Bull. Cupressus Conservation Proj. 11 (1): 52-69 (19.12.2022) **V.M. Dörken**[1] **& H. Nimsch**[2]

Morphology, anatomy and systematics of *Pseudotaxus* (Taxaceae, Coniferales) – *Taxus*-like, but not a *Taxus*

Abstract

Pseudotaxus is an endangered (IUCN status: vulnerable) monotypic Chinese coniferous genus belonging to the Taxaceae. At first glance, the sole species *Pseudotaxus chienii* appears quite similar to *Taxus*: it was first described as *Taxus chienii*. However, there are some distinct morphological features distinguishing *P. chienii* from *Taxus*. Superficially, the white aril is a striking difference to the red or orange *Taxus* aril, but is not the most important difference. More significant in distinguishing them are the branched pollen cones in *Pseudotaxus*. In addition there are numerous foliar features distinguishing both genera. Compared to *Taxus*, the leaves of *Pseudotaxus* are rigid, have a sharply-pointed leaf tip, a free petiole, and the stomatal bands marked by two longitudinal white wax bands with monocyclic stomata, whilst epidermal papillae are absent from the stomatal bands. The vegetative and reproductive structures of *P. chienii* are investigated in regards to species identification and systematics. Depending on the results, *Taxus* and *Pseudotaxus* share some common features but there are also numerous features distinguishing them which justify the treatment as two distinct, but closely related genera.

Introduction

Pseudotaxus W.C.Cheng is a monotypic coniferous genus in the family Taxaceae, comprising only the southeastern Chinese species *Pseudotaxus chienii* (W.C.Cheng) W.C.Cheng (≡ *Taxus chienii* W.C.Cheng), which was discovered in 1934 (ECKENWALDER 2009; FARJON 2010; DÖRKEN & NIMSCH 2018, 2019). It is rarely cultivated outside its natural Chinese habitat. At first glance, its morphology appears very similar to the closely related genus *Taxus* (ECKENWALDER 2009, FARJON 2010, DÖRKEN & NIMSCH 2018, 1019). One of the most striking differences between the genera is the colour of the aril, white in *Pseudotaxus*, red or orange in *Taxus* (ECKENWALDER 2009; FARJON 2010; DÖRKEN & NIMSCH 2018, 2019; DÖRKEN *et al.* 2019). The similarities of *Pseudotaxus* and *Taxus* indicate a close relationship between the genera as is suggested both on morpho-anatomical (e.g. GHIMIRE & HEO, 2014; GHIMIRE *et al.* 2014; ELPE *et al.* 2017, 2018) as well as in genetic studies (e.g. CHENG *et al.* 2000). Thus, it is not too surprising that it was initially treated as *Taxus chienii* W.C.Cheng. In 1947, CHENG created the new genus *Pseudotaxus* for it, an action repeated by FLORIN in 1948 as *Nothotaxus* (*nomen superfluum*), because of differences in the vegetative (epidermis and stomata) as well as in the reproductive structures (colour of the aril and branching pattern of pollen cones).

The major aim of this study is to find out if there are more reliable morpho-anatomic features, that distinguish both genera from each other. Therefore, the morphology and anatomy of leaves and cones of both genders were investigated with different methods.

Material und Methods

Material

Material of *Pseudotaxus chienii* was collected in the private living collection of Hubertus Nimsch, Bollschweil, St. Ulrich, Germany; material of *Taxus baccata* was collected from trees growing in the forests on the campus of the University of Konstanz, Germany.

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). Macrophotography was done with a digital camera (Canon PowerShot IS2) and microphotography with a digital microscope (Keyence VHX 500F) equipped with a

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high-precision VH mounting stand with X-Y stage and bright-field illumination (Keyence VH-S5). For scanning electron microscopy (SEM) analysis, the FAA-material was dehydrated in formaldehyde dimethyl acetal (FDA) for 24 hours (GERSTBERGER & LEINS 1978) and later critical point dried. Sputter coating was done with a Sputter Coater SCD 50 Bal-tec (Balzers). The specimens were examined with an Auriga Zeiss TM.

Special terms

Microsporangiophore: because the identity of the coniferous pollen sac carrying structure is not finally resolved, the terms "sporophyll" or "microsporophyll" are not used, because homologies that are *a priori* applied to it should be avoided. Thus, the neutral term "microsporangiophore" (carrier of the pollen sacs) is used instead.

Microsporangium (pollen sac): structure developed on a microsporangiophore producing the pollen grains.

Pollen cone: the pollen producing "male" cones; in the majority of conifers unbranched structures usually with numerous pollen sacs (microsporangia) carrying structures (microsporangiophores). Pollen cones of all conifers are non-woody and dry out during or shortly after pollen release; dry and empty pollen cones are quickly abscised.

Seed cone: the ovule producing "female" cones. Within all conifers they are compound, inflorescence-like structures or can be regarded as being derived from such once; in the majority of species they become woody. After seed release the empty seed cones of some species remain, at time for several years, on the tree, while in others they abscise, some quickly, some slowly.

Scutellum: small phylloid, distal structure, developed on the stalk of a microsporangiophore.

Cone: the more or less compact reproductive structures of conifers. The "male" cones producing the pollen are called pollen cones, the "female" cones producing the ovules, are called seed cones.

Systematics

The genus *Pseudotaxus* belongs to the Yew family Taxaceae, which also comprises the genera *Taxus* with 7 north hemispheric species, *Amentotaxus* with 5 species distributed in China, Taiwan, India and Vietnam, *Austrotaxus* with a single south hemispheric species in New Caledonia and *Torreya* with 6 species in N-America and SE-Asia. The systematic position of *Cephalotaxus* is still controversial today. Depending on the systematic view, it is part of the Taxaceae (ECKENWALDER 2009; FARJON 2010; LESLIE *et al.* 2012; GHIMIRE & HEO 2014; ELPE *et al.* 2017, 2018; DÖRKEN & NIMSCH 2018, 2019; DÖRKEN *et al.* 2011, 2019) or it is placed in the monogeneric distinct family Cephalotaxaceae (DALLIMORE & JACKSON 1966; PAGE 1990; HAO *et al.* 2008; PAN *et al.* 2011; YI *et al.* 2013; BYKOWSKA & KLIMKO 2018; MAJEED 2019). Cephalotaxaceae has also been treated as a trigeneric family, with *Amentotaxus*, *Cephalotaxus*, and *Torreya* (RUSHFORTH 1987).

Also the systematic position of the Taxaceae has been controversial for a long time. In earlier days it was treated in the distinct order "Taxales" (FLORIN 1948), because the typical coniferous bract/seed scale complex is absent or only hardly visible due to strong reduction of the female cones, in *Taxus* and *Pseudotaxus* often to a single ovule. The results of both numerous morpho-anatomic and molecular phylogenetic studies, show no doubt today, that Taxaceae belongs to the Pinales (syn. "Coniferales"), which are the largest group within living gymnosperms. In most recent phylogenies Taxaceae is placed as sister to the Cypress family Cupressaceae (e.g. CHAW *et al.* 2000; QUINN *et al.* 2002; BURLEIGH & MATTHEWS 2004; DÖRKEN *et al.* 2011; COLE *et al.* 2017).

The Taxaceae *s.str.* are formed by two tribes, 1) Taxeae (*Austrotaxus*, *Taxus* and *Pseudotaxus*) and 2) Torreyeae (*Amentotaxus* and *Torreya*). The genus *Cephalotaxus* is regarded either as sister to the Taxaceae *s.str.* (ELPE *et al.* 2018), or within Torreyeae as sister to *Amentotaxus* and *Torreya* (LESLIE *et al.* 2012; GHIMIRE & HEO 2014); additionally, GHIMIRE & HEO 2014 found *Austrotaxus* as basal, sister to all the rest of the family. Within the Taxaceae, *Taxus* is consistently the sister of *Pseudotaxus* (LESLIE *et al.* 2012; GHIMIRE & HEO 2014; ELPE *et al.* 2017, 2018). The genus *Pseudotaxus* comprises only the SE Chinese species, *P. chienii* (DÖRKEN & NIMSCH 2018, 2019).

Distribution

Pseudotaxus is a Chinese tertiary relict plant (FU *et al.* 1999). It has a scattered distribution pattern, in a quite large geographical area (FU *et al.* 1999; LIU *et al.* 2021) with a focus in the southern parts of the SE-Chinese Province Zhejiang (ECKENWALDER 2009). The distribution in the provinces Guandong, C-Guangxi, NW-Hunan and SW-Jiangxi is sporadic as it is restricted to suitable narrow high ridges. Consequently not only in horticulture, but also in its natural habitat, this species is rare and highly threatened. *Pseudotaxus* occurs in quite diverse habitats with a great heterogeneity of the biotic and abiotic environmental factors (LI 2020; LIU *et al.* 2021). In its main distribution area, there is a humid to subtropical climate, which is strongly influenced by the monsoon. *Pseudotaxus* occurs as single shrub or tree or in small groups disjunct in the understory of deciduous or evergreen mountain forests in about 900-1400 m above sea level. The annual mean temperature is about 12-15°C, the annual mean precipitation 1800-2000 mm, the air humidity about 80% (FU & JIN 1992). In genetic studies it could be shown that the adaptive genetic variation of local populations is significant and responses in most cases to abiotic and biotic stress factors (e.g. LIU *et al.* 2021), which can vary due to the distinct environmental heterogeneities existing between the different populations (LI 2020; LIU *et al.* 2021).

According to the IUCN (International Union for Conservation of Nature and Natural Resources) *Pseudotaxus* belongs to the category Vulnerable.

