Three Cambrian fossils assembled into an extinct body plan of cnidarian affinity

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The early Cambrian problematica Xianguangia sinica, Chengjiangopenna wangii, and Galeaplumosus abilus from the Chengjiang biota (Yunnan, China) have caused much controversy in the past and their phylogenetic placements remain unresolved. Here we show, based on exceptionally preserved material (85 new specimens plus type material), that specimens previously assigned to these three species are in fact parts of the same organism and represent an offshoot of the cnidarian stem lineage that diverged early during the Cambrian radiation of animal body plans.

Significance

New material from the Chengjiang fossil Lagerstätte clarifies the identity of three early Cambrian problematica. The presumed earliest hemichordate Galeaplumosus abilus and the putative ancient sea pen Chengjiangopenna wangii are in fact fragments of Xianguangia sinica. Here we demonstrate that X. sinica possessed a polypoid body, a blind gastric cavity partitioned by septum-like structures, a holdfast that contained an additional cavity functioning as a hydroskeleton, a basal pit used for anchorage, and a radial whorl of feather-like tentacles for ciliary suspension feeding. Phylogenetic analyses based on the new findings suggest that X. sinica represents an extinct body plan most closely allied to cnidarians and thus sheds light on their early evolution.

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densely distributed along both sides of each pinnule. Column with a central cavity partitioned by radiating septum-like structures. Column surface rough, with parallel longitudinal ridges. A circumferential constriction separates the column from the holdfast. Holdfast with a separate cavity. Underside of holdfast smooth and convex with a prominent, circular pit.

Description. A crown of feather-like tentacles projects from the marginal edge atop the column (Figs. 1A and 2A and SI Appendix, Figs. S1 and S2A) in a radial pattern (SI Appendix, Figs. S2F and S4A). The tentacles are straight proximally, but variably flexed distally (Fig. 1A and SI Appendix, Figs. S1D and S2A), ~18 in number (SI Appendix, Fig. S4A and B). Approximately 80 slender pinnules biserially branch off from the rachis of each tentacle at a variable angle of 30°–120° in an alternating pattern (Figs. 1B and 2A and C and SI Appendix, Figs. S1D and S3A and B). The pinnules appear flexible in shape (Figs. 1B and C and 2B and SI Appendix, Fig. S3). In exceptionally well-preserved specimens, long filaments are observed densely arrayed along both sides of each pinnule (Figs. 1C and 2D and SI Appendix, Fig. S3C–F). The filaments project nearly at the right angle from the surface of the pinnule, thus forming a comb-like arrangement (Figs. 1C and 2D and SI Appendix, Fig. S3C–F). In the middle portion of the tentacles of our largest specimens (e.g., Fig. 2C), the rachis is ~0.2 mm wide (SI Appendix, Table S2). The pinnules are spaced ~0.4 mm apart from each other; they reach a maximum length of 4.3 mm in the middle of the tentacle, and gradually decrease in length toward the distal tip (Figs. 1B and 2A).

