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RESEARCH ARTICLE

The structure and vasculature in pollen cones of *Taxus* (Taxaceae, Gymnospermae) and its evolutionary significance

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Abstract

The pollen cone structure and vasculature of *Taxus baccata* and *T. wallichiana* were investigated in the context of comparative studies in other conifers. The results indicate that the simple, flower-like pollen cone of *Taxus* could be derived from an ancestral compound inflorescence as in *Cephalotaxus* and *Pseudotaxus*. In *Cephalotaxus*, each lateral flower-like structure represents an entire pollen cone, compared with a single perisporangiate microsporangiophore in *Pseudotaxus*. The microsporangiophores are exclusively perisporangiate in both *Pseudotaxus* and *Taxus*, but in *Pseudotaxus* they are inserted in the axil of a pherophyll. By a simple reduction of these pherophylls, the *Taxus* pollen cone is formed—a strongly reduced inflorescence with flowers reduced to perisporangiate microsporangiophores, which are inserted directly at the cone axis, without pherophylls. Thus, the perisporangiate *Taxus* microsporangiophores correspond to the axillary flowers in *Cephalotaxus* and *Pseudotaxus*. The *Taxus* flower results from a strong reduction of an ancestral *Cephalotaxus*-like flower type (lateral pollen cones), where all remaining hyposporangiate microsporangiophores have fused to a radial structure during evolution. Thus, perisporangiate *Taxus* microsporangiophores represent radial synangia and not peltate microsporophylls, suggesting that hyposporangiate microsporangiophores are the ancestral condition in Taxaceae s.l.

KEYWORDS
conifer, evolution, flower, inflorescence, morphology, reproduction, sporophyll

1 | INTRODUCTION

This article represents part of a series describing pollen cone structure in different Taxaceae s.l. (Dörken et al., 2011; Dörken & Nimsch, 2016, 2023; compare the systematic note in Material and Methods for the taxa included in the Taxaceae s.l.). In the majority of conifers, the pollen cones are simple, flower-like structures consisting of numerous, hyposporangiate microsporangiophores that each lack a subtending bract-like pherophyll (e.g., Dallimore & Jackson, 1966; Eckenwalder, 2009; Farjon, 2010; Gifford & Foster, 1989; Krüssmann, 1955, 1983). Thus, these pollen cones fulfill the definition of a single "flower" (Hess, 2019; Hirmer, 1936; Stützel, 2021; Weberling, 1989; Wettstein, 1935; compare special terms in Material and Methods for the definition of "flower" used in in this study). In contrast, a few conifer taxa have compound pollen cones, including some Podocarpaceae (e.g., *Podocarpus*) and some Taxaceae s.l. (e.g., *Cephalotaxus*), which fulfill the definition of an "inflorescence" (Dörken et al., 2011; Mundry & Mundry, 2001; Schulz et al., 2014; compare special terms in Material and Methods for the definition of "inflorescence" used in in this study). Structural diversity within Taxaceae s.l., with both simple and compound pollen cones and both hyposporangiate and perisporangiate microsporangiophores is unique among extant conifers.

