



Amolops caelumnoctis Rao & Wilkinson, 2007, a Junior Synonym of *A. splendissimus* Orlov & Ho, 2007 (Amphibia: Anura: Ranidae)

YINPENG ZHANG^{1,2,3#}, JODI J. L. ROWLEY^{4,5,11#}, XIAOLONG LIU^{2,12}, TAO THIEN NGUYEN^{7,13}, HUY QUOC NGUYEN^{7,14}, BENJAMIN TAPLEY⁸, LUAN THANH NGUYEN^{9,10}, YUN YANG⁶, TIMOTHY CUTAJAR^{4,5,15}, YING ZHANG^{2,16} & ZHIYONG YUAN^{1,2*}

¹Key Laboratory of Freshwater Fish Reproduction and Development, Ministry of Education, Southwest University, Chongqing 400715, China

²Key Laboratory of Conserving Wildlife with Small Populations in Yunnan, Southwest Forestry University, Kunming 650224, Yunnan, China

³Michigan State University, East Lansing 48825, MI, United States

✉ zhan1198@msu.edu; <https://orcid.org/0000-0001-6857-7445>

⁴Australian Museum Research Institute, Australian Museum, 1 William St, Sydney, NSW, 2010, Australia

⁵Centre for Ecosystem Science, School of Biological, Earth and Environmental Sciences, University of New South Wales, Sydney NSW 2052, Australia

⁶Western National Nature Reserve of Yunnan Province, Wenshan 663000, Yunnan, China

✉ 1923677199@qq.com; <https://orcid.org/0000-0002-5060-7025>

⁷Department of Nature Conservation Curator of Herpetology, Vietnam National Museum of Nature, Academy of Science and Technology, Hanoi, Vietnam

⁸Zoological Society of London, Regent's Park, London, UK.

✉ ben.tapley@zsl.org; <https://orcid.org/0000-0002-9787-3793>

⁹EDGE of Existence Programme, Zoological Society of London, Regent's Park, London, UK

✉ nguyenluanbio@gmail.com

¹⁰Asian Turtle Program of Indo-Myanmar Conservation 1806, CT1-C14 Bac Ha Building, To Huu Street, Nam Tu Liem District, Hanoi, Vietnam

¹¹✉ jodi.rowley@australian.museum; <https://orcid.org/0000-0002-2011-9143>

¹²✉ 13888981969@163.com; <https://orcid.org/0000-0002-8417-5208>

¹³✉ taont@vnmn.vast.vn; <https://orcid.org/0000-0002-5640-4536>

¹⁴✉ huynguyen17295@gmail.com

¹⁵✉ timothy.cutajar@australian.museum; <https://orcid.org/0000-0001-7841-9205>

¹⁶✉ 2848289610@qq.com; <https://orcid.org/0000-0002-6143-8837>

[#]These authors contributed equally

*Corresponding author. ✉ yuanzhiyongkiz@126.com; <https://orcid.org/0000-0001-5991-3021>

Abstract

Amolops splendissimus Orlov and Ho, 2007 and *A. caelumnoctis* Rao and Wilkinson, 2007 were described almost simultaneously from either side of the China-Vietnam border. The two species share a strong morphological resemblance, and their taxonomic distinctiveness has been questioned, yet no one has confirmed the taxonomic relationship and status between the two taxa. To resolve this taxonomic issue, we collected additional topotypic and near-topotypic specimens of *A. splendissimus* and *A. caelumnoctis* from both China (*A. caelumnoctis*: Wenshan County, Yunnan Province; type locality Luchun County, Yunnan Province), and Vietnam (*A. splendissimus*: Tam Duong District, Lai Chau Province; type locality Mount Ky Quan San, Bat Xat, Lao Cai Province). Molecular analysis based on a 16S rRNA fragment revealed minimal genetic divergences between the two taxa (0.0%–0.4% uncorrected *p*-distance), and both species are closely related to *A. viridimaculatus* (2.1%–2.3%) and *A. medogensis* (3.5%–3.7%). Morphological comparisons between the newly collected specimens and the original descriptions of both species further support the lack of distinctiveness of the two species, hence, we conclude that *A. caelumnoctis* is a junior synonym of *A. splendissimus*.

Key words: Amphibian, Ranidae, species identification, taxonomy, China-Vietnam border

Introduction

The China-Vietnam border region straddles an area of high biodiversity, and regions on both sides of the national border share a similar species assemblage. Recent attention from taxonomists of both countries have led to increasing collaborative survey and research efforts, which have resulted in an increasing rate of new species discovery and new species distribution records on both sides of the national border (e.g. Luong *et al.*, 2019; Pham *et al.*, 2019; Yuan *et al.*, 2020; Wang *et al.*, 2020a).

Being one of the most diverse genera of the family Ranidae Batsch, 1796, the genus *Amolops* Cope, 1865, exhibits a high degree of adaptation to its microhabitat of swift-flowing mountainous streams. Adults typically have enlarged digital pads and hind feet with full webbing, while tadpoles are gastromyzophorous with abdominal suckers (Fei *et al.*, 2005). Currently 74 species are recognized in this speciose genus, distributed from Nepal, northern India, southern China, and mainland Indochina (Frost, 2021; Wang *et al.*, 2020b; Wu *et al.*, 2020), with eight species groups and two major geographic clades in China recently identified (Wu *et al.*, 2020). Among the *Amolops* species distributed in China, six species have been reported from the China-Vietnam border region (AmphibiaChina, 2021; Pham *et al.*, 2020), including *Amolops caelumnoctis* Rao and Wilkinson, 2007, *A. chungansis* Pope, *A. ricketti* Boulenger, 1899, *A. splendissimus* Orlov and Ho, 2007, *A. viridimaculatus* Jiang, 1983, and *A. wenshanensis* Yuan, Jin, Li, Stuart and Wu, 2018. Of these species, the taxonomic status of *A. caelumnoctis*, described from China (Qimaba Township, Luchun County, Honghe Prefecture, Yunnan Province), is particularly controversial (Biju *et al.*, 2010), with the morphologically similar *A. splendissimus*, described 150 km southwest in adjacent Vietnam (Ho Thau Village, Ho Thau Commune, Tam Duong District, Lai Chau Province). Both *A. caelumnoctis* and *A. splendissimus* share a strong morphological resemblance. However, neither *A. caelumnoctis* or *A. splendissimus* descriptions included molecular data (Orlov and Ho, 2007; Rao and Wilkinson, 2007). As a result, the validity of *A. splendissimus* has been questioned and it has been speculated that it is a possible junior synonym of *A. caelumnoctis* (Biju *et al.*, 2010). However, this assumption is based upon publication dates of November 10th 2007 for *A. caelumnoctis* and December 15th 2007 for *A. splendissimus* (Biju *et al.*, 2010), and the true publication date for *A. caelumnoctis* is December 28th 2007), giving priority to the name *A. splendissimus*.

We collected additional specimens of *A. caelumnoctis* and *A. splendissimus* from or near their type localities in the mountainous streams of Wenshan County and Luchun County (the type locality of *A. caelumnoctis*) in Yunnan Province, China, and Bat Xat District in Lao Cai Province and Tam Duong District (the type locality of *A. splendissimus*) in Lai Chau Province, Vietnam (Figure 1). We performed the first morphological and genetic analyses of these species using newly-collected specimens to determine the distinctiveness and validity of these two species.

Methods

Sampling

Three specimens (one female: AMS R188526; two juveniles: VNMN 010923 and HLNP2017100900016) of *A. splendissimus* were collected from Mount Ky Quan San, Bat Xat, Lao Cai Province, Vietnam (22.508°N, 103.615°E, 2118 m asl) by Chung Thanh Nguyen, Luan Thanh Nguyen, Luke Harding, Timothy Cutajar, Jodi J. L. Rowley, and Benjamin Tapley between 8–10 September 2017. Six specimens (four males: VNMN 10251, VNMN 10252, VNMN 10253, VNMN 10254; two females: VNMN 10250, VNMN 10255) of *A. splendissimus* were collected from Ho Thau Village, Ho Thau Commune, Tam Duong District, Lai Chau Province Vietnam by Tao T.N between 15–18 May 2019 (22.27255°N, 103.33216°E to 23.0346°N, 104.5009°E; altitude: 2060–2580 m asl). Two specimens of *A. caelumnoctis* were collected from Wenshan county, Yunnan Province, China (23.341832°N, 103.973461°E, 2027 m asl) (males: SWFU 003995, SWFU 004249) by Ying Zhang and Zhiyong Yuan on 29 April 2020, and three specimens of *A. caelumnoctis* from Luchun County, Honghe Prefecture, Yunnan Province, China (22.98372°N, 102.279485°E, 1840 m asl) (males: SWFU 004525, SWFU 004524, SWFU 004612) were collected by Xiaolong Liu, between 28–29 April 2020 (Figure 1). Specimens were preserved and deposited at the Vietnam National Museum of Nature (VNMN): VNMN 010923, VNMN 10250, VNMN 10251, VNMN 10252, VNMN 10253, VNMN 10254, VNMN 10255, Vietnam, Australian Museum (AMS): AMS R188526, Australia, and Southwest Forestry University (SWFU): SWFU 004612, SWFU 004249, SWFU 004524, SWFU 004525, SWFU 003995, China. A referred specimen was deposited at the Hoang Lien National Park headquarters (HLNP): HLNP2017100900016, in Vietnam as a reference for National Park scientists. (Table 1)

