# Taxonomic study of four closely-related species of the Pholcus yichengicus species group (Araneae, Pholcidae) from China's Qinling Mountains: An integrated morphological and molecular approach 

Lan Yang ${ }^{1}$, Qiaoqiao $\mathrm{He}^{1}$, Zhiyuan Yao ${ }^{1}$<br>1 College of Life Science, Shenyang Normal University, Shenyang 110034, Liaoning, China<br>https://zoobank.org/45A26952-6A37-4C42-99D6-DB5E6AD1BCC4<br>Corresponding author: Qiaoqiao He (heqq@synu.edu.cn); Zhiyuan Yao (yaozy@synu.edu.cn)

Academic editor: Danilo Harms * Received 24 November 2023 • Accepted 21 February 2024 • Published 14 March 2024


#### Abstract

Four morphologically similar species of the Pholcus yichengicus species group, occurring in geographic proximity of China's Qinling Mountains, were recognised, based on morphology and four methods of molecular species delimitation. They comprise two new species, namely Pholcus ankang sp. nov. and P. baoji sp. nov. and two previously described species: P. ovatus Yao \& Li, 2012 and P. taibaiensis Wang \& Zhu, 1992. Their DNA barcodes were obtained to estimate p-distances and K2P distances. In addition, an identification key for the four closely-related species is presented.


## Key Words

Biodiversity, daddy-long-legs spider, identification key, molecular species delimitation, new species

## Introduction

The family Pholcidae C.L. Koch, 1850 is a highly diverse group of spiders, with 97 genera and 1,937 species (World Spider Catalog 2023), classified under five subfamilies: Arteminae Simon, 1893, Modisiminae Simon, 1893, Ninetinae Simon, 1890, Pholcinae C.L. Koch, 1850 and Smeringopinae Simon, 1893 (Huber 2011a; Dimitrov et al. 2013; Huber et al. 2018). Pholcus Walckenaer, 1805 is the most diverse genus within Pholcinae, with 389 described species belonging to 21 species groups distributed mainly in the Palaearctic, Oriental, Afrotropical and Australasian biogeographic realms (Huber 2011b; Huber et al. 2018; World Spider Catalog 2023).

Bestriding the Palaearctic and Oriental Regions, China harbours a high diversity of Pholcus spiders. Recently, a large number of new species of Pholcus have been reported from northern China, based on morphological and molecular data. For instance, the extensive 2020 expedition into the Changbai Mountains, at the border between north-eastern China and North Korea, brings the species
count of Pholcus in the Changbai Mountains to 27 species, including 13 new species (Lu et al. 2021; Yao et al. 2021; Zhao et al. 2023a). The systematic investigation in the Yanshan-Taihang Mountains in northern China in 2021 recorded 36 Pholcus species, of which 14 species were new to science (Lu et al. 2022a, b). In 2022, Pholcus spiders were collected for the first time during an expedition to the Lüliang Mountains in Shanxi Province, northern China. The study identified one known species and eight new species (Zhao et al. 2023b). So far, 169 species, $43 \%$ of the genus, have been recorded in China (World Spider Catalog 2023).

China's Qinling Mountains is generally regarded as a geographical dividing line between northern China and southern China. It straddles the Provinces of Shanxi, Shaanxi and Henan. To date, 14 species of Pholcus have been found to be recorded in the Qinling Mountains (Zhang and Zhu 2009; Yao and Li 2012; Dong et al. 2016; World Spider Catalog 2023). This paper identifies four morphologically similar species from the Qinling Mountains (Shaanxi part, Fig. 1), based on
morphological and molecular evidence, including two new species to be described and all belong to the Pholcus yichengicus species group.

## Materials and methods

Specimens were examined and measured with a Leica M205 C stereomicroscope. Left male pedipalps were photographed. Epigynes were photographed before dissection. Vulvae were illustrated after treating them in a $10 \%$ warm solution of potassium hydroxide $(\mathrm{KOH})$ to dissolve soft tissues. Images were captured with a Canon EOS 750D wide zoom digital camera ( 24.2 megapixels) mounted on the stereomicroscope mentioned above and assembled using Helicon Focus v. 3.10.3 image stacking software (Khmelik et al. 2005). All measurements are given in millimetres (mm). Leg measurements are shown as: total length (femur, patella, tibia, metatarsus, tarsus). Leg segments were measured on their dorsal side. The distribution map was generated with ArcGIS v. 10.2 (ESRI Inc.). The specimens studied are preserved in $75 \%$ ethanol and deposited in the College of Life Science, Shenyang Normal University (SYNU) in Liaoning, China.

Terminology and taxonomic descriptions follow Huber (2011b) and Yao et al. $(2015,2021)$. The following abbreviations are used in the descriptions: ALE = anterior lateral eye, $\mathbf{A M E}=$ anterior median eye, $\mathbf{P M E}=$ posterior median eye, $\mathbf{L} / \mathbf{d}=$ length/diameter ratio; used in the illustrations: $\mathbf{a}=$ appendix, $\mathbf{b}=$ bulb, $\mathbf{d a}=$ distal apophysis, $\mathbf{e}=$ embolus, $\mathbf{f a}=$ frontal apophysis, $\mathbf{p a}=$ proximo-lateral apophysis, $\mathbf{p p}=$ pore plate, $\mathbf{p r}=$ procursus, $\mathbf{u}=$ uncus.

