

# **New information about spider variety (*Eriovixiapanigrahis* Archer 1951) (*Arachnidae: Araneidae*) emerged in India**

## **Abstract**

The purpose of this study is to provide the world new information about spider variety. During our survey, we found three female spiders and one male spider belonging to the *Eriovixia* genus under the bushes and shrubs of Kushadangri hill, which is close to Bhawanipatna town in Odisha, India. New information about the new species from genus *Eriovixia* are presented to India and the world. Out of the 33 species of *Eriovixia* that have been identified worldwide, this text describes a species that is new to science: *Eriovixiapanigrahis*, which is named after the author, and also registered in ZooBank.

**Keywords:** New species, *Eriovixiapanigrahis*, spider, ZooBank.

## **1. Introduction**

Spiders are an ancient group of arthropods and one of the most diverse groups, with 52,726 species, and can be found everywhere except Antarctica (World Spider Catalog, 2025). They have been studied as model organisms for ecological, developmental, evolutionary, and behavioural studies (Morehouse et al., 2017; George et al., 2019) because of their high abundance and rich diversity. The identification of this group based on morphology presents significant challenges and requires considerable time, primarily due to factors such as sexual dimorphism, polymorphism, and the absence of identification keys for juveniles (Coddington & Levi, 1991; Magalhaes et al., 2017). In light of these challenges, employing supplementary methods such as molecular analysis is crucial, as it can facilitate swift species identification and clarify taxonomic ambiguities (Tyagi et al., 2019).

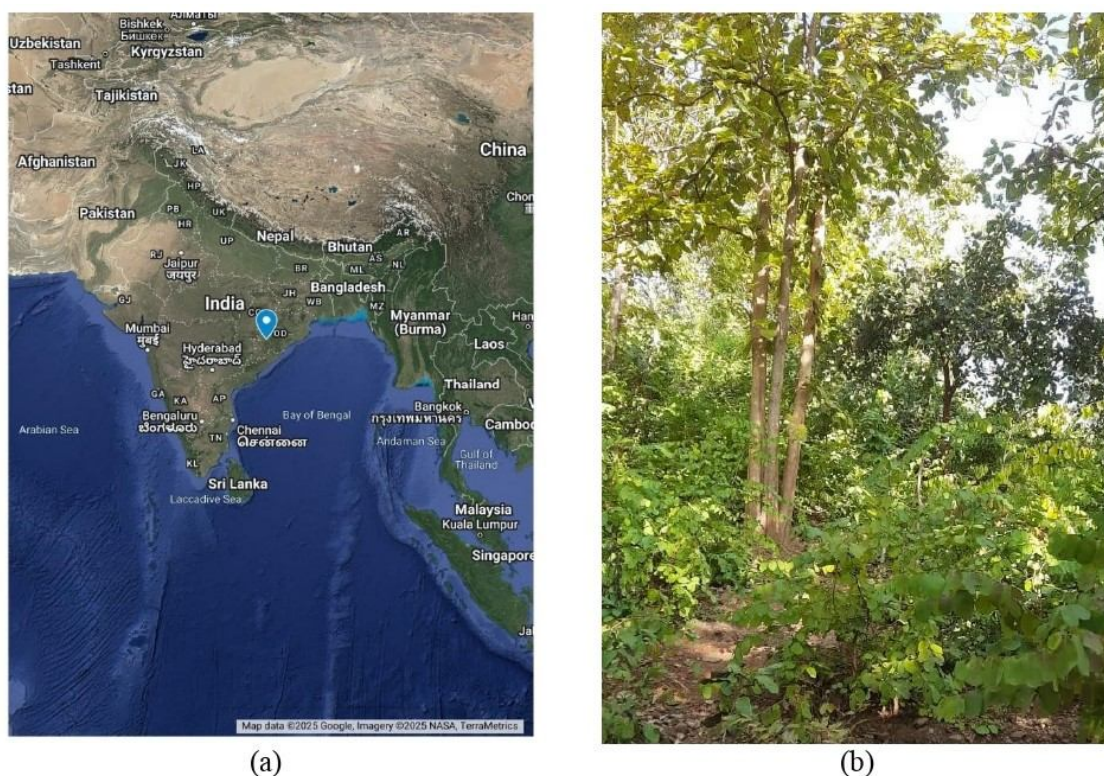
In this study, we employed molecular identification through the sequencing of a portion of the mitochondrial Cytochrome c Oxidase subunit I (COI) gene. There are 33 documented species of *Eriovixia* (Archer, 1951) globally (World Spider Catalog, 2025), and this text presents a description of one species that is new to science.