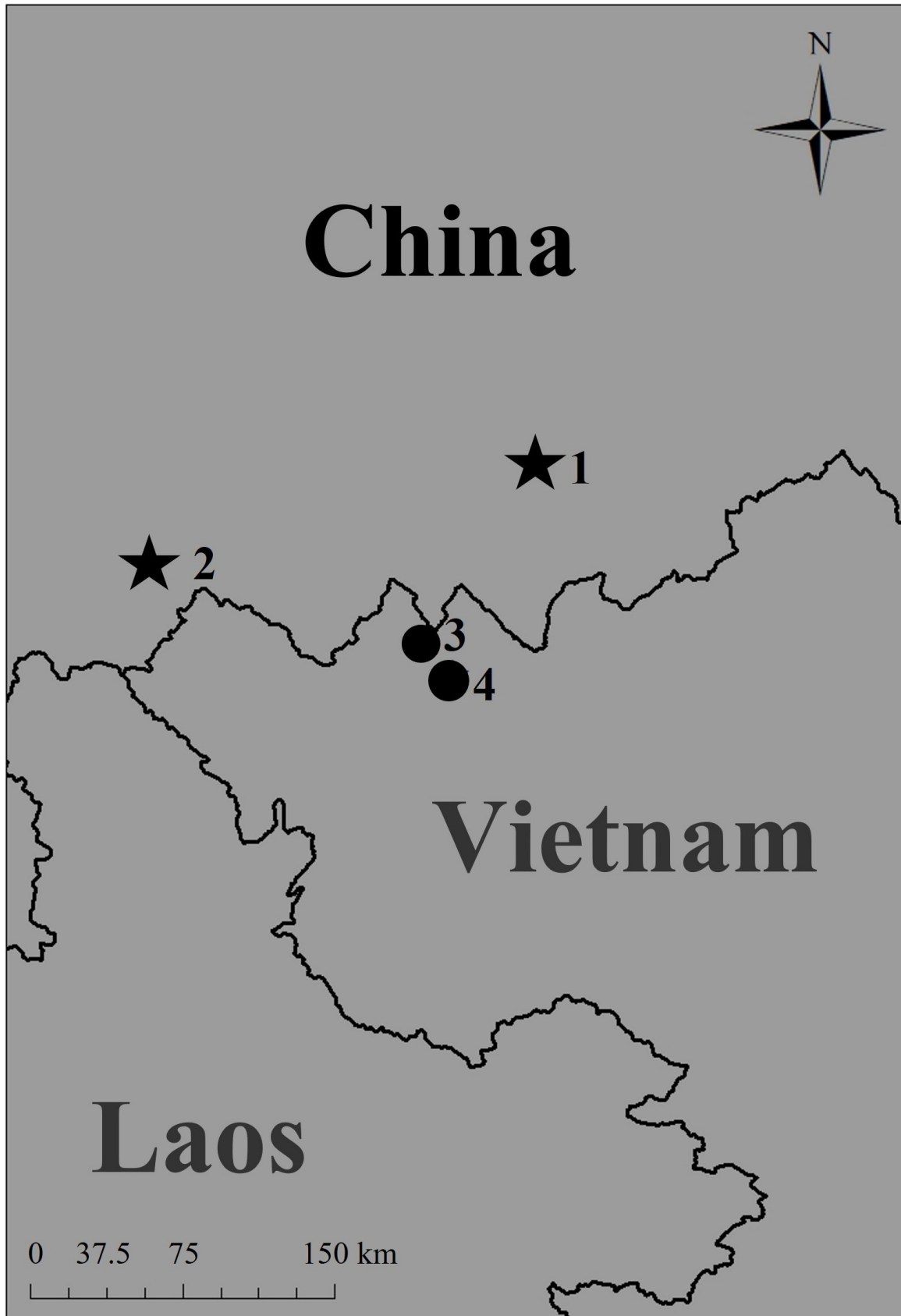


FIGURE 1. The known localities of *A. splendissimus* along the China-Vietnam bordering region. Localities marked by stars represent sites in China and solid circles represent sites in Vietnam. (1: Wenshan County, Yunnan, China; 2: Luchun County, Yunnan, China, *A. caelumnoctis* type locality; 3: Lai Chau Province, Vietnam, *A. splendissimus* type locality); 4: Bat Xat District, Lao Cai Province, Vietnam

TABLE 1. Detailed morphometric measurements of *Amolops splendidissimus* and *A. caelumnoctis* populations distributed through China-Vietnam bordering. The holotypes of *A. splendidissimus* and *A. caelumnoctis* description data were displayed here for partial morphological characteristics comparison. (M: male; JF: juvenile female; J: juvenile male; /: data not applicable or available).

| Species | <i>A. caelumnoctis</i> | | <i>A. caelumnoctis</i> | | <i>A. caelumnoctis</i> | | <i>A. caelumnoctis</i> | | <i>A. splendidissimus</i> | | <i>A. splendidissimus</i> | |
|---------------|------------------------|-------------|------------------------|-------------|------------------------|-------------|------------------------|-------------------|---------------------------|---|---------------------------|---|
| Accession no. | SWFU 004612 | SWFU 004249 | SWFU 004524 | SWFU 004525 | SWFU 003995 | AMS R188526 | VNMN 010923 | HLNP2017100900016 | | | | |
| Sex | M | M | M | M | M | JF | J | J | J | J | J | J |
| SVL(mm) | 77.8 | 83.1 | 80.3 | 81.7 | 80.4 | 50.1 | 39.1 | 36.4 | | | | |
| HW | 26.0 | 26.8 | 26.7 | 26.3 | 25.7 | 16.8 | 13.2 | 12.5 | | | | |
| HL | 25.9 | 27.4 | 26.8 | 27.1 | 25.5 | 18.6 | 13.6 | 12.8 | | | | |
| MN | 21.7 | 22.0 | 21.4 | 22.4 | 21.0 | 14.0 | 10.5 | 10.6 | | | | |
| MFE | 17.6 | 18.0 | 18.9 | 18.8 | 16.9 | 11.6 | 8.1 | 8.4 | | | | |
| MBE | 10.3 | 10.4 | 11.1 | 10.8 | 10.7 | 6.4 | 3.4 | 3.7 | | | | |
| SNL | 9.8 | 11.1 | 11.1 | 11.1 | 10.4 | 7.4 | 5.9 | 5.6 | | | | |
| ED | 9.1 | 8.6 | 9.4 | 9.0 | 8.2 | 5.8 | 4.5 | 4.4 | | | | |
| UEW | 6.0 | 6.8 | 7.1 | 7.1 | 6.8 | 5.3 | 3.2 | 2.7 | | | | |
| IN | 9.1 | 9.4 | 9.1 | 9.7 | 8.3 | 5.9 | 4.7 | 4.6 | | | | |
| NS | 5.7 | 5.9 | 5.8 | 6.0 | 5.7 | 3.8 | 3.0 | 3.2 | | | | |
| EN | 5.0 | 4.6 | 5.0 | 4.9 | 4.3 | 3.0 | 2.6 | 2.7 | | | | |
| TYD | 2.8 | 2.5 | 2.8 | 2.9 | 2.9 | 2.3 | 1.7 | 1.4 | | | | |
| TYE | 3.8 | 4.2 | 4.7 | 4.11 | 4.6 | 2.4 | 1.4 | 1.5 | | | | |
| FLL | 16.6 | 17.6 | 16.6 | 16.2 | 17.0 | 8.5 | 7.5 | 7.5 | | | | |
| HAL | 43.5 | 45.6 | 43.2 | 41.2 | 41.1 | 28.0 | 22.3 | 21.7 | | | | |
| FHL | 60.1 | 63.2 | 59.8 | 57.4 | 58.1 | 26.9 | 30.0 | 28.1 | | | | |
| NPL | 4.7 | 5.7 | 4.8 | 5.6 | 5.7 | / | / | / | | | | |