The filaments range from 540 to 630 μm in length and are spaced ~50 μm apart. The ring of tentacles surrounds a disk that bears a central oral opening leading into a central cavity filled with sediment (SI Appendix, Fig. S4A). Radiating bands of dark remains, which alternate with tentacle bases, extend from the wall of the column into the cavity (SI Appendix, Fig. S4B). The column is a robust cylindrical structure characterized by external longitudinal ridges and an internal central cavity preserved with dark remains of organic material (Fig. 3A and B and SI Appendix, Fig. S4A). A circumferential constriction delineates the column from the holdfast (Figs. 1A and 3A and B and SI Appendix, Fig. S2A, D, and E). In the lateral view, the holdfast tapers slightly toward the base (Fig. 3A and B) and is variable in length (compare Fig. 3A and B to SI Appendix, Fig. S2D and Table S2). A voluminous cavity occurs within the holdfast, which, in contrast to the central cavity of the column, does not show any dark remains (Fig. 3B). The holdfast has a smooth, convex, disk-shaped underside referred to as the basal disk (Fig. 3C and D and SI Appendix, Figs. S1A and S2F and G). The basal disk bears a prominent, bowl-shaped pit, which in most specimens occurs in an eccentric position and is filled with sediment (Fig. 3A, C, and D and SI Appendix, Fig. S2E–G). The pit varies in size, with a diameter accounting for ~40–60% of the basal disk (SI Appendix, Table S2). There is no evidence of any opening either on the basal disk or within the pit (Fig. 3D and SI Appendix, Fig. S4G). In some specimens, the holdfast is bent at the constriction, with the basal disk parallel to the bedding surface of the matrix rock (SI Appendix, Figs. S1A–C and S2G).
an associated piece of the column. The following lines of evidence support the interpretation of *G. abilus* as a fragment of *X. sinica*: (i) The tentacles of *X. sinica* and the purported “arms” of *G. abilus* show the same proportions, shape, and orientation; (ii) the number of pinnules [interpreted as “tentacles” in ref 18.] is consistent (~80 on each tentacle) in *X. sinica* and *G. abilus*, whereas it is lower in extant pterobranchs [up to 40 on each arm (33)]; (iii) the width and spacing of the tentacles of *G. abilus* correspond to the pinnules of *X. sinica*; and (iv) the size of *G. abilus* (~40 mm long) matches the tentacles of *X. sinica* rather than the pterobranch zooids, which typically do not exceed 1–2 mm (*Rhabdopleura* sp.) or 5 mm in length (*Cephalodiscus* sp.) (33). As has been pointed out previously (32), the putative evidence of tubes (coenecia) with fusellli—characteristic growth bands in tubes of pterobranchs and graptolites (32, 33)—is unconvincing in *G. abilus*. Given that the tubes are recalcitrant organic secretions of zooids that are expected to be better preserved than the zooids themselves, their absence in *G. abilus* is puzzling. It is also curious that the “feathered zooid arms” attach directly to the “cone-shaped zooid tube,” without any sign of the

Discussion

*C. wangii* and *G. abilus* Are Junior Synonyms of *X. sinica*. Our results revealed that specimens, which were previously assigned to three different taxa including *X. sinica*, *C. wangii*, and *G. abilus*, are in fact parts of the same species. Reexamination of specimen ELJ-Seapen-05–001 (*SI Appendix*, Fig. S4 C and D), originally named *C. wangii* and assigned to crown-group octocorals (17, 25), suggests that it is in fact an isolated tentacle of *X. sinica*. First, this specimen corresponds to the individual tentacles of *X. sinica* in overall shape and morphological details, both having a gradually tapering rachis flanked by alternating pinnules with filaments. Second, they show a similar size with the same proportions (*SI Appendix*, Table S2). Morphometric data revealed that the length of tentacles and the length and spacing of pinnules of *X. sinica* are proportional to those of *C. wangii* (*SI Appendix*, Table S2). The dark spots occurring along the pinnules in ELJ-Seapen-05–001 (*SI Appendix*, Fig. S4D) were originally interpreted as autozooids diagnostic of octocorals (17, 25). However, these spots are most likely preservation artifacts because they are irregularly shaped, unevenly spaced, and variable in size (40–70 μm across). Moreover, the spots are one to two orders of magnitude smaller than typical autozooids of modern octocorals (31). We therefore suggest that *C. wangii* is not an ancient sea pen but a junior synonym of *X. sinica*.

Similarly, we propose that the type and only specimen of the putative Cambrian hemichordate *G. abilus* (18) (*SI Appendix*, Fig. S4E), recovered from the same fossil Lagerstätte, is most likely a fragment of *X. sinica*. This fragment, originally interpreted as a pterobranch zooid (18), consists of a complete tentacle with pinnules, a proximal portion of a second tentacle, and

**Fig. 2.** Tentacle architecture of *X. sinica* (ELEL-SJ000827B, *SI Appendix*, Fig. S1). (A) Upper part showing numerous pinnate tentacles (arrowed). (B–D) Close-up of focus areas in A showing sinusoid pinnules (B), attachment sites (arrowheads) of pinnules branching from the rachis (C), and closely spaced cilia (arrows) fringing the pinnules (D), respectively. [Scale bars: 10 mm (A), 1 mm (B–D).]
reveal previously unknown features of X. *sinica*. This suggestion leaves the middle Cambrian tubicolous enteropneusts from the Burgess Shale (11, 34) and the alga-like pterobranchs (graptolites) from British Columbia and Utah (35, 36) among the oldest fossil record of hemichordates, and the rhopialium-bearing medusa (3) as the most convincing, fossil crown-group cnidarian (Medusozoa: Scyphozoa) from the Chengjiang biota.

