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A new species of pitviper of the genus *Protobothrops* from China (Squamata: Viperidae)

JIAN-HUAN YANG[1], NIKOLAI L. ORLOV[2] & YING-YONG WANG[1, 3, 4]
1 The Museum of Biology, Sun Yat-sen University, 135 Xingangxi Road, Guangzhou 510275, P. R. China.
E-mail: yangjh22@mail.sysu.edu.cn
2 Zoological Institute, Russian Academy of Sciences, Universitetskaya nab. 1, St. Petersburg 199034, Russia.
E-mail: azemiops@zin.ru
3 School of Life Sciences, Sun Yat-sen University, 135 Xingangxi Road, Guangzhou 510275, P. R. China
4 Corresponding author. E-mail: wangyy@mail.sysu.edu.cn

Abstract

A new species of pitviper, *Protobothrops maolanensis* **sp. nov.** is described from the forested karst region in Maolan National Nature Reserve, Guizhou, China based on scalation, body proportions and color pattern. Data on the natural history of the new species are provided and a new key to the currently recognized species of *Protobothrops* is given. The new discovery and other recent findings suggest that further research is desirable in the karst regions in southern China and adjacent Vietnam, Laos and Myanmar.

Key words: Crotalinae, karst region, *Protobothrops maolanensis* **sp. nov.**, snake, taxonomy, *Trimeresurus* sensu lato

Introduction

The Old World pitvipers of the genus *Trimeresurus* sensu lato are the most speciose group of Asian venomous snakes. They are widely distributed from northern peninsular India (Western Ghats) and Sri Lanka eastward to Taiwan, Japan (Ryukyu Archipelago) and the Philippines. They are recorded from the southern provinces of China southward through Indochina and the islands of the Sunda Shelf. The past ten years have witnessed the description of several new species of *Trimeresurus* sensu lato. Currently the group contains more than 50 species (Orlov *et al.*, 2002; Gumprecht *et al.*, 2004; Ziegler *et al.*, 2000; David *et al.*, 2002; Sanders *et al.*, 2004; Grismer *et al.*, 2006, 2008; Orlov *et al.*, 2009).

The taxonomy and phylogeny of *Trimeresurus* sensu lato is fairly well resolved. Several genera are now recognized on the basis of morphological and molecular analyses (Malhotra and Thorpe, 2004). Currently, ten genera are recognized: *Trimeresurus* Lacépède 1804, *Parias* Gray 1849, *Cryptelytrops* Cope 1860, *Peltopelor* Günther 1864, *Viridovipera* Malhotra and Thorpe 2004, *Garthius* Malhotra and Thorpe 2004, *Popeia* Malhotra and Thorpe 2004, *Himalayophis* Malhotra and Thorpe 2004, *Protobothrops* Hoge and Romano-Hoge 1983, and *Ovophis* Burger in Hoge and Romano-Hoge (Malhotra & Thorpe, 2004; Guo *et al.*, 2007; Malhotra *et al.*, 2010). The genus *Protobothrops* contains 13 species and subspecies as follows: *P. elegans* (Gray, 1849), *P. flavoviridis* (Hallowell, 1861), *P. sieversorum* (Ziegler, Herrmann, David, Orlov and Pauwels, 2000), *P. tokarensis* (Nagai, 1928), *P. trungkhanhensis* Orlov, Ryabov and Nguyen 2009, *P. jerdoni jerdoni* (Günther, 1875), *P. j. bourreti* (Klemmer, 1963), *P. j. xanthomelas* (Günther, 1889), *P. cornutus* (Smith, 1930), *P. kaulbacki* (Smith, 1940), *P. mangshanensis* (Zhao, 1990), *P. mucrosquamatus* (Cantor, 1939) and *P. xiangchengensis* (Zhao, Jiang and Huang, 1979). These species inhabit the central and northern regions of Indochina, eastern India, southern China, including Taiwan and Hainan, and the Ryukyu Archipelago of Japan. The latter six species occur in China (Zhao & Adler, 1993; Zhao *et al.*, 1998; David & Ineich, 1999; Gumprecht *et al.*, 2004; Guo *et al.*, 2006, 2007, 2009; Zhao, 2006; David et al, 2008; Orlov *et al.*, 2002, 2009). Monophyly of the genus *Protobothrops* has been corroborated by morphological and molecular studies, resulting in the synonymy of two genera, *Zhaoermia* Gumprecht and Tillack, 2004 and *Triceratolepidophis*

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Ziegler, Herrmann, David, Orlov and Pauwels, 2000. The phylogenetic relationships among most of the species of the genus are well established (Malhotra & Thorpe, 2004; Guo *et al.*, 2006, 2007, 2009).

