







Article

Micrurus nigrocinctus in Colombia: Integrating Venomics Research, Citizen Science, and Community Empowerment

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Abstract: Snakebite is a high-priority neglected tropical disease, and a strategic goal based on four pillars has been recommended to reduce mortality and morbidity. One is empowering rural communities through citizen science, education, and engagement. In this study, an integrative approach was used to expand our knowledge of *Micrurus nigrocinctus* status and characterize its venom. Using citizen science data and field visits to local communities, 99 records of *M. nigrocinctus* distributed in Antioquia, Chocó, and Córdoba were obtained. Children, young people, and adults recognized *M. nigrocinctus* as the most common coral snake species in their region, and two specimens were recovered for venom and Phylogenetic analyses. The *M. nigrocinctus* venom from Colombia exhibited similar chromatographic and electrophoretic profiles and biological activities and shared nearly identical protein families with Costa Rica. Commercial coral snake antivenoms also recognized and neutralized the whole venom from both countries. However, phylogenetic relationships showed greater divergence with specimens from Costa Rica. Involving communities helps prevent coral snake bites and facilitates access to rare specimens such as *M. nigrocinctus*, thereby enabling venom analyses, improving antivenom evaluation, and advancing toxinology research for medically significant species.

Keywords: *Micrurus nigrocinctus*; venomics research; citizen science; coralsnake; community empowerment; Colombia

Key Contribution: This study integrates citizen science with venom and phylogenetic analyses to update the knowledge of *Micrurus nigrocinctus* in Colombia. It highlights the value of community engagement for accessing coral snake specimens, preventing bites, and promoting the conservation of these medically important species.



Received: 13 April 2025
Revised: 13 May 2025
Accepted: 20 May 2025
Published: 27 May 2025

Citation: Rey-Suárez, P.; Rojo, L.P.; Gómez-Robles, J.; Parra-Moreno, S.; Pachón-Camelo, E.; Fuentes-Florez, Y.; Lomonte, B.; Fernández, J.; Sasa, M.; Núñez, V.; et al. *Micrurus nigrocinctus* in Colombia: Integrating Venomics Research, Citizen Science, and Community Empowerment. *Toxins* **2025**, *17*, 268. <https://doi.org/10.3390/toxins17060268>

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1. Introduction

Snakebite envenoming (SBE) represents a significant public health burden in many developing regions, particularly affecting resource-poor areas in sub-Saharan Africa, South Asia, Papua New Guinea, and Latin America. More than two million cases of SBE are reported annually, resulting in approximately 100,000 deaths and 300,000 cases of permanent disability [1]. Individuals at the highest risk live in rural areas, including children and young agricultural workers [2]. SBE occurs while people are engaged in everyday activities such as working in fields, collecting drinking water, sleeping in their homes at night, attending school, or even simply walking outdoors [3–6].

In 2017, the World Health Organization classified SBE as a high-priority neglected tropical disease [3,7], and in 2019, a strategic goal was established to reduce SBE-induced mortality and morbidity by 50% by 2030. This strategy is based on four pillars: (a) strengthening health systems, (b) ensuring safe and affordable access to quality antivenoms, (c) promoting research and innovation, and (d) community empowerment. According to WHO [8], empowering rural communities through citizen science, education, and engagement allows them to understand the associated risks better, adopt preventive measures, and seek timely and appropriate medical care, reducing the impact of snakebite [9,10]. By contributing their knowledge, communities not only manage to get involved in solving the problem that affects them but also participate in generating knowledge that promotes co-existence and the relationship between humans and snakes in these areas [8,11–13].

The involvement of local communities in a comprehensive approach to SBE could be key in situations where securing specimens for research programs or antivenom production is challenging. Such is the case of coral snakes (genus *Micrurus*), a group comprised of around 90 species [14], some of which are responsible for severe envenoming throughout the Americas [15–17]. Despite their medical relevance, knowledge about coral snake venoms remains limited, and the venoms of approximately two-thirds of the species remain uncharacterized. On the one hand, many species' secretive habits and semi-fossorial lifestyle mean that knowledge about their ecology and distribution is highly fragmented [18,19]. This prevents a clear view of the species that may be present around communities in specific geographic areas and, therefore, the identification of those species of interest in the context of SBE [20]. Adding to these drawbacks is the scarcity of coral antivenoms, with only a few countries on the continent producing them [21]. This situation results in part from the difficulties in finding and capturing coral snake specimens but also from the restrictions on their maintenance in captivity [22] and the low venom yield that these elapids generally possess [23].

To reverse this situation, some antivenom research and production centers, such as the Clodomiro Picado Institute at the University of Costa Rica, are turning to local communities to secure specimens and identify sites where species of interest are relatively more abundant. These joint efforts have consolidated the maintenance of an important collection of *M. nigrocinctus*, a widely distributed species in the country whose venom is also used as an immunogen in the production of anticoral antivenoms for the Central American region [24]. This species is characterized by its relatively slender body and tricolor ring pattern and is distributed in Central and South America, from Mexico to northwestern Colombia [18]. Throughout this distribution, *M. nigrocinctus* is considered a locally abundant species responsible for most regional coral-related accidents [25]. However, it is not a snake that is easily collected, especially in the southern limit of its distribution in South America, so knowledge about its ecology and venom composition is mainly limited to populations in Costa Rica. *M. nigrocinctus* venom exhibits presynaptic neurotoxic effects, inducing a concentration-dependent depolarization in isolated preparations of mouse phrenic nerve-diaphragm, accompanied by ultrastructural changes in nerve terminals, probably

due to the presynaptic action of PLA₂ [26] and post-synaptic blockade of three-finger toxins [27]. Severe local myotoxicity in mice and morphological alterations in myogenic cells *in vitro* have also been reported for this venom [28,29]. The venom proteome of *M. nigrocinctus* from Costa Rica is mainly composed of phospholipases A₂ (PLA₂; 48%) and three-finger toxins (3FTx; 38%), along with proteins belonging to the ohanin (OH; 3.8%), L-amino acid oxidase (LAAO; 2.3%), C-type lectin (C-Lec; 2.2%), serine proteinase (SVSP; 0.7%), and nucleotidase (Nuc; 0.5%) families. Recent analyses with more sensitive techniques also identified other protein families [30].

Here, we employ an integrative approach to expand our knowledge of the status of *M. nigrocinctus* and characterize its venom at the southern limit of its distribution. Using citizen science data and visits to local communities in the Urabá and Chocó regions in Colombia, we re-evaluated the species' known distribution in this country, analyzed its phylogenetic relationship with other coral snakes, characterized the biological activity and proteomic profile of its venom, and evaluated the ability of commercially available antivenoms to neutralize envenomation by this species. By including information on communities that coexist with *M. nigrocinctus*, our work also allows for an analysis of the perceptions of this and other coral snakes held by the people there. Importantly, our research provides practical insights into future research and public health strategies, enhancing our understanding of the species and its implications for public health.

2. Results

2.1. Distribution of *M. nigrocinctus* in Colombia Based on 'Citizen Science' and Scientific Database Reports

The data search and curation retrieved 99 records for *M. nigrocinctus* (January 2018–February 2025), distributed across three departments (Supplementary Table S1). The highest number of records came from the Department of Antioquia, accounting for 94% (93 records). The municipalities with the most records were Apartadó (27 records), Carepa (34 records), and Turbo (28 records) (Figure 1). Five percent were from the department of Chocó, and a single record was for the department of Córdoba. The latter represents the first record of the species for this department (an individual deposited in the collection of the Serpentarium of the University of Antioquia—SUA (Supplementary Figure S1). Of the total records, only 31% (31 records) were obtained from specimens in collections or reported through Biodiversity Information Systems (GBIF or SIB). The community contributed the remaining 69% (68 records) through the reviewed platforms. Regarding the origin of the records, four out of the five records from Chocó came from rural areas, as did the only Córdoba Department record. In contrast, 60 (64%) of the *M. nigrocinctus* records in Antioquia were obtained from urban areas, of which 45 (75%) were obtained through different groups on the Facebook website, representing the total number of records obtained through this source (Figure 2).

According to the distribution of *M. nigrocinctus*, the largest number of recorded snakes are clustered in the municipalities of Apartadó, Carepa, and Turbo (Figure 1); therefore, educational activities were conducted in these locations. A total of 802 individuals participated in the educational workshops. Among them, 403 were children, 215 were young people, and 184 were adults. Within the adult group, 20 belonged to indigenous communities, 72 were university students from various disciplines, and 17 were farmers. Notably, 50 healthcare personnel from the hospitals of Apartadó and Carepa attended the sessions, along with 25 members of rescue teams from Apartadó.

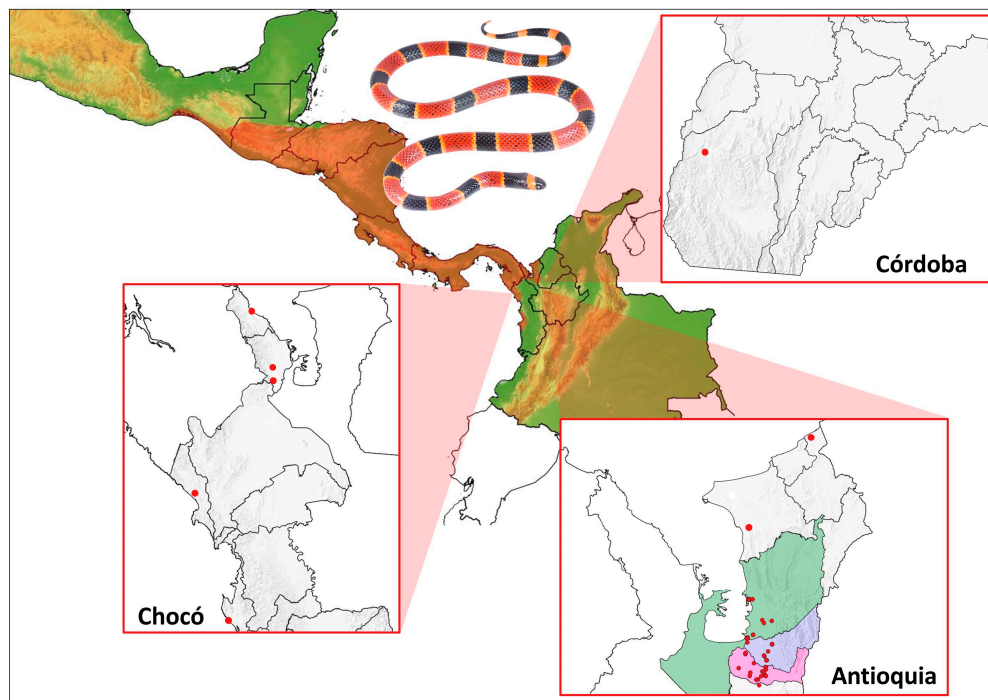


Figure 1. Distribution of *M. nigrocinctus* in Colombia based on ‘Citizen Science’ and scientific database reports. The species distribution is shaded in red. Red circles indicate recorded locations. In Antioquia, the municipalities with the most records were Apartadó (blue), Carepa (pink), and Turbo (green). The photograph shows an *M. nigrocinctus* specimen from Apartadó (Antioquia, Colombia).

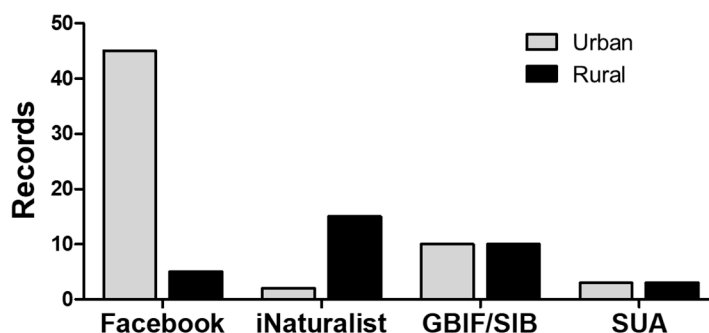


Figure 2. Reports for *M. nigrocinctus* in the Department of Antioquia based on ‘Citizen Science’ reports and scientific databases. GBIF/SIB: Biodiversity Information System/Biodiversity Information System; SUA: collection database of the Serpentarium of the University of Antioquia.

These educational initiatives raised awareness about snakebite prevention, the importance of snake conservation, and appropriate first aid responses in case of a bite. While *M. nigrocinctus* was the focal species, including other regional species such as *Bothrops asper*, *Porthidium nasutum*, and *Micrurus camilae* added crucial context for understanding snakebite risks in the community. The community showed great curiosity about coral snakes, actively engaged with the information provided, and demonstrated the ability to identify the venomous snake species in the area. Healthcare personnel received clear guidance on the specific treatment for coral snake bites and instructions on the importance of rapidly transferring patients to higher-level hospitals in cases of envenomation presenting with early signs of respiratory paralysis.

Overall, participants in the educational activities recognized *M. nigrocinctus* as the most common coral snake species in their region. As a result of this engagement, two *M. nigrocinctus* specimens were recovered, one from Apartadó and one from Carepa. These individuals were subsequently used for molecular and venomous analyses.

2.2. Molecular and Phylogenetic Analysis of *M. nigrocinctus* from Colombia

The phylogenetic reconstruction reveals a closer relationship between the *M. nigrocinctus* samples from Colombia and Panama, forming a highly supported monophyletic group. This clade is sister to *M. mosquitensis* from Costa Rica, *M. ruatanus* from Honduras, and *M. nigrocinctus* from the rest of Central America (Figure 3).



