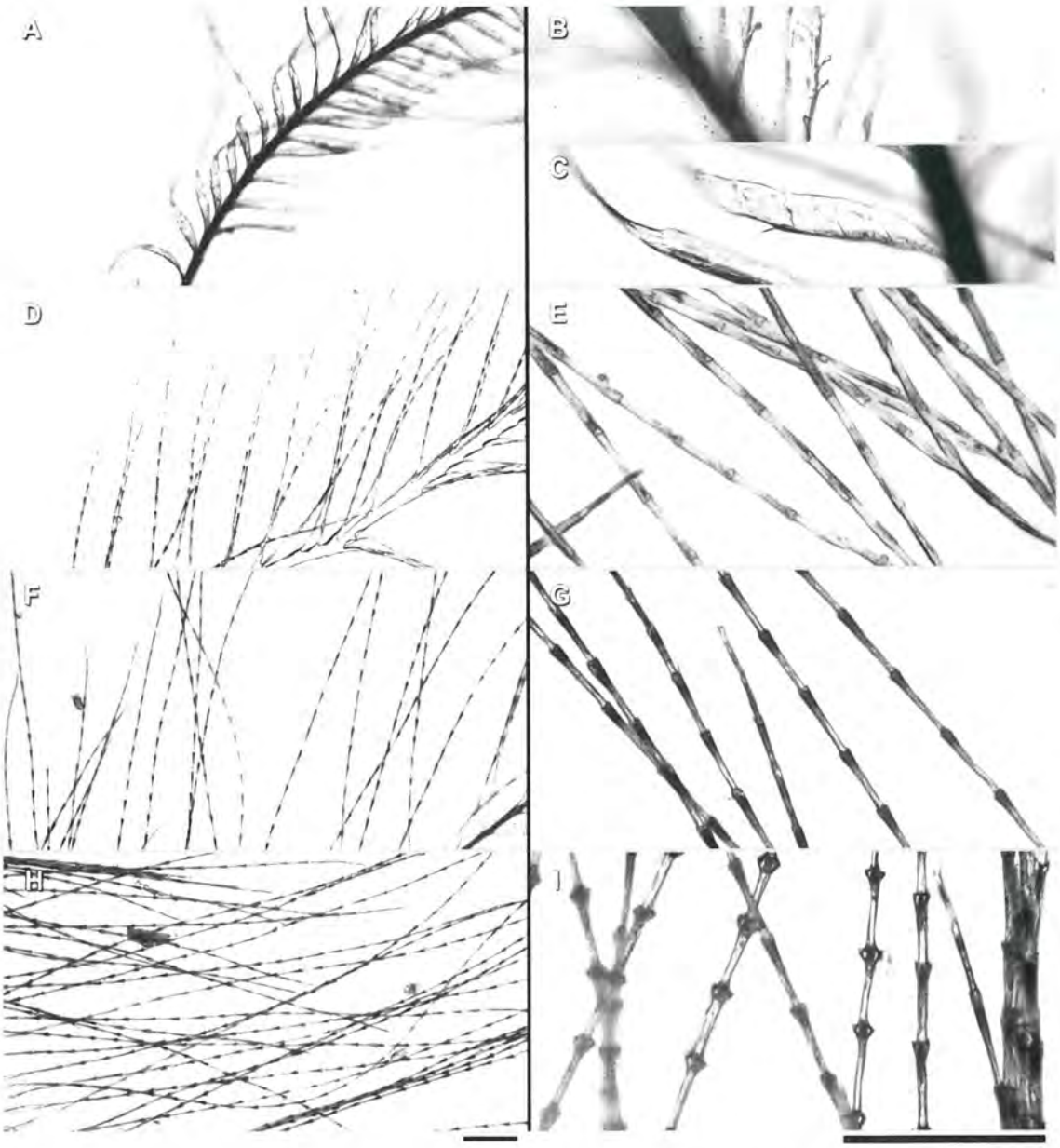

Image 4.6. Feather microstructures of *A. tristis*. A- Villi at 10X, B- Knobbed shape villi at 40X, C- Pointed villi at 40X, D- Plain nodes at 10X, E- Plain nodes at 40X, F- Plain pronged nodes at 10X, G- Plain pronged nodes at 40X, H- Quadrilobed nodes at 10X, I- Quadrilobed nodes at 40X. (Scale Bar: 0.1 mm in 10X and 40X).

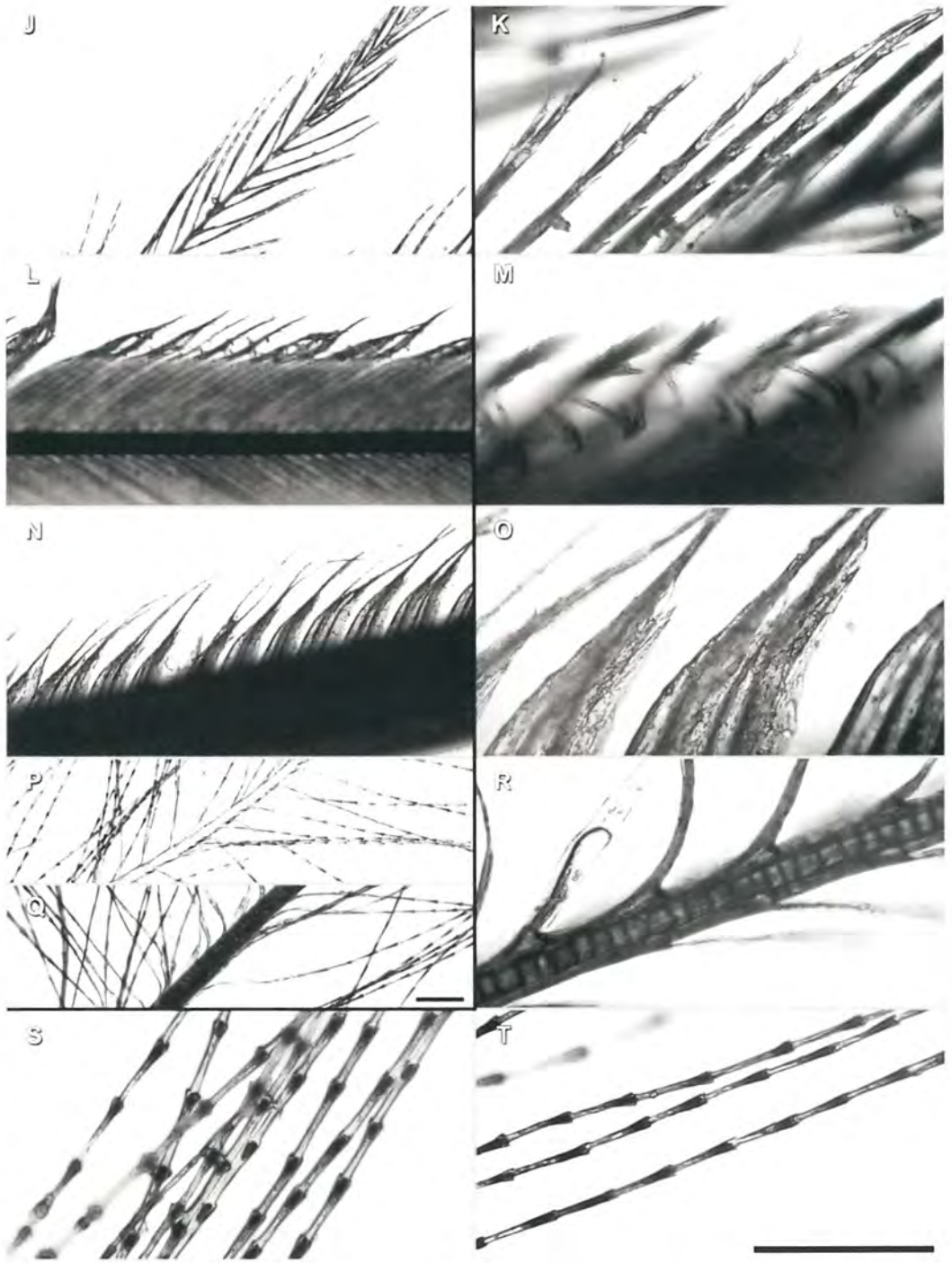

Image 4.7. Feather microstructures of *A. tristis*. J- Elongated prongs on nodes at 10X, K- Elongated prongs on nodes at 40X, L- Hooklets at 10X, M- Hooklets at 40X, N- Ventral teeth at 10X, O- Ventral teeth at 40X, P- Patchy pigmented ramus at 10X, Q- Densely pigmented ramus at 10X, R- Densely pigmented ramus at 40X, S- Patchy pigmentation on nodes at 40X, T- Densely pigmented nodes at 40X. (Scale Bar: 0.1 mm in 10X and 40X).

Image 4.8: Flight contour feather of *S. chinensis* from right wing and tail. A- Primary contour feather; B- Secondary contour feathers; C-Tail contour feathers (Only three numbers of feathers were retrieved from tail region of *S. chinensis* due to poor condition of road-kill carcass) (Scale bar: A, B & C= 1 cm).

4.5.2.2. Feather morphometry

The morphometric characteristics, namely calamus length, vane length and rachis length, measured for eight types of feathers in *S. chinensis,* are given in Table 4.8. The flight contour feathers from wings were the longest among all feather types. Flight contour feathers for *S. chinensis* were measured as calamus length range was 1.33±0.075 S.E. in cm, vane length range was 8.45±0.272 S.E. in cm, and rachis length range was 9.9±0.408 S.E. in cm. Down feathers from dorsal and ventral regions were the shortest in length in *S. chinensis*. In the case of the down feathers in *S. chinensis*, the calamus length was in the range of 0.25±0.029 S.E. in cm, the vane length range was 2.00±0.091 S.E. in cm, and the rachis length range was 2.25±0.065 S.E. in cm. Due to the absence of proper rachis, the vane and rachis length were not measured for powder down feathers in *S. chinensis*.

The measurements of barbs from different feathers of *S. chinensis* are shown in Table 4.9. In *S. chinensis*, the tail feathers have the longest barbs, i.e., 1.840±0.119 S.E. in cm. The shortest barb length was measured in powder down feathers in *S. chinensis,* i.e., 0.888±0.036 S.E. in cm.

4.5.2.3. Feather shaft-characters

The shaft characters of flight contour feathers of *S. chinensis* (Image 4.9) were as given below:

(a) Feather dorsal surface

The flight contour feathers (primary, secondary and tail contour feathers) of *S. chinensis* exhibited two distinct parallel fine lines running through the ventral surface of the rachis (Image 4.9A). The parallel lines are most prominent at the proximal region of the feather shaft, i.e., just after the calamus, and after reaching the intermediate region, it gradually diminishes as faint lines on the ventral surface of the shaft.

(b) Feather ventral surface

The dorsal surfaces of the rachis of *S. chinensis* had a distinct broad groove (Image 4.9B). These broad grooves were most prominent in the large contour feathers, i.e., primary, secondary and tail contour feathers. In these feathers, it was most prominent and well developed at the wide basal region of the rachis, just after the calamus, and it ran

through the rachis until the intermediate region. Towards the end of the intermediate region, this broad groove gradually diminished, finely merging with the rachis in the distal region of a feather.

(c) Cross-section of the rachis

We took the cross-sections at the proximal region of feathers above the end of the calamus of the secondary contour feathers rachis. The close-up views of the cross sections of *S. chinensis* exhibited the hollow rachis characteristics (Image 4.9C). Despite the presence of a broad groove in the under surface of the rachis, the cross sections confirm the hollowness of the rachis.

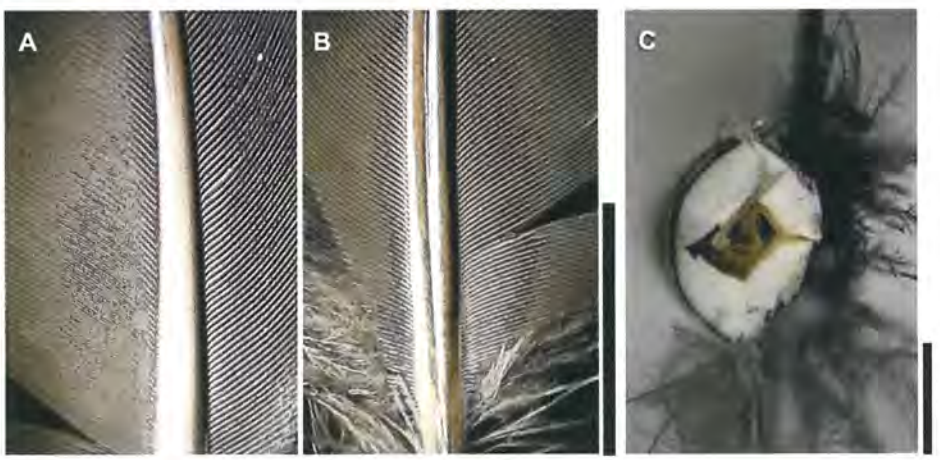

Image 4.9. Feather shaft characteristics of the secondary contour feather of *S. chinensis*. A-Dorsal surface, B-Ventral surface, C- Cross-section. (Scale Bar: A, B= 1 cm; C= 1 mm)

4.5.2.4. Feather microstructures

In *S. chinensis*, the microstructures observed (Table 4.10, Image 4.10, Image 4.11) include the following.

(a) Subpennaceous region: Subpennaceous regions are present on the barbs from the down region of contour feather (Dove, 2000). If present, this region is located at the very base of plumulaceous barbs. It contains barbules that have structural similarities to pennaceous feathers (long strap-like base and pennulum with hooklets on distal vanule). We recorded subpennaceous regions in the

plumulaceous barbs of the flight feathers (Wings and tail), body contour and semiplume feathers in *S. chinensis* (Image 4.10A, B).

(b) Nodes and nodes shape: Distinct swollen nodes were present in the barbules of plumulaceous barbs in all feathers. Three different node shapes, very large, flattened, plate-like nodes, i.e., crocus-shaped nodes (Image 4.10C, D), followed by several swollen but plain pronged nodes (Image 4.10E, F) that culminate into smaller, very minute and less conspicuous swollen nodes with or without prongs (Image 4.10G, H) present in the plumulaceous barbules. The barbules of *S. chinensis* presented asymmetrical vanules, i.e., with highly expanded and often flattened plate-like nodes (Crocus-shaped nodes) on the opposing vanules. The Crocus-shaped nodes were seen right after the basal cells, followed by plain-pronged nodes. The minute swollen nodes were present at the distal part of elongated filamentous barbules with tiny prongs or without prongs. All three different shaped nodes were present in the flight contour feathers, i.e., primary, secondary and tail contour feathers.

In *S. chinensis*, these three different nodal shapes were present in the barbules of all six different feathers. But in the body contour, semiplume, down, and powder-down feathers, these three differently shaped nodes are present together in the barbules of the same barb. Mostly the proximal plumulaceous barbules have crocus-shaped nodes with plain nodes, whereas the distal barbules have plain nodes with small prongs.

(c) Internode shape: The region between two nodes is the internode which is straight in shape in all the four Columbidae species (Image 4.10C, D, E, F, G, H).

(d) Prongs and prong size: Small prongs and nodes' different shapes were also observed at times (Table 4.10). The plain swollen nodes were present with very minute prongs (Image 4.10E, F). Occasionally, small prongs were also observed at the distal tip of elongated barbules with less conspicuous swollen nodes. In contrast, prongs were absent in powder down feathers of *S. chinensis*.

(e) **Hooklets and cilia:** Distinct hooklets were present in the pennaceous and plumulaceous barbs of all types of feathers of *S. chinensis* except powder-down feathers (Image 4.11K, L). In flight contours (primary, secondary and tail contour feathers), distinct hooklets were present in pennaceous barbs. In contrast, in body contours, semiplume and down feathers hooklets were present in plumulaceous barbs confirming the presence of sub plumulaceous region. Two differently shaped cilia (straight and spinelike elongated cilia and flower petal-shaped cilia) were present on the pennaceous and plumulaceous barbs of *S. chinensis* (Image 4.11I, J). Elongated, spinelike, and petal-shaped cilia were present in primary, secondary and tail contour feathers, i.e., in flight and body contour feathers. In comparison, short, straight, and spine-like cilia were present in the semiplume and down feathers of *S. chinensis*. Cilia was absent in powder down feather.

(f) **Ventral teeth:** Ventral teeth were present in the flight contour feathers (primary contour, secondary contour and tail contour), body contour, semiplume feathers and down feather (Image 4.11M, N). They were absent in powder down feathers.

(g) **Pigmentation:** The ramus of the barbs was pigmented with both dark and patchy types of pigmentation. In *S. chinensis*, ramus of flight contour feathers (primary, secondary and tail contour feathers) was present with both dark and patchy pigmentation (Image 4.11O); whereas ramus present in body contour, semiplume down and powder down feathers were densely pigmented. Nodes of *S. chinensis* had dark and patchy/slight pigmentation in all feather types. In *S. chinensis*, the nodes of tail contour feathers were pigmented densely, whereas the rest of the nodes had both pigmentation types (Image 4.11P). Internodes were patchy in pigmentation in all feather types except the down feathers, where both the pigmentations, dark and patchy, were present.

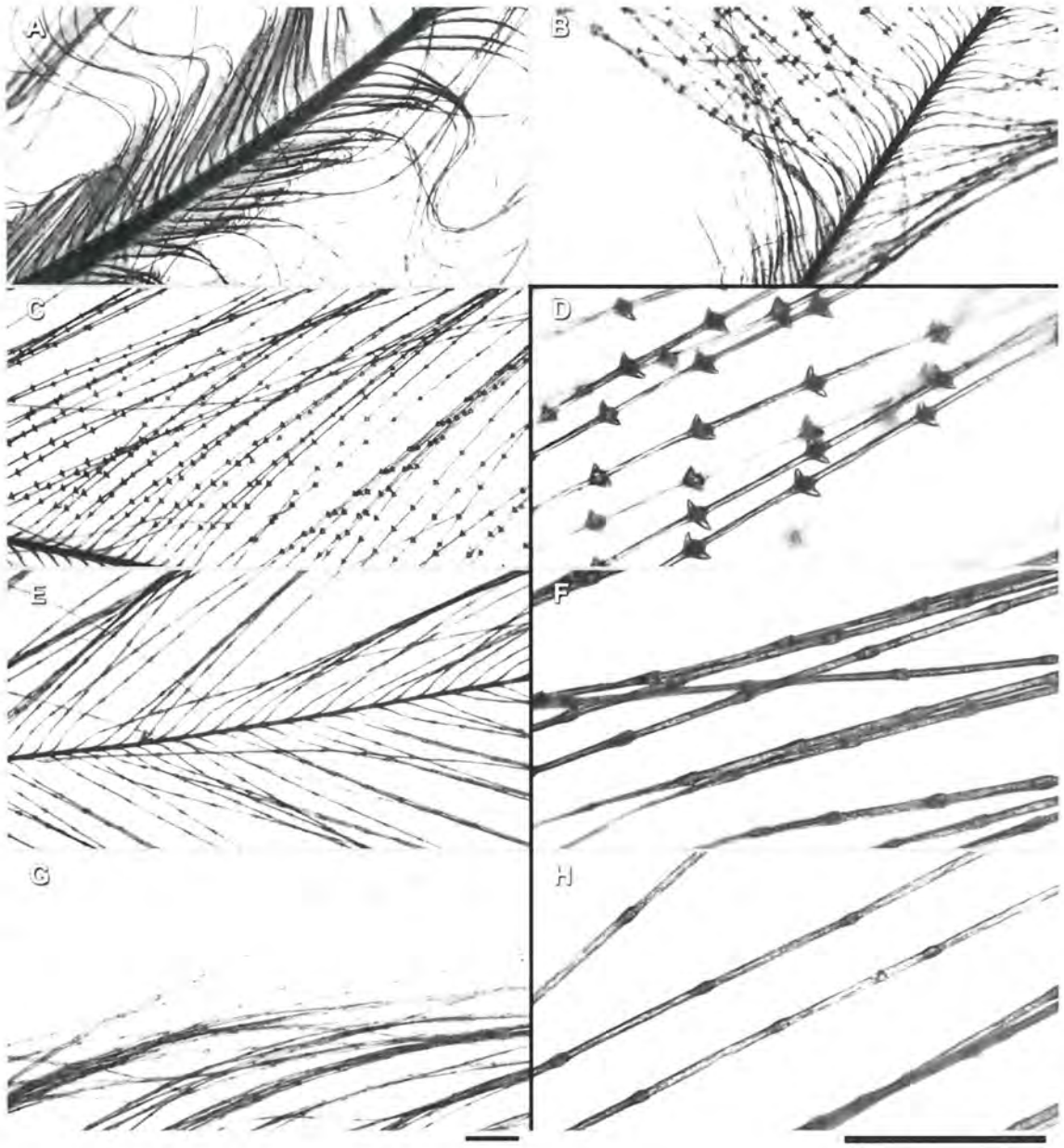

Image 4.10. Feather microstructures of *S. chinensis*. A-Subpennaceous region with plain nodes at 10X, B-Subpennaceous region with crocus-shaped nodes at 10X, C- Crocus shape nodes at 10X, D- Crocus shape nodes at 40X, E- Plain pronged nodes at 10X, F- Plain pronged nodes at 40X, G- Plain swollen nodes at 10X, H- Plain swollen nodes at 40X. (Scale Bar: 0.1 mm in 10X and 40X).

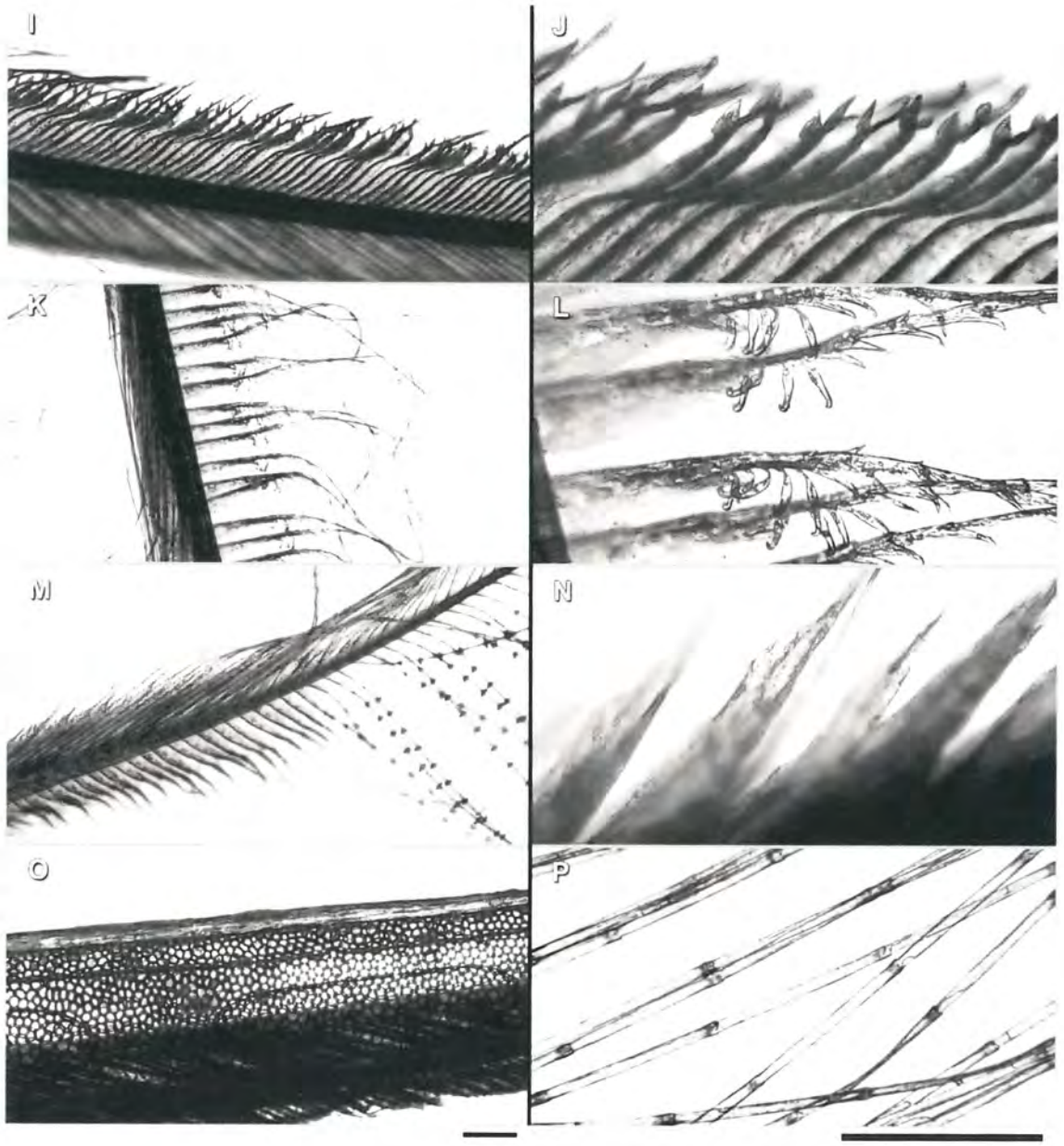

Image 4.11. Feather microstructures of *S. chinensis*. I- Petal-shaped cilia at 10X, J- Petal-shaped cilia at 40X, K- Hooklets at 10X, L- Hooklets at 40X, M- Ventral teeth at 10X, N- Ventral teeth at 10X, O- Patchy pigmentation on ramus at 10X, P- Patchy pigmentation on nodes at 40X. (Scale Bar: 0.1 mm in 10X and 40X).

4.6. Discussion

During this study, different morphological characteristics such as colour, pattern, and texture of different feather types from different body locations along with their morphometric measurements (i.e., length of calamus, vane, rachis and barb) from *A. tristis* and of *S. chinensis* were documented. Feather colour and texture differences are mainly based on their location on the avian body and functions (Ray *et al.*, 2021). Earlier, Chandler (1916) stated that colour is the most significant characteristic of avian species for their identification as it comes with specific patterns. During the present study, we observed silver-coloured filoplume feathers with pale black pigmentation on the barbs, a specific characteristic of *A. tristis*. However, it must be noted that it is difficult to retrieve the filoplume feathers due to their location and almost transparent nature. Except for the filoplume feathers, we recorded varying colours specific to the feather types of the three individuals of *A. tristis*; however, the feathers' colourations were similar in all the individuals. No specific patterns were present in the feathers of *A. tristis*. I have also not observed any specific pattern in the feathers of *S. chinensis*. Yet, the different colours in their different feathers signify their morphological difference.

Based on different functional aspects such as flight mechanism, thermoregulation, signalling ornaments, detection and protection, feather kinaesthetics, etc., and avian body locations, the feathers' texture varies (Lovette & Fitzpatrick, 2016; Ray *et al.*, 2021). The texture of the feathers of *A. tristis* mainly comprised of three types, i.e., rigid, semi-rigid and soft and fluffy, helping in the function of flight, protection and thermoregulation, respectively. Likewise, in *S. chinensis,* feather textures are also of three types: rigid, semi-rigid, soft and fluffy and soft serving various functions mentioned above (Gill, 1995; Lovette & Fitzpatrick, 2016).

Morphological variations depending upon different functions are important cues while studying avian speciation. Any changes in morphological appearances can indicate ongoing phenomena in the evolutionary framework of any species (Töpfer, 2018). Hence, documentation of various morphological characteristics is important for avian and other species. Although macro characteristics and morphometric measurements tend to change based on age and sex, the measurements are species-specific (Dove, 2000; Lee *et al.*, 2015).

The data on the feather morphometry can also be clues to the physical size of a species (Lee *et al.*, 2015). The present study provides the ranges for the feather morphometrics of *A. tristis* and *S. chinensis* that can be used for the same.