Recently, we collected specimens of a population of pitviper from Maolan National Nature Reserve, Guizhou, China. The specimens can be assigned to *Protobothrops* on the basis of the following characters: dorsal scales keeled, striated pattern of microdermatoglyphic structure in dorsal scales; palatine bone slightly elongate and triangle-shaped, not forked, without teeth; maxillary bones without projection on border of cavity; anterior edge of ectopterygoid not enlarged and Type 3 spinose hemipenis (Hoge & Romano-Hoge 1983; Malhotra & Thorpe, 2004; Guo & Zhao, 2006). These new snakes differ distinctly from all other species of *Protobothrops* in morphological and color pattern characteristics and are described herein as a new species.

Material and methods

Six individuals of the new species were collected from Maolan National Nature Reserve, Guizhou, China, from 17–22 May 2010 by Sheng Zheng and Jian-Huan Yang and from 9–11 August 2010 by Jian-Huan Yang, Yun Li and Run-Lin Li. Four specimens are designated as the type series, the remaining two specimens are poorly preserved. All the specimens were fixed in 10% formalin, then transferred to 75% ethanol and deposited at The Museum of Biology, Sun Yat-sen University (SYS), Guangzhou, China.

The following measurements were taken with a dial caliper to the nearest 0.01 mm: total length (TL) from the tip of the snout to the tip of tail; snout-vent length (SVL) from the tip of the snout to the anterior margin of the opening of the cloaca; tail length (TaL) from the posterior margin of the opening of the cloaca to the tip of tail; head length (HL) from the tip of the snout to the posterior margin of the mandible; head width (HW) at widest part of the head; the eye horizontal diameter (ED); snout length (SL) from the tip of the snout to the anterior margin of the eye; and distance from eye to nostril (EN) from the anterior margin of the eye to the posterior margin of the nostril.

Scale counts were taken as follows: number of supraocular (SO); number of preocular (PrO); number of postocular (PtO); number of subocular (SubO); number of scales between midsection of supraoculars in a line (SBSO); loreal (L); number of supralabials; number of infralabials; number of temporals; number of ventral scales (V, following Dowling 1951); number of subcaudals (Scd); number of anterior body scale rows (Sq1) at the level of the 15th ventral scale from the head; number of mid-body scale rows (Sq2) halfway between rear of head and opening of cloaca; and number of posterior body scale rows (Sq3) at the level of the 15th ventral scale anterior to the opening of the cloaca. Skull structure and sex were determined by dissection.

Comparative data for others species of *Protobothrops* were obtained from the literature (Maki, 1931; Ziegler *et al.*, 2000; Gumprecht *et al.*, 2004; Zhao, 2006; David *et al.*, 2008; Orlov *et al.*, 2009).

Protobothrops maolanensis sp. nov.

(Figs. 1–3, Table 1)

Holotype. SYS r000211, adult male (Fig. 1) from Maolan National Nature Reserve, Maolan town, Libo County, Guizhou, China (25°12'50.0" N, 107°59'56.04" E, 560 m a.s.l.), collected by Sheng Zheng and Jian-Huan Yang on 18 May 2010.

Paratypes. SYS r000210, adult male, same data as holotype. SYS r000276, adult male and SYS r000277, adult female, from the same locality as holotype, collected by Jian-Huan Yang, Yun Li and Run-Lin Li on 10 August 2010.

