



When light casts long shadows: The increasingly dark truth about *Pelodiscus axenaria* (Zhou, Zhang & Fang, 1991)

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Zhou *et al.* (1991) were the first to propose a scientific name for the soft-shelled turtles called 砂鳖 (shā biē = sand soft-shelled turtle) by the inhabitants of Hunan Province, China. The binomen *Trionyx axenaria* Zhou, Zhang & Fang, 1991 appears only in the English abstract of their otherwise fully Chinese-language paper, which provides few specifics allowing a distinction of their new species from the 中华鳖 (zhōng huá biē = Chinese soft-shelled turtle) other than that it reaches a smaller adult size, has a nearly round, sometimes warty or keeled but mostly smooth, heavily pigmented carapace, usually a central black spot on its yellow-white plastron, and very thin abdominal skin. In addition, the length of the snout was described as greater than the diameter of the eyes, and the ratios of eye diameter and interorbital distance to carapace length were indicated as smaller than those of the Chinese soft-shelled turtle. The entoplastron was described as relatively flat, straight, and with diverging posterior processes. The scientific species epithet is a misspelling of the Latin word *arenaria*, meaning “sandy”, and is not gender-correct with respect to the male generic name *Trionyx*. However, “*axenaria*” has been implicitly or explicitly treated as a noun in apposition (ICZN 1999: Articles 11.9.1.2, 31.2.1, and 31.2.2) by all later authors, and we do not intend to change the established usage of this name (see also Gong *et al.* 2022).

The original description makes no mention of any type material or any place of deposition for the then apparently live sand soft-shelled turtles. In the tables, numbers ranging from one and 11 (Zhou *et al.* 1991; table 2) to 73 turtles (Zhou *et al.* 1991: table 1) are listed, none of which is individually recognizable. The only figures accompanying the article show the shā biē in dorsal and ventral views in rather blurred and rasterized images (Fig. 1) but, being of live turtles in non-identical positions, it is impossible to tell whether the two photos actually depict the same or two different individuals. Exact locality data are not supplied, the range of shā biē is vaguely defined to encompass the counties of Taoyuan, Pingjiang, Rucheng, Lingling, and Shaoyang in Hunan Province.

Subsequent molecular genetic investigations by Chen *et al.* (2005, 2006), Fritz *et al.* (2010), Yang *et al.* (2011), Dong *et al.* (2016), and Gong *et al.* (2018) have confirmed the distinctiveness of the sand soft-shelled turtle, and it has been treated as a valid species of the genus *Pelodiscus* in all major turtle taxonomy checklists published after 2007 (Fritz & Havaš 2007; TTWG 2007, 2009, 2010, 2011, 2012, 2014, 2017, 2021; Rhodin *et al.* 2008).

Two studies employing nuclear and mitochondrial DNA sequences also revealed *P. axenaria* to be a member of a species complex comprising three morphologically challenging taxa (Gong *et al.* 2018, 2021). One of these was described as *Pelodiscus huangshanensis* Gong, Peng, Huang, Lin, Huang, Xu, Yang & Nie, 2021, whereas another clade (lineage I) discovered by Gong *et al.* (2018) was formally named *Pelodiscus shipian* Gong, Fritz, Vamberger, Gao & Farkas, 2022 (Gong *et al.* 2022). For the latter species, Hou *et al.* (2022), relying heavily on previous findings (Gong *et al.* 2018), introduced the name *Pelodiscus jiangxiensis* Hou, Lin, Li, Jiang & Shi, 2022 in a preprint. A planned paper in the International Journal of Zoology and Animal Biology was retracted before official publication, even though a PDF was circulated for a while on the internet. However, neither the preprint nor the PDF constitute published work according to the requirements of the International Code of Zoological Nomenclature (ICZN 1999, 2012: Articles 8.5.3, 9.9) because the name was not registered in ZooBank. Thus, *P. jiangxiensis* is an unavailable name in the sense of the Code, a point missed by Liu *et al.* (2024).

