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No. 12, Thiruvannamalai Nagar, Saravanampatti - Kalapatti Road, Saravanampatti,
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Ph: +91 9385339863 | www.threatenedtaxa.org

Email: sanjay@threatenedtaxa.org

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Cover: *Euphaea pseudodispar* shot at Kalindi River, Thirunelly, Wayanad district, Kerala. © Muneer P.K.



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

Swapna Devi Ray¹ , Goldin Quadros² , Prateek Dey³ , Padmanabhan Pramod⁴ & Ram Pratap Singh⁵

^{1,3,4,5} National Avian Forensic Laboratory, ² Wetland Ecology Division, Sálím 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.

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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 various research 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.

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INTRODUCTION

Feathers cover the body of birds (Gill 2007) and support their survival in a wide range of climatic conditions (Lovette & Fitzpatrick 2016). The study of the microscopic structures of feathers and their systematic description (i.e., plumology) has provided a useful tool in studies of bird evolution (Chandler 1916; Dove 1997), paleontology, archeology, ecology (e.g., examining feeding habits using prey remains) and in the forensic examination of bird strikes (Dove 1997), where feather microstructures support the identification of avian species (Chandler 1916; Lei et al. 2002; Dey et al. 2021). In India only a few recent plumology studies (Dey et al. 2021; Ray et al. 2021) have been reported.

The Common Myna *Acridotheres tristis* belongs to the family Sturnidae, and is widely distributed across the Indian subcontinent. It is a medium-sized (~25 cm) bird, with no distinct sexual dimorphism (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), *A. tristis* is among the top five most traded avian species in Indian pet markets and in the illegal pet/avian trade (Ahmed 1997, 2013). *A. tristis* is sold at a high price in both domestic and international illegal pet markets as Hill Myna *Gracula religiosa* by disguising its appearance with slight morphological modifications (Ahmed 1997). Without detailed examination it is difficult to distinguish these species (Ahmed, 1997; Lei et al. 2002), and the high demand for *G. religiosa* in the pet trade has put pressure on population of *A. tristis*. Plumology can be used to identify these birds from their feather microstructures (Dove 1997; Lei et al. 2002; Lee et al. 2016; Dey et al. 2021; Ray et al. 2021).

In the present study, we have focused on the systematic approach to document qualitative and quantitative feather characteristics of *A. tristis* useful for identifying species-specific feather signatures. We describe specific microstructures present in both pennaceous and plumulaceous barbs that can be used as baseline data for future plumology studies in India.

METHODS

Feathers from a specimen of *A. tristis* (26.60°N; 93.47°E) were collected during a road-kill survey in September 2019 from adjacent road-stretches of Kaziranga National Park, Assam, India (Figure 1). Permissions were obtained for the collection of avian

biological samples 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). Feathers from the collected individuals were sampled, and macro characteristics, microstructures and morphometric measurements were documented following methods described by Chandler (1916), Dove (1997), and Dey et al. (2021).

Nine different types of pennaceous and plumulaceous feathers were sampled from five different body locations (Image 1) as follows:

1. Primary contour feathers and secondary contour feathers were collected from the right wing;
2. Tail contour feathers were collected from the tail region;
3. Body contour, semiplume, down and powder down feathers were collected from dorsal, ventral, and tail regions.
4. Modified contour feathers known as bristle feathers were collected from specific locations near the eyes and beak.
5. Filoplume feathers, which are filamentous in structure, were retrieved from the right wing.

For primary contour, secondary contour, tail contour, body contour, semiplume, down and powder down types of feathers, two numbers from each type from their respective locations were retrieved for the study. Due to the location specificity, five each of bristle and filoplume feathers were collected. A total of 38 different feathers were studied to document macro characteristics and microstructures.

Based on morphometric measurements of rachis, the feathers were divided into three different regions, proximal, intermediate and distal, except for powder down and bristle feathers (Dey et al. 2021). Because of the absence of proper rachis, the powder down and bristle feathers were not divided into the three regions. From each region, five barbs were sampled for slide preparation. Five each of bristle and filoplume feathers were whole-mounted on slides. The slides were prepared using the dry mount method (Ray et al. 2021; Dey et al. 2021).