Diagnosis. *Protobothrops maolanensis* **sp. nov.** differs from other species of *Protobothrops* by the combination of the following characters: (1) a relatively small body size (TL 609–805 mm); (2) dorsal scales keeled throughout, in 23(rarely 25): 21(19): 17(15) rows; (3) ventral scales 186–193 in male, 190 in female; (4) subcaudals 78–85 pairs in male, 74 in female; (5) 7–8 supralabials; (6) 11–12 infralabials; (7) 10–12 scales between midsection of supraoculars in a line; (8) the 4th supralabials separated from suboculars by one row of scales; (9) 68–72 small and thin body bands across body and tail; (10) TaL/SVL ratio 0.238–0.269 in male, 0.190 in female; (11) SL/HL ratio 0.308–0.326 in male, 0.344 in female; (12) ED/HL ratio 0.164–0.176 in male, 0.152 in female; and (13) the unique color pattern. See below for differential diagnosis by way of comparison with all other species in the genus.

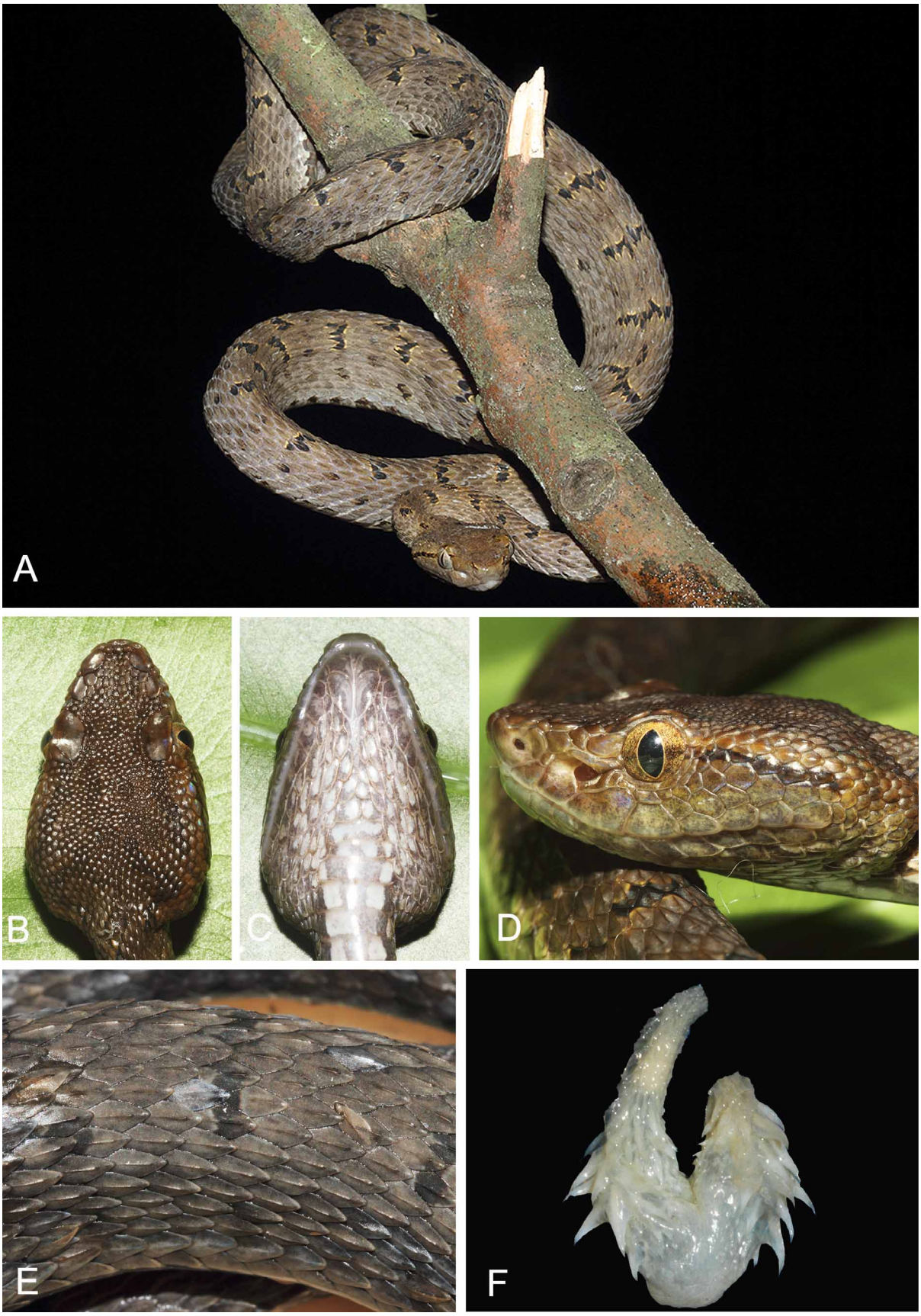

FIGURE 1. A–E (Holotype of *Protobothrops maolanensis* **sp. nov.**, SYSr000211). A: General aspect in life; B: dorsal views of head; C: ventral views of head; D: lateral views of head; E: dorsal view of midbody (showing thin dark brown bands and keeled scales). F (Paratype, SYS r000210): left hemipenis in asulcate view. Photographs by J.H. Yang.

FIGURE 2. A: paratype of *Protobothrops maolanensis* **sp. nov**. (female, SYS r000277); B: holotype of *P. trungkhanhensis* (male); C: paratype of *P. trungkhanhensis* (female); D: karst forest in Maolan National Nature Reserve; E: microhabitats of the type locality. Photographs by Sheng Zheng (A), N. L. Orlov (B, C) and J. H. Yang (D, E).

Description of holotype. Adult male with detailed measurements as listed in Table 1. Body moderately elongate, thin and slightly compressed; head elongate and triangular, distinctly wider than neck, covered with very small, convex and irregular shaped scales, 0.69 times wider than long; upper head scales smooth anteriorly, keeled on occipital, 10 scales between midsection of supraoculars in a transverse line; snout elongate, SL/HL ratio 0.31, nearly twice ED; eye convex, pupil vertical.

TABLE 1. Measurements and scale characters of the type specimens of *Protobothrops maolanensis* **sp. nov.** See text for abbreviations.

	SYS r000211 Holotype	SYS r000210 Paratype	SYS r000276 Paratype	SYS r000277 Paratype