Feather shaft, which is lightweight, stiff and strong, yet sufficiently flexible, were studied to understand better the avian flight mechanism (Wang & Meyers, 2017; Sullivan *et al.*, 2016). Different morphological characteristics, viz. pattern on rachis (in upper and lower surface), the structure of rachis (in upper and lower surface) and the cross-section of the rachis were studied to distinguish game bird and waterfowl's feathers from raptor feathers (Trail & Gaffney, 2021). In this study, we have described the characteristics of feather shafts of *A. tristis* and *S. chinensis*. In *A. tristis*, the absence of any specific patterns on the feather shaft but the presence of random dark to patchy pigmentation without any distinct groove was seen as a general characteristic of the feathers. In *S. chinensis*, two fine parallel lines on the upper surface of the rachis, the broad groove on the rachis's basal part and the rachis' unbroken hollowness are three specific characteristics that can differentiate *S. chinensis* from other birds at the family level.

Several studies have been conducted to know about the variation in diagnostic features among intra-species, inter-species and also among different feathers (Chandler, 1916; Dey, 1966; Robertson *et al.*, 1984; Brom, 1991; Dove, 2000; Dove & Peurach, 2002; Lee *et al.*, 2015, Dey *et al.*, 2021; Ray *et al.*, 2021). Previous studies on feather microstructures illustrate that despite individual differences in length and width, feather microstructures remain similar among individuals of a particular species (Dove, 1997; Lee *et al.*, 2015; Ray *et al.*, 2021). Hence, we have focussed more on different feather microstructures by increasing the number of the same types of feathers examined and sampling them from different body locations of the same species. Further, we found no difference among the individuals of *A. tristis* in macro-characteristics and microstructures. Even in different feathers of *S. chinensis*, the same types of feathers showed similar microstructures.

To identify Passerine birds, Chandler (1916) stated that the pennaceous barbs would contain three to four hooklets, while Lee *et al.*, (2015) observed the presence of the broadened shape of ventral teeth in *A. tristis*. However, Lee *et al.*, (2015) cautioned that

these microstructures could not be used as an exclusive character for identifying the species. At the same time, Dove (2000) suggested that pigmentation pattern gives diagnostic clues towards determining species group of birds. In contrast, our study of *A. tristis* feathers shows no peculiar uniform pigmentation pattern in nodes, internodes and ramus. However, we observe that dark and patchy pigmentation on different nodes can be used as a micro character for identifying *A. tristis*. Furthermore, from this study, we report three microstructures that can be used in the identification of *A. tristis* species; (i) the presence of specifically shaped finger-like villi, i.e., both knobbed and pointed on the border of the basal cells, (ii) presence of all the three types of nodes, i.e., quadrilobed nodes, pronged nodes and plain nodes, and (iii) the presence of sharply pointed pronged nodes on bristle and filoplume feathers.

A brief description of the microstructures on the barbules of the inner and outer vane of *Columba livia* was made by Chandler (1916). Different feather microstructures from five different species of Columbidae (Family Columbiformes) were also previously illustrated (Brom 1986b). Subsequently, different microstructures, such as barbules and nodes of a Columbidae species, Bronze wing pigeon (*Phaps chalcoptera*), were also described (Lee *et al.*, 2015). In our study, we have documented eight different microstructures on the barbules of *S. chinensis* at five different body locations. The presence of subpennaceous region on flight contour feathers (primary, secondary and tail contour feathers), body contour, semiplume and down feathers of *S. chinensis* is a significant family-level identification characteristic. More studies are required, especially focusing on the microstructure characteristic of a higher number of individuals of the same species. The presence of three different shaped nodes, i.e., crocus-shaped node, plain-pronged node and swollen node without prong on the plumulaceous barbules, can exclusively be used as family level specific characteristics of Columbidae (Brom, 1986b). The presence of these three differently shaped nodes on specific feather types of different body locations can be species-specific nodal structures. Pigmentation on nodes and internodes is also a diagnostic clue for determining if a species group is characterised by any pigmentation pattern (Dove, 2000). In this study, I have not observed any peculiar uniform pattern in *S. chinensis*, except for both dense and patchy pigmentations present randomly in both nodes and internodes.

By using different feather macro characters, morphometry and microstructures focusing on the pennaceous and plumulaceous barbs, plumology can give real insights into the specific characteristics of order, family and species of birds. During our study, we used a systematic approach towards identifying the bird species from the feathers. Among the macro characters of *A. tristis*, the filoplume feathers help determine the bird as a passerine species. While the feather macro characters and morphometry helped ascertain the species to the passerine group, the microstructure further facilitated identifying the bird to the species level. The microstructures present, like the finger-like projection of villi, three different types of nodes and elongated prongs on the nodes of bristle and filoplume feathers, help identify the species *A. tristis*. The study also provides the morphometry measurements for the feathers for future reference and is a baseline reference database for identifying *A. tristis* from India. Our study also provides key characteristics for identifying *S. chinensis* by focusing on different macro characteristics, morphometric measurements and microstructures. Especially the presence of asymmetrical vanules can be used for species identification when adequate feathers samples are available (Dove & Koch, 2010). The asymmetrical vanules are not always available to observe as it depends on the feather sampling. Still, in case available, that can be of much value as a means of identification using the specific microstructures. The systematic study on species-specific microstructures such as villi and nodes can be used in various scientific studies. The key characteristics of feather morphometrics and microstructures discussed here can also help in the future systematic study of plumology, avian forensics and even species evolution.

Tables:

Table 4.1. GPS points of the surveyed road-stretches in different districts of Assam.

S. No.	District	Starting point (Degree decimal)		Ending point (Degree decimal)	
		Latitude	Longitude	Latitude	Longitude
1	Dhubri	26.096276	89.979896	26.022215	89.995138
2	Chirang	26.488986	90.611877	26.486825	90.967446
3	Kamrup	26.100534	91.598195	26.11578	91.70855
4	Golaghat	26.64041	93.603723	26.577222	93.082373
5	Majuli	26.922475	94.166457	26.985472	94.153867
6	Jorhat	26.761102	94.213197	26.687827	94.276925
7	Tinsukia	27.396463	95.605647	27.317838	95.419596
	Tinsukia	27.396463	95.605647	27.286199	95.661759

Table 4.2. GPS locations of collected road-kill bird samples in Assam.

Sl. No.	Name of The Protected Area	Species Name	Scientific Name	Latitude (Degree Decimal)	Longitude (Degree Decimal)
1	Dehing-Patkai National Park	Hawk Spp.	*Accipitridae sp.*	27.345921	95.483298
2	Dehing-Patkai National Park	Hawk Spp.	*Accipitridae sp.*	27.355502	95.646274
3	Dehing-Patkai National Park	House Crow	*Corvus splendens*	27.370938	95.619660
4	Dehing-Patkai National Park	Common myna	*Acridotheres tristis*	27.325278	95.652222
5	Kaziranga National Park	Common myna	*Acridotheres tristis*	26.6075	93.470556
6	Kaziranga National Park	Common myna	*Acridotheres tristis*	26.574722	93.234167
7	Kaziranga National Park	Common myna	*Acridotheres tristis*	26.574444	93.231111
8	Kaziranga National Park	Common myna	*Acridotheres tristis*	26.640833	93.593611
9	Kaziranga National Park	Common myna	*Acridotheres tristis*	26.637222	93.559722
10	Kaziranga National Park	Common myna	*Acridotheres tristis*	26.61	93.480278
11	Kaziranga National Park	Common myna	*Acridotheres tristis*	26.578333	93.2675
12	Kaziranga National Park	Red-Vented Bulbul	*Pycnonotus cafer*	26.589167	93.408056
13	Kaziranga National Park	Spotted Dove	*Spilopelia chinensis*	26.575278	93.205556

Table 4.3. Feather macro-characteristics of three different individuals of *A. tristis*.

Sl. No.	Feather Type	Feather Location	Colour	Texture
1	Primary contour feather	Wing	Black and white	Rigid
2	Secondary contour feather	Wing	Dark brown	Rigid
3	Tail contour feather	Tail	Dark brown with white tinge	Rigid
4	Body Contour	Dorsal	Pale brown	Semi rigid
5	Body Contour	Ventral	Pale brown	Semi rigid
6	Semiplume	Dorsal	Pale brown	Soft and fluffy
7	Semiplume	Ventral	Pale brown	Soft and fluffy
8	Semiplume	Tail	White	Soft and fluffy
9	Down	Dorsal	Pale brown	Soft and fluffy
10	Down	Ventral	Pale white	Soft and fluffy
11	Down	Tail	Pale white	Soft and fluffy
12	Powder Down	Dorsal	White	Soft and fluffy
13	Powder Down	Ventral	White	Soft and fluffy
14	Powder Down	Tail	White	Soft and fluffy
15	Bristle	Near Eye and Beak	Black	Rigid
16	Filoplume	Wings	Silver	Soft

Table 4.4. Morphometric measurements of different feathers of all three individuals of *A. tristis*.

Sl. No.	Feather Type	Feather Location	Calamus Length ± S.E.			Vane Length ± S.E.			Rachis Length ± S.E.		
			IND-1	IND-2	IND-3	IND-1	IND-2	IND-3	IND-1	IND-2	IND-3
1	Primary contour feather	Wing	1.45±0.05	1.38±0.00	1.08±0.01	9.35±0.05	9.53±0.06	9.28±0.00	10.80±0.10	11.14±0.00	10.69±0.12
2	Secondary contour feather	Wing	1.35±0.05	1.019±0.14	1.11±0.03	7.80±0.00	9.24±0.39	8.67±0.09	9.25±0.05	10.19±0.03	10.32±0.16
3	Tail contour feather	Tail	0.80±0.10	0.82±0.05	0.63±0.00	7.3±0.10	7.03±0.37	7.25±0.06	8.25±0.05	8.19±0.48	8.74±0.03
4	Body Contour	Dorsal	0.20±0.00	0.27±0.03	0.35±0.04	3.85±0.05	3.62±0.04	3.86±0.23	4.25±0.05	3.98±0.05	3.98±0.05
5	Body Contour	Ventral	0.35±0.05	0.25±0.01	0.31±0.03	4.8±0.05	4.46±0.30	3.16±0.35	5.25±0.05	4.92±0.30	3.52±0.01
6	Semiplume	Dorsal	0.35±0.05	0.31±0.03	0.30±0.01	3.40±0.10	3.76±0.26	4.00±0.24	3.80±0.10	4.20±0.00	4.92±0.12
7	Semiplume	Ventral	0.45±0.05	0.28±0.01	0.26±0.00	4.51±0.39	3.66±0.01	3.60±0.02	4.58±0.42	4.25±0.06	4.18±0.07
8	Semiplume	Tail	0.45±0.05	0.35±0.05	0.37±0.02	4.95±0.15	4.45±0.35	3.59±0.39	5.50±0.10	5.00±0.04	3.92±0.36
9	Down	Dorsal	0.25±0.05	0.28±0.01	0.24±0.00	3.15±0.05	1.58±0.06	1.90±0.03	3.4±0.05	1.86±0.05	2.40±0.09
10	Down	Ventral	0.3±0.00	0.40±0.03	0.25±0.03	3.4±0.05	2.18±0.36	1.96±0.20	3.70±0.10	2.47±0.27	2.71±0.26
11	Down	Tail	0.2±0.00	0.37±0.01	0.35±0.00	3.25±0.05	3.38±0.07	3.31±0.01	3.45±0.05	3.75±0.06	3.66±0.01
12	Powder Down	Dorsal	0.2±0.00	0.25±0.00	0.25±0.05	NA	NA	NA	NA	NA	NA
13	Powder Down	Ventral	0.25±0.05	0.25±0.05	0.25±0.03	NA	NA	NA	NA	NA	NA
14	Powder Down	Tail	0.25±0.05	0.25±0.05	0.25±0.04	NA	NA	NA	NA	NA	NA
15	Bristle	Near Eye and Beak	0.26±0.02	0.19±0.01	0.16±0.01	1.00±0.03	0.53±0.109	0.59±0.01	1.26±0.05	0.72±0.04	0.89±0.05
16	Filoplume	Wings	NA	NA	NA	NA	NA	NA	1.94±0.26	1.22±0.14	1.34±0.23

IND – Individual.

Table 4.5. Average barb length of feathers of *A. tristis* in three individuals.

Sl. No.	Feather Type	Feather Location	Average Barb length in cm ± S.E.		
			IND-1	IND-2	IND-3
1	Primary contour feather	Wing	1.875±0.123	1.582±0.070	1.864±0.124
2	Secondary contour feather	Wing	1.821±0.111	1.409±0.0785	1.748±0.094
3	Tail contour feather	Tail	1.637±0.079	0.937±0.030	1.528±0.083
4	Body Contour	Dorsal	1.391±0.026	1.032±0.057	1.398±0.124
5	Body Contour	Ventral	1.646±0.043	1.131±0.023	1.611±0.105
6	Semiplume	Dorsal	1.532±0.033	1.035±0.086	1.349±0.079
7	Semiplume	Ventral	1.091±0.037	1.167±0.059	1.172±0.036
8	Semiplume	Tail	1.034±0.024	1.029±0.085	1.319±0.079
9	Down	Dorsal	1.415±0.068	1.106±0.053	1.383±0.044
10	Down	Ventral	1.604±0.064	1.158±0.021	1.502±0.061
11	Down	Tail	1.078±0.057	1.098±0.055	1.378±0.053
12	Powder Down	Dorsal	1.279±0.046	0.989±0.049	1.249±0.059
13	Powder Down	Ventral	1.032±0.043	1.058±0.022	0.958±0.035
14	Powder Down	Tail	0.765±0.028	1.054±0.037	1.407±0.060
15	Bristle	Near Eye and Beak	0.316±0.008	0.222±0.011	0.240±0.008
16	Filoplume	Wings	0.288±0.017	0.216±0.009	0.235±0.009

IND – Individual.

Table 4.6. Feather microstructures in three individuals of *A. tristis*.

Sl. No.	Feather Type	Feather Location	Villi	Villi shape	Nodes	Node shape	Prongs	Prong size	Hooklets	Ventral teeth	Internode shape	Pigmentation		
												Nodes	Internodes	Ramus
1	Wing Feather	Right Wing	1	KNB, PNT	1	2, 3	1	S	1	1	STR	6	5	6
2	Tail Contour	Tail	1	KNB, PNT	1	2, 3	1	S	1	1	STR	6	5	6
3	Body Contour	Dorsal &Ventral	1	KNB, PNT	1	2,3,4	1	S	1	0	STR	6	5	5, 6
4	Semiplume	Dorsal, Ventral & Tail	1	KNB, PNT	1	2,3,4	1	S	0	0	STR	6,5	5	5,6
5	Down	Dorsal, Ventral & Tail	1	KNB, PNT	1	2,3,4	1	S	0	0	STR	6	5	5, 6
6	Powder Down	Dorsal, Ventral & Tail	1	KNB, PNT	1	2, 3	0	NA	0	0	STR	5,6	5	6
7	Bristle	Near eye and beak	1	KNB, PNT	1	2	1	L	0	0	STR	6	5	6
8	Filoplume	Wings	0	NA	1	2	1	L	0	0	STR	5	5	5, 6

Legends: 0= Absent, 1= Present, KNB= Knobbed shape, PNT= Pointed shape, 2= Plain pronged node shape, 3=Plain nodes without prongs, 4= Quadrilobed node shape, S= Small, STR= Straight, 5= Patchy pigmentation, 6= Dark/dense pigmentation, NA=Not applicable.

Table 4.7. Feather macro-characteristics of *S. chinensis*.

Sl. No.	Feather Type	Feather Location	Colour	Texture
1	Primary Contour Feather	Right Wing	Dark Brown	Rigid
2	Secondary Contour Feather	Right Wing	Dark Brown	Rigid
3	Tail Contour	Tail	Dark Brown	Rigid
4	Body Contour	Ventral Side	Creamy with rusty tinge	Semi-Rigid
5	Body Contour	Dorsal Side	Creamy with black tinge	Semi-Rigid
6	Semiplume Feather	Ventral Side	Creamy with pale brown tinge	Soft and Fluffy
7	Semiplume Feather	Dorsal Side	Creamy with pale brown tinge	Soft and Fluffy
8	Down Feather	Ventral Side	Creamy with black tinge	Soft and Fluffy
9	Down Feather	Dorsal Side	Creamy with black tinge	Soft and Fluffy
10	Powder Down	Ventral Side	Pale brown	Soft and Fluffy
11	Powder Down	Dorsal Side	Pale brown	Soft and Fluffy
12	Bristle	Near eyes and beak	Pale brown	Rigid
13	Filoplume	Right and left wings	Transparent, slightly blackish barbs	Soft

Table 4.8. Feathers morphometric measurements of *S. chinensis*.

Sl. No.	Feather Type	Feather Location	Length of calamus (cm ± S.E.)	Length of Vane (cm ± S.E.)	Length of rachis (cm ± S.E.)
1	Wing Feather	Right Wing	1.33±0.07	8.45±0.27	9.90±0.41
2	Tail Contour	Tail	1.25±0.05	8.15±0.05	10.45±0.05
3	Body Contour	Dorsal & Ventral Side	0.30±0.04	3.25±0.06	3.53±0.06
4	Semiplume Feather	Dorsal & Ventral Side	0.20±0.00	3.25±0.52	3.45±0.52
5	Down Feather	Dorsal & Ventral Side	0.25±0.03	2.00±0.09	2.25±0.06
6	Powder Down	Dorsal & Ventral Side	0.25±0.03	NA	NA
7	Bristle	Near eyes and beak	0.36±0.02	2.20±0.07	2.50±0.06
8	Filoplume	Right and left wings	NA	NA	2.5±0.045

Table 4.9. Average barb length of *S. chinensis*.

Sl. No.	Feather Type	Feather Location	Average barb length ± S.E. in cm
1	Wing contour feather	Wing	1.653±0.054
2	Tail contour feather	Tail	1.840±0.119
3	Body Contour	Dorsal	1.119±0.026
4	Body Contour	Ventral	1.241±0.028
5	Semiplume	Dorsal	1.065±0.017
6	Semiplume	Ventral	1.312±0.037
7	Down	Dorsal	1.160±0.020
8	Down	Ventral	1.082±0.017
9	Powder Down	Dorsal	0.888±0.036
10	Powder Down	Ventral	1.067±0.022
11	Bristle	Near eyes and beak	0.985±0.034
12	Filoplume	Right and left wings	1.003±0.002

Table 4.10. Feather microstructures of *S. chinensis*.

Sl. No.	Feather Type	Feather Location	Villi	Nodes	Node shape	Prongs	Prong size	Hooklets	Ventral Teeth	Internode shape	Pigmentation		
											Nodes	Internodes	Ramus
1	Primary Contour Feathers	Right Wing	0	1	2,4	1	S	1	1	STR	5,6	5	5,6
2	Secondary Contour Feathers	Right Wing	0	1	2,4	1	S	1	1	STR	5,6	5	5,6
3	Tail Contour	Tail	0	1	2,4	1	S	1	1	STR	6	5	5,6
4	Body Contour	Dorsal &Ventral	0	1	2,3,4	1	S	1	1	STR	5,6	5	6
5	Semiplume	Dorsal & Ventral	0	1	2,3,4	1	S	1	1	STR	5,6	5,6	6
6	Down	Dorsal & Ventral	0	1	2,3,4	1	S	1	1	STR	5,6	5,6	6
7	Powder Down	Dorsal & Ventral	0	1	2,3,4	1	S	0	0	STR	5,6	5	6
8	Bristle	Near eye and beak	0	1	2, 3	1	S	0	0	STR	5,6	5	5,6
9	Filoplume	Right Wing & Left Wing	0	1	2	1	S	0	0	STR	5	5,6	5

Legends: 0= Absent, 1= Present, 2= Plain pronged node shape, 3=Plain nodes without prongs, 4= Crocus-shaped node shape, S= Small, STR= Straight, 5= Patchy pigmentation, 6= Dark/dense pigmentation.

Chapter 5

Chapter 5

Objective 3: Generate complete mitogenome sequences of selected traded avian species

5.1. Introduction

The mitochondrial genome (15–18 kb length) is a single circular extrachromosomal circular DNA consists of 13 protein coding genes (PCGs), two ribosomal RNA (rRNA), 22 transfer RNA (tRNA) genes, and a noncoding adenine(A) + thymine(T)-rich region (displacement loop region, D-loop) for most of the vertebrates (Satoh *et al.*, 2016). In comparison with nuclear genome; the mitogenomes possess multiple copies inside of a cell, they are small sized simple genomic organization which are maternally inherited with low sequence recombination and high rate of evolution, and almost unambiguous orthology (Liu *et al.*, 2014; Yan *et al.*, 2017; Sun *et al.*, 2020; Yu *et al.*, 2021; Zhang *et al.*, 2021; Yang *et al.*, 2022). The D-loop contains the mtDNA replication origin and promoters for mitochondrial RNA transcription and frequently used to discriminate individuals of a particular species or species with high phylogenetic divergence (Pereira *et al., 2010*). Low recombination ratio and high gene conservation rate projects mitogenomes as excellent molecular markers of a species for genetic studies (Brown *et al.*, 1979). The mitochondrial DNA (mtDNA) is considered an informative molecular tool widely used in a variety of genetic/evolutionary/forensic research (Duchene *et al.*, 2012; Wang *et al.*, 2016; Ko *et al.*, 2018).

The study of mitogenomes has a significant role in various fields such as biogeography, phylogenetics, evolution, forensics, population genetics and conservation biology (Sangster & Luksenburg, 2021; Zhang *et al.*, 2021). Even more, in the field of intensive palaeornithogical research too, DNA sequence-based analysis has proven a significant tool (Livezey & Zusi, 2007). Furthermore, mitogenomes transmit phylogenetic information more consistently throughout eruptive speciation episodes and are substantially preserved across speciation events (Rheindt & Edwards, 2011). Hence, mitogenomes present themselves as a dependable candidate to mirror the true phylogeny of a select taxon, in comparison to single/few nuclear gene-based phylogeny (Rheindt & Edwards, 2011; Campbell & Lapointe, 2011). Especially in the field of ornithology, several studies have been carried out on complete and partial mitogenome of birds in context of

their phylogenetics and evolution (Dey *et al.*, 2021; Sangster & Luksenburg, 2021; Sarkar *et al.*, 2021; Yu *et al.*, 2021; Zhang *et al.*, 2021). Authentic mitogenome sequences are very much crucial for identification of species which are used in a variety of research fields specially in taxonomy, systematics and wildlife forensic (Sangster & Luksenburg, 2021; Zhang *et al.*, 2021). However, a substantial number of sequenced mitogenomes of birds are faulty due to some issues such as misidentification, chimeras, poor sequence quality, or Nuclear mitochondrial DNA segments (NUMTs), incorrect sequence assembly and ID mislabelled in published papers and GenBank (Clark & Whittam, 1992; Arctander, 1995; Ruedas *et al.*, 1999; Sangster & Luksenburg, 2021). These types of published sequences are identified as the most rapidly growing problem for future studies on systematics, evolutionary biology and ecology.

In this chapter, we report mitogenome sequence and phylogenetic analyses of Common Myna (*Acridotheres tristis*) and Spotted Dove (*Spilopelia chinensis*). Both of these species are present in top 15 traded birds from Assam with high demand (Chapter 3). The mitogenome sequences are extremely useful for species identification in cases related to illegal wildlife trade/wildlife crimes. For instance, in absence of morphological data, insufficient evidence and high-quality bio-materials/samples (from wildlife crime sites), mitochondrial genes prove to be extremely useful for species identification. Further, mitogenomes of these two species will be useful for further studies on population genetics and to solve species distribution as well as taxonomic quarries.

Taking into account *A. tristis* commensal life history attributes, incongruent phylogeny and its potential as a biological pest, more ecological and genetic information in this regard is warranted. The influx of information on *A. tristis* species across its habitat range will assist in devising a healthy management plan for the above-mentioned species. Hence, we attempted to sequence the complete mitogenome of *A. tristis* species from India. Although a single sequence of *A. tristis* from China is available at NCBI GenBank, no descriptive mitogenomic analysis was added to its annexure. Further as mentioned above, genetic information from multiple geographic locations across its huge habitat range will efficiently aid in the management of *A. tristis* species. In this study, we decoded the complete mitogenome of *A. tristis* species using NGS tools, annotated and identified structural features of the newly sequenced mitogenome, compared the mitogenome with

previously available *A. tristis* mitogenome, and constructed a robust phylogenetic tree using select mitogenomes to elucidate a comprehensive phylogeny of Sturnidae and allied species.

Considering the unavailability of *S. chinensis* mitogenome from the Indian sub-continent, we attempted to sequence its complete mitogenome. Using NGS tools, successful sequencing of the *S. chinensis* mitogenome was achieved. *S. chinensis* mitogenome was annotated for various structural features, nucleotide composition/skew analysis and mitogenome based phylogenetic tree constructed to study its phylogenetic position within select species of Columbidae family.

The complete mitogenomes of *A. tristis* and *S. chinensis* would serve as reference, and provide species-specific mitochondrial DNA markers to identify species in case of wildlife crime.

5.2. Review of literature

Since the 1990s, mitochondrial genomes (mtDNA) have been most popularly used as molecular markers in phylogenetics and population genetics studies (Arctander, 1995; Palsboll *et al.*, 1995). The tremendous use of mitochondrial protein coding gene i.e., Cytochrome *b* (cyt *b*) for reconstructing the phylogenetic trees was really versatile (Arctander, 1995). However, Kocher *et al.* (1989) called the gene as "Universal" primers where the amplification of homologous fragments of cyt *b* occurs in mammals, birds, reptiles, fishes and invertebrates (Arctander, 1995). Although a few discrepancies were reported on the reconstruction of basal lineages based on mtDNA and nuclear genes which are affected by convergence, extinction of lineages, challenges of homology and alignment, selection and adaptation, and heterogeneity of evolutionary rates and branch-lengths (Meyer & Zardoya, 2003; Livezey & Zusi, 2007). Apart from using the inferring phylogeny (Hassanin *et al.*, 2005), the mitogenomes have been successfully used as molecular markers in the exploration of different avian phylogenies (Li *et al.*, 2016a; Mackiewicz *et al.*, 2019; Cai *et al.*, 2019). Recently, the use of high-throughput sequencing technology steps into the scenario with a real high number of mitogenome sequence generation for bird species (Morinha *et al.*, 2016; Yang *et al.*, 2018). Until 1st January, 2020, a total 1876 complete or partial mitogenomes of birds were published (Sangster & Luksenburg, 2021). As per the

previous research, a total of 1,559 numbers of available sequences are conspecific (Sangster & Luksenburg, 2021). Comparative studies of mitogenomes also show significant results for understanding mitogenomic characteristics and phylogeny for a particular order and family (Yang *et al.*, 2022).

Studies are limited on evolutionary, genetic and ecological aspects of *A. tristis*. The widely accepted Sturnidae phylogeny is based on concatenated ~5000 base pairs of mitochondrial and nuclear sequence sets, which needs further verification using whole mitochondrial genome phylogeny (Zuccon *et al.*, 2008; Lovette *et al.*, 2008). By using cytochrome *b* sequences Kang *et al.* (1994) conducted studies among three closely allied *Acridotheres* species for the better understanding of their phylogeny. A few recent studies have conducted on the phylogenetic relationships among songbird species (Barker *et al.*, 2004; Voelker & Spellman, 2004; Spicer & Dunipace, 2004; Eric&son & Johansson, 2003; Cibois & Cracraft, 2004; Alstro¨m *et al.*, 2006). Zuccon *et al.* (2006) reported on the relationship and the lineages of the family Sturnidae based on their nuclear as well as mitochondrial sequences. No published reportis available from India or Indian subcontinents on the complete mitogenome of *A. tristis* so far.

The family Columbidae consists of 300 extant species from 49 genera, yet the number of mitogenomes sequenced from Columbidae species are very scarce (Xu *et al.*, 2021). The complete mitogenome of Rock pegeon (*Columba livia*) (Kan *et al.*. 2010) and Egyptian swift Rock Pigeon (*Columba livia* breed Egyptian swift) (Li *et al.*, 2014) were reported from recent research. Till the year of 2015, mitogenomes of only five species from Columbidae family were available i.e., *Columba livia, Geotrygon violacea, Hemiphaga novaeseelandiae, Leptotila verreauxi* and *Zenaida auriculata*, and one extinct species (*Ectopiste migratorius*) (Huang *et al.*, 2015). Zhang *et al.* (2015) published the complete mitogenome of King pigeon (*Columba livia breed king*). Meanwhile, Yan *et al.* (2015) and Huang *et al.* (2015) reported about the complete mitogenome of *S. chinensis* from the locality of China. Further, multiple mitogenomes of a species from different geographic locations sre needed for comprehensive biogeographic and phylogenetic study of the same. In this aspect *S. chinensis* has no reported mitogenomes from India or Indian sub-continent.