Feather macro characters were observed by focusing on three main characters: colour, pattern and texture. Morphometric characters were measured from feathers' photographs for calamus length, vane length and rachis length using imageJ software. The feather microstructures were observed and documented using LaboMed Lx 500 compound light microscope. Slides

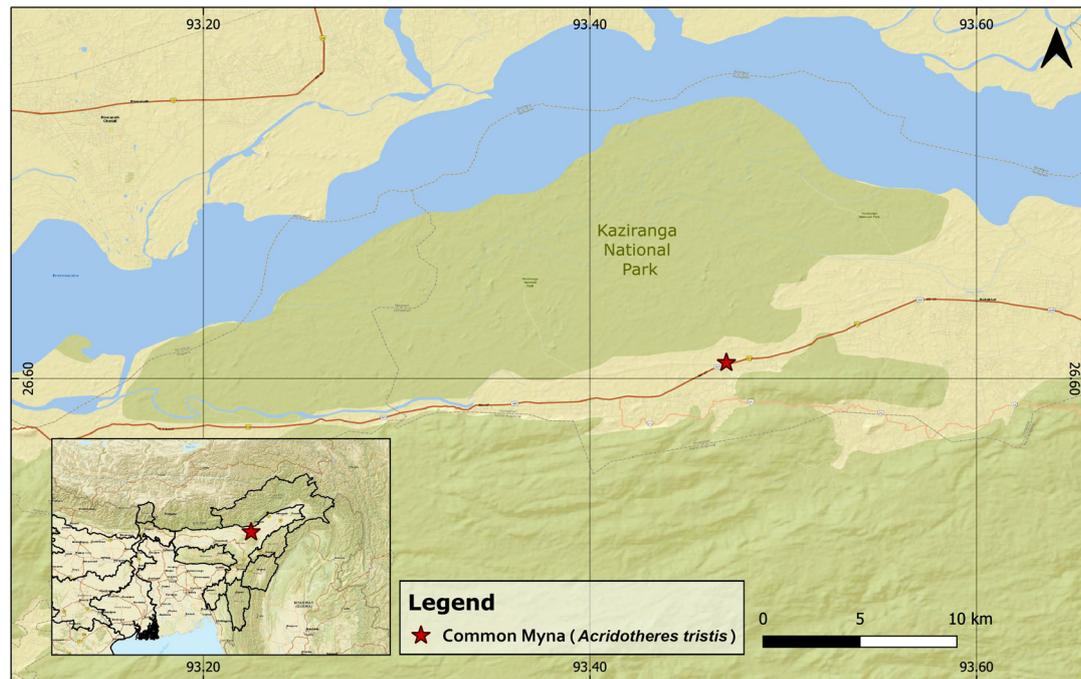


Figure 1. Geotag location of road-killed Common Myna.

were observed under 4X, 10X and 40X magnification for different characters, including presence of sub-pennaceous region, presence of villi, shape of villi, presence of nodes, shape of nodes, presence of hooklets, presence of prongs, size of prongs, presence of ventral teeth, shape of internodes, pigmentation on nodes, internodes, and ramus.

RESULTS

Feather macro characters

The feather macro characters documented for *A. tristis* are presented in Table 1. Feather color varied from black and white to dark brown to pale white and brown, even dark brown with a tinge of white. Only filoplume feathers showed a silvery appearance with pale black colored barbs at the tip. The texture of feathers varied. Flight contour feathers (primary contour, secondary contour and tail contour feathers) and bristles that represent modified contour feathers were firmly rigid, body contour feathers irrespective of their location were semi-rigid, and semiplume, down and powder down feathers were soft and fluffy.

Feather morphometry

Calamus length, vane length and rachis length of the nine different types of feathers were measured (Table

2). The primary contour feather from the wing was the longest; the average length for the calamus was 1.45 ± 0.050 cm, vane length 9.35 ± 0.050 cm and rachis 10.8 ± 0.100 cm. Bristles were the shortest feathers, with an average calamus length of 0.26 ± 0.024 cm, average vane length of 1 ± 0.032 cm and average rachis length of 1.26 ± 0.051 cm. The vane and rachis length was not measured for powder down due to the absence of rachis. As there was no quill present in filoplume, only the feather and barb lengths were measured.

The average length of barbs was measured. The longest feather type i.e. primary contour feathers followed with the longest barbs measured as 1.875 ± 0.123 cm while the barbs of filoplume feathers measured as the shortest with 0.288 ± 0.017 cm.

Feather microstructures

The barbs from the nine different feather types of *A. tristis* were dry-mounted onto slides to observe different microstructures (Table 3) under the microscope that included elongated barbules, distinct nodes, internodes, sub-pennaceous region, villi, prongs, hooklets, ventral teeth, pigmentation and other focused microstructures, elaborated below.