Sex	male	male	male	female
SVL	634	638	492	521
TaL	171	160	117	99
TaL/SVL	0.27	0.25	0.24	0.19
TL	805	798	609	620
HW	16.6	16.6	13.8	14.8
HL	24.0	24.5	19.6	21.7
HW/HL	0.69	0.68	0.70	0.68
SL	7.7	7.5	6.4	7.5
SL/HL	0.32	0.31	0.33	0.35
ED	4.2	4.3	3.2	3.3
ED/HL	0.18	0.18	0.16	0.15
EN	4.8	4.9	4.2	4.8
supralabials	8/8	7/8	8/8	8/8
3rd supralabials contact subocular	no	yes	yes	no
infralabials	11/12	11/12	12/12	11/11
SBSO	10	11	12	10
L	2	2	2	2
canthal scales	2/2	3/3	3/3	4/4
pre-V	2	2	1	2
V	186	188	193	190
Scd	85	78	80	74
Sq1 : Sq2 : Sq3	25 : 21 : 17	23 : 19 : 15	23 : 21 : 17	23 : 21 : 17
outermost row of dorsal scales	strongly keeled	strongly keeled	strongly keeled	smooth anteriorly, weakly keeled posteriorly
body bands	71	72	68	72

Rostral triangular, about 1.3 times broader than high, barely visible from above, broader posteriorly with three apical scales bordered laterally by internasals; internasals not in contact with rostral and separated from each other by four small scales; nasal trapezoid, undivided, with a round nostril opening in middle; 2/2 (L and R, hereafter) canthal scales between supraocular and internasal, distinctly larger than adjacent snout scales, anterior one largest, bordering sharp, raised *canthus rostralis*; two loreals, approximately equal size; supraocular large and elongate, wider than adjacent head scales, pointed anteriorly and 0.58 times as wide as interspace between supraoculars and surrounded by 13/13 small scales; two elongate upper-preoculars above loreal pit in contact with posterior loreal, lower one longer than upper and forming upper margin of loreal pit; one elongate lower-preocular, forming lower margin of loreal pit; two small postoculars; one elongate, thin, crescent-shaped subocular, separated from lower preocular by one scale; temporals numerous, upper ones keeled, lower ones smooth, those bordering supralabials larger than latter; anterior part of nasal large, turning over *canthus rostralis* and visible above; 8/8 supralabials, 2nd

forming anterior margin of loreal pit and separated from nasal by one small scale, 3rd largest, 3rd and 4th separated from subocular by a row of scales on both sides; 11/12 infralabials, 1st pair in contact with each other, first three in contact with anterior chin shield; posterior chin shield small, scarcely differentiated from postgenials.