.....continued on the next page

TABLE 1. (Continued)

| Species | <i>A. splendissimus</i> | | <i>A. splendissimus</i> | | <i>A. splendissimus</i> | | <i>A. splendissimus</i> | | <i>A. splendissimus</i> | | <i>A. splendissimus</i> | | <i>A. caelumnoctis</i> | |
|---------------|-------------------------|------------|-------------------------|------------|-------------------------|------------|-------------------------|---------------|-------------------------|--|-------------------------|--|------------------------|--|
| Accession no. | VNMN 10251 | VNMN 10252 | VNMN 10253 | VNMN 10254 | VNMN 10250 | VNMN 10255 | ZISP 7598 | KIZ-2003 A018 | (holotype) | | (holotype) | | (holotype) | |
| Sex | M | M | M | M | F | F | M | M | | | | | | |
| SVL(mm) | 62.1 | 72.7 | 68.4 | 73.4 | 89.8 | 88.3 | 76.6 | 71.3 | | | | | | |
| HW | 21.4 | 24.1 | 23.3 | 24.6 | 29.6 | 30.2 | 25.3 | 24.8 | | | | | | |
| HL | 21.7 | 24.8 | 22.4 | 25.9 | 30.2 | 28.0 | 26.0 | 25.3 | | | | | | |
| MN | 18.0 | 20.8 | 18.7 | 20.7 | 23.9 | 23.4 | / | / | | | | | | |
| MFE | 14.4 | 15.9 | 15.1 | 17.0 | 19.9 | 18.0 | / | / | | | | | | |
| MBE | 8.6 | 10.0 | 8.7 | 9.8 | 11.1 | 11.2 | / | / | | | | | | |
| SNL | 8.2 | 10.0 | 9.5 | 10.2 | 11.8 | 11.1 | 11.4 | 10.5 | | | | | | |
| ED | 6.2 | 6.5 | 7.0 | 7.9 | 8.8 | 8.5 | 8.8 | 8.8 | | | | | | |
| UEW | 5.5 | 5.6 | 5.9 | 5.9 | 7.8 | 7.5 | 7.2 | / | | | | | | |
| IN | 7.6 | 8.4 | 7.9 | 8.7 | 9.5 | 9.4 | 8.7 | / | | | | | | |
| NS | 4.7 | 5.5 | 5.5 | 5.9 | 6.8 | 6.3 | / | / | | | | | | |
| EN | 3.6 | 4.7 | 4.4 | 4.6 | 4.8 | 6.0 | 4.7 | / | | | | | | |
| TYD | 2.5 | 2.2 | 2.1 | 2.3 | 2.7 | 2.9 | 2.5 | 3.1 | | | | | | |
| TYE | 3.6 | 4.2 | 4.1 | 4.2 | 5.5 | 4.8 | 3.8 | / | | | | | | |
| FLL | 15.0 | 12.7 | 13.9 | 13.1 | 19.5 | 21.6 | / | 38.2 | | | | | | |
| HAL | 37.3 | 41.1 | 39.6 | 41.1 | 51.2 | 51.1 | / | 27.0 | | | | | | |
| FHL | / | / | 53.5 | 54.3 | 70.7 | 72.7 | 59.3 | 65.2 | | | | | | |
| NPL | 4.4 | 5.9 | 6.0 | 6.5 | 5.9 | 5.5 | 7.9 | / | | | | | | |

Morphometric analyses

Measurements were taken by Jodi Rowley, Xiaolong Liu and Tao Thien Nguyen from fourteen recently-collected specimens: three *A. splendissimus* (one female and two juveniles) from Lao Cai Province, Vietnam, six *A. splendissimus* (four males and two females) from Lai Chau Province, Vietnam, and five *A. caelumnoctis* (all males) from Yunnan province, China. Morphological characteristics and definitions follow Orlov and Ho (2007), with additional measurements of the head after Ninh *et al.* (2020). The following morphological data from preserved specimens were recorded to the nearest 0.1 mm using digital calipers (Table 1): snout-vent length (SVL); snout length, from anterior corner of eyes to tip of snout (SNL); maximum head width across the angle of jaws (HW); head length, from back of mandible to tip of jaws (HL); mandible jaw angle to nostril (MN); distance from back of mandible to front of the eye (MFE); distance from back of mandible to back of the eyes (MBE); horizontal diameter of the eye (ED); maximum width of upper eyelids (UEW); internarial space (IN); distance from nostril to snout tip (NS); distance from anterior corner of eye to nostril (EN); tympanum diameter (TYD); distance from posterior eye to anterior tympanum (TYE); femur length, from axilla to elbow (FLL); hand length, from elbow to tip of the third finger (HAL); forelimb hand length (FHL); and diameter of the nuptial pad of the first finger (NPL). We also considered the following relative ratios for comparison: SNL/SVL, HW/SVL, HL/SVL, HL/HW, SNL/HW, ED/SNL, ED/HL, EN/ED, TYE/TYD, TYD/ED (Table 3). The One Sample T-Test was conducted in R (RStudio Team, 2015) to examine possible differentiation between our collected specimens: *A. splendissimus* from Vietnam and *A. caelumnoctis* from China. Raw data were square root transformed for normality, and only males were included in the dataset as females were not collected in China.

Molecular analyses

A 699 bp fragment of 16S ribosomal RNA (16S rRNA) was sequenced from three *A. splendissimus* specimens from Bat Xat, Vietnam and three *A. caelumnoctis* specimens from Yunnan, China (Wenshan: $n = 1$, and Luchun: $n = 2$). To assess genetic similarity between *A. splendissimus* and *A. caelumnoctis* and determine preliminary phylogenetic relationships, we constructed a phylogenetic tree with the newly obtained sequences and comparable sequences for 28 *Amolops* species from Wu *et al.* (2020), downloaded from GenBank, with *A. panhai* Matsui and Nabhitabhata, 2006 and *A. lifanensis* (Liu, 1945) as outgroups (Table 2). Sequences from newly collected specimens were amplified using polymerase chain reaction (PCR) protocols and primers adapted from Wu *et al.* (2020). Amplification of crude nucleic acids extraction started with an initial denaturation for 4 min at 95 °C, followed by 35 cycles of 94 °C denaturation for 40 s, annealing at 50–55 °C for 1 min, extension at 72 °C for 1 min, ending with additional final extension at 72 °C for 10 min. Amplified products were sequenced for 16S rRNA using an ABI 3730 automated sequencer.

All sequences were aligned in ClustalX (Larkin *et al.*, 2007) and aligned sequences were compared using MEGA-X (Kumar *et al.*, 2018). The uncorrected genetic distances (p -distances) between *A. splendissimus* and *A. caelumnoctis*, along with genetic distance in selected *Amolops* species were calculated in MEGA-X using pairwise deletion of missing gaps. Phylogenetic trees were estimated with Maximum Likelihood (ML) using RaxML NG v0.9.0. (Kozlov *et al.*, 2019) and Bayesian inference (BI) using MrBayes 3.2.7a. We used JModelTest 2 (Guindon and Gascuel 2003; Darriba *et al.*, 2012) to infer the model of sequence evolution that best fit our 16S sequences. The model GTR+I+G was selected as the best fitted model for BI testing, and GTR was selected as the best fitted model for ML. For BI analyses, metropolis coupled Markov Chain Monte Carlo (MCMC) analyses were performed with 1 cold chain and 3 heated chains for 3,000,000 generations and sampled every 1,000 generations. Four independent MCMC runs were performed and the first 1,500 trees were discarded as burn-in. The potential scale reduction factor (PSRF) was > 1 and the average standard deviation of split frequencies (ASDSF) was < 0.01 . Bayesian statistics were used to evaluate tree and parameter convergence, respectively. Confidence of tree topology was assessed via posterior probability (PPr) (Huelsenbeck and Ronquist, 2001). ML analyses were conducted with RAXML NG v0.9.0. (Kozlov *et al.*, 2019). Branch support values (bootstrap supports [BS]) were determined based on 1,000 bootstrap replicates. We consider relationships with PPr ≥ 0.95 and BS ≥ 75 to be strongly supported (Alfaro *et al.*, 2003; Hillis and Bull, 1993).

TABLE 2. 16S mitochondrial fragments of *Amolops*, including outgroups used in our study. All *Amolops caelumnoctis* and *A. splendissimus* sequences were sequenced as part of this study. All other sequences were downloaded from GenBank.