## **2. Material and Methods**

### **2.1 Study Area**

Spiders were collected from different sites of Kushadangri Hill, which is located near Bhawanipatna town, the headquarters of the Kalahandi District, Odisha, India, and situated at

19°54'43.7"N 83°11'27.3"E (Fig. 1). The topography of Kalahandi consists of plain land, hills and mountains. The climate of this district is quite extreme dry except during monsoon. The average annual rainfall of 1,378.2 mm was recorded in this area.



**Fig. 1** Sample collection site (a) Kushadangri Hill near Bhawanipatna town is located in Kalahandi District, Odisha, Eastern India, (Map was developed by Google My Maps, 2025) (b) The site of collections are bushes, and lower regions of trees.

## 2.2 Sample Collection and Identification

Using a regular method of collection, a spider collecting survey was conducted between 6:00 am to 9:00 am and from 4:00 pm to 8:00 pm in the evening during August to December, 2024. Classification of the collected specimens had been carried out by the help of some pertinent literature (Siliwal, 2005; Sebastian & Peter, 2009) as well as online sources (World Spider Catalog, 2025; and Indian Biodiversity Portal). We used molecular identification since those specimens were difficult and confused to identify up to the species level. A stereo zoom binocular microscope was used for photography that was obtained. Some specimens were kept in 70% alcohol and deposited in the animal museum of the Department of Zoology, MaaManikeshwari University.

## 2.3 Molecular Study

### 2.3.1 DNA extraction and PCR

DNA extraction was carried out from single captured unidentified male spider using 'PureLink™ Genomic DNA Mini Kit, following the Kit protocol with some modification, like sample was homogenized in 100 µl of DNA Extraction Solution and incubated for 20 minutes at room temperature. The sample was added in the beaded vial containing XPLOREGEN gDNA Extraction Buffer™1. The homogenate was then vortexed at maximum speed for 10 minutes, followed by 300 µl of Xploregen gDNA Extraction Buffer™ 2 and horizontal vortexed the vial at maximum speed for 7 minutes. The homogenate was then centrifuged at 10,000 rpm for 3 minutes at room temperature. 800 µl of supernatant was transferred to a sterile 2 ml vial and 200 µl of Xploregen gDNA Extraction Buffer™ 3 solution was mixed by vortex for 5 seconds followed by centrifuge at 10,000 rpm for 2 minutes. Then the DNA was processed by using Xploregen gDNA Extraction Buffer™ 4, 5, 6. Lastly 30 µl of Xploregen gDNA Extraction Buffer™ 7 (Elution 1) was added to the center area of the spin column and centrifuged for 5 minutes at 10,000 rpm. After discarding the spin column and elution tubes were stored for further processing.