Morphology and anatomy

Pseudotaxus is an evergreen shrub of mostly just up to four metres in height; only in rare cases is it a small tree up to eight metres. However, these tree-like individuals usually have numerous stems without a dominant leading trunk. The dormancy buds are ovate with a distinct tip. There are numerous dry, brownish spreading bud scales showing a distinct midrib (Fig. 1B; cf. difference to *Taxus*, Fig. 9B), where the buds are inconspicuous, ovate, with green bud scales strongly appressed on each other.

The needle leaves are up to 2.5 cm long and 4.5 mm broad. They are helically set but distichously arranged. When sprouting, the new leaves are yellow-green. They turn to dark green while maturing (Figs 1A & E). The lamina is slightly convex. In comparison to the soft leaves of *Taxus*, the needles of *Pseudotaxus* are markedly ridged, and show a short pointed and acute tip (Fig. 1C; cf. difference to *Taxus*, Fig. 9C). There are two distinct white bands on the abaxial surface, marking the stomata fields (Figs 1A, D-E; cf. difference to *Taxus*, Fig. 9D). *Pseudotaxus* is strictly hypostomatic (Figs 1E & F), and stomata are exclusively developed abaxially, as in all the other Taxaceae (ELPE 2018). There are 23-28 abaxial stomatal rows (cf. difference to *Taxus*, only 13-15), which are arranged in two longitudinal bands. The stomata fields are separated by the raised, green midrib. The stomata fields of *Pseudotaxus* (Figs 1F & 2A) are free of epidermal papillae (in ELPE *et al.* 2017 called cellular protuberances) as in *Austrotaxus* and *Amentotaxus* (ELPE *et al.* 2017), whereas in *Taxus* (Fig. 9E) as well as in *Torreya* numerous epidermal papillae are developed. While in the other taxaceous genera the presence or absence of epidermal papillae is a constant feature throughout each genus, in *Cephalotaxus* there is some variation. In some species papillae are developed, in others they are absent (ELPE *et. al.* 2017). Another important difference between *Pseudotaxus* and *Taxus* is in the type of stomata. In *Pseudotaxus* they are monocyclic, showing just a single ring of isodiametric shaped subsidiary cells surrounding the guard cells of the stomata (Fig. 2B), a feature which is also developed in *Torreya* and *Amentotaxus* (ELPE *et al.* 2017). However, in *Taxus* the stomata are amphicyclic (Fig. 9F), as in *Austrotaxus* and *Cephalotaxus* (ELPE *et al.* 2017). There are four subsidiary cells surrounding the guard cells of the stomata, two elongated lateral ones and two polar ones (Fig. 9F). The short petiole is kneed and strongly appressed to the shoot axis (Fig. 1D), a distinct difference to *Taxus*, which has leaf bases strongly fused to the shoot axis (Fig. 9D). In *Pseudotaxus* the lamina is spreading more or less right-angled from the petiole (Fig. 1D). The leaf is supplied by a single vascular bundle strand (Figs 2C & E), which forms a distinct midrib, visible on both leaf surfaces as a longitudinal ridge. It is more prominently raised on the adaxial surface than on the abaxial surface. A thick cuticle covers the epidermis (Fig. 2D). The epidermis cells are thick walled and rich in phenolic substances (visible as the dark, intensive staining). A hypodermis is absent (Fig. 2D). The mesophyll is dimorphic with palisade parenchyma located towards the upper light exposed surface and spongy parenchyma placed towards the shaded lower surface (Figs 2C & D). In the collateral vascular bundle strand xylem is located towards adaxial and phloem towards abaxial (Fig. 2E). A transfusion tissue consisting of sclerenchymatic cells is well developed on both lateral sides of the vascular bundle strand (Fig. 2E). The leaves of *Pseudotaxus* lack a resin duct below the vascular bundle (Figs 2C & E) as in *Austrotaxus* and *Taxus*. In *Torreya* and *Amentotaxus* (tribe Torreyeae) a distinct resin duct is placed below the bundle strand, as also in *Cephalotaxus* (ELPE *et al.* 2018).

In most general texts or encyclopedias, the Taxaceae are described as dioecious (DALLIMORE & JACKSON 1966; GIFFORD & FOSTER 1989; PAGE 1990; COLE *et al.* 2017; ELPE *et al.* 2017), including *Pseudotaxus* (RUSHFORTH 1987; ECKENWALDER 2009; FARJON 2010). However our own experience based on six investigated individuals of *Pseudotaxus* have shown that the sex expression is not strictly dioecious as mentioned in this literature. The plants we investigated over a period of ten years showed a tendency to be functional either "male" or "female", however with a distinct variation between years. There were "female" plants which developed in one year exclusively seed cones and, in the subsequent year, also pollen cones in addition to seed cones; or in other years, even only just pollen cones. The situation in basically "male" individuals was more or less comparable to that of basically "female" individuals. In one year exclusively pollen cones were produced, in other years additional to pollen cones also some seed cones (and this even on the same branch), and in some years these individuals turned to exclusively "female". A similar phenomenon was also observed for Central European *Taxus baccata*. According to this, the genera *Pseudotaxus* and *Taxus* are not as strictly dioecious as frequently mentioned, because in addition to functional exclusively "male" and "female" individuals, "bisexual" ones also exist, and that sex expression can change markedly between years. These findings are in accordance with KRÜSSMANN (1983), who described *Taxus* as usually dioecious, rarely monoecious. MAERKI (2022) reported about a monoecious *Pseudotaxus* specimen.

As in all other conifers, the reproductive structures of *Pseudotaxus* are arranged in compact cones, the "male", pollen producing cones are called pollen cones, the "female" ovuliferous cones, seed cones. The cones of both sexes are always developed in an axillary position, in the axil of a typical shaped green needle leaf (Figs 2B & D). No terminal cones were observed. The majority of cones are developed on last year's lateral branchlets. They are placed on the lower side of the branchlets in a downward position (Figs 3A, 6A-B). The pollination drops, the receptive structure for pollen grains in seed cones, are always in a strictly downward position.

In *Pseudotaxus*, the pollen cones have a very short stalk, more or less sessile (Figs 3B-C), a difference to *Taxus* with its longer stalked pollen cones (Fig. 10A) The pollen cones consist of numerous pollen sac carrying structures, called microsporangiophores. *Pseudotaxus* microsporangiophores have a perisporangiate structure and consist of a central stalk, a shield-like terminal scutellum and numerous pollen sacs (= microsporangia) developed all around the stalk (Figs 3E-F). At a first glance, the pollen cone structure of *Pseudotaxus* (Fig. 3) and of *Taxus* appears quite similar (Fig. 10A); however there is one important striking difference. In *Taxus* the perisporangiate microsporangiophores are placed directly on the cone axis, and bracts are always absent (Fig. 10A). In *Pseudotaxus* however, the microsporangiophores are always inserted in the axil of a small scaly bract, showing a large skinny hyaline margin and a green central part. Thus, pollen cones of *Taxus* are simple unbranched structures, which correspond in total to a single flower, while *Pseudotaxus* pollen cones are compound and correspond to an inflorescence, and each bract-microsporangiophore-complex to a lateral flower (Fig. 3D; DLUHOSCH 1937; DUPLER 1919; WILDE 1944, 1975; KRÜSSMANN 1983; MUNDRY & MUNDRY 2001; DÖRKEN *et al.* 2011; DÖRKEN & NIMSCH 2016). In expanded morpho-anatomical studies it was found that each axillary perisporangiate microsporangiophore corresponds to a lateral, or axillary pollen cone, however strongly reduced. Thus, it is assumed that the pollen cone structure of *P. chienii* is derived from an ancestor showing, instead of axillary perisporangiate microsporangiophores, fully developed lateral pollen cones, similar to those developed within species of *Cephalotaxus* (DÖRKEN *et al.* 2011; DÖRKEN & NIMSCH 2016). This would explain the strictly axillary position of microsporangiophores in *Pseudotaxus*. If the perisporangiate microsporangiophores represented just simple pollen producing leaves in the sense of "microsporophylls", such an axillary position would not be possible, because ontogenetically a leaf can never be inserted in the axil of a leaf. Leaves are always formed as lateral structures on the shoot apex. In this regard, *Taxus* pollen cones, which are always free of bracts, could represent a progression of the *Pseudotaxus* situation. By a simple reduction of the *Pseudotaxus* microsporangiophore bract, the simple flower-like *Taxus* pollen cone is formed. In this case, the *Taxus* pollen cone is derived from a compound, inflorescence-like ancestor (DÖRKEN *et al.* 2011; DÖRKEN & NIMSCH 2016).

At pollination time, when the pollen grains are released, the bracts within *Pseudotaxus* pollen cones are strongly spreading, so the microsporangiophores – in particular the microsporangia – are best exposed to the ambient airflow, and the pollen grains can be efficiently shed from the cones. In immature pollen cones, the bracts are imbricate and cover the microsporangiophores entirely. Because *Pseudotaxus* pollen cones are not developed in a bud, the bracts within the cone carry out the protective function of the missing bud scales to protect the developing microsporangiophores (Fig. 3C). Thus, externally, only the bracts are visible during the early development of the cone.

The non-saccate pollen grains are spheroidal but slightly irregular in shape (Fig. 5), they are about 25-30 µm in diameter. The outer surface of the pollen grain (exine) is, as typical for Taxaceae, microverrucate-orbiculate. The density of the globose to oval orbicules is very high (Fig. 5). Earlier studies on pollen grains in Taxaceae have shown that they are useful to distinguish between the taxaceous geners, depending on the pollen grain size and the surface structure of the exine. BYKOWSKA & KLIMKO (2018) who investigated the pollen grains of *Cephalotaxus*, *Torreya* and *Taxus*, showed that pollen grains of *Cephalotaxus* and *Torreya* are significantly larger than these of *Taxus* and have a distinctly lower density of orbicules. In *Taxus* they were smaller and had many more orbicules. They also demonstrated that within *Taxus*, the exine sculpturing was so similar that it is not a good diagnostic feature for species identification. Interestingly, the structure of the exine sculpturing in *Pseudotaxus* is quite similar to *Taxus*. Compared to the images of *Taxus* presented in Figs 3-5 in BYKOWSKA & KLIMKO (2018), it seems that the density of orbicules is slightly higher in *Pseudotaxus* (Fig. 5). Comapring with the data on *Taxus* pollen grain size in PILCHER (1968) and BYKOWSKA & KLIMKO (2018), *Pseudotaxus* pollen grains, about 25-30 µm in diameter, are more or less the same size as in *Taxus*.

