

# Evaluating scenarios for the evolutionary assembly of the brachiopod body plan

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**SUMMARY** The fossil faunas of the Cambrian provide the only direct insight into the assembly of animal body plans. However, for many animal groups, their early fossil record is linked to disarticulated remains, interpretation of which is problematic since they possess few characters from which their affinity to phyla can be established and, indeed, few characters at all. One such group is the tommotiids, which has been interpreted, on the basis of skeletal anatomy, as a paraphyletic assemblage uniting brachiopods and phoronids, through the acquisition and subsequent modification, or loss, of an imbricated set of dorsal phosphatic sclerites. Here we

present a reexamination of the fossil evidence uniting the tommotiids and brachiopods, supplemented with new anatomical data from synchrotron radiation X-ray tomographic microscopy of key tommotiid taxa. The characters used to support the complex hypothesis of character evolution in the brachiopod stem lineage relies on scleritome reconstructions and inferred mode of life which themselves rely on brachiopods being chosen as the interpretative model. We advocate a more conservative interpretation of the affinity of these fossils, based a priori on their intrinsic properties, rather than the modern analogue in whose light they have been interpreted.

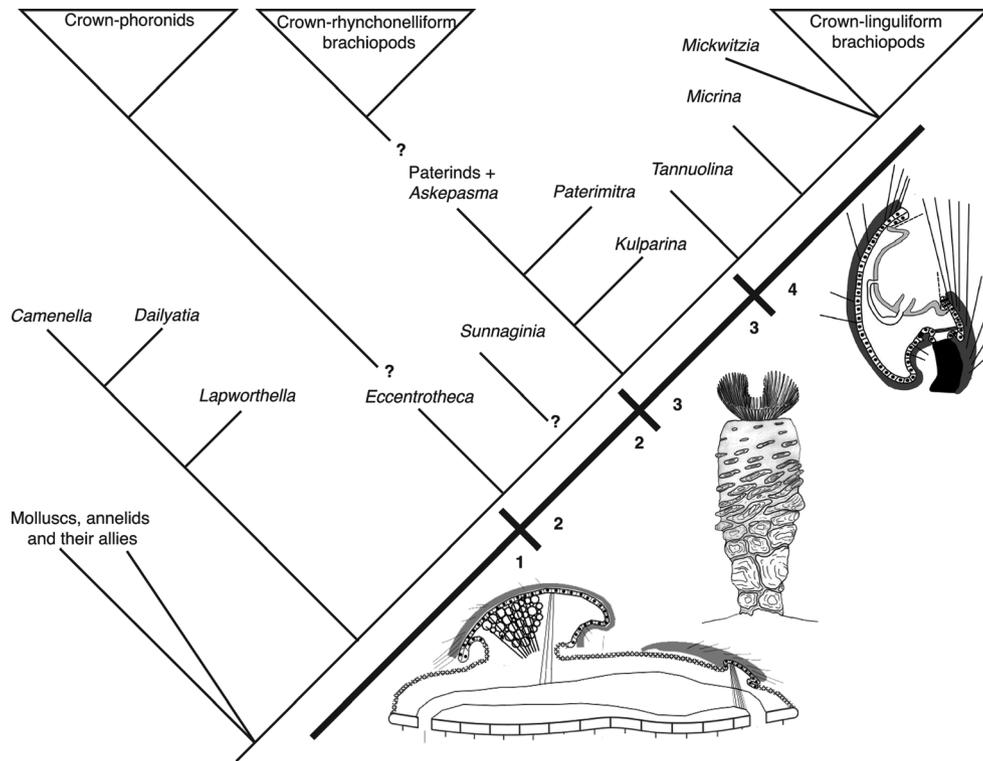
## INTRODUCTION

The origin of distinct animal body plans is at the heart of one of the most compelling enigmas in evolutionary paleobiology, the Cambrian Explosion. Comparative developmental biology can derive hypotheses for body plan evolution, but the fossil record is required to test these hypotheses and provide a temporal context. To date, the bulk of fossil evidence for the assembly of animal body plans has come from Cambrian *Konservat-Lagerstätten* which are restricted in their taxonomic, geographic, and temporal diversity. These deposits are preceded by the small shelly faunas that have played a minor role in the study of animal body plan evolution until comparatively recently, where they have been exploited to elucidate the evolutionary assembly of the brachiopod body plan in a radical new hypothesis.

The common ancestor of brachiopods and their immediate sister group, the phoronids, has been dated to the latest Ediacaran, and the brachiopod crown group to no later than the Middle Cambrian, through molecular divergence time estimates (Sperling et al. 2011). The skeletal remains in the small shelly faunas, therefore, provide the ideal dataset in which to search for insights into the assembly of the brachiopod body

plan. The discovery of exceptionally preserved fossils such as *Halkieria* (Conway Morris and Peel 1990, 1995) led to a hypothesis for the derivation of the brachiopod body plan from an armored slug-like ancestor, through the shortening and folding of the body axis (Holmer et al. 2002). *Halkieria* has since been recognized as a mollusc (Vinther & Nielsen 2005). Furthermore, recent examination of a group of early skeletal fossils, the tommotiids, has overturned this hypothesis, and a detailed sequence of character acquisition has been reconstructed through an array of stem-brachiopod lineages (Skovsted et al. 2008, 2009a, 2011; Balthasar et al. 2009; Holmer et al. 2011; Altenburger et al. 2013).

The emerging consensus of the role of the tommotiids in the assembly of the brachiopod body plan can be divided broadly into four key grades of organization (Fig. 1), each of which is represented by tommotiid taxa with progressively closer sister-relationships to the brachiopod crown. The plesiomorphic condition is represented by “camenellans” (*Camenella*, *Dailyatia*, *Lapworthella*, and their allies), possessing phosphatic sclerites with regular co-marginal ribs and retaining a vagile life-habit and a *Halkieria*-like body plan (Skovsted et al. 2009a). The key difference between this hypothesis and the halkieriid



**Fig. 1.** Hypothesis of the phylogenetic relationships between the tommotiids and crown-brachiopods, based on Skovsted et al. (2009a, 2011) and modified by Holmer et al. (2011). Phylogenetic positions for *Sunnaginina* based on Murdock et al. (2012). Four grades of organization represented by successively crown-ward placed tommotiid taxa: (1) unfused sclerites, vermiform; (2) fused sclerites, tubular body plan; (3), specialized sclerites, closed filtration chamber; (4), bimembrate scleritome, unmineralized attachment organism. Vermiform, tubular, and bimembrate body plan reconstructions shown, redrawn from Williams and Holmer (2002), Skovsted et al. (2011), and Holmer et al. (2008b), respectively.

model (Conway Morris and Peel 1995; Holmer et al. 2002) is the evolution of a tubular body plan and sessile life habit prior to the acquisition of many brachiopod apomorphies, whereas the halkieriid model suggests that the brachiopod body plan derived directly from the folding of a vagile slug-like ancestor. *Eccentrotheca* represents the first intermediate with a series of fused sclerites forming an irregular tube around an attachment organ (Skovsted et al. 2008, 2011), supporting a sister group relationship between phoronids and brachiopods. In this phylogenetic framework, phoronids are interpreted to have lost phosphatic sclerites but retained a tubular body plan (Skovsted et al. 2008, 2011).

