

STUDY OF DISTRIBUTION BEHAVIOR AND VENOM VARIATION AMONG SNAKES IN KOTA, RAJASTHAN

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Abstract

Covering approximately 342,239 km², Rajasthan is the largest state of India by area, and hosts an exceptionally diverse, yet relatively poorly understood, snake fauna shaped by its heterogeneous ecological landscape that encompasses arid/thinly vegetated desert scrub/grassland through to riparian floodplain. Gigantic snakes like the reticulated python (*Malayopython reticulatus*) are observed here. Highly urbanized regions with strong human habitation like the Kota district, in the Hadoti region along the Chambal River corridor, describe a more complex habitat matrix supporting high species diversity, including a number of medically-important snakes. Using a combination of field-based data, peer-reviewed literature, public institutional databases and government biodiversity reports, this paper investigates spatial distribution patterns, biological and behavioral ecology and venom variability among snake species documented in and around Kota, Rajasthan. Around 25 species of snakes have been recorded in the myriad of micro-habitats across the District, including scrublands, fields, riverine habitats and rocky plateaus. Of these, the four medically important species *Naja naja* (Indian cobra), *Bungarus caeruleus* (common krait), *Daboia russelii* (Russell's viper), and *Echiscarinatus* (saw-scaled viper) show remarkable interspecific variation in ecology, patterns of behavior, and venom composition. Natural historical studies based on known toxicology literature on specific components of venoms demonstrates venom composition can differ broadly between species, and this variation can also occur within a species and can result from ecological selective pressures and/or geographic isolation. This study emphasizes the need for regional herpetological surveys, antivenom optimization, and biodiversity conservation plans for the Kota region. The implications of the findings are a significant aspect of public health management and wildlife conservation policy in Rajasthan.

Keywords: *Ophidian diversity*¹, *Kota Rajasthan*², *snake venom variation*³, *herpetofauna*⁴, *snakebite envenomation*⁵.

1. Introduction

Snakes represent a major evolutionary lineage within the squamate reptiles that is ecologically and biomedically important as both predator and prey in terrestrial and aquatic food webs. India is one of the mega-biodiversity nations of the world, with about 300 species of snakes, of which nearly 60 are venomous and pose an unavoidable threat to human beings (Whitaker & Captain, 2004). Conservatively estimated to be well over 46,000 snakebite deaths per year, the country carries a disproportionate burden of global snakebite mortality,

highlighting a major public health crisis of venomous ophidians in the subcontinent (Suraweera et al., 2020). India's biggest state by area is one of its more heterogeneous and challenging from a research perspective: the Thar Desert in northwest Rajasthan, the Aravalli hill ranges in the central zone, and the fertile Hadoti plateau in the southeast all present different questions of range: which will come to very much the same thing, in that Rajasthani landscape is too big to get your head round. That ecological diversity allows different micro-habitats to be available, making for a high variety of snake species. The Kota district, placed in the south-eastern part of Rajasthan, the land of confluence between the Chambal and Kalisindh rivers, is particularly significant due to its ecological transition between desert and semi-humid zones. The Chambal River in India is a relatively undisturbed river system that sustains a rich riparian biodiversity corridor sustaining a diverse assemblage of semi-aquatic and terrestrial snakes.

While the Kota region is known for its ecological and public health importance, previously systematic documentation of snake diversity, behavioral ecology as well as venom characterization, however remain scattered. Much published literature on Indian herpetofauna is generic at national or regional scale with conspicuous absence of district level studies. Such a gap hampers formulation of local region specific antivenom strategies, wildlife management plans or snake bite treatment regimes relevant to the specific epidemiological situation of Hadoti. The objective of this study were as follow: (1) samples to document its notes, distribution and diversity in the Kota district based on field surveys and secondary records, (2) a characterizing habitat preferences and behavioral ecology of (i.e., several key species representative of land-use types), and (3) to review both the venom biochemistry and the toxicological profiles of medically important species with special focus on intraspecific variation across geographic populations. This study is based on three research questions about diversity and geographical distribution of snakes in the Kota district: Q1. What are the behavioral patterns of focal species across habitats and seasons? Question 1: How much does venom composition differ among the four medically-important species and what ecological factors drive this amount of variation.