Figure 3. Phylogenetic tree resulting from Bayesian inference of ND4 and *Cytb* gene fragments from 41 coral snake species. Posterior probabilities (>0.95) are indicated by pink circles. Lineage conformations are shown according to [31].

2.3. Comparative Biochemical and Biological Characterization of *M. nigrocinctus* Venom from Colombia and Costa Rica

The chromatographic profiles of *M. nigrocinctus* venoms from Colombia and Costa Rica were compared. Both venoms exhibited many prominent peaks eluting between 35 and 55 min. Notable differences were observed in the abundance of peaks eluting at 20 min and between 45 and 55 min when comparing both venoms (Figure 4). The comparative electrophoretic profiles for *M. nigrocinctus* venoms from Colombia and Costa Rica showed abundant proteins in the region between 10 and 20 kDa, with notable band intensity differences indicating variations in the relative abundance of such small proteins. On the contrary, the band pattern for the region between 50 and 75 kDa was similar for both venoms (Figure 4).

Using a shotgun proteomics approach, the venoms of *M. nigrocinctus* from Colombia and Costa Rica were compared, resulting in the detection of at least 75 and 134 proteins, respectively. Overall, both venoms presented almost the same protein families, except for a few minor components of the Cysteine proteinase (CysP) and Wapryn (Wap) families, which were not detected in the Costa Rican sample, and the CysP-inhibitor family, not detected in the Colombian sample (Figure 5; and Supplementary Table S2). Despite the conserved composition of these venoms in terms of protein families, the notable difference in the total number of proteins obtained in this comparison originates from the more significant number of variants belonging to the three-finger toxin (3FTx) and the phospholipase A₂ (PLA₂) protein families detected in the Costa Rican sample.

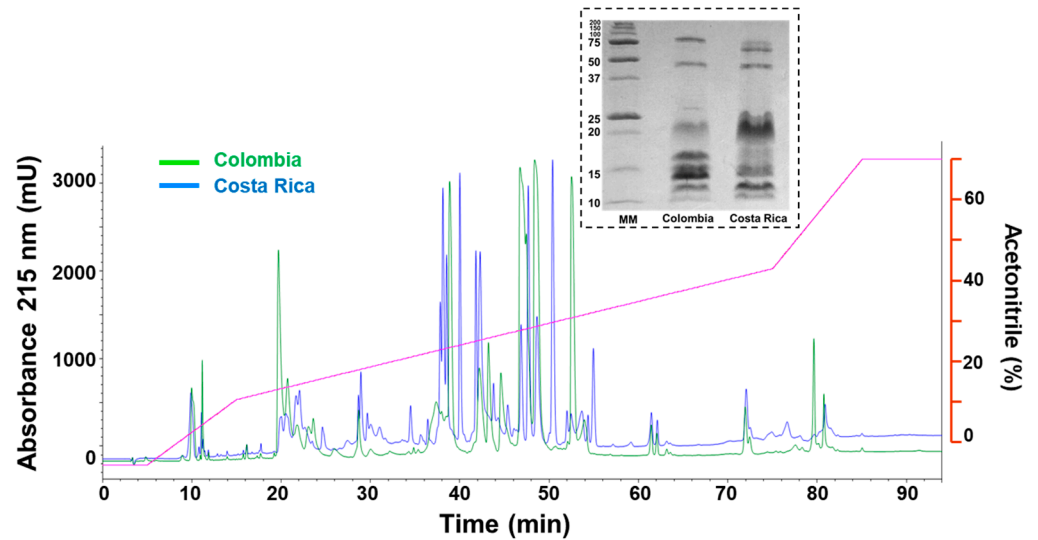


Figure 4. Comparison of the RP-HPLC profiles and SDS-PAGE (inset) of venom of *M. nigrocinctus* from Colombia (green) and Costa Rica (blue). Venom was fractionated on a C18 column and eluted with an acetonitrile gradient (purple line). The red axis corresponds to acetonitrile (%). For the SDS-PAGE, molecular weight markers are shown on the left in kDa.

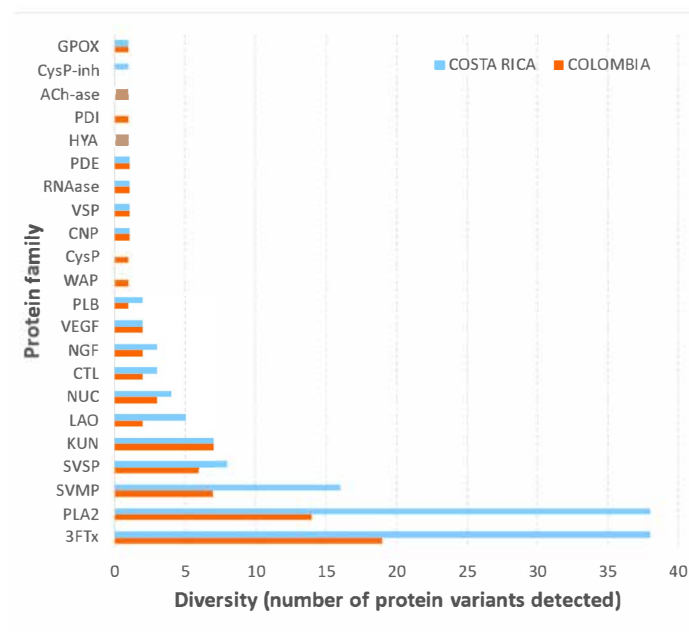


Figure 5. Diversity of proteins detected by shotgun proteomics in the venoms *Micrurus nigrocinctus* from Colombia and Costa Rica.

The PLA₂, myotoxic, and edema-forming activity of both venoms were comparable since statistically significant differences were not observed ($p > 0.05$) (Figure 6).

The intraperitoneal (i.p.) LD₅₀ for *M. nigrocinctus* venom from Colombia was estimated at 8.6 µg/mouse (95% CI = 3.18 to 15.8), equivalent to 0.5 µg/g of body weight.

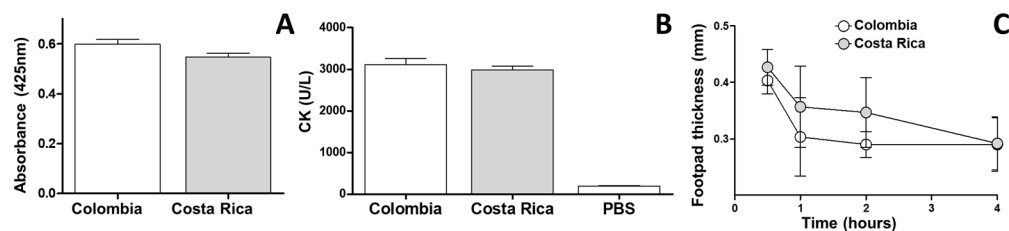


Figure 6. Comparative biological activities of *M. nigrocinctus* venom from Colombia and Costa Rica: (A) Phospholipase A₂ activity upon the monodisperse synthetic substrate 4-nitro-3-octanoyloxybenzoic acid (4-NOBA). (B) Myotoxic activity estimated by plasma creatine kinase (CK) activity. Each bar represents the mean ± SEM of triplicate assays. (C) Edema forming activity in the mouse footpad assay. Each point represents the mean ± SEM of triplicate assays. No statistically significant differences were found in any of the comparisons.

2.4. Immunorecognition and Neutralization by Commercial Coral Antivenoms

Commercial coral antivenoms demonstrated a clear recognition of whole *M. nigrocinctus* venoms from Colombia and Costa Rica when antibody titers were evaluated by ELISA. The anticoral-ICP antivenom exhibited a slightly stronger recognition of both venoms compared to the anticoral-INS antivenom, which was statistically significant ($p < 0.05$; Figure 7A), with the highest binding observed for *M. nigrocinctus* venom from Costa Rica, which is used in its production (Figure 7B).

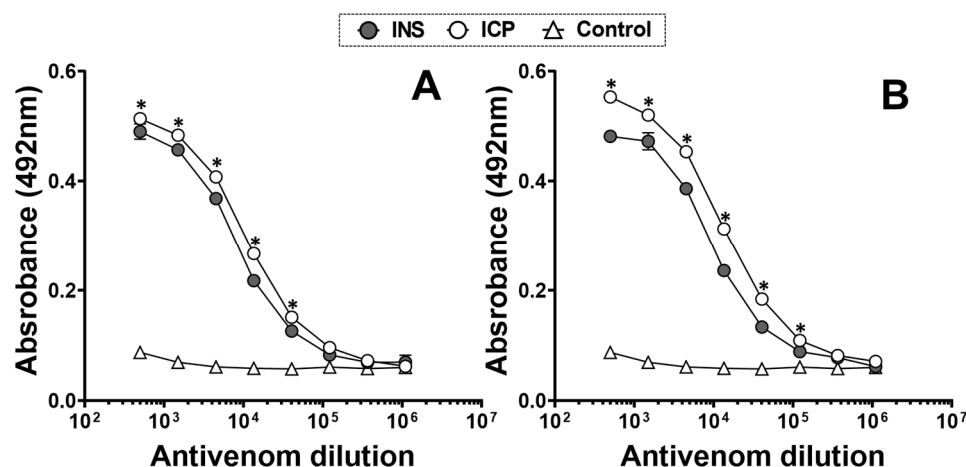


Figure 7. Recognition of commercial coral antivenoms against *M. nigrocinctus* venom from (A) Colombia and (B) Costa Rica. Each point on the graph represents the mean ± SD of the triplicate. * ($p < 0.05$) indicates a significant difference between both venoms.

In addition, by preincubation assay, the anticoral-ICP antivenom neutralized the lethal effect of *M. nigrocinctus* venom from Colombia in a proportion of 0.2 mg/mL. In contrast, anticoral-INS partially neutralized the lethal effect of this venom when tested at the same proportion (Table 1).

Table 1. The preincubation neutralizing ability of anticoral-INS antivenom and anticoral-ICP antivenom on the lethal effect of *M. nigrocinctus* venom from Colombia in mice.

Group	Venom/Antivenom	Dead/Inject ¹
<i>M. nigrocinctus</i> venom	0	4/4
<i>M. nigrocinctus</i> venom + anticoral-ICP	0.2 mg/mL	0/4
<i>M. nigrocinctus</i> venom + anticoral-INS	0.2 mg/mL	1/4

¹ The lethality-neutralizing ability of antivenom was evaluated by preincubating with the venom at 37 °C for 30 min and then injecting the mixture into mice (16–18 g of body weight) by i.p. route. Deaths were recorded at 48 h.

3. Discussion

In this study, we aimed to increase knowledge on the poorly studied population of *M. nigrocinctus* in Colombia by integrating information gathered from biological collections, as well as communities regarding its occurrence in different geographical areas, with data obtained from laboratory analyses on its venom characteristics such as protein composition, toxic activities, neutralization by commercially available antivenoms, and the phylogenetic relationships of this population with other coral snakes.

Data related to specimens preserved in biological collections constitutes a valuable source of information in terms of morphology, diet, reproduction, and distribution, among various other aspects [32,33]. Biological collections contain fundamental data that allow us to fill the gaps around these species [31,33–35]. However, elusive organisms that are difficult to find in the field, such as snakes of the *Micrurus* genus, often have significant gaps in many aspects mentioned above [19]. For these specimens, a helpful way to collect biological data is the information provided for local communities, such as photographs, information about geographic location, and date–time information [36]. In addition, developing and enhancing ‘citizen science’ platforms can potentially reduce the shortfall of information related to *Micrurus* species.

The distribution of *M. nigrocinctus* in Colombia has been thus far limited to the northern Chocó biogeographic region [20,37]. These records are associated with the departments of Chocó and Antioquia. Although the area of Antioquia with the highest number of records of *M. nigrocinctus* borders the department of Córdoba, to date, there were no records of the species for this department, to the best of our knowledge. Different studies have reported the presence of other species of coral snakes, such as *M. dissoleucus*, *M. dumerilii*, and *M. mipartitus*, in various areas of Córdoba [38–40]. Therefore, the record presented here constitutes the first report in the literature on species distribution for the Department of Córdoba.

Most reports of *M. nigrocinctus* in Colombia occurred in urban areas, indicating relatively frequent encounters between humans and coral snakes in such settings, similar to the occurrence in Costa Rica [41,42]. This epidemiological pattern has been observed in studies of coral snakebite incidence, where more than half of the events occurred in urban areas, contrasting with the pattern of viper bites [43]. It is known that some coral snake species in the Amazon, such as *M. surinamensis* and *M. lemniscatus*, live near or within urban perimeters, in fragments of vegetation, or even close to residential settlements [44], which increases the likelihood of snakebite incidents in these areas.

Thanks to this articulation with communities in regions with abundant *M. nigrocinctus*, several specimens were collected to determine phylogenetic relationships with other *Micrurus* species. Over time, the evolutionary relationships within the *Micrurus* genus, as well as species delimitations, have been complicated and unclear [45,46] because most systematic evaluations of *Micrurus* have been heavily based on phenotypic traits (e.g., coloration, scalation, hemipenis morphology, etc. [18]. Meanwhile, there is a limited number of molecular studies, which are restricted mainly in terms of the number of species and/or geographic sampling [47–51]. This has incorrectly classified many regional color variants as distinct species or subspecies within the genus [52]. Therefore, addressing species diversity within the *Micrurus* genus remains of vital importance from biological, medical, and conservation perspectives [51,52].