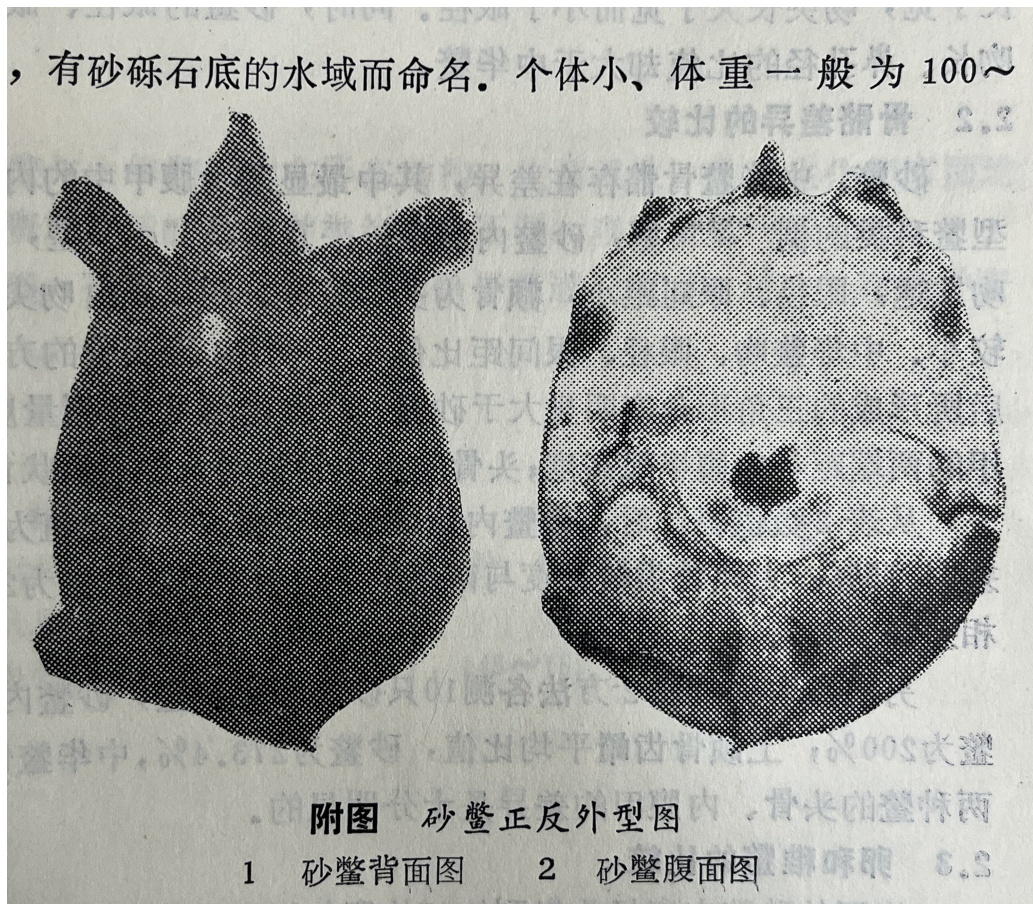


FIGURE 1. Illustrations from Zhou *et al.* (1991) showing one or two syntypes of *Trionyx axenaria*. Note the different outline of the “heart-shaped” central blotch on the plastron, the absence of dark markings on the left hyo–hypoplastra and suffusions on the front edge of the plastron, as well as the presence of black bands on the thighs on either side of the tail as opposed to the lectotype designated by Liu *et al.* (2024) claimed to be the same individual (Fig. 4, top). The dorsal-view image is totally unsuited for species (and specimen) identification.