| Species | Voucher | Locality | GenBank Acc. No. |
|---------------------------------|---------------------|---|------------------|
| <i>Amolops akhaorum</i> | FMNH 271355 | Vieng Phou Kha, Luang Namtha, Laos | FJ417158 |
| <i>A. aniqiaoensis</i> | KIZ011136 | Tibet, China | MN953658 |
| <i>A. archotaphus</i> | FMNH 271708 | Amphoe Chom Thong, Chiang Mai, Thailand | MN953659 |
| <i>A. bellulus</i> | KIZXPX9037 | Tengchong, Yunnan, China | MN953664 |
| <i>A. bellulus</i> | CAS 233986 | Tengchong, Yunnan, China | FJ417126 |
| <i>A. caelumnoctis</i> | SWFU 004524 | Luchun County, Yunnan, China | MZ484730 |
| <i>A. caelumnoctis</i> | SWFU 004525 | Luchun County, Yunnan, China | MZ484729 |
| <i>A. caelumnoctis</i> | SWFU 003995 | Wenshan County, Yunnan, China | MZ484728 |
| <i>A. chayuenensis</i> | KIZ014016 | Baxoi, Tibet, China | MN953666 |
| <i>A. chunganensis</i> | Tissue ID: YPX4947 | Bijie, Guizhou, China | MN953767 |
| <i>A. chunganensis</i> | KIZYPX18652 | Chengkou, Chongqing, China | MN953670 |
| <i>A. chunganensis</i> | KIZ03756 | Fangxian, Hubei, China | MN953671 |
| <i>A. compotrix</i> | FMNH 256500 | Nakai, Khammouan, Laos | FJ417141 |
| <i>A. cucae</i> | AMNH 168729 | Van Ban, Lao Cai, Vietnam | FJ417145 |
| <i>A. daiyunensis</i> | KIZ09023 | Daiyunshan, Fujian, China | MN953677 |
| <i>A. daorum</i> | KIZ047048 | Jingdong, Yunnan, China | MN953678 |
| <i>A. granulosis</i> | SCUM045823HX | Dayi, Sichuan, China | MN953680 |
| <i>A. jinjiangensis</i> | SCUM050435CHX | Deqing, Yunnan, China | EF453741 |
| <i>A. lifanensis</i> (outgroup) | SCUM045801HX | Maoxian, Sichuan, China | MN953702 |
| <i>A. loloensis</i> | SCUM045806HX | Xichang, Sichuan, China | MN953704 |
| <i>A. mantzorum</i> | SCUM045817HX | Wolong, Sichuan, China | MN953706 |
| <i>A. medogensis</i> | SYNU04II6216 | Medog, Tibet, China | MN953710 |
| <i>A. mengdingensis</i> | KIZ20160265 | Mengding, Yunnan, China | MK501808 |
| <i>A. nyingchiensis</i> | KIZ012633 | Mainling, Tibet, China | MN953719 |
| <i>A. panhai</i> (outgroup) | CAS 229600 | Pakchan Reserve Forest, Kawthoung, Tanintharyi, Myanmar | MN953722 |
| <i>A. shuichengicus</i> | SYS a004956 | Shuicheng County, Guizhou, China | MK604845 |
| <i>A. sinensis</i> | Tissue ID: YPX54730 | Mao'er Shan, Guangxi, China | MN953748 |
| <i>A. splendissimus</i> | VNMN 010923 | Bat Xat, Lao Cai, Vietnam | MZ484725 |
| <i>A. splendissimus</i> | AMS R188526 | Bat Xat, Lao Cai, Vietnam | MZ484726 |
| <i>A. splendissimus</i> | HLNP2017100900016 | Bat Xat, Lao Cai, Vietnam | MZ484727 |
| <i>A. tuberdepressus</i> | SCUM050433CHX | Jingdong, Yunnan, China | MN953729 |
| <i>A. viridimaculatus</i> | SCUM050403CHX | Pianma, Yunnan, China | MN953737 |
| <i>A. wenshanensis</i> | KU 292045 | Jinxiu, Guangxi, China | MN953725 |
| <i>A. wuyiensis</i> | KIZ04342 | Yiwu, Zhejiang, China | MN953761 |
| <i>A. yunkaiensis</i> | SYS a004681 | Yunkaishan Nature Reserve, Guangdong, China | MK263271 |

Results

Our final alignment of the 16S rRNA sequences including GenBank samples (Table 1) consisted of 699 bp including gaps and missing data. Among all sequences, there were 418 variable positions and 301 parsimony informative sites. Maximum Likelihood and Bayesian inference analysis recovered similar topologies and similar strongly supported divergences, there were some branches with low support values (BS < 75, PPr < 0.95) but the phylogenetic

relationship between *A. caelumnoctis* and *A. splendissimus* was well supported. Samples of *A. caelumnoctis* and *A. splendissimus* formed a well-supported correlation (BS = 100, PPr = 0.99) which was recovered as sister to *A. viridimaculatus* (Figure 2). There was some structuring within this well supported clade. *Amolops caelumnoctis* from Wenshan clustered with *A. splendissimus* from Bat Xat, Lao Cai closer compared to *A. caelumnoctis* from Luchun, Vietnam. Furthermore, the very low uncorrected *p*-distance (0.0%–0.4%) between *A. caelumnoctis* and *A. splendissimus* indicate that *A. caelumnoctis* and *A. splendissimus* represent a single operational taxonomic unit. Uncorrected *p*-distances were variable between sites (Wenshan vs. Luchun samples (~178.04 km): 0.2%–0.4%; Wenshan vs. Bat Xat samples (~98.25 km): 0.0%; Luchun vs. Bat Xat samples (~146.96 km): 0.2%–0.4%). The uncorrected *p*-distance between *A. splendissimus* and other *Amolops* species ranged from 2.3%–16.6% (*A. splendissimus* vs. *A. viridimaculatus*: 2.3%; *A. splendissimus* vs. *A. torentis*: 16.6%, Figure 2 and Table S1).

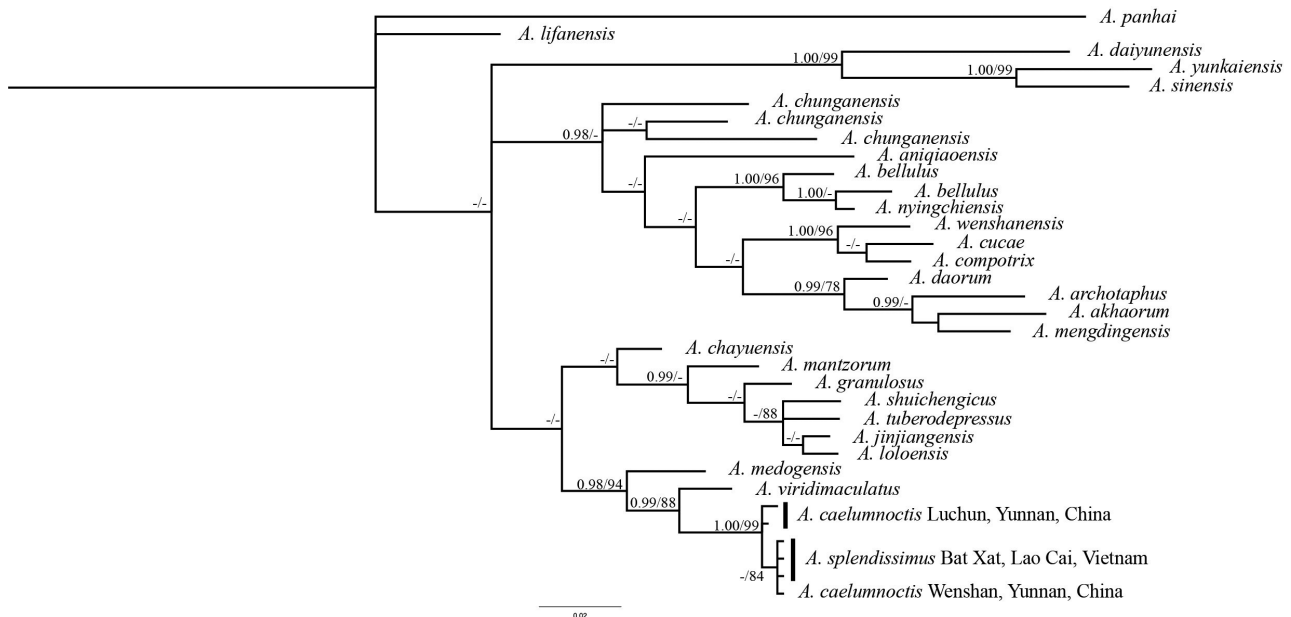


FIGURE 2. Bayesian inference and maximum-likelihood phylogenies based on 16S (mtDNA) gene, to display a clear monophyly of *Amolops caelumnoctis* and *A. splendissimus*. Numbers in forms above branches indicate bootstrap support values on right (BS \geq 75) and Bayesian posterior probabilities on left (PPr \geq 0.95), respectively; low support values (BS < 75, PPr < 0.95) are denoted by “-”. Specimens from China and Vietnam were labeled separately.

The original morphological descriptions of *A. caelumnoctis* and *A. splendissimus* are similar, particularly with respect to having a large body size, a large flattened and very broad head, near-equal head width and length, large eyes and a relatively short snout. (Table 3). In addition, our newly collected specimens from China (C) and Vietnam (V) share these characteristics in males having relatively short snouts (SNL/SVL: 12.6%–15.4%); broad and wide head (HW/SVL: 32.0%–34.8%; HL/SVL: 31.7%–37.1%); nearly equal ratio between head length and width (HL/HW: 95.8%–105.1% (V) vs. 99.2%–103.0% (C), One Sample *t*-test $p = 0.372$); relatively large eyes (ED/SNL: 65.0%–77.2% (V) vs. 77.5%–92.9% (C), One Sample *t*-test $p = 0.959$; EN/ED: 58.4%–72.4% (V) vs. 52.4%–54.9% (C), One Sample *t*-test $p = 0.026$); and a small sized tympanum (TYD/ED: 29.0%–40.3% (V) vs. 29.1%–39.4% (C), One Sample *t*-test $p = 0.103$). In addition to morphometric measurements, other morphological characters are similar between the type series of *A. caelumnoctis* and *A. splendissimus* and all newly collected specimens in: pectoral spines and gular pouches absent; invisible pineal body; two small oblique vomerine teeth; deeply notched tongue posteriorly; dorsolateral fold absent; temporal fold weakly developed; a vertical loreal region; distinct circummarginal grooves on fingers; lateral fringes on sides of fingers absent; fully webbed feet and outer metatarsal tubercle absent (Tables 3 and 4). However due to the lack of female specimens from China, comparisons with the newly collected specimens from Vietnam are somewhat limited due to the distinct sexual dimorphism known to be exhibited by congeneric species (e.g. Dever *et al.*, 2012; Lyu *et al.*, 2019a, b; Wang *et al.*, 2020a).