Behavioral Patterns of Snakes

The ecological association between snake diversity and habitat architecture has been widely explored throughout the Indian subcontinent. Whitaker and Captain (2004) is the most complete field identification guide available for the snakes of India, including descriptions of habitat preferences and behavior, as well as geographic ranges of almost every species known at the time. So they showed that most of the Indian snakes were highly micro-habitat tractable and that role of species assemblage were actually as different as forest interiors and ecotones. As reported by Daniel (2002), Rajasthan and other arid and semi-arid regions of the Indian subcontinent support characteristic communities of species that can cope with thermal stress and moisture stress. Number of Adaptations to Xeric Conditions: Specific adaptations of xeric reptiles, such as fossorial and nocturnal habits, as well as metabolic efficiency in sand boas (*Eryx conicus*, *Eryx johnii*), saw-scaled vipers (*Echiscarinatus*), and some colubrid species.

Multiple investigations have highlighted the importance of riparian corridors in maintaining levels of ophidian diversity. Moisture gradients offered by rivers like the Chambal (flowing through Kota), attract prey species like frogs, fish and small mammals reducing prey-predator gradients and thereby indirectly supporting higher snake densities along such bank regions (Das, 2002). The Chambal River system, in particular, has been identified as one of the more pristine ecosystems in the region due to its ravine terrain and lower levels of human disturbance, and it serves as an important wildlife refuge for snakes and gharial (*Gavialis gangeticus*). Based on the early classification of the Indian forest types as described by Champion and Seth (1968), the vegetation communities around Kota are classified primarily as the Southern Tropical Dry Deciduous Forest type, with patches of Tropical Thorn Forest in the relatively more dry areas. This structure of the vegetation contributes

both layered habitat complexity which supports cryptic snake species as well as a higher density of arboreal snakes above the leaf litter layer.

Venom Composition and Toxicological Variation

Snake venoms are diverse cocktails of proteins, enzymes, polypeptides, and non-protein components evolved primarily as tools of predation, and secondarily for defense (Calvete et al., 2009). The molecular level biochemical characterization of venom has greatly advanced with proteomic and transcriptomic technologies, allowing for large scale "venomics" characterization of venoms. The venom in *Najana naja* has been well studied, and it is mainly composed of postsynaptic neurotoxins (mainly three-finger toxins or 3FTx), cytotoxins (cardiotoxins) and phospholipase A₂ (PLA₂) enzymes (Fry et al., 2003). These effects result from the *cytotoxic* fraction, which plays a role in the necrosis of tissue at the bite site, and the neurotoxic fraction, which causes neuromuscular blockade, and can result in respiratory paralysis. Interestingly, large geographic variation in the cytotoxic vs. neurotoxic fractions of different populations of *N. naja* has been observed within the same species in India (Whitaker & Captain, 2004).

Bungarus caeruleus is reported to have one of the strongest neurotoxic venoms, among snakes in India. Its presynaptic mechanism is due to β -bungarotoxins and α -bungarotoxins act postsynaptically (Kini & Doley, 2010). These presynaptic toxins irreversibly inhibit the release of acetylcholine at the neuromuscular junction, resulting in difficult-to-reverse ascending paralysis that requires prolonged mechanical ventilation and is often only partially reversed by antivenom therapy. *Daboia russelii* venom is highly complex, and as in many other venoms, it is predominantly hemotoxic, with a full arsenal of serine proteases, metalloproteinases, and phospholipases that interact with coagulation cascades (Calvete et al., 2009). The Indian cobra, which accounts for the most provoking instances of human fatalities from snakebites in the country, displays dramatic differences in venom potency and composition throughout its range, such that antivenom raised against one geographic population may be ineffective against another (Warrell, 2010).

The venom of *Echiscarinatus* is cytotoxic, hemotoxic, prothrombin activator (ecarin), phospholipases, and hyaluronidases. In their comparison for the Mediterranean snake species *Echis*, Lomonte and Calvete (2017) found that inter-population divergence of venom composition is wide and probably due to the effect of local prey available and local co-evolution. Gutiérrez et al. Geographic venom heterogeneity is one of the most significant problems in developing potent polyvalent antivenoms effective in different parts of Indian sub-continent. This finding is particularly relevant to the Hadoti/Kota population as locally adapted venom phenotypes may differ from those used in the production of commercially available antivenoms.

2. Study Area Description

Geographic and Administrative Context: Kota district is located in Hadoti division of southeastern Rajasthan, approximately between 24°25'N–25°18'N latitude and 75°6'E–76°27'E longitude. Spanning 5,446 km², it is bordered by Bundi in the north, Jhalawar and Baran in the south, Madhya Pradesh to the east and Sawai Madhopur on the northwest. The eastern parts of the district are marked by the flow of the Chambal with the most intact and vibrant of the region's ravine systems.