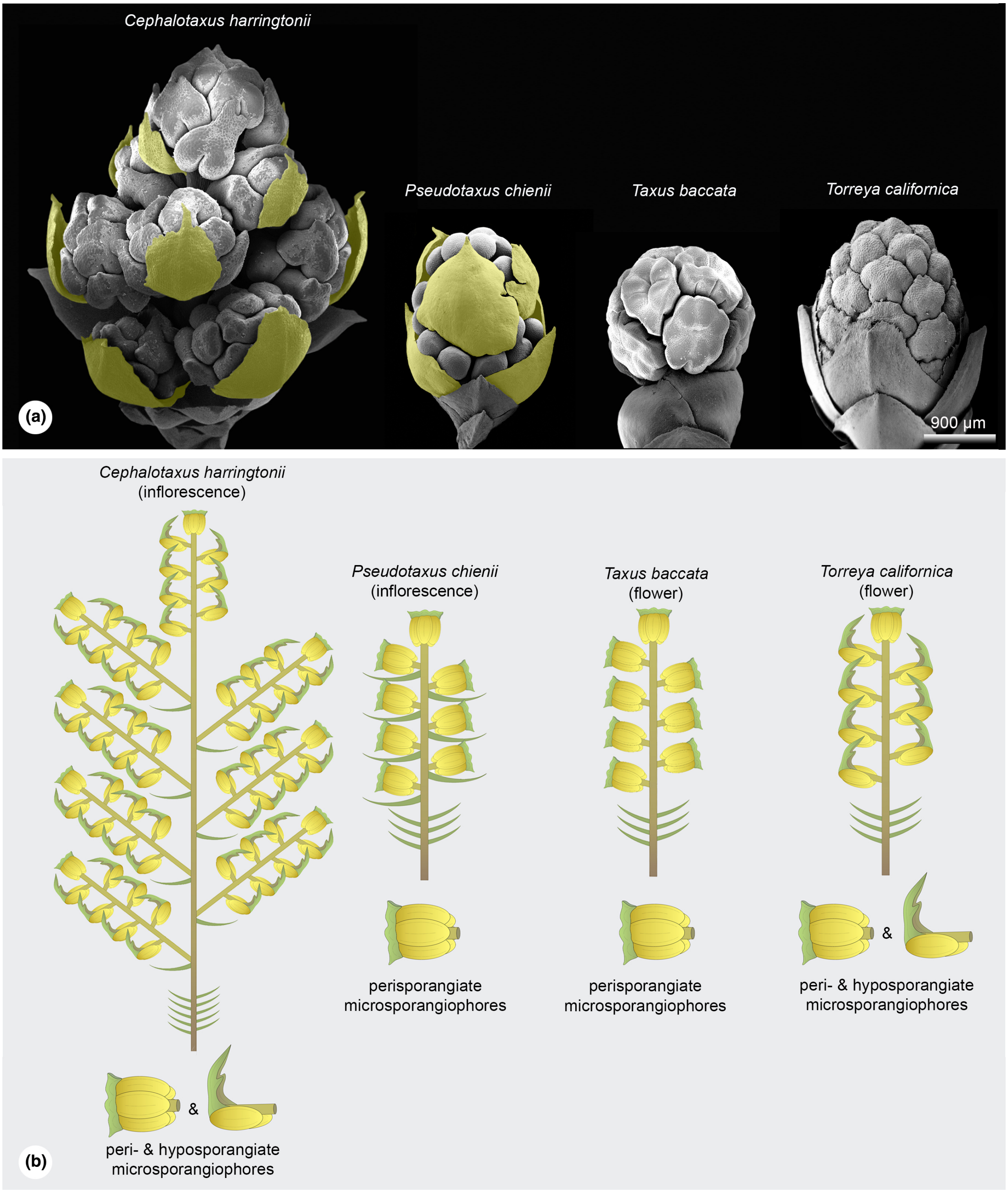


FIGURE 1 Pollen cones of Taxaceae s.l. (a) SEM images; (b) Schematic drawings of pollen cones and their microsporangiophores (note—the drawings do not reflect the natural size dimensions between the taxa, which can be taken from [a]); those of *Cephalotaxus* and *Pseudotaxus* are compound structures with scale-like subtending pherophylls (yellow), which bear axillary male reproductive units ("flowers"); each pherophyll bears an axillary pollen cone in *Cephalotaxus* and a single perisporangiate microsporangiophore in *Pseudotaxus*; pollen cones in *Taxus* and *Torreya* have a relatively simple structure; pherophylls are absent and the microsporangiophores are non-axillary; microsporangiophores of *Pseudotaxus* and *Taxus* are perisporangiate; in *Cephalotaxus* and *Torreya*, only the terminal one is perisporangiate; the lateral ones are hyposporangiate.

Therefore, investigations of taxaceous pollen cones seem to be a good starting point to get new insides into the evolution of coniferous male reproductive structures.

The *Cephalotaxus* inflorescence (Figure 1) consists of several lateral pollen cones (flower analogs), each inserted in the axil of a scale-like pherophyll (subtending leaf). In *Pseudotaxus* inflorescences (Figure 1), the lateral flowers are reduced to a single axillary perisporangiate microsporangiophore. Simple, flower-like pollen cones are formed in *Taxus* and *Torreya* (Figure 1). They consist of numerous non-axillary, microsporangiophores. Interestingly, the simple pollen cone of *Taxus* is structurally quite similar to the compound pollen cone of *Pseudotaxus*, the main difference being the absence of pherophylls in *Taxus*, so that all perisporangiate microsporangiophores are non-axillary. Leaves are always non-axillary structures, developed as lateral organs on the shoot apex, so the axillary position of the perisporangiate microsporangiophores in *Pseudotaxus* excludes a priori their interpretation as multisporangiate pollen-producing leaves in the sense of a peltate microsporophyll. This observation conflicts with the general interpretation of microsporangium-producing structures in conifers, which are widely accepted as microsporophylls (= pollen producing leaves) (e.g., Coulter & Chamberlain, 1917; Dallimore & Jackson, 1966; Eckenwalder, 2009; Farjon, 1984, 2005, 2010; Florin, 1951; Hirmer, 1936; Krüssmann, 1955, 1983; Liu, 1971; Lotsy, 1911; Mirov, 1967; Page, 1990; Spencer et al., 2015; Sporne, 1965; Williams, 2009). It seems most likely that perisporangiate microsporangiophores in *Pseudotaxus* represent strongly reduced lateral pollen cones with a markedly reduced axis (Dörken & Nimsch, 2023). The reduction of the axis is so strong that the primordia of all inserted hyposporangiate microsporangiophores get in contact and fuse to a radial structure—the perisporangiate microsporangiophore, which explains their axillary position. Thus, the perisporangiate microsporangiophores in *Pseudotaxus* are homologous to the flowers (lateral pollen cones) in the *Cephalotaxus* inflorescence, however strongly reduced, (Dörken et al., 2011; Dörken & Nimsch, 2016, 2023; Mundry & Mundry, 2001; Schulz et al., 2014).

These comparative observations raise two major questions, both have long been debated in the literature: (1) Does the simple, flower-like *Taxus* pollen cones represent a strongly reduced inflorescence-like structure, apparently derived from a compound *Pseudotaxus*-like ancestral condition? (2) Are the non-axillary, perisporangiate microsporangiophores of *Taxus* peltate microsporophylls, or are they strongly reduced lateral flowers, each representing a radial synangium (pollen cone)? To address these questions, pollen cones of *Taxus baccata* and *Taxus wallichiana* are here investigated, using both typical wild-type pollen cones and anomalous pollen cones with hyposporangiate microsporangiophores instead of perisporangiate ones or branched microsporangiophores instead of radial ones. A special focus addresses the vasculature within the pollen cones, in particular, the vascular supply of the microsporangiophores (mainly the number and position of vascular bundle strands).

2 | MATERIALS AND METHODS

2.1 | Materials

Material of *Taxus baccata* L. was collected from trees growing in the woodlands on the University Campus Konstanz, Konstanz (Germany). Material of *Taxus wallichiana* Zucc. was collected in the living collection of the Botanic Garden Konstanz, Konstanz (Germany), where specimens are cultivated as pot plants, which are stored in a temperate green house in winter.