TABLE 3. Comparison of morphometric measurements for *A. splendidissimus* and *A. caelumnoctis* using data reported for holotypes in the original species descriptions and data from our collected specimens. Numbers in parentheses indicate the average and standard deviation. (M: male; F,J: juvenile female; J: juvenile male; J: data not applicable or available).

| Species | <i>Amolops splendidissimus</i> | | | | | <i>Amolops caelumnoctis</i> | | | | |
|-------------|--------------------------------|-----------------------------|-----------------------------|---------|----------------------------|-----------------------------|-----------------------------|-----------------------------|-----------------------------|--|
| | Lai Chau, Vietnam | | Lai Chau, Vietnam | | Lao Cai, Vietnam | Yunnan, China | | Yunnan, China | | |
| Locality | (holotype) | | (holotype) | | (holotype) | (holotype) | | (holotype) | | |
| Sample size | M (1) | M (4) | F (2) | F,J (1) | J (2) | M (1) | M (5) | M (5) | M (5) | |
| SVL(mm) | 76.6 | 62.1–73.4 (69.0±5.2) | 88.3–89.8 (89.0±1.1) | 50.1 | 36.4–39.1 (37.8±1.9) | 71.3 | 77.8–83.1 (80.7±2.0) | 77.8–83.1 (80.7±2.0) | 77.8–83.1 (80.7±2.0) | |
| SNL/SVL(%) | 14.9 | 13.2–13.9 (13.6±0.4) | 12.6–13.1 (12.8±0.4) | 14.8 | 15.1–15.4 (15.2±0.2) | 14.7 | 12.6–13.8 (13.3±0.5) | 12.6–13.8 (13.3±0.5) | 12.6–13.8 (13.3±0.5) | |
| HW/SVL(%) | 33.0 | 33.2–34.4 (33.8±0.6) | 33.0–34.2 (33.6±0.9) | 33.5 | 33.8–34.3 (34.1±0.4) | 34.8 | 32.0–33.4 (32.6±0.7) | 32.0–33.4 (32.6±0.7) | 32.0–33.4 (32.6±0.7) | |
| HL/SVL(%) | 33.9 | 32.7–35.3 (34.2±1.1) | 31.7–33.7 (32.7±1.4) | 37.1 | 34.8–35.2 (35.0±0.3) | 35.5 | 31.7–33.4 (32.9±0.7) | 31.7–33.4 (32.9±0.7) | 31.7–33.4 (32.9±0.7) | |
| HL/HW(%) | 102.8 | 95.8–105.1 (101.2±3.9) | 92.8–102.1 (97.3±6.6) | 110.7 | 103.0–102.4 (102.7±0.4) | 102 | 99.2–103.0 (100.9±1.7) | 99.2–103.0 (100.9±1.7) | 99.2–103.0 (100.9±1.7) | |
| SNL/HW(%) | 45.1 | 38.3–41.5 (40.5±1.5) | 36.8–39.7 (38.2±2.0) | 44.0 | 44.7–44.8 (44.7±0.1) | 42.3 | 37.7–42.2 (40.7±1.8) | 37.7–42.2 (40.7±1.8) | 37.7–42.2 (40.7±1.8) | |
| ED/SNL(%) | 77.2 | 65.0–77.2 (72.8±5.5) | 75.0–76.1 (75.6±0.8) | 78.4 | 76.3–78.6 (77.4±1.6) | 83.8 | 77.5–92.9 (83.0±6.2) | 77.5–92.9 (83.0±6.2) | 77.5–92.9 (83.0±6.2) | |
| ED/HL(%) | 33.8 | 26.3–31.5 (29.1±2.3) | 29.2–30.2 (29.7±0.7) | 31.2 | 33.1–34.4 (33.7±0.9) | 34.8 | 31.4–35.1 (33.4±1.7) | 31.4–35.1 (33.4±1.7) | 31.4–35.1 (33.4±1.7) | |
| EN/ED(%) | 53.4 | 58.4–72.4 (62.9±6.5) | 54.8–70.4 (62.1±11.0) | 51.7 | 57.8–61.4 (59.6±2.5) | / | 52.4–54.9 (53.7±1.0) | 52.4–54.9 (53.7±1.0) | 52.4–54.9 (53.7±1.0) | |
| TYE/TYD(%) | 152 | 145.0–195.7 (176.8±22.8) | 166.4–206.7 (185.5±28.5) | 104.3 | 82.4–107.1 (94.7±17.5) | / | 135.7–168.0 (154.4±14.9) | 135.7–168.0 (154.4±14.9) | 135.7–168.0 (154.4±14.9) | |
| TYD/ED(%) | 28.4 | 29.0–40.3 (33.0±5.1) | 30.4–34.2 (32.3±2.7) | 39.7 | 37.8–31.8 (34.8±4.2) | 35.2 | 29.1–35.4 (31.4±2.5) | 29.1–35.4 (31.4±2.5) | 29.1–35.4 (31.4±2.5) | |

TABLE 4. General morphological comparisons between *A. splendissimus* and *A. caelumnoctis*, with original descriptions of the type series included (Orlov & Ho, 2007; Rao & Wilkinson, 2007).

| | <i>A. splendissimus</i> | <i>A. caelumnoctis</i> |
|---|---|---|
| Body size | Large | Large |
| Dorsal pattern | Many small bright yellow dots | Many small bright yellow dots |
| Pineal body | Not visible | Not visible |
| Male vocal sac | Absent | Absent |
| Eyes size | Large (ED/SNL: 65.0%–78.4%; ED/HL: 26.3%–34.4%; EN/ED: 51.7%–72.4%; TYD/ED: 28.4%–40.3%; n = 8) | Large (ED/SNL: 77.5%–92.6%; ED/HL: 31.4%–35.1%; EN/ED: 52.4%–54.6%; TYD/ED: 29.1%–35.4%; n = 6) |
| Dorsolateral fold | Absent | Absent |
| Loreal region | Vertical | Vertical |
| Skin texture | Smooth | Smooth |
| Transverse bands on dorsal surface of limbs | Absent | Absent |
| Webbing on feet | Full | Full |
| Nuptial pad on adult males | Present | Present |
| Tympanum | Small, slightly visible | Small, distinct |
| Vomerine teeth | Two, small, oblique vomerine teeth | Two small, oblique vomerine teeth near choanae |
| Snout | Round | Short and round |
| Tongue | Cordiform, deeply notched posteriorly | Deeply notched, posterior third free |
| Circum marginal grooves on fingers | Present | Present |
| Lateral fringes on hands | Absent | Absent |
| Outer metatarsal tubercle | Absent | Absent |
| Temporal/supratympanic fold | Present weakly | Present |
| Pectoral spines/gular pouches | Absent | Absent |

Discussion

Firstly, there is very low genetic divergence between *A. splendissimus* and *A. caelumnoctis* from or near their type localities, with low uncorrected *p*-distances between populations (Wenshan vs. Luchun samples: 0.2%–0.4%; Wenshan vs. Bat Xat samples: 0.0%; Luchun vs. Bat Xat samples: 0.2%–0.4%) (Table S1) at the 16S mitochondrial gene examined. Variation in uncorrected *p*-distance was lowest between populations that were closest to one another, but all variation was substantially lower than 3.0%, the threshold proposed for candidate species at the 16S gene (Vences *et al.* 2005). Secondly, morphological characteristics were similar among newly collected specimens and descriptions of the types of *A. splendissimus* and *A. caelumnoctis* (Table 3, Figures 3–5). The question then is priority. *A. caelumnoctis* is a junior synonym of *A. splendissimus* as the latter has nomenclatural priority by chronological order of description. The work describing *A. splendissimus* was officially published 13 days earlier than the description of *A. caelumnoctis*. Additionally, for Chinese populations, our sample from Wenshan, Yunnan displayed a closer genetic affinity to the Vietnamese Bat Xat population, where it is closer in geographic distance to Bat Xat, Lao Cai than to Luchun, Yunnan (Figure 1, ~98.25 km). The specimen from Wenshan was also more similar in general morphology to Bat Xat specimens in having larger irregular spots on the dorsum and limbs (Luchun specimens have smaller and denser spots; Figures 3–5). However, only one specimen was collected from Wenshan, hence, more specimens are needed to understand the genetic and geographic patterns of variation among populations. Based on evidence from the morphology and molecular data, we conclude that *A. splendissimus* and *A. caelumnoctis* are not distinct species.