Due to the strong reduction of the seed cone, a distinct bract/seed scale complex (as is typical in many other coniferous groups, e.g. Pinaceae or Sciadopityaceae) is absent or hardly visible in Taxaceae (MUNDRY 2000; GHIMIRE *et al.* 2014). Mature seed cones of *Pseudotaxus* are 5-8 mm long and 4-5 mm broad. There are 7-8 pairs of imbricate decussate membranaceous scale leaves at the stalk of the seed cone, similar to the situation in *Taxus* (DÖRKEN *et al.* 2019). At pollination time, these scale leaves surround the ovule nearly entirely, so that only the micropyle remains free. These scale leaves are persistent, and remain below the aril in mature cones as green scales (Figs 7A-B). In *Taxus*, however they dry out and become brownish (Fig. 10D). In most cases there is just a single ovule per seed cone in *Pseudotaxus* (Figs 7A-B); two ovules are exceptional (Fig. 6D). The ovules are supplied by two collateral vascular bundle strands (Fig. 6E). At maturity, they are surrounded by a fleshy white, cup-like aril (Figs 6A-B & 7B-E; cf. difference to *Taxus*, aril of all species are red or orange, Fig. 10C). The aril and the seed are free and not fused to each other. The mature aril is as long as the seeds or slightly longer. The distal part of the cup-shaped aril has an unlobed distal collar (Fig. 7D), as is also typical for *Taxus* (Fig. 10C). In the beginning, the aril is initially thin and green. As the seed cones mature, the aril becomes fleshy and changes to white. The aril formation starts after pollination time, and the first time that the juvenile aril is visible, about 2-3 months after pollination, it is still flat and greenish, visible between the scale leaves (Fig. 6a). After the aril has exceeded the scale leaves, the further development progresses faster.

Our investigations of *Pseudotaxus* seed cones – in particular about the origin of the aril – have shown that the aril is formed by the uppermost pair of scale leaves developed on the stalk of the cone (Fig. 6F). In the earliest ontogenetic stages, the primordia of these leaves are physically in contact and fuse subsequently to form a ring-like structure, which later grows apically and surrounds the seed cup-like and becomes fleshy. A detailed study is presented in DÖRKEN *et al.* 2019. In some rare cases these two lateral primordia forming the aril do not become fused or only fuse partly to each other, so that the mature aril consists of 2 free halves or a cup like aril with a distinct two-lobed collar (Fig. 7E). Such arils are also described in DÖRKEN *et al.* (2019) and MAERKI (2022). The formation of the aril represents an adaptation to seed dispersal by birds, as in *Taxus* (FARJON, 2007; DÖRKEN & HETZEL 2017; DÖRKEN *et al.* 2019). Due to this, the mature seed cones of *Pseudotaxus* and *Taxus* are structurally quite similar to each other. In both, the aril is strongly fleshy, cup-like and not fused to the seed. In *Pseudotaxus* the surface of the aril is smooth and weakly covered with short cylindrical cellular protuberances (Fig. 7F); in *Taxus* there are numerous densely developed irregularly shaped cellular protuberances (Fig. 10E). As in *Taxus*, the cones mature in the year of pollination.

The seeds are ovate and brownish and 0.5-1 cm in length (Fig. 8A). There is a large ovate hilum at the base of the seed. The hilum is surrounded by a swollen collar like ring (Fig. 8B). Even on mature seeds the micropyle is still visible (Figs 8A & C). The testa is characterised by longitudinal cells showing distinctly raised cell walls (Fig. 8D). In *Taxus*, the testa is smooth with numerous small flat holes (Fig. 10F).

Conclusion

In summary, *Pseudotaxus* and *Taxus* appear quite similar to each other at first glance, but numerous distinct distinguishing characteristics exist which are summarised in Table 1 (p. 59). From the results gained in the present study, the treatment of *Pseudotaxus* as a distinct monotypic genus is justified. Despite the fact that there are numerous distinct morpho-anatomical differences between *Taxus* and *Pseudotaxus*, there are also numerous common features which both taxa share. Thus, we strongly support the systematic treatment of the two genera as distinct taxa but as closely related sister groups in the tribe Taxeae.

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Bibliography

BURLEIGH, J. G. & S. MATHEWS (2004). Phylogenetic signal in nucleotide data from seed plants: implications for resolving the seed plant tree of life. *Amer. J. Bot.* 91: 1599-1613.

BYKOWSKA, J. & M. KLIMKO (2018). Pollen morphology of selected species of the Cephalotaxus Sieb. et Zucc., Taxus L. and Torreya Arn. genera. *Acta Biol. Cracov., Ser. Bot.* 60(2).

CHAW, S.M., C.L. PARKINSON, Y. CHENG, T.M. VINCENT & J.D. PALMER (2000). Seed plant phylogeny inferred from all three plant genomes: Morphology of extant gymnosperms and origin of Gnetales from conifers. *PNAS* 97: 4086-4091.

CHENG, W.C. (1934). Enumeration of Chekiang Plants III. *Contr. Biol. Lab. Sci. Soc. China, Bot. Ser.* 4: 240.

CHENG, W.C. (1947). *Res. Notes Forest. Inst. Natl. Centr. Univ. Nanking, Dendrol. Ser.* 1: 1.

CHENG, Y., R.G. NICOLSON, K. TRIPP & S.M. CHAW (2000). Phylogeny of Taxaceae and Cephalotaxaceae genera inferred from chloroplast matK gene and nuclear rDNA ITS region. *Mol. Phylogen. Evol.* 14: 353-365.

COLE, T.C., J.B. BACHELIER & H.H. HILGER (2017). Tracheophyte phylogeny poster - Vascular plants: systematics and characteristics. *PeerJ Preprints*.

DALLIMORE, W. & A.B. JACKSON (1966). *A handbook of Coniferae and Ginkgoaceae*. Arnold: London.

DLUHOSCH, H. (1937). Entwicklungsgeschichtliche Untersuchungen über die Mikrosporophylle der Koniferen. *Biblioth. Bot.* 114(3): 1-24.

DÖRKEN, V.M. & H. NIMSCH (2019). *Morphology and Identification of Coniferous Genera*. Verlag Kessel: Remagen-Oberwinter.

DÖRKEN, V.M. & H. NIMSCH (2018). *Differentialdiagnostik in Koniferen - Ein illustrierter Gattungsschlüssel*. Verlag Kessel: Remagen-Oberwinter.

DÖRKEN, V.M. & I. HETZEL (2017). Taxus baccata - Europäische Eibe (Taxaceae). *Jahrb. Bochumer Bot. Ver.* 8: 298-305.

DÖRKEN, V.M., H. NIMSCH & P.J. RUDALL (2019). Origin of the Taxaceae aril: Evolutionary implications of seed-cone teratologies in Pseudotaxus chienii. *Ann. Bot.* 123: 133-143.

DÖRKEN, V.M., Z.X. ZHANG, I. MUNDRY & TH. STÜTZEL (2011). Morphology and anatomy of male reproductive structures in Pseudotaxus chienii (W.C. Cheng) W.C. Cheng (Taxaceae). *Flora* 206(5): 444-450.

Dörken, V.M. & H. Nimsch (2016). Some new aspects about the evolution of pollen cones and perisporangiate microsporangiophores in Taxaceae. *Bull. Cupressus Conserv. Proj.* 5: 3-21.

Dupler, A.W. (1919). Staminate strobilus of Taxus canadensis. *Bot. Gaz.* 68: 345-366.

Eckenwalder, J.E. (2009). *Conifers of the world*. Timber Press: Portland.

Elpe, C., P. Knopf, Th. Stützel & C. Schulz (2017). Cuticle micromorphology and the evolution of characters in leaves of Taxaceae s.l. *Bot. J. Linn. Soc.* 184(4): 503-517.

Elpe, C., P. Knopf, Th. Stützel & C. Schulz (2018). Diversity and evolution of leaf anatomical characters in Taxaceae s.l. - Fluorescence microscopy reveals new delimitating characters. *J. Pl. Res.* 131(1): 125-141.

Farjon, A. (2007). *A natural history of conifers*. Timber Press: Portland.

Farjon, A. (2010). *A Handbook of the World´s Conifers*. Brill: Leiden & Boston.

Florin, R. (1948). On Nothotaxus, a new genus of the Taxaceae, from Eastern China. *Acta Hort. Berg.* 110: 385-395.

Fu, L.I. & J.M. Jin (1992). *China plant red data book - Rare and endangered plants*. Vol. 1. Science Press: Beijing.

Fu, L.G., N. Li & R.R. Mill (1999). Taxaceae. *In:* Wu, ZY & Ph. Raven (eds.). *Flora of China*. Vol. 4. Beijing & St. Louis: Science Press & Missouri Botanical Garden Press; p. 89-98.

Gerlach, D. (1984). *Botanische Mikrotomtechnik, eine Einführung*. 2nd ed. Stuttgart,Thieme.

Gerstberger, P. & P. Leins (1978). Rasterelektronenmikroskopische Untersuchungen an Blütenknospen von Physalis philadelphia (Solanaceae). *Ber. Deutsch. Bot. Ges.* 91: 381-387.

Ghimire, B. & K. Heo (2014). Cladistic analysis of Taxaceae s.l. *Pl. Syst. Evol.* 300: 217-223.

