Confirmation of Naja oxiana in Himachal Pradesh, India

VISHAL SANTRA^{1,2*}, JOHN BENJAMIN OWENS², STUART GRAHAM³, WOLFGANG WÜSTER³, SOURISH KUTTALAM³, OMESH BHARTI⁴, MELVIN SELVAN⁵, NILANJAN MUKHERJEE¹ & ANITA MALHOTRA³

¹Simultala Conservationists (Foundation for Wildlife), Nalikul, Hooghly, West Bengal, India
²Captive & Field Herpetology, Wales, UK
³MEFGL School of Natural Sciences, Bangor University, Wales, UK
⁴State Institute of Health & Family Welfare, Shimla, Himachal Pradesh, India
⁵Endangered Wildlife Trust, Dingdudal, Tamil Nadu, India
*Corresponding author e-mail: vscaptiveandfieldherpetology@gmail.com

.....

The Central Asian cobra *Naja oxiana* (Eichwald, 1831), also called the Caspian, Oxus, or Russian cobra, is a species of venomous snake in the family Elapidae. *Naja oxiana* exhibits considerable ontogenetic variation in its pattern; however, juveniles tend to be pale, with a faded appearance.

Juveniles also have noticeable dark and light cross-bands of approximately equal width around the body. Adults are completely light to chocolate brown or yellowish, with some specimens retaining traces of juvenile banding, especially the first few dark ventral bands (Wüster, 1998).

Naja oxiana was originally considered a subspecies of the Indian, spectacled, Asian or binocellate cobra Naja naja (Wüster & Thorpe, 1992) and little was known about its distribution in India (Mahajan & Agrawal, 1976). Whilst N. *naja* is confidently recorded throughout the most of India, with the exception of the north-eastern states, the distribution of N. oxiana is still in need of confirmation (Whitaker & Captain, 2008). The older literature frequently used the names Naja n. oxiana or N. naja for several species of Asiatic cobras prior to their reclassification (Wüster, 1998), and included north-west India for the distribution of *N. oxiana*, but with no specific locations (Biswas & Sanyal, 1977). The frequent absence of hood marks in many N. naja from northern India often led to these specimens being misidentified as N. oxiana, further adding to the confusion (Wüster, 1998). In Wüster & Thorpe's (1992) review of published records of N. oxiana from northern India, the only records considered reliable were from Jammu (Murthy & Sharma, 1976; Murthy et al., 1979), and from Solan District, Himachal Pradesh (Mahajan & Agrawal, 1976). Reports from elsewhere in India appear to be due to misidentification of the melanistic morph of N. naja (Saikia et al., 2007). The status of the species in India has been all the more mysterious since there have been no confirmed records from India since the 1970s.

Here, we report new observations of *N. oxiana* (Fig. 1) from Himachal Pradesh in 2017 and 2018. Two live *N. oxiana* specimens were encountered in the area surrounding Majoga, in the district of Chamba (Fig. 2) during a research expedition undertaken as part of the Bangor University Biodiversity Informatics and Technology Exchange for the Management of Snakebite (BITES) project in 2017. Two further *N. oxiana* specimens were encountered between June and September 2018 during a collaborative research expedition by BITES,



Figure 1. Female N. oxiana displaying juvenile hood markings

Simultala Conservationists (Foundation for Wildlife) and Captive & Field Herpetology teams. All individuals were found on grassy / rocky slopes with sparse mixed alpine shrub and tree cover with occasional large rocks, boulders and rock faces, or on agricultural land and pasture. The habitat in which *Naja naja* can be found in includes agricultural fields around villages at lower elevations (<900 m), up into pine forests. This habitat differs from *N. oxiana* which appears to frequent higher altitude and more open, rocky habitats, which consist of hemp (*Cannabis sativa*) cover along with some, less dense, pine/tree cover.

Identification was confirmed using morphological characters from Wüster (1998) and phylogenetic analysis. Morphological data are provided in Table 1. Interestingly, the ventral and subcaudal scale counts of the new Himachal Pradesh specimens are slightly below the range previously reported for *N. oxiana* (ventral scales 199 - 207 in males and 191 - 210 in females, subcaudal scales 66 - 71 in males and 62 - 70 in females), and within the range of corresponding counts in *N. naja*. However, the low dorsal scale row counts around the neck and the pattern are characteristic of *N. oxiana*.

For 2017 samples, the evolutionary history was inferred using the Maximum Likelihood method based on the Tamura 3-parameter model (Tamura, 1992) in MEGA7 (Kumar, 2016). The analysis involved 20 nucleotide sequences comprising 421 base pairs of 16S large subunit ribosomal DNA,

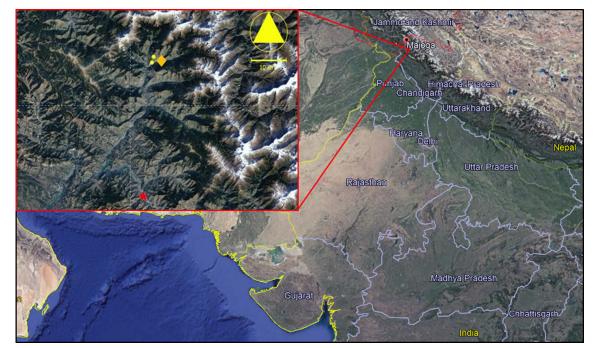


Figure 2. Naja oxiana Indian distribution (Yellow dots = N. oxiana observation locations, red star = Chamba, orange diamond = Majoga,)