The mitochondrial gene fragment encoding COI and two nuclear gene fragments encoding H3 and wnt were obtained for 19 samples (Table 1). Primers are listed in Table 2. Two species Pholcus paralinzhou and
P. taishan were selected as outgroups. DNA sequences were checked and edited with BioEdit 7.2.2 (Hall 1999). P-distances and K2P distances from COI were computed using MEGA 5 (Tamura et al. 2011). Phylogenetic trees were constructed using the Maximum Likelihood (ML) method for molecular species delimitation, using both COI and a combined dataset. ML analyses were conducted using RAxML 8.2.9 under a GTRCAT model for all partitions, with 500 rapid bootstrap replicates followed

Table 1. Voucher specimen information.

| Species | Voucher | GenBank accession number |  | Collection |  |
| :--- | :---: | :---: | :---: | :---: | :---: |
|  | code | COI | H3 | wnt | locality |
| P. ankang | W265 | PP082941 | PP349964 | PP349983 | China, |
| sp. nov. | W266 | PP082942 | PP349965 | PP349984 | Shaanxi, |
|  | W267 | PP082943 | PP349966 | PP349985 | Ankang |
|  | W268 | PP082944 | PP349967 | PP349986 |  |
|  | W269 | PP082945 | PP349968 | PP349987 |  |
| P. baoji | W270 | PP082946 | PP349969 | PP349988 | China, |
| sp. nov. | W271 | PP082947 | PP349970 | PP349989 | Shaanxi, |
|  | W272 | PP082948 | PP349971 | PP349990 | Baoji |
|  | W273 | PP082949 | PP349972 | PP349991 |  |
| P. ovatus | W220 | PP082951 | PP349955 | PP349974 | China, |
|  | W221 | PP082952 | PP349956 | PP349975 | Shaanxi, |
|  | W222 | PP082953 | PP349957 | PP349976 | Xian |
|  | W223 | PP082954 | PP349958 | PP349977 |  |
| P. | W224 | PP082955 | PP349959 | PP349978 | China, |
| taibaiensis | W225 | PP082956 | PP349960 | PP349979 | Shaanxi, |
|  | W226 | PP082957 | PP349961 | PP349980 | Baoji |
|  | W227 | PP082958 | PP349962 | PP349981 |  |
| P. | W228 | PP082959 | PP349963 | PP349982 |  |
| paralinzhou | y046 | MW721825 | ON375203 | ON375294 | China, |
|  |  |  |  |  | Henan, |
| P. taishan | y133 | MW721826 | ON375204 | ON375293 | China, |
|  |  |  |  |  | Shandong, |
|  |  |  |  | Taian |  |



Figure 1. Distribution records of four closely-related species of Pholcus from the Qinling Mountains, China. 1. Pholcus ankang sp. nov.; 2. P. baoji sp. nov.; 3. P. ovatus; 4. P. taibaiensis.

Table 2. Primers used for amplification and sequencing.

| Gene | Primer | F/R | Sequence 5'-3' | Reference |
| :---: | :--- | :---: | :---: | :---: |
| COI | LCO1490 | F | GGTCAACAAATCATAAAGATATTGG | Folmer et al. <br> $(1994)$ |
|  | C1-N-2776 | R | GGATAATCAGAATANCGNCGAGG | Vink et al. <br> (2005) |
| H3 | H3af | F | ATGGCTCGTACCAAGCAGACVGC | Colgan et al. <br> wnt <br>  <br>  <br> wht <br>  <br> H3ar |
| Spwgf1 | R | ATATCCTTRGGCATRATRGTGAC | (1998) |  |
|  | Spwgr1 | R | ACTTGRCAACACCARTGAAAWG | et al. (2009) |
|  | Wnt2f | F | CAGTGRAATGTRCARTTG |  |
|  | Wnt2r | R | CNGTTCAAACTTGYTGGATG |  |

by a thorough Maximum Likelihood tree search (Stamatakis 2014). The sequences are deposited in GenBank. For additional information on extraction, amplification and sequencing procedures, see Yao et al. (2016).

We applied four methods for molecular species delimitation. The Automatic Barcode Gap Discovery (ABGD) analyses were conducted using both Jukes-Cantor and Kimura 2-P distance matrices with options: Pmin $=$ $0.001, \operatorname{Pmax}=0.1$, Steps $=10, \mathrm{X}=1.0, \mathrm{Nb}$ bins $=20$ (Puillandre et al. 2012). The Bayesian implementation of the Poisson Tree Processes (bPTP) analysis was run for 100,000 generations, with a thinning of 100 and burnin of 0.25 (Zhang et al. 2013). The Generalised Mixed Yule Coalescent (GMYC) analysis was performed under the single threshold model using the R 4.2.2 package SPLITS (R Development Core Team 2023). The phylogenetic tree was converted to an ultrametric format for GMYC analysis using BEAST 1.8.2 (Drummond et al. 2012). Bayesian Phylogenetics and Phylogeography (BPP) requires data from multiple genes and pre-defined candidate species. We used BPP to test the results between the analyses mentioned above. Like Yang (2015), we conducted four different sets of analyses with different values of $\alpha$ and $\beta: G \theta(2,1000)$ and $G \tau(2,2000)$, $G \theta(2,100)$ and $G \tau(2,200), G \theta(2,100)$ and $G \tau(2,2000)$, $G \theta(2,1000)$ and $G \tau(2,200)$. The analyses were performed using the following settings: species delimitation $=1$, algorithm $=0$, finetune $=5$. The reversible-jump MCMC analyses were run for 100,000 generations and sampled every two generations, with 25,000 samples being discarded as burn-in.