5.3. Materials and methods

5.3.1. Sample collection and DNA extraction

Road-kill samples of *A. tristis* tissue was obtained from 26°34'28"N; 93°13'52"E (Figure 5.1) and tissue samples of *S. chinensis* was obtained from 26°34'31"N; 93°12'20"E (Figure 5.2) of the adjacent road stretches of Kaziranga National Park, Assam, India during a road-kill survey in September 2019 (Same individuals of the species used in Chapter-4, Objective-2); with due permission from the Office of the Principal Chief Conservator of Forests, Assam Forest Department (Ref. no. WL/FG.31/Pt/Technical Committee/2018) and Office order (No. 258, date: 11/01/2019) and Assam State Biodiversity Board (Ref. no: ABB/Permission/2012/82). The species identification was performed using a field guide (Grimmett *et al.,* 2016) and then re-verified using photographic evidence of the sample by professional ornithologists. The tissue samples were stored in a DMSO salt-buffer (Seutin *et al.,* 1991) and later under suitable conditions, it was transported to National Avian Forensic Laboratory (NAFL) at SACON, Coimbatore Tamil Nadu. At NAFL, samples were stored at -80°C avian bio-banking facility till further use. The tissue sample used for sequencing was stored in the avian bio-bank at NAFL under the accession code "NAFL/0277.7/T/24/06/19" for *A. tristis* and "NAFL/0286.3/T/27/9/19" for *S. chinensis*. About 25 mg of sampled muscle tissue were digested in lysis buffer (10 mMTris, 10 mM EDTA, 10% SDS and 40µg Proteinase K) and subsequent DNA extraction was carried out using the Phenol-Chloroform-Isoamyl Alcohol method with minor modifications (Sambrook *et al.,* 1989). The quality and quantity of the extracted DNA were estimated using DeNovix Spectrophotometer (DeNovix Inc., Delaware, USA) and Qubit-4 Fluorometer (Thermo Fisher Scientific, USA).

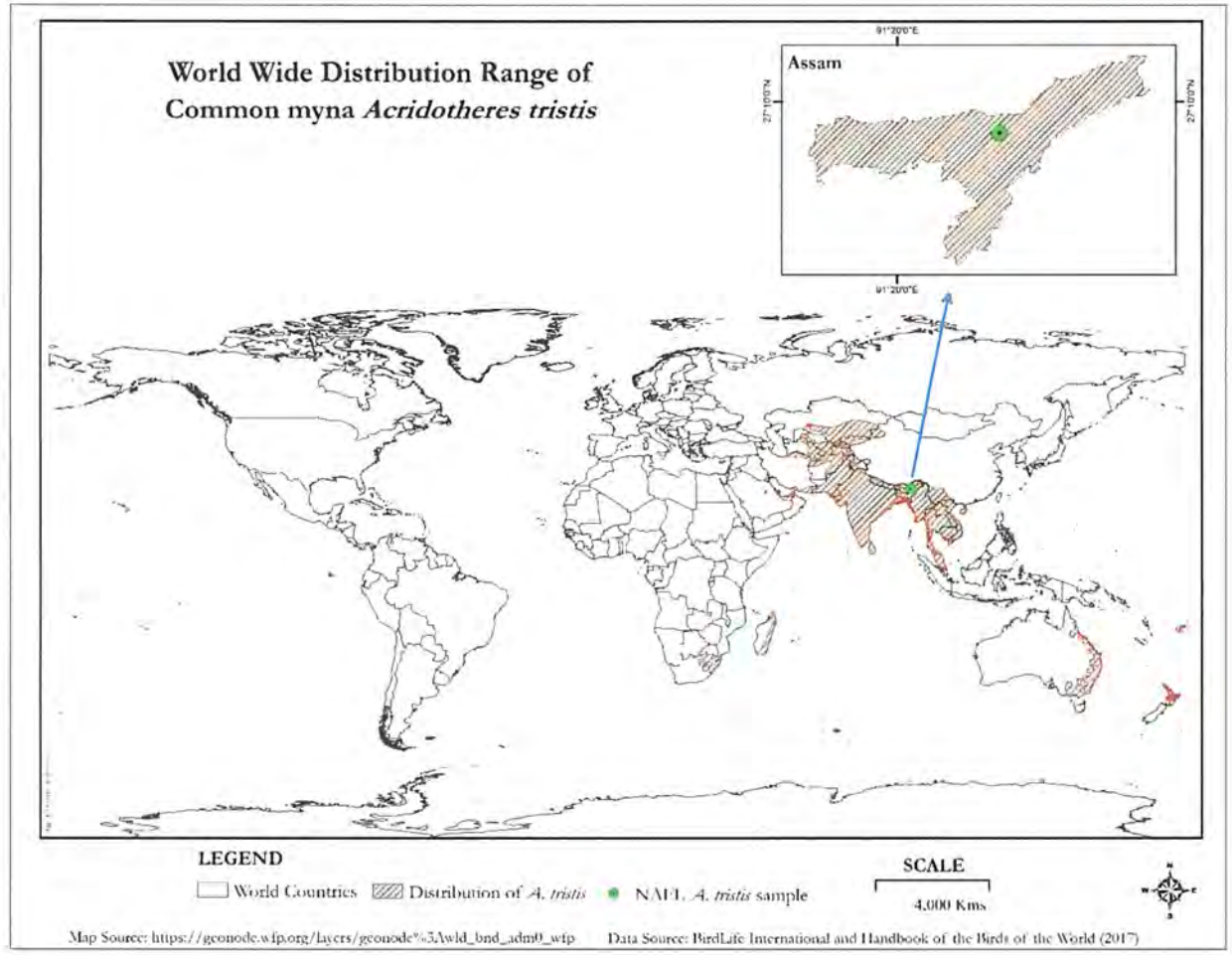

Figure 5.1. Spatial distribution and geographic range map of native residential distribution of *A. tristis* with the location of collected road-kill sample.

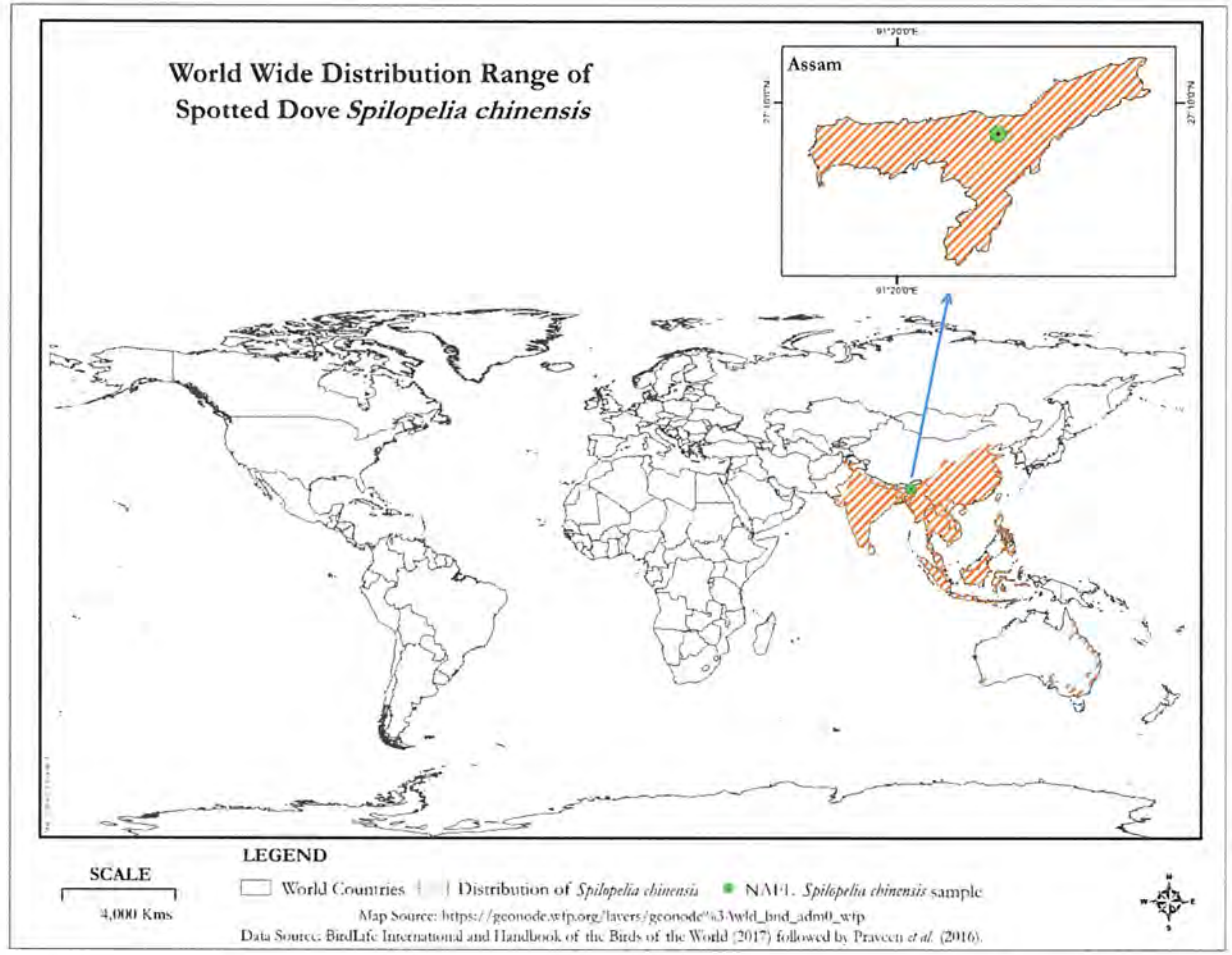

Figure 5.2. Spatial distribution and geographic range map of native residential distribution of *S. chinensis* with the location of collected road-kill sample.

5.3.2. Library preparation

High molecular weight DNA extracted from *A. tristis* and *S. chinensis* samples were utilized to construct a genomic library for sequencing. Paired-end 350 bp genomic libraries were prepared using a TruSeq DNA PCR-Free library preparation kit (Illumina Inc., USA). About 1.1 micrograms of the extracted DNA was fragmented to the targeted size using a focused ultrasonicator (Covaris M220, USA). Subsequent end-repair, poly-"A" adenylation and adapter ligation were accomplished following the manufacturer's protocol. A detailed protocol is outlined in previous studies by our group (Sarkar *et al.*, 2020, Dey *et al.*, 2021). The desired peak size of the library fragments was estimated using Fragment Analyzer (Agilent, USA) and quantification was carried out using QIAseq Library Quant Assay Kit (Qiagen N.V., Germany). The prepared library was sequenced using NextSeq550 (Illumina Inc., USA) housed at NAFL, SACON.

5.3.3. Mitogenome assemblage and gene annotation

(a) Common Myna (*Acridotheres tristis*)

The quality of the raw sequence obtained from NextSeq550 was checked using FastQC (http://www.bioinformatics.babraham.ac.uk/projects/fastqc/). High-quality raw reads were mapped directly onto the reference mitogenome (*A. tristis* accession no. NC_015195.1, hereafter mentioned as *A. tristis*_NC_015195.1) using Bowtie2 (Langmead & Salzberg, 2012). *A. tristis* mitogenome sequenced in this study was extracted out from the mapped reads as a single contig using IGV (Robinson *et al.*, 2011).

Structural features of the complete mitogenome of *A. tristis* sequenced in this study were initially predicted using MITOS2 (Donath *et al.*, 2019). Protein coding regions were identified using NCBI ORF Finder (Tatusov & Tatusov, 2011). Subsequently, boundaries of protein-coding genes (PCGs) were manually annotated by combining output from MITOS2 and gene boundaries of reference mitogenome. The ribosomal RNA (rRNA) genes were initially predicted by MITOS2 and the boundaries were confirmed by comparison with rRNA genes of the reference mitogenome. Transfer RNA (tRNA) positions and their putative secondary structures were surmised using tRNAscan-SE2.0 (Lowe & Chan, 2016). Nucleotide composition skew, AT-skew (A-T)/ (A+T) and GC-

skew (G-C)/ (G+C) was calculated manually (Perna & Kocher, 1995). A circular mitogenome map was constructed using the CGView server (Grant & Stothard, 2008).

(b) Spotted Dove (*Spilopelia chinensis)*

Read generated by NextSeq550 was assessed using FastQC (http://www.bioinformatics.babraham.ac.uk/projects/fastqc/). High quality reads were mapped directly onto the reference mitogenome (NCBI GenBank accession no. NC_026459.1) using Bowtie2 (Langmead & Salzberg, 2012). The assembled mitogenome was fished out as a single contig using IGV (Robinson *et al.*, 2011) and further analysis was conducted.

Utilizing MITOS2, structural characteristics of *S. chinensis* mitogenome sequenced in this study was predicted (Donath *et al.*, 2019). Further these predictions were verified using manually mapping to reference mitogenome (rRNAs), boundaries of protein coding genes (PCG) using NCBI ORF Finder (Tatusov & Tatusov, 2011) and tRNAs using tRNAscan-SE2.0 (Lowe & Chan, 2016). AT-skew {(A-T)/(A+T)} and GC-skew {(G-C)/(G+C)} of the nucleotide composition were manually estimated (Perna & Kocher, 1995) and using the CGView server, a circular mitogenome map was created (Grant & Stothard, 2008).

5.3.4. Phylogenetic analysis

For the phylogenetic analysis of *A. tristis,* Complete mitochondrial genomes retrieved from the NCBI GenBank database included the genera of *Passer, Melaenornis, Copsychus, Niltava, Ficedula, Myophonus, Phoenicurus, Monticola, Oenanthe, Luscinia, Buphagus, Sturnus, Acridotheres, Leucopsar, Toxostoma, Rhabdornis, Gracula, Myadestes, Turdus, Zoothera* and *Catharus* (Table 5.1). A total of 30 mitogenomes from 21 genera, along with the newly sequenced *A. tristis* mitogenome were analysed for phylogenetic topology. All the select mitogenomes were concatenated to their 13 PCGs for phylogenetic tree construction.

For the phylogenetic analysis of *S. chinensis*, mitogenomes from *Streptopelia, Columba, Patagioenas, Ectopistes, Hemiphaga, Didunculus, Goura, Zenaida* and *Gallus* genera were retrieved from NCBI GenBank database (Table 5.2). All the select

mitogenomes along with *S. chinensis* mitogenome sequenced in this study were concatenated to their 13 PCGs for phylogenetic tree construction.

For both of the species, sequence alignment was carried out using the MUSCLE program in Unipro UGENE (Okonechnikov *et al.*, 2012). At first, the best-fit nucleotide substitution model for the aligned sequences was identified as GTR+F+I+R4 using ModelFinder (Kalyaanamoorthy *et al.*, 2017). Subsequently, a phylogenetic tree was constructed employing the Maximum Likelihood algorithm (utilizing the GTR+F+I+R4 model) in IQ-TREE version 1.6.12 (Nguyen *et al.*, 2015) and ran for 10000 bootstrap replicates. FigTree ver 1.4.4 was used to visualize and edit the resulting phylogenetic tree (Rambaut, 2007).

5.4. Results

5.4.1. Common Myna (*Acridotheres tristis*)

5.4.1.1. Mitogenome organization and gene arrangement

The complete mitogenome of *A. tristis* was successfully sequenced, assembled, annotated and submitted to NCBI GenBank (accession no. OK542103, hereafter mentioned as *A. tristis*_ OK542103). *A. tristis* mitogenome was found to be 16822 base pairs in length. A total of thirteen PCGs, twenty-two tRNA, two rRNA and one control region (CR) constitute the *A. tristis* mitogenome (Table 5.3). Except for eight tRNAs (trnQ, trnA, trnN, trnC, trnY, trnS2, trnP and trnE) and one PCG (nad6) which were located on the light (-) strand, all the other genes were located on heavy (+) strand (Figure 5.3). The *A. tristis* mitogenome (OK542103) exhibited similar mitogenome length and gene arrangement as reported previously (Sarkar *et al.*, 2020; Sun *et al.*, 2020; Dey *et al.*, 2021) including mitogenome reports of a Sturnidae species (NCBI accession no. NC_014455) (Qian *et al.*, 2013). Following Sangster & Luksenburg (2021), the identity of our mitogenome sequence of *A. tristis*_ OK542103 was verified with reference sequences of three commonly used markers in Muscicapoidea systematics: nad2 (1041 bp; n=2466, incl. six of *A. tristis*), part of cox1 (696 bp; n=2263, incl. 23 of *A. tristis*), and cytb (1143 bp; n=744, incl. two of *A. tristis*). In each of these analyses, our sequence of *A. tristis* clustered with the reference sequences of *A. tristis*, indicating that our sample was correctly identified.

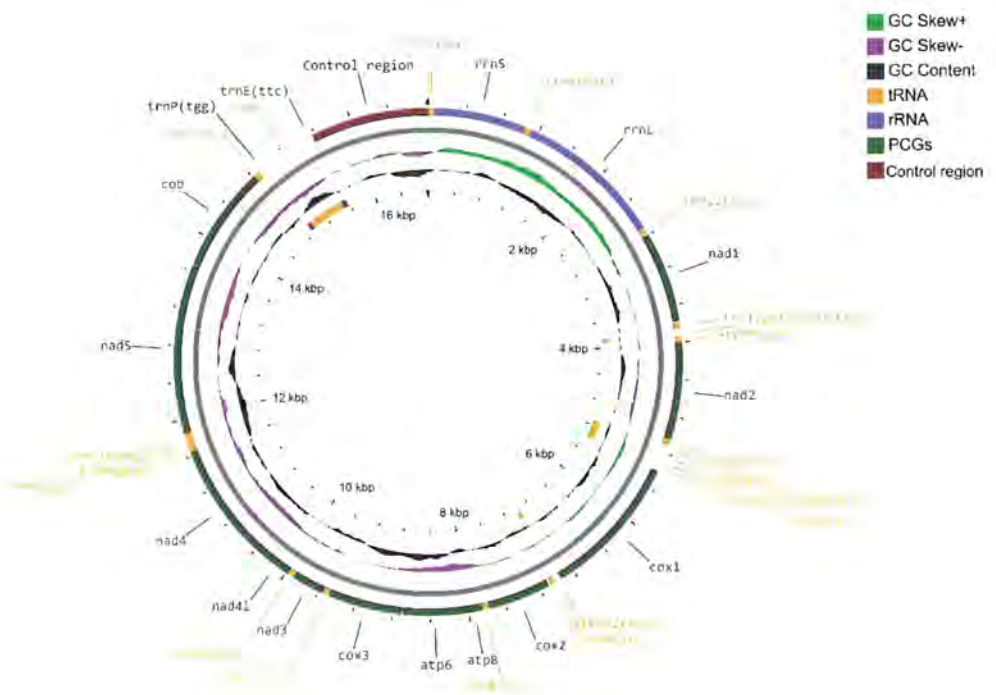

Figure 5.3. Circular map of *A. tristis* mitogenome. Various genes are represented with different colour blocks legend for which is situated in the top right corner of the image. Black sliding window indicates GC content of the regions and GC skew is represented through purple sliding window.

Nucleotide composition of *A. tristis*_ OK542103 mitogenome was estimated as A: 4960 (29.5%), C: 5475 (32.5%), G: 2588 (15.4%) and T: 3799 (22.6%) (Table 5.4). The (A+T)% and AT-skew were estimated at 52.1% and 0.13 respectively, showing AT richness and nucleotide composition skewed towards AT richness (Table 5.4). The nucleotide base composition and base skewness of our *A. tristis* mitogenome were nearly identical with minute variations with the already existing mitogenome. The single nucleotide variations between the new and old *A. tristis* mitogenomes (OK542103 and NC015195.1) were noticed in the genes: nad2 (3 bp), cox1 (2 bp), atp6 (2 bp), cox3 (3 bp), nad4l (1 bp), nad4 (1 bp), nad6 (2 bp), 12s rRNA (1 bp), 16s rRNA (3 bp), tRNAD (1 bp), tRNAS1 (1 bp) and CR region (1 bp). Further pairwise differentiation of the two *A. tristis* mitogenomes (OK542103 and NC015195.1) estimated a sequence similarity of 99.9% with only 22 nucleotides variation among them. High AT bias is typical of most vertebrate

mitogenomes, including the members of the Sturnidae family (Qian *et al.*, 2013; Sarkar *et al.*, 2020; Sun *et al.*, 2020; Dey *et al.*, 2021).

5.4.1.2. Protein Coding Genes, ribosomal and transfer RNA genes

The total length of PCGs concatenated together was estimated at 11395 base pairs (approximately 3798 codons) (Table 5.4). Among the 13 annotated PCGs of *A. tristis*_ OK542103 mitogenome, 12 PCGs initiated with the start codon ATG except for cox1 gene which started with GTG codon. TAA was the most abundant stop codon, apart from PCGs such as nad1, cox1, nad5 and nad6 which terminated with stop codon AGG, AGG, AGA and TAG respectively. In nad2, cox3 and nad4 PCGs the TAA stop codon was incomplete and required poly "A" adenylation at their 3' end to form a complete TAA stop codon. Overlapping of sequences was observed between atp8/atp6 and nad4l/nad4 PCGs to the degree of 10 and 7 nucleotides respectively. Generally, many attributes of PCGs in avian mitogenomes mentioned above are conserved across species and families including members of the Sturnidae family (Qian *et al.*, 2013; Sarkar *et al.*, 2020; Sun *et al.*, 2020; Dey *et al.*, 2021). Interestingly frameshift mutation observed in nad3 PCGs of numerous previously reported avian mitogenomes was not identified in *A. tristis* mitogenomes (OK542103 and NC015195.1) (Mindell *et al.*, 1998; Dey *et al.*, 2021). Such an arrangement of the nad3 gene may hint at possible independent evolution of the nad3 gene in the *Acridotheres* genus, as opposed to the frameshift mutation reported from previous avian mitogenome studies.

*A. tristis*_ OK542103 mitogenome housed two rRNAs on its heavy chain, namely the smaller 12s rRNA and larger 16s rRNA. The 12s and 16s rRNAs were comprised of 979 and 1598 base pairs respectively. The rRNAs were situated between trnF and trnL2, with trnV separating the 12s and 16s rRNAs. The nucleotide composition of the rRNAs was characterized by slightly higher A+T% (53.22%) and positive AT-skew (0.22), showing an AT bias in its composition (Table 5.4). *A. tristis*_NC015195.1 mitogenome also exhibits similar positioning of rRNAs, length and nucleotide composition. The positioning, length and high AT-skew of rRNAs are similar across wide variety of avian taxa including Sturnidae species (Qian *et al.*, 2013; Sarkar *et al.*, 2020; Sun *et al.*, 2020; Dey *et al.*, 2021). A total of 22 tRNAs were identified in *A. tristis*_ OK542103

mitogenome and their putative secondary structures were predicted (Figure 5.4). The length of tRNAs predicted in the sequenced mitogenome ranged between 63 to 75 base pairs (Table 5.3). Total length of all tRNAs concatenated together was estimated to be 1542 base pairs. The A+T% of tRNA sequences was calculated at 57.2% and AT-skew of 0.13, indicating an A+T biased nature. The putative tRNA structures showed a conserved cloverleaf model in *A. tristis*_OK542103 mitogenome. It was found that tRNAs for alanine, cysteine, glutamic acid, histidine, leucine (L2), methionine, serine (S1 and S2) and tyrosine had wobble base pair in their acceptor arm whereas trnD, trnM, trnP and trnS (S2) had wobble base pairing in T loop. The DΨU loop of trnA, trnC, trnE, trnQ, trnP and trnW also had wobble base pairs (Figure 5.4).

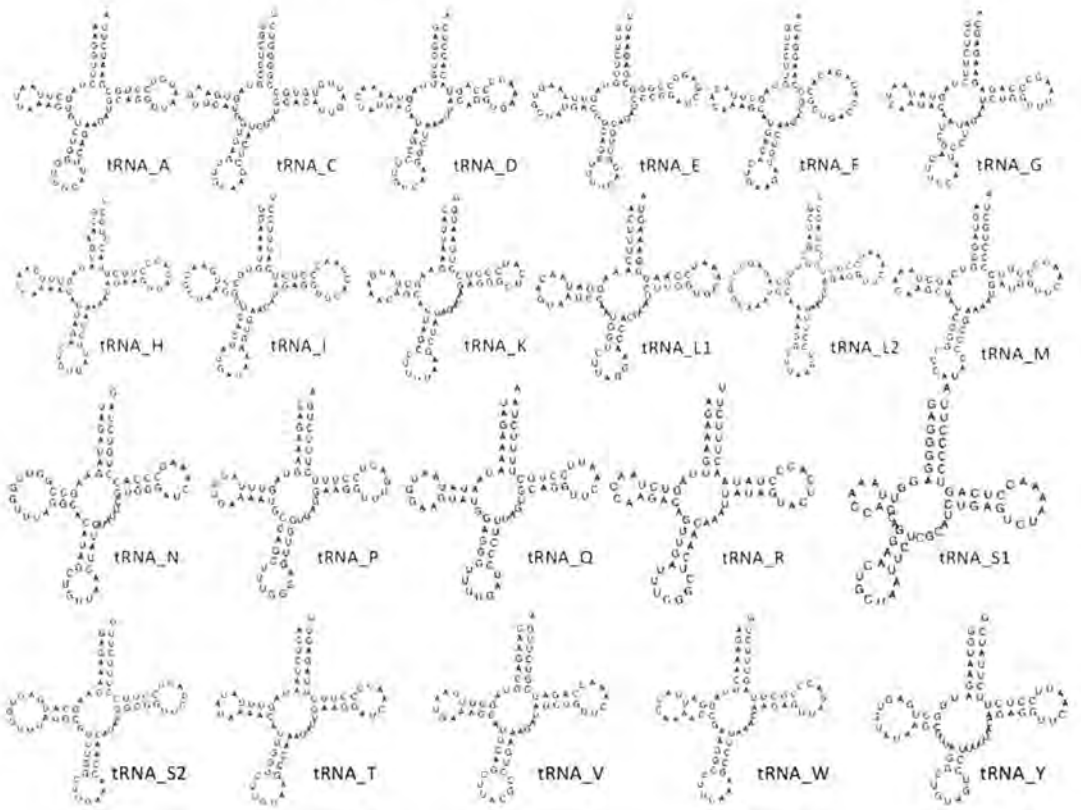

Figure 5.4. Putative secondary structure for 22 tRNAs predicted in *A. tristis* mitogenome.

5.4.1.3. Mitochondrial control region

The D-loop or mitochondrial control region (CR) of *A. tristis*_ OK542103 mitogenome was estimated to be 1251 base pairs in length. The nucleotide composition of the CR region was calculated at A+T% at 54.7%, indicating AT richness similar to previously reported *A. tristis*_NC015195.1 mitogenome and in other avian mitogenomes including members of the Sturnidae family (Qian *et al.*, 2013; Sarkar *et al.*, 2020; Sun *et al.*, 2020; Dey *et al.*, 2021). This high A+T% in the CR plays an important role in the initiation, replication and transcription related activities of the mitogenome (Urantówka *et al.*, 2018). Further the genes trnT, trnP, trnE and nad6 along CR region are believed to evolve under events of duplication, pseudogenization and genome rearrangement, making it an active avenue of evolution studies (Urantówka *et al.*, 2018). Hence, the arrangement of genes in and around the mitochondrial CR region has been of particular interest to biologists. We report through this study the *A. tristis* mitogenomes (OK542103 and NC015195.1) display ancestral avian CR gene order (Desjardins & Morais, 1990; Urantówka *et al.*, 2018). Several avian families (Charadriiformes, Accipitriformes, Psittaciformes, Galliformes, and Passeriformes) include species that may display different CR gene orders respectively, including ancestral and various duplicated CR gene orders. It may be inferred that the 'typical' or 'ancestral' avian gene order observed in multiple species randomly across wide varieties of avian families (Charadriiformes, Accipitriformes, Psittaciformes, Galliformes, Passeriformes) may hint at convergent evolution of gene arrangement in and around mitochondrial CR region.