Ghimire, B., C. Lee & K. Heo (2014). Leaf anatomy and its implication for phylogenetic relationships in Taxaceae s.l. *J. Pl. Res.* 127: 373-388.

Gifford, E.M. & A.S. Foster (1989). *Morphology and Evolution of Vascular Plants*. 3rd ed. Freeman & Company: New York.

Hao, D.C., P.G. Xiao, B. Huang, G.B. Ge & L. Yang (2008). Interspecific relationships and origins of Taxaceae and Cephalotaxaceae revealed by partitioned Bayesian analyses of chloroplast and nuclear DNA sequences. *Pl. Syst. Evol.* 276(1): 89-104.

IUCN (2001). *Red List of Threatened Species*, Version 3.1; http://www.iucnredlist.org/search.

Krüssmann, G. (1983). *Handbuch der Nadelgehölze*. 2. Aufl. Parey: Hamburg & Berlin.

Leslie, A.B., J.M. Beaulieu, H.S. Rai & S. Mathews (2012). Hemisphere-scale differences in conifer evolutionary dynamics. *PNAS* 109 (40): 16217–16221; supplementary data.

Li, S., Z. Wang, Y. Su & T. Wang (2021). EST- SSR- based landscape genetics of Pseudotaxus chienii, a tertiary relict conifer endemic to China. *Ecology and Evolution* 11: 9498-9515.

Liu, L., Z. Wang, Y. Su, & T. Wang (2021). Population transcriptomic sequencing reveals allopatric divergence and local adaptation in Pseudotaxus chienii (Taxaceae). *BMC genomics* 22(1): 1-21.

Majeed, A., A. Singh, S. Choudhary & P. Bhardwaj (2019). RNAseq-based phylogenetic reconstruction of Taxaceae and Cephalotaxaceae. *Cladistics* 35(5): 461-468.

Maerki, D. (2022). Observation of a monoecious Pseudotaxus chienii specimen. *Bull. Cupressus Conservation Proj.* 11: 70-71.

Mundry, I. (2000). Morphologische und morphogenetische Untersuchungen zur Evolution der Gymnospermen. *Biblioth. Bot.* 152.

Mundry, I. & M. Mundry (2001). Male cones in Taxaceae s.l. - An example of Wettstein's Pseudanthium concept. *Pl. Biol.* 3: 405-416.

Page, C.N. (1990). Gymnosperms. *In:* Kubitzki, K. (ed.), *The families and genera of vascular plants*. Vol. 1. Springer: Berlin, Heidelberg, New York.

Pan, H.W., Y.R. Guo, Y.J. Su & T. Wang (2011). Development of microsatellite loci for *Cephalotaxus oliveri* (Cephalotaxaceae) and cross-amplification in Cephalotaxus. *Amer. J. Bot.* 98(8): e229-e232.

Pilcher, J.R. (1968). Some applications of scanning electron microscopy to the study of modern and fossil pollen. *Ulster Journal of Archaeology* 31: 87-91.

Quinn, C.J., R.A. Price & P.A. Gadek (2002). Familial concept and relationships in the conifers based on rbcL and matK sequence comparisons. *Kew Bull.* 57: 513-531.

Rushforth, K. (1987). *Conifers*. Christopher Helm, London.

Wilde, M.H. (1944). A new interpretation of coniferous cones: I. Podocarpaceae (Podocarpus). *Ann. Bot.* 8: 1-41.

Wilde, M.H. (1975). A new interpretation of microsporangiate cones in Cephalotaxaceae and Taxaceae. *Phytomorph.* 25: 434-450.

Yi, X., L. Gao, B. Wang, Y.J. Su & T. Wang (2013). The complete chloroplast genome sequence of Cephalotaxus oliveri (Cephalotaxaceae): evolutionary comparison of Cephalotaxus chloroplast DNAs and insights into the loss of inverted repeat copies in gymnosperms. *Genome biology and evolution* 5(4): 688-698.

Table 1: Distinguishing characters between *Taxus* and *Pseudotaxus*.

feature	***Taxus***	***Pseudotaxus***
Bud		
Shape	ovate or roundish (Fig. 9B)	ovate, pointed tip (Fig. 1B)
bud scales	green, strongly appressed (Fig. 9B)	brown, dry & spreading (Fig. 1B)
needle leaf		
structure	soft	Rigid
adaxial colour	dark green (Fig. 9A)	shiny dark green (Figs 1A, B & E)
abaxial colour	bright green (Figs 9A & D)	2 longitudinal white wax bands (Figs 1A, D & E)
Tip	not acute (Fig. 9C)	pointed and acute (Fig. 1C)
Petiole	decurrent on the green shoot (Fig. 9D)	not decurrent on the brown [non.photosynthetic] shoot (Fig. 1D)
Base	adnate to the shoot axis (Fig. 9D)	appressed to the shoot axis (Fig. 1D)
stomata		
Type	amphicyclic (Fig. 9F)	monocyclic (Fig. 2B)
total number of rows	13-15	23-28
papillae between stomata	present (Fig. 9E)	absent (Figs 1F, 2A)
pollen cone		
structure	simple (Fig. 10A)	compound (Fig. 3D)
Stalk	long (Fig. 10A)	very short (Figs 3B & D)
bracts in the pollen cones	absent (Fig. 10A)	present (Fig. 3D)
seed cone		
scale leaves on stalks of mature cones	dry & brownish; apex rounded (Fig. 10D)	green; apex acute (Fig. 7B)
Seed		
Colour	black	brown
Testa	smooth with numerous small holes (Fig. 10F)	longitudinal cells with raised walls (Fig. 8D)
Aril		
Colour	orange or red (Figs 10C & D)	white (Figs 7B-E)
surface	numerous irregularly shaped cellular protuberances (Fig. 10E)	few short cylindric cellular protuberances (Fig. 7F)

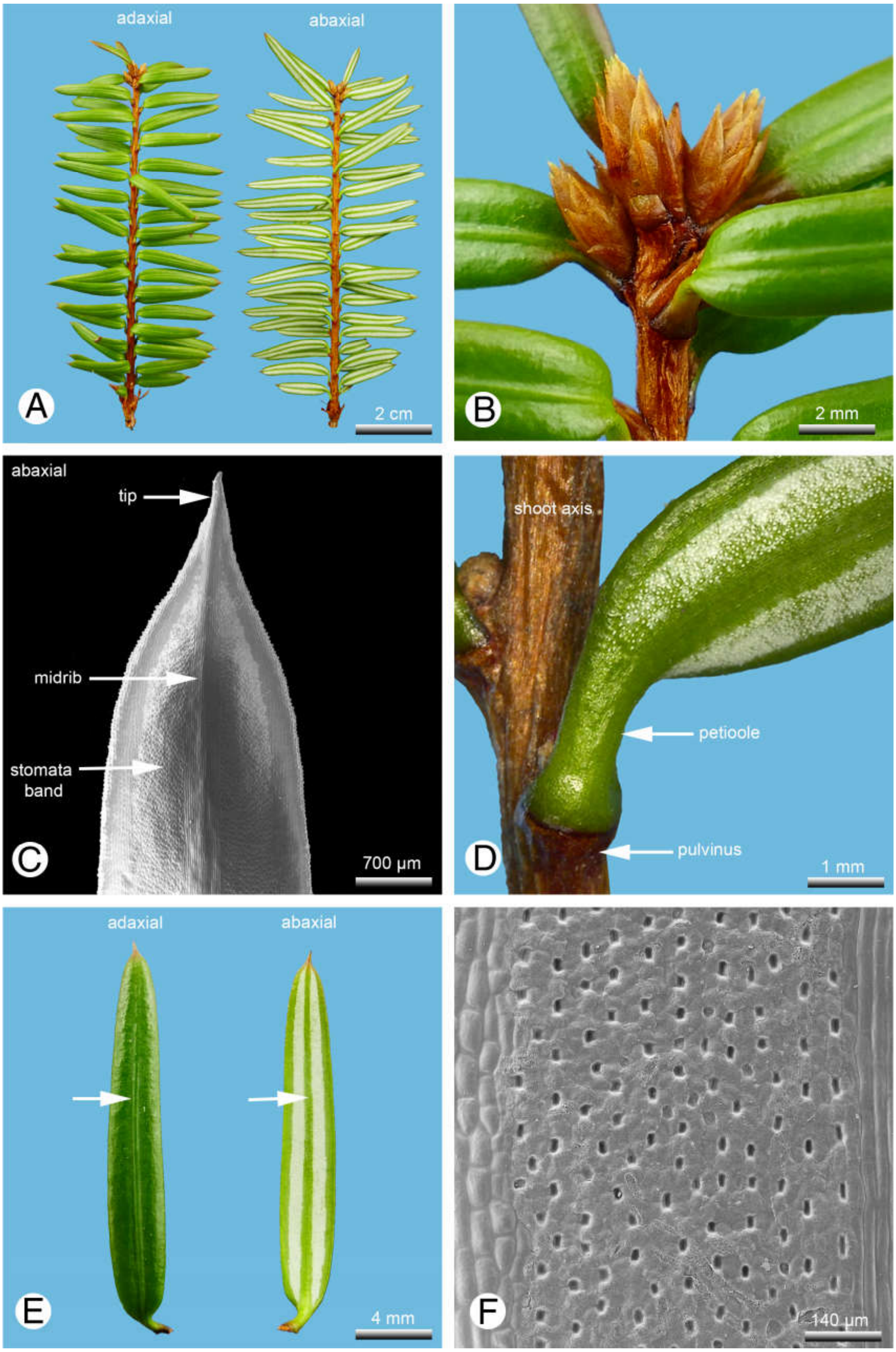

Fig. 1: *Pseudotaxus chienii*. Vegetative structures; morphology of shoots and leaves.