## Results

We obtained a concatenated alignment of 1767 bp (COI, 1184 bp; H3, 293 bp; wnt, 290 bp). Separate phylogenetic analyses of the individual gene COI and concatenated data found compatible topologies. Fig. 2 presents the phylogenetic tree from the concatenated data. The tree clearly divided the samples into four deeply divergent clades. We defined the four major clades as four candidate species, because the ABGD, GMYC and bPTP analyses unambiguously support their status as separate species and the results were fairly consistent with morphology. We used BPP to validate the four species. The BPP analyses found speciation probabilities of one for all of the nodes tested using all four prior combinations. Furthermore, the smallest p-distance and K2P distance amongst the species is 0.068 and 0.072 , respectively (between $P$. ovatus and $P$. taibaiensis) (Table 3). Of the four species, two are new and descriptions are provided below.

Table 3. The average uncorrected p-distances (below diagonal) and K2P distances (above diagonal) amongst the species and the maximum p-distances (on diagonal) within each species.

|  | P. ankang <br> sp. nov. | P. baoji <br> sp. nov. | P. ovatus | P. taibaiensis |
| :--- | :---: | :---: | :---: | :---: |
| P. ankang sp. nov. | $\mathbf{0 . 0 0 1}$ | 0.099 | 0.102 | 0.119 |
| P. baoji sp. nov. | 0.092 | $\mathbf{0}$ | 0.096 | 0.102 |
| P. ovatus | 0.094 | 0.090 | $\mathbf{0 . 0 0 1}$ | 0.072 |
| P. taibaiensis | 0.109 | 0.095 | 0.068 | $\mathbf{0 . 0 0 1}$ |

## Taxonomy

Family Pholcidae C.L. Koch, 1850
Subfamily Pholcinae C.L. Koch, 1850

## Genus Pholcus Walckenaer, 1805

Type species. Aranea phalangioides Fuesslin, 1775

## Pholcus yichengicus species group

Diagnosis and description. See Huber (2011b).

## Identification key

Note that males and females must be present for this key to work.
1 Sclerotised prolatero-subdistal apophysis of procursus prolatero-proximally strongly widened (figs 134C, 137A in Yao and Li (2012)); raised prolatero-subdistal membranous edge of procursus laterally strongly curved (figs 134D, 137B in Yao and Li (2012)); appendix with slender median branch (branch length/appendix length ratio: 0.5); vulval anterior arch postero-medially strongly protruding (figs 135B, 137D in Yao and Li (2012)). . P. ovatus

- Sclerotised prolatero-subdistal apophysis of procursus not widened prolatero-proximally (e.g. fig. 169C in Yao and Li (2012)); raised prolatero-subdistal membranous edge of procursus laterally angular or rectangular (e.g. fig. 169D in Yao and Li (2012) and Fig. 5D); appendix with slender median branch (branch length/appendix length ratio: 1 or 0.2 ) or angular branch; vulval anterior arch not protruding postero-medially (e.g. Fig. 4B). 2

2 Raised prolatero-subdistal membranous edge of procursus laterally rectangular (Fig. 5D); appendix with slender median branch (branch length/appendix length ratio: 1, arrow 2 in Fig. 6C); epigynal plate nearly triangular and posteriorly straight (Fig. 6A); vulval pore plates relatively close to each other (Fig. 6B) $\qquad$ P. baoji sp. nov.

- Raised prolatero-subdistal membranous edge of procursus laterally angular; epigynal plate posteriorly strongly curved (e.g. Fig. 4A); vulval pore plates widely separated (e.g. Fig. 4B)

3 Sclerotised prolatero-subdistal apophysis of procursus not protruding latero-distally (arrow 4 in Fig. 3D); appendix with angular median branch (arrow 2 in Fig. 4C) $\qquad$ P. ankang sp. nov.

- Sclerotised prolatero-subdistal apophysis of procursus latero-distally protruding (fig. 169D in Yao and Li (2012)); appendix with slender median branch (branch length/appendix length ratio: 0.2).......
P. taibaiensis


Figure 2. The results of species delimitation conducted by the ABGD, GMYC, bPTP and BPP analyses; different colours of the bars represent the different species (Phylogenetic tree was inferred from ML analysis, bootstrap values are provided at the nodes).