2.2 | Methods

Freshly collected material was photographed and then fixed in FAA (100 mL FAA = 90 mL ethanol 70% + 5 mL acetic acid 96% + 5 mL formaldehyde solution 37%) before being stored in 70% ethanol. The anatomy was studied from sections using the classical paraffin technique and subsequent astrablue/safranin staining (Gerlach, 1984). For scanning electron microscopy (SEM), the FAA-fixed material was dehydrated in formaldehyde dimethyl acetal (FDA) for 24 h (Gerstberger & Leins, 1978) and subsequently critical point-dried. Sputter-coating was performed using a sputter coater SCD 50 Bal-tec (Balzers). The specimens were examined with an Auriga Zeiss TM. Macrophotography was carried out with a digital camera (Canon PowerShot IS2) and microphotography with a digital microscope (Keyence VHX 500F) equipped with a high-precision VH mounting stand with X-Y stage and bright-field illumination (Keyence VH-S5).

2.3 | Special terms

Flower: The definition of the term "flower" has been intensively debated in the literature and numerous definitions exist. A representative spectrum of definitions is available in Bateman et al. (2006). In this study, the term "flower" is used for a morphological clearly delimitable unbranched short shoot-like axis, which carries fertile leaves (sporophylls), bears the reproductive organs, and their dimorphism is related to sexual reproduction (Hess, 2019; Stützel, 2021; Weberling, 1989; Wettstein, 1935) (Figure 1).

Inflorescence/compound pollen cones: In this study, the terms "inflorescence" and "compound" are used only for branched male reproductive units which differ strongly from the vegetative parts; they develop from a single bud in the taxaceous genera *Cephalotaxus* and

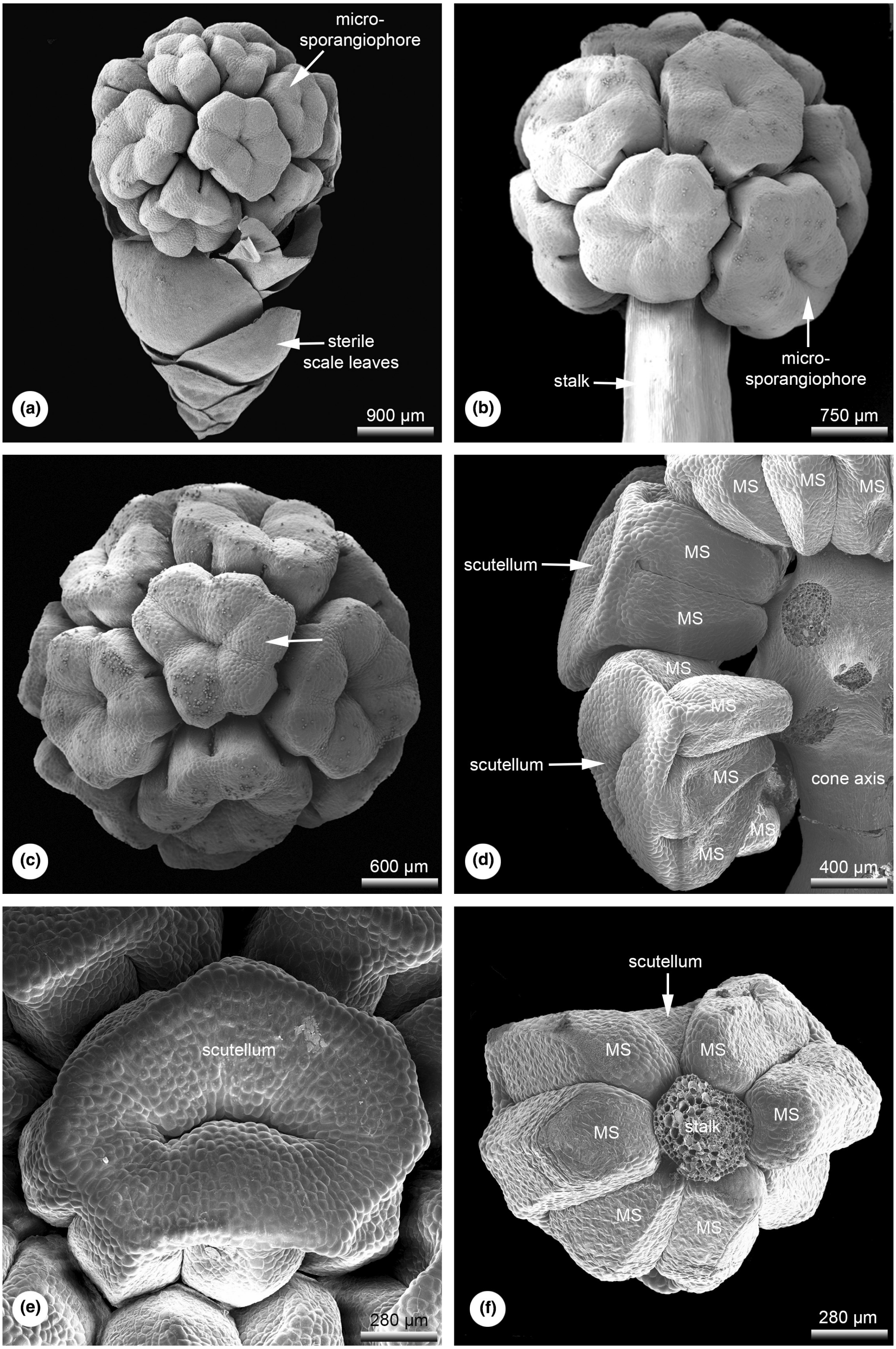


FIGURE 2 Pollen cone structure (SEM images); (a–c) *Taxus baccata*; (d–f) *T. wallichiana*; (a) Lateral view of a pollen cone with several basal pairs of scale leaves and numerous distal perisporangiate microsporangiophores; (b) The cone axis elongates at pollination time; (c) Top view; cone axis with a terminal perisporangiate microsporangiophore (arrow); (d) Pherophylls within the pollen cones are absent; (e) Top view of a perisporangiate microsporangiophore with a shield-like scutellum; (f) Bottom view of a perisporangiate microsporangiophore with numerous microsporangia arranged all around the central stalk. MS, microsporangium.