A: Ad- and abaxial view of a last year’s shoot. **B:** Vegetative buds. **C:** Leaf tip pointed and acute. **D:** Leaf base. **E:** Ad- and abaxial view of a leaf showing a well developed midrib on both sides. **F:** Abaxial stomata field.

Images: V.M. DÖRKEN

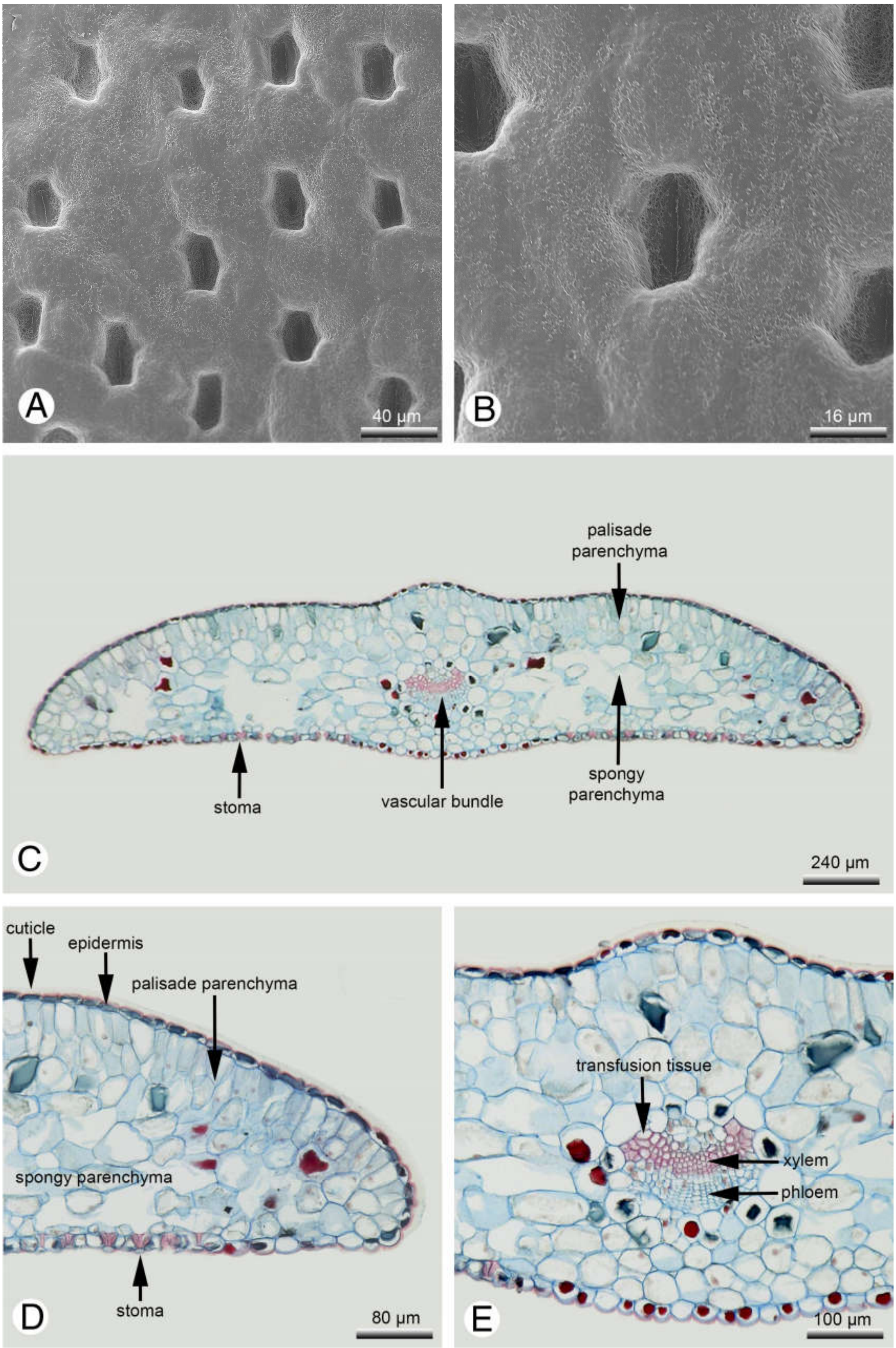

Fig. 2: *Pseudotaxus chienii*. Vegetative structures; morphology and anatomy of leaves.

A: Abaxial stomata fields showing no epidermal papillae. **B:** Monocyclic stoma. **C:** Cross section of a leaf showing the bifacial structure. **D & E:** Details of C. **D:** Detail of the different leaf tissues. **E:** Detail of the vascular bundle.

Images: V.M. DÖRKEN

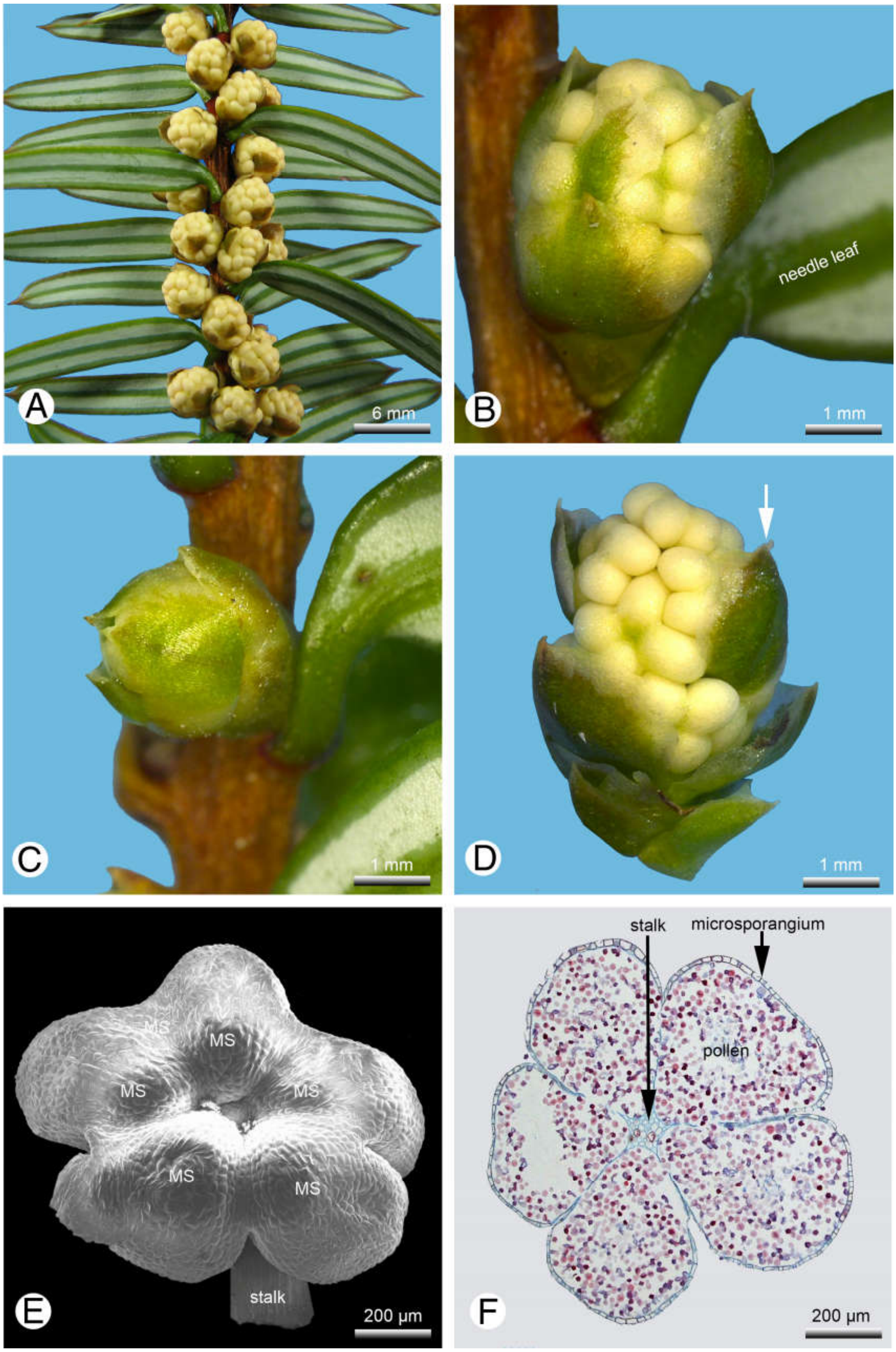

Fig. 3: *Pseudotaxus chienii*. Reproductive structures: morphology and anatomy of pollen cones.

A: Abaxial pollen cones. **B:** Pollen cones axillary. **C:** Immature pollen cone; the microsporangiophores are covered by their bracts; bud scales are absent. **D:** Mature pollen cone; bracts of the microsporangiophores are spreading; **E:** Perisporangiate microsporangiophore (SEM-image); **F:** Cross section of a perisporangiate microsporangiophore.