To clarify the taxonomy, Gong *et al.* (2022) designated a neotype for *P. axenaria*. However, Liu *et al.* (2024) reported to have located ten of the 73 original syntypes of *T. axenaria* in the collection of the Animal Museum of Life Sciences College of Hunan Normal University, Changsha (HNNU), and used these to invalidate the neotype designation for *T. axenaria* (Gong *et al.* 2022). Surprisingly, Liu *et al.* (2024) used the previous redescription of *T. axenaria* and its neotype (Gong *et al.* 2022) for erecting yet another species. Liu *et al.* (2024) designate the neotype of *T. axenaria* as the name-bearing type of their new species *Pelodiscus huike* Liu, Fu, Ma, Zhou, Blanck, Zhou, Xiao & Mo, 2024, without having examined the specimen. In doing so, Liu *et al.* (2024: fig. 4) republished the respective pictures of the neotype of *T. axenaria* from Gong *et al.* (2022: fig. 1; Jinan University, Guangzhou, JNU 20210001), without crediting them to the original source or notifying the photographer (Shiping Gong, SG). At the same time, Liu *et al.* (2024) relegated *P. shipian* into the synonymy of *P. axenaria*. In the following, we reexamine their conclusions in the light of additional evidence.

The syntypes of *Trionyx axenaria*. Liu *et al.* (2024) state that Gong-Jian Zhou (GJZ; a member of their author team) deposited syntypes of *T. axenaria* at HNNU upon their death. However, despite repeated attempts, we were unable to locate any of these specimens earlier (Xiang Xu and Shuqiang Li, *in litt.* to SG, 14 February 2020; see also Gong *et al.* 2022). Ting Zhou (another coauthor of Jia-Yu Liu) and GJZ had lost track of the specimens in 2012 (pers. comm. to SG, 23 March 2022). Nevertheless, Zhou & Li (2013: p. 156) provided photos of a preserved sand soft-shelled turtle bearing a specimen tag with the number HNNU-1991001ZGJ, which they erroneously labeled as holotype (Fig. 2). However, in the absence of a holotype designation by Zhou *et al.* (1991), this specimen cannot be regarded as such (ICZN 1999: Article 72). The whereabouts of this individual were not indicated and still remain unknown to the scientific community. Both the TTWG (2021) turtle checklist and Iverson (2022) list the “holotype” as “not located”.



FIGURE 2. Photos from Zhou & Li (2013) of a preserved soft-shelled turtle erroneously labeled as the holotype of *Pelodiscus axenaria*. The specimen bears the same registration number (i.e., HNNU-1991001ZGJ) as another specimen designated as a paralectotype by Liu *et al.* (2024). The whereabouts of the specimen figured in Zhou & Li (2013) remain unknown.

Seven days after our neotype designation for *P. axenaria* was published (8 April 2022), GJZ notified us (pers. comm. to SG, 15 April 2022) that, with the help of Xiao-Yang Mo (XYM; yet another of Liu’s coauthors), nine syntypes were suddenly rediscovered on 13 April 2022 in the late Youhui Shen’s office at HNNU (Shen died on 9 December 2018). We also received two photographs of these freshly found specimens displayed on a tray, of which one (lower row, second from right; Fig. 3) was indicated to represent the “holotype”. However, this individual is clearly not the same as the one figured as “holotype” by Zhou & Li (2013), nor is it identical with the specimen later designated as the lectotype by Liu *et al.* (2024). The proposed lectotype is also not consistent with the live turtle pictured in Zhou *et al.* (1991: fig. 1, our Fig. 3), although Liu *et al.* (2024) declared them to be indistinguishable (see also below). Unfortunately, our subsequent request to collaborate and to examine the purported types were declined by XYM.

Liu *et al.* (2024) mention ten (abstract, p. 337: four males and six females; p. 340: six males and four females) instead of nine adult specimens in their paper, all preserved in 10% buffered formalin and deposited at HNNU, bearing the registration numbers HNNU-1991001ZGJ to HNNU-1991010ZGJ. Number HNNU-1991001ZGJ is assigned to a paralectotype (Liu *et al.* 2024: figs. 2a, b; reproduced here as our Fig. 4), which is obviously not the same individual depicted by Zhou & Li (2013) as the “holotype” of *P. axenaria*, despite having the same registration number. Although both turtles are males, the two have been fixed in highly dissimilar postures and exhibit different neck patterns and ventral markings. Furthermore, the plastron is apparently detached in the “holotype” (Fig. 2), while it is intact in the paralectotype (Fig. 4, bottom), and the specimen label is attached to the right hindleg of the former, but to the left foreleg of the latter.