Recent studies have described *Micrurus nigrocinctus* as a species complex due to its taxonomic diversity, forming polyphyletic groups that include species such as *M. mosquitensis*, *M. ruatanus*, and *M. nigrocinctus* from several localities across Central America [31]. These authors emphasize that the complex comprises at least three distinct lineages at the species level. In this way, the phylogenetic analysis of this study revealed a well-supported clade

formed by *M. nigrocinctus* from Colombia and Panama, suggesting that *M. nigrocinctus* from Colombia is part of lineage 1. This clade is monophyletic and highly divergent from all other *M. nigrocinctus* lineages from Central America and, until now, has been restricted exclusively to Panama [31].

The proteomic profiling of *M. nigrocinctus* venom from Colombia showed a qualitatively similar protein family composition when compared to venom from Costa Rica, and the identified components are in general agreement with previous studies on the proteome of the latter venom [29,30]. However, the notable difference observed in the diversity of protein variants for 3FTx and PLA₂ components between both venoms is intriguing and would demand further studies to establish the underlying mechanisms. Our findings suggest the possibility that a larger array of genes for these toxins could be either (a) present, (b) expressed, or (c) differentially regulated in the Costa Rican specimens, as compared to Colombian counterparts. Since our proteomic analyses were conducted using a general database for snakes (Uniprot Serpentes), future studies should address this question by complementing the proteomic data with specific venom gland transcriptomic data of *M. nigrocinctus*, which are currently unavailable.

The venom of Costa Rican *M. nigrocinctus* is known to be PLA₂-rich in its composition [29] and has demonstrated strong local myotoxic activity in the mouse model [28,53,54]. In this context, the venom of *M. nigrocinctus* from Colombia exhibited PLA₂ activity (4-NOBA), edema-forming, and myotoxic activities similar to the Costa Rican venom of this species. Likewise, the LD₅₀ of *M. nigrocinctus* from Colombia was similar to that reported for the Costa Rican population [22,24], *M. mosquitensis* [55], and *M. ruatanus* [56]—species that belong to the *M. nigrocinctus* complex—further supporting the phylogenetic relationships described within this group.

As shown by the ELISA titration curves, the anticoral-ICP antivenom exhibited somewhat stronger recognition of *M. nigrocinctus* venom from Colombia, resulting in antibody binding signals nearly as high as those obtained for the homologous venom (*M. nigrocinctus* from Costa Rica) used in its production. The results highlight the high degree of antigenic conservation between the venoms from the two populations. On the other hand, the anticoral-INS antivenom showed good cross-recognition of *M. nigrocinctus* venom from Colombia despite this venom not being included in the immunizing mixture for its production [57]. Our findings align with previous studies, which observed cross-recognition of *M. nigrocinctus* and *M. mosquitensis* venoms from Costa Rica by the anticoral-INS antivenom [58].

The observed immunorecognition and neutralization are likely due to the antigenic similarity reported for venoms that exhibit a PLA₂-predominant phenotype. It has been previously noted that coral snake venoms with a PLA₂ predominance are better recognized and neutralized by ICP antivenom than those dominated by a high proportion of 3FTx proteins [55,59–61]. Thus, the dichotomous grouping of *Micrurus* venoms according to their relative abundance of 3FTx and PLA₂ protein families is not only a quantitative feature but also a qualitative one by possibly reflecting the antigenic divergence of proteoforms between the two groups [30]. Nevertheless, it has been shown that the ICP antivenom can neutralize the venom of *M. ruatanus*, a highly lethal species with 3FTx predominance [56] that inhabits Roatán Island and belongs to lineage 2 of the *M. nigrocinctus* complex [31]. According to our findings, *M. ruatanus* shows a close phylogenetic relationship with *M. nigrocinctus* from Colombia, suggesting possible antigenic similarities between their venom components.

The anticoral-ICP antivenom and anticoral-INS antivenom neutralized the lethal activity of *M. nigrocinctus* venom from Colombia in a preincubation-type murine model. This preclinical assay suggests that treatment with these antivenoms is likely effective in cases of envenomations by this coral snake.

Micrurus snakes are generally docile and colorful and are known for their striking combination of red, yellow, and black rings [62]. This appearance can spark curiosity, especially in young children [43,63,64], often placing them among the populations most affected by coral snake bites [64–66]. Envenomations by *Micrurus* in children are particularly serious, not only due to the severe neurotoxic syndrome these bites induce, which can lead to rapid respiratory failure [7,17,63], but also due to myotoxic effects, which result in significant increases in plasma creatine kinase levels and are rarely observed in adults [17].

The WHO strategy to decrease the burden of snakebites hinges on four pillars [3]. This research is based on two of these: to empower and engage communities through knowledge about snake biology, snake bite prevention, and management, and to ensure safe and effective treatment through the preclinical evaluation of the antivenom available in Colombia and Costa Rica for the treatment of *M. nigrocinctus* envenomation. Adequate knowledge of snake behavior habits and appropriate first aid can reduce the likelihood and consequences of snakebites among people at high risk of encountering them [67]. Considering the distribution of *M. nigrocinctus* in Colombia, the frequency of sightings reported by communities, and the severity of coral snake envenomation in children, the high participation of infant and young population from Apartadó in the workshop “Coral snakes Are Not What People Think They Are” was particularly important, since educating new generations (children and youth) is essential to fostering a new relationship with coral snakes, one that prioritizes conservation, prevention, and sustainable co-existence between humans and snakes.

In addition, this educational approach was implemented with local adult populations and health professionals. Several studies have found that many healthcare workers in snakebite-endemic regions have poor general knowledge about the prevention and management of snakebite envenoming [67,68]. The implemented educational program addressed important issues such as how to recognize the main *Micrurus* species in the region, the differentiation of venomous and non-venomous mimic snakes, how to avoid snakebites, administration of first aid, as well as the importance of seeking immediate medical attention [43,63,69]. Finally, the involvement of communities not only helps prevent coral snake bite, but increases the likelihood that scientists gain access to otherwise difficult-to-find specimens, such as *M. nigrocinctus*. This, in turn, facilitates venom analyses of rare species, the evaluation of antivenoms, and advances the toxinology knowledge for medically significant species.

4. Conclusions

This study demonstrates the effectiveness of citizen science and community empowerment as tools to enhance knowledge about *Micrurus nigrocinctus* in Colombia. The involvement of local communities not only aids in the prevention of coral snake bites through education and awareness but also significantly increases the likelihood of accessing rare and elusive species such as *M. nigrocinctus*.

The venom profile of Colombian specimens closely resembles that of Costa Rican populations in terms of protein composition and biological activity and was similarly recognized and neutralized by both anticoral-INS and anticoral-ICP antivenoms, despite underlying phylogenetic divergence. This, in turn, contributes to advancing toxinological knowledge for medically important snakes.

5. Materials and Methods

5.1. Distribution of *M. nigrocinctus* in Colombia Based on ‘Citizen Science’ and Scientific Database Reports

Given the elusive nature of *M. nigrocinctus* and the scarcity of records in Colombia, we conducted a detailed assessment of its distribution. We integrated various sources of

information on records and occurrences, supplementing the records of localities registered in databases of scientific collections with information from public participation, both from data provided by visited communities and from online databases with verifiable photographic records. Records were reviewed in the main herpetological collections linked to the national system of biological collections of the Alexander von Humboldt Institute for Biological Resources Research through the Biodiversity Information System (BIS) and the Global Biodiversity Information System (GBIF) and in the collection database of the Serpentarium of the University of Antioquia (SUA). Additionally, secondary information available in books, herpetological inventory, characterization reports, reports and information on snakebite accidents, scientific articles, and other academic documents, among others, that may offer information on *M. nigrocinctus* in Colombia, was reviewed.

Citizen science information comes from online databases with verifiable photographic records and from photograph recognition by members of the communities visited. We reviewed photograph records posted in groups created on the Facebook website for this purpose (Aliados de las serpientes—Colombia, Serpientes de Colombia, Identificación de serpientes, Serpientes de Colombia/Fauna ofídica colombiana) and on the biodiversity registry site iNaturalist until February 2025. Keywords such as “*Micrurus nigrocinctus*”, “*nigrocinctus*”, “Coral centroamericana”, and “Coral Urabá” were used. In all cases, only reliable records with accurate taxonomic determinations, clear and precise information on location, non-redundant records, and information consistent with the ecological characteristics of this species were considered. The data were deposited into a database for further analysis.

In addition, to engage communities living in regions with frequent *M. nigrocinctus* sightings, a workshop titled “Coral Snakes Are Not What People Think They Are” was held. This workshop covered key topics such as general information about snakes (both venomous and non-venomous), snakebite prevention (how to avoid bites and what to do in case of an incident), and snake conservation, with emphasis on the ecological and scientific importance of snakes, especially the role venomous species play in antivenom production and medical research. Special attention was given to coral snakes during the training. The workshop featured a mix of vivid photographs, sounds, and videos integrated into an engaging presentation. Hands-on interaction was a key component, with participants using 3D models (including snake skeletons, skulls, and fangs) and real specimens (snake skins, sheds, eggs, and authentic fangs). Participants were allowed to touch a live snake at the end, fostering a more direct connection with these animals.

5.2. Molecular and Phylogenetic Analysis of *M. nigrocinctus* from Colombia

Genomic DNA was extracted from shed skin and blood of two *M. nigrocinctus* specimens from Apartadó and Carepa (Antioquia—Colombia) using E.Z.N.A.[®] Omega Tissue DNA Kit (Cat. D3396-01) and following the manufacturer’s protocols. The primers, listed in supporting information Supplementary Table S3, allowed us to obtain sequences for two genes (*cytb* and *nd4*). PCR reactions were set up to a final volume of 25 µL, using 1 µL genomic DNA (2 ng/µL), 0.5 µL of each primer (0.2 µM), 2.5 µL of 10X PCR buffer, 0.5 µL total dNTPs (0.2 mM), 0.75 µL of MgCl₂ (1.5 mM), 0.1 µL of Platinum[®] Taq DNA Polymerase (1 U), and 19.15 µL of H₂O. Typical amplification conditions involved initial denaturation at 94 °C for 5 min, followed by 35 cycles with a denaturation step at 95 °C for 45 s, an annealing stage at 55 °C for 45 s, an extension at 72 °C for one min, and a final extension at 72 °C for 10 min. Amplicons were separated by electrophoresis on 1.5% agarose gels in 1X TAE buffer, dyed with GelRed[™] Nucleic Acid Gel Stain (Biotium, Inc., Fremont, CA, USA), and visualized under UV light. We performed Sanger sequencing in a capillary automated ABI3500 sequencer (Applied Biosystems[®], Thermo Scientific; Waltham,

MA, USA) at the AUSTRAL omics (Santiago, Chile). The DNA sequences were edited (Trim Ends and de Novo Assemble) and aligned in Geneious Prime v2025.0.3 [70]. For *nd4* and *cytb* genes, the nucleotide sequences were translated into proteins to evaluate the reading frame and ensure the absence of premature stop codons or other nonsense mutations in GeneDoc [71]. Novel sequences were deposited in GeneBank (accession numbers are shown in Supplementary Table S3 and Figure 3).

Data from a total of 41 coral snakes (Supplementary Table S4) were used in the phylogenetic analysis, including *Micruroides euryxanthus* as an outgroup, according to the phylogenetic analysis performed by Jowers et al. [31]. PhyloSuite v 1.2.3 [72] was used for data standardization and concatenation. The CDS genes were aligned using the MACSE algorithm [73] with the vertebrate mitochondrial genetic code and standard code for nuclear genes. We concatenated the *cytb* and *nd4* genes for species with at least two genes sequenced. Bayesian inference (BI) tree was reconstructed using MrBayes 3.2.7 [74], and fitting substitution models were determined in MEGA v 11.0.13 [75] with the Bayesian information criterion (BIC). BI analysis using the default settings by four simultaneous Markov chains was run for five million generations in two independent runs, with sampling every 1000 generations, and the initial 25% of samples were discarded as burn-in. Posterior probabilities were calculated from the consensus of the remaining trees. The confidence of the Bayesian sampling was verified for the free parameters using the effective sample size statistic (ESS) implemented in the software Tracer v.1.5 [76]. All parameters showed ESS greater than 300, and the analyses converged asymptotically, indicating reliable performance.

5.3. Characterization of *M. nigrocinctus* Venom from Colombia and Its Immunorecognition by Commercial Coral Snake Antivenoms

5.3.1. Venoms and Antivenoms

Venom was manually extracted from two *M. nigrocinctus* individuals from the municipalities of Apartadó and Carepa Colombia, located in the department of Antioquia, Colombia. Both specimens were maintained in captivity at the institutional serpentarium of the University of Antioquia, collection Licensee: No. 0524 27 May 2014) and No. 001566 (24 July 2024). A venom pool from Costa Rican specimens of *M. nigrocinctus* was kindly provided by Instituto Clodomiro Picado, University of Costa Rica. Two commercially available equine anticoral antivenoms were evaluated: (a) INS-antivenom (produced by Instituto Nacional de Salud, Colombia; batch N°23AMP01, expiry date November 2025) and ICP-antivenom (produced by Instituto Clodomiro Picado, Costa Rica; batch 7040723ACLQ, expiry date 26 July). Both antivenoms were used before their expiration dates.