### 2.3.2 Genetic analysis

The PCR reaction was carried out in a volume of 25 µl containing 10 µl of 10X Taq polymerase buffer with 3.2 mM MgCl<sub>2</sub>, 2.5 mM of dNTPs (2.5 mM each), 1 µl units of TaqDNA Polymerase Enzyme (3U/ml), 2 µl nM of each primer, and 1.5 µl of the template DNA. The COI primers COI forward GGTCAACAAATCATAAAGATATTGG and COI reverse TAAACTTCAGGGTGACCAAAAAATCA were used to identify the spider species. The thermal profile used to amplify COI region from spider species was initial denaturation at 94°C for 3 minutes followed by 30 cycles of denaturation at 94°C for 1 minutes, annealing at 50°C for 1 minutes, extension at 72°C for 2 minutes and final extension at 72°C for 7 minutes. The amplicons were visualized in a 1.2% agarose gel stained with ethidium bromide on a UV transilluminator. Bi-directional DNA sequencing reaction of PCR-amplified DNA was carried out with LCO 1490 & chelicerate reverse 2 primers using BigDye™ Terminator v3.1 Cycle Sequencing Kit on ABI 3130 Genetic Analyzer. The sequences were aligned with Clustal W (Thompson JD et al. 1994), and then edited using the BioEdit sequence analysis tool (Hall TA. 1999).

### 2.3.3 Bioinformatics Study

The consensus sequence was generated for each sample using BioEdit version 6.0.7 and were searched over the GenBank database using Basic Local Alignment Search Tool (BLAST) against the spider genomes previously present in GenBank (NCBI WEB SITE) (Table 1). The complete sequences were deposited in GenBank with accession number PQ846871.1. The mtCOI sequence of spider (our isolated spider strain KSP04) were compared with the whole world samples of gene COI of spider species by Maximum likelihood method (Table 1) by using Multiple Sequence Alignment (MSA) based on the sequences available in NCBI GenBank. In this study, we placed the species having more than 88% similarity with our discovered species.

## 3. Results

As seen in Fig. 1(b), three female spiders and one male spider were captured beneath the bushes and shrubs during our survey. We verified that these were members of the genus *Eriovixia* based on the identification key; nonetheless, they differed from other species of the genus, making it challenging to determine their exact taxa. As a result, we used the COI sequencing for molecular identification, concentrating on unidentified caught specimens (*Eriovixia* species) and conducting additional taxonomic research on them. Further some spiders were identified as *Eriovixialaglaizei*, collected from same site during our study.

**Table 1: Blast results of *Eriovixia* species**

Sl. No.	Spider Name	Accession Number	% of similarity
1	Eriovixiapoonaensis isolate ADB027 cytochrome oxidase subunit I (COI) gene.	KT383689.1	96.41%
2	Eriovixiapoonaensis isolate ADB087 cytochrome oxidase subunit I (COI) gene.	KT383762.1	96.41%
3	Eriovixialaglaizei isolate 11 cytochrome c oxidase subunit I (COX1) gene.	OQ821698.1	96.17%
4	Eriovixiapoonaensis voucher ARAMP067 cytochrome c oxidase subunit I (COX1) gene.	PQ651558.1	96.07%
5	Eriovixiapoonaensis voucher AA_527 cytochrome c oxidase subunit I gene.	MK392775.1	96.03%
6	Eriovixiapoonaensis voucher AA_703 cytochrome c oxidase subunit I gene.	MK392777.1	95.81%
7	Eriovixiapoonaensis voucher AA_965 cytochrome c oxidase subunit I gene.	MK392776.1	95.81%
8	Eriovixialaglaizei voucher 196 cytochrome c oxidase subunit I (COI) gene.	MK420106.1	95.79%
9	Eustala sp. isolate CARBII047-18 cytochrome c oxidase subunit I (COX1) gene.	OR892997.1	88.49%
10	Eustala sp. isolate CARBII066-18 cytochrome c oxidase subunit I (COX1) gene.	OR893011.1	88.33%

Following receipt of the BLAST report from the NCBI site, there was strong confirmation that the specimen, using morphological keys, was indeed *Eriovixia* species. According to phylogenetic analysis and nucleotide homology, the entire BLAST result, which is displayed in Table 1, indicated a high degree of similarity with the top. Three species, two from *Eriovixia* and one from *Eustala*, exhibit more than 88% identity with our isolated spider strain KSP04.