The transition from an *Eccentrotheca*-like total-group ancestor to the crown-brachiopod condition is perceived to have been achieved through two further grades of organization. Firstly, the specialization of sclerites to house not just the attachment organ but the rest of the body cavity (Holmer et al. 2008b) is manifest as the large symmetrical “S” sclerites of *Paterimitra* fused to smaller, asymmetrical, *Eccentrotheca*-like “L” sclerites (Skovsted et al. 2009b; Larsson et al. 2013). Similarities between the microstructure of *Eccentrotheca*, *Paterimitra*, and the paterinid brachiopod *Askepasma* (Balthasar

et al. 2009) have been marshalled in support of a brachiopod affinity for the tommotiids (Larsson et al. 2013), furthermore *Paterimitra* has been interpreted as a stem-rhynchonelliform brachiopod (and, therefore, a crown-brachiopod; Holmer et al. 2011). Finally, the most derived clade of tommotiids, the tannuolinids *Tannuolina* and *Micrina*, have been interpreted to exhibit a number of brachiopod and, indeed, linguliform brachiopod apomorphies; both taxa are interpreted to have lost mineralized sclerites around the attachment organ and have sclerites punctured by setal tubes. In addition, *Micrina* is reconstructed with a bimembrate scleritome housing a closed filtration chamber (Holmer et al. 2008b).

Perhaps the most valuable aspect of this phylogenetic hypothesis is its predictive power, both in terms of presenting search criteria for membership to the total-group Brachiopoda and in the expected sequence of first appearances of the defining characteristics of the group. We set out to test the distribution of characters implied in the taxa within this phylogenetic scheme, and use the predictive nature of the hypothesis to constrain the affinity of other tommotiid taxa not explicitly resolved by these authors. Our data on the internal architecture of tommotiid sclerites focuses on the nature of growth lamellae and shell-

penetrating pores. These observations do not agree with the predictions of existing hypotheses, and raise questions about the evidence supporting tommotiids as putative stem-brachiopods.

## MATERIALS AND METHODS

The material on which this study is based came from reference collections from the Swedish Museum of Natural History, Stockholm, Sweden, with additional new material of *Sunnaginia* recovered from ex situ blocks of Comley Limestone from the Lower Cambrian of Comley, Shropshire, UK. Approximately 16 kg of rock was digested using 7% buffered acetic acid following Jeppsson & Anehus (1995). The acid-insoluble residue was size-sorted in water using 63  $\mu\text{m}$  and 1 mm sieves, and the fraction passing the 1 mm and retained in the 63  $\mu\text{m}$  sieve was density-separated using bromoform (specific gravity of 2.889) to concentrate the phosphatic material. Fossils were recovered by manual picking of the resulting heavy fraction under a binocular microscope. Specimens were mounted on 3 mm brass stubs using clear nail varnish. Their internal 3D structure was characterized using synchrotron radiation X-ray tomographic microscopy (SRXTM) (Donoghue et al. 2006); measurements were taken using 10 $\times$  and 20 $\times$  objective lenses, exposure times of 55–800 msec, energy of 12–24 keV. For each dataset, 1501 projections over 180 $^\circ$  were acquired from which tomographic reconstruction (Marone and Stampanoni 2012) results in volumetric data with voxel sizes of 0.74 and 0.37  $\mu\text{m}$ , respectively. These experiments were performed on the TOMCAT beamline (Stampanoni et al. 2006) at the Swiss Light Source, Paul Scherrer Institut, Villigen, Switzerland. Specimens are deposited at the Swedish Museum of Natural History, Stockholm.

The taxon and character lists in the phylogenetic hypothesis proposed by Skovsted et al. (2011) and Larsson et al. (2013), were used to generate a character matrix for 13 taxa (10 characters). Characters were coded using only direct fossil evidence. Parsimony analysis was run on this dataset in “Tree analysis using new technology” (Goloboff et al. 2008). Characters were treated as unordered and unweighted and all most parsimonious trees were found using implicit enumeration. This resulted in 50 most parsimonious trees, with tree length 11. The resulting tree set was then subjected to a splits analysis using the package *Splitstree4* (Huson & Bryant 2006). A consensus network was calculated from all trees, and splits were included above weight thresholds of 0.10 and 0.50.

## RESULTS

### Shell-penetrating tubes

Coalescing setal tubes have been described in sellate sclerites of both *Tannuolina pavlovi* (Kouchinsky et al. 2010) and *Micrina*

*etheridgei* (Holmer et al. 2008a, Fig. 1, A). We encountered comparable structures in two tannuolinid taxa, *M. etheridgei*, and *Tannuolina multifora*, from both mitral and sellate sclerites (Fig. 2). In sellate sclerites of both *Micrina* and *Tannuolina*, these coalescent pores are found along the sclerite margins (Fig. 2, A–E, P–T), comprising coarse aperturally directed pores (approx. 10  $\mu\text{m}$  in diameter) coalescing with finer pores (approx. 5  $\mu\text{m}$  in diameter) at angles of approximately 35–45 $^\circ$ , 30–50  $\mu\text{m}$  below the outer surface of the sclerite. Coarse pores broaden towards the outer surface and penetrate the entire sclerite. These are comparable in size to other known shell-penetrating tubes of brachiopods, e.g., those described by Jin et al. (2007). The finer pores run sub-parallel to the outer sclerite surface and do not open to the inner surface. Pores of the same geometry are present in mitral sclerites of *Micrina* around the adapical margin, and both canals are seen penetrating the inner surface (Fig. 2, F–O). Pores in mitral sclerites of *Tannuolina* dominate the accrescent side and can be very densely packed and up to approximately 50  $\mu\text{m}$  in diameter. No coalescent pores have been identified in examined specimens (Fig. 3).