Sub-pennaceous region: The barbs of all the feathers showed the absence of a sub-pennaceous region in both pennaceous and plumulaceous barbules in all feather types.

Table 1. Feather macro-characteristics.

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

Table 2. Feather morphometric measurements.

	Feather type	Feather location	Length (in cm)			
			Calamus ± S.E.	Vane ± S.E.	Rachis ± S.E.	Barb ± S.E.
1	Primary contour feather	Wing	1.45±0.050	9.35±0.050	10.8±0.100	1.875±0.123
2	Secondary contour feather	Wing	1.35±0.050	7.80±0.000	9.25±0.050	1.821±0.111
3	Tail contour feather	Tail	0.8±0.100	7.3±0.100	8.25±0.050	1.637±0.079
4	Body contour	Dorsal	0.2±0.000	3.85±0.050	4.25±0.050	1.391±0.026
5	Body contour	Ventral	0.35±0.050	4.85±0.050	5.25±0.050	1.646±0.043
6	Semiplume	Dorsal	0.35±0.050	3.40±0.100	3.80±0.100	1.532±0.033
7	Semiplume	Ventral	0.45±0.050	4.51±0.395	4.58±0.425	1.901±0.037
8	Semiplume	Tail	0.45±0.050	4.95±0.150	5.50±0.100	1.034±0.024
9	Down	Dorsal	0.25±0.050	3.15±0.050	3.45±0.050	1.415±0.068
10	Down	Ventral	0.3±0.000	3.45±0.050	3.70±0.100	1.604±0.064
11	Down	Tail	0.2±0.000	3.25±0.050	3.45±0.050	1.078±0.057
12	Powder down	Dorsal	0.2±0.000	N/A	N/A	1.2799±0.046
13	Powder down	Ventral	0.25±0.050	N/A	N/A	1.032±0.043
14	Powder down	Tail	0.25±0.050	N/A	N/A	0.765±0.028
15	Bristle	Near Eye and Beak	0.26±0.024	1±0.032	1.26±0.051	0.316±0.008
16	Filoplume	Wings	N/A	N/A	1.94±0.262	0.288±0.017

Villi: Villi are the unique diagnostic microstructural characteristic of passerine birds that extend out from the basal cell of the barbules, only present in the basal cell region of the plumulaceous barbs. The shape of villi was

either knobbed or pointed, but sometimes both were present in the basal cells forming finger-like structures (Image 2A–B).

Nodes and their shape: The barbules of all feathers

had nodes that were swollen, with three different shapes: plain nodes (Image 2C–D), plain pronged nodes (Image 2E–F) and quadrilobed nodes (Image 2G–H). The plumulaceous barbs have all three node types, which were absent in pennaceous barbs. The quadrilobed nodes were mainly present in the proximal region of barbules (Image 2), while the distal region had plain nodes either with prongs or without prongs. These nodes were present in all the different feather types, except in powder down, bristle and filoplume feathers.

Internode shape: The region between two nodes is the internode, which is straight in shape and present in the barbules of plumulaceous barbs (Image 2C–H).

Prongs and their 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 2I–J) and filoplume (Image 3K–L) feathers, elongated and large-sized prongs are present. Prongs were totally absent in powder down feathers.

Hooklets: Distinct hooklets were present in pennaceous barbs of primary contour, secondary contour and tail contour feathers, and were present after the basal cells of the barbules (Image 3M–N). Hooklets were completely absent in all plumulaceous barbs of *A. tristis*.

Ventral teeth: Pennaceous barbs had ventral teeth at the end of basal cells that were less broadened (Image 3O–P).

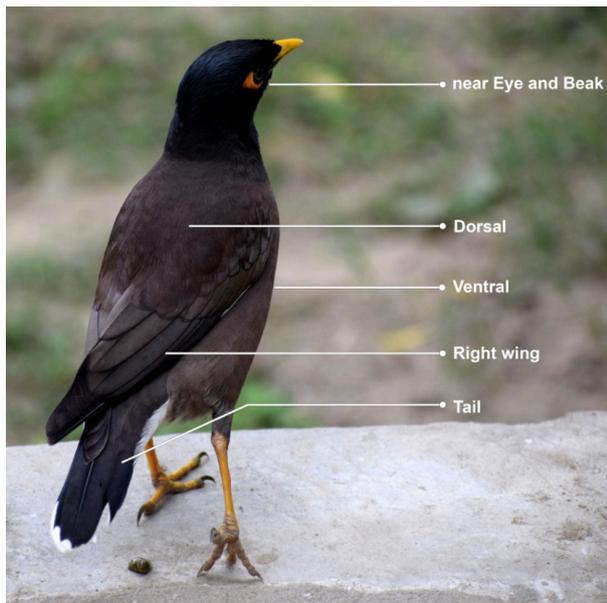


Image 1. Common Myna with locations of feathers sampled. ©Rajesh Kumar.