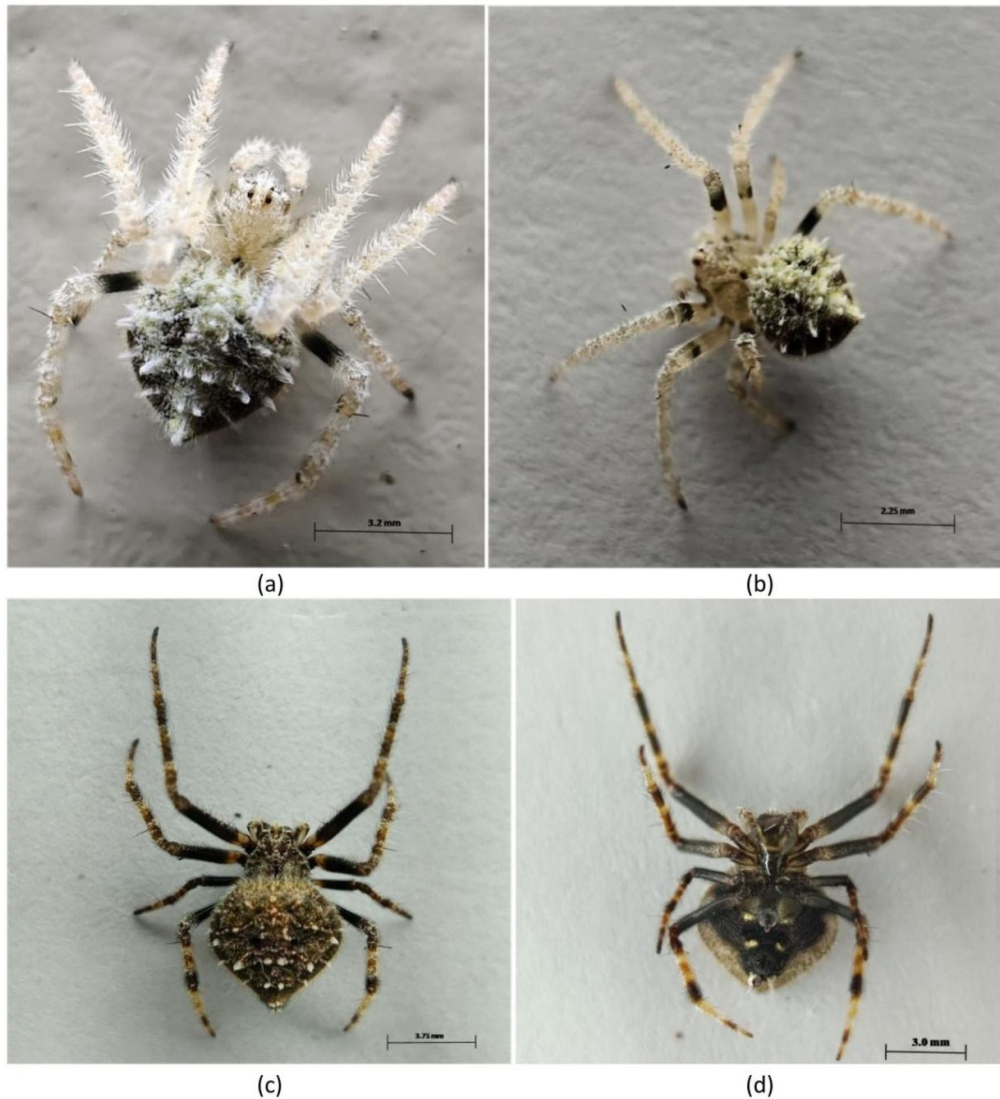
#### 4. Discussion

Molecular identification is more accurate than morphological features, particularly when it comes to spider identification. It has been confirmed that the captured unidentified spider may belong to the genus *Eriovixia* based on certain common characteristics, such as a small, triangular body, a small cephalothorax, and a slightly shiny, colour-patterned abdomen that resembles like leaves.