5.4.1.5. Phylogenetic analysis

A robust phylogenetic tree was constructed using mitogenomes from 7 families, 21 genera and 30 species. The tree included 9 species from the Sturnidae family and the other 21 from allied families of the Passeriformes order whose mitogenome was retrieved from NCBI GenBank. The clustering in the phylogenetic tree generated in this study was supported by high nodal values (~100). Species of the Sturnidae family analysed in the phylogenetic tree clustered together in a separate node with the nearest phylogenetic member being *Buphagus erythrorhynchus* (NC053077.1) from the Buphagidae family (Figure 5.5). However, inclusion of *Toxostoma redivivum* (NC053053.1) species from the

Mimidae family was also noticed within the clustering of the Sturnidae family analysed in the phylogenetic tree. Amongst the Sturnidae genera *Acridotheres*, *Sturnus*, *Gracula*, *Leucopsar* and *Rhabdornis* clustered together. Members of *Acridotheres* genus clustered closely in the same clade with high nodal values indicating successful mitogenome sequencing of *A. tristis*_ OK542103. All the three *Acridotheres* mitogenomes are clustered together originating from a single node. Select members of *Acridotheres* were phylogenetically close to *Sturnus cineraceus* and *Sturnus sericeus*.

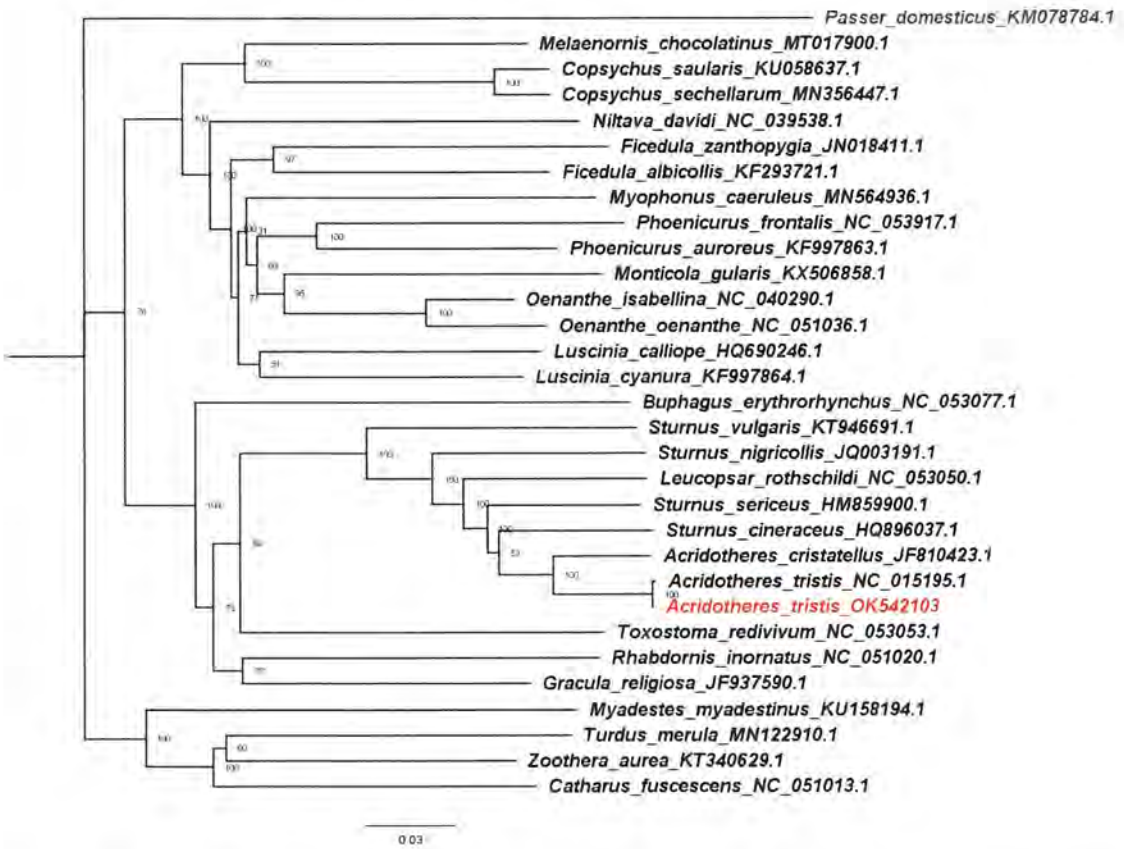

Figure 5.5. ML tree based on the phylogenetic relationships of 31 mitogenomes determined using concatenated PCGs. Tree construction was achieved in IQ-TREE with TVM+F+R4 model with 10000 bootstrap replicates. *A. tristis* mitogenome sequenced in this study is marked in bold red colours and out-group in green.

A previous study on phylogeny of starlings and mockingbirds included 120 species with evidence from ~8000 bp of mitochondrial and nuclear gene sequences, found no

incongruence between members of Mimidae and Sturnidae family (Lovette & Rubenstein, 2007). The phylogenetic incongruence of Mimidae family with Sturnidae observed in this study may be attributed to inadequate taxon sampling (30 species, 1 Mimidae), owing to low number of available mitogenomes of starlings and allied species. Further utilization of only mitochondrial PCGs for phylogenetic analysis on such a dataset may have induced clustering of *T. redivivum* within members of the Sturnidae family. Although initial phylogenetic studies had strongly recommended monophyly of the *Sturnus* genus by subsuming *Acridotheres* into *Sturnus*, subsequent studies have clearly separated the two genera into *Acridotheres* and *Sturnus* (Zuccon *et al.*, 2007). As such in our phylogenetic tree, we also re-iterated the same evidence through whole mitogenome phylogeny. Further, in previous reports (Zuccon *et al.*, 2007; Lovette *et al.*, 2008) *Acridotheres* were estimated to be closely related to *Sturnus cineraceus* and *Sturnus sericeus*, the same was also elucidated from the whole mitogenome phylogeny reconstructed in this study.

5.4.2. Spotted Dove (*Spilopelia chinensis*)

5.4.2.1. Mitogenome organization and gene arrangement

S. chinensis mitogenome was successfully sequenced, assembled and annotated in this study. The mitogenome of *S. chinensis* was deciphered to be 16966 nucleotides in length, encasing thirteen PCGs, twenty-two tRNAs, two rRNA and one control region (CR) (Table 5.5). Majority of the structural attributes (PCGs, tRNAs and rRNAs) are located on the heavy (+) strand whereas eight tRNAs (trnQ, trnA, trnN, trnC, trnY, trnS2, trnP and trnE) and one PCG (nad6) is located on the light (-) strand of the mitogenome (Figure 5.6). The length, structural attributes and arrangement of genic features of *S. chinensis* mitogenome is strikingly similar to previously reported mitogenomes of Columbidae family (Huang *et al.*, 2015; Chen *et al.*, 2020) and of other avian families (Sarkar *et al.*, 2020, Sun *et al.*, 2020, Dey *et al.*, 2021). *S. chinensis* mitogenome contained nucleotides in the following ratio: A: 5096 (30.0%), C: 5400 (31.8%), G: 2371 (14.0%), T: 4097 (24.1%) and 2 unresolved bases (1'N', 1'R'). The nucleotide composition of *S. chinensis* mitogenome was estimated to be AT rich (68.1% and AT skew at 0.10) as compared to GC content of 38.2% (GC skew at -0.38) (Table 5.6). Previously reported mitogenomes of Columbidae and other avian families also displayed high AT richness in their nucleotide composition (Huang

et al., 2015; Chen *et al.*, 2020, Sarkar *et al.*, 2020; Sun *et al.*, 2020; Dey *et al.*, 2021). Similar trend was observed by *S. chinensis* mitogenome analysed in this study.

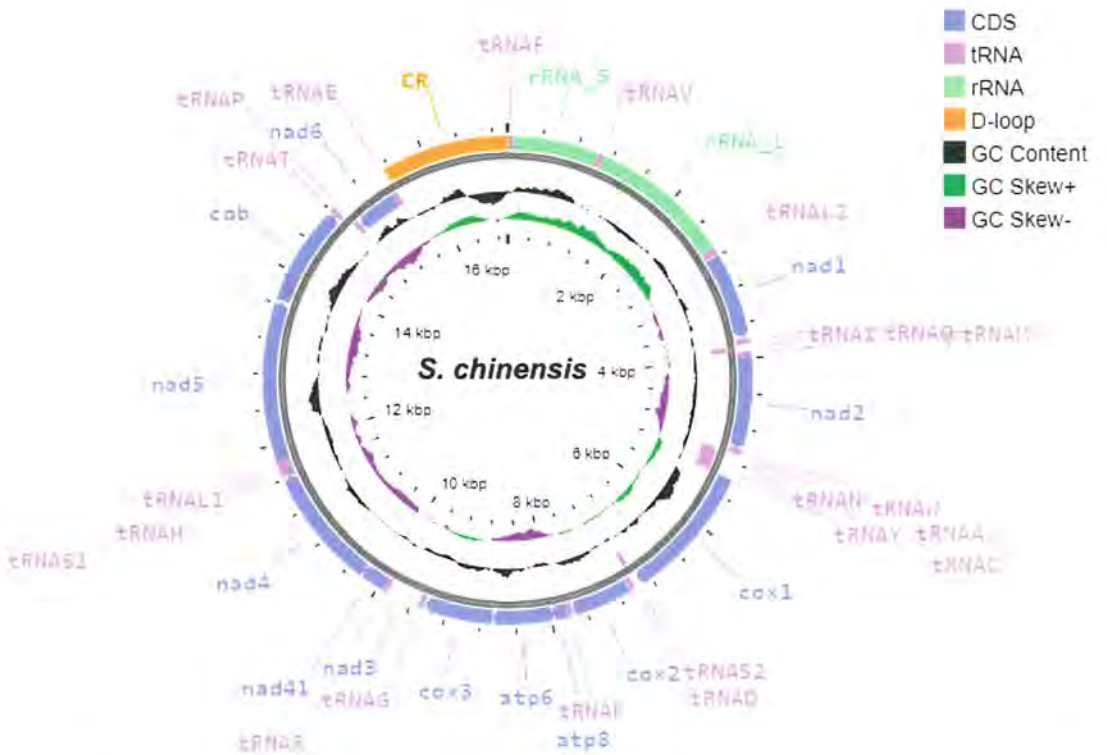

Figure 5.6. Circular map of *S. chinensis* mitogenome. Various genes are represented with different colour blocks legend for which is situated in the top right corner of the image. Black sliding window indicates GC content of the regions and GC skew is represented through purple sliding window.

5.4.2.2. Protein Coding Genes, ribosomal and transfer RNA genes

It was estimated that the combined length of the PCGs was 11395 base pairs (Table 5.6) in *S. chinensis* mitogenome. Concatenated PCGs were estimated to contain 63.5% 'A+T' nucleotides, displaying AT richness (Table 5.6). Amongst the 13 PCGs, 10 were coded with ATG as the most common start codon, except for nad5, cox1 (GTG as start codon) and nad3 (ATT as the start codon). PCGs such as nad1, nad2, cox1, nad3 and nad5 which terminated with stop codon AGA, TAG, AGG, TAA and TAG, respectively. Remaining PCGs terminated with TAA as the most common stop codon, including cox3 and nad4 which terminated with incomplete TAA stop codons. In previously reported avian

mitogenomes similar characteristics of PCGs have been observed (Sarkar *et al.*, 2020; Sun *et al.*, 2020; Dey *et al.*, 2021). Frame shift mutation has been observed in nad3 gene across avian families and taxa, and also reported in newly generated *S. chinensis* mitogenome in this study.

The 12s rRNA and 16s rRNA present in *S. chinensis* mitogenome, comprised of 971 and 1569 bp in length. They were placed between trnF and trnL2, with trnV separating them and were situated on the heavy chain. rRNAs annotated in *S. chinensis* mitogenome displayed high A+T% (59.7%) and a positive AT skew (0.22). Many different avian taxa, including those belonging to the Columbidae family, share similarities in the placement, length, and high AT-skew of rRNAs as identified in *S. chinensis* mitogenome (Huang *et al.*, 2015; Chen *et al.*, 2020; Sarkar *et al.*, 2020; Sun *et al.*, 2020; Dey *et al.*, 2021).

S. chinensis mitogenome was searched for tRNAs and a total of 22 tRNAs were identified and their secondary structures predicated (Figure 5.7). The length of tRNAs ranged between 69-74 bp and concatenated to a total 1427 bp. The A+T richness was estimated at 56.7% with a skew value of -0.001. Except for trnS2 which had a 'D-arm' missing, all other tRNAs displayed normal clover leaf shaped structure.

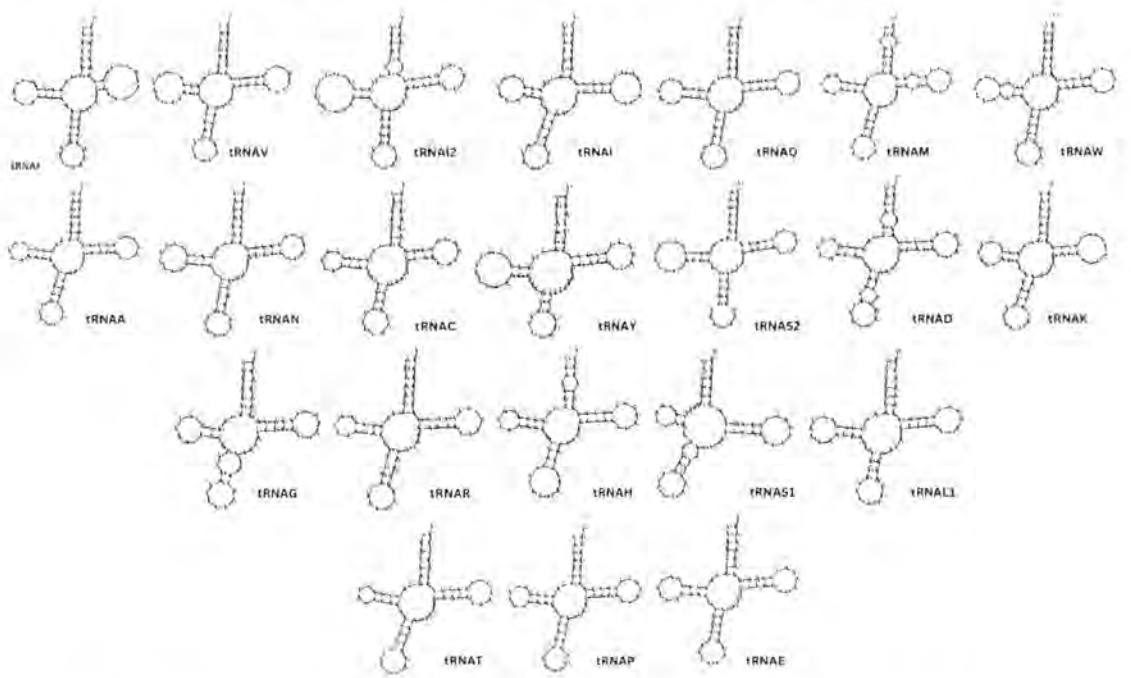

Figure 5.7. Putative secondary structure for 22 tRNAs predicted in *S. chinensis* mitogenome.

S. chinensis mitogenome sequenced in this study was compared with previously available mitogenomes of Spotted dove submitted in NCBI GenBank repository (NC_026459 and KP636801), and a dissimilarity of 5% in nucleotide sequences with difference of 763 to 767bp was observed in newly generated mitogenome. Upon further comparison, it was noticed that all the differences in nucleotide sequence of *S. chinensis* mitogenome sequenced in this study with NC_026459 and KP636801 were only point mutations and were distributed throughout the length of the mitogenome.

5.4.2.3. Mitochondrial control region

The *S. chinensis* mitogenome sequenced in this study housed a D-loop or mitochondrial regulatory region (CR) that was around 1251 base pairs long. The CR region's nucleotide makeup was determined to be 56.7% A+T, showing high AT richness. Generally the mitochondrial CR region displays high A+T content, which is believed to be essential for initiation, replication and transcription related activities of the mitogenome (Urantówka *et al.*, 2018). Birds are characterized by a unique arrangement of genes (trnT, trnP, trnE and nad6) in and around mitochondrial control region. Such arrangement of genes (trnT, trnP, trnE and nad6) in and around mitochondrial control region is an active site of rapid evolution (Urantówka *et al.*, 2018). Birds across taxa display different CR gene orders, the *S. chinensis* mitogenome sequenced in this study displayed typical' or 'ancestral' avian gene order.

5.4.2.4. Phylogenetic analysis

We utilized 17 mitogenomes from the Columbidae family to construct mitogenome based phylogeny (Figure 5.8). The retrieved phylogenetic topology shows high nodal values for the clustered species in the phylogenetic tree. In this analysis the genus *Streptopelia* was considered similar to *Spelopelia*, as in absence of a comprehensive Columbidae phylogeny, segregation of the above-mentioned genera is beyond the scope of the study. We reported that the *S. chinensis* mitogenome sequenced in this study was monophyletic with other *Streptopelia* species. All the three *Spilopelia/Streptopelia* mitogenomes clade together on a single node and are closest to *S. decaocto, S. orientalis* and *S. tranquebarica*. Similar clustering has been reported by previous studies (Johnson *et al.*, 2001; Huang *et al.*, 2015; Chen *et al.*, 2020). Further, *Columba* is more closely related to *Streptopelia/Spelopelia*

genus than any other Columbidae genus. Similar observations were also reported in previous studies (Johnson *et al.*, 2001; Huang *et al.*, 2015; Chen *et al.*, 2020). In the absence of a large number of mitogenomes from the Columbidae family, we present only limited phylogenetic inference using the available data set. However, our inferences align with previously reported studies on Columbidae phylogeny.

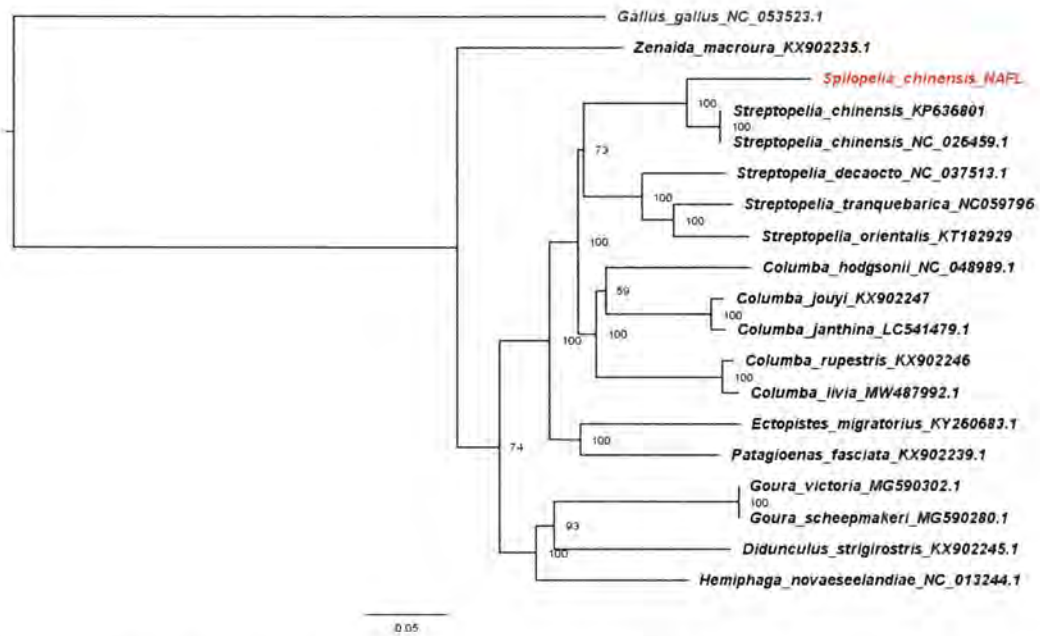

Figure 5.8. ML tree based on the phylogenetic relationships of 17 mitogenomes determined using concatenated PCGs. Tree construction was achieved in IQ-TREE with TVM+F+R4 model with 10000 bootstrap replicates. The sequenced mitogenome of *S. chinensis* is marked in bold red colours and out-group in green.

5.5. Discussion

This chapter provides for the first time detailed comparative mitogenomic and phylogenetic elucidation of *A. tristis* mitogenome and *S. chinensis* mitogenome.

The newly sequenced mitogenome of *A. tristis* was slightly biased towards AT richness. The mitochondrial CR region of *A. tristis*_OK542103 was arranged as their ancestral avian CR gene order. Topology inferred from the whole mitogenome phylogeny provided evidence for clustering of *Mimidae* species within the Sturnidae family. Further, *Sturnus* genus also was inferred to be monophyletic. Select *Acridotheres* members were

found to be phylogenetically close to *Sturnus cineraceus* and *Sturnus sericeus*. The complete mitogenome of *A. tristis*_OK542103 sequenced in this study provides important genetic resources for not only *A. tristis* species but also for *Acridotheres* as a genus. The mitogenome sequence generated here can be utilized to design genetic markers for forensic, evolutionary and life history studies of *A. tristis* and co-generic species. Further comparative mitogenomic analyses performed on select mitogenomes in this study, provides intriguing information regarding the evolution of Sturnidae species.

We elucidated the genetic features and structural characteristics of the *S. chinensis* mitogenome sequenced in this study. Further, we also present a mitogenome PCGs based phylogeny that highlights close relation of *Streptopelia/Spilopelia* genera to *Columba* amongst species of Columbidae family. We also compared our findings with previously available mitogenomes of *S. chinensis* species (NCBI accession no. NC026459.1, KP636801) and report variability of ~700 bp. Based on morphological cues *Spilopelia chinensis* is generally believed to include two major subspecies of *Spilopelia chinensis suratensis* and *Spilopelia chinensis chinensis* (Garrett & Walker, 2022). *Spilopelia chinensis chinensis* has an estimated distribution across the regions of Myanmar, central and eastern China, and Taiwan; whereas *Spilopelia chinensis suratensis* is predominantly distributed across the Indian sub-continent (Garrett & Walker, 2022). No detailed genetic or phylogeographic study has been published delimiting the sub-species and their respective geographic ranges of *S. chinensis* species complex (Garrett & Walker, 2022). In light of such intriguing sub-species distribution of *S. chinensis* (*sensu stricto*), the difference of ~5% in mitogenome sequences noticed in this study is valid and warrants further investigation. Such sequence dissimilarity points in the direction that the *S. chinensis* mitogenome sequenced in this study may belong to a different sub-species of *S. chinensis* (possibly *S. chinensis suratensis*) than the previously sequenced mitogenomes of *S. chinensis* species (NCBI accession no. NC026459.1, KP636801; possibly *S. chinensis chinensis*). However, detailed morphological and population genetic studies encompassing various geographic ranges of *S. chinensis* species are needed to comprehensively shed light on queries related to *S. chinensis* sub-species distribution and delimitation. Hence, in this study we have considered the old available name/combination of the species as *S. chinensis* (*sensu stricto*) following Grimmett *et al.* (2014) and Praveen *et al.* (2016, 2021). Thus, the

mitogenome sequence generated in this study can be used to develop genetic and forensic markers of *S. chinensis* species from India. Further descriptive understanding of *S. chinensis* mitogenome provides valuable insights into evolutionary biology and life history of Columbidae family.

Tables

Table 5.1. Family, genus and species information of mitogenomes analysed for *A. tristis* phylogenetic study along with their respective GenBank accession number.

Family	Genus	Species	GenBank Accession no.
Passeridae	*Passer*	*Passer domesticus*	KM078784
Muscicapidae	*Melaenornis*	*Melaenornis chocolatinus*	MT017900.1
Muscicapidae	*Copsychus*	*Copsychus sechellarum*	MN356447.1
Muscicapidae	*Copsychus*	*Copsychus saularis*	KU058637.1
Muscicapidae	*Niltava*	*Niltava davidi*	NC039538.1
Muscicapidae	*Ficedula*	*Ficedula zanthopygia*	JN018411.1
Muscicapidae	*Ficedula*	*Ficedula albicollis*	KF293721.1
Muscicapidae	*Myophonus*	*Myophonus caeruleus*	MN564936.1
Muscicapidae	*Phoenicurus*	*Phoenicurus frontalis*	NC053917.1
Muscicapidae	*Phoenicurus*	*Phoenicurus auroreus*	KF997863.1
Muscicapidae	*Monticola*	*Monticola gularis*	KX506858.1
Muscicapidae	*Oenanthe*	*Oenanthe oenanthe*	NC051036.1
Muscicapidae	*Oenanthe*	*Oenanthe isabellina*	NC040290.1
Muscicapidae	*Luscinia*	*Luscinia cyanura*	KF997864.1
Muscicapidae	*Luscinia*	*Luscinia calliope*	HQ690246.1
Buphagidae	*Buphagus*	*Buphagus erythrorhynchus*	NC053077.1
Sturnidae	*Sturnus*	*Sturnus vulgaris*	KT946691.1
Sturnidae	*Sturnus*	*Sturnus nigricollis*	JQ003191.1
Sturnidae	*Leucopsar*	*Leucopsar rothschildi*	NC053050.1
Sturnidae	*Sturnus*	*Sturnus sericeus*	HM859900.1
Sturnidae	*Sturnus*	*Sturnus cineraceus*	HQ896037.1
Sturnidae	*Acridotheres*	*Acridotheres cristatellus*	JF810423.1
Sturnidae	*Sturnus*	*Acridotheres tristis*	NC015195.1
Mimidae	*Toxostoma*	*Toxostoma redivivum*	NC053053.1
Sturnidae	*Rhabdornis*	*Rhabdornis inornatus*	NC051020.1
Sturnidae	*Gracula*	*Gracula religiosa*	JF937590.1
Turdidae	*Myadestes*	*Myadestes myadestinus*	KU158194.1
Turdidae	*Turdus*	*Turdus merula*	MN122910.1
Turdidae	*Zoothera*	*Zoothera aurea*	KT340629.1
Turdidae	*Catharus*	*Catharus fuscescens*	NC051013.1

Table 5.2. Family, Genus and species information of mitogenomes analysed for *S. chinensis* phylogenetic study along with their respective GenBank accession number.

Family	Genus	Species	GenBank Accession no.
Columbidae	*Streptopelia*	*Streptopelia tranquebarica*	NC059796
Columbidae	*Columba*	*Columba jouyi*	KX902247
Columbidae	*Streptopelia*	*Streptopelia orientalis*	KT182929
Columbidae	*Columba*	*Columba rupestris*	KX902246
Columbidae	*Columba*	*Columba livia*	MW487992.1
Columbidae	*Columba*	*Columba hodgsonii*	NC_48989.1
Columbidae	*Patagioenas*	*Patagioenas fasciata*	KX902239.1
Columbidae	*Ectopistes*	*Ectopistes migratorius*	KY260683.1
Columbidae	*Streptopelia*	*Streptopelia decaocto*	NC_37513.1
Columbidae	*Hemiphaga*	*Hemiphaga novaeseelandiae*	NC_13244.1
Columbidae	*Didunculus*	*Didunculus strigirostris*	KX902245.1
Columbidae	*Goura*	*Goura victoria*	MG590302.1
Columbidae	*Columba*	*Columba janthina*	LC541479.1
Columbidae	*Zenaida*	*Zenaida macroura*	KX902235.1
Columbidae	*Goura*	*Goura scheepmakeri*	MG590280.1
Columbidae	*Streptopelia*	*Streptopelia chinensis*	NC_026459.1
Columbidae	*Streptopelia*	*Streptopelia chinensis*	KP636801.1
Phasianidae	*Gallus*	*Gallus gallus*	NC_53523.1

Table 5.3. Summary of *A. tristis* mitogenome.