## Pholcus ankang sp. nov.

https://zoobank.org/D586922C-E2C7-4C52-BE9F-A9AA9F38BD06 Figs 3, 4

Type material. Holotype $\delta^{\lambda}$ (SYNU-Ar00398) and Paratypes $2 \widehat{\lambda}$ (SYNU-Ar00399-400) $2 \circ$ (SY-NU-Ar00401-02), China, Shaanxi, Ankang, Shiquan County, Chengguan Town, Guiguling Scenic Spot ( $33^{\circ} 12.32^{\prime} \mathrm{N}, 108^{\circ} 20.15^{\prime} \mathrm{E}, 1585 \mathrm{~m}$ elev.), 24 July 2022, Z. Yao, L. Yang \& L. Zhang leg.

Etymology. The specific name refers to the type locality and is a noun in apposition.

Diagnosis. The species resembles $P$. baoji sp. nov. with similar male chelicerae and uncus (Fig. 4C, D), but it can be distinguished by raised prolatero-subdistal membranous edge of procursus laterally angular (arrow 3 in Fig. 3D; laterally rectangular in P. baoji sp. nov., arrow 3 in Fig. 5D), by appendix with angular median branch (arrow 2 in Fig. 4C; slender median branch, same length as appendix in $P$. baoji sp. nov., arrow 2 in Fig. 6C), by epigynal plate posteriorly strongly curved (Fig. 4A;
posteriorly straight and epigynal plate nearly triangular in $P$. baoji sp. nov., Fig. 6A) and by vulval pore plates widely separated (Fig. 4B; relatively close to each other in $P$. baoji sp. nov., Fig. 6B).

Description. Male (holotype): Total length 5.17 (5.26 with clypeus), carapace 1.53 long, 1.62 wide, opisthosoma 3.64 long, 1.58 wide. Leg I: 39.46 (10.13, $0.66,9.94$, 16.54, 2.19), leg II: 26.96 (7.50, 0.63, 6.79, 10.57, 1.47), leg III: 18.30 ( $5.38,0.60,4.45,6.79,1.08$ ), leg IV: 24.35 (7.24, 0.61, 6.03, 9.29, 1.18); tibia I L/d: 70. Eye interdistances and diameters: PME-PME 0.25, PME 0.19 , PME-ALE 0.06, AME-AME 0.06, AME 0.15. Sternum width/length: 1.12/0.94. Habitus as in Fig. 4E, F. Carapace yellowish, with brown radiating marks and marginal brown bands; ocular area yellowish, with anterior brown marks; clypeus brown; sternum yellowish, with brown marks. Legs yellowish, but dark brown on patellae and whitish on distal parts of femora and tibiae, with darker rings on subdistal parts of femora and proximal and subdistal parts of tibiae. Opisthosoma yellowish, with dorsal and lateral spots. Chelicerae (Fig. 4D) with pair of


Figure 3. Pholcus ankang sp. nov., holotype male A, B. Pedipalp (A. Prolateral view; B. Retrolateral view); C, D. Distal part of procursus (C. Prolateral view, arrow 1 indicates distal membranous process, arrow 2 indicates sclerotised prolatero-subdistal apophysis; D. Dorsal view, arrows 1, 2 indicate dorsal spines, arrow 3 indicates angular part of raised prolatero-subdistal membranous edge, arrow 4 indicates latero-distal part of sclerotised prolatero-subdistal apophysis). Abbreviations: $a=$ appendix, $b=b u l b, e=$ embolus, $\mathrm{pr}=$ procursus, $\mathrm{u}=$ uncus. Scale bars: $0.20 \mathrm{~mm}(\mathbf{A}, \mathbf{B}) ; 0.10 \mathrm{~mm}(\mathbf{C}, \mathbf{D})$.


Figure 4. Pholcus ankang sp. nov., holotype male (C-F) and paratype female (A, B, G, H) A. Epigyne, ventral view; B. Vulva, dorsal view; C. Bulbal apophyses, prolateral view, arrow 1 indicates latero-median protrusion, arrow 2 indicates angular median branch; D. Chelicerae, frontal view; E-H. Habitus (E, G. Dorsal view; F. Lateral view; H. Ventral view). Abbreviations: $a=$ appendix, $\mathrm{b}=$ bulb, $\mathrm{da}=$ distal apophysis, $\mathrm{e}=$ embolus, $\mathrm{fa}=$ frontal apophysis, $\mathrm{pa}=$ proximo-lateral apophysis, $\mathrm{pp}=$ pore plate, $\mathrm{u}=$ uncus. Scale bars: $0.20 \mathrm{~mm}(\mathbf{A}-\mathbf{D}) ; 1.00 \mathrm{~mm}(\mathbf{E}-\mathbf{H})$.
proximo-lateral apophyses, pair of distal apophyses with two teeth each and pair of frontal apophyses. Pedipalp as in Fig. 3A, B; trochanter with long (longer than wide), retrolaterally strongly bulged ventral apophysis; femur with small retrolatero-proximal apophysis and distinct ventral protuberance; tibia with prolatero-ventral projection; procursus simple proximally, but complex distally, with raised prolatero-subdistal membranous edge bearing distal membranous process (arrow 1 in Fig. 3C), sclerotised prolatero-subdistal apophysis (arrow 2 in Fig. 3C) and two strong dorsal spines (arrows 1 and 2 in Fig. 3D); uncus latero-medially protruding (arrow 1 in Fig. 4C), with proximal apophysis and distal scaly edge; appendix curved, with angular median branch (arrow 2 in Fig. 4C); embolus weakly sclerotised, with some transparent distal projections (Fig. 4C). Retrolateral trichobothrium of tibia I at 7\% proximally; legs with short vertical setae on tibiae, metatarsi and tarsi; tarsus I with 38 distinct pseudosegments.