Pseudotaxus. Cases of male reproductive structures consisting of several lateral units (pollen cones) that each develop from a single bud, should not be termed compound or inflorescence; they represent a dense pollen cone cluster, typical of, for example, *Sciadopitys verticillata* (Sciadopityaceae) and *Juniperus drupacea* (Cupressaceae s.str.) (Dörken, 2019; Dörken & Nimsch, 2023) (Figure 1).

Microsporangiophore: Structure bearing the microsporangia; here, the term "sporophyll" or "microsporophyll" is not used to avoid making an a priori homology statement (Figure 1).

Hyposporangiate: Hyposporangiate microsporangiophores are dorsiventral; they consist of a central stalk, and a terminal small-scale leaf-like scutellum; and microsporangia are developed exclusively on the lower side of the stalk (Figure 1).

Perisporangiate: Perisporangiate microsporangiophores are radial; they consist of a central stalk and a terminal shield-like scutellum; and microsporangia are developed all around the central stalk (Figure 1).

Pherophyll: A lateral shoot bearing leaf (Stützel & Trovó, 2013); mostly simple; often markedly smaller when developed in the floral region above the typical trophophylls (Wagenitz, 2008) (Figure 1).

Stalk: The term "stalk" is used for the central axis of a microsporangiophore (Figure 1).

Scutellum: The terminal phylloid-like structure developed on the central stalk of the microsporangiophores is termed "scutellum" (Figure 1).

Synangium: Group of fused sporangia (Wagenitz, 2008).

2.4 | Systematic note

The systematic relationships of the taxaceous genera, in particular the position of *Cephalotaxus* and *Amentotaxus*, is still highly controversial. Previously, *Cephalotaxus* and *Amentotaxus* were often treated in a separate family (Cephalotaxaceae) and placed in the Coniferopsida (e.g., Chamberlain, 1935; Engler, 1954; Florin, 1948; Krüssmann, 1955, 1983; Pilger, 1926). All other genera (*Austrotaxus*, *Pseudotaxus*, *Taxus* and *Torreya*) were placed in a separate family called Taxaceae, which represent the Taxaceae s. str. They were placed in a distinct order called Taxales. However, subsequent studies showed a close relation between Taxaceae and Cephalotaxaceae (Keng, 1969, Chen & Wang, 1985, Stützel & Röwekamp, 1999, Restemeyer, 1999, Farjon, 1998, 2001, Mundry, 1999, Mundry & Mundry, 2001), which was later confirmed by several molecular studies (Chaw et al., 2000, Cheng et al., 2000, Quinn et al., 2002; Ran et al., 2010). Thus, Stevens (2001) and Eckenwalder (2009) merged the Cephalotaxaceae in the Taxaceae s.l. This family concept is accepted in this study, despite that in one of the latest molecular phylogenetic studies (Yang et al., 2022), the genus *Cephalotaxus* was separated from the Taxaceae s.l. and placed again in the monogeneric family Cephalotaxaceae.