Images: V.M. DÖRKEN

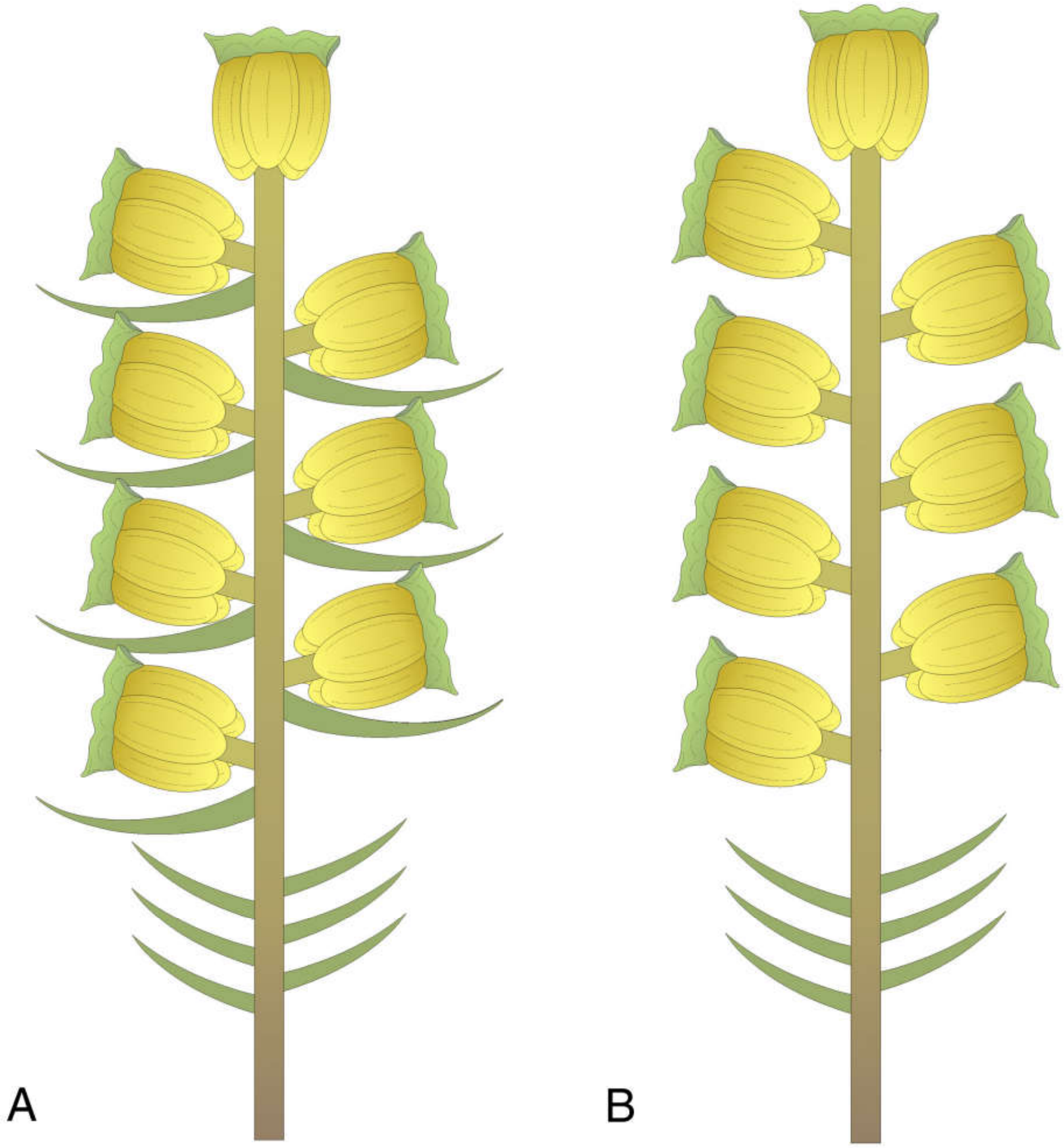

Fig. 4: Schematic drawing of pollen cones.

A: Compound pollen cone of *Pseudotaxus chienii*, the microsporangiophores are inserted in the axil of a scaly bract; **B:** Simple pollen cone of *Taxus baccata*; bracts within the cone are absent.

Drawings: V.M. DÖRKEN

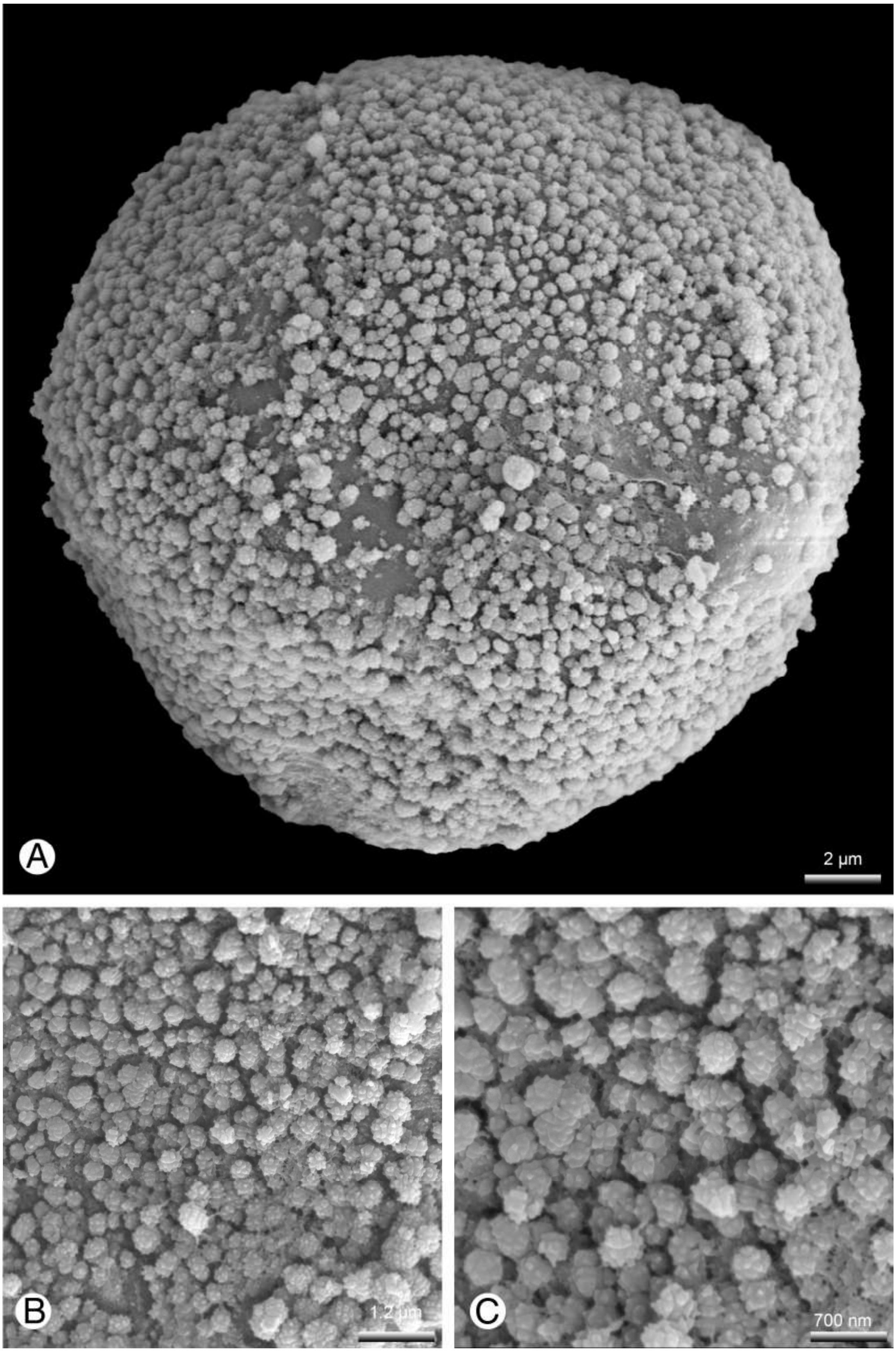

Fig. 5: *Pseudotaxus chienii*. Reproductive structures; morphology of pollen grains (SEM-images).

A: Pollen grain. **B & C:** Pollen grain with microverrucate-orbiculate exine sculpturing showing a very high density of global to oval orbicules (SEM-image).