Dorsal scales narrow, pointed and strongly keeled throughout; in 25 rows on anterior body, 21 rows at mid-body, 17 rows at posterior of body; pre-ventral scales two; ventral scales 186; anal scale undivided; subcaudals divided, 85 pairs.

Coloration in life. Grayish brown above, with 71 dark brown transverse bands with discontinuous pale yellow rough edges, across body and tail. The dark brown bands are relatively small and thin, occupy about six dorsal scale rows in width and 0.5–1.5 dorsal scales in length and often broken into spots. The bands become gradually closer towards tail; tip of tail dark brown with scatted pale spots. Underside gray, with a series of pale brown blotches on both sides. Dorsal head gray brown, with indistinctly symmetrical dark streaks. A postorbital thin dark brown stripe with discontinuous pale yellow edge, starting from posterior margin of eye, extending across upper temporals posteriorly, ending above corner of lip.

Variation. Measurements and scale characters of all the specimens are listed in Table 1. All the paratypes match the holotype in overall morphology and scalation. Dorsal scales in 23 rows on anterior part of body in all three paratypes, 19 rows on mid-body in SYS r000210, 15 rows on posterior part of body in SYS r000210; 3rd supralabials touching the subocular in SYSr000210; 3/3 canthal scales in SYS r000210 and 000276, 4/4 in SYS r000277.

Sexual dimorphism. No significant sexual dimorphism occurs, other than the female having fewer subcaudals than the males (74 vs. 78–85) and the outermost row of dorsal scales are weakly keeled in the female, nearly invisible at anterior body, while the dorsal scales are strongly keeled throughout in male.

Hemipenis. The everted left hemipenis (paratype, SYS r000210) is 18.5 mm long, robust and forked, spinous proximally and calyculate distally; it bears two lobes covered with spines and microspines, extending to the 8th subcaudal. *Sulcus spermaticus* forked at the truncus 6.5 mm from the base of the hemipenis. Strong spines on asulcate side at the base part of each lobe, followed by smaller spines more distally; sulcate side covered with microspines. The hemipenis of *P. maolanensis* closely resembles the Type 3 spinose hemipenis of Malhotra & Thorpe (2004).

FIGURE 3. The type locality of *Protobothrops maolanensis* **sp. nov.**, Maolan National Nature Reserve (circle, "MNNR"), Maolan town, Guizhou, China.

Distribution and natural history. Currently, *P. maolanensis* is known only from the type locality, Maolan National Natural Reserve, Maolan town, Guizhou, China. All individuals were collected at 550–600 m elevation in a subtropical karst evergreen forest. They were found on the ground under scrub at night; no individuals were observed during the day. Other species of amphibians and reptiles at the same habitat in the type locality during the survey included *Polypedates megacephalus* (SYS a000959-962), *Kurixalus odontotarsus* (SYS a000926-931), *Microhyla fissipes* (SYS a000940), *Microhyla heymonsi* (SYS a000938-939), *Goniurosaurus* cf. *luii* (SYS r000217-219), *Takydromus* sp., *Sphenomorphus* cf. *indicus*, *Protobothrops mucrosquamatus* (SYS r000238), *Dinodon flavozonatum* (SYS r000274), *Achalinus spinalis* (SYS r000273) and *Bungarus multicinctus* (SYS r000305).

Etymology. The specific epithet refers to the type locality, Maolan Town, Guizhou, China. We propose the English name "Mao-lan pitviper".

Comparisons. *Protobothrops maolanensis* **sp. nov.** can be readily distinguished from *P. cornutus* and *P. sieversorum* by the absence of horn-shaped projections on the supraocular and a different color pattern.