Climate: The district is situated in a semi-arid to sub-humid tropical climate characterized by three seasons namely hot dry summer (March–June) in which temperatures may reach 45°C and sometimes exceeding > 45°C, a monsoon season (July–September), with mean annual rainfall of approximately 750–850 mm; and a mild, dry

winter (November–February). This climatic regime has direct implications for snake activity because of its effects on thermoregulation, prey availability, and timing of reproduction.

Topography and Vegetation: Kota is located on the Vindhyan Plateau, at an elevation of 200 to 500 metres above sea level. Three principal forms of land characterize the landscape: the Chambal ravines (*beehad* in local dialect), sharply dissected with seasonal streams and steep earthen cliffs; flat agricultural plains covered with wheat, soybean, and mustard; and rocky hillocks with scrub vegetation. According to Champion and Seth (1968), the dominant vegetation is Southern Tropical Dry Deciduous Forest, while *Anogeissus pendula*, *Buteamonosperma*, *Boswelliaserrata*, *Acacia* spp. being characteristic trees. Areas of degradation and fallow land just outside agricultural bounds are dominated by thorn-scrub vegetation.

Biodiversity Significance: The Chambal River corridor, on the eastern limit of the district, is one of the least indurated river systems of peninsular India and harbors gharial (*Gavialis gangeticus*), mugger crocodile (*Crocodylus palustris*), Gangetic dolphin (*Platanista gangetica*), Indian skimmer and variety of waterbirds. Although a part of this corridor is formally protected as the National Chambal Wildlife Sanctuary (NCWS), the sanctuary is situated along the tri-state boundary of Rajasthan, Madhya Pradesh and Uttar Pradesh and offers a similar, if less formal refuge for diversity of reptiles including, snakes. The ecological heterogeneity of the district, given its mix of riparian zones, rocky outcrops, scrubland, agricultural fields, and periurban areas, presents varied micro-habitats that host functionally distinct snake assemblages.

3. Research Methodology

Study Design: We used a combination of formal systematic field-based surveys over two annual cycles and a synthesis of secondary data based on published herpetological records, institutional collections, and regional biodiversity reports to pursue this study. Seasonal field surveys (pre-monsoon (April–May), monsoon (July–September), post-monsoon (October–November), and winter (December–January) were thus carried out to capture temporal variation in species occurrence and activity.

Field Survey Methods

- **Transect Walks:** Visual encounter surveys (VES) were carried out across systematically spaced transects of uniform length (500 m) over each of six representative habitat types: Chambal riparian zone, Agricultural fields, Dry deciduous scrubland, Rocky hillocks, Human habitation periphery and Road verges. Diurnal, crepuscular and nocturnal species were captured by accordingly walking transects at dawn, dusk and at night (headlamps). GPS coordinates, time, microhabitat information, temperature and relative humidity were recorded for each survey event.
- **Habitat-Specific Search:** Along with transects, directed search was performed in specific micro-habitats that included rocky crevices, leaf litter, termite mounds, compost and refuse piles near agriculture-based villages, and shallow water margins. These small-scale funnel traps and pitfall traps with drift fences were used in some locations where fossorial species might be present.
- **Road Mortality Surveys:** Systematic surveys of major and minor roads within the district were conducted to record information on specimens killed on the road- these records do not need to be accompanied with behavioural detection probability data as they provide only presences.
- **Community Interviews:** We pooled geo-referenced encounter records, behavioral observations, and historical snakebite incidence data via structured interviews with local snake rescuers, farmers, veterinarians, and community health workers. Interview-based data were subjected to verification cross-checks.

Species Identification

Field identification of encountered snakes was done by standard taxonomic keys from Whitaker and Captain (2004) and Daniel (2002), and confirmed via photography. Photographs were validated through scrutiny and review by expert herpetologists. Morphometric measurements (total length, snout-vent length, head measures) were recorded from each animal just before release. Specimens were not collected or killed. Identification was based on scalation, color, body proportions, and distributional data.

Venom Characterization

Abstract Venom composition data were collated from the peer-reviewed literature on proteomic and toxicological studies instead of independent extraction of venoms, since the collection of venoms from free-living specimens requires adequate laboratory facilities and permits (both regulatory and ethical approval) that are outside the limits of the current biodiversity survey. Species-specific venom profiles were compiled using published databases of venomics (e.g., UniProtKB/Swiss-Prot protein database and peer-reviewed literature).

Data Analysis

Standard ecological indices (species richness (S), Shannon diversity index (H'), and Sorenson's similarity coefficient) were used to assess species diversity and to compare habitats. For habitat associations, we ran chi-square tests against random expectations in our data to identify significant preferences. Temperature and rainfall data from the Indian Meteorological Department (IMD) were used for correlation between climatic variables and detection frequency, and seasonal activity data were plotted (Chawla et al., 2017).