Female (paratype, SYNU-Ar00401): Similar to male, habitus as in Fig. 4G, H. Total length 4.60 ( 4.75 with clypeus), carapace 1.31 long, 1.44 wide, opisthosoma 3.29 long, 1.22 wide; tibia I: 7.18 ; tibia I L/d: 51. Eye interdistances and diameters: PME-PME 0.23, PME 0.16 , PME-ALE 0.05 , AME-AME 0.05 , AME 0.13 . Sternum width/length: 0.90/0.76. Epigyne (Fig. 4A) posteriorly strongly curved, laterally and medially strongly sclerotised, with column-shaped knob. Vulva (Fig. 4B) with laterally strongly curved, sclerotised anterior arch and pair of nearly elliptic pore plates.

Variation. Tibia I in two paratype males (SY-NU-Ar00399-400): 9.04, 9.36. Tibia I in another paratype female (SYNU-Ar00402): 7.12.

Natural history. The species was found on the underside of an overhang on rocky cliffs.

Distribution. China (Shaanxi, type locality; Fig. 1).

## Pholcus baoji sp. nov.

https://zoobank.org/EE2818D8-A3D6-4E90-BE29-91278647C486 Figs 5, 6

Type material. Holotype $\overparen{ }$ (SYNU-Ar00403) and Paratypes $1 \delta^{\Uparrow}$ (SYNU-Ar00404) 2 (SYNU-Ar00405-06), China, Shaanxi, Baoji, Long County, Xinjichuan Town, Longmendong Scenic Spot ( $35^{\circ} 2.33^{\prime} \mathrm{N}, 106^{\circ} 40.22^{\prime} \mathrm{E}$, 1489 m elev.), 29 July 2022, Z. Yao, L. Yang \& L. Zhang leg.

Etymology. The specific name refers to the type locality and is a noun in apposition.

Diagnosis. The species resembles $P$. ankang sp. nov. with similar male chelicerae and uncus (Fig. 6C, D), but it can be distinguished by raised prolatero-subdistal membranous edge of procursus laterally rectangular (arrow 3 in Fig. 5D; laterally angular in $P$. ankang sp. nov., arrow 3 in Fig. 3D), by appendix with slender median branch (same length as appendix, arrow 2 in Fig. 6C; angular median branch in P. ankang sp. nov., arrow 2 in Fig. 4C), by
epigynal plate nearly triangular and posteriorly straight (Fig. 6A; posteriorly strongly curved in P. ankang sp. nov., Fig. 4A) and by vulval pore plates relatively close to each other (Fig. 6B; widely separated in P. ankang sp. nov., Fig. 4B).

Description. Male (holotype): Total length 4.96 (5.05 with clypeus), carapace 1.48 long, 1.78 wide, opisthosoma 3.48 long, 1.52 wide. Leg I: 42.86 ( $10.77,0.75,11.15$, 17.56, 2.63), leg II: 28.13 (7.82, 0.70, 6.98, 10.96, 1.67), leg III missing, leg IV: 25.71 (7.69, 0.65, 6.28, 9.62, 1.47); tibia I L/d: 70. Eye interdistances and diameters: PMEPME 0.26, PME 0.16, PME-ALE 0.06, AME-AME 0.05 , AME 0.12. Sternum width/length: $1.08 / 0.93$. Habitus as in Fig. 6E, F. Carapace yellowish, with brown radiating marks and marginal brown bands; ocular area yellowish, with anterior brown marks; clypeus brown; sternum yellowish, with brown marks. Legs yellowish, but dark brown on patellae and whitish on distal parts of femora and tibiae, with darker rings on subdistal parts of femora and proximal and subdistal parts of tibiae. Opisthosoma yellowish, with dorsal and lateral spots. Chelicerae (Fig. 6D) with pair of proximo-lateral apophyses, pair of distal apophyses with two teeth each and pair of frontal apophyses. Pedipalp as in Fig. 5A, B; trochanter with long (longer than wide), retrolaterally strongly bulged ventral apophysis; femur with small retrolatero-proximal apophysis and distinct ventral protuberance; tibia with prolat-ero-ventral projection; procursus simple proximally, but complex distally, with raised prolatero-subdistal membranous edge bearing distal membranous process (arrow 1 in Fig. 5C), sclerotised prolatero-subdistal apophysis (arrow 2 in Fig. 5C) and two strong dorsal spines (arrows 1 and 2 in Fig. 5D); uncus latero-medially protruding (arrow 1 in Fig. 6C), with proximal apophysis and distal scaly edge; appendix curved, with slender median branch (same length as appendix, arrow 2 in Fig. 6C); embolus weakly sclerotised, with some transparent distal projections (Fig. 6C). Retrolateral trichobothrium of tibia I at $8 \%$ proximally; legs with short vertical setae on tibiae, metatarsi and tarsi; tarsus I with 35 distinct pseudosegments.