Orb-shaped webs, which are typical web characteristics of the genus *Eriovixia*, were formed by this caught spider in vegetation like bushes and shrubs. After six o'clock in the evening, these spiders were caught. The members of this genus are nocturnal and typically avoid daylight. Even yet, it was more difficult to identify the species (our interest of male spider) because of some morphological characteristics, such as the subtriangular's spiny abdomen, pear-shaped carapace, and lack of caudal appendage, as seen in fig. 2(a) and (b).



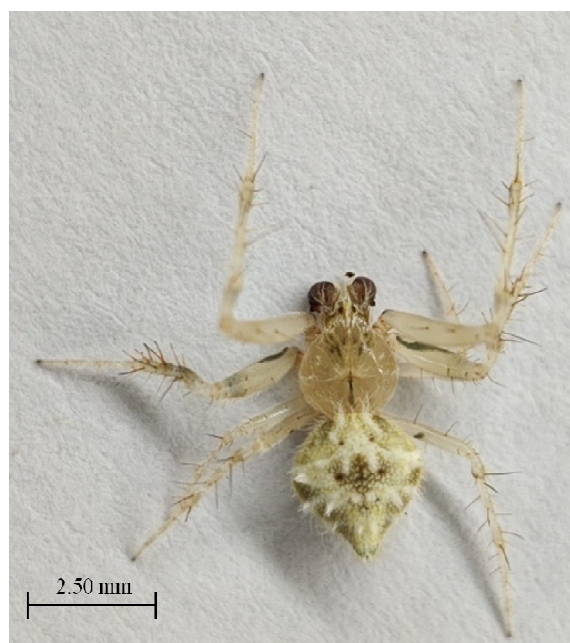
Additionally, the male of this species was almost as large as the female of *Eriovixialaglaizei*, with a white, slightly translucent cephalothorax, leg-pale translucent with black-coloured bands, and a mixed white and grey abdomen (Fig. 2(a) & (b)). Males had a bulbous, strongly textured abdomen, while females had lighter abdomens, which resulted in a rough look with darker or moss-like areas (Fig. 2). Both sexes had hairs and spines on their legs (Fig. 2).



**Figure 2**(a) & (b) Male *Eriovixiapanigrahis*; (c) & (d) Female *Eriovixiapanigrahis*

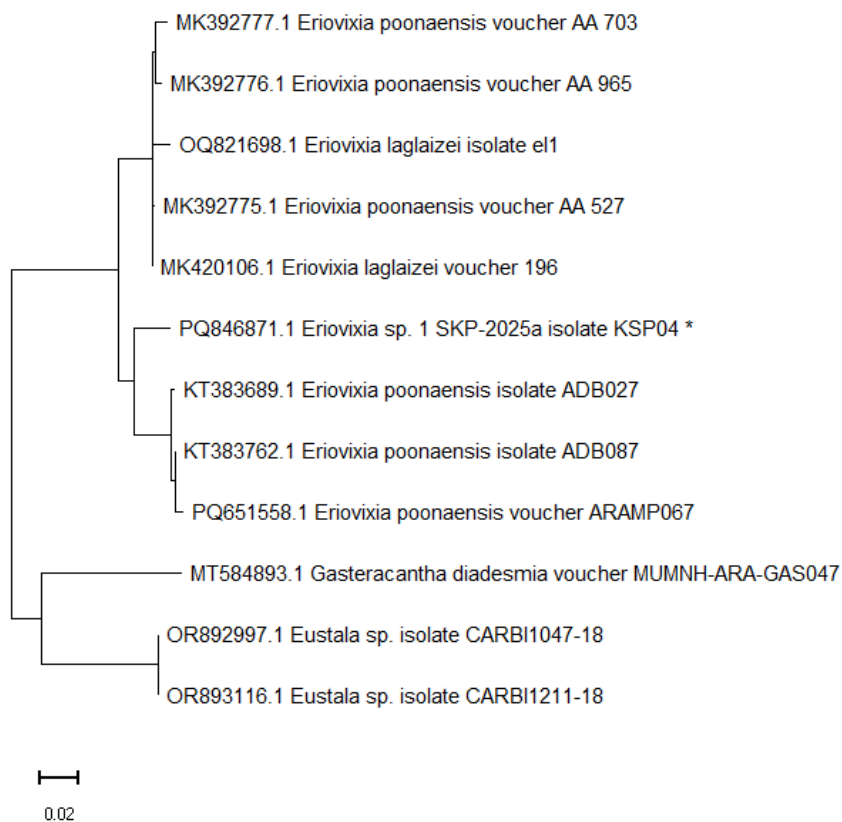
For more conformation and accuracy, we went for molecular identification by sequencing of part of the mitochondrial (mt) Cytochrome c Oxidase subunit I (COI) gene. It has been widely used in the last decade in biodiversity research for accurate species identification (Naseem et al., 2018); it can be integrated in the phylogenetic studies also (Wheeler et al., 2017). The complete sequences were deposited in GenBank with our isolated strain KSP04, and we got the comparative data (BLAST Result) with the whole world samples (the 10 most likely samples) by the maximum likelihood method as shown in Table 1. It showed more similarity with *Eriovixiapoonaensis* (96.41%) and less similarity (88.33%) with