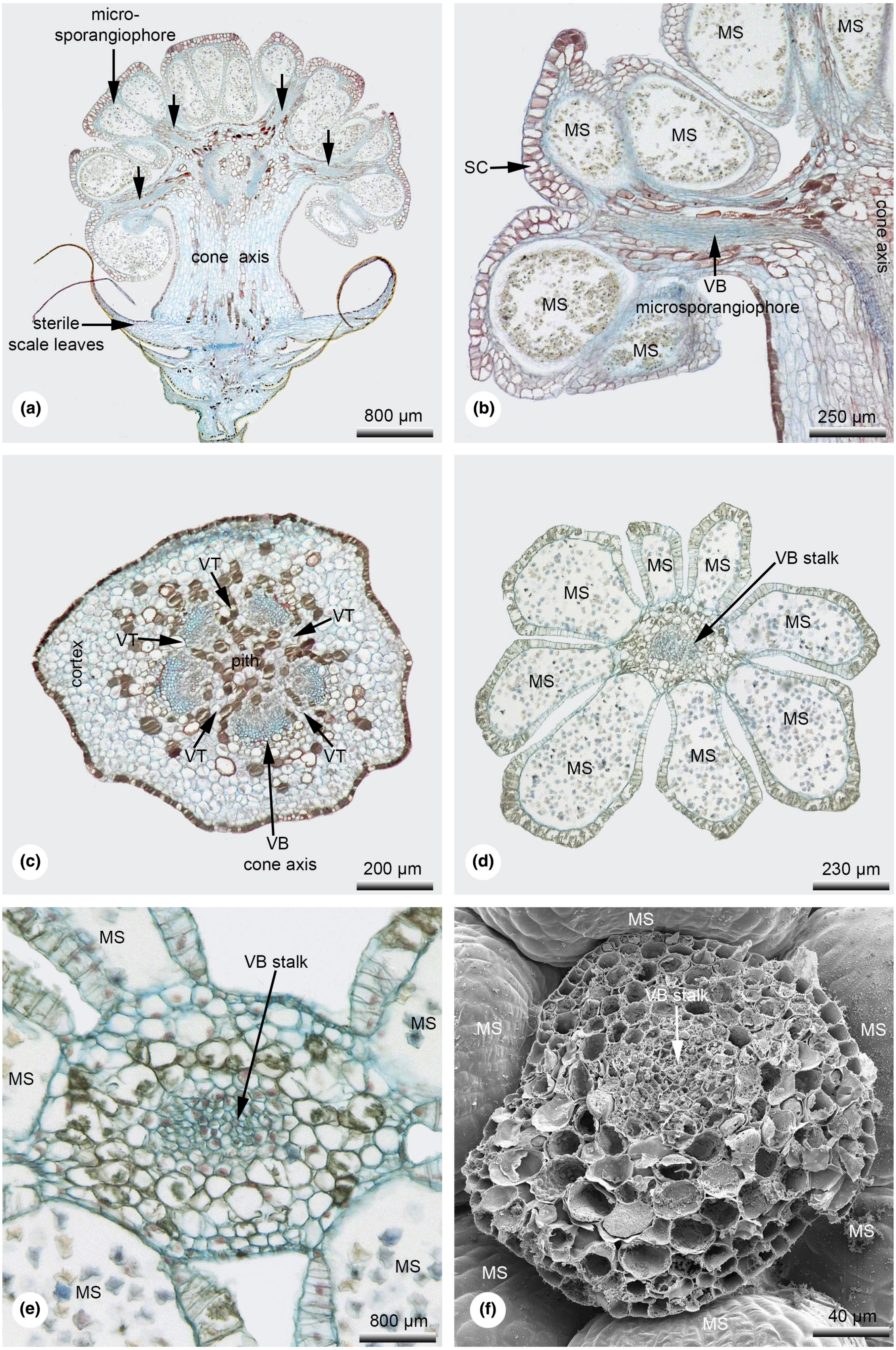
3 | RESULTS

3.1 | Wild-type pollen cones

Pollen cones of *Taxus* are already initiated in the year before pollination. They develop on lateral, plagiotropic shoots and are inserted in the axil of a typical-shaped needle leaf. The needle leaves are inserted spirally, but arranged in two lateral rows. The stalks of the pollen cones become incurved so that most pollen cones point downward on the lower side of lateral branches. Several pairs of decussate, membranous scale leaves are inserted at the base of the pollen cone (Figures 2a and 3a). The distal fertile unit is formed by numerous, perisporangiate microsporangiophores (Figures 2a–d and 3a,b). The cone axis terminates with a perisporangiate microsporangiophore (Figure 2c). At early developmental stages, the basal scale leaves cover and protect the distal microsporangiophores. Shortly before pollination, the stalk of the pollen cone elongates, so that the microsporangiophores become uncovered and well exposed to the ambient airflow (Figure 2b). Each microsporangiophore consists of (4-) 6–9 (-12) microsporangia (pollen sacs) (Figures 2f and 3d). The distal scutellum is shield-like (Figure 2e) and consists of 1–3 layers of large cells (Figure 3b). The microsporangia are attached to the stalk of the microsporangiophore as well as to the scutellum (Figure 3b,d). They are oriented parallel to the stalk and arranged radially around it (Figures 2f and 3b,d). The microsporangiophore is supplied by a single vascular strand; vascular bundles supplying each microsporangium were not found (Figure 3d–f). Due to the dense arrangement of

FIGURE 3 Vasculature of pollen cones; (a, b, c, and f) *Taxus wallichiana*; (d, e) *T. baccata* (a) Longitudinal section; microsporangiophores are supplied by a single vascular bundle strand; (b) Longitudinal section of a single perisporangiate microsporangiophore; numerous microsporangia are inserted all around the stalk; microsporangia covered by a distinct scutellum; (c) Cross-section of the cone axis; the concentric bundle is strongly fragmented by the vascular traces of the microsporangiophores; (d) Cross-section of a perisporangiate microsporangiophore; (e, f) Cross-section of the stalk of a perisporangiate microsporangiophore; there is usually a single vascular bundle strand; (e) Microtome section; (f) SEM image. MS, microsporangium; SC, scutellum; VB, vascular bundle; VT, vascular trace.

microsporangiophores, the concentric vascular bundle of the cone axis becomes strongly interrupted by the numerous vascular traces of the microsporangiophores (Figure 3c).



3.2 | Anomalous-shaped pollen cones

In addition to wild-type microsporangiophores, some anomalous forms were found; some were branched (Figure 4a–c), others supplied by more than one vascular bundle strand (Figure 4d,e), or they have a dorsiventral shape instead of a radial one (Figure 4f).

Branched microsporangiophores are rare and very few were found (Figure 4a–c). They are inserted in a lateral position on the cone axis. They consist of a common short stalk which branches once dichotomously so that two lateral fertile units are formed (Figure 4a–c). Each lateral unit has a hyposporangiate structure, consisting of a central stalk and a small distal scutellum. Up to four microsporangia are inserted on the lower side of the stalk (Figure 4a,b). A single common vascular bundle enters the basal parts of the common stalk (Figure 4a,b), and branches into two strands (Figure 4a,b), each entering the stalk of the lateral fertile units (Figure 4a–c). The vascular bundles supplying each lateral unit branch from the common stalk in different layers. Vascular bundles supplying each microsporangium were not found.

Wild-type perisporangiate microsporangiophores are supplied by a single vascular bundle strand (Figure 3d–f). In several investigated pollen cones, at least one perisporangiate microsporangiophore was found showing two vascular bundle strands (Figure 4d,e) instead of one. In other respects, these two-veined microsporangiophores appear similar to more typical single-veined microsporangiophores (Figure 3a).

In several pollen cones, dorsiventral hyposporangiate microsporangiophores were found instead of radial ones (Figure 4f). These hyposporangiate microsporangiophores each consists of a stalk and 3–4 microsporangia, are inserted on the lower side of the stalk (Figure 4f). The distal scutellum is strongly reduced. The hyposporangiate microsporangiophores are supplied by a single vascular bundle strand (Figure 4f).