Table 3. Feather microstructures.

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	0/1	KNB/PNT	0/1	2,3,4	0/1	S/L	0/1	0/1	STR/KNK	6	5	6
2	Tail Contour	1	KNB, PNT	1	2, 3	1	S	1	1	STR	6	5	6
3	Body Contour	1	KNB, PNT	1	2, 3	1	S	1	0	STR	6	5	5, 6
4	Semiplume	1	KNB, PNT	1	2,3,4	1	S	0	0	STR	6,5	5	5,6
5	Down	1	KNB, PNT	1	2,3,4	1	S	0	0	STR	6	5	5, 6
6	Powder Down	1	KNB, PNT	1	3	0	NA	0	0	STR	5,6	5	6
7	Bristle	1	KNB,PNT	1	2	1	L	0	0	STR	6	5	6
8	Filoplume	0	NA	1	2	1	L	0	0	STR	5	5	5, 6

0—Absent | 1—Present | KNB—Knobbed | PNT—Pointed | 2—Plain pronged node | 3—Plain unpronged node | 4—Quadrilobed node | 5—Small | L—Large | STR—Straight | KNK—Kinked | 5—Patchy pigmentation | 6—Dark pigmentation | NA—Not applicable.

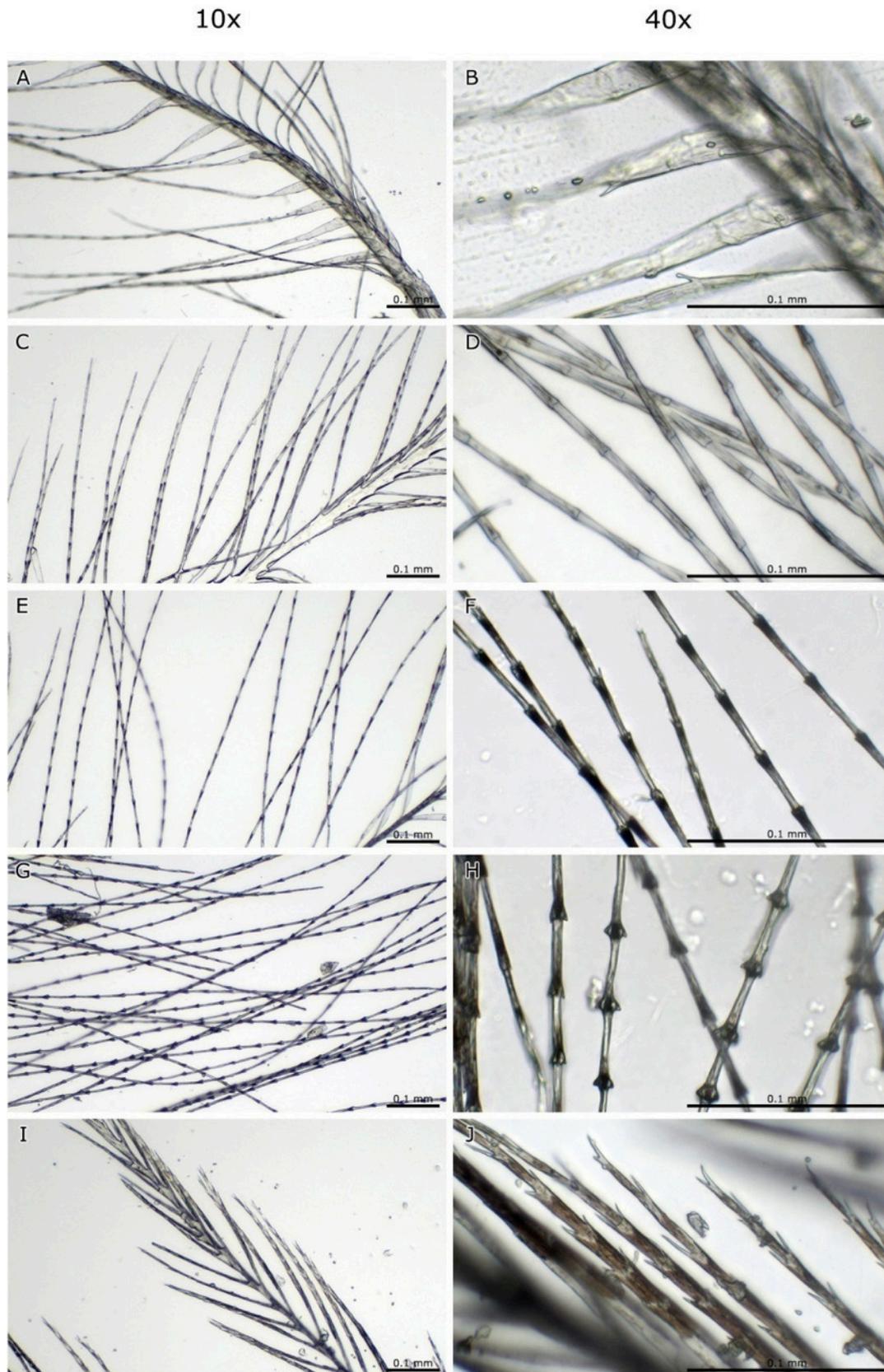


Image 2. Feather microstructures of *A. tristis*. A—Villi at 10X | B—Villi at 40X | C—Plain unpronged nodes at 10X | D—Plain unpronged nodes at 40X | E—Plain pronged nodes at 10X | F—Plain pronged nodes at 40X | G—Quadrilobed nodes at 10X | H—Quadrilobed nodes at 40X | I—Elongated prongs on bristle feathers bars at 10X | J—Elongated prongs at bristle feathers bars at 40X. © Swapna Devi Ray.