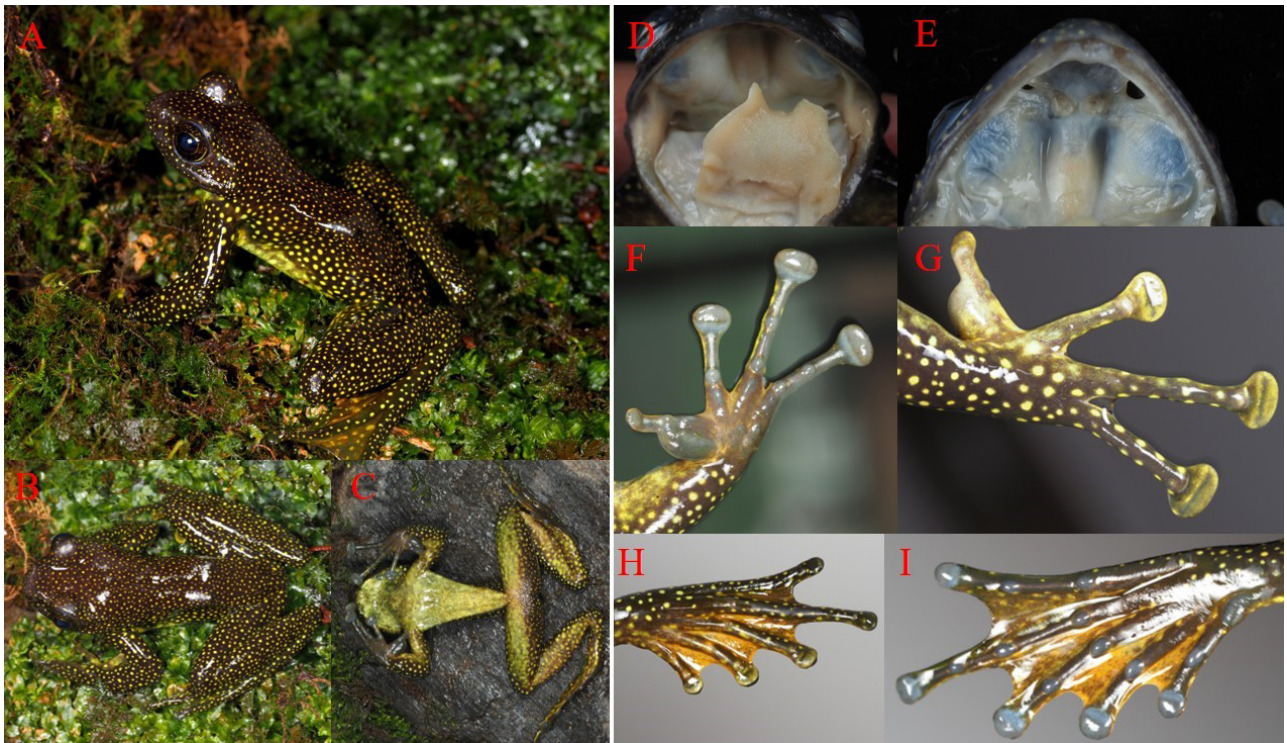


FIGURE 3. *Amolops caelumnoctis* in life from Wenshan County (SWFU 004443, A–I, Photos by Zhiyong Yuan). (A) adult male *in situ*, (B) and same individual in dorsal and (C) ventral view, and (D) inside of mouth showing tongue and (E) maxillary and vomerine teeth, plus (F) ventral and (G) dorsal views of hand and, (H) ventral and (I) dorsal view of foot showing webbing.

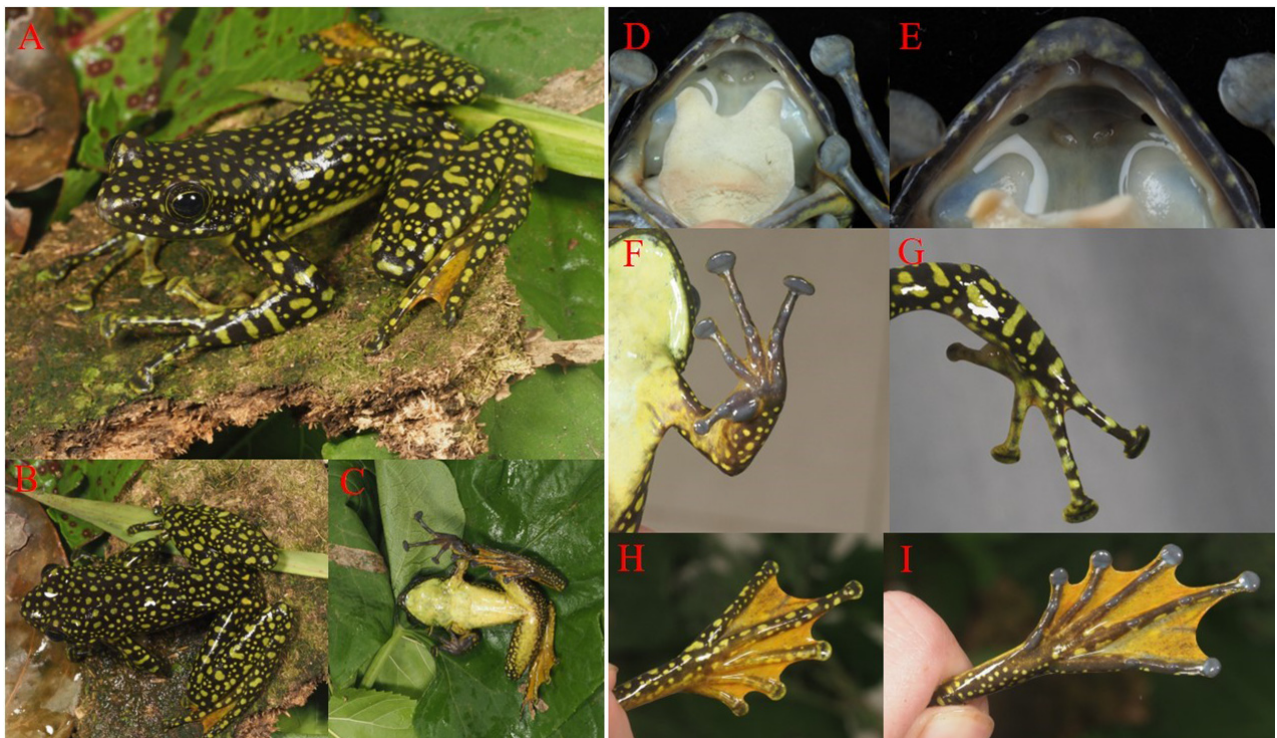


FIGURE 4. *Amolops caelumnoctis* in life from Luchun County (SWFU 004612, A–I, Photos by Xiaolong Liu). (A) adult male *in situ*, (B) and same individual in dorsal and (C) ventral view, and (D) inside of mouth showing tongue and (E) maxillary and vomerine teeth, plus (F) ventral and (G) dorsal views of hand and, (H) ventral and (I) dorsal view of foot showing webbing.

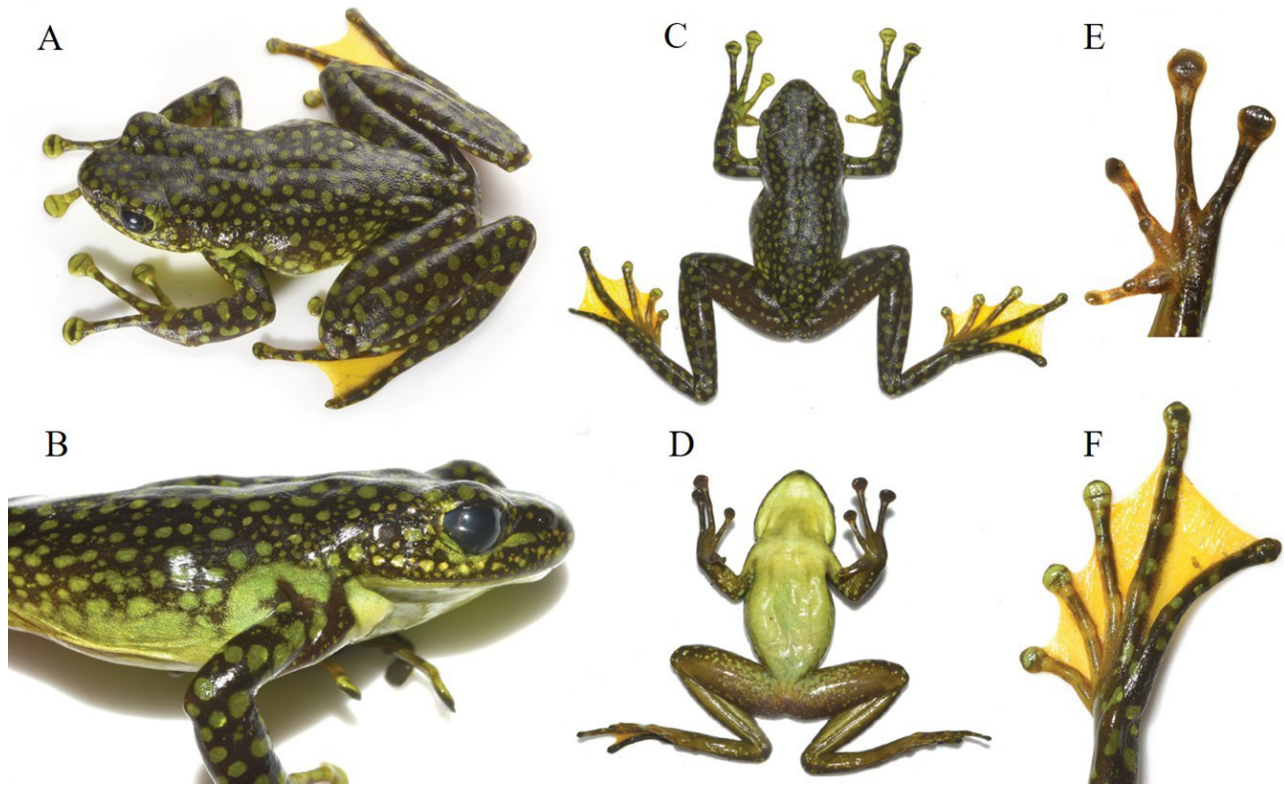


FIGURE 5. *Amolops splendissimus* in life from Bat Xat District Lao Cai Province (AMS R188526, A–F, Photos by Jodi Rowley). (A) adult female, dorsolateral, (B) head lateral, and (C) dorsal view, and (D) ventral view, (E) ventral view of hand, (F) and ventral view of foot showing webbing.