Ethical Considerations

All fieldwork were conducted in accordance with the Wildlife Protection Act, 1972 (Government of India) and other relevant Rajasthan Forest Department rules and regulations. Before the start of the survey necessary permissions were obtained from the Chief Wildlife Warden, Rajasthan. No Snakes were wounded, obtained or retained captive. Snakes were not handled unless necessary for identification purposes (handled using approved hook-and-tube techniques for handling venomous snakes). Verbal informed consent was obtained from interview participants in community contexts before any data collection took place.

4. Results and Analysis

Species Diversity and Composition

A Field surveys and verified secondary records documented 24 snake species from 6 families and 18 genera in Kota district. This survey comprises a large proportion of the known ophidian diversity of Rajasthan as a whole. Of the assemblage recorded, non-venomous species were the most common (62.5%), followed by mildly (rear-fanged colubrids) and highly (elapids and viperids) venomous species (37.5%). Species listed in the table with taxonomic affiliation, venom status, and primary habitat associations (number of studies that included the species).

Table 1: Documented Snake Species of Kota District, Rajasthan: Taxonomic Status, Venom Classification, and Primary Habitat Association

S. No.	Scientific Name	Common Name	Family	Venom Status	Primary Habitat
1	<i>Najanaja</i>	Indian cobra	Elapidae	Highly venomous (neurotoxic + cytotoxic)	Agricultural fields, human habitation, scrubland
2	<i>Bungaruscaeruleus</i>	Common krait	Elapidae	Highly venomous (neurotoxic)	Agricultural fields, human habitation
3	<i>Daboiarusselii</i>	Russell's viper	Viperidae	Highly venomous (hemotoxic)	Open scrubland, agricultural edges
4	<i>Echiscarinatus</i>	Saw-scaled viper	Viperidae	Highly venomous (hemotoxic + cytotoxic)	Rocky hillocks, dry scrubland
5	<i>Ptyas mucosa</i>	Indian rat snake	Colubridae	Non-venomous	Agricultural fields, scrubland, riparian
6	<i>Gongylophisconicus</i>	Rough-scaled sand boa	Boidae	Non-venomous	Sandy/loamy soil, scrubland
7	<i>Eryxjohnii</i>	Smooth-scaled sand boa	Boidae	Non-venomous	Sandy soil, arid scrubland
8	<i>Lycodonaulicus</i>	Common wolf snake	Colubridae	Mildly venomous (rear-fanged)	Human habitation, rocky areas
9	<i>Boigatrigonata</i>	Indian gamma snake	Colubridae	Mildly venomous (rear-fanged)	Scrubland, agricultural fields
10	<i>Xenochrophispiscator</i>	Checkered keelback	Colubridae	Non-venomous	Riparian, water margins
11	<i>Ahaetullanasuta</i>	Green vine snake	Colubridae	Mildly venomous (rear-fanged)	Scrubland, secondary forest
12	<i>Amphiesmastolatium</i>	Striped keelback	Colubridae	Non-venomous	Agricultural fields, riparian
13	<i>Oligodonarnensis</i>	Common kukri snake	Colubridae	Non-venomous	Scrubland, agricultural
14	<i>Oligodontaeniolatus</i>	Streaked kukri snake	Colubridae	Non-venomous	Rocky hillocks, arid areas
15	<i>Dendrelaphistrictis</i>	Indian bronzeback	Colubridae	Non-venomous	Riparian, secondary forest edges

16	<i>Coelognathushelena</i>	Trinket snake	Colubridae	Non-venomous	Rocky terrain, agricultural edges
17	<i>Platycepsrhodorhachis</i>	Jan's cliff racer	Colubridae	Non-venomous	Rocky hillocks, dry areas
18	<i>Psammophiscondanarus</i>	Indian sand snake	Colubridae	Mildly venomous (rear-fanged)	Arid scrubland, open ground
19	<i>Spalerosophisdiadema</i>	Royal snake	Colubridae	Non-venomous	Arid scrubland, sandy terrain
20	<i>Bungarussindanus</i>	Sind krait	Elapidae	Highly venomous (neurotoxic)	Rocky areas, scrubland
21	<i>Trimeresurusgramineus</i>	Bamboo pit viper	Viperidae	Venomous (hemotoxic)	Riparian vegetation (local, limited)
22	<i>Najaoxiana</i>	Oxus cobra	Elapidae	Highly venomous (neurotoxic + cytotoxic)	Arid rocky terrain (peripheral records)
23	<i>Hemorrhaisravergeri</i>	Spotted whip snake	Colubridae	Non-venomous	Open rocky terrain
24	<i>Coronellabrachyura</i>	Indian smooth snake	Colubridae	Non-venomous	Rocky terrain, scrubland

Source: Compiled from Whitaker & Captain (2004); Daniel (2002); Das (2002); Sharma (1981); Zoological Survey of India records.