Images: V.M. DÖRKEN

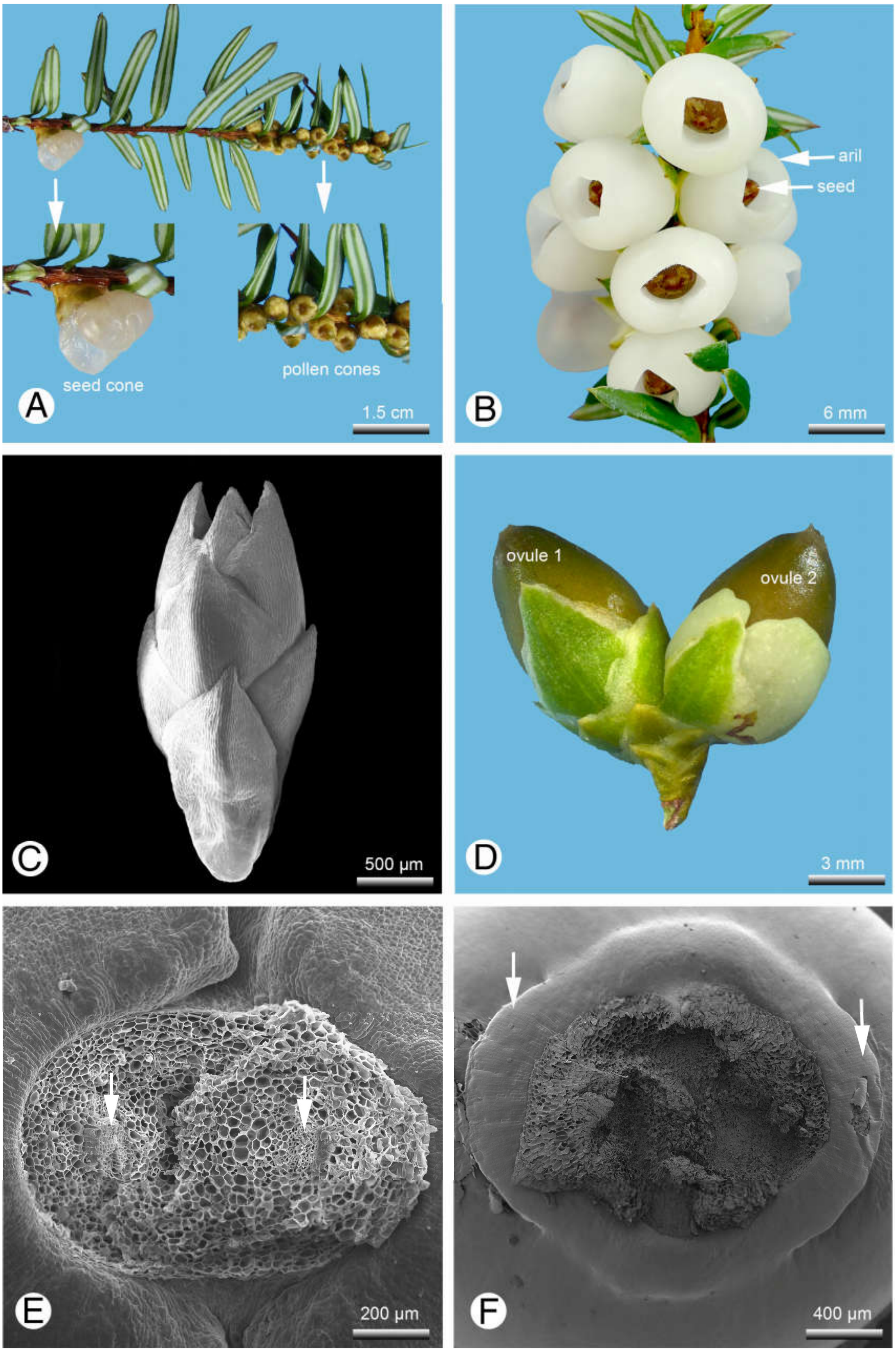

Fig. 6: *Pseudotaxus chienii*. Reproductive structures; morphology of seed cones.

A: Last year´s shoot with a basal seed cone and several distal pollen cones. **B:** Abaxial seed cones. **C:** Immature seed cone; the ovule is covered by the scale leaves of the stalk. **D:** Seed cone with two instead of the typical single ovule. **E:** Basal part of an ovule showing two vascular bundles (arrows) supplying the ovule (SEM-image). **F:** The aril is formed by two lateral primordia (arrows) developed below the ovule (SEM-image).

Images: V.M. DÖRKEN

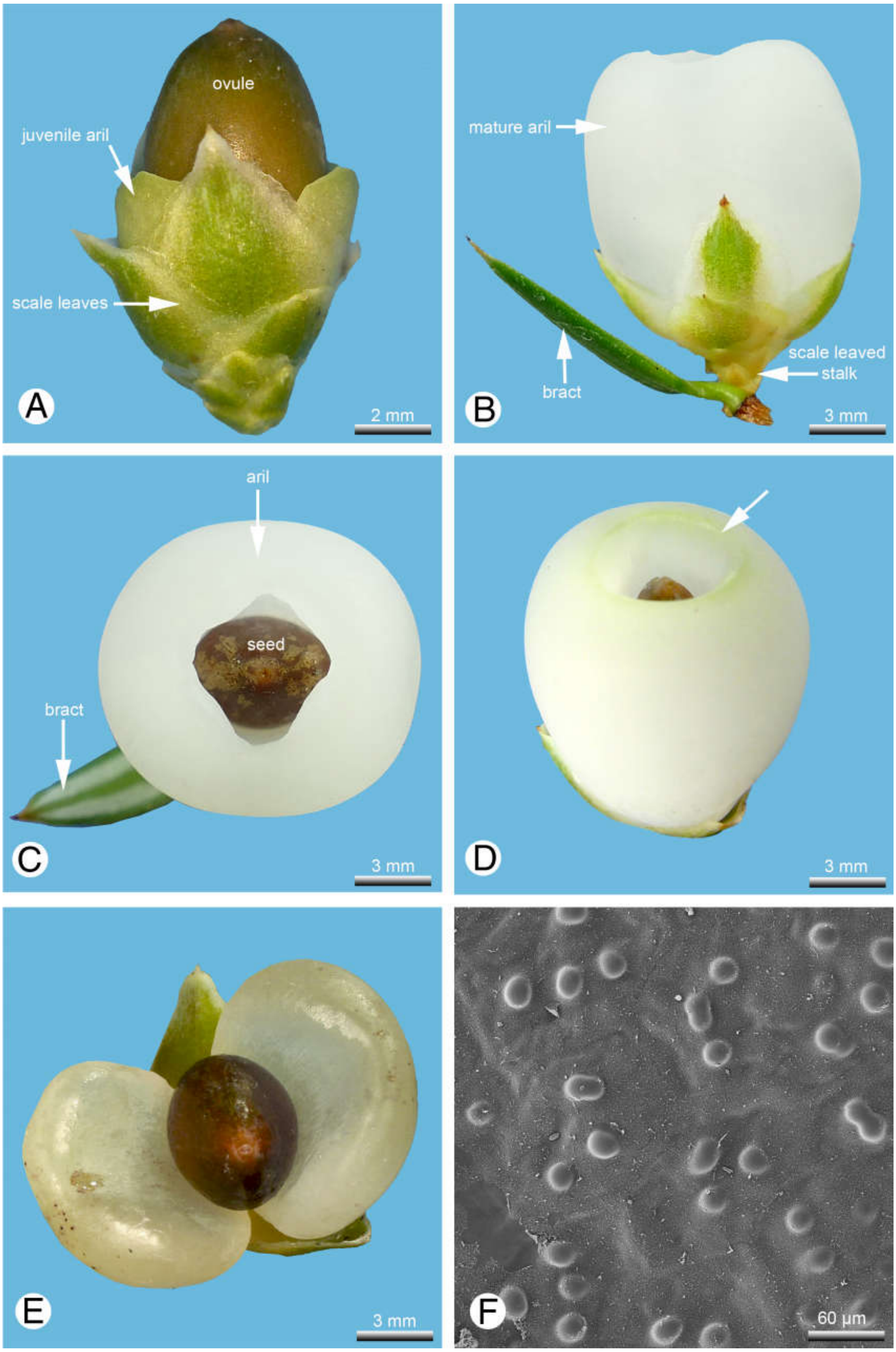

Fig. 7: *Pseudotaxus chienii*. Reproductive structures; morphology of seed cones.

A: Immature seed cone; about three month after pollination the aril is visible the first time. **B-E:** Mature seed cones. **B:** Lateral view. **C:** Top view; seed and aril not fused. **D:** Lateral view; aril with an unlobed distal collar. **E:** Anomalous seed cone; aril consists of two free halves. **F:** Microsculpturing of the outer aril surface (SEM-image).

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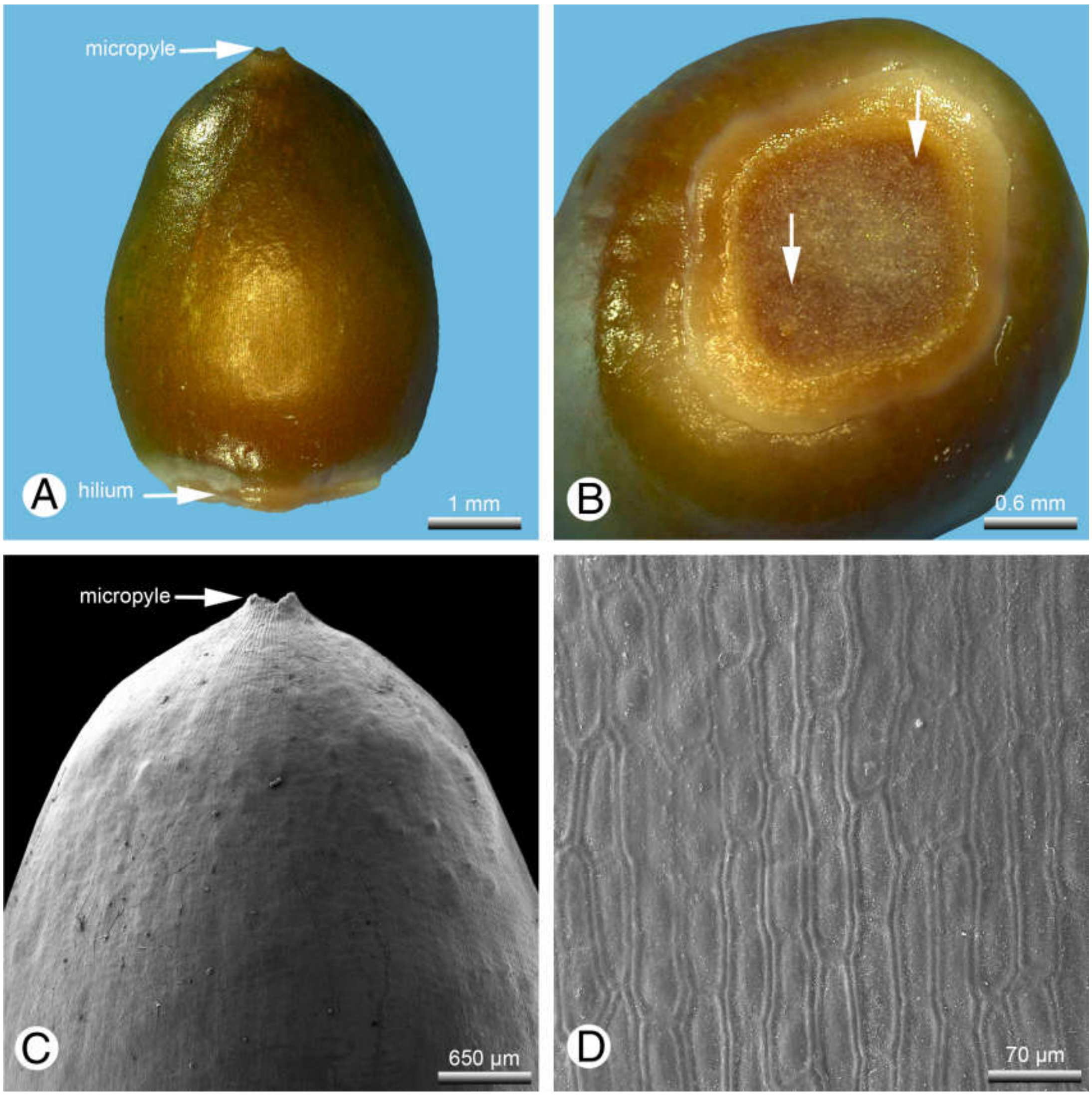

Fig. 8: *Pseudotaxus chienii*. Reproductive structures; morphology of seeds.