In spite of Liu *et al.* (2024) claiming otherwise, the lectotype HNNU-1991002ZGJ does not match up to the live turtle shown in Zhou *et al.* (1991) (Figs. 1 and 4). The “heart-shaped” central blotch on the plastron has a different outline, and the dark markings on both hyo–hypoplastra as well as the suffusions on the front edge of the plastron of the lectotype are lacking in Zhou *et al.*’s (1991) ventral-view figure, whereas the black bands on the thighs of the latter are absent in the former specimen. This means that the specimens presented by Liu *et al.* (2024) and the original type material of *T. axenaria* cannot be equated.



FIGURE 3. The purported type series of *Trionyx axenaria* shortly after its “rediscovery”. Photographer unknown. The lectotype and a paralectotype designated by and figured in Liu *et al.* (2024) are seen in the upper row, first and second from right, respectively. The specimens are bearing no or an obviously different specimen tag. Also note that there are only nine individuals, not ten, in this photo.

As revealed by Liu *et al.*'s (2024) figure 2 (our Fig. 4), both the lectotype and the paralectotype now bear linen labels not associated with either of them at the time of their “rediscovery” (they are seen in the upper row, first and second from right, respectively, on the tray; Fig. 3), whereas the original paper tag attached to the left hindleg of the specimen designated as paralectotype has since been removed. This suggests that the specimens have been relabeled, compromising their identity and identification with authentic type material.

Genetics. Liu *et al.* (2024) reported to have sequenced DNA extracted from the intestinal tissue of three of their putative type specimens (HNNU-1991001ZGJ, HNNU-1991002ZGJ, HNNU-1991003ZGJ), to prove that the interpretation of *P. axenaria* in Gong *et al.* (2022) was based on a taxonomic misconception. However, there are no cuts facilitating tissue removal in their photos of the specimens (Fig. 4), suggesting either that the turtles were later sampled or that other turtles were used.

According to Liu *et al.* (2024), the putative type specimens of *T. axenaria* were preserved in 10% buffered formalin for more than 30 years. It is well known that sequencing DNA from formalin-fixed, in particular long-term preserved, specimens is challenging. DNA extracted from formalin-preserved samples is typically low-yield, highly fragmented, and heavily cross-linked. PCR amplification and sequencing of formalin-degraded DNA templates is generally restricted to few and short DNA fragments of less than 200 bp length, and the usage of sophisticated approaches for historical DNA, involving DNA extraction in clean room facilities, special protocols, and Illumina sequencing is recommended (e.g., Kehlmaier *et al.* 2020; Hahn *et al.* 2022, 2024). In contrast, Liu *et al.* (2024) had reportedly obtained 988-bp-long

mtDNA sequences using standard DNA extraction and Sanger sequencing approaches (see GenBank accession numbers OR759009–OR759011). The presented results are surprising and suggest that the specimens were either not formalin-preserved for more than 30 years (i.e., not representing syntypes of *T. axenaria*) or that the sequences originate from fresh material (contamination?) and have been erroneously assigned.