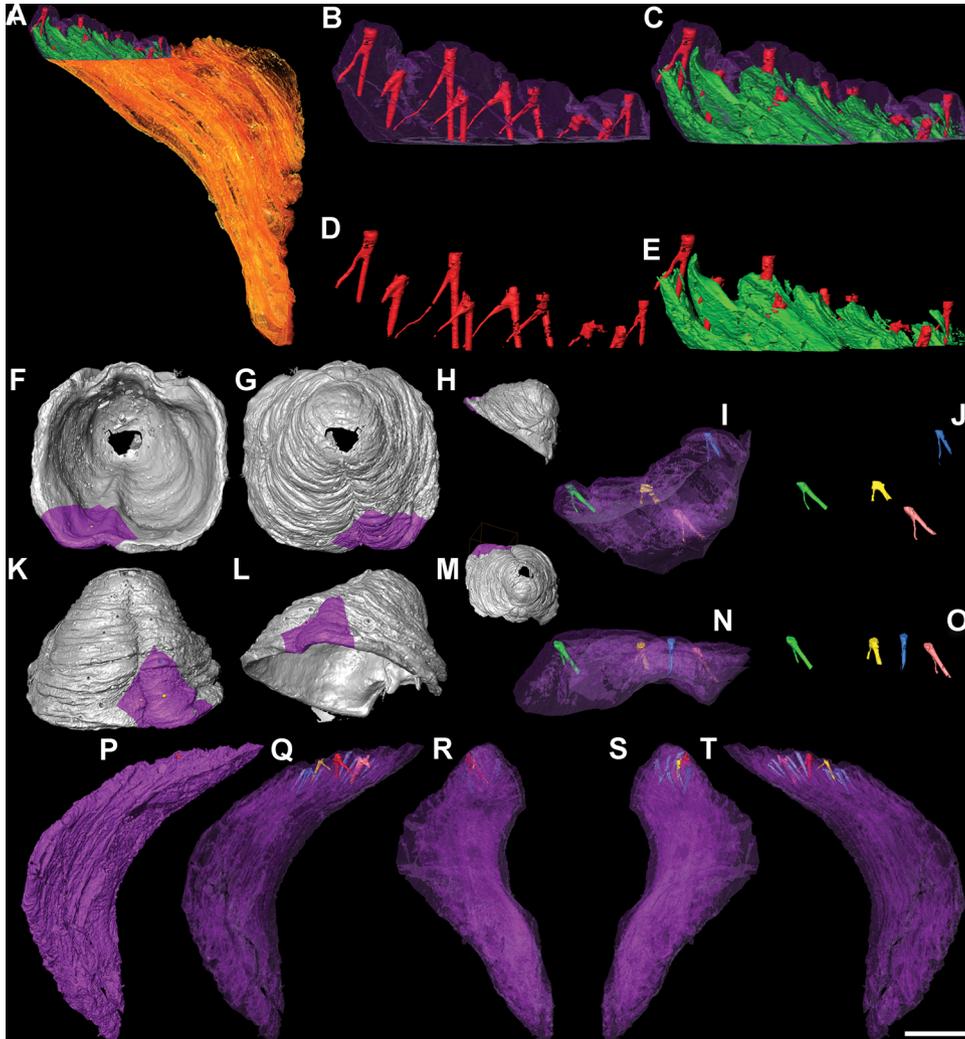
### Tommotiid microstructure

*Kulparina* sclerites display a series of alternating dense and porous layers, packaged into first and second order lamination, with a pervasive polygonal fabric (Fig. 4). This is directly comparable to the internal structures described for *Paterimitra* (Larsson et al. 2013), *Eccentrotheca* (Balthasar et al. 2009), and *Askepasma* (Topper et al. 2013). *Sunnaginia imbricata* also displays an alternation of dense and porous layers in the Siberian material, but no polygonal fabric. However, sclerites of *Sunnaginia ?imbricata* from the Comley Limestone, Shropshire, UK, have an architecture unique among the tommotiids, consisting of alternating dense and porous layers with inter-lamellar cavities spanned by pillars resulting in a colonnaded architecture (Fig. 5, D and Murdock et al. 2012). These cavities are not simply inflated porous laminae but, rather, the laminae are deflected around them. However, our data demonstrate significant differences in the sclerite structure of *Tannuolina* and *Micrina* versus *Sunnaginia*. Tannuolinid sclerites are dominated by cavities and penetrating tubes that obscure much of the original lamination (Fig. 5, C). In addition, the cavities are not spanned by pillars, as in *Sunnaginia*, but are instead punctured by shell-penetrating tubes.

## DISCUSSION

### Shell-penetrating tubes

A significant component of the case for the brachiopod affinity of the tommotiids is built upon the similarity between shell-penetrating tubes in sclerites of members of the family Tannuolinidae (Conway Morris and Chen 1990; Williams and

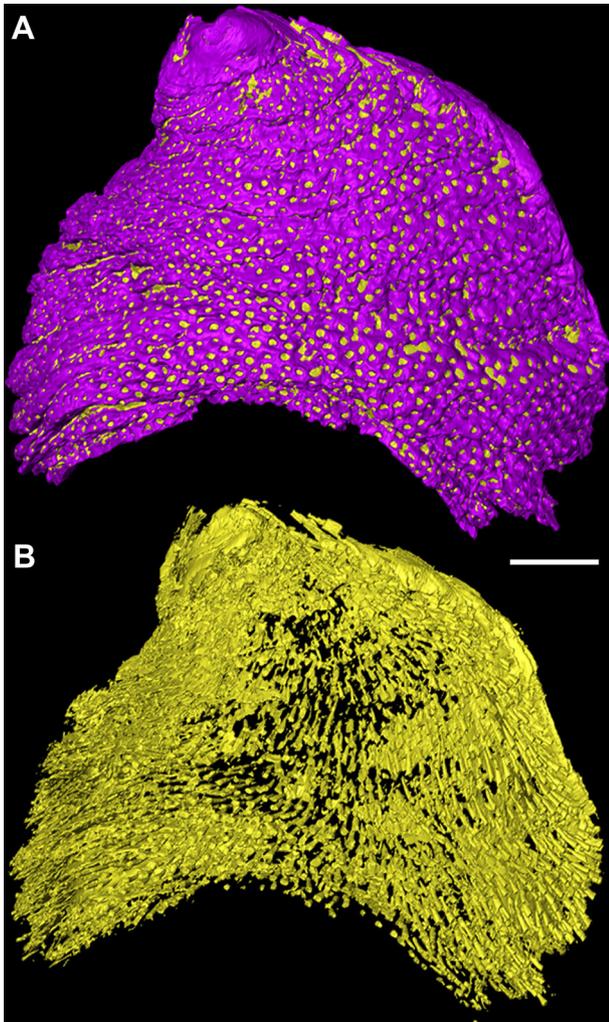


**Fig. 2.** Bifurcating pores in tannuolinid sclerites. A–O: *M. etheridgei* from Sample UNEL1858, Todd River Dolomite, Ca 70 km SE of Alice Springs, Northern Territory, 24°13'S, 134°19'E (A–E) and Sample UNEL1845 Ajax Lst, Mt. Scott Range, South Australia (F–O), see Bengtson et al. (1990) for further details. P–T: *T. multifora* from Ulug-Shangan River, Tannu-Ola Range, Altaj-Sajan, Siberia. SRXTM images. A–E: SMNH X5032 reconstruction of sellate sclerite with transparent section showing rendering of infilled cavities (green) and pores (red); close-up of transparent section with cavities (B and D) and/or outline (D and E) removed. F–O: SMNH X5033 reconstruction of mitral sclerite (white) with expanded section near adapical margin (purple) with selected pores infilled (colored). F, G, K, and L: Complete sclerite in adapical, apical, lateral, and oblique lateral views. H–J and M–O. Expanded section with rendering of infilled pores, transparent sclerite removed for clarity (J and O) and orientation of sclerite in each case shown (H and M). P–T: SMNH X2334 reconstruction of sellate sclerite with renderings of selected pores (colored), sclerite transparent and shown rotated successively by 90° through (Q–T). Scale bar represents 220  $\mu\text{m}$  (A); 100  $\mu\text{m}$  (B–E); 360  $\mu\text{m}$  (F, G, K, and L); 720  $\mu\text{m}$  (H and M); 140  $\mu\text{m}$  (I, J, N, and O); and 180  $\mu\text{m}$  (P–T).