4 | DISCUSSION

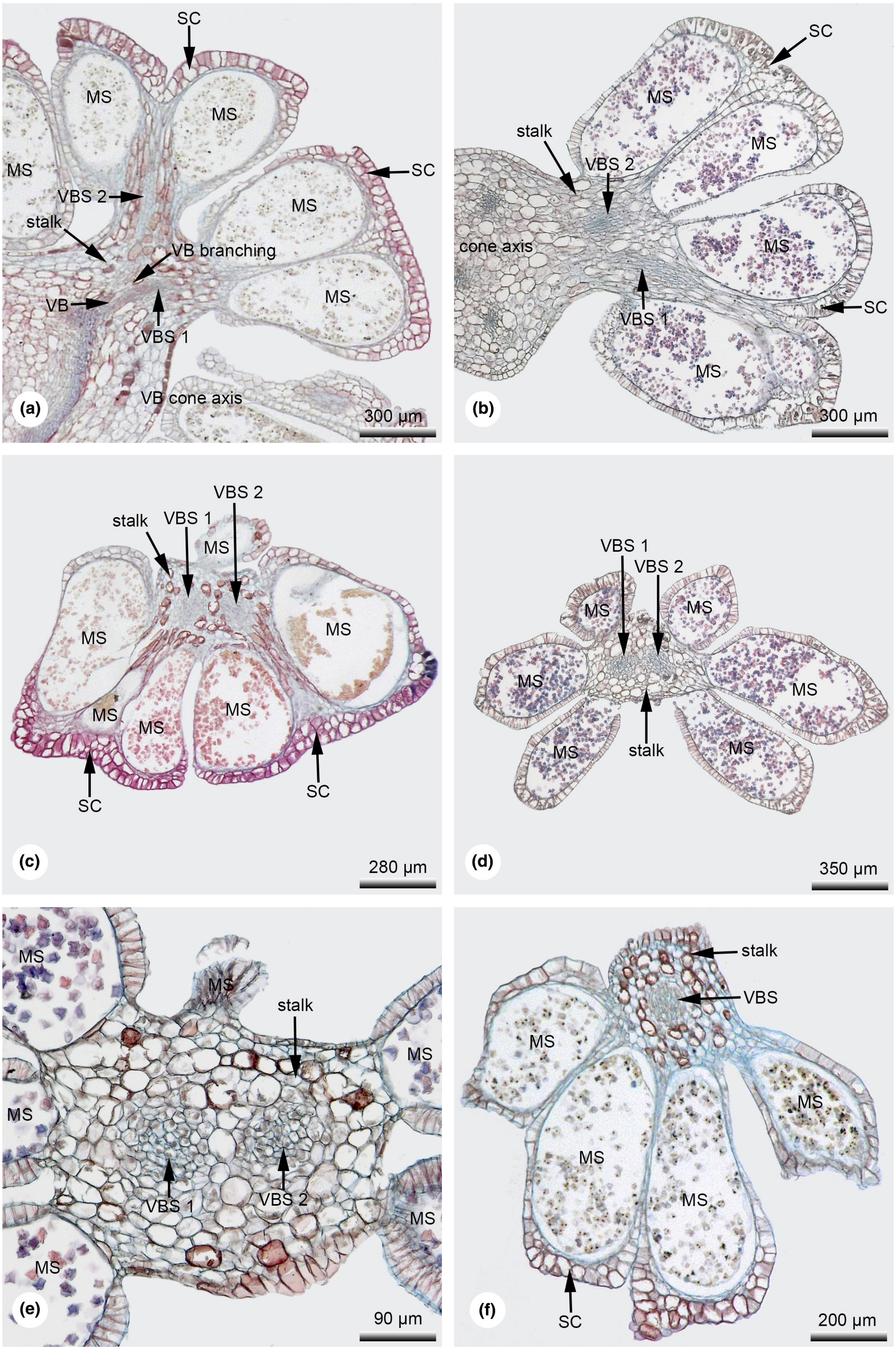
Different genera of Taxaceae s.l. display either compound inflorescence-like pollen cones (e.g., *Cephalotaxus* and *Pseudotaxus*) or simple flower-like ones (e.g., *Taxus* and *Torreya*) (Figure 1). In previous studies, an evolutionary transition was suggested, from the inflorescence-like pollen cones in *Cephalotaxus* to the simple "flowers" of *Torreya* (Dörken et al., 2011; Dörken & Nimsch, 2016, 2023; Mundry & Mundry, 2001). In the *Cephalotaxus* inflorescence, each flower corresponds to a fully developed pollen cone, and in *Pseudotaxus*, the flowers are reduced to single perisporangiate microsporangiophores. In both genera, each lateral flower is inserted in the axil of a pherophyll (Figure 1). Thus, the perisporangiate microsporangiophores of *Pseudotaxus* are homologous with an entire lateral pollen cone in the *Cephalotaxus* inflorescence.

The flower-like pollen cone of *Taxus* represents a simple structure consisting of numerous, perisporangiate microsporangiophores and lacking pherophylls. It could have evolved by a sudden loss of pherophylls in the *Pseudotaxus* inflorescence. In this concept, the perisporangiate *Taxus* microsporangiophores are homologous with a lateral, axillary flower in the *Pseudotaxus* and *Cephalotaxus* inflorescences. This evolutionary scenario therefore supports a pseudanthial origin of the *Taxus* pollen cone (Mundry & Mundry, 2001, Dörken et al., 2011; Dörken & Nimsch, 2016, 2023). Furthermore, it may help to explain the presence of two types of microsporangiophore in Taxaceae s.l., both hyposporangiate (dorsiventral) and perisporangiate (radial), whereas all other coniferous families are exclusively hyposporangiate (e.g., Dluhosch, 1937; Dupler, 1919; Eckenwalder, 2009; Farjon, 2010; Mundry & Mundry, 2001; Nozeran, 1955; Schulz et al., 2014; Thomson, 1940; Wilde, 1975; Worsdell, 1901).