**X. sinica Was a Suspension Feeder with an Extinct Body Plan.** In addition to a polypoid body with a holdfast and numerous tentacles demonstrated previously (e.g., refs. 16, 23, 26, and 27), our study revealed previously unknown features of *X. sinica* (Fig. 4A), not least, a feather-like organization of tentacles with alternating pinnules that bear dense, biserial filaments. These filaments are interpreted here as cilia involved in suspension feeding. Fossilized cilia of similar size were reported from the tentacles of the brachiopod *Heliomedusa orienta* from the Chengjiang biota (37). Apart from brachiopods, dense arrays of cilia occur in suspension feeding apparatuses of various other aquatic invertebrates, including phoronids, entoprocts, bryozoans, polychaetes (sabellids and serpulids), pterobranchs, and rotifers (e.g., *Stephanoceros fimbriatus*) (38, 39). Compared with typical cilia of extant metazoans [axoneme diameter ~0.25 μm (40)], the cilia of *X. sinica* appear rather thick (19–23 μm; SI Appendix, Table S2). This might be a result of secondary thickening during preservation or diagenesis. Alternatively, these filamentous structures might have been modified for filter feeding, with numerous long cilia bound together in a functional unit reminiscent of macrocilia of living ctenophores, measuring 15 μm in diameter and ~100 μm in length (41). The alternate branching of pinnules in *X. sinica* (Fig. 4A) might have facilitated the uptake of food by preventing direct confluence of transported particles from opposite pinnules. A similar staggered arrangement of branches occurs in the tentacles of some living suspension feeders, such as sabellid polychaetes (42) and feather stars (43). Thus, the distinctive architecture of tentacles in *X. sinica* strongly implicate a suspension-feeding behavior using a ciliary system.

We further identified a spacious cavity within the holdfast, which most likely did not have a connection to the central cavity in the column of *X. sinica* because in contrast to the latter, it does not show dark organic remains (Fig. 3B). Reexamination of the specimen RCCBYU 10216 (23, 26) revealed that *X. sinica* most likely possessed septum-like structures within the central cavity (SI Appendix, Fig. S4A), corresponding in position to indentations between longitudinal ridges of the column (Fig. 3A and B and SI Appendix). Like in anthozoan polyps, the septum-like structures might have served for stabilizing the column and increasing the surface area of the central cavity, which might have been part of the gastrovascular system.

In all specimens of *X. sinica* with a preserved holdfast, the basal disk appears as a convex structure, which bears a circular pit filled with sediment. This anatomy suggests that the animal was most likely able to anchor itself in the soft sediment of the Cambrian seafloor using its holdfast. The holdfast might have acted as a burrowing physa, embedded in the sediment in living condition. The holdfast cavity might have served as a hydrostatic skeleton and antagonist to the body wall musculature, whereas the basal pit might have been used for engulfing mud or sand—an anchoring strategy possibly also used by some Ediacaran (635–542 Mya) organisms (44).

In summary, our data revealed a unique combination of characters for *X. sinica*, including (i) a polypoid body bearing feather-like tentacles with densely ciliated, alternating pinnules used for suspension feeding; (ii) a blind gastrovascular cavity.

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**Fig. 4.** Reconstruction and phylogenetic position of *X. sinica*. (A) Three-dimensional model generated with 3ds Max. (B) Summary of metazoan relationships inferred from Bayesian analyses based on 111 characters and 37 taxa under Mkvi + Γ model (see SI Appendix, Figs. S5–S7 and Text S2 and S3 for details). Numbers at nodes indicate posterior probabilities. *X. sinica* is resolved as a stem-group cnidarian. Eumetazoa, Neuralia, Bilateria, Nephrozoa, Protostomia, and Deuterostomia are monophyletic, whereas the monophyly of Spiralia is unresolved. Animal silhouettes are by courtesy of PhyloPic (www.phylopic.org).