Name	Start	Stop	Strand	Length	Intragenic nucleotide	Start Codon	Stop Codon
trnF(gaa)	1	69	+	69	0		
rrnS	70	1048	+	979	0		
trnV(tac)	1049	1118	+	70	0		
rrnL	1119	2716	+	1598	0		
trnL2(taa)	2717	2791	+	75	7		
nad1	2799	3776	+	978	10	ATG	AGG
trnI(gat)	3787	3858	+	72	6		
trnQ(ttg)	3865	3935	-	71	-1		
trnM(cat)	3935	4003	+	69	0		
nad2	4004	5043	+	1040	0	ATG	TA(A)
trnW(tca)	5044	5113	+	70	1		
trnA(tgc)	5115	5183	-	69	7		
trnN(gtt)	5191	5263	-	73	0		
trnC(gca)	5264	5330	-	67	-1		
trnY(gta)	5330	5400	-	71	1		
cox1	5402	6952	+	1551	-9	GTG	AGG
trnS2(tga)	6944	7016	-	73	3		
trnD(gtc)	7020	7088	+	69	10		
cox2	7099	7782	+	684	1	ATG	TAA
trnK(ttt)	7784	7851	+	68	1		
atp8	7853	8020	+	168	-10	ATG	TAA
atp6	8011	8694	+	684	6	ATG	TAA

Name	Start	Stop	Strand	Length	Intragenic nucleotide	Start Codon	Stop Codon
cox3	8701	9484	+	784	0	ATG	T(AA)
trnG(tcc)	9485	9553	+	69	0		
nad3	9554	9904	+	351	1	ATG	TAA
trnR(tcg)	9906	9975	+	70	1		
nad4l	9977	10273	+	297	-7	ATG	TAA
nad4	10267	11644	+	1378	0	ATG	T(AA)
trnH(gtg)	11645	11714	+	70	1		
trnS1(gct)	11716	11779	+	64	0		
trnL1(tag)	11780	11850	+	71	0		
nad5	11851	13668	+	1818	8	ATG	AGA
cob	13677	14819	+	1143	3	ATG	TAA
trnT(tgt)	14823	14892	+	70	11		
trnP(tgg)	14904	14973	-	70	7		
nad6	14981	15499	-	519	0	ATG	TAG
trnE(ttc)	15500	15571	-	72	0		
Control region	15572	16822	+	1251			

Table 5.4. Nucleotide composition and skew values of *A. tristis* mitogenomes.

*A. tristis*_OK542103: nucleotide composition and skew values of different genic features									
	Size	A%	C%	G%	T%	A+T%	G+C%	AT-skew	GC-skew
mtDNA	16822	29.5	32.5	15.4	22.6	52.1	47.9	0.13	-0.36
PCGs	11395	28.7	35	14.1	22.2	50.9	49.1	0.13	-0.43
tRNA	1542	32.4	25.1	17.8	24.8	57.2	42.8	0.13	-0.17
rRNA	2577	32.5	26	20.7	20.7	53.2	46.8	0.22	-0.11
CR	1251	27.2	31.5	13.8	27.5	54.7	45.3	-0.01	-0.39
Comparative nucleotide composition of *A. tristis* mitogenomes (OK542103 and NC015195.1)									
	Size	A%	C%	G%	T%	A+T%	G+C%	AT-skew	GC-skew
OK542103	16822	29.5	32.5	15.4	22.6	52.1	47.9	0.13	-0.36
NC015195.1	16822	29.4	32.6	15.4	22.5	51.9	48.1	0.13	-0.36

Table 5.5. Summary of *S. chinensis* mitogenome.

Name	Start	Stop	Strand	Length	Start codon	Stop codon
trnF(gaa)	1	69	+	69		
rrnS	70	1038	+	971		
trnV(tac)	1039	1111	+	73		
rrnL	1112	2705	+	1569		
trnL2(taa)	2706	2779	+	74		
nad1	2791	3756	+	966	ATG	AGA
trnI(gat)	3773	3843	+	71		
trnQ(ttg)	3849	3919	-	71		
trnM(cat)	3919	3987	+	69		
nad2	3988	5026	+	1041	ATG	TAG
trnW(tca)	5027	5097	+	71		
trnA(tgc)	5099	5167	-	69		
trnN(gtt)	5170	5242	-	73		
trnC(gca)	5243	5309	-	67		
trnY(gta)	5309	5380	-	72		
cox1	5382	6932	+	1551	GTG	AGG
trnS2(tga)	6924	6997	-	74		
trnD(gtc)	7000	7068	+	69		
cox2	7071	7754	+	684	ATG	TAA
trnK(ttt)	7756	7825	+	70		
atp8	7827	7994	+	168	ATG	TAA
atp6	7985	8668	+	684	ATG	TAA
cox3	8668	9451	+	784	ATG	T(AA)
trnG(tcc)	9452	9520	+	69		
nad3	9521	9700	+	180	ATT	TAA
	9657	9872	+	216		

Name	Start	Stop	Strand	Length	Start codon	Stop codon
trnR(tcg)	9874	9942	+	69		
nad4l	9944	10240	+	297	ATG	TAA
nad4	10234	11611	+	1378	ATG	T(AA)
trnH(gtg)	11612	11680	+	69		
trnS1(gct)	11681	11746	+	66		
trnL1(tag)	11746	11816	+	71		
nad5	11796	13634	+	1839	GTG	TAG
cob	13646	14788	+	1143	ATG	TAA
trnT(tgt)	14788	14856	+	69		
trnP(tgg)	14864	14933	-	70		
nad6	14944	15465	-	522	ATG	TAG
trnE(ttc)	15469	15539	-	71		
Control region	15540	16966	+	266		

Table 5.6. Nucleotide composition and skew values of *S. chinensis* mitogenome.

***S. chinensis*–NAFL: nucleotide composition and skew values of different genic features**									
	Size	**A%**	**C%**	**G%**	**T%**	**A+T%**	**G+C%**	**AT-skew**	**GC-skew**
mtDNA	16966	30	31.8	14	24.1	61.8	38.1	0.10	-0.38
PCGs	11399	29.4	34.1	12.3	24.2	63.5	36.5	0.09	-0.46
tRNA	1545	32.1	25.6	17.3	25	57.7	42.3	0.12	-0.19
rRNA	2540	32.8	26.9	19.5	20.9	59.7	40.4	0.22	-0.15
CR	1427	28.7	28	14.6	28.8	56.7	43.4	-0.001	-0.31

Chapter 6

Chapter 6

Summary and Conclusion

6.1. Summary

The illicit trade of birds is widespread across the globe, causing the major concern of population declines of several elusive and endemic species. It is the fastest growing business with ease of moneymaking with minimum effort. The major driving factors of the illegal bird trades vary and include traditional beliefs, traditional medicines, different socio-cultural issues, superstitions, and the easiest connectivity among the participants in the trade. It is a serious matter of concern where the ongoing scenario always remains clandestine.

This study provides information for the first time on illegally traded avian species in Assam (Chapter 3). The state of Assam is a "Gate-way" of travel and transport and is marked as the "Transitory Route" for illegally traded birds in India due to its transboundary nature. Moreover, different socio-cultural practices followed by the local tribes also play a significant role in the illegal bird trade scenario in Assam. Using the snowball sampling method, a systematic market survey was conducted using a semi-structured questionnaire for illegally traded birds and local people's perceptions towards issues associated with the traded birds. In total, 130 local markets from 16 districts of Assam were surveyed by interacting with 1003 respondents. A total 82 bird species belonging to 33 families representing 12 orders including native and migratory birds were found to be involved in trade. Among 82 bird species, 13 were winter migrants. The respondents reported nine different trapping gears used by the bird trappers. According to the respondents, the number of traded species varied significantly (χ^2= 765.798, df= 81, p=0.001). Based on the local trade price and the demand, the bird species were divided into three categories, namely high (Rs. 300–500), moderate (Rs. 200–300), and low (Rs.1–200). The percentage of highly demanded bird species was 58% whereas 26% of bird species were of moderate demand and 16% were of low demand. The results showed a strong correlation between the traded avian species and their market price (r=0.60, p<0.001) largely reflecting the demand. The reasons behind the ongoing illegal bird trade in Assam were categorised into five major categories viz. Bush-meat, Plume trade, Pet trade, Superstition and Black magic

and Traditional/Cultural use. Bush-meat turns out for the reason for trading the highest numbers of species (specially, i.e., duck, other water birds, paddy field birds) from Assam during the winter season. Local people's perceptions towards the issues of illegal bird trade were also measured by using a Likert scale. People from different districts across Assam agreed that they have frequently encountered or know about birds being traded. The local people from Assam strongly agree about stopping the ongoing bird trade. The majority of them strongly agreed that the appropriate execution of strong legislation is the way to stop such cruel activities with the help of local NGOs as well as forest departments.

The lacuna in law enforcement combined with traditional/cultural ethics drives the ongoing bird trade in Assam. However, to book wildlife crimes in court, one has to have substantial evidence of the individuals and the wild bird species being traded. The main concern is the specific identification of the species and their categorization per the wildlife laws, especially when the criminals are very much aware of the pros and cons and always take care not to be caught by any law and legislation. In such case-scenarios, wildlife forensics is an efficient method for identifying the species using very minute amounts of available biological samples. In this study, microstructure and molecular methods for forensic analysis were opted for two illegally traded avian species from Assam in high demand in the trade. With due permission from the Forest department of Assam and Assam State Biodiversity Board, opportunistic road-kill surveys were conducted to collect biological samples of bird species. 13 individuals of five different bird species were collected, Common Myna (*Acridotheres tristis*) and Spotted Dove (*Spilopelia chinensis*) were the two species selected for generating detailed feather microstructure signature and molecular markers for identification of species from scarce biological materials.

Three individuals of *A. tristis* and a single individual of *S. chinensis* (due to the unavailability of more samples as road kill) were used to develop feather microstructure atlas (Chapter 4). The different types of feathers (i.e., Contour and non-contour feathers) were selected from five different body locations (Dey *et al.*, 2021) of the road-kill specimen of *A. tristis* and *S. chinensis*. Morphological characteristics (colour, pattern, texture) were quantified from images of feathers. Permanent slides were prepared using the dry mount method and observed under a light microscope. Different feather microstructures were documented using LaboMed Lx500 light microscope with image aR software. Both

pennaceous and plumulaceous barbs were studied for six different feathers from four different body regions of each species.

The colour of the *A. tristis* feathers varied from black and white to dark brown to pale white and brown, even dark brown with a tinge of white. Among all the feathers of *A. tristis*, only filoplume feathers showed an additional silvery appearance with pale black-colored barbs at its tip. The texture of feathers varied from being firmly rigid for flight contour feathers, i.e., primary contour, secondary contour, and tail contour feathers. Body contour feathers were semi-rigid, whereas, irrespective of their location on the body, the semiplume, down and powder down feathers were soft and fluffy in texture, while the bristles that are modified contour feathers are of rigid texture. Calamus length, vane length, and rachis length of the nine different types of feathers were measured for feather morphometry of the three individuals of *A. tristis*. Primary contour feathers from the wings were the longest feather among all feathers in *A. tristis*. All three individuals of *A. tristis* had the same morphometric pattern, and of the three individuals, the one referred to as IND-2 had the longest and shortest feather morphometry. Primary contour feathers possess the longest rachis in (11.15±0.00 S.E. in cm), and bristle being the shortest in (0.73±0.05 S.E. in cm). Among all the considered characteristics of the feathers, barb lengths were the longest in primary contour feathers (1.88±0.12 S.E. in cm). In contrast, the shortest barbs were present in filoplume feathers (0.25±0.02 S.E. in cm). The present study showed three types of nodal structures, the elongated prongs in bristle and filoplume feathers being significant characteristics of *A. tristis*. The barbules of all feathers had nodes, swollen in shape like a bulb, with three different shapes viz. plain nodes, plain pronged nodes, and quadrilobed nodes. The presence of both sharply pointed and knobbed-shaped villi were also important micro-characteristics of A. tristis. Pennaceous barbs are present with distinct hooklets and ventral teeth. All the nodes and ramus of the plumulaceous barbs are dark with patchy pigmentation.

The colour of *S. chinensis* varied from dark brown to pale brown, creamy, with varying tinges of rusty, pale brown, and black. The common texture, ranging from firmly rigid to soft and fluffy, was observed in different feather types of *S. chinensis*. In *S. chinensis*, flight contour feathers (i.e., primary and secondary contour feathers) and tail feathers were firmly rigid in texture. Irrespective of their different body locations, body contour feathers were

semi-rigid in texture. On the other hand, the semiplume, down, and powder down feathers were soft and fluffy in texture in all four Columbidae species. The length of flight contour feathers from wings was the longest among the different feather types. Flight contour feathers (Primary & Secondary contour feathers) of *S. chinensis* were with the longest rachis (9.90±0.41 S.E. in cm), and down feathers were the shortest (2.25±0.07 S.E. in cm). The barbs of tail feathers were the longest (i.e., 1.84±0.12 S.E. in cm), and that of powder down feathers were the shortest (i.e., 0.89±0.04 S.E. in cm). Distinct microstructures (i.e., hooklets, cilia, and ventral teeth) were present in the pennaceous barbs. Three different node shapes were present in the plumulaceous barbules viz. very large, flattened, plate-like nodes (i.e., Crocus-shaped nodes), followed by many swollen but plain-pronged nodes that culminate in smaller, very minute and less conspicuous swollen nodes with or without prongs. All the nodes, internodes, and ramus of the barbs were present within both dark and patchy pigmentation.

The collected tissue samples from the single road-kill individual of *A. tristis* and *S. chinensis* were used to generate the complete mitogenome sequence (Chapter 5). DNA was isolated by the Phenol-Chloroform-Isoamyl Alcohol method, with minor modifications (Sambrook *et al.*, 1989; Dey *et al.*, 2021). The whole mitochondrial genomes were generated from the isolated DNA samples using the Next Generation Sequencing technique, and data were analyzed for various parameters. Estimation of the evolutionary divergences between the sequences was conducted using the Kimura 2-parameter model (Kimura, 1980), using MEGA X for the genus-level distances of the same genus members (Kumar *et al.*, 2018).

The complete sequence data of *A. tristis* was submitted to GenBank under the GenBank Accession number BankIt2510010 Common_myna_NAFLOK542103. Our study reported a 16822 base pair long mitogenome sequence of *A. tristis*. The total content of *A. tristis* was A: 4960 (29.5%), C: 5475 (32.5%), G: 2588 (15.4%), and T: 3799 (22.6%). The base composition of the sequence was dominated by higher A+T (53%) than G+C (47%) content. The skew values were AT-skew ((A-T)/(A+T)) 0.1325 and GC-skew ((G-C)/(G+C))-0.3581. Using mitogenomes from 7 families, 21 genera, and 30 species, the phylogenetic tree was constructed for *A. tristis*. The used mitogenome sequences were retrieved from NCBI GenBank, including 9 species from the Sturnidae family and the other 21 from allied families of the Passeriformes order. The phylogenetic tree reveals the clustered nature of the Sturnus genus in their respective clades. From the Sturnidae genera,

Acridotheres, *Sturnus*, *Gracula*, *Leucopsar*, and *Rhabdornis* clustered together. The phylogenetic tree reveals that select members of *Acridotheres* were phylogenetically close to *Sturnus cineraceus* and *Sturnus sericeus*. This study, for the first time, provides the complete mitogenome of *A. tristis* from the Indian subcontinent.

Our study reported a 16966 base pair long mitogenome sequence of *S. chinensis* encasing thirteen PCGs, twenty-two tRNAs, two rRNA, and with one mitochondrial control region (CR). The total content of *S. chinensis* was A: 5096 (30.0%), C: 5400 (31.8%), G: 2371 (14.0%), T: 4097 (24.1%), and 2 unresolved bases (1'N', 1'R'). The composition of nucleotides in *S. chinensis* mitogenome was estimated to be AT rich (68.1% and AT skew at 0.10) compared to GC content of 38.2% (GC skew at -0.38). The phylogenetic tree for the sequenced mitogenome of *S. chinensis* was prepared using 17 other mitogenomes from the Columbidae family. The retrieved phylogenetic topology shows the clustered species with high nodal values. In this study, the genus *Streptopelia* is considered similar to *Spelopelia* in the absence of a comprehensive Columbidae phylogeny. The phylogenetic tree reveals that the sequenced mitogenome of *S. chinensis* clades monophyletically with other *Streptopelia* species. All three *Spilopelia/Streptopelia* mitogenomes retrieved from NCBI GenBank clade together on a single node and are closest to *S. decaocto*, *S. orientalis*, and *S. tranquebarica*. In this study, only limited phylogenetic inference using the available data set could be drawn due to the absence of large numbers of mitogenomes from the Columbidae family. This is the first sequenced complete mitogenome of *S. chinensis* reported from India, specifically from the North-East India.

6.2. Conclusion

This study provides for the first time an insight into ongoing avian trade in Assam, so far not done systematically. Hence, this study's results will help map bird trade hot spots in Assam. The usage of traded avian species and other information related to avian trade in Assam can be used by the forest departments, conservationists, and law enforcement authorities to act against illegal wildlife crimes. The study of microscopic characteristics of feathers can assist in the identification of avian species in forensic investigations for the two selected traded avian species, i.e., *A. tristis* and *S. chinensis*, especially in cases when nothing remains but only feathers. Feather microstructures for different feathers from

different body locations were documented with selected parameters that can directly help in forensic investigations for any available feather fragments. The complete mitogenomes of *A. tristis* and *S. chinensis* generated from the study would serve as a reference and provide species-specific mitochondrial DNA markers to identify species in case of wildlife crime. This study offers a baseline for future studies on illegal avian trade and their forensic investigations.

References

References

1. Ahmed, A. (2013). 'Wildlife on sale': An insight into the Sonepur Mela, Bihar. *Traffic Post, 17*, 12–13.
2. Ahmed, A. (1997). *Live bird trade in northern India.* New Delhi: WWF Traffic-India.
3. Ahmed, A. (1999). *Fraudulence in Indian live bird trade: An identification monograph for control of illegal trade.* New Delhi: WWF Traffic-India & Government of India, Ministry of Environment and Forests.
4. Ahmed, A. (2002). *Live Bird Trade in India, Unpublished report.* New Delhi: WWF Traffic-India.
5. Ahmed, A. (2002, 31st July). "Bird trappers make good zoo keeper". Down to Earth.
6. Ahmed, A. (2010). *Imperilled custodians of the night: A study of the illegal trade, trapping, and utilization of Owls in India.* New Delhi: WWF Traffic-India.
7. Ahmed, A. (2013). The Trade, Trapping, and Utilization of Lorises in India. *Journal of the Bombay Natural History Society, 110*(3), 210–213.
8. Ahmed, A. (2014). Weaved in illegal wildlife trade, future for Bayas appears bleak. *Traffic Post, 21*, 18–19.
9. Aiyadurai, A. (2011). Wildlife hunting and conservation in Northeast India: a need for an interdisciplinary understanding. *International Journal of Galliformes Conservation, 2*, 61–73.
10. Aiyadurai, A. (2012). Bird hunting in Mishmi hills of Arunachal Pradesh, north-eastern India. *Indian Birds*, *7*(5), 134–137.
11. Aiyadurai, A., Singh, N. J., & Milner-Gulland, E. J. (2010). Wildlife hunting by indigenous tribes: a case study from Arunachal Pradesh, north-east India. *Oryx, 44*(4), 564–572.
12. Ali, S., & Ripley, S. D. (1987). *Handbook of the birds of India and Pakistan.* Vol. 5. Bombay: Oxford University Press.

13. Alström, P., Ericson, P. G., Olsson, U., & Sundberg, P. (2006). Phylogeny and classification of the avian superfamily Sylvioidea. *Molecular Phylogenetics and Evolution, 38*(2), 381–397. https://doi.org/10.1016/j.ympev.2005.05.015

14. Arctander, P. (1995). Comparison of a mitochondrial gene and a corresponding nuclear pseudogene. *Proceedings of the Royal Society of London. Series B: Biological Sciences, 262*(1363), 13–19. https://doi.org/10.1098/rspb.1995.0170

15. Atkinson, R., & Flint, J. (2001). Accessing hidden and hard-to-reach populations: Snowball research strategies. *Social Research Update, 33*(1), 1–4.

16. Baker, A. J., & Moeed, A. (1987). Rapid genetic differentiation and founder effect in colonizing populations of common mynas (*Acridotheres tristis*). *Evolution, 41*(3), 525–538.

17. Barker, F. K., Cibois, A., Schikler, P., Feinstein, J., & Cracraft, J. (2004). Phylogeny and diversification of the largest avian radiation. *Proceedings of the National Academy of Sciences, 101*(30), 11040–11045. https://doi.org/10.1073/pnas.040189210

18. Bezerra, D. M. M., Araújo, H. F., & Alves, R. R. N. (2019). Understanding the use of wild birds in a priority conservation area of Caatinga, a Brazilian tropical dry forest. *Environment, Development and Sustainability, 22*(6), 5297–5316. https://doi.org/10.1007/s10668-019-00425-1

19. Bhargava, R. (2000). A preliminary survey of the western population of Finn's Weaver in Kumaon terai, Uttar Pradesh, Northern India. *Oriental Bird Club Bulletin, 32*, 21–29.

20. Bhargava, R. (2017). *Status of Finn's Weaver in India: Past & Present.* Mumbai: Bombay Natural History Society.

21. Bhatt, J. P., & Pandit, M. K. (2019). Local hunting practices and wildlife conservation in Arunachal Pradesh, India. *Animal Conservation, 22*(6), 525–526.

22. Bhupathy, S., Kumar, S. R., Thirumalainathan, P., Paramanandham, J., & Lemba, C. (2013). Wildlife exploitation: A market survey in Nagaland, North-eastern India. *Tropical Conservation Science, 6*(2), 241–253.

23. Bhupathy, S., Srinivas, G., Kumar, N. S., Karthik, T., & Madhivanan, A. (2011). Herpetofaunal mortality due to vehicular traffic in the Western Ghats, India: a case study. *Herpetotropicos*, *5*(2), 119–126.

24. BirdLife International. (2016). *Spilopelia chinensis*. The IUCN red list of threatened species 2016: e.T60482887A95160992. https://dx.doi.org/10.2305/IUCN.UK.2016-3.RLTS.T60482887A95160992.en. [Accessed on 02nd December, 2022].

25. Brack, D. (2004). The growth and control of international environmental crime. *Environmental Health Perspectives*, *112*(2), A80–A81.

26. Broad, S., Mulliken, T., & Roe, D. (2014). The nature and extent of legal and illegal trade in wildlife. In S., Oldfield (Ed.), *The Trade in Wildlife: Regulation for Conservation* (pp. 25–44). London: Routledge.

27. Brom, T. G. (1986a). Identification of bird remains for bird strikes analysis: a literature synopsis. *Proceeding of the 18th Meeting Bird Strike Committee Europe, Copenhagen*, 255–261.

28. Brom, T. G. (1990). Villi and the phyly of Wetmore's order Piciformes (Aves). *Zoological journal of the Linnean Society, 98*(1), 63–72. https://doi.org/10.1111/j.1096-3642.1990.tb01219.x

29. Brom, T. G. (1991). *The diagnostic and phylogenetic significance of feather structures*. Doctoral dissertation. Instituut voor Taxonomische Zoölogie, Universiteit van Amsterdam, Amsterdam.

30. Brom, T. G., & Frank, U. (1992). The diagnostic and phylogenetic significance of widened and pronged hamuli in feathers. *Scanning Microscopy, 6*(2), 597–602.

31. Brown, W. M., George Jr., M., & Wilson, A. C. (1979). Rapid evolution of animal mitochondrial DNA. *Proceedings of the National Academy of Sciences, 76*(4), 1967–1971. https://doi.org/10.1073/pnas.76.4.1967

32. Bušina, T., Kouba, M., & Pasaribu, N. (2021). What is the reliability of visually based animal trade census outcomes? A case study involving the market monitoring of the Sumatran laughingthrush *Garrulax bicolor*. *Bird Conservation International*, *31*(2), 326–336. DOI:10.1017/S095927092000026X

33. Butchart, S. H. (2008). Red List Indices to measure the sustainability of species use and impacts of invasive alien species. *Bird Conservation International, 18*(S1), S245–S262. https://doi.org/10.1017/S095927090800035X

34. Cai, T., Cibois, A., Alström, P., Moyle, R. G., Kennedy, J. D., Shao, S., Zhang, R., Irestedt, M., Ericson, PGP., Gelang, M., Qu, Y. H., Lei, F. M., Fjeldså, J. (2019). Near-complete phylogeny and taxonomic revision of the world's babblers (Aves: Passeriformes). *Molecular Phylogenetics and Evolution, 130*, 346–356. https://doi.org/10.1016/j.ympev.2018.10.010

35. Campbell, V., & Lapointe, F. J. (2011). Retrieving a mitogenomic mammal tree using composite taxa. *Molecular Phylogenetics and Evolution, 58*(2), 149–156. https://doi.org/10.1016/j.ympev.2010.11.017

36. Cardoso, P., Amposah-Mensah, K., Barreiros, J.P., Bouhuys, J., Cheung, H., Davies, A., Kumschick, S., Longhorn, J. S., Martínez-Munoz, A. C., Morcatty, Q. T., Petersl, G., Ripple, J. W., Rivera-T´ellez, E., Stringham, C. O., Toomes, A., Tricorache, P., & Fukushima, S. C. (2021). Scientists' warning to humanity on illegal or unsustainable wildlife trade. *Biological Conservation, 263*, 109341. https://doi.org/10.1016/j.biocon.2021.109341.

37. Ceballos, G., Ehrlich, P. R., & Dirzo, R. (2017). Biological annihilation via the ongoing sixth mass extinction signaled by vertebrate population losses and declines. *Proceedings of the National Academy of Sciences, 114*(30), E6089–E6096. http://www.pnas.org/cgi/doi/10.1073/pnas.1704949114

38. Chandler, A. C. (1916). *A study of the structure of feathers, with reference to their taxonomic significance*. California: University of California Press.

39. Chen, Y. X., Sun, C. H., Li, Y. K., Fei, Y. L., Xue, X. M., Hou, S. L., Zhou, Y. W., Jiang, J., & Guo, H. T. (2020). Complete mitogenome of *Treron sphenurus* (Aves, Columbiformes): the first representative from the genus *Treron*, genomic comparisons and phylogenetic analysis of Columbidae. *Animal Biotechnology, 33*(6), 1–11. https://doi.org/10.1080/10495398.2020.1862135

40. Chhangani, A. K. (2004). Frequency of avian road-kills in Kumbhalgarh Wildlife Sanctuary, Rajasthan, India. *Forktail, 20*, 110–111.

41. Choudhury, K., & Ghosh, S. (2008). Mortality of butterfly fauna due to vehicular traffic and their conservation in Ripu-Chirang Reserve Forests of western Assam, India. *2nd Asian Lepidoptera Conservation Symposium 2008, Penang, Malaysia*, 1–16. https://www.researchgate.net/publication/330345758.

42. Chutia, P., & Solanki, G. S. (2013). Patterns of bird hunting in Arunachal Pradesh and implications for biodiversity conservation. *Tropical Ecology*, *54*(2), 263–267.

43. Cibois, A., & Cracraft, J. (2004). Assessing the passerine "Tapestry": phylogenetic relationships of the Muscicapoidea inferred from nuclear DNA sequences. *Molecular Phylogenetics and Evolution, 32*(1), 264–273. https://doi.org/10.1016/j.ympev.2003.12.002

44. Clark, A. G., & Whittam, T. S. (1992). Sequencing errors and molecular evolutionary analysis. *Molecular Biology and Evolution, 9*(4), 744–752. https://doi.org/10.1093/oxfordjournals.molbev.a040756

45. Clark, H. L., & Allen, J. A. (1893). A Neglected Branch of Ornithology. *Auk, 10*(1): 93–95. https://doi.org/10.2307/4067928

46. Compton, J., Khounboline, K., & Quang, Le. H. (1999). *Vanishing point: an investigation into cross-border wildlife trade between Laos and Vietnam.* Hanoi: WWF Indochina Programme.