This taxonomic revision has consequences to our understanding of extinction risk. *Amolops splendissimus* is currently assessed as Vulnerable (VU) due to its limited range and ongoing decline in the extent and quality of its habitat (IUCN SSC, 2017). *Amolops caelumnoctis* is assessed as Least Concern (LC) by the IUCN as some of the associated habitat is protected and in good condition and there are no known major threats (IUCN SSC, 2020). It is likely that *Amolops splendissimus* qualifies for being reassessed as Least Concern as this synonymization results in additional known locations of the species and an increase in the Extent of Occurrence, including within protected areas in China.

A significant barrier to the understanding of biodiversity in border areas is insufficient communication and data exchange between neighboring countries. This collaboration between researchers from both Vietnam and China resolves the disputed taxonomic status of this torrent frog species, and we hope that many more taxonomic uncertainties along the China-Vietnam can be resolved through similar collaboration. For instance, though lacking solid evidence, *Varanus nebulosus* (Gray) is suspected to be a junior synonym of *V. vietnamensis* (De Lisle) (Wang *et al.*, 2020b; Böhme and Ziegler, 1997; Rao and Yang, 1996), and *Sylvirana hekouensis* (Fei, Ye, and Jiang) and *S. menglaensis* (Fei, Ye, and Xie) are now junior synonyms of *S. nigrovittata* (Sheridan and Stuart, 2018; AmphibiaChina, 2021). We believe that future collaborations using integrative methods such as morphological, ecological, and molecular data will strongly promote fauna diversity research along China-Vietnam border and that the resolution of taxonomic uncertainty may facilitate the robust prioritization of amphibians for subsequent conservation intervention.

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TABLE S1. The pairwise distance (*p*-distance) results between one population of *A. splendidissimus* from Vietnam, *A. caelumnotis* from China, and other selected *Amolops* species. The distance between *A. splendidissimus* and *A. caelumnotis* marked in bold in their close relationship.

| GenBank ID | Species | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 |
|------------|----------------------------|-------|-------|-------|-------|-------|-------|-------|-------|-------|
| 1 | <i>Amolops daiyunensis</i> | | | | | | | | | |
| 2 | <i>A. lifanensis</i> | 0.099 | | | | | | | | |
| 3 | <i>A. aniqiaoensis</i> | 0.105 | 0.075 | | | | | | | |
| 4 | <i>A. chunganensis</i> | 0.081 | 0.060 | 0.052 | | | | | | |
| 5 | <i>A. yunkaiensis</i> | 0.073 | 0.102 | 0.110 | 0.085 | | | | | |
| 6 | <i>A. sinensis</i> | 0.065 | 0.108 | 0.108 | 0.089 | 0.038 | | | | |
| 7 | <i>A. panhai</i> | 0.137 | 0.111 | 0.140 | 0.136 | 0.137 | 0.145 | | | |
| 8 | <i>A. granulosis</i> | 0.089 | 0.047 | 0.064 | 0.044 | 0.079 | 0.089 | 0.111 | | |
| 9 | <i>A. chuichengicus</i> | 0.089 | 0.055 | 0.058 | 0.041 | 0.156 | 0.094 | 0.126 | 0.019 | |
| 10 | <i>A. tuberodepressus</i> | 0.093 | 0.062 | 0.058 | 0.046 | 0.089 | 0.092 | 0.126 | 0.021 | 0.017 |
| 11 | <i>A. mantorum</i> | 0.093 | 0.058 | 0.058 | 0.042 | 0.093 | 0.094 | 0.125 | 0.024 | 0.024 |
| 12 | <i>A. jinjiangensis</i> | 0.091 | 0.058 | 0.058 | 0.041 | 0.151 | 0.083 | 0.125 | 0.015 | 0.023 |
| 13 | <i>A. loloensis</i> | 0.091 | 0.055 | 0.056 | 0.044 | 0.087 | 0.091 | 0.123 | 0.019 | 0.011 |
| 14 | <i>A. chayuenis</i> | 0.089 | 0.049 | 0.051 | 0.039 | 0.083 | 0.089 | 0.123 | 0.021 | 0.028 |
| 15 | <i>A. bellulus</i> | 0.091 | 0.051 | 0.049 | 0.041 | 0.153 | 0.098 | 0.130 | 0.036 | 0.111 |
| 16 | <i>A. bellulus</i> | 0.095 | 0.062 | 0.051 | 0.048 | 0.091 | 0.098 | 0.134 | 0.041 | 0.041 |
| 17 | <i>A. nyingchiensis</i> | 0.097 | 0.059 | 0.045 | 0.049 | 0.100 | 0.102 | 0.132 | 0.041 | 0.041 |

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TABLE S1. (Continued)

| GenBank ID | Species | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 |
|------------|--|-------|-------|-------|-------|-------|-------|-------|-------|-------|
| 18 | <i>A. akhaorum</i> | 0.089 | 0.064 | 0.060 | 0.056 | 0.172 | 0.100 | 0.125 | 0.053 | 0.133 |
| 19 | <i>A. mengdingensis</i> | 0.091 | 0.068 | 0.047 | 0.042 | 0.173 | 0.092 | 0.134 | 0.045 | 0.131 |
| 20 | <i>A. archotaphus</i> | 0.105 | 0.073 | 0.060 | 0.056 | 0.096 | 0.100 | 0.136 | 0.051 | 0.051 |
| 21 | <i>A. daorum</i> | 0.093 | 0.064 | 0.043 | 0.046 | 0.091 | 0.094 | 0.128 | 0.041 | 0.045 |
| 22 | <i>A. wenshanensis</i> | 0.097 | 0.062 | 0.062 | 0.052 | 0.087 | 0.097 | 0.131 | 0.043 | 0.047 |
| 23 | <i>A. cucae</i> | 0.093 | 0.064 | 0.062 | 0.054 | 0.152 | 0.098 | 0.126 | 0.049 | 0.121 |
| 24 | <i>A. compotrix</i> | 0.095 | 0.055 | 0.062 | 0.048 | 0.151 | 0.095 | 0.119 | 0.036 | 0.118 |
| 25 | <i>A. chunganensis</i> | 0.093 | 0.064 | 0.068 | 0.041 | 0.101 | 0.108 | 0.131 | 0.057 | 0.064 |
| 26 | <i>A. chunganensis</i> | 0.099 | 0.064 | 0.056 | 0.042 | 0.095 | 0.096 | 0.126 | 0.049 | 0.045 |
| 27 | <i>A. medogensis</i> | 0.097 | 0.062 | 0.064 | 0.054 | 0.091 | 0.100 | 0.128 | 0.036 | 0.043 |
| 28 | <i>A. viridimaculatus</i> | 0.103 | 0.060 | 0.068 | 0.056 | 0.085 | 0.102 | 0.125 | 0.034 | 0.038 |
| 29 | <i>A. splendissimus</i> (Bat Xat, Lao Cai) | 0.103 | 0.066 | 0.070 | 0.054 | 0.089 | 0.106 | 0.131 | 0.045 | 0.040 |
| 30 | <i>A. splendissimus</i> (Bat Xat, Lao Cai) | 0.103 | 0.066 | 0.070 | 0.054 | 0.089 | 0.106 | 0.131 | 0.045 | 0.040 |
| 31 | <i>A. splendissimus</i> (Bat Xat, Lao Cai) | 0.103 | 0.066 | 0.070 | 0.054 | 0.089 | 0.106 | 0.131 | 0.045 | 0.040 |
| 32 | <i>A. caelumnoctis</i> (Wenshan, Yunnan) | 0.103 | 0.066 | 0.070 | 0.054 | 0.088 | 0.106 | 0.130 | 0.045 | 0.041 |
| 33 | <i>A. caelumnoctis</i> (Luchun, Yunnan) | 0.099 | 0.066 | 0.070 | 0.054 | 0.088 | 0.104 | 0.132 | 0.041 | 0.039 |
| 34 | <i>A. caelumnoctis</i> (Luchun, Yunnan) | 0.101 | 0.064 | 0.068 | 0.052 | 0.086 | 0.104 | 0.130 | 0.043 | 0.040 |