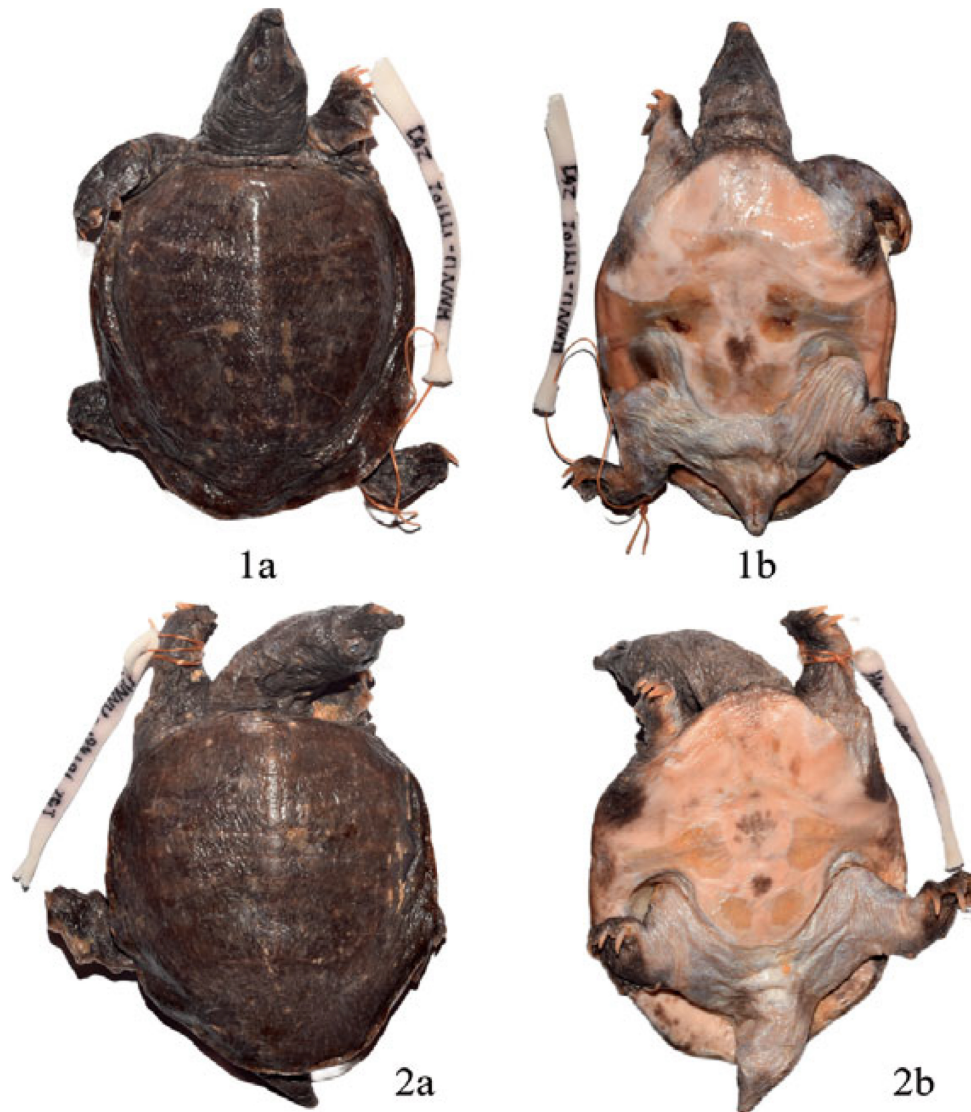


Fig. 2. Dorsal and ventral aspects of the lectotype and paralectotype of *Pelodiscus axenaria*: 1a, dorsal view of the lectotype (HNNU-1991002ZGJ, adult female); 1b, ventral view of the lectotype (HNNU-1991002ZGJ, adult female); 2a, dorsal view of the paralectotype (HNNU-1991001ZGJ, adult male); 2b, ventral view of the paralectotype (HNNU-1991001ZGJ, adult male).

FIGURE 4. The lectotype of *Pelodiscus axenaria* and a paralectotype designated by and figured in Liu *et al.* (2024: fig. 2); reproduced from that study, including the original caption. Note that both specimens bear different labels compared to our Figure 3. The specimens are fully intact without scars or cuts facilitating intestinal tissue removal for genetic studies.

Conclusions. There is no evidence that the specimens presented by Liu *et al.* (2024) as type material of *T. axenaria* are part of the original type series of that species. The obvious fact that specimen labels have been secondarily attached and changed raises serious doubts about their authenticity and identity. We therefore refute that any of these specimens represent syntypes of *T. axenaria*. The purported mtDNA sequences cannot be firmly associated with the type material either. In

the light of the evidence presented, the neotype designation for *T. axenaria* by Gong *et al.* (2022) stands, and *Pelodiscus huike* Liu, Fu, Ma, Zhou, Blanck, Zhou, Xiao & Mo, 2024, based on the same name-bearing type (JNU 20210001), is an objective junior synonym of *Trionyx axenaria* Zhou, Zhang & Fang, 1991 (ICZN 1999: Article 61.3.4).

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