47. Das, A., Ahmed, F. M., Lahkar, B. P., & Sharma, P. (2007). A preliminary report of reptilian mortality on road due to vehicular movement near Kaziranga National Park, Assam, India. *Zoos' Print, 22* (7): 2742–2744. DOI: 10.11609/JoTT.ZPJ.1541.2742-4

48. Datta, A., Anand, M. O., & Naniwadekar, R. (2008). Empty forests: Large carnivore and prey abundance in Namdapha National Park, north-east India. *Biological Conservation, 141*(5), 1429–1435.

49. Davies, A. (1970). Micromorphology of feathers using the scanning electron microscope. *Journal of the Forensic Science Society, 10*(3), 165–174. https://doi.org/10.1016/S0015-7368(70)70591-4

50. Day, M. G. (1966). Identification of hair and feather remains in the gut and faeces of stoats and weasels. *Journal of Zoology, 148*(2), 201–217. https://doi.org/10.1111/j.1469-7998.1966.tb02948.x

51. de Oliveira, W. S. L., de Faria Lopes, S., & Alves, R. R. N. (2018). Understanding the motivations for keeping wild birds in the semi-arid region of Brazil. *Journal of Ethnobiology and Ethnomedicine, 14*(1), 1–14. https://doi.org/10.1186/s13002-018-0243-6

52. Deedrick, D. W., & Mullery, J. P. (1981). Feathers are not lightweight evidence. *FBI Law Enforcement Bulletin, 50*(9), 22–23.

53. Desjardins, P., & Morais, R. (1990). Sequence and gene organization of the chicken mitochondrial genome: A novel gene order in higher vertebrates. *Journal of Molecular Biology, 212*(4), 599–634. https://doi.org/10.1016/0022-2836(90)90225-B

54. Destro, G. F. G., Pimentel, T. L., Sabaini, R. M., Borges, R. C., & Barreto, R. (2012). Efforts to combat wild animals trafficking in Brazil. In G. A. Lameed (Ed.) *Biodiversity enrichment in a diverse world* (pp. 421–436). New York: IntechOpen. https://doi.org/10.5772/3088

55. Develey, P. F., & Stouffer, P. C. (2001). Effects of roads on movements by understory birds in mixed-species flocks in central Amazonian Brazil. *Conservation Biology, 15*(5), 1416-1422. https://doi.org/10.1111/j.1523-1739.2001.00170.x

56. Devi, O. S. (2019). *Tradable Bioresources of Assam*. Guwahati: Assam State Biodiversity Board.

57. Dey, P., Ray, S. D., Sharma, S. K., Pramod, P., & Singh, R. P. (2021). Identification of a unique barb from the dorsal body contour feathers of the Indian Pitta *Pitta brachyura* (Aves: Passeriformes: Pittidae). *Journal of Threatened Taxa, 13*(8), 19029–19039. https://doi.org/10.11609/jott.6362.13.8.19029-19039

58. Dey, P., Sharma, S. K., Sarkar, I., Ray, S. D., Pramod, P., Kochiganti, V. H. S., Quadros, G., Rathore, S. S., & Singh, R. P. (2021). Complete mitogenome of endemic plum-headed parakeet *Psittacula cyanocephala*–characterization and phylogenetic analysis. *PLoS ONE, 16*(4), e0241098. https://doi.org/10.1371/journal.pone.0241098

59. Dhindsa, M. S., Sandhu, J. S., Sandhu, P. S., & Toor, H. S. (1988). Roadside birds in Punjab (India): relation to mortality from vehicles. *Environmental Conservation, 15*(4), 303–310. https://doi.org/10.1017/S0376892900029799

60. Donath, A., Jühling, F., Al-Arab, M., Bernhart, S. H., Reinhardt, F., Stadler, P. F., Middendorf, M., & Bernt, M. (2019). Improved annotation of protein-coding genes boundaries in metazoan mitochondrial genomes. *Nucleic Acids Research, 47*(20), 10543–10552. https://doi.org/10.1093/nar/gkz833

61. Dove, C. J. (1997). Quantification of microscopic feather characters used in the identification of North American plovers. *The Condor, 99*(1), 47–57. https://doi.org/10.2307/1370223

62. Dove, C. J. (1998a). *Microscopic variation in downy feathers characters of Charadriiformes (Aves): A descriptive and phylogenetic analysis*. Doctoral dissertation. George Mason University, Virginia.

63. Dove, C. J. (1998b). Feather evidence helps clarify locality of anthropological artifacts in the Museum of Mankind. *Pacific Studies, 21*(3), 73–84.

64. Dove, C. J. (2000a). A descriptive and phylogenetic analysis of plumulaceous feather characters in Charadriiformes. *Ornithological Monographs, 51*, 1–163. https://doi.org/10.2307/40166844

65. Dove, C. J. (2000b). *Forensic Ornithology*. Washington: Smithsonian Institute.

66. Dove, C. J., & Agreda, A. (2007). Differences in plumulaceous feather characters of dabbling and diving ducks. *The Condor, 109*(1), 192–199. https://doi.org/10.1093/ condor/109.1.192

67. Dove, C. J., & Coddington, C. P. (2015). Forensic techniques identify the first record of Snowy Owl (*Bubo scandiacus*) feeding on a Razorbill (*Alca torda*). *The Wilson Journal of Ornithology, 127*(3), 503–506. https://doi.org/10.1676/14-176.1

68. Dove, C. J., & Koch, S. L. (2011). Microscopy of feathers: A practical guide for forensic feather identification. *The Microscope, 59*(2), 51–71.

69. Dove, C. J., & Peurach, S. C. (2002). Microscopic Analysis of Feather and Hair Fragments Associated with Human Mummified Remains from Kagamil Island, Alaska. In B. Frohlich, R. Gilberg, & A. B. Harper (Eds.), *To the Aleutians and Beyond: The Anthropology of William S. Laughlin* (pp. 51–62). Copenhagen: Department of Ethnography, The National Museum of Denmark.

70. Dove, C. J., & Wickler, S. (2016). Identification of bird species used to make a Viking Age feather pillow. *Arctic, 69*(1), 29–36.

71. Dove, C. J., Dahlan, N. F., & Heacker, M. (2009). Forensic bird-strike identification techniques used in an accident investigation at Wiley Post Airport, Oklahoma, 2008. *Human-Wildlife Conflicts. 3*(2), 179–185. https://doi.org/10.26077/ace5-s883

72. Dove, C. J., Hare, P. G., & Heacker, M. (2005). Identification of ancient feather fragments found in melting alpine ice patches in southern Yukon. *Arctic, 58*(1), 38–43. https://doi.org/10.14430/arctic387

73. Duchene, S., Frey, A., Alfaro-Núñez, A., Dutton, P. H., Gilbert, M. T. P., & Morin, P. A. (2012). Marine turtle mitogenome phylogenetics and evolution. *Molecular Phylogenetics and Evolution, 65*(1), 241–250. https://doi.org/10.1016/j.ympev. 2012.06.010

74. Ericson, P. G., & Johansson, U. S. (2003). Phylogeny of Passerida (Aves: Passeriformes) based on nuclear and mitochondrial sequence data. *Molecular Phylogenetics and Evolution, 29*(1), 126–138. https://doi.org/10.1016/S1055-7903(03)00067-8

75. Esmail, N., Wintle, B. C., Sas-Rolfes, M., Athanas, A., Beale, C.M., Bending, Z., Dai, R., Fabinyi, M., Gluszek, S., Haenlein, C., Harrington, A, L., Hinsley, A., Kariuki, K., Lam, J., Markus, M., Paude, K., Shukhova, S., Sutherland, J. W., Verissimo, D., Wang, Y., & Waugh, J. (2020). Emerging illegal wildlife trade issues: A global horizon scan. *Conservation Letter, 13*(4), e12715. https://doi.org/10.1111/conl.12715

76. Food and Agriculture Organization of the United Nations [FAO.] (2011). International trade in wild birds, and related bird movements, in Latin America and the Caribbean. *Animal Production and Health Paper*, 166. http://www.fao.org/3/ ai0708e.pdf

77. Feare, C., & Craig, A. (1999). *Starlings and Mynas*. New Jersey: Princeton University Press.

78. Fleischer, R. C., Williams, R. N., & Baker, A. J. (1991). Genetic variation within and among populations of the common myna (*Acridotheres tristis*) in Hawaii. *Journal of Heredity, 82*(3), 205–208. https://doi.org/10.1093/ oxfordjournals. jhered.a111066

79. Forest Survey of India. (2021). *India State of Forest Report*. Dehradun: Ministry of Environment Forest and Climate Change.

80. Forman, R. T., Reineking, B., & Hersperger, A. M. (2002). Road traffic and nearby grassland bird patterns in a suburbanizing landscape. *Environmental Management. 29*(6), 782–800. https://doi.org/10.1007/s00267-001-0065-4

81. Fukushima, C. S., Tricorache, P., Toomes, A., Stringham, O. C., Rivera-Téllez, E., Ripple, W. J., Peters, G., Orenstein, R. I., Morcatty, T. Q., Longhorn, S. J., Lee, C., Kumschick, S., de Freitas, M. A., Duffy, R. V., Davies, A., Cheung, H., Cheyne, S. M., Bouhuys, J., Barreiros, J. P., Amponsah-Mensah, K., & Cardoso, P. (2021).

Challenges and perspectives on tackling illegal or unsustainable wildlife trade. *Biological Conservation, 263*, 109342. https://doi.org/10.1016/j.biocon. 2021. 109342

82. Fuller, M. E. (2015). *The structure and properties of down feathers and their use in the outdoor industry*. Doctoral dissertation. University of Leeds, Leeds.

83. Gadow, D. H. (1882). On the Colour of Feathers as affected by their Structure. *Proceedings of the Zoological Society of London 50*(3), 409–422.

84. Gamborg, C., Parsons, R., Puri, R. K., & Sandøe, P. (2012). Ethics and research methodologies for the study of traditional forest-related knowledge. In J. A. Parrotta, & R. L. Trosper (Eds.), *Traditional Forest-Related Knowledge* (Vol. 12, pp. 535–562). Dordrecht: Springer. https://doi.org/10.1007/978-94-007-2144-9_14

85. Garnier, F. (1996). *The Mekong Exploration Commission Report, 1866–1868: Travels in Cambodia and part of Laos* (Vol. 1). Cambodia: White Lotus Press.

86. Garrett, K. L., & Walker, R. L. (2022). Spotted Dove (*Spilopelia chinensis*), version 1.1. In Birds of the World (N. D. Sly, Ed.). Cornell Lab of Ornithology, Ithaca, NY, USA. https://doi.org/10.2173/bow.spodov.01.1 [Accessed on 02nd December, 2022].

87. Gilardi, J. D. (2006). Captured for conservation: will cages save wild birds? A response to Cooney & Jepson. *Oryx, 40*(1), 24–26. https://doi.org/10.1017/ S0030605306000160

88. Gilbert, F. F., & Nancekivell, E. G. (1982). Food habits of mink (*Mustela vison*) and otter (*Lutra canadensis*) in northeastern Alberta. *Canadian Journal of Zoology, 60*(6), 1282–1288. https://doi.org/10.1139/z82-172

89. Gill, F. B. (1995). *Ornithology*. New York: Macmillan publishers.

90. Government of Assam. (2021a). Assam at a Glance 2021. Guwahati: Directorate of Economics and Statistics, Government of Assam.

91. Government of Assam. (2021b). Statistical Handbook Assam, 2021. Guwahati: Directorate of Economics and Statistics, Government of Assam.

92. Grant, J. R., & Stothard, P. (2008). The CGView Server: a comparative genomics tool for circular genomes. *Nucleic Acids Research, 36*(suppl_2), W181–W184. https://doi.org/10.1093/nar/gkn179

93. Grieser-Johns, A., & Thomson, J. (2005). *Going, going, gone: the illegal trade in wildlife in East and Southeast Asia.* Washington: World Bank.

94. Grimmett, R., Inskipp, C., & Inskipp, T. (2016). *Birds of the Indian Subcontinent: India, Pakistan, Sri Lanka, Nepal, Bhutan, Bangladesh and the Maldives*. London: Bloomsbury Publishing.

95. Guhathakurta, P., Bandgar, A., Menon, P., Prasad, A. K., Sangwan, N., & Advani, S. C. (2020). *Observed Rainfall Variability and Changes over Assam State*. Pune: India Meteorological Department.

96. Gupta, H. S. (2004). Waterbirds diversity of Ranchi district. *Zoos' Print Journal*, *19*(9), 1630.

97. Harris, J. B. C., Tingley, M. W., Hua, F., Yong, D. L., Adeney, J. M., Lee, T. M., Marthy, W., Prawiradilaga, D. M., Sekercioglu, C. H., Suyadi., Winarni, N., & Wilcove, D. S. (2017). Measuring the Impact of the Pet Trade on Indonesian Birds. *Conservation Biology, 31*(2), 394–405. https://doi.org/10.1111/cobi.12729

98. Harris, M. P., Williamson, S., Fallon, J. F., Meinhardt, H., & Prum, R. O. (2005). Molecular evidence for an activator–inhibitor mechanism in development of embryonic feather branching. *Proceedings of the National Academy of Sciences, 102*(33), 11734–11739. https://doi.org/10.1073/pnas.050078110

99. Hassanin, A., Leger, N., & Deutsch, J. (2005). Evidence for multiple reversals of asymmetric mutational constraints during the evolution of the mitochondrial genome of Metazoa, and consequences for phylogenetic inferences. *Systematic Biology, 54*(2), 277–298. https://doi.org/10.1080/10635150590947843

100. Helldin, J. O., & Seiler, A. (2003). Effects of roads on the abundance of birds in Swedish forest and farmland. *Habitat Fragmentation due to Transportation Infrastructure – IENE 2003*, 1–9.

101. Hilaluddin, Kaul, R., & Ghosh, D. (2005). Extraction and use of Galliformes by indigenous ethnic groups in Northeast India. *Proceedings of the 3rd International Galliformes Symposium, World Pheasant Association*, 220–225.

102. Hodson, N. L. (1960). A survey of vertebrate road mortality 1959. *Bird Study, 7*(4), 224–231. https://doi.org/10.1080/00063656009475974

103. Hodson, N. L. (1962). Some notes on the causes of bird road casualties. *Bird Study, 9*(3), 168–173. https://doi.org/10.1080/00063656209476024

104. Huang, Z., Tu, F., & Liu, X. (2015). Determination of the complete mitogenome of Spotted Dove, *Spilopelia chinensis* (Columbiformes: Columbidae). *Mitochondrial DNA Part A, 27*(6), 4224–4225. https://doi.org/10.3109/19401736.2015.1022750

105. Imchen, A., & Joglekar, P. P. (2015). Traditional Animal Hunting Practices among the Ao Nagas: A Case Study of Mangmetong Village, Nagaland. *Heritage: Journal of Multidisciplinary Studies in Archaeology*, *3*, 507–525.

106. Islam, K. & Williams, R. N. (2020). Red-vented Bulbul (*Pycnonotus cafer*), version 1.0. In Birds of the World (S. M. Billerman, Ed.). Cornell Lab of Ornithology, Ithaca, NY, USA. https://doi.org/10.2173/bow.revbul.01 [Accessed on 02nd December, 2022]

107. Islam, M. Z., & Rahmani, A. R. (2004). *Important Bird Areas in India: priority sites for conservation.* Mumbai: Indian Bird Conservation Network, Bombay Natural History Society and Birdlife International (UK).

108. Islam, M., & Saikia, P. K. (2014). A study on road-kill Herpetofauna in Jaypore Reserve Forest, Assam. *NeBIO, 5*(1), 78–83.

109. IUCN Red List (2021). Summary statistics. Threatened species in each major taxonomic group by country. https://www.iucnredlist.org/statistics. [Accessed on 02nd June 2021].

110. Jeganathan, P., Mudappa, D., Kumar, A., & Raman, T. S. (2018). Seasonal variation in wildlife roadkills in plantations and tropical rainforest in the Anamalai Hills, Western Ghats, India. *Current Science, 114*(3), 619–626. DOI: 10.18520/cs/v114/i03/619-626

111. Jepson, P. (2010). *Towards an Indonesian bird conservation ethos: Reflections from a study of bird-keeping in the cites of Java and Bali.* In S. Tidemann & A. Gosler (Eds.), Ethno-ornithology: Birds, indigenous peoples, culture and society (pp. 313–330). London: Routledge.

112. Jiang, J. (2019). Analysis of microstructure and difference of 6 species of remiges in Galliformes. E3S Web of Conferences, 131, 01130. https://doi.org/10.1051/e3sconf/ 201913101130

113. Johnson, K. P., de Kort, S., Dinwoodey, K., Mateman, A. C., ten Cate, C., Lessells, C. M., & Clayton, D. H. (2001). A molecular phylogeny of the Dove genera *Streptopelia* and *Columba*. *The Auk, 118*(4), 874–887. https://doi.org/10.1093/auk/118.4.874

114. Johnson, P. T. (2005). Snowball Sampling. In P. Armitage & T. Colton (Eds.). *Encyclopedia of Biostatistics*, 2nd ed. (pp. 1–2). New Jersey: John Wiley and Sons.

115. Kalyaanamoorthy, S., Minh, B. Q., Wong, T. K., Von Haeseler, A., & Jermiin, L. S. (2017). ModelFinder: fast model selection for accurate phylogenetic estimates. *Nature Methods, 14*(6), 587–589. https://doi.org/10.1038/nmeth.4285

116. Kan, X. Z., Li, X. F., Zhang, L. Q., Chen, L., Qian, C. J., Zhang, X. W., & Wang, L. (2010). Characterization of the complete mitochondrial genome of the Rock pigeon, *Columba livia* (Columbiformes: Columbidae). *Genetics and Molecular Research, 9*(2), 1234–1249.

117. Kang, C. L., Chan, W. K., Feare, C. J., Wells, D., Kang, N., & Kang, N. (1994). The relationship between mynas: an application of DNA sequencing. *Journal of Ornithology, 135*, 33.

118. Kannan, R. & James, D. A. (2020). Common Myna (*Acridotheres tristis*), version 1.0. In Birds of the World (S. M. Billerman, Ed.). Cornell Lab of Ornithology, Ithaca, NY, USA. https://doi.org/10.2173/bow.commyn.01 [Accessed on 20th March, 2022].

119. Ko, A. M. S., Zhang, Y., Yang, M. A., Hu, Y., Cao, P., Feng, X., Zhang, L., Wei, F., & Fu, Q. (2018). Mitochondrial genome of a 22,000-year-old giant panda from southern China reveals a new panda lineage. *Current Biology, 28*(12), R693–R694. https://doi.org/10.1016/j.cub.2018.05.008

120. Kocher, T. D., Thomas, W. K., Meyer, A., Edwards, S. V., Pääbo, S., Villablanca, F. X., & Wilson, A. C. (1989). Dynamics of mitochondrial DNA evolution in animals: amplification and sequencing with conserved primers. *Proceedings of the National Academy of Sciences, 86*(16), 6196–6200. https://doi.org/10.1073/pnas.86.16.6196

121. Krishnan, A. (2022). *Why is India a major hub for wildlife trafficking?* Mongabay, Mongabay Series: Beyond Protected Areas, Environment Explained. https://india.mongabay.com/2022/06/explainer-why-is-india-a-major-hub-for-wildlife-trafficking/ [Accessed on 15th November 2022].

122. Krishnasamy, K. & Zavagli, M. (2020). *Southeast Asia: At the heart of wildlife trade.* Malaysia: Traffic.

123. Kroll, G. (2015). An environmental history of road kill: Road ecology and the making of the permeable highway. *Environmental History, 20*(1), 4–28. https://doi.org/10.1093/envhis/emu129

124. Kumara, H. N., & Singh, M. (2004). The influence of differing hunting practices on the relative abundance of mammals in two rainforest areas of the Western Ghats, India. *Oryx, 38*(3), 321–327.

125. Kumara, H. N., Sharma, A. K., Kumar, A., & Singh, M. (2000). Roadkills of wild fauna in Indira Gandhi wildlife sanctuary, Western Ghats, India: implications for management. *Biosphere Conservation: for nature, wildlife, and humans, 3*(1), 41–47. https://doi.org/10.20798/biospherecons.3.1_41

126. Langmead, B., & Salzberg, S. L. (2012). Fast gapped-read alignment with Bowtie 2. *Nature Methods, 9*(4), 357–359.

127. Lavorgna, A., Middleton, S. E., Pickering, B., & Neumann, G. (2020). Flora Guard: Tackling the online illegal trade in endangered plants through a cross-disciplinary ICT-enabled methodology. *Journal of Contemporary Criminal Justice, 36*(3), 428–450. https://doi.org/10.1177/1043986220910297

128. Laybourne, R. C. (1974). Collision between a vulture and an aircraft at an altitude of 37,000 feet. *Wilson Bulletin, 86*(4), 461–462.

129. Laybourne, R. C. (1984). Identification of bird remains from bird-aircraft incidents by the microstructure of the downy part of the feather. *Proceeding of the 17th meeting Bird Strike Committee Europe, (Rome)*, 282–286.

130. Laybourne, R. C. (1986). The variation of the nodal structures on downy barbules of some species of birds. *Proceeding of the 18th meeting Bird Strike Committee Europe, Copenhagen*. 24.

131. Laybourne, R. C., & Dove, C. J. (1994). Preparation of birdstrike remains for identification. *Proceedings and working papers of the 22nd Bird Strike Committee Meeting Europe, Vienna, 22*, 531–534.

132. Laybourne, R. C., Sabo, B. A., & Morningstar, A. (1992). Basic technique for preparation of down for examination with the scanning electron microscope. *The Auk, 109*(1), 195–197. https://doi.org/10.2307/4088284

133. Lee, J. G. H., Chng, S. C. L., & Eaton, J. A. (2016). *Conservation Strategy for Southeast Asian Songbirds in Trade. Recommendations from the First Asian Songbird Trade Crisis Summit 2015*. Singapore: Wildlife Reserves Singapore & TRAFFIC,. https://doi.org/10.13140/RG.2.2.12805.96483

134. Lee, J., Sarre, S. D., Joseph, L., & Robertson, J. (2015). Microscopic characteristics of the plumulaceous feathers of Australian birds: a preliminary analysis of taxonomic discrimination for forensic purposes. *Australian Journal of Forensic Sciences, 48*(4), 421–444. http://dx.doi.org/10.1080/00450618.2015.1076034

135. Lei, F. M., Qu, Y. H., Gan, Y. L., Gebauer, A., & Kaiser, M. (2002). The feather microstructure of Passerine sparrows in China. *Journal für Ornithologie, 143*(2), 205–213. https://doi.org/10.1007/BF02465449

136. Leupen, B. T., Shepherd, L., Shepherd, C. R., Damianou, E., & Nijman, V. (2022). Market surveys in Mataram, Lombok, illustrate the expanse of legal and illegal Indonesian bird trade networks. *Indonesian Journal of Applied Environmental Studies, 3*(1), 42–52. DOI: 10.33751/injast.v3i1.5127

137. Lewis, R., Deshpande, K., Mendis, A., Patankar, V., Mendiratta, U. (2022). *Illegal trade of marine species in India: 2015-2021.* Karnataka: Wildlife Conservation Society-India. https://doi.org/10.19121/2022.Report.43707

138. Li, C. H., Shi, W., & Shi, W. Y. (2014). Mitochondrial genome sequence of Egyptian swift Rock Pigeon (*Columba livia* breed Egyptian swift). *Mitochondrial DNA, 26*(3), 479–480. https://doi.org/10.3109/19401736.2013.873931

139. Li, X., Lin, L., Cui, A., Bai, J., Wang, X., Xin, C., Zhang, Z., Yang, C., Gao, R. R., Huang, Y., & Lei, F. (2016). Taxonomic status and phylogenetic relationship of tits based on mitogenomes and nuclear segments. *Molecular Phylogenetics and Evolution, 104*, 14–20. https://doi.org/10.1016/j.ympev.2016.07.022

140. Lin, J. (2005). Tackling Southeast Asia's illegal wildlife trade. *Sybil, 9*, 191.

141. Lipske, M. (1982). To down detective Roxie Laybourne, a feather's the clue. *Smithsonian, 12*(12), 98–105.

142. Liu, G., Zhou L, Li B, Zhang L. 2014. The complete mitochondrial genome of *Aix galericulata* and *Tadorna ferruginea*: Bearings on their phylogenetic position in the Anseriformes. *PLoS ONE, 9*(11):e109701. https://doi.org/10.1371/ journal. pone.0109701

143. Livezey, B. C., & Zusi, R. L. (2007). Higher-order phylogeny of modern birds (Theropoda, Aves: Neornithes) based on comparative anatomy. II. Analysis and discussion. *Zoological Journal of the Linnean Society, 149*(1), 1–95. https://doi.org/10.1111/j.1096-3642.2006.00293.x

144. Lovette, I. J. & Fitzpatrick, J. W. (Eds.). (2016). *The Cornell Lab of Ornithology's Handbook of Bird Biology*. West Sussex: John Wiley & Sons.

145. Lovette, I. J., & Rubenstein, D. R. (2007). A comprehensive molecular phylogeny of the starlings (Aves: Sturnidae) and mockingbirds (Aves: Mimidae): congruent mtDNA and nuclear trees for a cosmopolitan avian radiation. *Molecular Phylogenetics and Evolution, 44*(3), 1031–1056. https://doi.org/10.1016/j.ympev.2007.03.017

146. Lovette, I. J., McCleery, B. V., Talaba, A. L., & Rubenstein, D. R. (2008). A complete species-level molecular phylogeny for the "Eurasian" starlings (Sturnidae: *Sturnus*, *Acridotheres*, and allies): Recent diversification in a highly social and dispersive avian group. *Molecular Phylogenetics and Evolution, 47*(1), 251–260. https://doi.org/10.1016/j.ympev.2008.01.020

147. Lowe S., Browne M., Boudjelas S., De Poorter M. (2000). *100 of the World's Worst Invasive Alien Species A selection from the Global Invasive Species Database.* Auckland: The Invasive Species Specialist Group (ISSG) a specialist group of the Species Survival Commission (SSC) of the World Conservation Union (IUCN). www.issg.org/booklet.pdf

148. Lowe, T. M., & Chan, P. P. (2016). tRNAscan-SE On-line: integrating search and context for analysis of transfer RNA genes. *Nucleic Acids Research, 44*(W1), W54–W57. https://doi.org/10.1093/nar/gkw413

149. Lucas, A. M., & Stettenheim, P. R. (1972). *Avian Anatomy-Integument. Agriculture Handbook 362*. Washington: U.S. Department of Agriculture.

150. Mackiewicz, P., Urantówka, A. D., Kroczak, A., & Mackiewicz, D. (2019). Resolving phylogenetic relationships within Passeriformes based on mitochondrial genes and inferring the evolution of their mitogenomes in terms of duplications. *Genome Biology and Evolution, 11*(10), 2824–2849. https://doi.org/10.1093/gbe/evz209

151. Madge, S. (2020). House Crow (*Corvus splendens*), version 1.0. In Birds of the World (J. del Hoyo, A. Elliott, J. Sargatal, D. A. Christie, & E. de Juana, Eds.). Cornell Lab of Ornithology, Ithaca, NY, USA. https://doi.org/10.2173/bow.houcro1.01 [Accessed on 22nd November, 2022].

152. Mahananda, P., & Jelil, S. N. (2017). Small Indian Civet-A report on a road kill of Viverricula indica from Guwahati, Assam. *Zoo's Print, 32*(1), 26–28. DOI: 10.13140/RG.2.2.12628.19845

153. Majumdar, A., Gopinathan, M., Alam, I., Chandra, K., Alfred, J. R. B., & Chowdhury, B. R. (2022). *Field Guide Birds of India*. Kolkata: Zoological Survey of India.