5.3.2. Venom Chromatographic and Electrophoretic Profiles

The chromatographic profiles of the venoms of *M. nigrocinctus* from Colombia and Costa Rica were compared. Two mg of each venom were dissolved in 200 μ L of 0.1% trifluoroacetic acid (Solution A; TFA), centrifuged at 1250 \times g for 5 min, and fractionated on a C₁₈ column (250 \times 4.6 mm, 5 μ m particle size; Phenomenex, Torrance, CA, USA) using an Agilent 1220 equipment, with monitoring at 215 nm. Elution was performed at a flow rate of 1 mL/min, applying the following gradient toward Solution B (acetonitrile containing 0.1% TFA): 5% B for 5 min, 5–15% B for 10 min, 15–45% B for 60 min, and 45–70% B for 12 min [77]. Also, 30 μ g of each venom was analyzed by SDS-PAGE under non-reducing conditions using a 15% gel in a Mini-Protean Tetra Cell electrophoretic system (Bio-Rad, Hercules, CA, USA) at 150 volts. The Precision Plus Protein™ Standards (Broad Range, Bio-Rad, Hercules, CA, USA) were used as molecular weight markers, and the proteins were visualized by Coomassie Blue R-250 staining.

5.3.3. Shotgun Proteomic Profiling

Venom samples of 15 µg of *M. nigrocinctus* from Colombia or Costa Rica, respectively, were concentrated into a single band by SDS-PAGE under reducing conditions after entering the stacking gel. The concentrated bands were visualized with Coomassie R-250 staining, excised from the gel, and subjected to reduction with 10 mM of dithiothreitol for 30 min at 56 °C and alkylation with 50 mM of iodoacetamide for 20 min in the dark, followed by overnight digestion with sequencing-grade trypsin at 37 °C in an automated workstation (Intavis., Waldhäuser, Tübingen, Germany). The resulting peptides were analyzed by nESI-MS/MS using a nano-Easy[®] 1200 chromatograph in line with a Q-Exactive Plus[®] mass spectrometer (Thermo Scientific; Waltham, MA, USA). An amount of 5 µL of each digest was loaded onto a C₁₈ trap column (75 µm × 2 cm, 3 µm particle size; PepMap, Thermo Scientific; Waltham, MA, USA), washed with 0.1% formic acid (solution A), and separated at a flow rate of 200 nL/min using a C₁₈ Easy-spray[®] column (15 cm × 75 µm, 3 µm particle size). Separation was achieved with a gradient toward solution B (80% acetonitrile, 0.1% formic acid) developed in a total of 120 min (1–5% B in 1 min, 5–26% B in 84 min, 26–80% B in 30 min, 80–99% B in 1 min, and 99% B for 4 min). MS spectra were acquired in positive mode at 1.9 kV, with a capillary temperature of 200 °C, using 1 µscan in the range 400–1600 m/z, maximum injection time of 50 msec, AGC target of 1 × 10⁶, and resolution of 70,000. The top 10 ions with 2–5 positive charges were fragmented with an AGC target of 3 × 10⁶, minimum AGC 2 × 10³, maximum injection time 110 ms, dynamic exclusion time 5 s, and resolution 17,500. MS/MS spectra were processed against protein sequences contained in the Serpentes UniProt/SwissProt database (<https://www.uniprot.org/blast>, accessed on January 2024) using PEAKS X (Bioinformatics Solutions), and matches were assigned to known protein families by similarity. Cysteine carbamidomethylation was set as a fixed modification, while deamidation of asparagine or glutamine and methionine oxidation were set as variable modifications, allowing up to 3 missed cleavages by trypsin. Parameters for match acceptance were set to FDR < 0.1%, detection of at least 1 unique peptide, and −10lgP protein score ≥ 30.

5.3.4. Biological Activities of *M. nigrocinctus* Venom from Colombia

The PLA₂ activity of *M. nigrocinctus* venom from Colombia was tested using the monodisperse synthetic substrate 4-nitro-3-octanoyloxy-benzoic acid (4-NOBA). An amount of 20 µg of venom was dissolved in 25 µL of buffer (10 mM Tris, 10 mM CaCl₂, 100 mM NaCl, pH 8.0) and added to microplate wells (in triplicate), mixed with 25 µL of the substrate (1 mg/mL in acetonitrile) and 200 µL of the same buffer. After incubation for 60 min at 37 °C, absorbances were recorded at 405 nm using a microplate reader, and activity was expressed as the absorbance change relative to the negative control (substrate alone) [78]. The venom of *M. nigrocinctus* from Costa Rica was compared under the same conditions.

The edema-forming activity was evaluated in the mouse footpad assay [79]. In brief, 5 µg of *M. nigrocinctus* venom from Colombia or Costa Rica, dissolved in 50 µL of saline solution, was injected subcutaneously into the right footpad in groups of three mice of 18–20 g body weight. As a negative control, the left footpad was injected with the same volume of saline solution. The progression of edema was evaluated by measuring the footpad thickness with a caliper at 0.5, 1, 2, and 4 h after the injection.

The myotoxicity activity was evaluated using a group of three mice that received an intramuscular injection containing 10 µg of *M. nigrocinctus* venom from Colombia or Costa Rica (in 50 µL Buffer PBS) into the gastrocnemius. Control mice received an injection of PBS. Blood was collected after 1.5 h from the tip of the tail into heparinized capillaries, and the activity of creatine kinase (CK) in plasma was determined using a UV kinetic assay (CK-Nac, Wiener) [80].

To evaluate the lethal activity, various amounts of *M. nigrocinctus* venom from Colombia (from 3.5 to 56 µg), dissolved in 250 µL of saline solution, were injected intraperitoneally (i.p.) in groups of four mice (16–18 g of body weight). Deaths were recorded after a 48 h observation period, and the median lethal dose (LD₅₀) was calculated by probits [81]. Animal experiments were conducted under a study protocol approved by the Institutional Committee for the Care and Use of Laboratory Animals (CICUA) from the University of Antioquia (License No. 160 of 2024).

5.3.5. Venom Immunorecognition and Neutralization by Commercial Anticoral Antivenoms

The antibody titers of the ICP-antivenom and INS-antivenom, respectively, were assessed against whole *M. nigrocinctus* venoms from Colombia and Costa Rica using an enzyme-linked immunosorbent assay (ELISA). Each microplate well was coated with 0.1 µg of complete venom diluted in 100 µL of coating buffer (0.1 M Tris, 0.15 M NaCl, pH 9.0) and incubated overnight at room temperature. The wells were blocked with 100 µL of 1% bovine serum albumin in phosphate buffer (BSA-PBS; 0.04 M phosphates, 0.12 M NaCl, pH 7.2) for 90 min. Serial dilutions of each antivenom or a non-immune equine serum as a negative control (1:500 to 1:1.093.500) were added to the wells and incubated for 90 min at room temperature. After washing, a peroxidase-labeled anti-horse IgG conjugate (1:8000; Sigma-Aldrich., Darmstadt, Germany) was added as the secondary antibody and incubated for 90 min at room temperature. Following a final wash, 100 µL of peroxidase substrate (2 mg/mL of *o*-phenyldiamine diluted in 0.1 M sodium citrate, pH 5.0; 4 µL of 30% H₂O₂ per 10 mL of final solution) was added for color development. The absorbance was measured at 492 nm using a Multiskan Sky spectrophotometer (Thermo Scientific; Waltham, MA, USA).

The ability of commercial equine antivenoms to neutralize the lethal effect of *M. nigrocinctus* was evaluated by preincubation experiments. Groups of four mice (16–18 g of body weight) were injected intra-peritoneally (i.p.) with 250 µL of a solution containing 25.8 µg of *M. nigrocinctus* venom (equivalent to a challenge of 3 × LD₅₀), previously incubated for 30 min at 37 °C with the antivenoms (ICP or INS) in a proportion of 0.2 mg of venom per mL of antivenom. A group of control mice received the same dose of venom and were incubated only with saline solution. Deaths were recorded after 48 h.

Supplementary Materials: The following supporting information can be downloaded at: <https://www.mdpi.com/article/10.3390/toxins17060268/s1>, Figure S1: *M. nigrocinctus* from Córdoba. Individual deposited in the collection of the Serpentarium of the University of Antioquia—SUA #1930; Table S1: Database of distribution of *M. nigrocinctus* based on “citizen science” and scientific database reports; Table S2: Protein matches obtained by nESI-MS/MS shotgun analysis of tryptic peptides derived from *Micrurus nigrocinctus* (Colombia) whole venom; Table S3. Primers and accession numbers for the new sequences deposited in GenBank; Table S4. Species, countries, localities, and GenBank accession numbers used in this study. The order of the species corresponds from top to bottom in the phylogenetic tree.

Author Contributions: Conceptualization, P.R.-S. and J.G.-R.; methodology, P.R.-S., L.P.R., J.G.-R., S.P.-M., Y.F.-F., E.P.-C., M.S.-C., B.L. and J.F.; formal analysis, P.R.-S. and B.L.; resources, P.R.-S. and V.N.; data curation, J.G.-R. and S.P.-M.; writing—original draft preparation, P.R.-S., M.S.-C, L.P.R., and J.G.-R.; writing—review and editing, P.R.-S., B.L., L.P.R. and M.S.; funding acquisition, P.R.-S. and M.S.-C. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by the Ministerio de Ciencia y Tecnología of Colombia (Min-Ciencias) under Grant Number 101750 (228-2023) from the “Programa Orquídeas, Mujeres en la Ciencia: Agentes para la Paz”, the Agencia Nacional de Investigación y Desarrollo (ANID) with Grant FONDECYT REGULAR No. 1220921, and the University of Antioquia (UdeA).

Institutional Review Board Statement: The animal study protocol was approved by the Institutional Committee for the Care and Use of Laboratory Animals (CICUA) from the University of Antioquia (License No. 160 of 2024).

Informed Consent Statement: Not applicable.

Data Availability Statement: The original contributions presented in this study are included in this article and Supplementary Material. Further inquiries can be directed to the corresponding author.

Acknowledgments: The authors thank the communities of Apartadó, Carepa, and Turbo, as well as the educational institutions of Apartadó, for the participation of children and young people. Henry Roso, from the Center for Ecological and Agro-Environmental Studies and the University of Antioquia.

Conflicts of Interest: The authors declare no conflicts of interest.

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M. nigrocinctus from Córdoba

Individual deposited in the collection of the Serpentarium of the University of Antioquia - SUA