Protobothrops maolanensis **sp. nov.** differs from *P. elegans* by having fewer mid-body dorsal scales (19–21 vs. 23–25), 4th supralabials separated from suboculars by one row of scales (two in *P. elegans*), two loreals (one in *P. elegans*), dorsal scales keeled throughout (outermost row of dorsal scales smooth in *P. elegans*) and a different color pattern.

Protobothrops maolanensis **sp. nov.** differs from *P. flavoviridis* by having fewer mid-body dorsal scales (19–21 vs. 33–40), two loreals (one in *P. flavoviridis*), lower number of ventral scales (186–193 vs. 222–237 in *P. flavoviridis*), dorsal scales keeled throughout (outermost row of dorsal scales smooth in *P. flavoviridis*) and a different color pattern.

Protobothrops maolanensis **sp. nov.** differs from the *P. jerdonii* complex (*P. j. jerdonii*, *P. j. bourreti*, *P. j. xanthomelas*) by its smaller size, slender body, dorsal scales keeled throughout (outer one or two rows of dorsal scales smooth in *P. jerdonii*) and by a clearly different color pattern.

Protobothrops maolanensis **sp. nov.** differs from *P. kaulbacki* by its smaller size, fewer mid-body dorsal scales (19–21 versus 25), dorsal scales keeled throughout (outermost row of dorsal scales smooth in *P. kaulbacki*), fewer ventral scales (186–193 vs. 201–212 in *P. kaulbacki*), and by a clearly different color pattern.

Protobothrops maolanensis **sp. nov.** differs from *P. mangshanensis* by its smaller size (largest TL 805 mm in *P. maolanensis* sp. nov. vs. largest TL 2375 mm in *P. mangshanensis*), internasals separated by scales (touching each other in *mangshanensis*), two loreals (one in *mangshanensis*), fewer infralabials, (11–12 vs. 13–16), more subcaudals (74–85 vs. 60–67), fewer mid-body dorsal scales (19–21 vs. 25), dorsal scales keeled throughout (outermost row of dorsal scales smooth in *P. mangshanensis*) and a different color pattern.

Protobothrops maolanensis **sp. nov.** differs from *P. mucrosquamatus* by its smaller size (largest TL 805 mm vs. largest TL 1310 mm in *P. mucrosquamatus*), fewer mid-body dorsal scales (19–21 vs. 23–27), fewer ventral scales (186–193 vs. 194–233 in *P. mucrosquamatus*), dorsal scales keeled throughout (outermost row of dorsal scales smooth in *P. mucrosquamatus*) and a different color pattern.

Protobothrops maolanensis **sp. nov.** differs from *P. tokarensis* by having fewer mid-body dorsal scales (19–21 vs. 31–32), two loreals (one in *P. tokarensis*), fewer ventral scales (186–193 vs. 203–208 in *P. tokarensis*), dorsal scales keeled throughout (outermost row of dorsal scales smooth in *P. tokarensis*) and a different color pattern.

Protobothrops maolanensis **sp. nov**. differs from *P. xiangchengensis* by having fewer mid-body dorsal scales (19–21 vs. 25), more subcaudals (74–85 vs. 50–66), dorsal scales keeled throughout (outer two rows of dorsal scales smooth in *P. xiangchengensis*), absence of a dark brown blotch under loreal pit (present in *P. xiangchengensis*).

Protobothrops maolanensis **sp. nov.** is superficially most similar in appearance to *P. trungkhanhensis* from Vietnam, from which it differs by having more dorsal scale rows on the anterior part of the body (23–25 vs. 19), outermost row of dorsal scales on body strongly keeled in males and weakly keeled in females (dorsal scales strongly keeled throughout in both male and female *P. trungkhanhensis*), lower ED/HL ratio (0.15–0.18 vs. 0.23), higher HW/HL ratio (0.68–0.70 vs. 0.65–0.66), and fewer body bands (68–72 vs. 76–84).