Female (paratype, SYNU-Ar00405): Similar to male, habitus as in Fig. 6G, H. Total length 4.75 ( 4.80 with clypeus), carapace 1.35 long, 1.62 wide, opisthosoma 3.40 long, 1.70 wide; tibia I: 7.10; tibia I L/d: 51 . Eye interdistances and diameters: PME-PME 0.22 , PME 0.15 , PMEALE 0.06, AME-AME 0.05 , AME 0.11. Sternum width/ length: 1.01/0.79. Epigyne (Fig. 6A) nearly triangular, laterally and medially strongly sclerotised, with col-umn-shaped knob. Vulva (Fig. 6B) with laterally strongly curved, posteriorly sclerotised anterior arch and pair of nearly elliptic pore plates.

Variation. Tibia I in paratype male (SYNU-Ar00404): 10.44. Tibia I in another paratype female (SY-NU-Ar00406): 7.18.

Natural history. The species was found on the underside of an overhang on rocky cliffs.

Distribution. China (Shaanxi, type locality; Fig. 1).


Figure 5. Pholcus baoji sp. nov., holotype male A, B. Pedipalp (A. Prolateral view; B. Retrolateral view); C, D. Distal part of procursus (C. Prolateral view, arrow 1 indicates distal membranous process, arrow 2 indicates sclerotised prolatero-subdistal apophysis; D. Dorsal view, arrows 1, 2 indicate dorsal spines, arrow 3 indicates rectangular part of raised prolatero-subdistal membranous edge, arrow 4 indicates latero-distal part of sclerotised prolatero-subdistal apophysis). Abbreviations: $\mathrm{a}=$ appendix, $\mathrm{b}=\mathrm{bulb}, \mathrm{e}=\mathrm{embolus}$, $\mathrm{pr}=$ procursus, $\mathbf{u}=$ uncus. Scale bars: $0.20 \mathrm{~mm}(\mathbf{A}, \mathbf{B}) ; 0.10 \mathrm{~mm}(\mathbf{C}, \mathbf{D})$.


Figure 6. Pholcus baoji sp. nov., holotype male (C-F) and paratype female (A, B, G, H) A. Epigyne, ventral view; B. Vulva, dorsal view; C. Bulbal apophyses, prolateral view, arrow 1 indicates latero-median protrusion, arrow 2 indicates slender median branch; D. Chelicerae, frontal view; E-H. Habitus (E, G. Dorsal view; F. Lateral view; H. Ventral view). Abbreviations: $\mathrm{a}=$ appendix, $\mathrm{b}=$ bulb, da $=$ distal apophysis, $\mathrm{e}=$ embolus, $\mathrm{fa}=$ frontal apophysis, $\mathrm{pa}=$ proximo-lateral apophysis, $\mathrm{pp}=$ pore plate, $\mathrm{u}=$ uncus. Scale bars: $0.20 \mathrm{~mm}(\mathbf{A}-\mathbf{D}) ; 1.00 \mathrm{~mm}(\mathbf{E}-\mathbf{H})$.

## Pholcus ovatus Yao \& Li, 2012

Pholcus ovatus Yao and Li (2012: 28), figs 134A-D, 135A-E, 136A-D, 137A-D.

Material examined. $1 \delta^{\widehat{ }}$ (SYNU-Ar00120F) 1 (SY-NU-Ar00121F), China, Shaanxi, Xi'an, Zhouzhi County, Banfangzi Town (type locality), roadside of G108 ( $33^{\circ} 48.02^{\prime} \mathrm{N}, 107^{\circ} 59.08^{\prime} \mathrm{E}, 1165 \mathrm{~m}$ elev.), 31 July 2022, Z. Yao, L. Yang \& L. Zhang leg.

Diagnosis. The species resembles P. taibaiensis Wang \& Zhu, 1992 (Yao and Li 2012: 34, figs 169A-D, 170AC) with similar male chelicerae and epigyne (figs 135A, 136B, C, 137C in Yao and Li (2012)), but it can be distinguished by sclerotised prolatero-subdistal apophysis of procursus prolatero-proximally strongly widened (figs 134C, 137A in Yao and Li (2012); not widened in P. taibaiensis, fig. 169C in Yao and Li (2012)), by raised pro-latero-subdistal membranous edge of procursus laterally strongly curved (figs 134D, 137B in Yao and Li (2012); laterally angular in P. taibaiensis, fig. 169D in Yao and Li 2012), by appendix median branch length/appendix length ratio: 0.5 (figs 134A, 136A in Yao and Li (2012); 0.2 in P. taibaiensis, fig. 169A in Yao and Li (2012)) and by vulval anterior arch postero-medially strongly protruding (figs 135B, 137D in Yao and Li (2012); not protruding in P. taibaiensis, fig. 170B in Yao and Li (2012)).

Natural history. The species was found on the underside of an overhang on rock cliffs.