154. Marshall, H., Collar, N. J., Lees, A. C., Moss, A., Yuda, P., & Marsden, S. J. (2020a). Spatio-temporal dynamics of consumer demand driving the Asian Songbird Crisis. *Biological Conservation, 241*, 108237. https://doi.org/10.1016/j.biocon. 2019.108237

155. Marshall, H., Collar, N. J., Lees, A. C., Moss, A., Yuda, P., & Marsden, S. J. (2020b). Characterizing bird-keeping user-groups on Java reveals distinct behaviours, profiles and potential for change. *People and Nature, 2*(4), 877–888. https://doi.org/10.1002/pan3.10132

156. Martin, E. B., & Phipps, M. (1996). A review of the wild animal trade in Cambodia. *TRAFFIC Bulletin-Wildlife Trade Monitoring Unit, 16*(2), 45–60.

157. May, T. (Ed.). (2002). *Qualitative research in action*. London: SAGE Publications.

158. Mazumdar, K., Samal, P. K., & Gupta, A. (2014). Hunting of avifauna in proposed Tsangyang Gyatso Biosphere Reserve, Western Arunachal Pradesh. *Zoo's Print, 19*(4) 3–7.

159. McClenachan, L., Cooper, A. B., & Dulvy, N. K. (2016). Rethinking trade-driven extinction risk in marine and terrestrial megafauna. *Current Biology, 26*(12), 1640–1646. http://dx.doi.org/10.1016/j.cub.2016.05.026

160. McNamara, J., Robinson, E. J., Abernethy, K., Midoko Iponga, D., Sackey, H. N., Wright, J. H., & Milner-Gulland, E. J. (2020). COVID-19, systemic crisis, and possible implications for the wild meat trade in Sub-Saharan Africa. *Environmental and Resource Economics, 76*(4), 1045–1066. https://doi.org/10.1007/s10640-020-00474-5

161. McNeely, J. A., Kapoor-Vijay, P., Zhi, L., Olsvig-Whittaker, L., Sheikh, K. M., & Smith, A. T. (2009). Conservation biology in Asia: the major policy challenges. *Conservation Biology*, *23*(4), 805–810. https://doi.org/10.1111/j.1523-1739.2009.01284.x

162. Menon, V., (1996). Impact of trade on Himalayan biodiversity. In G. S. Gujral & V. Sharma (Eds.), *Changing perspectives of biodiversity status in the Himalaya.* New Delhi: British High Commission, British Council Division.

163. Messinger, N. G. (1965). Methods Used for Identification of Feather Remains from Wetherill Mesa. *Memoirs of the Society for American Archaeology, 19*, 206–215. https://doi.org/10.1017/S0081130000004585

164. Meyer, A., & Zardoya, R. (2003). Recent advances in the (molecular) phylogeny of vertebrates. *Annual Review of Ecology, Evolution, and Systematics, 34*, 311–338.

165. Mills, J. A., & Servheen, C. (1990). *The Asian trade in bears and bear parts.* Washington: World Wildlife Fund and Traffic USA.

166. Mills, J., & Servheen, C. (1994). The Asian trade in bears and bear parts: impacts and conservation recommendations. *Bears: Their Biology and Management, 9*, 161–167. https://doi.org/10.2307/3872697

167. Mindell, D. P., Sorenson, M. D., & Dimcheff, D. E. (1998). An extra nucleotide is not translated in mitochondrial ND3 of some birds and turtles. *Molecular Biology and Evolution, 15*(11), 1568–1571. https://doi.org/10.1093/oxfordjournals.molbev.a025884

168. Morcatty, T. Q., Feddema, K., Nekaris, K. A. I., & Nijman, V. (2021). Online trade in wildlife and the lack of response to COVID-19. *Environmental Research, 193*, 110439. https://doi.org/10.1016/j.envres.2020.110439.

169. Morinha, F., Clemente, C., Cabral, J. A., Lewicka, M. M., Travassos, P., Carvalho, D., Dávila, J. A., Santos, M., Blanco, G., & Bastos, E. (2014). Next-generation sequencing and comparative analysis of *Pyrrhocorax pyrrhocorax* and *Pyrrhocorax graculus* (Passeriformes: Corvidae) mitochondrial genomes. *Mitochondrial DNA Part A, 27*(3), 2278–2281. https://doi.org/10.3109/19401736.2014.984179

170. Morton, O., Scheffers, B. R., Haugaasen, T., & Edwards, D. P. (2021). Impacts of wildlife trade on terrestrial biodiversity. *Nature Ecology & Evolution, 5*(4), 540–548. https://doi.org/10.1038/s41559-021-01399-y

171. Naderifar, M., Goli, H., & Ghaljaie, F. (2017). Snowball sampling: A purposeful method of sampling in qualitative research. *Strides in Development of Medical Education*, *14*(3), e67670. https://doi.org/10.5812/sdme.67670

172. Name, L. (2016, April). *Inside the Secret Trade That Threatens Rare Birds.* Wildlife Watch, National Geography Society.

173. Naro, E., Mero, E. L., Naro, E., Kapfo, K., Wezah, K., Thopi, K., Rhakho, K., Akami, L., Thopi, L., Chirhah, M., Chirhah, T., Tsuhah, T., Thopi, T., Wezah, W., Mero, W. L., Thopi, W., Thopi, K., Naro, T., Tsuhah, W., Dahanukar, N., & Molur, P. B. (2015). Project hunt: an assessment of wildlife hunting practices by local community in Chizami, Nagaland, India. *Journal of Threatened Taxa, 7*(11), 7729–7743; http://dx.doi.org/10.11609/JoTT.o4219.7729-43

174. Nash, S. V. (1997). *Fin, Feather, Scale and Skin: observations on the wildlife trade in Lao PDR and Vietnam.* Malaysia: TRAFFIC Southeast Asia.

175. Nekaris, K. A. I., Shepherd, C. R., Starr, C. R., & Nijman, V. (2010). Exploring cultural drivers for wildlife trade via an ethnoprimatological approach: a case study of slender and slow lorises (*Loris* and *Nycticebus*) in South and Southeast Asia. *American Journal of Primatology*, *72*(10), 877–886. https://doi.org/10.1002/ajp.20842

176. Nguyen, L. T., Schmidt, H. A., Von Haeseler, A., & Minh, B. Q. (2015). IQ-TREE: A fast and effective stochastic algorithm for estimating maximum-likelihood phylogenies. *Molecular Biology and Evolution, 32*(1), 268–274. https://doi.org/10.1093/molbev/msu300

177. Nijman, V. (2010). An overview of international wildlife trade from Southeast Asia. *Biodiversity and Conservation, 19*(4), 1101–1114. https://doi.org/10.1007/s10531-009-9758-4

178. Nijman, V., Zhang, M. X., & Shepherd, C. R. (2016). Pangolin trade in the Mong La wildlife market and the role of Myanmar in the smuggling of pangolins into China. *Global Ecology and Conservation, 5*, 118–126.

179. Niraj, K. S. & Ghosh, S. (2014, March). *Alarm bells over rising pet trade in wild animals in India (Unregulated pet trade causes conservation concern).* Traffic Post, TRAFFIC India.

180. Niraj, S. K. (2009). *Sustainable development, poaching, and illegal wildlife trade in India*. Doctoral Dissertation. The University of Arizona, USA.

181. Nitzsch, C. L. (1876). *System der Pterylographie: Nach seinen handschriftlich aufbewahrten Untersuchungen verfasst*. Halle: Eduard Anton.

182. Nooren, H., & Claridge, G. (2001). *Wildlife trade in Laos: the end of the game*. Gland: IUCN-The World Conservation Union.

183. Ogra, M. (2009). Attitudes toward resolution of human–wildlife conflict among forest-dependent agriculturalists near Rajaji National Park, India. *Human Ecology, 37*(2), 161–177. https://doi.org/10.1007/s10745-009-9222-9

184. Okonechnikov, K., Golosova, O., Fursov, M., & UGENE Team. (2012). Unipro UGENE: a unified bioinformatics toolkit. *Bioinformatics, 28*(8), 1166–1167. https://doi.org/10.1093/bioinformatics/bts091

185. Oldfield, S. (Ed.). (2002). *The Trade in Wildlife: Regulation for conservation*. London: Routledge.

186. Palsbøll, P. J., Clapham, P. J., Mattila, D. K., Larsen, F., Sears, R., Siegismund, H. R., Sigurjónsson, J., Vasquez, O., & Arctander, P. (1995). Distribution of mtDNA haplotypes in North Atlantic humpback whales: the influence of behaviour on population structure. *Marine Ecology Progress Series, 116*, 1–10.

187. Pap, P. L., Osváth, G., Daubner, T., Nord, A., & Vincze, O. (2020). Down feather morphology reflects adaptation to habitat and thermal conditions across the avian phylogeny. *Evolution, 74*(10), 2365–2376. https://doi.org/10.1111/evo.14075

188. Pap, P. L., Osvath, G., Sandor, K., Vincze, O., Bărbos, L., Marton, A., Nudds, R. L., & Vagasi, C. I. (2015). Interspecific variation in the structural properties of flight feathers in birds indicates adaptation to flight requirements and habitat. *Functional Ecology, 29*(6), 746–757. https://doi.org/10.1111/1365-2435.12419

189. Pereira, F., Carneiro, J., & Van Asch, B. (2010). A guide for mitochondrial DNA analysis in non-human forensic investigations. *The Open Forensic Science Journal, 3*(1), 33–44. DOI:10.2174/1874402801003020033

190. Perna, N. T., & Kocher, T. D. (1995). Patterns of nucleotide composition at fourfold degenerate sites of animal mitochondrial genomes. *Journal of Molecular Evolution, 41*(3), 353–358. https://doi.org/10.1007/BF00186547

191. Praveen J., Jayapal, R., & Pittie, A. (2021[2016]). A checklist of the birds of India. *Indian Birds*, *11*, 113–170.

192. Praveen, J., Jayapal, R., & Pittie, A. (2020). Taxonomic updates to the checklists of birds of India and the South Asian region—2020. *Indian Birds*, *16*(1), 12–19.

193. Prum, R. O., & Dyck, J. (2003). A hierarchical model of plumage: Morphology, development, and evolution. *Journal of Experimental Zoology Part B: Molecular and Developmental Evolution, 298B*(1), 73–90. https://doi.org/10.1002/jez.b.27

194. Qian, C., Ren, Q., Kan, X., Guo, Z., Yang, J., Li, X., Yuan, J., Qian, M., Hu, Q., & Zhang, L. (2013). Complete mitochondrial genome of the Red-billed Starling, *Sturnus sericeus* (Aves: Passeriformes): The first representative of the family Sturnidae with a single control region. *Mitochondrial DNA, 24*(2), 129–131. https://doi.org/10.3109/19401736.2012.731407

195. Qiang, J., Currie, P. J., Norell, M. A., & Shu-An, J. (1998). Two feathered dinosaurs from northeastern China. *Nature, 393*(6687), 753–761. https://doi.org/10.1038/31635

196. Rahmani, A. R., Islam, M. Z. U., & Kasambe, R. M. (2016). *Important Bird and Biodiversity Areas in India. Priority Sites for Conservation.* Second Edition: Revised and Updated. Volume I. Mumbai: Bombay Natural History Society, Indian Bird Conservation Network, Royal Society for the Protection of Birds and BirdLife International (UK).

197. Ramachandran, R., Kumar, A., Gopi Sundar, K. S., & Bhalla, R. S. (2017). Hunting or habitat? Drivers of waterbird abundance and community structure in agricultural wetlands of southern India. *Ambio, 46*(5), 613–620. https://doi.org/10.1007/s13280-017-0907-9

198. Rambaut A. (2007). FigTree, a graphical viewer of phylogenetic trees. http://tree.bio.ed. ac.uk/software.figtreetree.

199. Ray, S. D., Dey, P., Islam, N., Sharma, S. K., Pramod, P., & Singh, R. P. (2021). Comparative study of Yellow-billed Babbler (*Turdoides affinis*) feathers reveals uniformity in their microstructures among individuals. *Journal of Experimental Biology and Agricultural Sciences, 9*(1), 51–64. https://doi.org/10.18006/2021.9(1).51.64

200. Ray, S. D., Quadros, G., Dey, P., Pramod, P., & Singh, R. P. (2022). Feather characteristics of Common Myna *Acridotheres tristis* (Passeriformes: Sturnidae) from India. *Journal of Threatened Taxa, 14*(6), 21161–21169. https://doi.org/10.11609/jott.7936.14.6.21161-21169

201. Rentschlar, K. A., Miller, A. E., Lauck, K. S., Rodiansyah, M., Bobby, Muflihati, & Kartikawati. (2018). A silent morning: the songbird trade in Kalimantan, Indonesia. *Tropical Conservation Science, 11*, 1–10. https://doi.org/ 10.1177/1940082917753909

202. Rheindt, F. E., & Edwards, S. V. (2011). Genetic introgression: An integral but neglected component of speciation in birds. *The Auk, 128*(4), 620–632. https://doi.org/10.1525/auk.2011.128.4.620

203. Robertson, G. R. (2002). Birds of a feather stick: microscopic feather residues on stone artifacts from Deep Creek Shelter, New South Wales. In S. Ulm, C. Westcott, J. Reid, A. Ross, I. Lilley, J. Prangnell, & L. Kirkwood (Eds.), *Barriers, Borders, Boundaries: Proceedings of the 2001 Australian Archaeological Association Annual Conference* (Vol. 7, pp. 175–182). Australia: Anthropology Museum, The University of Queensland.

204. Robertson, J., Harkin, C., & Govan, J. (1984). The identification of bird feathers. Scheme for feather examination. *Journal of the Forensic Science Society, 24*(2), 85–98. https://doi.org/10.1016/S0015-7368(84)72301-2

205. Robinson, J. G., & Bennett, E. L. (2002). Will alleviating poverty solve the bushmeat crisis? *Oryx, 36*(4), 332–332. https://doi.org/10.1017/ S0030605302000662

206. Robinson, J. T., Thorvaldsdóttir, H., Winckler, W., Guttman, M., Lander, E. S., Getz, G., & Mesirov, J. P. (2011). Integrative genomics viewer. *Nature Biotechnology, 29*(1), 24–26. https://doi.org/10.1038/nbt.1754

207. Roe, D., Dickman, A., Kock, R., Milner-Gulland, E. J., Rihoy, E., & Sas-Rolfes, M. (2020). Beyond banning wildlife trade: COVID-19, conservation and development. *World Development, 136*, 105121. https://doi.org/10.1016/ j.worlddev.2020.105121

208. Rohwer, V. G., Rohwer, S., & Kane, L. (2021). Filoplume morphology covaries with their companion primary suggesting that they are feather-specific sensors. *Ornithology, 138*(3), ukab024. https://doi.org/10.1093/ornithology/ukab024

209. Ruedas, L. A., Salazar-Bravo, J., Dragoo, J. W., & Yates, T. L. (1999). The Importance of Being Earnest: What, if Anything, Constitutes a "Specimen Examined?". *Molecular Phylogenetics and Evolution, 17*(1), 129–132. https://doi.org/10.1006/mpev.2000.0737

210. Sambrook, J., Fritsch, E. F., & Maniatis, T. (1989). *Molecular cloning: a laboratory manual* (2nd ed.). Cold spring harbor: Cold spring harbor laboratory press.

211. Sangster, G., & Luksenburg, J. A. (2021). Sharp increase of problematic mitogenomes of birds: Causes, consequences, and remedies. *Genome Biology and Evolution, 13*(9), 1–14. https://doi.org/10.1093/gbe/evab210

212. Sarkar, I., Dey, P., Sharma, S. K., Ray, S. D., Kochiganti, V. H. S., Singh, R., Pramod, P., & Singh, R. P. (2020). *Turdoides affinis* mitogenome reveals the translational efficiency and importance of NADH dehydrogenase complex-I in the Leiothrichidae family. *Scientific Reports, 10*, 16202. https://doi.org/10.1038/ s41598-020-72674-4

213. Satoh, T. P., Miya, M., Mabuchi, K., & Nishida, M. (2016). Structure and variation of the mitochondrial genome of fishes. *BMC Genomics, 17*(1), 719. https://doi.org/10.1186/s12864-016-3054-y

214. Selvan, K. M., Veeraswami, G. G., Habib, B., & Lyngdoh, S. (2013). Losing threatened and rare wildlife to hunting in Ziro Valley, Arunachal Pradesh, India. *Current Science, 104*(11), 1492–1495.

215. Seutin, G., White, B. N., & Boag, P. T. (1991). Preservation of avian blood and tissue samples for DNA analyses. *Canadian Journal of Zoology, 69*(1), 82–90. https://doi.org/10.1139/z91-013

216. Sharma, S. (2014). *Protecting India's Wilderness, Introduction to wildlife laws in India.* Panda, WWF Traffic Newsletter, 19–24 pp.

217. Sharma, S. K. (1988). Bird casualties in road accidents. *Journal of Bombay Natural History Society, 85*(1), 195–197.

218. Silaeva, O. L., & Chernova, O. F. (2022). The Current State of Identification Ptilology in Russia. Biology Bulletin Reviews, 12(2), 149–163. https://doi.org/10.1134/s2079086422020098

219. Silaeva, O. L., Chernova, O. F., Varaksin, A. N., & Bukreev, S. A. (2022). Metric coordination between dimensional characteristics of birds and microscopic characters of their downy feathers on the example of some Anseriformes species. *Materials on flora and fauna of the Republic of Bashkortostan, 35*, 50–64.

220. Singh, R., Slade, L., & Ilyas, O. (2011). Trapped and traded—an insight into the field realities of the live bird trade in Uttar Pradesh, India. *Birding ASIA, 16*, 45–47.

221. Siva, T., & Neelanarayanan, P. (2020). Impact of vehicular traffic on birds in Tiruchirappalli District, Tamil Nadu, India. *Journal of Threatened Taxa, 12*(10), 16352–16356. https://doi.org/10.11609/jot.5532.12.10.16352-16356

222. Sollund, R., & Maher, J. (2015). *The illegal wildlife trade: A case study report on the illegal wildlife trade in the United Kingdom, Norway, Colombia and Brazil.* A study compiled as part of the EFFACE project. University of Oslo and University of South Wales.

223. Souto, W. M. S., Torres, M. A. R., Sousa, B. F. C. F., Lima, K. G. G. C., Vieira, L. T. S., Pereira, G. A., Guzzi, A., Silva, M. V., & Pralon, B. G. N. (2017). Singing for cages: The use and trade of Passeriformes as wild pets in an economic center of the Amazon—NE Brazil route. *Tropical Conservation Science, 10,* 1–19. https://doi.org/10.1177/1940082917689898

224. Spicer, G. S., & Dunipace, L. (2004). Molecular phylogeny of songbirds (Passeriformes) inferred from mitochondrial 16S ribosomal RNA gene sequences. *Molecular Phylogenetics and Evolution, 30*(2), 325–335. https://doi.org/10.1016/S1055-7903(03)00193-3

225. Stettenheim, P. R. (2000). The integumentary morphology of modern birds—an overview. *American Zoologist, 40*(4), 461–477. https://doi.org/10.1093/icb/40.4.461

226. Su, S., Cassey, P., & Blackburn, T. M. (2014). Patterns of non-randomness in the composition and characteristics of the Taiwanese bird trade. *Biological Invasions, 16*(12), 2563–2575. https://doi.org/10.1007/s10530-014-0686-1

227. Sullivan, T. N., Pissarenko, A., Herrera, S. A., Kisailus, D., Lubarda, V. A., & Meyers, M. A. (2016). A lightweight, biological structure with tailored stiffness: The feather vane. *Acta Biomaterialia, 41*, 27–39. https://doi.org/10.1016/j.actbio.2016.05.022

228. Sun, C. H., Liu, H. Y., & Lu, C. H. (2020). Five new mitogenomes of *Phylloscopus* (Passeriformes, Phylloscopidae): Sequence, structure, and phylogenetic analyses. *International Journal of Biological Macromolecules, 146*, 638–647. https://doi.org/10.1016/j.ijbiomac.2019.12.253

229. Sun, C. H., Liu, H. Y., Min, X., & Lu, C. H. (2020). Mitogenome of the little owl *Athene noctua* and phylogenetic analysis of Strigidae. *International Journal of Biological Macromolecules, 151*, 924–931. https://doi.org/10.1016/j.ijbiomac.2020.02.238

230. Sundar, G. K. S. (2004). Mortality of herpetofauna, birds and mammals due to vehicular traffic in Etawah District, Utter Pradesh, India. *Journal of the Bombay Natural History Society, 101*(3), 392–398.

231. Sundar, K. G., & Kittur, S. (2013). Can wetlands maintained for human use also help conserve biodiversity? Landscape-scale patterns of bird use of wetlands in an agricultural landscape in north India. *Biological Conservation, 168*, 49–56. https://doi.org/10.1016/j.biocon.2013.09.016

232. Sur, S., Saikia, P. K., & Saikia, M. K. (2022). Speed thrills but kills: A case study on seasonal variation in roadkill mortality on National Highway 715 (new) in Kaziranga-Karbi Anglong Landscape, Assam, India. *Nature Conservation, 47*, 87–104. https://doi.org/10.3897/natureconservation.47.73036

233. Taher, M., & Ahmed, P. (2021). *Assam: A geographical profile, 6th edition.* Guwahati: Mani Manik Prakash.

234. Talukdar, S., & Gupta, A. (2018). Attitudes towards forest and wildlife, and conservation-oriented traditions, around Chakrashila Wildlife Sanctuary, Assam, India. *Oryx, 52*(3), 508–518.

235. Tatusov T, Tatusov R. (2011). NCBI ORF Finder, Maryland: National Center for Biotechnology Information. http://www.ncbi.nlm.nih.gov/gorf/gorf. 2011

236. Terrill, R. S., & Shultz, A. J. (2022). Feather function and the evolution of birds. Biological Reviews, 1–22. https://doi.org/10.1111/brv.12918

237. Thomsen, J. B., Edwards, S. R., & Mulliken, T. A. (1992). *Perceptions, conservation and management of wild birds in trade.* Traffic International, WWF International, IUCN .

238. Scott, T. G. (1938). Wildlife mortality on Iowa highways. *American Midland Naturalist, 20*(3), 527–539. https://doi.org/10.2307/2420289

239. Töpfer, T. (2018). Morphological variation in birds: Plasticity, adaptation, and speciation. In D. T. Tietze (Ed.), *Bird Species, How They Arise, Modify and Vanish*, (pp. 63–73). Cham: Springer. https://doi.org/10.1007/978-3-319-91689-7_4

240. Trail, P. W., & Gaffney, A. M. (2021). Distinguishing gamebird and waterfowl feathers from raptor feathers. *Forensic Science International: Animals and Environments, 1*, 100009. https://doi.org/10.1016/j.fsiae.2021.100009

241. Tynsong, H., Tiwari, B. K., & Dkhar, M. (2012). Bird hunting techniques practiced by War Khasi community of Meghalaya, North-east, India. *Indian Journal of Traditional Knowledge, 11*(2), 334–341.

242. United Nations Office on Drugs and Crime (UNODC). (2020). *World Wildlife Crime Report 2020: Trafficking in Protected Species*. New York: United Nations Publication. https://www.unodc.org/documents/data-and analysis/wildlife/2020/ World_Wildlife_Report_2020_9July.pdf

243. United Nations Office on Drugs and Crime (UNODC). (2022). *Illegal wildlife trade and climate change: Joining the dots*. New York: United Nations Publication. https://www.unodc.org/documents/data-and-analysis/wildlife/llegal_wildlife_trade_and_climate_change_2022.pdf

244. Uprety, Y., Chettri, N., Dhakal, M., Asselin, H., Chand, R., & Chaudhary, R. P. (2021). Illegal wildlife trade is threatening conservation in the transboundary landscape of Western Himalaya. *Journal for Nature Conservation, 59*, 125952.

245. Urantówka, A. D., Kroczak, A., Silva, T., Padrón, R. Z., Gallardo, N. F., Blanch, J., Blanch, B., & Mackiewicz, P. (2018). New insight into parrots' mitogenomes indicates that their ancestor contained a duplicated region. *Molecular Biology and Evolution, 35*(12), 2989–3009. https://doi.org/10.1093/molbev/msy189

246. Vaithianathan, K. (2022). Flyways and Migratory Birds of India. *Science Reporter, 59*(6), 40–42.

247. Van der Zande, A. N., Ter Keurs, W. J., & Van der Weijden, W. J. (1980). The impact of roads on the densities of four bird species in an open field habitat—evidence of a long-distance effect. *Biological Conservation, 18*(4), 299–321. https://doi.org/10.1016/0006-3207(80)90006-3

248. van Uhm, D., & Spapens, T. (2018). Illegal trade in protected birds in the Netherlands. *Tijdschrift voor Criminologie, 60*(2), 231–245.

249. Vasquez, J. C. (2014). Compliance and enforcement mechanisms of CITES. In S. Oldfield (Ed.), *The Trade in Wildlife, Regulation for Conservation* (pp. 85–91). London: Routledge.

250. Velho, N., & Laurance, W. F. (2013). Hunting practices of an Indo-Tibetan Buddhist tribe in Arunachal Pradesh, north-east India. *Oryx, 47*(3), 389–392. https://doi.org/10.1017/S0030605313000252

251. Veríssimo, D., Challender, D. W., & Nijman, V. (2012). Wildlife trade in Asia: start with the consumer. *Asian Journal of Conservation Biology, 1*(2), 49–50.

252. Vijayakumar, S. P., Vasudevan, K., & Ishwar, N. M. (2001). Herpetofaunal mortality on roads in the Anamalai Hills, southern Western Ghats. *Hamadryad, 26*(2), 253–260.

253. Vinther, J., Briggs, D. E., Clarke, J., Mayr, G., & Prum, R. O. (2010). Structural coloration in a fossil feather. *Biology Letters, 6*(1), 128–131. https://doi.org/10.1098/rsbl.2009.0524

254. Voelker, G., & Spellman, G. M. (2004). Nuclear and mitochondrial DNA evidence of polyphyly in the avian superfamily Muscicapoidea. *Molecular Phylogenetics and Evolution, 30*(2), 386–394. https://doi.org/10.1016/S1055-7903(03)00191-X

255. Wang, Y., Shen, Y., Feng, C., Zhao, K., Song, Z., Zhang, Y., Yang, L., & He, S. (2016). Mitogenomic perspectives on the origin of Tibetan loaches and their adaptation to high altitude. *Scientific Reports, 6*, 29690. https://doi.org/10.1038/srep29690

256. Watt, G. (1908). *The commercial products of India: being an abridgment of the dictionary of the economic products of India*. London: John Murray.

257. WCS India. (2020). *Workshop report: WCS India and Assam Forest department Hold workshop to Fight against Wildlife Trafficking*. Guwahati: Wildlife Conservation Society.

258. Wilson-Wilde, L. (2010). Wildlife crime: a global problem. *Forensic Science, Medicine, and Pathology, 6*(3), 221–222.

259. Woolloff, A., Nkoke, S., Musing, L., & Svensson, M. S. (2022). *Cyber enabled wildlife trade in Central African countries and Nigeria*. United Kingdom: TRAFFIC International, Cambridge.

260. Traffic India (2016). *Workshop Proceedings First National Workshop on Capacity Building for Combating Wildlife Crime in India*. New Delhi: WWF TRAFFIC India.

261. World Bank Group (2018). *Tools and Resources to Combat Illegal Wildlife Trade*. Washington: World Bank.

262. Traffic (2008). *What's Driving the Wildlife Trade? A Review of Expert Opinion on Economic and Social Drivers of the Wildlife Trade and Trade Control Efforts in Cambodia, Indonesia, Lao PDR, and Vietnam*. Washington: East Asia and Pacific Regional Office, Rural Development, Natural Resources and Environment Sector Unit, & International Traffic Network, World Bank.

263. Wray, R. S. (1887). On the Structure of the Barbs, Barbules, and Barbicels of a Typical Pennaceous Feather. *Ibis, 29*(4), 420–423. https://doi.org/10.1111/j.1474-919X.1887.tb06624.x

264. WWF Traffic India. (2016). *Workshop Proceedings*: *First National Workshop on Capacity Building for Combating Wildlife Crime in India*. Coimbatore: Traffic India.