#1930



19	C6JUP3	56.7	20	6.76E+06	1	1	3	9070	3FTx	Three-finger toxin McTx3, <i>Micrurus corallinus</i>	E.PPGTLETC(+57.02)PDDFTC(+57.02)VK.K	56.7	1835.81	16	3.2	918.91	2	CAM			
20	A0AGF7YYW:	65.3	7	2.13E+08	2	2	10	15817	CNP	Natriuretic peptide, <i>Micrurus fulvius</i>	L.GDGC(+57.02)FGQRIDR.I G.DGC(+57.02)FGQRIDR.I	41.9 44.3	1279.57 1222.55	11 10	0.6 0.7	427.53 408.52	3 3	CAM CAM			
21	A0A194ARC7	288.6	68	6.60E+09	37	7	423	16197	PLA2	phospholipase A2, <i>Micrurus tener</i>	R.VHKC(+57.02)FPSMTMYSYDC(+57.02)SEGKLT(+57.02)K K.C(+57.02)FPSMTMYSYDC(+57.02)SEGKLT(+57.02)KDNH R.VHKC(+57.02)FPSMTMYSYDC(+57.02)SEGKLT(+57.02)SE K.RVHKC(+57.02)FPSMTMYSYDC(+57.02)SEGK.L K.C(+57.02)FPSMTMYSYDC(+57.02)SEGKLT(+57.02)K.D R.VHKC(+57.02)FPSMTMYSYDC(+57.02)SEGK.L K.C(+57.02)FPSMTMYSYDC(+57.02)SEGK.L	53.0 79.4 68.2 76.6 50.7 61.7 55.8	3400.47 3038.24 2860.20 2482.05 2463.99 2325.95 1961.73	28 25 23 20 20 19 16	1.7 2.5 2.4 2.8 2.7 3.8 4.1	851.13 760.07 716.06 621.52 822.34 582.50 981.88	4 4 4 4 3 4 4			CAM CAM CAM CAM CAM CAM CAM	
22	P21790	168.6	56	2.68E+10	7	3	192	3314	PLA2	Phospholipase A2 1 (Frag), <i>Micrurus nigrocinctus</i>	NLYQFKNMIQC(+57.02)ITTKR.S N.LYQFKNMIQC(+57.02)ITTKR.S L.YQFKNMIQC(+57.02)ITTKR.S	68.9 46.8 48.4	1943.97 1829.93 1716.84	15 14 13	1.1 0.6 0.2	972.99 458.49 573.29	2 4 3			CAM CAM CAM	
23	C0HKB8	248.0	52	8.09E+09	21	6	228	13188	PLA2	Acidic phospholipase A2, <i>Micrurus dumerilii</i>	R.TAAIC(+57.02)FAKAPYDDNFMNPNRC(+57.02)Q A.IC(+57.02)FAKAPYDDNFMNPNRC(+57.02)Q R.TAAIC(+57.02)FAKAPYDDNFMNPNRC F.AKAPYDDNFMNPNRC(+57.02)Q K.APYDDNFMNPNRC(+57.02)Q K.APYDDNFMNPNRC	48.7 47.7 48.4 47.7 73.3 80.2	2830.26 2603.14 2542.17 2182.95 1967.83 1679.74	24 21 22 18 16 14	10 1.3 12 0.5 2.4 3	944.44 868.72 848.41 728.66 984.92 640.88	3 3 3 3 2 2			CAM CAM CAM CAM CAM CAM	
24	A0A194AP65	228.3	52	6.71E+07	17	1	187	15781	PLA2	phospholipase A2, <i>Micrurus tener</i>	N.C(+57.02)KAFVC(+57.02)NC(+57.02)DRTAALC(+57.02)Fg	58.0	2176.96	18	1.4	726.66	3	CAM	A0A194AT46; A0A194AT52; A0A194AS63; A0A194AS63; A0A194AT50		
25	A0A194ARA5	211.0	52	8.77E+06	14	2	145	15708	PLA2	phospholipase A2, <i>Micrurus tener</i>	N.C(+57.02)KAFVC(+57.02)N(+98)C(+57.02)GRTAALC(+57.0 K.AFVC(+57.02)N(+98)C(+57.02)GRTAALC(+57.02)FGKA	41.7 47.9	2119.94 1831.82	18 16	3.1 1	530.99 611.61	4 3	CAM; Deam (NQ) CAM; Deam (NQ)			
26	A0A2D4N7P6	196.2	61	1.74E+06	16	1	124	11841	PLA2	phospholipase A2 (Frag), <i>Micrurus spixii</i>	K.DHGC(+57.02)WPK.W	48.6	898.38	7	-1.9	450.19	2	CAM			
27	A0A2D4Q1F6	232.3	34	9.29E+08	15	4	110	17844	PLA2	PLA2 domain-containing prot (Frag), <i>Micrurus surinamensis</i>	K.QC(+57.02)YDEAEKVHGC(+57.02)KPLVMFYSEC(+57.02 G1.CC(+57.02)YDEAEKVHGC(+57.02)KPL.V R.C(+57.02)YDEAEKVHGC(+57.02)YDEAEK.V K.QC(+57.02)YDEAEKVHGC	49.0 61.0 55.7 39.4	3052.33 1832.82 1816.72 1334.56	24 15 14 11	3.3 -0.4 1.5 0.7	764.09 459.21 606.58 445.86	4 4 3 3			CAM CAM CAM CAM	
28	G9I930	229.5	34	3.73E+08	14	3	100	16793	PLA2	Basic PLA2 homolog MitX-beta, <i>Micrurus tener tener</i>	D.FVDYGC(+57.02)YIC(+57.02)VAR.D F.VDYGC(+57.02)YIC(+57.02)VAR.D D.YGC(+57.02)YIC(+57.02)VAR.D	47.2 49.3 47.7	1408.59 1281.52 1047.43	11 10 8	1.5 0.7 0.7	705.30 631.77 524.72	2 2 2			CAM CAM CAM	
29	A0A194ARA9	222.8	52	4.34E+07	17	1	88	16003	PLA2	phospholipase A2, <i>Micrurus tener</i>	R.C(+57.02)YIC(+57.02)QVHDDC(+57.02)YGEAEKIDGC(+57.02 K.C(+57.02)KDFVC(+57.02)NC(+57.02)DRVAANC(+57.02)FAJ	40.8 44.4	2554.98 2233.95	21 18	1.7 14.8	852.67 559.50	3 4			CAM CAM	
30	A0A2D4PZ69	141.7	30	0.00E+00	7	1	60	14834	PLA2	phospholipase A2 (Frag), <i>Micrurus surinamensis</i>	K.C(+57.02)KDFVC(+57.02)NC(+57.02)DRVAANC(+57.02)FAJ	40.8	2233.95	18	14.8	559.50	4	CAM			
31	A0A2H6MVX1	196.7	54	2.32E+08	12	1	67	12150	PLA2	phospholipase A2 (Frag), <i>Micrurus lemniscatus carvalhoi</i>	R.RSAWDFTNVGC(+57.02)YIC(+57.02)GAAGSGTPVDELDR.	56.0	3010.26	27	6.2	1004.43	3	CAM	A0A2H6MVP9		
32	A0A289ZBS3	156.3	42	9.93E+07	7	4	16	13449	PLA2	phospholipase A2 (Frag), <i>Micrurus laticollaris</i>	K.C(+57.02)KDFVC(+57.02)NC(+57.02)DLVAANC(+57.02)FAJ H.YNDNYNDLKR.C Y.NDDNYNDLKR.C D.DNYNDLKR.C	61.8 48.0 49.9 40.6	2190.93 1541.71 1378.65 1149.58	18 12 11 9	-1.2 3.1 0 0	731.32 514.91 690.33 575.80	3 3 2 2			CAM CAM CAM CAM	
33	Q8JFB2	118.3	13	2.70E+07	5	1	7	16430	PLA2	Phospholipase A2 GL16-1, <i>Laticauda semifasciata</i>	A.DYGC(+57.02)YIC(+57.02)GAGSGTPVDELDR.C	43.4	2147.85	20	3.6	1074.94	2	CAM			
34	A0A2H6N4A4	77.8	36	6.86E+06	3	1	5	11369	PLA2	phospholipase A2 (Frag), <i>Micrurus lemniscatus carvalhoi</i>	Y.SYEC(+57.02)SEGKLT(+57.02)KNDTKC(+57.02)K.E	43.2	2321.99	19	-11	775.00	3	CAM			
35	G9I929	171.0	43	4.51E+09	7	7	100	9498	KUN	Kunitz-type neurotoxin MitX-alpha, <i>Micrurus tener tener</i>	L.TPVSSQIRPAPFC(+57.02)YEDPPFFQK.C S.SQIRPAPFC(+57.02)YEDPPFFQK.C S.QIRPAPFC(+57.02)YEDPPFFQK.C Q.NHFTTMSEC(+57.02)NRCV(+57.02)HG R.PAPFC(+57.02)YEDPPFFQK.C A.FC(+57.02)YEDPPFFQK.C Q.NHFTTMSEC(+57.02)NRC.V	63.4 66.1 64.6 54.1 58.9 45.6 61.1	2513.20 2129.60 2041.97 1848.75 1644.73 1476.64 1395.57	21 17 16 15 13 11 11	0.8 1.6 3.2 1.7 12.5 3 0.9	838.74 710.68 681.67 617.26 823.38 739.33 698.79	3 3 3 3 2 2 2				CAM CAM CAM CAM CAM CAM CAM
36	U3EPJ0	192.0	30	7.05E+09	8	7	71	8912	KUN	Kunitz inhibitor 6, <i>Micrurus fulvius</i>	K.QFVYGGC(+57.02)GGNANNFKTIDEC(+57.02)KR.T K.QFVYGGC(+57.02)GGNANNFKTIDEC(+57.02)KR.R R.KC(+57.02)KQFVYGGC(+57.02)GGNANNFK.T K.C(+57.02)KQFVYGGC(+57.02)GGNANNFK.T K.QFVYGGC(+57.02)GGNANNFK.T Q.FVYGGC(+57.02)GGNANNFK.T F.VYGGC(+57.02)GGNANNFK.T	80.8 59.5 54.8 51.8 65.8 65.4 49.7	2534.14 2378.04 2047.94 1919.84 1631.72 1503.66 1356.59	22 21 18 17 15 14 13	1.6 2.6 2.1 3.9 0.4 0.4 3.2	634.54 793.69 683.65 640.96 544.91 752.84 679.30	4 3 3 3 3 2 2			CAM CAM CAM CAM CAM CAM CAM	
37	A0A194AR89	124.8	49	1.35E+08	4	4	13	9147	KUN	Kunitz-type protease inhibitor 5, <i>Micrurus tener</i>	L.RRPEFC(+57.02)NLPVAVTGPC(+57.02)K.A R.IC(+57.02)QEFVYGGC(+57.02)KGNANK.F R.RPEFC(+57.02)NLPVAVTGPC(+57.02)K.A R.AFYNSVLR.I	56.3 45.2 43.8 64.0	1900.94 1857.85 1744.84 1131.57	16 16 15 9	-3.6 2.1 0.9 1.7	476.24 620.29 582.62 566.79	4 3 3 2			CAM CAM CAM CAM	
38	U3F589	91.6	20	1.77E+08	3	3	12	9303	KUN	Kunitz inhibitor 7, <i>Micrurus fulvius</i>	L.RGPKYCI(+57.02)YLPADPGPC(+57.02)RR.Y K.YC(+57.02)YLPADPGPC(+57.02)RR.Y K.YC(+57.02)YLPADPGPC(+57.02)RR.R	41.9 52.0 51.2	2062.00 1623.73 1467.63	17 13 12	1.2 0.2 2.4	516.51 542.25 734.82	4 3 2			CAM CAM CAM	
39	A0A194ARF4	79.1	17	1.10E+07	1	1	5	8961	KUN	Kunitz-type protease inhibitor 3, <i>Micrurus tener</i>	K.ANFPAPYDPASHK.C	63.8	1626.75	14	0.8	543.26	3				
40	U3FAC8	65.0	13	1.81E+07	1	1	2	9563	KUN	Kunitz inhibitor 5, <i>Micrurus fulvius</i>	K.TMREC(+57.02)NRCV(+57.02)HG	42.5	1418.60	11	0.7	473.87	3	CAM	A0A194ARF8; A0A194ASB8; U3EPJ7; U3FAD2; A0A0F7Y2Z3; A0A194AT92; A0A194AR93		
41	A0A194AT88	61.4	14	2.57E+06	1	1	1	9537	KUN	Kunitz-type protease inhibitor 4, <i>Micrurus tener</i>	C.SQFNVGGC(+57.02)DGNK.N	44.2	1345.54	12	0.6	673.78	2	CAM	A0A0F7YYW6; U3F5C3		
42	U3FYN7	191.9	34	8.87E+08	10	10	61	22516	VEGF	Vascular endothelial growth factor 1, <i>Micrurus fulvius</i>	F.QEYDVEVEYFKPSC(+57.02)LLMK.C K.LKHFSQHIHMFQOHSK.C K.HFOSQHIHMFQOHSK.C K.HFOSQHIHMFQOHS.S R.RKHL.YKQDPLTC(+57.02)K.C R.KQENHC(+57.02)EPC(+57.02)SER.R I.KQENHC(+57.02)EPC(+57.02)SER.R R.KHLYKQDPLTC(+57.02)K.C K.QEN(+98)HC(+57.02)EPC(+57.02)SER.R K.HLYKQDPLTC(+57.02)K.C	49.1 42.6 63.2 47.5 35.9 47.4 38.1 59.6 51.4 72.5	2497.21 2344.16 2102.99 1887.86 1685.90 1685.73 1572.64 1529.80 1445.53 1401.71	20 19 17 15 13 13 12 12 11 11	8.8 0.2 1.8 1.6 -0.2 0.7 -0.1 1.1 11.3 -0.2	830.08 587.05 526.75 472.97 422.48 562.92 525.22 510.94 723.78 701.86	3 4 4 4 4 3 3 3 3 2			CAM CAM CAM CAM CAM CAM CAM CAM CAM CAM	
43	U3FAK1	124.9	6	1.76E+07	3	7	47564	VEGF	Vascular endothelial growth factor 2, <i>Micrurus fulvius</i>	K.GGVTSPSC(+57.02)GIHKELDR.T R.FHHQTC(+57.02)SC(+57.02)YR.R K.GGVTSPSC(+57.02)GIHKE	54.0 68.5 46.9	1711.83 1394.56 1198.58	16 10 12	1 -0.2 0.2	428.97 465.86 400.53	4 3 3			CAM CAM CAM		

Supplemental Table S2. Protein matches obtained by nESI-MS/MS shotgun analysis of tryptic peptides derived from *Micurus nigrocinctus* (Costa Rica) whole venom.