Key to the species of *Protobothrops*

This new key was established by building on previous work (after Boulenger, 1896; Stejneger, 1907; Maki, 1931; Pope, 1935; Bourret, 1936; Smith, 1943; Zhao *et al.*, 1998; Ziegler *et al.*, 2000; Gumprecht *et al.*, 2004; Herrmann *et al.*, 2004; Guo *et al.*, 2006, 2007; Zhao, 2006; Orlov *et al.*, 2009):

1	horn-shaped projection on supraocular present	2
-	horn-shaped projection on supraocular absent	3
2	187–193 ventrals and 71–78 pairs of subcaudals	*P. cornutus*
-	228–235 ventrals and 79–82 pairs of subcaudals	*P. sieversorum*
3	outermost row of dorsal scales keeled	4
-	outermost row of dorsal scales smooth	5
4	dorsal scale rows 19-19-17	*P. trungkhanhensis*
-	dorsal scale rows 25(rarely 23)-19(21)-15(17)	*P. maolanensis* **sp. nov.**
5	dorsal scale rows at mid-body ≤ 21	6
-	dorsal scale rows at mid-body ≥ 23	7
6	160–173 ventrals and 44–57 pairs of subcaudals	*P. jerdoni jerdoni*
-	176–188 ventrals and 54–67 pairs of subcaudals	*P. jerdoni xanthomelas*
-	189–192 ventrals and 65–72 pairs of subcaudals	*P. jerdoni bourreti*
7	one loreal	8
-	two loreals	11
8	dorsal scale rows at mid-body ≥	9
-	dorsal scale rows at mid-body ≤ 25	10
9	31–32 dorsal scale rows at mid-body (average 31) and 203–209 ventrals	*P. tokarensis*
-	33–40 dorsal scale rows at mid-body (average 35) and 220–237 ventrals	*P. flavoviridis*
10	6–9 scales between supraoculars, dorsum with green background colour	11
-	11–15 scales between supraoculars, dorsum with yellow or red-brown background colour	*P. elegans*
11	201–212 ventrals and 66–82 pairs of subcaudals	*P. kaulbacki*
-	187–198 ventrals and 60–67 pairs of subcaudals	*P. mangshanensis*
12	10–12 scales between supraoculars, 175–194 ventrals, and 44–66 pairs of subcaudals	*P. xiangchengensis*
-	14–18 scales between supraoculars, 194–233 ventrals, and 70–108 pairs of subcaudals	*P. mucrosquamatus*

Discussion

All individuals of the new species were found only within the karst region. The grayish brown coloration of the new species appears to camouflage these snakes in their habitat. Field and captive observations indicate that the new pitviper is terrestrial and does not climb; we believe that this new small-sized pitviper prefers to use limestone caves as shelter and is perhaps highly specialized for life on karst.

The herpetological diversity of karst regions is high and typified by many endemic species. Such areas contain many types of microhabitats, such as scrub, marsh, farmland, forest, caves and limestone, all of which are inhabited by numerous species of amphibians and reptiles. Herpetological surveys in karst regions of Vietnam have led to surprising recent discoveries, including new species of geckos, skinks and snakes (Ziegler *et al.*, 2007; Ziegler & Nguyen, 2010; Orlov *et al.*, 2008, 2009, 2010). Similar discoveries might be expected elsewhere. Southern China has large areas of karst, in Guangdong, Guangxi, Yunnan, Guizhou and Hainan Island. Recent surveys in this region have yielded a new bird, vipers and geckos (Grismer *et al.*, 1999, 2002; Guo *et al.*, 2006; Zhou & Jiang, 2008; Wang *et al.*, 2010). These Chinese regions are linked to the huge formations of continental and insular northern and central Vietnam, where new records of rare species (e.g., *Boiga kraepelini*, *B. guangxiensis*, *Goniurosaurus lichtenfelderi*, *G.* cf. *luii*, *Shinisaurus crocodylurus*, *P. cornutus*,) and new species (*Goniurosaurus catbaensis*, *G. huuliensis*, *P. sieversorum*, *P. trungkhanhensis*, *Cryptelytrops truongsonensis*, *Boiga bourreti*) have also been documented (Grismer *et al.*, 1999; Orlov *et al.*, 2003, 2004, 2008, 2009; Vu *et al.*, 2006; Tillack *et al.*, 2004; Ziegler & Nguyen, 2010; Ziegler *et al.*, 2000, 2004; 2007, 2008).

Biodiversity explorations, especially herpetological surveys, in the karst regions of southern China have just begun. Further research in the karst regions in southern China and adjacent Vietnam, Laos and Myanmar requires additional attention.

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