Kota district, Rajasthan was surveyed for snake diversity in the months of February-March and the comprehensive list of 24 species is provided in table 1 with field survey evidences along with some of the records from other studies collected through tertiary standards. Species are displayed in ascending taxonomic family order and include the common English name, family placement, venom status (Toxicinjury.net toxicology classification system), and the approximate associated micro-habitat types with which each species show the greatest association. Rows 1–4 depict the "Big Four" of medically important species that are emphasised by the fact that they have regional public health significance. Using classic and standard toxicological, medical literature, Venom status is way, beyond Species.

Habitat-wise Distribution

Distribution of snake species was significantly different between the six habitat types surveyed. Highest number of species encounters (n = 9 species, 38% of overall recorded diversity), followed by dry deciduous scrubland (n = 7 species, 29%) and riparian/water margin habitats along the Chambal River (n = 6 species 25%). We recorded 5 species of narrow or specialized distribution from rocky hillocks and the Chambal ravine terrain. The periphery of human habitation produced relatively low species richness but disproportionately high encounter rates of *N. naja*, *B. caeruleus* and *L. aulicus*, species that benefit from the abundance of rodent prey associated with grain storage and domestic refuse. Habitat-based distribution of species encounter frequency across the six surveyed habitat categories is displayed in Table 2.

Table 2: Habitat-wise Relative Encounter Frequency (%) of Selected Snake Species Across Six Habitat Types in Kota District, Rajasthan

Species	Agricultural Fields	Dry Scrubland	Riparian Zone	Rocky Hillocks	Human Habitation	Road Verges
<i>Najanaja</i>	35%	18%	8%	5%	28%	6%
<i>Bungaruscaeruleus</i>	40%	10%	12%	3%	30%	5%
<i>Daboiarusselii</i>	42%	32%	6%	10%	5%	5%
<i>Echiscarinatus</i>	10%	50%	2%	35%	1%	2%
<i>Ptyas mucosa</i>	38%	25%	20%	5%	5%	7%
<i>Gongylophisconicus</i>	30%	45%	2%	18%	3%	2%
<i>Xenochrophispiscator</i>	5%	3%	82%	2%	1%	7%
<i>Lycodonaulicus</i>	15%	15%	5%	20%	40%	5%
<i>Boigatrigonata</i>	30%	38%	8%	12%	8%	4%

Source: Compiled from field survey data, cross-validated against Whitaker & Captain (2004) and Alirol et al. (2010).

Table 2: The relative encounter frequency (given as percentage of total encounters per species per habitat type) of nine representative species of snakes across six main habitat types surveyed in Kota district, Rajasthan. For each species, encounter frequency was calculated by dividing the number of individuals recorded for each habitat type by the total number of individuals of that species recorded, and multiplied by 100. Data are pooled from four seasonal survey rounds. The percentages are indicative of clear habitat preferences; 82% of the individuals of *Xenochrophispiscator* were found in riparian environments, 85% combined for both *Echiscarinatus* in dry scrub and rocky habitats, and 90% of *Najanaja* and *Bungaruscaeruleus* in human changed habitats such as agricultural fields and human settlements, (Dulvy et al., 2017).

Behavioral Traits: Activity Patterns, Feeding Ecology, and Defensive Behavior

After surveying all sessions, we found the greatest majority of species (more than 70%) exhibited a clear bimodal distribution pattern, with activity peaks occurring principally at dawn (05:30–07:00 hr) and post-sunset (19:30–22:00 hr). Both *B. caeruleus* and *L. aulicus* were confirmed strictly nocturnal; this was consistent with published accounts throughout their distributions. *Ptyas mucosa*, *Dendrelaphistristis* and *Ahaetullanasuta* were mainly diurnal, while the medically important viperids (*D. russelii* and *E. carinatus*) showed mainly crepuscular to nocturnal activity in summer, giving way to partial diurnality from September to February. The variation in encounter rates over the seasons were positively correlated with the onset of monsoons. The species encountered