Distribution. China (Shaanxi, Fig. 1).

## Pholcus taibaiensis Wang \& Zhu, 1992

Pholcus taibaiensis Wang and Zhu (1992: 20), figs 1-6. Song, Zhu and Chen (1999: 63), fig. 25I-K. Zhang and Zhu (2009: 90), fig. 52A-I. Huber (2011b: 451), figs 2097-2099, 2124, 2125, 2178-2183, 2185 and 2199. Yao and Li (2012: 34), figs 169A-D, 170A-C.

Material examined. 3§ (SYNU-Ar00130F-Ar00132F) 3 ? (SYNU-Ar00133F-Ar00135F), China, Shaanxi, Baoji, Mei County, Yingtou Town, near Haopingsi Temple (type locality) $\left(34^{\circ} 5.32^{\prime} \mathrm{N}, 107^{\circ} 42.33^{\prime} \mathrm{E}, 1101 \mathrm{~m}\right.$ elev.), 30 July 2022, Z. Yao, L. Yang \& L. Zhang leg.

Diagnosis. The species resembles $P$. ovatus Yao \& Li, 2012 (Yao and Li 2012: 28, figs 134A-D, 135A-E, 136A-D, 137A-D) with similar male chelicerae and epigyne (fig. 170A in Yao and Li (2012)), but it can be distinguished by sclerotised prolatero-subdistal apophysis of procursus not widened prolatero-proximally (fig. 169C in Yao and Li (2012); strongly widened in P. ovatus, figs 134C, 137A in Yao and Li (2012)), by raised prolatero-subdistal membranous edge of procursus laterally angular (fig. 169D in Yao and Li (2012); laterally strongly curved in $P$. ovatus, figs 134D, 137B in Yao and Li (2012)), by appendix median branch length/appendix length ratio: 0.2 (fig. 169A in Yao and Li (2012); 0.5 in P. ovatus, figs 134A, 136A in Yao and Li (2012)) and by
vulval anterior arch not protruding postero-medially (fig. 170B in Yao and Li (2012); strongly protruding in P. ova$t u s$, figs 135B, 137D in Yao and $\operatorname{Li}(2012)$ ).

Natural history. The species was found on the underside of an overhang on rocky cliffs.

Distribution. China (Shaanxi, Fig. 1).

## Acknowledgements

The manuscript benefits greatly from comments by Danilo Harms, Bernhard Huber, Yanfeng Tong and an anonymous reviewer. Joseph KH Koh checked the English. This study was supported by the Science \& Technology Fundamental Resources Investigation Program of China (2023FY100200) and the National Natural Science Foundation of China (NSFC-32170461, 31872193).

## References

Blackledge TA, Scharff N, Coddington JA, Szüts T, Wenzel JW, Hayashi CY, Agnarsson I (2009) Reconstructing web evolution and spider diversification in the molecular era. Proceedings of the National Academy of Sciences of the United States of America 106(13): 5229-5234. https://doi.org/10.1073/pnas. 0901377106
Colgan DJ, McLauchlan A, Wilson GDF, Livingston SP, Edgecombe GD, Macaranas J, Cassis G, Gray MR (1998) Histone H3 and U2 snRNA DNA sequences and arthropod molecular evolution. Australian Journal of Zoology 6(5): 419-437. https://doi.org/10.1071/ZO98048
Dimitrov D, Astrin JJ, Huber BA (2013) Pholcid spider molecular systematic revisited, with new insights into the biogeography and the evolution of the group. Cladistics 29(2): 132-146. https://doi. org/10.1111/j.1096-0031.2012.00419.x
Dong T, Zheng G, Yao Z, Li S (2016) Thirteen new species of the spider genus Pholcus Walckenaer, 1805 (Araneae: Pholcidae) from China. Zootaxa 4170(1): 1-40. https://doi.org/10.11646/zootaxa.4170.1.1
Drummond AJ, Suchard MA, Xie D, Rambaut A (2012) Bayesian Phylogenetics with BEAUti and the BEAST 1.7. Molecular Biology and Evolution 29(8): 1969-1973. https://doi.org/10.1093/molbev/ mss075
Folmer O, Black M, Hoeh W, Lutz R, Vrijenhoek R (1994) DNA primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates. Molecular Marine Biology and Biotechnology 3(5): 294-299.
Hall TA (1999) BioEdit: A user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. Nucleic Acids Symposium Series 41: 95-98.
Huber BA (2011a) Phylogeny and classification of Pholcidae (Araneae): An update. The Journal of Arachnology 39(2): 211-222. https://doi. org/10.1636/CA10-57.1
Huber BA (2011b) Revision and cladistic analysis of Pholcus and closely related taxa (Araneae, Pholcidae). Bonner Zoologische Monographien 58: 1-509.
Huber BA, Eberle J, Dimitrov D (2018) The phylogeny of pholcid spiders: A critical evaluation of relationships suggested by molecular data (Araneae, Pholcidae). ZooKeys 789: 51-101. https://doi. org/10.3897/zookeys.789.22781