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**References:**

1. Ou et al. (2021) *PNAS* 118(41), 12808–12813.
partitioned by septum-like structures into confluent compartments; (iii) a holdfast harboring a separate cavity that might have served as a hydroskeleton; and (iv) a basal disk with a circular pit probably used for anchoring the animal in the soft substrate. Furthermore, X. sinica was most likely a radially symmetric animal, as inferred from the radial arrangement of tentacles and septum-like structures and the absence of any features that would indicate bilateral symmetry. This combination of features reveals an extinct body plan that is unknown from any other fossil or living animal group but might have emerged in the Cambrian “explosion” of evolutionary innovations, along with some other unusual body plans that have since become extinct (4, 10, 45–47).

**X. sinica Represents a Stem-Group Cnidarian.** X. sinica has been alternatively interpreted as a crown-group cnidarian (16, 19–22), a ctenophore (23, 26), a lophophorate (27), or a survivor of vendobions (28). We have shown here that X. sinica had a relatively simple anatomy with a radial symmetry, a polypoid body, and a blind gut containing septum-like structures. This simple anatomy suggests that the animal was most likely composed of two germ layers corresponding to the organizational level of cnidarians and ctenophores. The overall external and internal anatomy also corresponds well to coelenterates rather than to the more complexly organized lophophorates with a U-shaped through-gut (48) or to Ctenophora was based on the observation of total-group Cnidaria. This finding also implies that the radial symmetry displayed by X. sinica might be an ancestral trait of Cnidaria, which persisted in hydrozoans (60, 61) but was modified to bilateral or biradial symmetry in anthozoans (62, 63) and tetraradial symmetry in most medusozoans (52, 64). The evolution of the alternating, densely ciliated pinnules (“filtering combs”) on the tentacles adapted for suspension feeding can be interpreted by two equally parsimonious scenarios. In the first scenario, the ciliated pinnules evolved before the divergence of X. sinica from the cnidian stem group. This scenario would suggest that suspension feeding is an ancestral feature (plesiomorphy) of total-group cnidarians and that the pinnules were lost in crown cnidarians, which instead coopted the raptorial feeding strategy using unbranched, cnida-laden tentacles. In the second scenario, the last common ancestor of X. sinica and crown cnidarians was a predator armed with cnidocytes, suggesting that suspension feeding is a derived feature (apomorphy) of X. sinica. Whichever of the two scenarios is favored, the revealed body plan embodied by X. sinica suggests that the feeding behavior of stem-group cnidarians may have been strikingly different from that of their crown-group descendants. The distinctive suite of characters revealed in X. sinica significantly augments the morphological disparity of total-group Cnidaria.

**Materials and Methods**

All fossil specimens (n = 88) of X. sinica examined in this study were collected from the Cambrian Chengjiang Lagerstätte and deposited in four institutes in Beijing, Xi’an, Nanjing, and Kunming (SI Appendix, Table S1 and Text S1). Mechanical preparation of fossils was performed using a Zeiss Stemi 2000 stereomicroscope under various lighting conditions. Specimens were photographed using a Canon EOS 5D Mark II optical camera under sunlight and a Zeiss Stemi 2000-C microscope equipped with a digital camera under LED light. Backscattered electron (BSE) imaging and energy-dispersive X-ray spectroscopy (EDS) analysis of uncoated specimens were conducted on an FEI Quanta 650 FEG field emission scanning electron microscope in low-vacuum mode. Morphological reconstructions were conducted using Carls Zeiss AxioVision 4.1. Three-dimensional reconstruction of X. sinica was performed using 3ds Max 9.0. Phylogenetic analyses were conducted using MrBayes 3.2, TNT 1.1, and PAUP* 4.0. Application of phylogenetic terminology, including stem, crown, and total group, follows Xiao et al. (65).