Holmer 2002; Li and Xiao 2004) and in more derived stem-brachiopods, viz. the mickwitziiids (Skovsted and Holmer 2003; Balthasar 2004). The tubes in the sclerites of *Mickwitzia* are inferred to have housed setae in vivo. Mickwitziid shell-penetrating tubes are characteristically straight-walled (not tapering), open to the external surface, and are surrounded by significantly inward bending layers (Balthasar 2004). These features contrast with the perforations found in *Micrina* sclerites, specifically in the lack of inwardly deflected layers, and the presence of longitudinal striations (Williams and Holmer 2002).

Balthasar (2004) ascribed these differences to alternative modes of setal secretion.

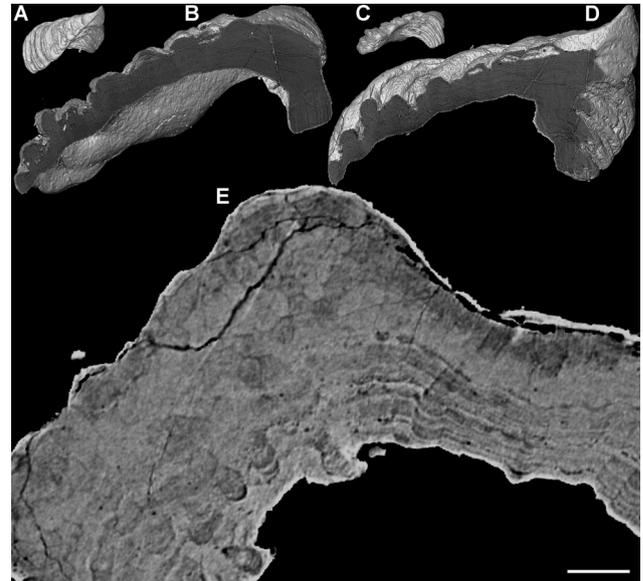
Aside from being present in sclerites of *M. etheridgei*, shell-penetrating tubes have previously been identified in sclerites of *T. multifora* (Fonin and Smirnova 1967; Qian and Bengtson 1989) and *T. pavlovi* (Kouchinsky et al. 2010). Fine (1–2  $\mu\text{m}$ ) pores that end in bulb-like swellings seen in *T. pavlovi* may be interpreted as abandoned fine setae that were sealed from the inside when new growth lamellae were deposited (Kouchinsky et al. 2010). These are comparable to bundles of phosphatized



**Fig. 3.** Pores in mitral sclerite of *T. multifora* from Ulug-Shangan River, Tannu-Ola Range, Altaj-Sajan, Siberia. SRXTM images. A: SMNH X2333 reconstruction of mitral sclerite, with rendering of infilled pores. B: Sclerite removed for clarity. Scale bar represents 500  $\mu\text{m}$ .

setae within blind-ended striated epipunctae of Ordovician orthide brachiopods (Jin et al. 2007), the fine striation of the tube walls representing the impressions of setal canals. The coalescence of shell-penetrating tubes has been described in sellate sclerites of both *T. pavlovi* (Kouchinsky et al. 2010) and *M. etheridgei*, in the latter referred to as "...a penetrative striated setal column and a *Micrina*-mickwitziid column" (Holmer et al. 2008a, Fig. 1, A). To this we add homologous structures in both the mitral and the sellate sclerites of the two tannuolinid taxa, *M. etheridgei* and *T. multifora*.

The interpretation of shell-penetrating pores of tannuolinid sclerites as housing setae *in vivo* is integral to the proposed close affinity between tommotiids and brachiopods. However, the coarse pores that bifurcate basally, seen in *Micrina* and *Tannuolina*, cannot readily be explained as openings for setae.



**Fig. 4.** Comparison of the microstructure of *K. rostrata* and "*E. guano*". A–D: *K. rostrata* and E. "*E. guano*" from Sample UNEL1858 Kulpara Lst, ca 7.5 m below base of Parara Lst, Horse Gully, Ardrossan, South Australia, 34°28'S, 137°53'E, see Bengtson et al. (1990) for further details. SRXTM images. A–D: SMNH X5031 reconstruction of sclerite with cross section combined with surface rendering, in oblique apical (A and B) and lateral (C and D) views. Orientation of sclerite shown in each case (A and C). E: SMNH X5035 Cross section through sclerite, orthogonal to outer surface. Scale bar represents 300  $\mu\text{m}$  (A and C); 100  $\mu\text{m}$  (B and D); and 25  $\mu\text{m}$  (E).

Distally branching setae in lophotrochozoans are not uncommon (e.g., Hausam and Bartolomaeus 1998), but branching in a basal direction would have to be explained as coalescing double setae or double-rooted setae. The large angle between the basal branches of the pores in the tannuolinids are difficult to reconcile with a merging of two adjacent setae, and the known mode of formation of lophotrochozoan setae does not suggest any mechanism to produce double roots. Pores in brachiopod shells not housing setae are considerably more complex (e.g., Pérez-Huerta et al. 2009), and likely polyphyletic; we are not aware of any direct analogue to the basally bifurcating type described here and in *T. pavlovi* (Kouchinsky et al. 2010). We suggest that a non-setal nature of the coarse pores is more likely (although the function of these pores is unclear), as the proposed presence of setae imposes morphological constraints that do not seem to be present.