*Eustalas* species. So from molecular analysis it was confirmed that the male captured spider was belonged to genus *Eriovixia*. It exhibited 96.41% similarity to *E. poonaensis*; however, *E. poonaensis* differs from our captured spider (*Eriovixia* sp.) based on common characteristics such as (♀) approximately 5-8 mm in length; (♂) smaller, usually 3-5 mm in length; carapace 2.50 mm long & 2 mm wide; abdomen 4 mm long & 4 mm wide; cephalothorax brownish, leg yellow with black band, and abdomen-yellowish; the ventral side should have a large black patch shaped like a star, and one pair of white spots between the epigastric furrow and the spinnerets (Tikader, B, 1981). Therefore, we verified that the male spider that was collected was not *E. poonaensis*.



**Figure 3** Male *Eriovixia laglaizei*

Next to *E. poonaensis*, the our species of interest showed 96.17% similarity with *E. laglaizei*, but it morphologically differ having the characters like (♀) approximately 8.50 mm in length; (♂) smaller typically around 5.0 mm in length; carapace-3.10 mm long & 2.60 mm wide; abdomen-6.20 mm long & 4.85 mm wide, caudal appendages seen, cephalothorax-yellowish, leg- yellowish black, abdomen- greyish yellow with 12 white spots (Fig. 3). The epigyne is dark brown in colour, below that dark yellow patches are present. The palps does not have the bands (Fig. 3). Morphologically *E. laglaizei* are larger in size as compare to *E. poonaensis* and the colour of *E. laglaizei* is variable with small hairs on the abdomen.



**Figure 4** Phylogenetic Tree of our interest of spider species

On the basis of phylogeny analysis, it was confirmed that *E. poonaensis* and *E. laglaizei* are genetically more similar rather than morphologically because they were present in the same clade, whereas our species of interest (*Eriovixia* sp.) and *E. poonaensis* were originated from the same ancestor, but our *Eriovixia* species was different from both *E. poonaensis* and *E. laglaizei* (Fig. 4). So our interest of *Eriovixia* species was a newly discovered spider species under the genus *Eriovixia*, which is new to science. As per the surname of the discoverer (Dr. S K Panigrahi), the new spider species was named as “*panigrahis*”, and registered in ZooBank with LSID: [zoobank.org/pub:C1E55439-EA9F-44BD-825A-68185075941F](https://zoobank.org/pub:C1E55439-EA9F-44BD-825A-68185075941F).

## 5. Conclusion

New information about the new species from genus *Eriovixia* are presented to India and the world. There are 33 documented species of *Eriovixia* globally, and this text presents a description of another one species that is new to science, is named based on the discoverer name as *Eriovixia panigrahis*.