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TABLE S1. (Continued)

| | GenBank ID | Species | 10 | 11 | 12 | 13 | 14 | 15 | 16 | 17 | 18 | 19 |
|----|------------|----------------------------|-------|-------|-------|-------|-------|-------|-------|----|----|----|
| 1 | MN953677 | <i>Amolops daiyunensis</i> | | | | | | | | | | |
| 2 | MN953702 | <i>A. lifanensis</i> | | | | | | | | | | |
| 3 | MN953658 | <i>A. aniqiaoensis</i> | | | | | | | | | | |
| 4 | MN953671 | <i>A. chunganensis</i> | | | | | | | | | | |
| 5 | MK263271 | <i>A. yunkaiensis</i> | | | | | | | | | | |
| 6 | MN953748 | <i>A. sinensis</i> | | | | | | | | | | |
| 7 | MN953722 | <i>A. panhai</i> | | | | | | | | | | |
| 8 | MN953680 | <i>A. granulatus</i> | | | | | | | | | | |
| 9 | MK604845 | <i>A. chuichengicus</i> | | | | | | | | | | |
| 10 | MN953729 | <i>A. tuberodepressus</i> | | | | | | | | | | |
| 11 | MN953706 | <i>A. mantzorum</i> | 0.024 | | | | | | | | | |
| 12 | EF453741 | <i>A. jinjiangensis</i> | 0.013 | 0.021 | | | | | | | | |
| 13 | MN953704 | <i>A. loloensis</i> | 0.017 | 0.024 | 0.008 | | | | | | | |
| 14 | MN953666 | <i>A. chayuenis</i> | 0.030 | 0.023 | 0.024 | 0.028 | | | | | | |
| 15 | FJ417126 | <i>A. bellulus</i> | 0.041 | 0.038 | 0.093 | 0.040 | 0.038 | | | | | |
| 16 | MN953664 | <i>A. bellulus</i> | 0.043 | 0.047 | 0.041 | 0.041 | 0.047 | 0.023 | | | | |
| 17 | MN953719 | <i>A. nyingchiensis</i> | 0.043 | 0.043 | 0.041 | 0.041 | 0.047 | 0.016 | 0.012 | | | |

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TABLE S1. (Continued)

| GenBank ID | Species | 10 | 11 | 12 | 13 | 14 | 15 | 16 | 17 | 18 | 19 |
|------------|----------|--|-------|-------|-------|-------|-------|-------|-------|-------|-------|
| 18 | FJ417158 | <i>A. akhaorum</i> | 0.066 | 0.062 | 0.116 | 0.051 | 0.055 | 0.093 | 0.056 | 0.059 | |
| 19 | MK501808 | <i>A. mengdingensis</i> | 0.051 | 0.047 | 0.114 | 0.043 | 0.047 | 0.091 | 0.040 | 0.041 | 0.042 |
| 20 | MN953659 | <i>A. archotaphus</i> | 0.056 | 0.055 | 0.045 | 0.053 | 0.053 | 0.045 | 0.056 | 0.055 | 0.043 |
| 21 | MN953678 | <i>A. daorum</i> | 0.047 | 0.043 | 0.040 | 0.040 | 0.040 | 0.028 | 0.036 | 0.033 | 0.038 |
| 22 | MN953725 | <i>A. wenshanensis</i> | 0.049 | 0.049 | 0.043 | 0.043 | 0.045 | 0.042 | 0.045 | 0.047 | 0.057 |
| 23 | FJ417145 | <i>A. cucae</i> | 0.055 | 0.051 | 0.102 | 0.045 | 0.047 | 0.072 | 0.047 | 0.049 | 0.086 |
| 24 | FJ417141 | <i>A. compotrix</i> | 0.045 | 0.049 | 0.099 | 0.040 | 0.042 | 0.069 | 0.045 | 0.047 | 0.080 |
| 25 | MN953670 | <i>A. chunganensis</i> | 0.066 | 0.060 | 0.064 | 0.068 | 0.051 | 0.053 | 0.060 | 0.063 | 0.064 |
| 26 | MN953767 | <i>A. chunganensis</i> | 0.049 | 0.047 | 0.045 | 0.049 | 0.040 | 0.043 | 0.045 | 0.047 | 0.068 |
| 27 | MN953710 | <i>A. medogensis</i> | 0.045 | 0.041 | 0.040 | 0.043 | 0.034 | 0.043 | 0.041 | 0.045 | 0.068 |
| 28 | MN953737 | <i>A. viridimaculatus</i> | 0.047 | 0.043 | 0.041 | 0.045 | 0.036 | 0.047 | 0.049 | 0.053 | 0.064 |
| 29 | MZ484725 | <i>A. splendissimus</i> (Bat Xat, Lao Cai) | 0.051 | 0.045 | 0.044 | 0.042 | 0.042 | 0.051 | 0.051 | 0.055 | 0.070 |
| 30 | MZ484726 | <i>A. splendissimus</i> (Bat Xat, Lao Cai) | 0.051 | 0.045 | 0.044 | 0.042 | 0.042 | 0.051 | 0.051 | 0.055 | 0.070 |
| 31 | MZ484727 | <i>A. splendissimus</i> (Bat Xat, Lao Cai) | 0.051 | 0.045 | 0.044 | 0.042 | 0.042 | 0.051 | 0.051 | 0.055 | 0.070 |
| 32 | MZ484728 | <i>A. caelumnoctis</i> (Wenshan, Yunnan) | 0.053 | 0.045 | 0.045 | 0.043 | 0.041 | 0.050 | 0.051 | 0.055 | 0.070 |
| 33 | MZ484729 | <i>A. caelumnoctis</i> (Luchun, Yunnan) | 0.049 | 0.041 | 0.041 | 0.040 | 0.041 | 0.047 | 0.047 | 0.051 | 0.066 |
| 34 | MZ484730 | <i>A. caelumnoctis</i> (Luchun, Yunnan) | 0.051 | 0.043 | 0.043 | 0.041 | 0.040 | 0.048 | 0.049 | 0.053 | 0.068 |

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TABLE S1. (Continued)

| GenBank ID | Species | 20 | 21 | 22 | 23 | 24 | 25 | 26 | 27 | 28 | 29 | 30 | 31 | 32 | 33 |
|------------|--|-------|-------|-------|-------|-------|-------|-------|-------|-------|--------------|--------------|--------------|--------------|--------------|
| 20 | <i>A. archotaphus</i> | | | | | | | | | | | | | | |
| 21 | <i>A. daorum</i> | 0.032 | | | | | | | | | | | | | |
| 22 | <i>A. wenshanensis</i> | 0.043 | 0.040 | | | | | | | | | | | | |
| 23 | <i>A. cucae</i> | 0.058 | 0.041 | 0.028 | | | | | | | | | | | |
| 24 | <i>A. compotrix</i> | 0.049 | 0.043 | 0.023 | 0.025 | | | | | | | | | | |
| 25 | <i>A. chunganensis</i> | 0.064 | 0.055 | 0.061 | 0.059 | 0.057 | | | | | | | | | |
| 26 | <i>A. chunganensis</i> | 0.055 | 0.045 | 0.051 | 0.049 | 0.051 | 0.051 | | | | | | | | |
| 27 | <i>A. medogensis</i> | 0.062 | 0.045 | 0.055 | 0.055 | 0.053 | 0.068 | 0.053 | | | | | | | |
| 28 | <i>A. viridimaculatus</i> | 0.058 | 0.049 | 0.058 | 0.053 | 0.051 | 0.068 | 0.056 | 0.028 | | | | | | |
| 29 | <i>A. splendissimus</i> (Bat Xat, Lao Cai) | 0.072 | 0.055 | 0.060 | 0.056 | 0.054 | 0.070 | 0.059 | 0.034 | 0.023 | | | | | |
| 30 | <i>A. splendissimus</i> (Bat Xat, Lao Cai) | 0.072 | 0.055 | 0.060 | 0.056 | 0.054 | 0.070 | 0.059 | 0.034 | 0.023 | 0.000 | | | | |
| 31 | <i>A. splendissimus</i> (Bat Xat, Lao Cai) | 0.072 | 0.055 | 0.060 | 0.056 | 0.054 | 0.070 | 0.059 | 0.034 | 0.023 | 0.000 | 0.000 | | | |
| 32 | <i>A. caelumnoctis</i> (Wenshan, Yunnan) | 0.072 | 0.055 | 0.060 | 0.056 | 0.054 | 0.070 | 0.058 | 0.034 | 0.023 | 0.000 | 0.000 | 0.000 | | |
| 33 | <i>A. caelumnoctis</i> (Luchun, Yunnan) | 0.068 | 0.051 | 0.057 | 0.055 | 0.054 | 0.074 | 0.058 | 0.034 | 0.023 | 0.004 | 0.004 | 0.004 | 0.004 | |
| 34 | <i>A. caelumnoctis</i> (Luchun, Yunnan) | 0.070 | 0.053 | 0.058 | 0.054 | 0.052 | 0.072 | 0.056 | 0.032 | 0.021 | 0.002 | 0.002 | 0.002 | 0.002 | 0.002 |