### Scleritome reconstructions and mode of life

The tommotiids are reconstructed with a diverse range of scleritomes and modes of life. A number of configurations for the sclerites of *Camenella* (and by inference *Dailyatia* and *Lapworthella*) have been proposed (see Li & Xiao 2004 for discussion), but the consensus rests invariably on a vagile

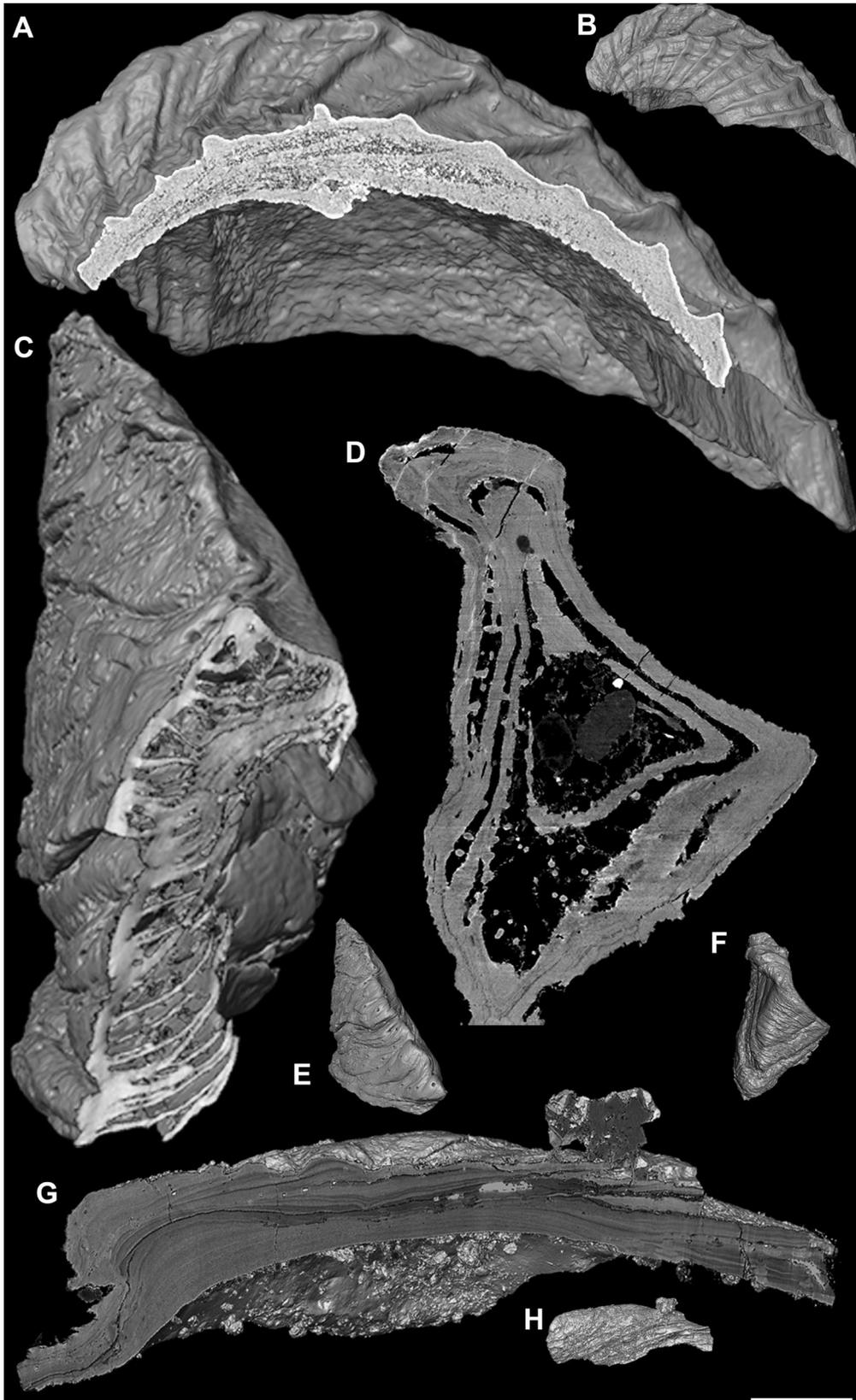


Fig. 5. Continued.

vermiform bilaterian with a dorsal covering of imbricating sclerites, supported by evidence of mechanical wear on sclerites of *Camenella reticulosa* (Skovsted et al. 2009a). In contrast, recent discoveries of partially articulated specimens of *Eccentrotheca* (Skovsted et al. 2008, 2011) and *Paterimitra* (Skovsted et al. 2009b; Larsson et al. 2013) have provided an alternative, tubular scleritome surrounding an attachment organ of a sessile organism.

The *Eccentrotheca* scleritome consists of a series of rings of sclerites comprising a tube expanding gradually toward the adapical end, open at both ends. The scleritome is inferred to have formed as a series of disconnected rings surrounding a sessile vermiform organism, which fused progressively to form a tube. Although it was attached to a substrate, Skovsted et al. (2011) note that the attachment structures were unlike the pedicle of Cambrian brachiopods. The scleritome of *Paterimitra* is known from conjoined symmetrical (“S”) sclerites combined with smaller asymmetrical (“L”) sclerites, and it is interpreted to have been derived from an *Eccentrotheca*-like scleritome (Skovsted et al. 2009b; Larsson et al. 2013). Similarities between *Kulparina rostrata* and *Eccentrotheca guano* led Skovsted et al. (2011) to suggest synonymising these taxa, since *K. rostrata* sclerites equivalent to *Paterimitra* “S” sclerites and “*E. guano*” as “L” sclerites occur in a tubular *Kulparina* scleritome. This synonymy is supported by the close similarity in shell structure of the two sclerite types (Fig. 4). An *Eccentrotheca*-like scleritome has, therefore, been proposed as the basal condition for all more derived tommotiids. The co-occurrence and similar size, of sclerites of *Sunnaginina ? imbricata* and *Eccentrotheca kanesia* (Murdock et al. 2012) is consistent with *Sunnaginina* conforming to the same gestalt, supported by a single fused specimen of *Sunnaginina imbricata* (Landing 1995).

Direct evidence for a bivalved scleritome in the tannuolinids is restricted to the relative abundance of mitral versus sellate sclerites of *Micrina*, and supported by an artificially produced bivalved “scleritome” (Holmer et al. 2008b). Holmer et al. (2008b) proposed that the merged mitral sclerites of *Tannuolina* described by Li and Xiao (2004) are “probably homologous [to *Micrina* mitral sclerites]...with the deltoid area in *Micrina* corresponding to the combined carinate sides in *Tannuolina*” (Holmer et al. 2008b, p. 727). Composite sclerites of *Tannuolina zhangwentangi* (Qian and Bengtson 1989; Li and Xiao 2004)

favor a multisclerite scleritome with the decrescent side and a surface consisting of sella, and duplicature of the sclerites forming imbricating surfaces. In addition, a *Micrina*-like scleritome according to Holmer et al.’s (2008b) reconstruction, is problematic for *T. pavlovi* (Kouchinsky et al. 2010) because the deltoid (or combined carina in *Tannuolina* sclerites) area of the mitral and duplicature of the sellate sclerites have to provide an opening for an attachment organ (pedicle). Since *T. pavlovi* lacks a carina on the mitral and a duplicature on the sellate sclerites, and the apex of the duplicatural side overhangs to such a degree, this inferred opening would be greatly reduced or absent.