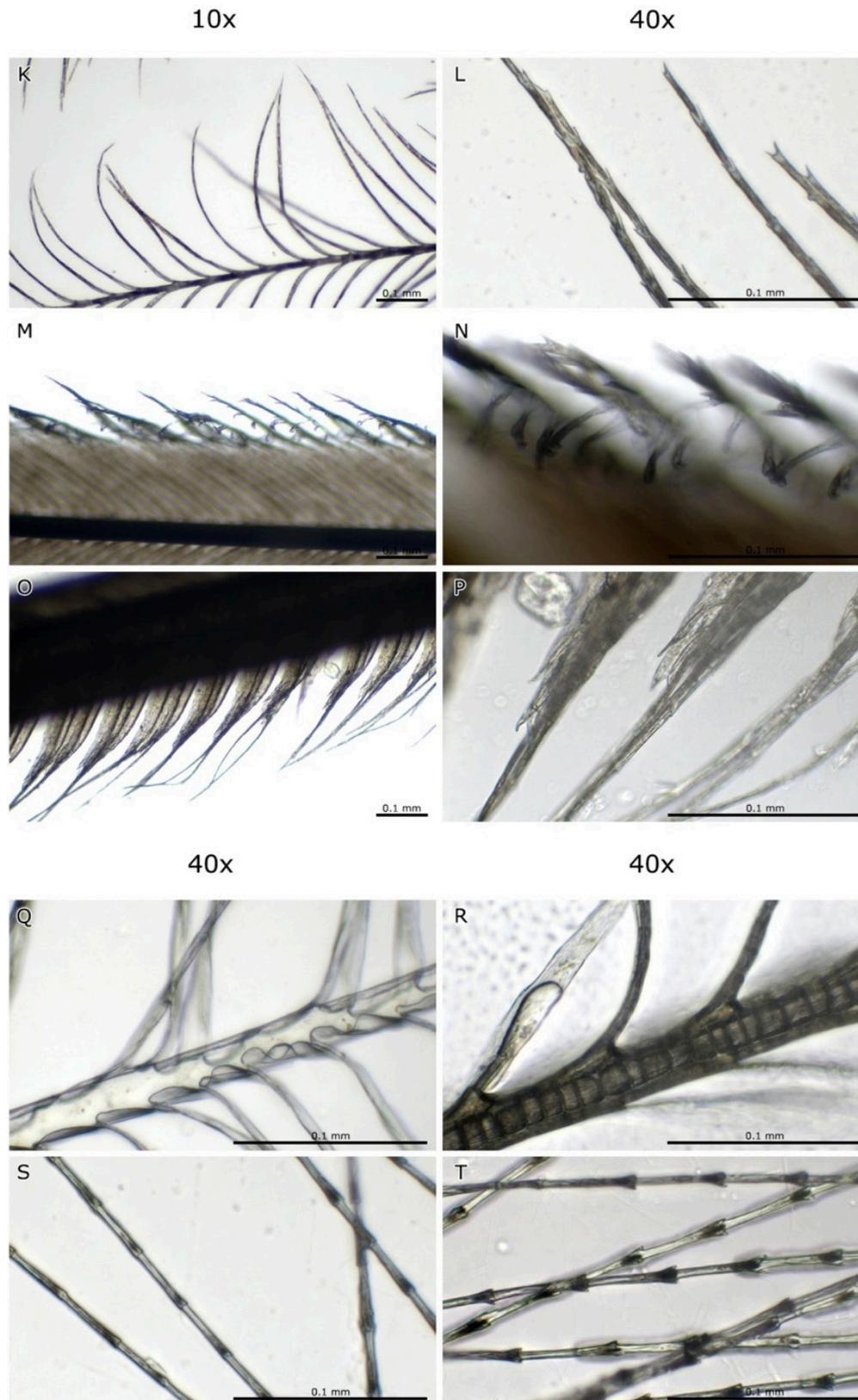


Image 3. Feather microstructures of Common Myna (*A. tristis*): K—Elongated prongs on filoplume feathers at 10X | L—Elongated prongs on filoplume feathers at 40X | M—Hooklets at 10X | N—Hooklets at 40X | O—Ventral teeth at 10X | P—Ventral teeth at 40X | Q—Patchy pigmentation on ramus 40X | R—Dark pigmentation on ramus at 40X | S—Patchy pigmentation on nodes at 40X | T—Dark pigmentation on nodes at 40X. © Swapna Devi Ray.

Pigmentation: Dark pigmentation was mainly present on the nodes where internodes mostly had patchy pigmentation. However, in the semiplume and powder down feathers, nodes had both types of pigmentation (Image 3S–T). Ramus was present with both patchy (Image 3Q) and dark pigmentation (Image 3R).

DISCUSSION

In this study we have documented feather macro-characters, morphometry and microstructures of *A. tristis*. The colour and texture of feathers mainly depends on their location in the body, and also their functional aspects (Ray et al. 2021). According to Chandler (1916), colour is the most important characteristic in species identification, and we observed silver-colored filoplume feathers with pale black pigmentation on the barbs as a specific character of *A. tristis*. It must be noted, however, that it is difficult to retrieve filoplume feathers due to their location and almost transparent nature. Except for the filoplume feathers, we recorded varying colors specific to feather types.

The texture of feathers is known to vary based on their body location and functions, such as flight, thermoregulation, signaling and protection (Lovette & Fitzpatrick 2016). The texture of the feathers of *A. tristis* mainly comprised of three types: rigid, semi-rigid, and soft and fluffy, associated with flight, protection and thermoregulation respectively. While macro characteristics and morphometric measurements tend to vary according to bird age and sex, the measurements are species-specific (Dove 2000; Lee et al. 2015). Data on feather morphometry can also provide clues about physical size (Lee et al. 2015). The present study provides ranges for feather morphometry of *A. tristis* that can be used for these purposes.