**Disclaimer (Artificial intelligence)**

All authors of this manuscript hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

## 6. References

1. Coddington, J. A., & Levi, H. W. (1991). Systematics and evolution of spiders (Araneae). *Annual review of ecology and systematics*, 565-592.
2. Google. [Google Maps directions to Kushadangri Hill near Bhawanipatna]. Retrieved January 21, 2025, from [https://www.google.com/maps/d/u/0/edit?mid=1\\_jCGXd2lrsUrcoetnpnRb7VSOCdHeQY&ll=19.912527064514045%2C83.19101190000002&z=18](https://www.google.com/maps/d/u/0/edit?mid=1_jCGXd2lrsUrcoetnpnRb7VSOCdHeQY&ll=19.912527064514045%2C83.19101190000002&z=18)
3. Hall, T. A. (1999, January). BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. In *Nucleic acids symposium series* (Vol. 41, No. 41, pp. 95-98).
4. Hilbrant, M., Damen, W. G., & McGregor, A. P. (2012). Evolutionary crossroads in developmental biology: the spider Parasteatodepidariorum. *Development*, 139(15), 2655-2662.
5. Magalhaes, I. L., Martins, P. H., Nogueira, A. A., & Santos, A. J. (2017). Finding hot singles: matching males to females in dimorphic spiders (Araneidae: Micrathena) using phylogenetic placement and DNA barcoding. *Invertebrate Systematics*, 31(1), 8-36.
6. Morehouse, N. I., Buschbeck, E. K., Zurek, D. B., Steck, M., & Porter, M. L. (2017). Molecular evolution of spider vision: new opportunities, familiar players. *The Biological Bulletin*, 233(1), 21-38.
7. Naseem, S., & Tahir, H. M. (2018). Use of mitochondrial COI gene for the identification of family Salticidae and Lycosidae of spiders. *Mitochondrial DNA Part A*, 29(1), 96-101.
8. Siliwal, M., Molur, S., & Biswas, B. K. (2005). Indian spiders (Arachnida: Araneae): updated checklist 2005. *ZOOSPRINT JOURNAL*, 20(10), 1999-2049.
9. Tahir, H. M., Summer, M., Mehmood, S., Ashraf, S., & Naseem, S. (2019). DNA barcoding of spiders from agricultural fields. *Mitochondrial DNA Part B*, 4(2), 4144-4151.
10. Thompson, J. D., Higgins, D. G., & Gibson, T. J. (1994). CLUSTAL W: improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position-specific gap penalties and weight matrix choice. *Nucleic acids research*, 22(22), 4673-4680.
11. Tikader, B. K. (1981). Studies on some orb-weaving spiders of the genera Neoscona Simon and AraneusClerck of the family Araneidae (= Argiopidae) from India. *Rec. zool. Survey, India, Occ. Pap.*, 24, 1-60.
12. Tyagi, K., Kumar, V., Kundu, S., Pakrashi, A., Prasad, P., Caleb, J. T., & Chandra, K. (2019). Identification of Indian spiders through DNA barcoding: cryptic species and species complex. *Scientific Reports*, 9(1), 14033.



13. Uetz, G. W., Clark, D. L., & Roberts, J. A. (2016). Multimodal communication in wolf spiders (Lycosidae)—an emerging model for study. In *Advances in the Study of Behavior* (Vol. 48, pp. 117-159). Academic Press.
14. Wheeler, W. C., Coddington, J. A., Crowley, L. M., Dimitrov, D., Goloboff, P. A., Griswold, C. E., ... & Zhang, J. (2017). The spider tree of life: phylogeny of Araneae based on target□gene analyses from an extensive taxon sampling. *Cladistics*, 33(6), 574-616.
15. World Spider Catalog (2025). World Spider Catalog. Version 25.5. Natural History Museum Bern, online at <http://wsc.nmbe.ch>, accessed on 20.01.2025. doi: 10.24436/2
16. Sebastian, P. A., & Peter, K. V. (Eds.). (2009). *Spiders of India*. Universities press.
17. Indian Biodiversity Portal, <https://indiabiodiversity.org/>