However, the origin and structure of coniferous microsporangiophores is still controversial and there are two major conflicting theories: (1) the hyposporangiate type is derived from an ancestral perisporangiate condition (e.g., Dluhosch, 1937; Dupler, 1919; Worsdell, 1901), and (2) perisporangiate microsporangiophores are radial synangia—a fusion of dorsiventral microsporangiophores (e.g., Dörken et al., 2011; Dörken & Nimsch, 2023; Mundry & Mundry, 2001; Thomson, 1940; Wilde, 1975). The latter concept was strongly supported by investigations on perisporangiate microsporangiophores (Dörken & Nimsch, 2016, 2023) of wild-type and anomalous taxaceous pollen cones and microsporangiophores in *Cephalotaxus*, *Pseudotaxus*, *Taxus* and *Torreya*. These studies indicated that the perisporangiate microsporangiophores of *Pseudotaxus* represent a fusion product of

FIGURE 4 Pollen cone anomalies; (a, c, and f) *Taxus wallichiana*; (b, d, and e) *T. baccata*; (a–c) Branched microsporangiophores consisting of two fertile lateral units; (a, b) Longitudinal sections; there is a common basal stalk with a single vascular bundle strand, which branches one time; each vascular bundle strands enters one of the stalks of the lateral units; note that these strands branches from the common stalk in different layers. (c) Cross-section showing the two vascular bundles before they fuse in basal parts to a common one; (d) Cross-section of a perisporangiate microsporangiophore which is supplied by two vascular bundle strands; (e) Stalk of a microsporangiophore with two vascular bundle strands; (f) Cross-section of a hyposporangiate microsporangiophore; the microsporangiophore is supplied by a single vascular bundle strand. MS, microsporangium; SC, scutellum; VB, vascular bundle; VBS, vascular bundle strand).

several hyposporangiate microsporangiophores. In wild-type pollen cones of *Pseudotaxus*, the axis of the lateral flowers is usually so strongly reduced that the primordia of all inserted hyposporangiate microsporangiophores become in contact and fuse entirely to a radial structure with a single common stalk. Thus, these



strongly reduced flowers correspond to a radial synangium and not to a peltate microsporophyll (Dörken & Nimsch, 2023). This idea was supported by the fact that axillary hyposporangiate microsporangiophores were also found in many of the investigated *Pseudotaxus* pollen cones. The lateral flowers were so strongly reduced that only a single hyposporangiate microsporangiophore remained, its axillary position indicating that it is not merely a simple dorsiventral microsporophyll (Dörken & Nimsch, 2023).

While in *Pseudotaxus* and *Taxus* all microsporangiophores are perisporangiate, in *Cephalotaxus* and *Torreya* only the terminal one is perisporangiate, the lateral ones being hyposporangiate. The terminal microsporangiophore is also a fusion product of several hyposporangiate microsporangiophores. Usually the apex of the pollen cone is quite small, so that the primordia of several distal hyposporangiate microsporangiophores become in contact and subsequently fuse to a radial synangium (Dörken & Nimsch, 2016). Lateral hyposporangiate microsporangiophores are each supplied by a single vascular bundle strand, so it is not surprising that the terminal perisporangiate microsporangiophores of *Cephalotaxus* and *Torreya* have up to four vascular strands, each belonging to one of the hyposporangiate microsporangiophores involved in the formation of the terminal radial synangium (Dörken & Nimsch, 2016). In *Pseudotaxus* and *Taxus*, the reduction of the lateral flowers (perisporangiate microsporangiophores) seems to be so strong that just a single central bundle strand is needed for a sufficient vascular supply (Dörken & Nimsch, 2023).

At first glance, perisporangiate microsporangiophores in *Taxus* seem to be quite homogenous and differ only slightly in their size and in the number of inserted microsporangia. However, anatomical investigations of the vasculature show two distinct different types. The first type, the common one, is supplied by a single vascular bundle strand. The second type is supplied by two vascular bundles strands. It seems that in these “two-veined” perisporangiate microsporangiophores, the fusion of the hyposporangiate microsporangiophores (in this case two) was not as strong as usual. This idea is supported by the fact that the hyposporangiate microsporangiophores found in some of the investigated *Taxus* pollen cones were also each supplied by a single vascular bundle strand.

Further supportive arguments elucidating the perisporangiate *Taxus* microsporangiophores as radial synangia (lateral cones) were found in the anomalous branched microsporangiophores showing two lateral, hyposporangiate fertile units, which are inserted on a common stalk (Figure 4a–c). Interestingly, the vascular bundles of the fertile lateral units enter the common stalk in different layers, which excludes a priori a whorled identity for this structure. This seems to provide good evidence that each of these units represents a single hyposporangiate microsporangiophore and the entire branched structure a lateral flower (cone) and not a radial microsporangiophore. The branched lateral flowers are not as strongly reduced as is typical for the wild type. In this case, the common stalk of the two lateral units can be understood best as representing a strongly reduced rudimentary pedicel. The fact that each lateral subunit is supplied by a distinct vascular bundle strand, while the inserted microsporangia lack vasculature, might indicate that each of the lateral hyposporangiate subunits represents a dorsiventral microsporophyll.