most frequently occurred in post-monsoon surveys (October–November), when prey activity (amphibians (Order: Anura); rodents; squamates) was at a peak following the system-wide pulse of productivity from the monsoon season. The lowest detection rates occurred in the hottest months of the year (May–June), when surface activity was limited to nocturnal behaviour due to extreme heat (>43°C). Species differed considerably in their behavioral repertoires used defensively. When provoked *N. naja* displayed typical behaviours including hood spreading and hissing loudly, whilst the aquatic *E. carinatus* always elicited its characteristic stridulatory coiling display (body coiling followed by friction between the raised scales generating a rasping sound) across all encounters. Though *P. mucosa* is non-venomous, bluff aggression was displayed in the form of lateral flattening of the body and rapid striking, behavior which has been widely documented in the species. Table 3 provides an overview of the behaviour and activity patterns of the key medically important and ecologically important snake species in Kota.

Table 3: Behavioral and Activity Profiles of Key Snake Species in Kota District, Rajasthan

Species	Activity Pattern	Primary Prey	Feeding Strategy	Defensive Display	Reproduction
<i>Najana naja</i>	Crepuscular/Nocturnal	Rodents, frogs, lizards	Active pursuit	Hood spread, hissing	Oviparous (30–45 eggs)
<i>Bungarus caeruleus</i>	Strictly nocturnal	Small snakes, lizards, rodents	Nocturnal active forager	Concealment, flattening	Oviparous (8–15 eggs)
<i>Daboia russelii</i>	Crepuscular/Nocturnal	Rodents, frogs	Ambush	Hissing, body inflation	Viviparous (20–40 neonates)
<i>Echis carinatus</i>	Crepuscular/Nocturnal	Scorpions, lizards, frogs	Ambush	Rasping coil display	Viviparous (4–16 neonates)
<i>Ptyas mucosa</i>	Diurnal	Rodents, frogs, lizards	Active pursuit	Bluff aggression	Oviparous (6–14 eggs)
<i>Xenochrophis piscator</i>	Diurnal/Crepuscular	Fish, frogs, tadpoles	Active pursuit near water	Musk secretion, biting	Viviparous (10–40 neonates)
<i>Gongylophis conicus</i>	Nocturnal	Rodents, lizards	Constriction/ambush	Ball-rolling defensive posture	Viviparous
<i>Boiga trigonata</i>	Nocturnal	Lizards, geckos, small rodents	Arboreal ambush	Lateral compression, tail vibration	Oviparous (5–9 eggs)

Source: Compiled from Whitaker & Captain (2004); Daniel (2002); Sharma (1981); Alirol et al. (2010).

Summary of some key behavioural features of eight snake species of ecological and medical importance from Kota district, Rajasthan (column explanation in the following manner- common name/scientific name) Data include timing of activity (e.g. diurnal, crepuscular, or nocturnal), general prey categories targeted, foraging strategy (active pursuit vs ambush predators), behavioral repertoire of defensive behavior when encountered or disturbed, and reproductive mode (oviparous egg-laying vs. viviparous livebearing). Abstract: These profiles were developed based on field observations coupled with rudimentary species accounts summarized in the scientific literature. Knowledge of activity and behaviour profiles is vital for the planning of awareness-raising and snakebite prevention strategies for the area.

Venom Characteristics and Toxicological Profiles

We classified the recorded venomous wildlife in Kota district into three main groups according to the predominant venom types: (1) *neurotoxic* species, including the elapids (*N. naja*, *B. caeruleus*, and *B. sindanus*); (2) *hemotoxic/cytotoxic* species, including the viperids (*D. russelii* and *E. carinatus*); and (3) rear-fanged mildly venomous colubrids with a low clinical significance (*L. aulicus*, *B. trigonata*, *A. nasuta*, and *P. condanarus*). Comparison of venom composition and clinical effects about the five venomous species are summarized in Table 4.

Table 4: Venom Composition, Toxicological Classification, and Clinical Manifestations of the Five Highly Venomous Snake Species Documented in Kota District, Rajasthan

Species	Venom Type	Key Components	Venom	LD ₅₀ (µg/g, mouse, IV)	Primary Effects	Clinical	Antivenom Available
<i>Najanaja</i>	Neurotoxic + Cytotoxic	Three-finger toxins (3FTx), cytotoxins/cardiotoxins	PLA ₂	~0.40	Ptosis, respiratory paralysis, necrosis	local	Yes (polyvalent)
<i>Bungaruscaeruleus</i>	Neurotoxic (pre + postsynaptic)	β-bungarotoxin, α-bungarotoxin, PLA ₂		~0.09	Descending/ascending paralysis, failure	respiratory	Yes (polyvalent)
<i>Daboiarusselii</i>	Hemotoxic + Cytotoxic	Serine proteases, metalloproteinases, PLA ₂ , lectins		~0.40	Coagulopathy (DIC), renal failure, swelling	local	Yes (polyvalent)
<i>Echiscarinatus</i>	Hemotoxic + Cytotoxic	Ecarin (prothrombin activator), hyaluronidase	PLA ₂	~2.30	Coagulopathy, haemorrhage, tissue destruction	local	Yes (polyvalent)
<i>Bungarussindanus</i>	Neurotoxic	β-bungarotoxin variants, PLA ₂		~0.11	Neuromuscular blockade, paralysis		Yes (polyvalent, cross-reactive)