A: Mature seed in lateral view. **B:** Hilium; there are two collateral vascular bundles supplying the seed. **C:** Distal part of the seed showing the micropyle (SEM-image). **D:** Microsculpture of the testa (SEM-image).
Images: V.M. DÖRKEN

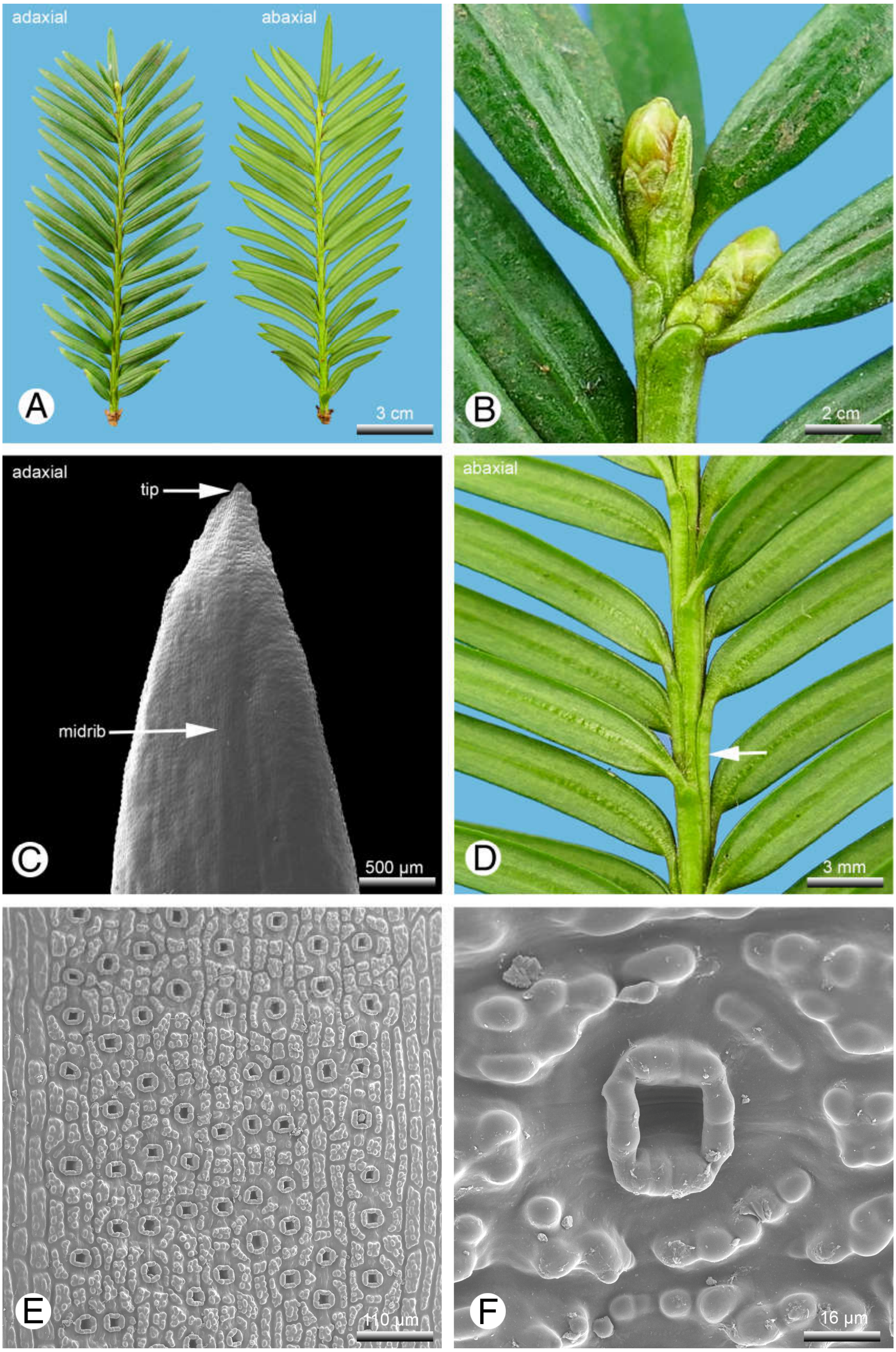

Fig. 9: *Taxus baccata*. Distinguishing characters of *Taxus* and *Pseudotaxus*.

A: Ad- and abaxial view of a last year's shoot; abaxial leaf surfaces without conspicuous wax markings. **B:** Vegetative buds roundish and appressed. **C:** Leaf tip pointed but not acute. **D:** Leaf bases fused to the stem. **E:** Stomata placed between numerous epidermal papillae. **F:** Amphicyclic stomata.

Images: V.M. DÖRKEN

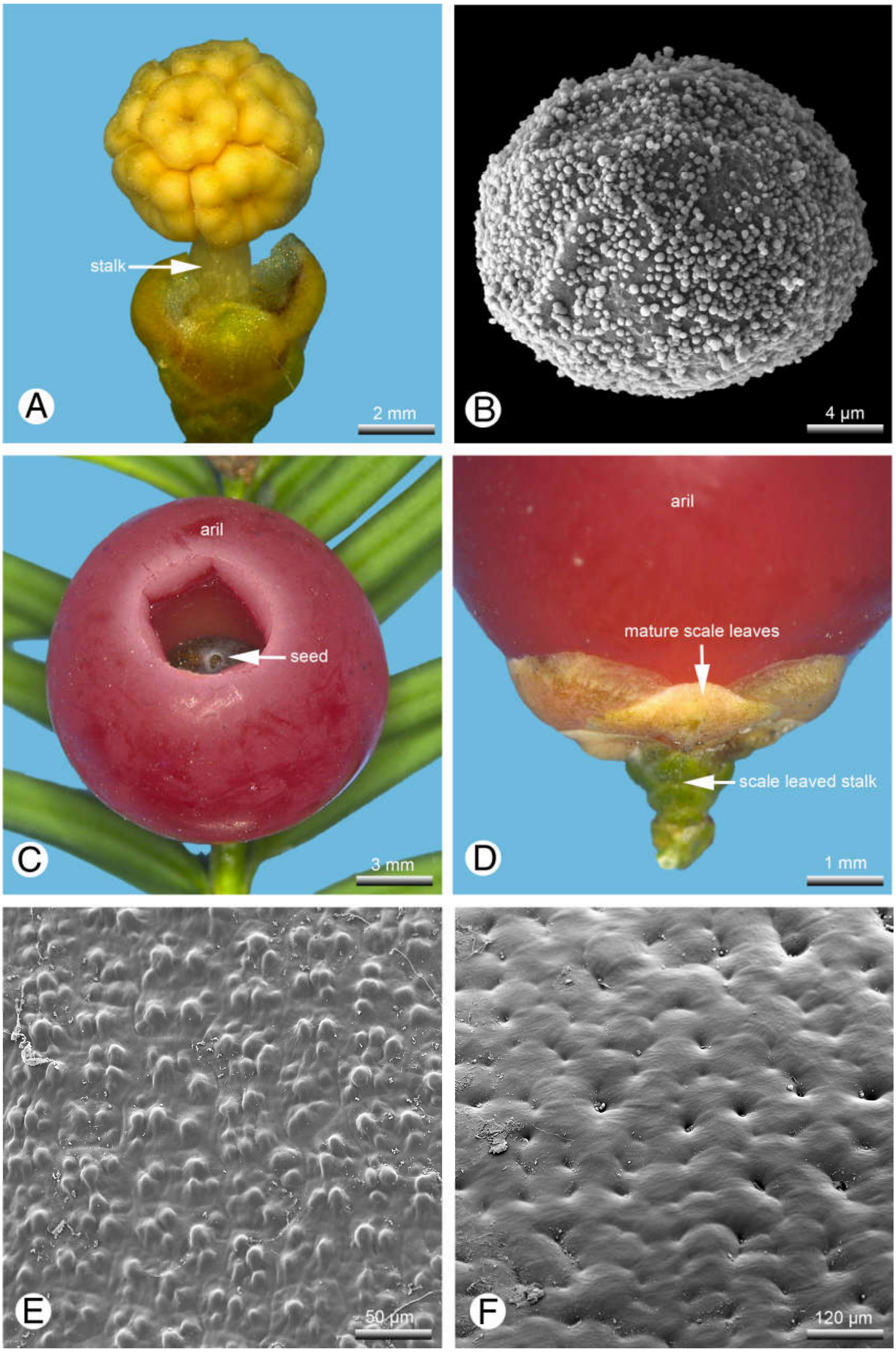

Fig. 10: *Taxus baccata*. Distinguishing characters of *Taxus* and *Pseudotaxus*.

A: Pollen cone long stalked and simple; bracts within the pollen cone absent. **B:** Pollen grain with microverrucate-orbiculate exine sculpturing (SEM-image). **C:** Mature seed cone with red aril. **D:** At maturity the scale leaves on the stalk of the seed cone are dry and brown. **E:** Surface of the aril with numerous densily arranges epidermal papillae (SEM-image). **F:** Surface of the testa smooth with numerous small holes (SEM-image).

Images: V.M. DÖRKEN