Even today, a pseudanthial origin for the simple *Taxus* pollen cone is often doubted. The main argument of the critics against a pseudanthial interpretation is that in seed plants, the regularly branching pattern is axillary, so that lateral shoots are always inserted in the axil of a bract. Thus, the “non-axillary” *Taxus* microsporangiophores are still widely regarded as homologous to a leaf in the sense of a microsporophyll and not to a strongly reduced lateral flower, of which the pherophyll was lost during evolution. However, these critics have no problem to accept non-axillary flowers as shoots, as they can be found in the inflorescences (heads) of some taxa such as Asteraceae (Engler, 1954), Ericaulaceae (Ruhland, 1903; Stützel, 1984; Stützel & Trovó, 2013; Trovó et al., 2010), or those developed in the spicate inflorescences of Araceae, *Acorus* (Acoraceae), and several alismatids (Buzgo, 2001). Furthermore, the flowers of most Brassicaceae lack noticeable bracts (Nikolov, 2019). It is widely accepted that these flowers are derived from an axillary branching pattern and that the ancestral condition were flowers inserted in the axil of a bract.

Thus, non-axillary flowers as developed in some angiospermous inflorescences clearly demonstrate that bracts within fertile units can be strongly reduced or entirely lost during evolution, forming compact inflorescences. The axillary position of the perisporangiate microsporangiophores in *Pseudotaxus* represents a strong argument for the flower-like (shoot-like) nature of the microsporangiophores in *Taxus* and can be regard as the ancestral condition (Dörken et al., 2011; Dörken & Nimsch, 2023).

Some authors interpret the pherophylls in *Pseudotaxus* pollen cones as sterile bracts. For example, Page (1990) wrote “*Male cones present singly in the axils of foliage leaves, the microsporophyll whorls vertically separated by broad, interposed, sterile bracts*.” But within a pollen cone, such sterile pherophylls can only be explained as being modified microsporangiophores. In this case, the sterile bracts and the microsporangiophores (here representing microsporophylls) have to strictly follow the phyllotaxis, which is not the case in *Pseudotaxus* (Dörken et al., 2011).

Dörken and Nimsch (2023) demonstrated that the microsporangiophores of *Pseudotaxus* are inserted directly in the axil of the pherophyll, as is typical for a lateral branch. Interestingly, it is also not uncommon that in the *Pseudotaxus* inflorescence, the distal-most microsporangiophores (flowers, cones) have already lost their pherophyll (compare Figures 3 and 4 in Dörken et al., 2011, Figure 2 in Dörken & Nimsch, 2022, and Figure 1 in this study). This arrangement is in accordance with the situation in some angiosperm inflorescences, as discussed above, indicating that bracts can become entirely reduced in coniferous pollen cones. Therefore, it is just a small evolutionary step from *Pseudotaxus* to *Taxus*, by a simple reduction of all pherophylls.

5 | CONCLUSIONS AND OUTLOOK

The simple, flower-like pollen cone of *Taxus* could be regarded as derived from an ancestral compound inflorescence. In this concept, the perisporangiate *Taxus* microsporangiophore represents a strongly reduced flower, which corresponds to the axillary flowers in the *Cephalotaxus* and *Pseudotaxus* inflorescences. The perisporangiate *Taxus* microsporangiophore consists of at least two strongly fused hyposporangiate microsporangiophores, which are inserted on a strongly reduced pedicel, so that finally a radial synangium is formed. Thus, no evidences were found, that could explain the anomalous hyposporangiate *Taxus* microsporangiophores as derived from an ancestral perisporangiate, radial condition, as it was suggested in earlier studies (e.g., Dluhosch, 1937; Dupler, 1919; Worsdell, 1901). Quite the contrary, the perisporangiate condition appears to be derived. It is formed by several fused hyposporangiate microsporangiophores, which indicates that in Taxaceae s.l., the hyposporangiate condition is ancestral. Thus, the simple, flower-like *Taxus* pollen cones are probably of pseudanthial origin and the lateral flowers are strongly reduced radial synangia, most likely derived from a fully developed lateral pollen cone (flower) as they are still developed in the *Cephalotaxus* inflorescence.

The relatively homogenous situation in all other coniferous pollen cones, mostly representing simple, unbranched structures consisting of numerous hyposporangiate microsporangiophores, makes the evolutionary transitions suggested here for Taxaceae s.l. impossible. However, searching for such anomalies as described for *Taxus* above or those found in *Pseudotaxus* (Dörken & Nimsch, 2023) might allow new insights into the evolutionary history of coniferous pollen cones, in particular, when these results will be taken in the context of a well-supported conifer phylogeny, ideally including both extant and fossil taxa, as it was done, for example, in Spencer et al. (2015) for the evolution of conifer seed cones. It could also help to provide evidence which could explain also pollen cones in other coniferous groups as possibly derived from an ancestral compound condition.

As this study shows for *Taxus* and *Pseudotaxus*, the vasculature of the microsporangiophores can play a key role in elucidating the origin of pollen cones. When there is more than one vascular bundle supplying the microsporangiophore and each bundle strand enters the common stalk in different layers, it seems highly likely that such microsporangiophores represent a fusion product of several strongly reduced dorsiventral structures and would therefore represent a lateral flower and not a single microsporophyll. There are some useful evidence that may indicate the simple pollen cone structure in other conifers as derived from a pseudanthial origin. For example, in Pinaceae, there are several cases documented where instead of the typical bisporangiate microsporangiophores, multisporangiate ones were found, or even radial microsporangiophores instead of hyposporangiate ones (Dörken, 2017; Dörken & Nimsch, 2015). However, in Pinaceae, the number of microsporangiophores per pollen cone is mostly significantly higher than in Taxaceae s.l., except in *Tsuga*, which is more or less comparable to the number of microsporangiophores in *Taxus* and *Pseudotaxus* (Dörken, 2017; Farjon, 2010). Thus, new insights could result from further investigations in other conifer families (both living and fossil), focusing on the position of the pollen cones in the branching pattern, the position of microsporangiophores in the pollen cones and their vasculature. Also, pollen cone anomalies, which have been mostly entirely neglected, do not inevitably represent random mutations.

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CONFLICT OF INTEREST STATEMENT

The author declares that there is no conflict of interest.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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