Species	Naja oxiana	Naja oxiana	Naja oxiana	Naja oxiana
Reference number	17.V18	17.V19	18.16	18.51
Date	21.09.2017	22.09.2017	11.07.2018	16.08.2018
Time (GMT + 5.5 h)	Not recorded	Not recorded	21:56	22:13
Altitude (m a.s.l.)	1900	2100	2021	1565
Morphology				
Sex	Female	Female	Female	Female
Snout-vent length (cm)	99	98	91	106
Tail length (cm)	14	18.5	18.3	21.5
Visible neck banding	Present	Absent	Present	Absent
Hood markings	Present	Absent	Present	Absent
Cuneate scale	Absent (The left 3rd and 4th infralabial scales were damaged)	1 cuneate scale on the left side between 3rd and 4th infralabials	1 cuneate scale between 3rd and 4th infralabial on both sides	Not known
Scalation				
Scale rows at mid-body	21	21	21	21
Scale rows around neck	23	23	23	22
Scale rows before vent (one head length ahead of vent)	16	15	15	15
Ventrals ¹	189	190	189	189
Subcaudals (all paired)	37 (Tail tip missing)	57	60	61
First V of first dark band (ventral scale no.)	6th	6th	6th	5th
Last V of first dark band (ventral scale no.)	8th	9th	9th	8th

Table 1.	Sample data and	l morphological	I characteristics of N	<i>. oxiana</i> from Hima	chal Pradesh, India
----------	-----------------	-----------------	------------------------	---------------------------	---------------------

¹ Recorded using the Dowling method (Dowling, 1950)

including three sequences of *N. oxiana* and one of *N. naja* generated specifically for this study. Other sequences were downloaded from GenBank. All positions with less than 90 % site coverage were eliminated. The tree with the highest log likelihood (-1114.13) is shown in Figure 3. A discrete Gamma

distribution was used to model evolutionary rate differences among sites (5 categories, parameter = 0.1132). All novel sequences used in this study are deposited on GenBank, with the accession numbers: MN548728-30 and MN648780.

These observations confirm the continued presence of

N. oxiana in northern India, and represent a species range extension of approximately 120 km east-north-east and 220 km north-west from the two previous Indian records from Jammu and Solan District in Himachal Pradesh, respectively, and an elevation increase of 100 m from 2000 m a.s.l. (Wüster, 1998) to 2100 m a.s.l.

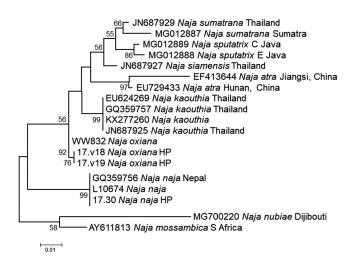


Figure 3. *Naja oxiana* molecular phylogenetic analysis by Maximum Likelihood method. The percentage of trees in which the associated taxa clustered together is shown next to the branches (only when >50 %). Tips are labelled with associated GenBank Accession numbers. The tree is drawn to scale, with branch lengths measured in the number of substituti ons per site.

ACKNOWLEDGEMENTS

We would like to thank the Forest Department of Himachal Pradesh for permission to carry out the survey under permit number FFE-FB-F(10)-3/2017. Funding for this work was provided by the European Union Seventh Framework Programme (under the grant agreement number PIRSES-GA-2013-612131) to the BITES consortium in 2017, and a small grant from The Rufford Foundation to Vishal Santra (Offer Letter: 25313-1) in 2018. We would also like to extend our thanks to Anatoli Togridou, Richard Southworth, Jasmine Torrez, Molla Talhauddin Ahmed, Sankha Suvra Nandy, Shyamal Kumar Ghosh and Anweshan Patra for volunteering their help during fieldwork in 2018.

REFERENCES

- Biswas, S. & Sanyal, D.P. (1977). Fauna of Rajasthan, India, Reptilia. *Records of the Zoological Survey of India* 73: 247-269.
- Dowling, H.G. (1951). A proposed standard system of counting ventrals in snakes. *British Journal of Herpetology* 1: 97-99.
- Eichwald, E. (1831). Zoologia Specialis Quam Expositis Animalibus tum Vivis, tum Fossilibus Potisimum Rossiae in Universum, et Poloniae in Species, In Usum Lectionum Publicarum in Universitate Caesareae Vilnensi Habendarum. Part 3, Josephi Zawadski, Vilnius. 314 pp.
- Kumar S., Stecher, G. & Tamura, K. (2016). MEGA7: Molecular evolutionary genetics analysis version 7.0 for bigger datasets. *Molecular Biology and Evolution* 33:1870-1874.
- Mahajan, K.K. & Agrawal, H.P. (1976). Report on a collection of reptile fauna from Simla Hills, Himachal Pradesh. *Zoological Survey of India* 71: 301-312.
- Murthy, T.S.N. & Sharma, B.D. (1976). A contribution to the herpetology of Jammu and Kashmir. *British Journal of Herpetology* 5: 533-538.
- Murthy, T.S.N., Sharma, B.D. & Sharma, T. (1979). Second report on the herpetofauna of Jammu and Kashmir, India. *Snake* 11: 234-241.
- Saikia, U., Sharma, D.K., & Sharma, R.M. 2007. Checklist of the reptilian fauna of Himachal Pradesh, India. *Reptile Report* 8: 6-9.
- Tamura, K. (1992). Estimation of the number of nucleotide substitutions when there are strong transitiontransversion and G + C-content biases. *Molecular Biology* and Evolution 9: 678-687.
- Whitaker, R. & Captain, A. (2008). *Snakes of India: The Field Guide*. Draco Books, Chennai, India. 385 pp.
- Wüster, W. & Thorpe, R.S. (1992). Asiatic cobras: Population systematics of the *Naja naja* species complex (Serpentes: Elapidae) in India and central Asia. *Herpetologica* 48: 69-85.
- Wüster, W. (1998). The cobras of the genus *Naja* in India. *Hamadryad* 23: 15-32.

Accepted: 11 November 2019