The transition from the slug-like, vagile plesiomorphic lophotrochozoan condition through a tubular scleritome to the bivalved, sessile condition of crown brachiopods is central to the interpretation of tommotiids as stem-brachiopods. Scleritome reconstructions for taxa with articulated specimens are not in dispute, and it is reasonable to infer similar body plans for related taxa. However, the homology between, for example, *Camenella* mitral/sellate sclerites, *Paterimitra* S-sclerites, and *Micrina* mitral/sellate sclerites (which is inferred by this phylogenetic hypothesis), has not been demonstrated. Taking into consideration the lack of direct evidence for the bivalved condition in *Micrina*, we suggest the uncertainty surrounding scleritome reconstructions negates their use in assessing the phylogenetic affinity of the tannuolinids. In addition, considering that the scleritome reconstruction for most taxa is inferred based on hypotheses of sister relationships, the distribution of differing scleritomes should not be used to support the phylogenetic scheme.

### Sclerite architecture

The tommotiids are characterized by growth via internal accretion, which is recorded in the sclerites as a series of laminae marking variations in the nature of the apatite secreted. Usually there is an alternation of dense and porous laminae. However, there is significant variation on this basic pattern among the tommotiids. The sclerites of *Paterimitra* and *Eccentrotheca* are dominated by this lamellar structure with characteristic first- and second-order laminations, and they have been compared closely with the stem-paterinid brachiopod *Askepasma* (Balthasar et al. 2009; Larsson et al. 2013). In

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**Fig. 5.** Microstructure of selected tommotiids. A and B: *Camenella garbowskae* from Lena River, Siberia, Russia; Tommotian, Cambrian. C and E: *M. etheridgei* from Sample UNEL1863a, Todd River Dolomite, Ca 70 km SE of Alice Springs, Northern Territory, 24°13’S, 134°19’E, see Bengtson et al. (1990) for further details. D and F: *Sunnaginina ? imbricata* Missarzhevsky 1969 from Comley Limestone, Shropshire, UK; Lower Cambrian. G and H: *Sunnaginina imbricata* Missarzhevsky 1969 from Aldan River, Siberia, Russia; Uppermost Tommotian, Cambrian SRXTM images. A and B. SMNH X2241 surface rendering of (?sellate) sclerite (B) and cross section combined with surface rendering (A), in lateral view. C and E: SMNH X5034 surface rendering of sellate sclerite (E) and cross section combined with surface rendering (C), in lateral view. D and F: NRM-PZ X 4455 rendering of complete sclerite (F) and cross section (B), in apical view. G and E: NRM-PZ X 4464 rendering of complete sclerite (H) with cross section combined with surface rendering (B), in lateral view. Scale bar represents 100 µm (A and G); 300 µm (B); 200 µm (C and H); 1000 µm (E and G); 500 µm (D); and 2000 µm (F).

addition to this, all three taxa possess a polygonal ornament that is reflected in underlying laminae. The principal character distinguishing the camenellans is the presence of co-marginal ribs produced by the inflation of specific laminae close to the sclerite margin, as well as their sharing a basic cone-shaped sclerite (Skovsted et al. 2009a). Otherwise, the camenellans are directly comparable to *Eccentrotheca* and *Paterimitra*, the inflated laminae homologous to porous laminae in these taxa. *Kulparina* sclerites display the same internal structures described by Balthasar et al. (2009) for *Paterimitra* and *Eccentrotheca*, supporting the suggestion that “*E. guano*” sclerites are, in fact, part of the *Kulparina* scleritome.

The similarities between sclerite architecture of *Askepasma* and the tommotiids *Paterimitra*, *Eccentrotheca*, and *Kulparina* support a close affinity of at least some tommotiids and paterinid brachiopods (Larsson et al. 2013). Balthasar et al. (2009) argue that the brachiopod total-group is best reconstructed as “solidly rooted within tommotiids,” i.e., at least some tommotiids lie on the brachiopod stem. Nevertheless, to avoid placing undue confidence in this phylogenetic interpretation, the tommotiids should, at most, be assigned to the brachiopod total-group, as opposed to the brachiopod stem (see Donoghue and Purnell 2009 for discussion of total-group versus stem). Without demonstrating homology among the sclerites of other tommotiid genera, which is intractable without more complete scleritome models, a more detailed phylogenetic hypothesis cannot be supported. Even considering similarities in microstructure, direct comparison of disarticulated skeletal remains of organisms with apparently diverse body plans, remains problematic.

### The brachiopod affinity of the tommotiids

The re-evaluation of the distribution of characters in key tommotiid taxa, and the addition of the new data presented here, uncovers problems with several aspects of the phylogenetic hypothesis that has been presented by previous authors. The phylogeny that has been proposed for the brachiopod stem (Fig. 1) predicts a hierarchical set of characters supporting nested clades. However, the distribution of characters in this scheme (Fig. 6, A) reveals a pattern of non-, or only partially overlapping sets of characters that cannot provide intrinsic support for the phylogenetic hypothesis onto which they have been mapped. To demonstrate this, we used the taxon and character lists in the phylogenetic hypothesis proposed by Skovsted et al. (2011) and Larsson et al. (2013) to generate a character matrix, and performed a parsimony analysis, which resulted in 50 most parsimonious trees (tree length 11). We then generated consensus networks from these trees.