Source: Calvete et al. (2009); Kini&Doley (2010); Warrell (2010); Gutiérrez et al. (2006); WHO (2019).

A comparative toxicological profile of the five clinically relevant venomous snake species noted in Kota district, Rajasthan, is shown in Table 4. Venom type describes the dominant pharmacological mechanism and key venom compounds represent the main protein families characterized by proteomic analysis as described in published venomomics studies. LD₅₀ values represent the lethal dose required to kill 50% of experimental mouse models through intravenous injection (quantified in micrograms of venom protein per gram body weight) and are informative of relative venom potency, with lower values being indicative of higher toxicity. The clinical effects are opposed the systematic and local manifestations of envenomation reported in a clinical case series. Availability of antivenom : CRI & VINS Bioproducts polyvalent antivenom available in India.

Intraspecific Venom Variation

For all four members of India's "Big Four," published proteomic data show significant intraspecific venom variation. *N. naja* populations from peninsular India have a higher relative component of cytotoxic constituents compared to their higher proportion of classical postsynaptic neurotoxins from northern India (Fry et al., 2003). Distinguished by one of the most extreme examples of geographic venom variation known in any snake species worldwide, four *D. russelii* subspecies display different proteomic profiles; (Warrell, 2010). Russell's viper (*D. russelii*) which in Rajasthan are all supposed to be *D. r. russelii*, populations of which have venoms with a less neurotoxic and a greater hemorrhagic component than those of Sri Lankan populations. Desert-margin populations (e.g., in Kota) have been invoked in the literature for *E. carinatus* as perhaps exemplifying intraspecific variation due to diet-induced divergence (Lomonte & Calvete, 2017).

5. Discussion

Species Diversity and Habitat Associations

The report of 24 species of snake species from Kota district adds to current understanding of ophidian diversity across the Hadoti region of Rajasthan and is comparable to diversity expected from a landscape of such ecological heterogeneity. In particular, with both *Xenochrophispiscator* and *Dendrelaphistristis*, the association with permanent water indicates that the Chambal River corridor acts as a moisture-retaining, prey-rich structural component within the assemblage. The relative abundance of the 'Big Four' species in the agricultural habitats is in accordance with the findings reported by Alirol et al. (2010), which reported that proximity to agricultural fields is among the strongest predictors of snakebite occurrence in South Asia. High abundance of this species in human habitat fringe is alarming from the public health standpoint due to its position as the first major snakebite mortality in India (Kasturiratne et al., 2008) and its nocturnal activity which renders humans undetectable before being bitten at night (typically sleeping) (Steidley et al., 2020). The preference of *E. carinatus* for rocky habitat and dry scrubland at Kota is in accordance with the known ecology of the species throughout Rajasthan and western India (Whitaker & Captain, 2004). Such large expanse of rocky hillocks and ravine terrain of Chambal badlands provide suitable thermal *refugia* and the shelter sites for this species. Finally, the open agricultural fields associated with *D. russelii* in the current study are in contrast to the lower risk of human contact reported in agricultural fields or other occupied areas indicating a reliance on top-down control in populations, evidenced by high densities of rats usually dependent on high rodent prey resources represented by crop productive landscapes (Daniel, 2002).

Behavioral Ecology and Its Implications

In Kota, the recorded diurnal activity patterns and the majority of species being nocturnal and/or crepuscular suggest that human snakebite prevention and management needs to consider localized snake activity patterns.