Prot Group	Accession	-10lgP	Cov (%)	Area	#Pept	#Unique	#Spectr	Avg. Mass	Pr.family	Matching protein, Species	Supporting unique peptides	-10lgP	Mass	Length	ppm	m/z	z	PTM	Indistinguishable matching proteins
1	COHK04	253.4	100	1.77E+08	36	3	369	7543	3FTx	Clark toxin-1, <i>Micurus clarki</i>	K.GC(+57.02)ASSC(+57.02)PKNGFKFKIEC(+57.02)C(+57.02)TK.D K.NGFIKFKIEC(+57.02)C(+57.02)TKDNC(+57.02)I K.NGFIKFKIEC(+57.02)C(+57.02)TK.D	64.8 60.7 72.5	2604.23 2259.09 1756.90	22 18 14	2.0 10.6 5.1	652.07 754.04 586.64	4 3 3	CAM	U3EPK7; A0A0F7YYX2; A0A0F7YZ34;
2	COHLK5	240.5	100	4.63E+08	35	6	369	7544	3FTx	Clark toxin-I-Mdm, <i>Micurus dumenilli</i>	K.DGFIKFKIEC(+57.02)C(+57.02)YKNC(+57.02)I K.GC(+57.02)ASSC(+57.02)PKDGF.KI K.GC(+57.02)ASSC(+57.02)PKDGF.KI K.KG.C(+57.02)ASSC(+57.02)PKDGF.I K.DGFIKFK.I K.DGFIKFK.K	39.8 66.4 57.6 29.5 39.5 25.6	2260.07 1553.73 1425.64 1312.55 966.55 838.46	18 14 13 12 8 7	6.0 2.6 1.0 3.2 9.6 9.5	566.03 518.92 713.83 438.53 484.29 420.24	4 3 2 3 2 2	CAM	
3	A0A0H4BEF7	280.4	75	1.11E+10	31	20	366	9303	3FTx	Three-finger toxin D.I, <i>Micurus diastema</i>	K.SAVFETTC(+57.02)LPGLHKVC(+57.02)YKRWHPGAVGC(+57.02)AVTC(+57.02)F R.WHIGHPGVAGC(+57.02)AVTC(+57.02)PRRYLIEVEC(+57.02)C(+57.02)ATDKC(+57.02)I K.VC(+57.02)YKRWHPGAVGC(+57.02)AVTC(+57.02)AVTC(+57.02)PRR R.RYLIEVEC(+57.02)C(+57.02)ATDKC(+57.02)NR R.YLIEVEC(+57.02)C(+57.02)ATDKC(+57.02)NR R.RYLIEVEC(+57.02)C(+57.02)ATDKC(+57.02)NR K.RWHPGAVGC(+57.02)AVTC(+57.02)PRR R.RYLIEVEC(+57.02)C(+57.02)ATDKC(+57.02)NR Y.LIEVEC(+57.02)C(+57.02)ATDKC(+57.02)NR R.WHIGHPGVAGC(+57.02)AVTC(+57.02)PRR R.RYLIEVEC(+57.02)C(+57.02)ATDKC W.WHIGHPGVAGC(+57.02)AVTC(+57.02)PRR R.YLIEVEC(+57.02)C(+57.02)ATDKC R.WHIGHPGVAGC(+57.02)AVTC Y.LIEVEC(+57.02)C(+57.02)ATDKC K.RWHPGAVGC(+57.02)A R.WHIGHPGVAGC(+57.02)A.V R.WHIGHPGVAGC(+57.02)A R.WHIGHPGVAGC R.WHIGHPGVA.G	29.2 25.3 57.5 77.3 66.2 40.2 68.7 40.2 56.5 82.4 71.0 75.1 62.9 32.6 40.3 46.5 58.7 37.4 40.4 38.9	4351.10 4054.90 2580.24 2199.02 2042.92 2042.92 2029.98 1886.82 1879.86 1873.88 1768.85 1687.80 1612.75 1460.70 1449.68 1345.65 1260.58 1189.55 1029.51 972.49	38 34 22 17 16 16 18 15 15 14 17 16 13 14 12 12 12 11 10 9	14.0 13.9 4.0 1.2 12.5 14.6 3.9 7.6 4.1 9.4 1.2 1.5 8.0 2.0 6.6 -0.6 0.9 2.1 2.7 4.7	1088.80 812.00 646.07 550.76 681.99 1022.48 508.51 629.95 627.63 625.64 590.63 553.61 538.59 487.91 725.85 449.56 421.20 595.78 515.77 487.26	4 5 4 4 3 2 2 3 3 3 3 3 3 2 2 2 3 2 2 2	CAM	
4	A0A0H4BD8	249.6	69	1.08E+10	22	9	271	10798	3FTx	Three-finger toxin B.B, <i>Micurus browni</i>	R.AIEGFC(+57.02)AASC(+57.02)PKVGLPHVTC(+57.02)C(+57.02)SADNC(+57.02)NSR.I K.VGLGPHVTC(+57.02)C(+57.02)SADNC(+57.02)NSRN(+98)FLK K.VGLGPHVTC(+57.02)C(+57.02)SADNC(+57.02)NSRN.F K.VGLGPHVTC(+57.02)C(+57.02)SADNC(+57.02)NSRN.F K.VGLGPHVTC(+57.02)C(+57.02)SADNC(+57.02)NSRN K.VGLGPHVTC(+57.02)C(+57.02)SADN.C K.VGLGPHVTC(+57.02)C(+57.02)SAD.N K.VGLGPHVTC(+57.02)C(+57.02)S K.VGLGPHVTC(+57.02)C	40.7 25.1 34.9 48.6 67.1 32.2 36.0 48.5 38.9	3294.41 2506.12 2263.95 2116.88 2002.84 1485.63 1371.59 1098.50 938.46	30 22 20 19 18 14 13 10 9	3.6 1.8 2.6 2.1 1.5 0.6 2.6 0.2 1.6	824.61 627.54 755.66 706.64 1002.43 743.82 686.80 550.25 470.24	4 4 3 3 2 2 2 2 2	CAM	Deam (NQ)
5	P80548	225.9	90	1.36E+10	19	6	233	6583	3FTx	Three-finger toxin Mm I, <i>Micurus nigrocinctus</i>	MIC(+57.02)HMQSSQPPTIKT(+57.02)SEGCG(+57.02)YK.K MIC(+57.02)HMQSSQPPTIKT(+57.02)SEGCG(+57.02)YK.K MIC(+57.02)HMQSSQPPTIKT M.IC(+57.02)HMQSSQPPTIKT H.NQSSQPPTIKT N.QQSSQPPTIKT	65.3 58.7 64.6 43.8 37.5 35.3	3009.36 2881.26 1767.84 1636.80 1226.63 1112.58	25 24 15 14 11 10	3.1 1.7 1.8 2.8 -0.1 3.3	753.35 721.32 884.93 546.61 614.32 557.30	4 4 2 3 2 2	CAM	
6	A0A194ARK4	220.0	73	2.48E+09	17	2	172	8761	3FTx	Three-finger toxin 16d, <i>Micurus tener</i>	K.YAVGLMPTGTMVYHGG(+57.02)ASTC(+57.02)HR.G R.GKYVVC(+57.02)C(+57.02)STDLCC(+57.02)NK	74.7 66.4	2596.12 1717.72	22 14	7.1 2.0	650.04 859.87	4 2	CAM	
7	A0A0H4IS74	180.3	68	5.84E+08	12	12	168	8737	3FTx	Three-finger toxin B.F, <i>Micurus browni</i>	R.IGKDGFSVTC(+57.02)TEKENLC(+57.02)FTM(+15.99)FSARNPAQIER.G R.IGKDGFSVTC(+57.02)TEKENLC(+57.02)FTMFSAR.N K.DGFSVTC(+57.02)TEKENLC(+57.02)FTMFSAR.N R.GC(+57.02)ASSC(+57.02)SSRYM(+15.99)KC(+57.02)C(+57.02)STDSK(+57.02)JNG R.IGKDGFSVTC(+57.02)TEK.E R.NMKC(+57.02)C(+57.02)STDSK(+57.02)JNG R.GC(+57.02)ASSC(+57.02)SSRYM.K.C K.ENLC(+57.02)FTM(+15.99)FSAR.N K.DGFSVTC(+57.02)TEK.E N.LC(+57.02)FTMFSAR.N K.ENLC(+57.02)FTMF.S R.GC(+57.02)ASSC(+57.02)SSR.Y	43.5 65.5 38.2 21.5 73.5 25.2 43.2 53.7 67.4 34.4 20.9 48.1	3897.85 2960.35 2662.15 2449.85 1603.76 1481.50 1392.56 1390.60 1305.55 1131.52 1060.44 970.36	33 25 22 21 14 12 12 11 11 9 8 9	10.6 2.7 6.3 1.4 1.1 3.2 2.4 6.0 2.3 0.8 11.4 -2.0	975.48 741.10 888.40 817.62 802.89 741.76 697.29 696.31 653.79 566.77 531.23 486.19	4 4 3 3 2 2 2 2 2 2 2 2	CAM; Ox (M)	A0A194ARE8
8	A0A194ARD0	204.2	43	4.75E+08	16	1	162	9142	3FTx	Three-finger toxin 17a, <i>Micurus tener</i>	E.GAYNVC(+57.02)C(+57.02)STDLCC(+57.02)NK.S	51.7	1660.66	14	2.2	831.34	2	CAM	A0A194ARU; A0A194APH0; A0A194ARD4; A0A194ATC3; A0A194ATC0
9	A0A194ATB0	188.2	70	7.05E+06	15	1	157	8925	3FTx	Three-finger toxin 5a, <i>Micurus tener</i>	R.GC(+57.02)GC(+57.02)PTVPGIHSIC(+57.02)C(+57.02)TSDK.C	23.7	2232.97	20	3.4	559.25	4	CAM	
10	U3F5E9	226.4	49	2.35E+09	14	6	137	8666	3FTx	Three-finger toxin 13, <i>Micurus fulvius</i>	R.GRYQPMGC(+57.02)GC(+57.02)PESRR.G R.GRYQPMGC(+57.02)GC(+57.02)PESRR.G R.YQPMGC(+57.02)GC(+57.02)PESRR.G R.YQPMGC(+57.02)GC(+57.02)PESRR.G R.GRYQPMGC(+57.02)G.C R.GRYQPMGC	62.8 72.9 50.4 66.7 30.9 25.8	1938.82 1782.72 1725.70 1569.60 1153.46 936.41	16 15 14 13 10 8	1.4 2.9 3.6 1.3 -0.7 1.3	647.28 595.25 576.24 785.81 577.74 469.21	3 3 3 2 2 2	CAM	
11	A0A194APE1	180.2	51	1.65E+08	12	4	123	9898	3FTx	Three-finger toxin 9a, <i>Micurus tener</i>	K.NQNLC(+57.02)YKM(+15.99)FTTFPGFGWTQK.G T.RC(+57.02)LKQNLC(+57.02)YKM R.C(+57.02)LKQNLC(+57.02)YKM K.NQNLC(+57.02)YKM	20.7 33.9 65.7 44.2	2596.22 1495.74 1339.64 938.43	21 11 10 7	11.9 0.8 2.1 0.5	866.43 499.59 670.83 470.22	3 3 2 2	CAM; Ox (M)	A0A0H4BEG1
12	A0A0H4ISB0	217.0	75	1.64E+07	13	2	117	9282	3FTx	Three-finger toxin T.B, <i>Micurus tener</i>	R.RYLEVEC(+57.02)C(+57.02)ATDKC(+57.02)NR R.WHMGPGVAGC(+57.02)AVTC(+57.02)PRR	45.5 67.0	2182.99 1868.82	17 17	5.1 2.4	728.68 623.95	3 3	CAM	
13	A0A194ARH7	190.0	59	2.48E+08	13	7	115	9897	3FTx	Three-finger toxin 8a, <i>Micurus tener</i>	R.C(+57.02)PESTPKKVC(+57.02)C(+57.02)ATNCC(+57.02)I T.ANTLMC(+57.02)DNSNPSIRTPK.R.C T.ANTLMC(+57.02)DNSNPSIRTPK.R K.IFTTFPEFGWQK.G	39.8 30.2 64.0 66.8	2280.96 2173.07 2016.97 1728.89	19 19 18 14	-0.9 -2.4 3.1 7.2	1141.49 544.28 673.33 577.31	2 4 3 3	CAM	