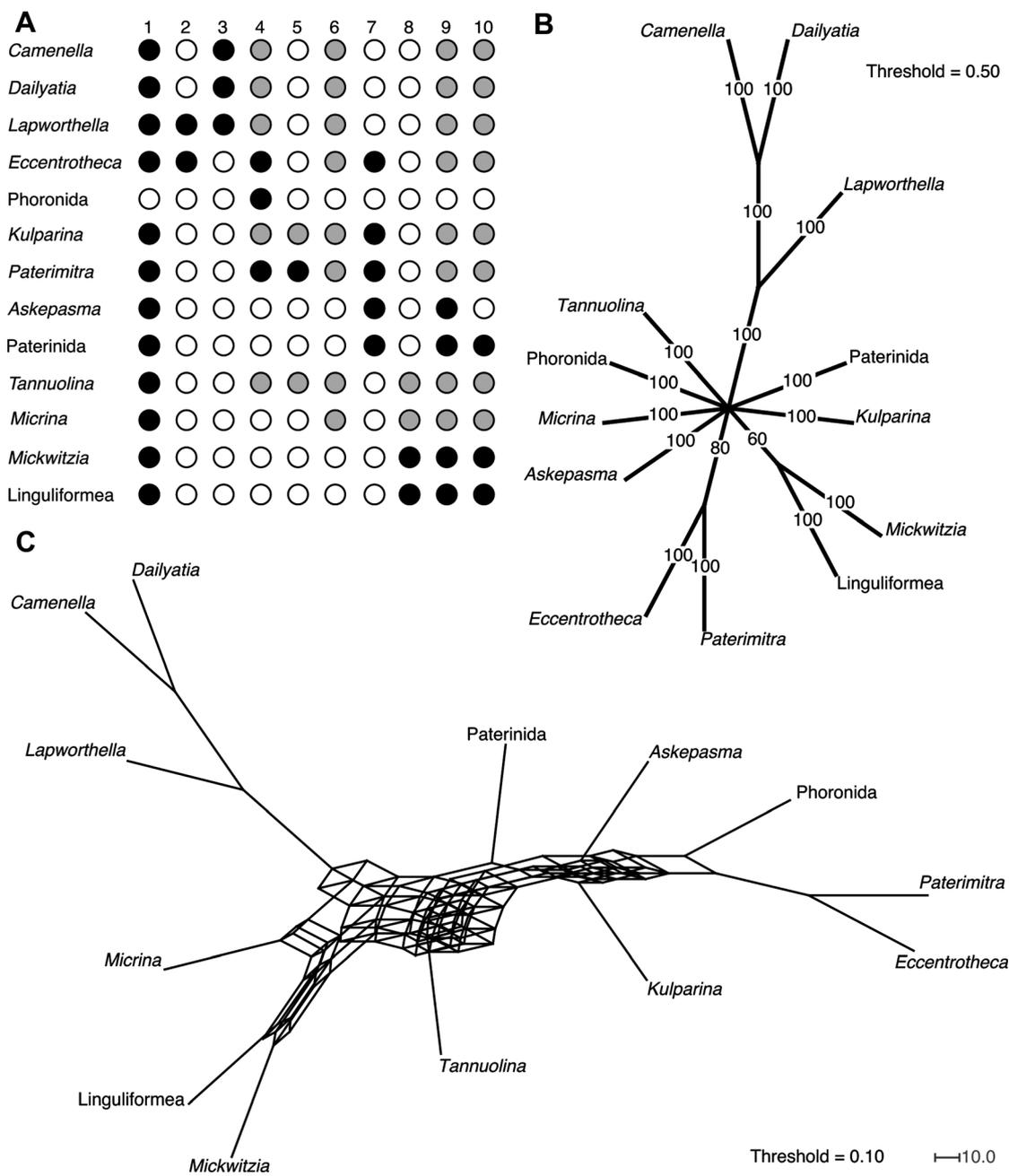
Consensus networks are a type of splits-network, which extend the amount of information portrayed by a consensus tree (Holland and Moulton, 2003). As with trees, edges within a network represent the relationships between taxa and internal nodes the possible common ancestors, while edge weights

correspond to the number of input trees in which the split is present (Holland and Moulton, 2003; Moulton and Huber, 2009). However, a phylogenetic network aims to also represent the conflict within the data, by computing and representing a collection of bifurcations present in a set of input trees, including those that may be incompatible with one another. The more tree-like a network, the less conflict is present in the dataset; in fact, a phylogenetic tree is just a special case of a phylogenetic network where the data do not conflict (Huson, 1998). Whereas consensus trees represent areas of low resolution with low support values, networks are able to represent less frequent, alternative topologies, along with their relative support (Holland and Moulton, 2003).

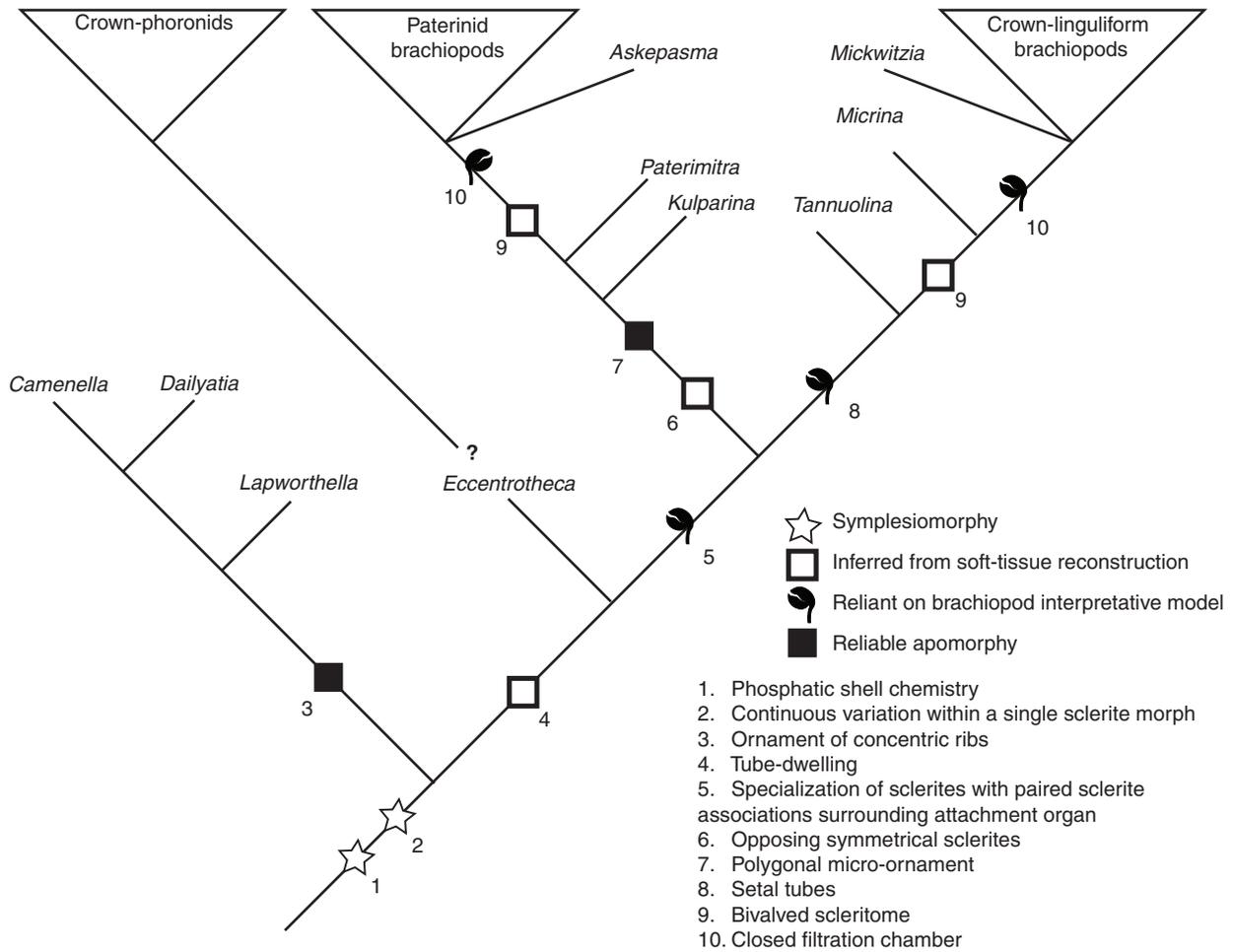
Among the 50 most parsimonious trees recovered in the exhaustive search, the hypothesis proposed by Skovsted et al. (2011) and Larsson et al. (2013) was not recovered. A “camenellan” clade comprising *Lapworthella*, *Dailyatia* and *Camenella* is recovered in all most parsimonious trees, but beyond this, where splits are drawn above a weight threshold of 0.50 (i.e., including only relationships supported by half of the most parsimonious trees), the splits analysis recovers a largely unresolved network (Fig. 6, B). Drawing the relationships present in at least 10% of most parsimonious trees demonstrates the high amount of conflict between different equally parsimonious scenarios as demonstrated by the net-like appearance of the consensus network (Fig. 6, C).