Notably, times of peak snake activity align with agricultural practices of the mostly rural population field-entry in the early morning before the heat of the day, crop-watching or harvesting for several hours after sunset, and nighttime irrigation work leading to high encounter probabilities. These results lend support to the arguments of Gutiérrez et al. (2006) for targeting public health behavior interventions encompass the use of protective footwear, torchlight, and community-wide educational initiatives to discuss peak points of risk. The defensive behavior repertoire of a given species also translated directly to clinical relevance. The comparatively cryptic, ambush-predator mode of defense of *B. caeruleus* vs. the greater aposematic warning coloration displays of *N. naja* may help explain the frequent accidental envenomation by this species (especially as individuals contact or attempt to reach through grass and/or forego visual contact by sleeping on the ground). Their viviparous reproductive mode and large litter sizes allow *D. russelii* and *E. carinatus* to produce very high densities of offspring in a single reproductive season, possibly creating localized population spikes that could increase the risk of snakebite around localized great birthing sites. That narrow time frame of gravid female aggregations close to rocky outcrops and scrubland edges during the post-monsoon period is a time of risk that community health programs should target.

Venom Variation and Ecological Significance

The significant differences in venom composition that we document between species and populations in Kota and across the broader Rajasthan region have important implications for clinical care and antivenom manufacture. This antivenom is a stock polyvalent antivenom derived from the venoms of *N. naja*, *B. caeruleus*, *D. russelii*, and *E. carinatus* specimens collected mainly from Tamil Nadu and Maharashtra, which raises concerns regarding the neutralizing ability of the commercial antivenom against the different venom phenotypes expected in northern/northwestern Indian populations. This problem on the larger subcontinental scale has been recognized (Warrell, 2010) with perception that geographic venom variation constitutes one of the most underdressed problems in clinical snakebite management in India. Calvete et al. These authors (Zelanis et al., 2009) have also critically discussed this issue and proposed that a venomomics-informed antivenom production approach i.e. proteomic characterization of geographically distinct venom populations is one way of making antivenoms more effective. This argument is especially powerful for a geographic area distinct from that of the regions of the commercial antivenom source populations which, ecologically, this separates from the areas where this population exists such that the populations in question are unlikely interchangeable for this specific characteristic. Moreover, the prey-driven evolution hypothesis of venom variation (Lomonte and Calvete, 2017) may have particular relevance in the Kota. The differences between the scorpion, sandy-terrain lizard, and rodent prey obtainable in the Kota region compared with the amphibian-based diet of this species in wetter areas may result in selection for different venom components (e.g., increases in phospholipases and hyaluronidases directed at arthropod prey) from the venom used to produce antivenom in the laboratory.

Conservation Status and Threats

Based on the Wildlife Protection Act, 1972 of India, all snake species recorded in Kota, Rajasthan are also protected under Schedule II that prohibits any collection, trade, or killing {but not consumption} of the animals. But human type of persecution motivated by fear, and cultural ideal, is still a big deal for snakes dying for all of the part. Apart from this, habitat destruction due to conversion of scrubland and fallow land into agricultural land, quarrying on rocky hillocks and expansion of road network across the Chambal ravines is leading to fragmentation of habitat and population decline. Habitat specialists, such as the rocky hillock specialist *E. carinatus*, sandy loam soil *Gongylophis conicus*, and arid scrubland *Spalerosophis diadema* species emerge as the most susceptible to habitat loss).

6. Conclusion

An integrative inventory of 24 snake species, their habitat associations and behavioral ecology, along with a synthesis of available toxicological literature is provided as a foundation for future studies of the snake fauna of the Kota district of Rajasthan, India. Widely represented among habitats, the "Big Four" venomous species (*Naja naja*, *Bungarus caeruleus*, *Daboia russelii*, and *Echiscarinatus*) are almost universally encountered in agricultural fields and the human habitation periphery contributing zones of maximum encounter probability and therefore maximum snakebite potential. The behavioral activity patterns, predominantly crepuscular to nocturnal for the dangerous species, coincide with human rural activity cycles to promote high snakebite risk especially during and just after the monsoon. Within the context of this study, venom samples can vary in an interspecific as well as in an intraspecific way which becomes a relevant aspect both clinically and ecologically (i.e. conservation). The difference between venom phenotypes observed in regional populations relative to those used in commercial antivenom production indicate an urgent need for regional venom characterization studies and perhaps the inclusion of Rajasthan-origin specimens into antivenoms production protocols. Based on these findings, the following recommendations are made: (1) the establishment of a state-level herpetological monitoring program to provide long-term species occurrence data for the state under the Rajasthan Forest Department, (2) commissioning of comparative venom proteomic studies involving the use of specimens from Rajasthan (especially *D. russelii* and *E. carinatus*), (3) implementation of community-based snakebite prevention programs particularly in agricultural communities in Kota, *Bundi* and the *Jhalawar* districts; (4) school curriculum and rural health program content on snake ecology and snakebite management; and (5) enhancement of legal protection for snake habitats within the Chambal ravine system, preferably as an extension of buffer zones in the National Chambal Wildlife Sanctuary.

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