26	C6IUP2	143.7	43	1.01E+08	7	2	26	9984	3FTx	Three-finger toxin 3FTx-2, <i>Micurus corallinus</i>	K.GC(+57.02)HRC(+57.02)PESTPNEK.Y R.C(+57.02)PESTPNEK.Y	68.7 52.2	1683.75 1060.45	14 9	-0.4 -0.5	562.26 531.23	3 2	CAM	
27	A0A0G3VLL2	71.1	19	3.83E+08	3	3	20	8592	3FTx	Three-finger toxin LB, <i>Micurus laticollaris</i>	R.RGVQVSC(+57.02)C(+57.02)M(+15.99)IDKC(+57.02)JNG R.GVQVSC(+57.02)C(+57.02)M(+15.99)IDKC(+57.02)JNG R.GVQVSC(+57.02)C(+57.02)M(+15.99)IDK.C	41.4 41.2 27.4	1798.76 1642.66 1311.56	15 14 11	2.3 1.8 1.7	600.59 822.34 656.79	3 2 2	CAM; Ox (M) CAM; Ox (M) CAM; Ox (M)	
28	A0A194ATB7	134.4	50	1.51E+08	5	2	20	9220	3FTx	Three-finger toxin 1, <i>Micurus tener</i>	K.GGC(+57.02)TQEC(+57.02)PKETGMVLVC(+57.02)R.T K.ETTGMVLVC(+57.02)R.T	74.1 50.9	2195.98 1178.58	19 10	2.2 0.5	733.00 590.30	3 2	CAM	
29	P01425	68.0	25	7.39E+07	1	1	19	6838	3FTx	Short neurotoxin 1, <i>Hemachatus haemachatus</i>	LEC(+57.02)HNNQSSOPPTTK.S	68.0	1753.81	15	-9.0	585.60	3	CAM	P01426; P68417; P68418; P68419; P01427
30	U3FYQ6	137.1	52	3.74E+06	6	3	18	9302	3FTx	Three-finger toxin 4, <i>Micurus fulvius</i>	K.TC(+57.02)SKEKFC(+57.02)YLMVYVPSLNP.K.G K.EEKFC(+57.02)YLMVYVPSLNP.K.G K.FC(+57.02)YLMVYVPSLNP.K.G	64.7 44.9 59.6	2550.20 2073.99 1687.81	21 17 14	10.5 9.6 8.5	638.56 692.34 844.92	4 3 2	CAM CAM CAM	
31	A0A0H4B1D1	110.6	39	1.05E+05	4	1	18	9640	3FTx	Three-finger toxin L.F., <i>Micurus laticollaris</i>	K.C(+57.02)YEGETR.K.S	23.9	1098.48	9	-1.4	550.24	2	CAM	
32	U3FYR0	90.3	34	3.34E+09	4	4	15	8691	3FTx	Three-finger toxin 10, <i>Micurus fulvius</i>	T.VC(+57.02)LDLGHLLQC(+57.02)YVGR.D R.GC(+57.02)ASSC(+57.02)SPFYR.T R.GC(+57.02)ASSC(+57.02)SPF.Y T.LQC(+57.02)YVGR.D	22.6 58.0 22.4 38.4	1902.94 1290.51 971.35 894.44	16 11 9 7	-12.3 2.3 3.4 1.0	476.74 646.26 486.68 448.23	4 2 2 2	CAM CAM CAM CAM	
33	C6IUP1	67.5	38	1.04E+08	4	3	8	9311	3FTx	Three-finger toxin 3FTx-1, <i>Micurus corallinus</i>	S.LIC(+57.02)YNTM(+15.99)MQKVTCT(+57.02)PEGKDKC(+57.02)EK.Y I.C(+57.02)YNTMM(+15.99)QI(+98)KVTCT(+57.02)PEGKDKC(+57.02)EK.Y S.LIC(+57.02)YNTM(+15.99)QI(+15.99)QIQTCT(+57.02)PEGKDK.C	22.5 23.6 25.9	2748.24 2523.06 2347.07	22 20 19	-2.6 -3.0 -2.2	917.09 631.77 587.77	3 4 4	CAM; Ox (M) CAM; Ox (M); Deam (NQ) CAM; Ox (M)	
34	F5CPE2	65.3	18	6.12E+07	3	3	6	9083	3FTx	Three-finger toxin MALT0063C, <i>Micurus altirostris</i>	K.AGAYNIC(+57.02)C(+57.02)STDL.C(+57.02)NK.I A.GAYNIC(+57.02)C(+57.02)STDL.C(+57.02)NK.I A.YNIC(+57.02)C(+57.02)STDL.C(+57.02)NK.I	45.6 21.0 25.5	1745.72 1674.68 1546.62	15 14 12	9.4 2.6 0.4	582.92 838.35 774.32	3 2 2	CAM CAM CAM	
35	COHR2	79.5	88	1.56E+07	4	4	5	7196	3FTx	Micurotoxin 2, <i>Micurus mipartitus</i>	K.TC(+57.02)PFTTC(+57.02)PMSCC(+57.02)PGGOSIC(+57.02)YQR.K R.C(+57.02)IVANCI(+57.02)PFGSHDTSLLC(+57.02)C(+57.02)TR.D R.KWEEHGERIERR.C R.KWEEHGERIERR	23.4 42.3 31.9 41.5	2706.09 2324.98 1760.88 1604.78	23 20 13 12	5.5 -0.1 -0.1 -0.3	903.04 776.00 441.23 402.20	3 3 4 4	CAM CAM CAM CAM	
36	Q53B59	63.7	10	8.05E+07	2	1	5	9853	3FTx	Long neurotoxin OH-37, <i>Ophiophagus hannah</i>	S.FGC(+57.02)AATC(+57.02)PK.V	46.0	1010.43	9	2.0	506.22	2	CAM	P82662
37	R4G2K1	64.3	19	2.03E+07	2	1	2	8768	3FTx	3FTx-Pse-180, <i>Pseudonaja modesta</i>	P.GGMRSVC(+57.02)C(+57.02)STDL.C(+57.02)JNK	45.9	1743.72	15	-3.9	872.86	2	CAM	R4G332; R4G7K8
38	F5CPE7	74.5	16	3.96E+06	3	1	12	9051	3FTx	Putative three finger toxin, <i>Micurus altirostris</i>	T.TC(+57.02)SEGQC(+57.02)YKKSWR.D	31.8	1688.74	13	2.2	563.92	3	CAM	
39	A0A0F7YVW3	109.6	15	5.71E+08	4	4	26	15817	CNP	Natriuretic peptide, <i>Micurus fulvius</i>	R.IDRIGSVSGMGC(+57.02)N.R.V L.GDGC(+57.02)FGQRIDR.I G.DGCG(+57.02)FGQRIDR.I R.IGVSVMGC(+57.02)N.R.V	49.1 39.2 43.7 60.6	1520.72 1279.57 1222.55 1136.51	14 11 10 11	1.4 0.4 1.2 5.3	507.91 427.53 408.52 569.26	3 3 3 2	CAM CAM CAM CAM	
40	P81167	292.8	92	2.78E+06	54	1	614	13312	PLA2	Basic phospholipase A2 nigroxin B, <i>Micurus nigrocinctus</i>	NLIDFKNMICK(+57.02)TNR.H	64.3	1866.94	15	5.1	467.75	4	CAM	
41	C0HK8	254.0	73	2.11E+09	29	3	421	13188	PLA2	Acidic phospholipase A2, <i>Micurus dumerilii</i>	R.TAALC(+57.02)FAKAPYDNNFMNINPR.C K.APYDNNFMNINPRC(+57.02)Q K.APYDNNFMNINPR.C	73.2 67.6 80.1	2542.17 1967.83 1679.74	22 16 14	2.8 3.0 1.7	848.40 984.92 840.88	3 2 2	CAM CAM CAM	
42	A0A0F7YZT8	242.2	62	3.97E+08	37	3	404	16122	PLA2	phospholipase A2, <i>Micurus fulvius</i>	K.WTLYSYC(+57.02)SN(+98)GQLTC(+57.02)KDNN(+98)TKC(+57.02)K.D K.WTLYSYC(+57.02)SN(+98)GQLTC(+57.02)KDNN(+98)TK.C K.WTLYSYC(+57.02)SN(+98)GQLTC(+57.02)K.D	37.0 27.2 21.3	2742.17 2454.05 1880.81	22 20 15	-5.1 -5.2 -4.9	686.55 819.02 941.41	4 3 2	CAM; Deam (NQ) CAM; Deam (NQ) CAM; Deam (NQ)	
43	A0A0F7YZR9	252.0	61	0.00E+00	35	1	368	16128	PLA2	phospholipase A2, <i>Micurus fulvius</i>	K.WTPYSYDC(+57.02)SEGKLT.C(+57.02)KDNN(+98)TK.C	23.4	2567.09	21	-2.7	856.70	3	CAM; Deam (NQ)	
44	U3F572	248.4	48	5.18E+06	35	1	361	16223	PLA2	phospholipase A2, <i>Micurus fulvius</i>	K.WTLYSYDC(+57.02)SKGQ(+98)LT.C(+57.02)KDNN(+98)TK.C	32.8	2583.13	21	3.8	646.79	4	CAM; Deam (NQ)	
45	A0A0F7YZM3	230.6	67	4.52E+08	29	2	343	16066	PLA2	phospholipase A2, <i>Micurus fulvius</i>	R.TAALC(+57.02)FAKAPYDKNYNDPSRC(+57.02)Q R.TAALC(+57.02)FAKAPYDKNYNDPSR.C	35.9 21.7	2816.30 2528.21	24 22	0.2 0.7	705.08 843.75	4 3	CAM CAM	
46	A0A0F7Z3J9	230.7	52	5.29E+07	30	1	339	16067	PLA2	phospholipase A2, <i>Micurus fulvius</i>	K.GNDTKC(+57.02)KDFVC(+57.02)NCI(+57.02)DRTAALC(+57.02)FAKA	25.1	2750.20	23	8.3	551.05	5	CAM	
47	A0A194ARS9	226.4	66	5.11E+07	27	2	295	16124	PLA2	phospholipase A2, <i>Micurus tener</i>	K.WTLYSYDC(+57.02)SNGLTC(+57.02)KDNTKC(+57.02)KDFVC(+57.02)NCI(+57.02)D K.WTLYSYDC(+57.02)SN(+98)GQLTC(+57.02)KDNTK.C	25.6 21.4	3922.61 2569.07	31 21	1.4 10.3	981.66 1285.56	4 2	CAM CAM; Deam (NQ)	
48	A0A2D4N7P6	223.0	66	1.53E+08	23	1	294	11841	PLA2	phospholipase A2 (Frag), <i>Micurus spixii</i>	A.KAPYNN(+98)KN(+98)YN(+98)IDPKR.C	20.9	1836.90	15	7.0	919.46	2	Deam (NQ)	
49	A0A0F7YVW1	187.9	49	1.48E+09	19	3	274	16162	PLA2	phospholipase A2, <i>Micurus fulvius</i>	R.TAALC(+57.02)FAKAPYNN(+98)DKN(+98)YNN(+98)DLSRC(+57.02)Q R.TAALC(+57.02)FAKAPYNN(+98)DKN(+98)YNN(+98)DLSR.C K.APYNN(+98)DKN(+98)YNN(+98)DLSRC(+57.02)Q	31.2 35.7 40.9	2835.28 2547.20 1972.85	24 22 16	-8.0 0.3 -13.4	946.09 850.07 987.42	3 3 2	CAM; Deam (NQ) CAM; Deam (NQ) Deam (NQ); CAM	
50	A0A194ARB9	213.0	51	0.00E+00	20	1	272	16077	PLA2	phospholipase A2, <i>Micurus tener</i>	R.TAALC(+57.02)FAKAPYNNKNYNDPSRC(+57.02)Q	28.6	2815.32	24	-7.6	704.83	4	CAM	
51	U3EPF6	197.0	61	1.23E+09	16	4	229	15844	PLA2	phospholipase A2, <i>Micurus fulvius</i>	R.TAALC(+57.02)FAKAPYDNNFMNMSKR.C R.TAALC(+57.02)FAKAPYDNNFMNMSKR K.APYDNNFMNMSKR.C K.APYDNNFMNMSKR	46.4 23.8 70.9 66.1	2564.16 2408.06 1701.72 1545.62	22 21 14 13	1.0 7.3 2.9 3.0	642.05 1205.05 851.87 516.22	4 2 2 3	CAM CAM CAM CAM	
52	A0A194ARA9	180.9	58	1.28E+08	19	3	186	16003	PLA2	phospholipase A2, <i>Micurus tener</i>	R.C(+57.02)C(+57.02)QVHDDC(+57.02)YGEAEKIDGCI(+57.02)DPK.A R.C(+57.02)C(+57.02)QVHDDC(+57.02)YGEAEKIDGCI(+57.02)D.P D.PKATHYSYDC(+57.02)SEGQLTC(+57.02)K.D	50.1 24.7 35.3	2554.98 2329.83 2143.93	21 19 18	1.1 -1.2 -2.1	852.67 777.62 536.99	3 3 4	CAM CAM CAM	
53	A0A194AT57	180.5	60	8.39E+06	16	1	182	15864	PLA2	phospholipase A2, <i>Micurus tener</i>	K.DNDTKC(+57.02)QDFVC(+57.02)NCI(+57.02)DRTAALC(+57.02)FAKA	39.9	2808.17	23	13.5	937.08	3	CAM	A0A194AS73; A0A194AT61
54	Q6SLM2	97.8	15	3.74E+09	3	1	148	15058	PLA2	Acidic phospholipase A2 1 (Frag), <i>Bungarus caeruleus</i>	M.NMIQC(+57.02)ANTR.T	36.2	1106.50	9	2.6	554.26	2	CAM	
55	A0A194AP70	222.3	59	5.74E+08	26	4	145	15865	PLA2	phospholipase A2, <i>Micurus tener</i>	R.C(+57.02)C(+57.02)KVHDDC(+57.02)YAAAEKYHGC(+57.02)WPK.L	71.9	2654.09	21	-11.3	664.52	4	CAM	