Several studies have examined the variation of diagnostic feather features among species, and 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). These studies illustrate that the feather microstructures of a species remain the same irrespective of individual variation (Dove 1997; Lee et al. 2015; Ray et al. 2021). 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 cannot be used as an exclusive character for the

identification of species, while Dove (2000) suggested that pigmentation patterns provide diagnostic clues for determining species groups. From our study of *A. tristis* feathers, we observed that there is no particular uniform pigmentation pattern present in nodes, internodes, and ramus. However, the presence of dark and patchy pigmentation on different shapes of nodes can be used as a micro character for the identification of *A. tristis*. Also from this study we report three microstructures that can be used in the identification of *A. tristis* species: (i) the presence of finger-like villi that are distinctively knobbed and pointed on the border of the basal cells, (ii) the presence of all three types of nodes: quadrilobed, pronged and plain, and (iii) the presence of sharply pointed pronged nodes on bristle and filoplume feathers.

CONCLUSION

Plumology uses feather macro characters, morphometry, and microstructures to aid the identification of order, family and species of birds. During our study we used a systematic approach towards identification of *A. tristis*. Macro-characters including filoplume feathers helped to identify this as a passerine species, while examination of microstructures including finger-like projection of villi, the presence of three node types and the presence of elongated prongs on the nodes of bristle and filoplume feathers were identified as specific to *A. tristis*. This study provides feather morphometry measurements for future reference as a baseline for the identification of *A. tristis* from India.

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