There is support for some commonly accepted clades in the network, such as the placement of *Mickwitzia* on the linguliform stem and a possible sister relationship between *Eccentrotheca* and *Paterimitra* in 80% of most parsimonious trees, that is also well supported by microstructural data (see above). However, although a close affinity between *Micrina* and linguliform brachiopods is recovered in 50% of trees, 30% place *Micrina* with the camenellans, *Tannuolina* also groups with camenellans in 30% of trees and with phoronids in another 30%, but a relationship with linguliform brachiopods is uncommon. Evidently, these character distributions can only be established post hoc from a prior hypothesis of relationships. In summary, the inferred relationships are weakly supported by an evolutionary narrative, rather than hierarchical suites of shared derived characters, based in intrinsic fossil evidence. Furthermore, as we have demonstrated, many of the characters used in support of clades are dubious in nature.

Donoghue and Purnell (2009) highlighted the importance of the intrinsic properties of fossil data in attempting to avoid the circularity of identifying anatomical homologies for phylogenetic analysis. They advocated a “bottom up” approach, in which interpretative milieu are considered and justified on the basis of intrinsic fossil evidence from among broad groupings of organisms. The tommotiids and the brachiopods have been unified largely on the basis of commonalities in their sclerite architecture. At a gross level, features such as phosphatic shell chemistry, basal internal accretion, and alternating dense and



**Fig. 6.** Phylogenetic analysis of key tommotiid taxa and their relationship to brachiopods. A: Character matrix for the characters and taxa in the hypothetical reconstruction of the phylogenetic relationships between the tommotiids and crown-brachiopods, based on Skovsted et al. (2011) and Larsson et al. (2013). Presence of a character indicated by a filled circle, absence by an open circle. Uncertain characters assignments indicated by a gray circle. Character distribution implied by direct evidence from the fossil record. Characters: (1) Phosphatic shell chemistry; (2) continuous variation within a single sclerite morph; (3) ornament of concentric ribs; (4) tube-dwelling; (5) specialization of sclerites with paired sclerite associations surrounding attachment organ; (6) two opposing symmetrical sclerites; (7) micro-ornament of polygonal compartments; (8) setal tubes; (9) bivalved scleritome; (10) closed filtration chamber. B: Consensus networks calculated from all most parsimonious trees, splits included above a weight threshold of 0.50, edge lengths quoted. C: Consensus networks calculated from all most parsimonious trees, splits included above a weight threshold of 0.10. In both cases, edge lengths reflect weight, i.e., percentage of most parsimonious trees (mpts) which recover any given edge.



**Fig. 7.** Hypothesis of the phylogenetic relationships between the tommotiids and crown-brachiopods, based on Skovsted et al. (2011) and Larsson et al. (2013). Characters used to reconstruct relationships shown below the nodes for which they are proposed as apomorphies. All, bar two, can be discounted or refuted as genuine apomorphies for one of three reasons, see text for details.

porous laminae support this affinity. However, these features are not exclusive to Brachiopoda, nor is the tubular scleritome or sessile life habit. Also, multiple switches from phosphatic to calcitic mineralization are implied by a crown-Brachiopoda rooted in the tommotiids. As noted by Sperling et al. (2011), this is inconsistent with the pattern observed in other biomineralizing groups.

The chiral symmetry of tommotiid sclerites clearly supports membership of Bilateria, and a lophotrochozoan affinity is suggested by the presence of at least some structures consistent with setal tubes. However, there is little consensus over the relationships among lophotrochozoan phyla and, therefore, character evolution in this clade. Indeed, the plesiomorphic condition for Lophotrochozoa has been proposed to be typified by other taxa, with distributions of characters that cannot be reconciled with the tommotiid–brachiopod model; such as *Cotyledion* recently interpreted as a stem-entoproct (Zhang et al., 2013). How, then, can we polarize character evolution among

brachiopods and phoronids and, indeed, justify the interpretation of tommotiids in light of brachiopods? This will remain a limiting factor until relationships among extant lophotrochozoans are resolved and more anatomical information on the tommotiids is recovered, to distinguish better, through congruence, anatomical homologies from simple similarities that are the consequence of convergence, not vertical descent.

## CONCLUSION

In light of these new data we believe that the detailed phylogenetic scheme that has been proposed for the tommotiids is not strongly supported. This concerns, in particular, the choice of characters defining the acquisition of the brachiopod body plan. The phylogenetic scheme that has been proposed for the brachiopod and phoronid stems relies largely on a mosaic of only partially overlapping suites of characters among putative sister

taxa, rather than on hierarchically nested suites of characters defining successively inclusive clades. Almost exclusively, these characters can be rejected for one of three reasons: (i) the characters are not exclusive to the clade they are proposed to support, and/or are symplesiomorphies of higher clades; (ii) the characters are inferred from soft-tissue reconstructions which remain to be tested; (iii) circularity in the relationship between the interpretation of the homologies and phylogenetic inference (Fig. 7).

Although a placement within the Lophotrochozoa can be supported for the tommotiids, their precise affinities with the crown-Brachiopoda remain to be demonstrated. Instead, based on the available evidence, we find no support for resolving the phylogenetic affinity of the tommotiids beyond total-group Brachiopoda. This is in no small part due to the paucity of phenotypic characters that is an intrinsic property of a record consisting largely of disarticulated skeletal remains; there are simply not enough data to exact congruence tests of hypotheses of homology. Statements affirming tommotiids as de facto brachiopods should therefore be avoided. The interpretation of the tommotiids serves as a model for many other Cambrian fossil groups which are known largely, or exclusively, from disarticulated skeletal micro remains.

Nevertheless, it is clear that there is a wealth of information to be extracted from the small shelly faunas for the reconstruction of phylum-level body plan evolution. Discoveries of new taxa (Kouchinsky et al. 2010), new articulated specimens (Skovsted et al. 2008, 2009b; Larsson et al. 2013) and new microstructural data (Balthasar et al. 2009; Skovsted et al. 2009a; Murdock et al. 2012; Larsson et al. 2013) have been crucial for informing our understanding of the assembly of the brachiopod body plan, and with greater taxonomic sampling coupled with further discoveries, these hypotheses can be further refined. However, material progress in elucidating the evolutionary insights afforded by the tommotiid fossil record is unlikely until the evolutionary relationships of the living lophotrochozoans are resolved.

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