104	A0A194AS8	303.1	49	7.89E+06	38	2	500	58757	LAO	Amine oxidase, <i>Micrurus tener</i>	R.RFDEIVGGFDRLPNSMYQAIANNMVR.L R.TPANTLSVVTADYVIVC(+57.02)STSR.A	72.7 83.8	2898.43 2317.13	25 21	12.3 10.7	725.62 773.39	4 3	CAM	A0A194ARE6
105	A0A8CS5978	138.9	14	5.65E+06	5	1	28	39163	LAO	L-amino-acid oxidase, <i>Laticauda laticaudata</i>	K.SSADIVINDLSLHQLPKK.E	71.8	2090.17	19	10.4	523.56	4		
106	A0A2D4KYM4	63.6	29	1.77E+06	2	1	3	13430	LAO	Amine oxidase domain-containing prot (Frag), <i>Micrurus paraensis</i>	R.EYIKRFRPLSEFVQEN(+98)ENAWYYIK.N	50.5	3235.63	25	5.4	809.92	4	Deam (NQ)	
107	A0A194APD1	193.4	25	5.40E+07	12	12	27	52018	HVA	Hyaluronidase, <i>Micrurus tener</i>	R.APMPYNEPFLVFNWAPPTQC(+57.02)QJLR.Y K.TFHGLGVVIDENWRPQWDNL.N R.SIQFAKELHPDLSHAIKR.L R.KHSDSNALHLFPESFR.I K.HSDSNALHLFPESFR.I K.ELHPDLSHAIKR.L K.ELHPDLSHAIKR.R R.NDQLWLWR.D K.KC(+57.02)SHFLC(+57.02)K.G R.LAKEEFEK.A L.AKEEFEK.A K.LELENLK.Y	47.6 77.7 73.6 50.8 64.0 63.6 44.1 36.3 35.9 37.7 23.2 33.1	2779.33 2411.16 2218.19 2031.00 1902.90 1543.81 1387.71 1242.65 1063.49 992.52 879.43 857.49	23 19 19 17 16 13 12 9 8 8 7 7	10.9 7.1 -0.2 8.9 6.5 2.1 9.5 3.5 0.8 3.2 0.4	927.46 603.80 555.55 508.76 476.74 515.61 463.58 622.34 532.76 497.27 440.73 429.75	4 4 4 4 4 3 3 2 2 2 2 2	CAM	
108	A0A194AT39	224.7	55	1.38E+09	12	9	114	29105	SP	Serine proteinase 1a, <i>Micrurus tener</i>	K.VPHC(+57.02)IVDINILHNPVC(+57.02)QAAVPTMSVK.N E.HIAPLSLSPKPPSMGSDC(+57.02)R.V K.NILC(+57.02)AGLEGEKDC(+57.02)K.G R.FPC(+57.02)ALVLEPQVYK.V L.SLSPKPPSMGSDC(+57.02)R.V K.NILC(+57.02)AGLEGEK.D E.GIAGNSVMC(+57.02)P R.VMGWGTITTPK.A G.DDIMLRL.L	71.0 73.9 70.4 67.3 51.6 53.8 31.8 43.0 24.6	2862.40 2049.01 1805.87 1562.82 1517.70 1315.68 1204.56 1189.62 874.46	25 19 16 14 14 12 12 11 7	11.0 2.8 13.1 11.2 2.6 8.7 6.2 -1.9 0.0	716.61 513.26 903.95 792.42 506.91 658.85 603.29 595.81 438.24	4 4 2 2 3 2 2 2 2	CAM	A0A194AP49
109	A0A61VE32	153.7	22	1.32E+08	4	1	29	30967	SP	Serine protease harobin, <i>Notechis scutatus</i>	E.HIAPLSLPSNPPSM(+15.99)GSVC(+57.02)R.V	43.1	2035.00	19	-13.3	679.33	3	Ox (M); CAM	A0A670ZDU7; A0A670ZG3; A0A670ZGB3
110	A0A646QDU0	53.1	6	1.91E+07	1	1	8	28299	SP	Kallikrein, <i>Thelotornis mossambicus</i>	R.VMGWGTITTPK.V	53.1	1808.87	16	11.8	905.45	2		
111	U3F8T6	77.2	6	7.36E+06	3	3	3	48122	SP	Coagulation factor VII, <i>Micrurus fulvius</i>	R.LGKYHSHRDEGEQER.R K.YHSHRDEGEQER.R R.RVDLHHEK.Y	54.5 28.7 25.1	1952.95 1654.74 1250.70	16 13 10	3.1 -0.6 -1.5	489.25 414.69 417.91	2 4 3		A0A194AG8
112	A0A346C96	96.7	3	1.03E+07	5	5	10	185667	SP	Cobra venom factor (Frag), <i>Spilotes sulphureus</i>	Q.YFTYLLTK.G R.ITPDLPSFR.F K.YIQEGDAC(+57.02)K.A R.QNQPVYK.V R.VGLVAVDK.A	39.4 49.3 30.3 44.1 23.5	1160.65 1157.64 1082.47 976.53 799.48	9 10 9 8 8	3.3 8.8 1.0 3.3 1.0	581.33 579.83 542.24 489.28 400.75	2 2 2 2 2		
113	A0A2D4PRX2	159.6	20	3.04E+08	8	8	32	29231	SP	Peptidase S1 domain-containing prot (Frag), <i>Micrurus surinamensis</i>	R.LPPYADWIKENPEVQLHEVTVPHTH.A R.LPPYADWIKENPEVQLHEVTVPHT.H K.ENVPEVQLHEVTVPHTH.A M.TLQLEVPGLDTC(+57.02)R.S R.LPPYADWIK.E R.LKVPVQYSR.I K.VPVQYSR.I K.LKVPVQY.S	83.1 29.3 39.1 54.6 44.5 33.5 51.6 26.4	3132.59 2924.50 1999.00 1854.99 1151.60 1088.63 847.46 845.50	27 25 18 16 9 9 7 7	7.0 11.3 14.8 9.8 5.3 1.1 1.6 1.6	784.16 732.14 1000.52 619.34 576.81 545.33 424.74 423.76	4 4 2 3 2 2 2 2		A0A2D4PRY7; A0A2D4LPB6
114	A0A8C6Y285	152.1	21	0.00E+00	4	1	12	28110	SP	Peptidase S1 domain-containing prot, <i>Naja naja</i>	R.IIGGFEC(+57.02)NENEHR.S	36.9	1573.69	13	2.4	525.57	3	CAM	A0A8C6Y2J5
115	A0A2D4H4D5	50.2	5	5.37E+05	1	1	1	33871	SP	Peptidase S1 domain-containing prot (Frag), <i>Micrurus corallinus</i>	K.VTNYVNVINQVIQRPP.K	50.2	1940.03	16	8.4	647.69	3		
116	A0A194AS47	313.4	53	2.78E+09	67	12	402	69138	MP	Metalloproteinase type III 2, <i>Micrurus tener</i>	K.TSVAVVQDYGKGTSMVAVTMAHEMGNLGNHDK.G R.KRNDAQLTRIDPFGNTLGLAHGSLC(+57.02)SPK.T K.GTSMVAVTMAHEMGNLGNHKGSC(+57.02)TC(+57.02)GSNK.C K.GTSMVAVTMAHEMGNLGNHDK.G R.RTKPAYQFSSC(+57.02)SVQEHQR.Y R.IDPFGNTLGLAHGSLC(+57.02)SPK.T R.TKPAYQFSSC(+57.02)SVQEHQR.Y K.PAYQFSSC(+57.02)SVQEHQR.Y R.VYEMVNLNKM.YR.H R.VYEMVNLNKM.M R.VYEMVNLNKM.M K.TSVAVVQDYGK.G	30.6 31.7 42.7 73.9 78.2 76.9 75.8 54.0 62.9 62.1 54.5 71.9	3596.72 3422.78 3400.48 2449.13 2208.05 2113.06 2051.95 1822.81 1671.85 1377.74 1221.64 1165.60	34 31 32 23 18 20 17 15 13 11 10 11	10.6 13.3 0.9 0.8 0.5 7.5 2.7 -1.2 7.4 3.5 11.2 2.0	900.20 856.71 851.13 613.29 553.02 705.37 514.00 608.61 558.29 460.26 611.84 583.81	4 4 4 4 4 3 3 3 3 3 2 2		
117	A0A2D4K510	247.8	35	3.18E+08	27	2	133	42159	MP	Peptidase M128 domain-containing prot (Frag), <i>Micrurus paraensis</i>	R.ASLVASVMAHELGNLGR.H R.IDPFGDTIGR.A	86.0 56.0	1974.05 1106.54	19 10	2.1 0.5	494.52 554.28	4 2		
118	A0A2D4K4X3	193.9	59	3.05E+07	17	1	75	16463	MP	Disintegrin domain-containing prot (Frag), <i>Micrurus paraensis</i>	R.SAEC(+57.02)PTDSFQSGHPCC(+57.02)QNNQGYC(+57.02)YNGK.C	42.9	3119.22	27	13.1	780.82	4	CAM	
119	A0A2H6MYK4	155.2	40	3.05E+07	9	1	57	19047	MP	Disintegrin domain-containing prot (Frag), <i>Micrurus lemniscatus carvalhoi</i>	R.AAKDDC(+57.02)DLPEFC(+57.02)TGQSAEC(+57.02)PTDSFOR.N	40.0	3004.23	26	-1.0	1002.42	3	CAM	
120	A0A2D4K530	180.4	57	1.75E+08	10	2	57	15566	MP	Disintegrin domain-containing prot (Frag), <i>Micrurus paraensis</i>	R.AAKDDC(+57.02)DLPEFC(+57.02)TGQSAEC(+57.02)PTSLHQR.N K.DDC(+57.02)DLPEFC(+57.02)TGQSAEC(+57.02)PTSLHQR.N	75.3 22.8	3121.32 2851.15	27 24	2.3 1.5	781.34 951.39	4 3	CAM	
121	R4FIC4	141.6	8	7.13E+06	12	1	53	68297	MP	SVMP-Den-9, <i>Denisonia devisi</i>	R.NGHPCC(+57.02)QNNKGYC(+57.02)YNGKC(+57.02)P.I	23.3	2166.88	18	-14.9	723.29	3	CAM	
122	PDDJ43	127.3	24	1.32E+06	12	1	49	18439	MP	Zinc metalloproteinase-disintegrin-like mikanin (Frag), <i>Micropechis ikaheca</i>	K.LQREHQ(+98)C(+57.02)DSGEC(+57.02)C(+57.02)EK.K	23.8	1935.75	15	-3.9	968.88	2	Deam (NQ); CAM	
123	A0A2D4KIW9	123.8	35	1.56E+07	5	1	25	12910	MP	ADAM cysteine-rich domain-containing protein (Frag), <i>Micrurus paraensis</i>	R.GQDC(+57.02)SFC(+57.02)R.I	46.9	1028.38	8	0.6	515.20	2	CAM	
124	A0A0B4SX85	92.2	4	5.37E+07	3	1	20	68466	MP	Venom metalloprotease, <i>Philodryas chammisani</i>	R.DGHPC(+57.02)QNNQGYC(+57.02)YNGK.C	55.8	1910.74	16	0.3	637.92	3	CAM	A0A098M208
125	Q10749	135.3	10	1.33E+08	7	2	19	68176	MP	SVMP-disintegrin-like mocharhagin, <i>Naja mossambica</i>	R.AAKNDC(+57.02)DFPELC(+57.02)TGR.S K.NDC(+57.02)DFPELC(+57.02)TGR.S	59.7 22.3	1752.76 1482.59	15 12	2.7 3.8	877.39 742.30	2 2	CAM	
126	A0A2D4KKB8	151.4	36	1.61E+08	5	1	19	11174	MP	Peptidase M128 domain-containing prot (Frag), <i>Micrurus paraensis</i>	D.RPQ(+98)C(+57.02)ILNKPLSR.N	50.5	1481.81	12	-0.3	494.95	3	Deam (NQ); CAM	A0A2D4KKA6

127	A0A6P9DR30	121.4	5	1.88E+07	6	4	10	110261	MP	Aminopeptidase, <i>Pantherophis guttatus</i>	R.YISFNSYGK.T R.IYAQPLQIK.T K.ITEIPELRL.T K.SNIEWLWK.Q	39.3 51.5 37.0 39.6	1077.51 1072.63 969.55 888.47	9 9 8 7	3.4 2.4 1.2 -0.1	539.77 537.32 485.78 445.24	2 2 2 2		
128	A0A6P9DVP2	134.2	5	7.79E+04	4	1	10	106081	MP	Aminopeptidase, <i>Pantherophis guttatus</i>	R.ASLINNWFQLVSAGK.L	69.5	1559.87	15	13.6	780.95	2		
129	A0A8C6XE13	100.0	3	6.37E+05	3	1	5	107157	MP	Aminopeptidase, <i>Naja naja</i>	K.VSVYAAPHK.I	41.2	970.52	9	0.6	486.27	2	V8P2K1	
130	A0A8C6XUX5	81.3	4	7.63E+06	3	1	4	73206	MP	X-prolyl aminopeptidase 2, <i>Naja naja</i>	R.QVIGPELQRR	28.5	1038.58	9	2.8	520.30	2	A0A8C6XX20	
131	A0A9F2N2V2	85.4	4	1.10E+06	3	1	4	77598	MP	Xaa-Pro aminopeptidase 2, <i>Python bivittatus</i>	R.RQLEEEYR.W	37.1	1121.55	8	0.4	561.78	2		
132	A0A2D4F8G4	85.4	6	1.81E+06	2	2	2	46082	ACh-ase	Carboxylic ester hydrolase (Frag), <i>Micrurus corallinus</i>	K.KLMEEGQIQKPVIIQK.N K.MMHWYAEFAR.T	64.3 42.1	1909.11 1340.58	17 10	2.7 7.2	478.29 447.87	4 3	A0A6P9CCH2; A0A8C5S1S7	
133	A0A8C6YH4	172.4	36	1.63E+09	10	10	92	22033	GPOX	Glutathione peroxidase, <i>Naja naja</i>	Q.LFQKGDVNGENEQKIYTLK.N K.LFWSPKIHDIKWVFEK.F K.GDVNGENEQKIYTLK.N Q.LFQKGDVNGENEQK.I K.FLVNPGQKPVIMR.W K.IHDIKWVFEK.F K.NSCI(+57.02)PPVIVETFGD.T Q.NI(+98)SEIQGK.H K.FLVNPGQK.P K.LFWSPK.I	59.7 61.9 65.0 58.0 61.8 58.2 41.7 28.2 39.5 50.5	2370.22 2200.18 1853.92 1604.78 1384.76 1328.69 1320.57 1001.54 901.50 889.51	20 17 16 14 12 10 12 9 8 7	4.1 11.3 -1.2 2.1 0.5 2.8 4.4 1.9 -0.1 8.5	791.08 551.06 618.98 803.40 462.60 665.35 651.29 501.78 451.76 445.76	3 4 3 2 3 2 2 2 2 2		V8P395
134	A0A611VT21	105.9	12	1.73E+07	3	3	5	38143	CysP-inh	Antihemorrhagic factor cHLP-B-like, <i>Notechis scutatus</i>	K.EGHSHTLIEHHYQK.N K.LLSDGINKVVASK.C K.AAGEAHIGFC(+57.02)R.A	32.6 65.1 59.9	1780.84 1471.82 1187.55	15 14 11	-0.2 1.7 2.1	446.22 491.62 594.78	4 3 2		CAM