265. Wyatt, T. (2009). Exploring the organization of Russia Far East's illegal wildlife trade: two case studies of the illegal fur and illegal falcon trades. *Global Crime, 10*(1–2), 144–154.

266. Wyler, L. S., & Sheikh, P. A. (2008). *International illegal trade in wildlife: threats and US policy*. Washington: Congressional Research Service.

267. Xu, N., Ding, J., Que, Z., Xu, W., Ye, W., & Liu, H. (2021). The mitochondrial genome and phylogenetic characteristics of the Thick-billed Green-Pigeon, *Treron curvirostra*: The first sequence for the genus. *ZooKeys, 1041*, 167–182. https://doi.org/10.3897/zookeys.1041.60150

268. Yan, C., Mou, B., Meng, Y., Tu, F., Fan, Z., Price, M., Yue, B., & Zhang, X. (2017). A novel mitochondrial genome of *Arborophila* and new insight into *Arborophila* evolutionary history. PLoS ONE, 12(7), e0181649. https://doi.org/10.1371/journal.pone.0181649

269. Yan, S. Q., Guo, P. C., Li, Y. M., Qi, S. M., Bai, C. Y., Zhao, Z. H., & Sun, J. H. (2015). Complete mitochondrial genome of the Spotted dove (*Streptopelia chinensis*). *Mitochondrial DNA Part A, 27*(5), 3067–3068.

270. Yang, C., Du, X., Liu, Y., Yuan, H., Wang, Q., Hou, X., Gong, H., Wang, Y., Huang, Y., Li, X., & Ye, H. (2022). Comparative mitogenomics of the genus *Motacilla* (Aves, Passeriformes) and its phylogenetic implications. *ZooKeys, 1109*, 49–65. https://doi.org/10.3897/zookeys.1109.81125

271. Yang, C., Yang, M., Wang, Q., Lu, Y., & Li, X. (2018). The complete mitogenome of *Falco amurensis* (Falconiformes, Falconidae), and a comparative analysis of genus *Falco*. *Zoological Science, 35*(4), 367–372. https://doi.org/10.2108/zs170182

272. Yu, J., Liu, J., Li, C., Wu, W., Feng, F., Wang, Q., Ying, X., Qi, D., & Qi, G. (2021). Characterization of the complete mitochondrial genome of *Otus lettia*: exploring the mitochondrial evolution and phylogeny of owls (Strigiformes). *Mitochondrial DNA Part B, 6*(12), 3443–3451. https://doi.org/10.1080/23802359.2021.1995517

273. Zhang, R. H., He, W. X., & Xu, T. (2015). Characterization of the complete mitochondrial genome of the king pigeon (*Columba livia breed king*). *Mitochondrial DNA, 26*(3), 491–492. https://doi.org/10.3109/ 19401736.2014.1003906

274. Zhang, Z., Li, S., Zhang, J., Song, W., Yang, J., & Mu, J. (2021). The complete mitochondrial genome of an endangered minnow *Aphyocypris lini* (Cypriniformes: Xenocyprididae): genome characterization and phylogenetic consideration. *Biologia, 76*(11), 3311–3321. https://doi.org/10.1007/s11756-021-00811-z

275. Zuccon, D., Cibois, A., Pasquet, E., & Ericson, P. G. (2006). Nuclear and mitochondrial sequence data reveal the major lineages of starlings, mynas and related taxa. *Molecular Phylogenetics and Evolution, 41*(2), 333–344. https://doi.org/10.1016/j.ympev.2006.05.007

276. Zuccon, D., Pasquet, E., & Ericson, P. G. (2008). Phylogenetic relationships among Palearctic–Oriental starlings and mynas (genera *Sturnus* and *Acridotheres*: Sturnidae). *Zoologica Scripta, 37*(5), 469–481. https://doi.org/10.1111/j.1463-6409.2008.00339.x

Appendix

Appendices

Appendix-1: Questionnaire used for snowball sampling of illegally traded avifauna in Assam.

Questionnaire for Collecting Information on Illegal Avian Trade in Assam

Sálim Ali Centre for Ornithology & Natural History

Anaikatty, Coimbatore, TamilNadu-641108

(Affiliated to Bharathiar University, Coimbatore, Tamil Nadu)

Data collector:

Q. Sheet no._____ Date_____

1. General Details of Surveyed Market

Name of the market	District	Nearby	Area
GPS:……°……'……."/……°……'……."			

2. Additional Details of the Market

Nearby NP/WLS/Forest area	Division &Range	Beat

3. Details of Surrounding Villages

Name of Village	Adj. forest area	Adj. wetland	Major earning source	Area (urban/semi urban)

4. General details of Live species/Bushmeat/ Miscellaneous item in the Market

Mammals	Birds (Local / Vernacular Name)	Wild Birds Eggs	Known Bush-Meat	Unknown Bush-Meat	Miscellaneous Item

5. Detail of the Avian species

Sl. No	Species Name	Local / Vernacular Name	Male	Price	Female	Price	Juvenile	Price

Note:

6. Use of the Avian species being hunted and sold in the market

Medicinal	
Rituals	
Superstition	
Bush-Meat	
Local Demand	
High Profit	
Use as Pet	
Others	

7. Collection area of the avian species and distance of that area from the market

NP	WLS	Forest area	Fringe village	Wetland	Riverside	Agricultural area	Locally trapped
Dist.	Dist.	Dist.	Dist.	Dist.	Dist.	Dist.	Dist.

8. Collection Methods of Avian species for Market selling

Traditional trap	Hunting	Collection from the nest	Using net	Poisoning	Others

9. Collectors Details

Sl. No.	Name	Age	Address	Occupation	Professional / Occasional Trader	Freq. Of Species Collection	Approx. Income Rate

10. Any other Species which are commonly seen in the market by other vendors?

Sl. No.	Species Name	Local / Vernacular Name	Age group	Price

11. Sellers interested to continue trading of wild species?

12. Sellers aware about conservation laws of wild species?

13. Any local NGO's or any individual working in that area working for conservation of wild species?

Note:

Publications

Journal of Threatened Taxa | www.threatenedtaxa.org | 26 June 2022 | 14(6): 21161–21169
ISSN 0974-7907 (Online) | ISSN 0974-7893 (Print)
https://doi.org/10.11609/jott.7936.14.6.21161-21169
#7936 | Received 27 March 2022 | Final received 01 May 2022 | Finally accepted 10 June 2022
OPEN ACCESS
COMMUNICATION

Feather characteristics of Common Myna *Acridotheres tristis* (Passeriformes: Sturnidae) from India

Swapna Devi Ray [1], Goldin Quadros [2], Prateek Dey [3], Padmanabhan Pramod [4] & Ram Pratap Singh [5]

[1,3,4,5] National Avian Forensic Laboratory, [2] Wetland Ecology Division,
Sálim Ali Centre for Ornithology and Natural History, Anaikatty, Coimbatore, Tamil Nadu 641108, India.
[1,3] Bharathiar University, Coimbatore, Tamil Nadu 641046, India.
[5] Department of Life Science, School of Earth, Biological and Environmental Sciences, Central University of South Bihar, Gaya, Bihar 824236, India.
[1] swapnadray555@gmail.com, [2] goldinq@gmail.com, [3] pratikdey23@gmail.com, [4] neosacon@gmail.com,
[5] rampratapsingh81@gmail.com (corresponding author)

Abstract: The systematic study of feather microstructure supports species identification, which is important in cases of illegally traded birds and bird-aircraft strikes. Our study focused on morphometric, macro- and micro-characters of feathers of Common Myna *Acridotheres tristis* from India. Among macro-characters, silver-colored filoplume feathers with pale black pigmentation on the barbs are specific for *A. tristis*. Morphometric measurements revealed that primary contour feathers (10.8±0.100 cm) were the longest and bristle feathers (1.26±0.051 cm) the shortest among all feathers. The longest (average) barb is found in primary contour feathers (1.875±0.123 cm), and the shortest in filoplume feathers (0.288±0.017 cm). We observed 3 types of nodal structures, and elongated prongs in bristle and filoplume feathers are significant characteristics of *A. tristis*. These insights into feather microstructures of *A. tristis* will aid species identification using plumology.

Keywords: Micro-structure, macro-structure, morphometry, plumology, Sturnidae.

Editor: P.A. Azeez, Coimbatore, Tamil Nadu, India. **Date of publication:** 26 June 2022 (online & print)

Citation: Ray, S.D., G. Quadros, P. Dey, P. Pramod & R.P. Singh (2022). Feather characteristics of Common Myna *Acridotheres tristis* (Passeriformes: Sturnidae) from India. *Journal of Threatened Taxa* 14(6): 21161–21169. https://doi.org/10.11609/jott.7936.14.6.21161-21169

Funding: This work was supported by the Ministry of Environment, Forest and Climate Change (MoEFCC).

Competing interests: The authors declare no competing interests.

Author details: Swapna Devi Ray is doctoral student of Bharathiar University, Coimbatore and affiliated to SACON. She is a researcher from ecology and environmental Science background. For her doctoral degree she is working on wildlife crime, plumology and molecular markers of avian species at National Avian Forensic Laboratory, SACON. Dr. Goldin Quadros is working as Principal Scientist at the Wetland Ecology Division of SACON. His area of specialization is the benthic fauna from the intertidal regions of coasts, estuaries and creeks. He is also working as the coordinator of ENVIS Centre at SACON. Prateek Dey is a doctoral student of Zoology (Avian Genetics) at SACON. He has Integrated MSc in life sciences from Central University of Tamil Nadu. Currently he is working on whole genome sequencing of birds. Dr. Padmanabhan Pramod is a Senior Principal Scientist and head of Nature Education programme at SACON. Over a period of 27 years he has carried out variousresearch projects in the field of ecosystem assessments, eco-development and mitigation measures for the bird hazards to aircrafts. Dr. Ram Pratap Singh is Associate Professor and Head, Department of Life Science at Central University of South Bihar. The primary focus of Dr. Singh's research is Avian Genetics and Avian Forensic. He established the National Avian Forensic Laboratory and started the Genome Resource Bank at SACON.

Author contributions: S.D.R. collected the sample; R.P.S., G.Q. and P.P. conceived the idea and supervised the research; R.P.S., and P.P. generated the funds for the study. S.D.R, G.Q., P.D. and R.P.S. standardized the methodology; S.D.R. generated the data. S.D.R., G.Q. and R.P.S. wrote the manuscript and analyzed the data. All the authors reviewed the manuscript.

Acknowledgements: We are thankful to the Ministry of Environment, Forest and Climate Change (MoEFCC) for providing funding to the project. We thank to the Assam Forest Department and the Assam State Biodiversity Board for providing necessary permissions. We are greatly thankful to the field assistance Mr. Niren Singh for his assistance during field works in Assam.

Biologia
https://doi.org/10.1007/s11756-023-01327-4

ORIGINAL ARTICLE

Complete mitogenome of common myna (*Acridotheres tristis*) – characterization and phylogenetic implications

Swapna Devi Ray[1,2] · Prateek Dey[1,2] · Indrani Sarkar[1] · Sanjeev Kumar Sharma[1] · Goldin Quadros[1] · Padmanabhan Pramod[1] · Venkata Hanumat Sastry Kochiganti[3] · Ram Pratap Singh[4]

Received: 7 June 2022 / Accepted: 17 January 2023

Abstract

Common myna (*Acridotheres tristis*) is a member of the Sturnidae family with widespread natural and introduced habitat range across many continents. *Acridotheres tristis* population thrives in close association with human settlements and agricultural fields making it one of the most invasive bird species worldwide. For better management and conservation of *A. tristis* species, genetic information collected from various locations throughout its habitat range is necessary. Hence, we sequenced *A. tristis* mitogenome (NCBI accession no. OK542103) using NGS tools and characterized 37 genes (including the Protein Coding Genes, tRNA and rRNA) in the newly sequenced mitogenome. Codon usage analysis of selected mitogenomes investigated in this study revealed the third position of codons were mostly predominated by cytosine (C3) followed by adenine (A3). From the dN/dS analysis the strongest purifying selection was observed on nad4l (0.04 units) and the lowest on nad4 (0.34 units). The mitochondrial control region (CR) displayed identical ancestral avian CR gene order. Topology inferred from the concatenated mitogenome PCGs based phylogeny provided evidence for the clustering of Mimidae species within the Sturnidae family. *Acridotheres* as a genus is closely related to *Sturnus cineraceus* and *Sturnus sericeus*. The complete mitogenome of *A. tristis* sequenced in this study along with a comprehensive bioinformatics analysis of selected Sturnidae mitogenomes provides invaluable insights and genetic resources for designing future studies on *A. tristis* species with conservation and management implications.

Keywords *Acridotheres tristis* · Mitogenome · Codon usage · Sturnidae phylogeny

Introduction

Common myna (*Acridotheres tristis* (Linnaeus, 1766)) is a medium-sized passerine bird of the Sturnidae family found abundantly across the wide distribution area of south and central Asia including the Indian subcontinent (Feare and Craig 1999). *Acridotheres tristis* dwell comfortably across environments and are easily introduced and bred in a wide variety of ecosystems (Kannan and James 2020; Feare and Craig 1999). Owing to such a life-history trait *A. tristis* is recognized as a model organism for evolutionary biologists to study the evolution of avian species across a wide range of biogeographic realms (Baker and Moeed 1987; Fleischer et al. 1991; Kannan and James 2020). However, widespread range and easy adaptation to new environments have also led to *A. tristis* being recognized as one of the top 100 most invasive species (Lowe et al. 2000). Across their extant habitat *A. tristis* feed on insect or plant resources

Swapna Devi Ray and Prateek Dey contributed equally.

✉ Ram Pratap Singh
rampratapsingh81@gmail.com

1 Sálim Ali Centre for Ornithology and Natural History, 641108 Anaikatti, Coimbatore, Tamil Nadu, India

2 Bharathiar University, 641046 Coimbatore, Tamil Nadu, India

3 National Institute of Animal Nutrition and Physiology, 560030 Bengaluru, India

4 Department of Life Science, Central University of South Bihar, 824236 Gaya, Bihar, India

Published online: 24 January 2023

Department of Environmental Science, K.R.T. Arts, B.H. Commerce & A.M. Science (KTHM), College, Nashik, Maharashtra, India.

In Collaboration With

Brazil Chapter – International Youth Society

Organized

Online International Multidisciplinary Conference

On

Recent Trends in Environmental Science and Management

Conference Date: August 6 & 7, 2021

Conference Website: https://kailasevs.wixsite.com/imcrtesm2021

IMCRTESM/LAA/79 Date: 27/07/2021

Letter of Abstract Acceptance

(Abstract ID: IMCRTESM202179)

Dear Swapna Devi Ray

We cordially invites you to attend the "Online International Multidisciplinary Conference on Recent Trends in Environmental Science and Management (IMCRTESM 2021) to be held on August 6 & 7, 2021.

We welcome you to join us and share your knowledge and views on the theme "Environmental Science and Management". We are glad to inform you that your abstract has been accepted by the review/scientific committee under Oral Presentation. In this regard we are pleased to welcome you to join us and give a presentation on "Quantification of Different Feather Characteristics of Common Myna (Acridotheres tristis)."

The Online International Multidisciplinary Conference on Recent Trends in Environmental Science and Management (IMCRTESM 2021) jointly organized by Department of Environmental Science, K.R.T. Arts, B.H. Commerce & A.M. Science (KTHM), College, Nashik, Maharashtra, India and Brazil Chapter - International Youth Society, Australia.

We look forward to seeing you in IMCRTESM 2021.

For more details about IMCRTESM 2021: https://kailasevs.wixsite.com/imcrtesm2021 / Or feel free to write us on imcrtesm2021@kthmcollege.ac.in

Thanks and Regards

Prof. K. D. Ahire

Dr. H.C. Tiago Soares

Quantification of Different Feather Characteristics of Common Myna (*Acridotheres tristis*).

Swapna Devi Ray, Goldin Quadros, Sanjeev Kumar Sharma, Prateek Dey, Ram Pratap Singh

Abstract

The systematic study of feather microstructure is very convenient for analyzing paleontology, archeology, food habits of prey species, forensic and bird-aircraft hit. But, it has not been explored much in India. Feather forensic has great importance in species identification using feathers in case of illegal bird trade and bird-aircraft hit.

In our study, we have followed a systematic approach to document qualitative and quantitative characteristics of Common Myna (*Acridotheres tristis*) feathers for identifying species-specific feather signatures. *A. tristis* belongs to family Sturnidae which is widely distributed all over India. Although, IUCN has declared *A. tristis* as one of the world's most invasive species; still this bird species is found to be a popular "cage bird" because of their singing ability. In India, *A. tristis* comes under one of the top five mostly traded avian species in pet markets.

Feather samples of *A. tristis* were collected during road-kill survey in Assam from the adjacent road-stretches of Kaziranga National Park in the month of September, 2019.with due permission of Assam Forest Department and Assam State Biodiversity Board. Samples were collected from wings, dorsal, ventral and tail regions of*A. tristis*. For each feather type viz. primary contour feather, secondary contour feather, tail contour feather, body contour feather, semiplume feather, down feather and powder down feather, two feathers were collected. Filoplume feathers could not be retrieved due to the poor condition of carcass. A total 28 feathers were selected for the microstructure study of the species. Feather samples were scanned by using Samsung SCX-4x21Series printer for their morphometric measurements. Each of these feathers weredivided into threedifferent regions as proximal, intermediate and distal. From each region, fivenumbers of feather barbs were sampled for slide preparation. Using dry mount method permanent slides were prepared for this study. From one single feather, 15 numbers of slides were prepared for thestudy. All the slides were observed under LaboMed LX 500 light microscope in 40X, 100X

and 400X resolutions. Different feather microstructures were documented and studied by using Mia Cam software.

Macro-characteristics viz. colour, pattern and texture were also observed for the studied feathers. Based on the location of feathers, colour varied from white, white-brown to different shades of brown. There was nosingle pattern present on the feathers. Flight contour feathers i.e. primary contour feather, secondary contour feather, tail feather and bristle feather were rigid in texture where body contour, semiplume, down and powder down feathers were present as soft and fluffy in texture.

From morphometric measurements, primary contour feather (10.9 cm) is the longest whereas down feather (3.3 cm) is the shortest among all studied feathers. Average longest barb length is present in primary contour feather (1.875±0.123 S.E. in cm) and the shortest barb length present in bristle feather (0.316±0.008 S.E. in cm).

Both pointed and knobbed shaped villi were present in the basal cell of all types of studied feathers. Three different nodal shapeswere present on the barbules of feather barbs, as plain nodal shape without prong, plain nodal shape with prong and quadrilobed shaped nodes with dark pigmentation. Distinct hooklets and ventral teeth are present only on proximal region of feather barbs of all contour feathers i.e. primary contour, secondary contour, tail contour and body contour. Internodes are straight in structure and patchy in pigmentation. Both patchy and dark brown pigmentation present on the ramus of the barbs except powder down and bristle where ramus is only present with dark pigmentation.

The microstructure data showed the presence of specific microstructures i.e. knobbed shaped and pointed villi, plain darkly pigmented nodal structures; which are the significant characteristics of passerine birds. The possible utility of our study is to use the data as tool in both forensic study and bird-strike case studies. This is the first study on *A. tristis* from India which provides the insights of feather microstructures with its significance in species identification.

Key Words: *Feather, microstructure, Plumology, Sturnidae.*

STRIDE
SCHEME FOR TRANS-DISCIPLINARY RESEARCH FOR INDIA'S DEVELOPING ECONOMY

INTERNATIONAL YOUTH SOCIETY

Certificate

Maratha Vidya Prasarak Samaj's

K.R.T. Arts, B.H. Commerce and A.M. Science (KTHM) College, Nashik, (MS), India,

Department of Environmental Science & UGC-STRIDE Cell

In Collaboration With

Brazil Chapter - International Youth Society, Australia

Organized

IMCRTESM 2021

Abstract ID: IMCRTESM202179

International Multidisciplinary Conference on Recent Trends in Environmental Science and Management

This is to certify that Prof./Dr./Mr./Ms. *SWAPNA DEVI RAY* of *Bharathiar University, Coimbatore; Salim Ali Centre for Ornithology and Natural History (SACON), Anaikatty, Coimbatore, TN* has participated & presented paper on *Quantification of Different Feather Characteristics of Common Myna (Acridotheres tristis)* in the International Multidisciplinary Conference on Recent Trends in Environmental Science and Management organized by Department of Environmental Science & UGC-STRIDE Cell, K. R. T. Arts, B. H. Commerce, A. M. Science (KTHM) College, Nashik, Maharashtra, India in collaboration with Brazil Chapter, International Youth Society, Australia held during 6th & 7th August 2021.

Mr. K. D. Ahire
Convener
IMCRTESM 2021

Dr. H. C. Tiago Soares
Convener, IMCRTESM, 2021
Country Director Brazil, IYS

Dr. Piya Ratna Maharjan
Global President,
International Youth Society

Dr. P. M. Nalawade
Head, Dept. of Envt. Science,
KTHM College, Nashik

Dr. N. D. Gaikwad
IQAC Coordinator
KTHM College, Nashik

Dr. V. B. Gaikwad
Principal
KTHM College, Nashik

STRIDE
SCHEME FOR TRANS-DISCIPLINARY RESEARCH FOR INDIA'S DEVELOPING ECONOMY

INTERNATIONAL YOUTH SOCIETY

Certificate of Recognition

Maratha Vidya Prasarak Samaj's

K.R.T. Arts, B.H. Commerce and A.M. Science (KTHM) College, Nashik, (MS), India,

Department of Environmental Science & UGC-STRIDE Cell

In Collaboration With

Brazil Chapter - International Youth Society, Australia

Organized

International Multidisciplinary Conference on Recent Trends in Environmental Science and Management

The International Award Selection Committee is proudly conferred **"YOUNG SCIENTIST AWARD"** to Dr. / Ms. *SWAPNA DEVI RAY* of *Bharathiar University, Coimbatore; Salim Ali Centre for Ornithology and Natural History (SACON), Anaikatty, Coimbatore, TN* for the outstanding presentation on *"Quantifi-cation of Different Feather Characteristics of Common Myna (Acridotheres tristis)"* in the International Multidisciplinary Conference on Recent Trends in Environmental Science and Management organized by Department of Environmental Science & UGC-STRIDE Cell, K. R. T. Arts, B. H. Commerce, A. M. Science (KTHM) College, Nashik, Maharashtra, India in collaboration with Brazil Chapter, International Youth Society, Australia held during 6th & 7th August 2021.

Mr. K. D. Ahire
Convener
IMCRTESM 2021

Dr. H. C. Tiago Soares
Convener, IMCRTESM, 2021
Country Director Brazil, IYS

Dr. Piya Ratna Maharjan
Global President,
International Youth Society

Dr. P. M. Nalawade
Head, Dept. of Envt. Science,
KTHM College, Nashik

Dr. N. D. Gaikwad
IQAC Coordinator
KTHM College, Nashik

Dr. V. B. Gaikwad
Principal
KTHM College, Nashik

Illegal Trade of Avian Species-A Case Study from Assam

[1,2] Swapna Devi Ray, [1,2] Prateek Dey, [1] Sanjeev Kumar Sharma, [3] P A Azeez, [4] Goldin Quadros and [1,5] Ram Pratap Singh

[1]National Avian Forensic Laboratory, Sálim Ali Centre for Ornithology and Natural History, Anaikatty, Coimbatore, Tamil Nadu, India

[2]Bharathiyar University, Maruthamalai main road, Coimbatore North taluk, Coimbatore, Tamil Nadu, India

[3]Environmental Impact Assessment Division, Sálim Ali Centre for Ornithology and Natural History (SACON), Anaikatty (PO), Coimbatore, Tamil Nadu, India

[4]Wetland Ecology Division, Sálim Ali Centre for Ornithology and Natural History, Anaikatty, Coimbatore, Tamil Nadu, India

[5]Dept. of Life Science, School of Earth, Biological and Environmental Sciences, Central University of South Bihar, Gaya, Bihar, India

Globally, the trade of wild species, a multi-billion dollar business, is attributed to be a major cause for the decline of numerous wild flora and fauna. Of the many wild species traded, the trafficking of avifauna is huge, with approximately 3300 species being subject to illegal trade. In India, reportedly around 450 bird species are traded in more than 900 markets, making the country the 3rd highest in global bird trade. The unlawful trade is perhaps the main reason for at least 176 bird species becoming 'threatened'. The illegal bird trade in India is greatly lucrative, relatively fail-safe and has developed into a well-organized business.

The documentation of illegal trade and trafficking has been mainly focused in the central and northern states of India, while there is a huge information lacuna on illegal avian trade, especially from North-East parts. It is speculated that northeastern states would be more vulnerable to illegal avian trade because of geographical, geopolitical, and socio-cultural reasons. Hence, Assam was chosen as the study area for the present investigation since it is a known "transitory route" for wildlife trafficking along the northeastern region. However, there is a serious dearth of systematic literature on wildlife trafficking in the state.

The market survey was undertaken in the vicinity of the IBA sites as well as protected areas across different districts of Assam. The snowball sampling method with a semi-structured questionnaire survey was conducted covering two seasons, winter and monsoon, in select markets for gaining insights on the traded avian species. In total 130 local markets from 17 districts of Assam were surveyed by interacting with 1003 respondents. A total of 82 avian species representing 33 families and 12 orders were found in the illegal trade in the region. Species belonging to the order Passeriformes (35%) and family Anatidae (13%) were highly frequent in the trade. The major reasons for the trade of avian species were as bush-meat (51%, N=62), plume-trade (14%, N=17), pet

trade (13%, N=16), superstition and black magic (12%, N=14), and cultural/traditional uses (10%, N=12). The respondents reported about nine trapping methods commonly used to collect birds from their natural habitats. Of these, the use of bait (18%) was the highest, while pitfall trap (2%) was used only for a few species. The number of traded species, according to the respondents, varied significantly (X^2= 765.798, df= 81, p=0.001). Our result showed that a strong correlation between the traded avian species and their market price (Pearson correlation coefficient: r=0.60, p<0.001), the price largely reflecting the demand.

Our work gives important insight into the level of illegal avian trade in Assam and the potential threat it poses to the wild avifauna in the region. The information on trapping methods, the use of the traded species and their prices can give a perception of the intensity of the ongoing illegal trade. The information provides very critical cues to the forest departments, other law enforcement agencies, and researchers to identify and execute appropriate action plans to curb the illegal wildlife trade.

Keywords: *Avifauna, Illegal Trade, Bush-meat, trapping, Plume-trade, Pet trade, Superstition and black magic, Cultural/Traditional use.*

7TH INTERNATIONAL CONFERENCE ON ENVIRONMENT AND ECOLOGY

Jointly Organised by

International Foundation for Environment and Ecology, Kolkata, West Bengal

Dept. of Environmental Science, Bharathiar University, Tamil Nadu

Dept. of Zoology, Govt. Degree College, Bhadarwah, Jammu & Kashmir

Dept. of Zoology, Mahishadal Raj College, West Bengal

Date: 26th, 27th and 28th November, 2021

Certificate of Participation and Paper Presentation

This is to certify that Swapna Devi Ray of Sálim Ali Centre for Ornithology and Natural History (SACON), Coimbatore, Tamil Nadu has participated in the 7th International Conference on Environment and Ecology (ICEE 2021) held on 26th, 27th and 28th November, 2021 through Google Meet and also presented a paper on "Illegal Trade of Avian Species-A Case Study from Assam."

Dr. Tridib Bandopadhyay
Chairman
International Foundation for Environment and Ecology, Kolkata, West Bengal

Dr. Manimekalan A.
HoD of Environmental Science
Bharathiar University, Coimbatore, Tamil Nadu

Dr. Kuldeep Kumar Sharma
Principal
Government Degree College, Bhadarwah, Jammu and Kashmir

Dr. Asim Kumar Bera
Principal
Mahishadal Raj College, Dist.- Purba Medinipur, West Bengal

Date: 28th November, 2021

Certificate No.: *ICEE/168/2021*