Khmelik VV, Kozub D, Glazunov A (2005) Helicon Focus 3.10.3. https://www.heliconsoft.com/heliconsoft-products/helicon-focus/ [Accessed 1 November 2023]
Lu Y, Yang F, He Q (2021) Pholcus maxian sp. nov., the fifth endemic spider species of Pholcus phungiformes species-group (Araneae: Pholcidae) at the border between Jilin, China and North Korea. Biodiversity Data Journal 9: e72464 [1-7]. https://doi.org/10.3897/ BDJ.9.e72464
Lu Y, Chu C, Zhang X, Li S, Yao Z (2022a) Europe vs. China: Pholcus (Araneae, Pholcidae) from Yanshan-Taihang Mountains confirms uneven distribution of spiders in Eurasia. Zoological Research 43(4): 532-534 [\& Suppl. 1-78]. https://doi.org/10.24272/j. issn.2095-8137.2022.103
Lu Y, Yao Z, He Q (2022b) A new species of Pholcus yichengicus spe-cies-group (Araneae, Pholcidae) from Hebei Province, China. Biodiversity Data Journal 10: e81800 [1-7]. https://doi.org/10.3897/ BDJ.10.e81800
Puillandre N, Lambert A, Brouillet S, Achaz G (2012) ABGD, Automatic barcode gap discovery for primary species delimitation. Molecular Ecology 21(8): 1864-1877. https://doi.org/10.1111/j.1365294X.2011.05239.x
R Development Core Team (2023) R Foundation for Statistical Computing. http://www.R-project.org/ [Accessed on 1 November 2023]
Song D, Zhu M, Chen J (1999) The spiders of China. Hebei Science and Technology Publishing House Shijiazhuang, 640 pp.
Stamatakis A (2014) RAxML Version 8: A tool for phylogenetic analysis and post-analysis of large phylogenies. Bioinformatics (Oxford, England) 30(9): 1312-1313. https://doi.org/10.1093/bioinformatics/ btu033
Tamura K, Peterson D, Peterson N, Stecher G, Nei M, Kumar S (2011) MEGA5: Molecular evolutionary genetics analysis using maximum likelihood, evolutionary distance, and maximum parsimony methods. Molecular Biology and Evolution 28(10): 2731-2739. https:// doi.org $/ 10.1093 / \mathrm{molbev} / \mathrm{msr} 121$
Vink CJ, Thomas SM, Paquin P, Hayashi CY, Hedin M (2005) The effects of preservatives and temperatures on arachnid DNA. Invertebrate Systematics 19(2): 99-104. https://doi.org/10.1071/ IS04039

Wang X, Zhu M (1992) One new species of the genus Pholcus from China (Araneae: Pholcidae). Acta Arachnologica Sinica 1(1): 20-22.
World Spider Catalog (2023) World Spider Catalog, Version 24.5. Natural History Museum Bern. http://wsc.nmbe.ch [Accessed 12 November 2023]
Yang Z (2015) The BPP program for species tree estimation and species delimitation. Current Zoology 61(5): 854-865. https://doi. org/10.1093/czoolo/61.5.854
Yao Z, Li S (2012) New species of the spider genus Pholcus (Araneae: Pholcidae) from China. Zootaxa 3289(1): 1-271. https://doi. org/10.11646/zootaxa.3289.1.1
Yao Z, Pham DS, Li S (2015) Pholcid spiders (Araneae: Pholcidae) from northern Vietnam, with descriptions of nineteen new species. Zootaxa 3909(1): 1-82. https://doi.org/10.11646/zootaxa.3909.1.1
Yao Z, Dong T, Zheng G, Fu J, Li S (2016) High endemism at cave entrances: a case study of spiders of the genus Uthina. Scientific Reports 6: 35757 [1-9 \& Suppl. 1-52]. https://doi.org/10.1038/ srep35757
Yao Z, Wang X, Li S (2021) Tip of the iceberg: species diversity of Pholcus spiders (Araneae, Pholcidae) in the Changbai Mountains, Northeast China. Zoological Research 42(3): 267-271 [\& Suppl. 1-60]. https://doi.org/10.24272/j.issn.2095-8137.2021.037
Zhang F, Zhu M (2009) A review of the genus Pholcus (Araneae: Pholcidae) from China. Zootaxa 2037(1): 1-114. https://doi.org/10.11646/ zootaxa.2037.1.1
Zhang J, Kapli P, Pavlidis P, Stamatakis A (2013) A general species delimitation method with applications to phylogenetic placements. Bioinformatics (Oxford, England) 29(22): 2869-2876. https://doi. org/10.1093/bioinformatics/btt499
Zhao F, Jiang T, Yang L, He Q, Zheng G, Yao Z (2023a) Pholcid spiders of the Pholcus phungiformes species group (Araneae, Pholcidae) from Liaoning Province, China: An overview, with description of a new species. ZooKeys 1156: 1-14. https://doi.org/10.3897/zookeys.1156.98331
Zhao F, Yang L, Zou Q, Ali A, Li S, Yao Z (2023b) Diversity of Pholcus spiders (Araneae: Pholcidae) in China's Lüliang Mountains: an integrated morphological and molecular approach. Insects 14(4): 364 [1-34]. https://doi.org/10.3390/insects14040364

