Short Communication

Pollen of *Ceratostema* Jussieu (Ericaceae, Vaccinieae): tetrads without septa

Abstract The pollen morphology of two species of the Neotropical genus *Ceratostema* (Ericaceae) was examined by light, scanning and transmission electron microscopy. The *Ceratostema* species examined have 3-colporate pollen grains united in permanent tetrahedral tetrads that show a common condition encountered in the Ericaceae. But the septal exine was absent between two neighboring grains in each pollen tetrad of *Ceratostema*. The pollen tetrads without septa are the first report for the Ericaceae as well as other angiosperm families.

Key words *Ceratostema*, pollen morphology, tetrad without septum

Introduction

Ceratostema Jussieu (Ericaceae: Vaccinioideae, Vaccinieae) is a genus composed of about 33 species of Neotropical blueberries, ranging from southern Colombia to northern Peru, with a disjunct species in Guyana (Lutyen 2002). Previously, pollen morphology of two *Ceratostema* species has been studied under only the scanning electron microscope (Lutyen 1978; Maguire et al. 1978), and was described as having tetrahedral tetrads, with viscin threads absent. The tetrad diameter for one species; *C. glandulifera*, was 22 – 23 µm, with an exine sculpture that was microverrucate to microrugulate with aperture margins and distal poles somewhat psilate.

Compound pollen grains in the form of dyad, triad, tetrad, pseudomonad, polyad, or pollinium are not an uncommon phenomenon and occur in more than 56 families of the

angiosperms (Erdtman 1945; Knox and McConchie 1986). Of these, at least 52 families produce compound pollen in the form of tetrad (Copenhaver 2005). Hitherto, the pollen tetrads without septa have not been reported in any of these families. During a detailed study on pollen morphology of the tribe Vaccinieae, we have discovered the pollen tetrads without septa in two species of *Ceratostema*: *C. lanigerum* and *C. loranthiflorum*.

Materials and methods

Pollen grains of two species of *Ceratostema* were examined with the light microscope (LM) and scanning electron microscope (SEM), and with the transmission electron microscope (TEM) for *C. loranthiflorum*. Polliniferous materials were provided from the dried herbarium specimens of the Herbarium of Swedish Museum of Natural History, Stockholm, Sweden (S). Voucher information for the specimens examined is given below:

- ***C. lanigerum*** (Sleum.) Luteyn Ecuador: Prov. Napo-Pastaza, Mera, in rastrijo, 1100m alt. 21.12.1955. E. Asplund 18937 (S)
- ***C. loranthiflorum*** Benth. Ecuador: Prov. Loja, Cerro Villonaco, 20 km to Catomyo, 2600m alt. 22.05.1967. B. Sparre 16627 (S)

Pollen preparation methods for LM, SEM, and TEM follow the protocols described in Sarwar et al. (2005). Pollen slides of both collections are deposited at the Hokkaido University Museum, Japan. Descriptive terminology follows Praglowski and Grafström (1985) and Punt et al. (1994).

Results

In LM, the acetolysed pollen grains of *Ceratostema* are minute and oblate, united in compact tetrahedral tetrad (Fig. 1a, b); they are homodynamosporus, i.e., all four pollen grains of the tetrad are of same size; the viscin threads are absent; the tetrad diameter is (26.4–) 29.9 – 31.5 (–33.0) µm; the polar and equatorial diameter of individual pollen grain is (13.5–) 15.3 – 16.4 (–20.1) µm and (20.6–) 21.9 – 24.0 (–26.4) µm, respectively. Pollen grains are 3-aperturate, with the apertures arranged according to "Fischer's Law", i.e., the apertures form in pairs at six points in the tetrad. The apertures are colporate, colpus distinct, (19.8–) 20.7 – 21.7 (–23.1) µm long in total, (0.3–) 1.0 (– 2.0) µm wide; the ora are lalongate, i.e., the endoaperture is transversely elongated. The apocolpial exine is (0.5–) 1.1 – 1.5 (–2.0) µm thick, the septum absent (Fig. 1a, b), and the exine sculpture verrucate to rugulate or sometimes psilate.

In SEM, exine sculpture along the colpi is similar to that of appearing at distal pole (Fig. 1c–e), but at the mesocolpial exine, it has a tendency to decrease in lateral extension of the rugulae with more distinct unit (Fig. 1d, g). The apocolpial exine sculpture is coarsely rugulate-psilate, with the rugulae faintly striate (Fig. 1f), or the exine sculpture is coarsely rugulate, with the rugulae finely to clearly striate (Fig. 1h). The colpus membrane is granular to granuloid.

In TEM, no septum was found between the neighboring individual pollen grains (Fig. 2a, b), although the boundaries of cytoplasm of each grain were not distinct within a tetrad as the pollen sample was of herbarium origin. But the aborted and convoluted, vestigial septal exine (Fig. 2c–e) was found at the equatorial region of original individual pollen grain. The vestigial septal exine is connected with columellae, with the foot layer and endexine of the outer exine wall covering the whole tetrad. The apocolpial exine is composed of thick ektexine: tectum (T), columellae (rod-like element distinct) (C), foot layer (F), and thin endexine (En) with higher

electron density than ektexine. The ectexine is ca. 1.5 – 1.7 µm, and a total exine is ca. 1.9 – 2.2 µm in thickness. Intine (I) is usually thin, showing lower electron density than the endexine, but sometimes is thick near vestigial septal exine (Fig. 2b-c).

Discussion

Our pollen morphological observations on *Ceratostema* largely agree with the earlier descriptions (Lutyen 1978; Maguire et al. 1978), except for the absence of septa between neighboring grains in each pollen tetrad. The septum has not been observed in pollen tetrads of both *Ceratostema lanigerum* and *C. loranthiflorum* under LM (Fig. 1a, b), which was later confirmed under TEM (Fig. 2a–e). One of the reasons for the lack of reference to the absence of a septum in previous studies on *Ceratostema* tetrads (Lutyen 1978; Maguire et al. 1978) may be that the pollen tetrads were observed by only SEM, but not under either LM or TEM.

These specialized pollen tetrads of *Ceratostema* are probably due to the abortion of the septum between two neighboring grains during the late developmental stages of permanent tetrads. The aborted septal exine composed of both the ektexine and the endexine (Fig. 2c–e) may support the supposition. On the other hand, there is a possibility of pseudomonad pollen of these two *Ceratostema* species. In the pseudomonad pollen, only one pollen grain is fully developed, and the other three are aborted during ontogenetic development. The TEM photographs of the aborted septal exine in the *Ceratostema* tetrads (Fig. 2c–e) shows a similar appearance to "the collapse of the proximal wall" in the S-type monad (= pseudomonad) of *Leucopogon fraseri* (Figs. 30 and 31 in McGlone 1978) of the tribe Styphelieae in the same family Ericaceae (Kron et al. 2002). But under LM, the *Styphelia* type (S-Type) shows the monad-like pollen with three vestigial pollen grains (Fig. 1 in McGlone 1978). The pollen of *Ceratostema* appears to have the standard

homodynamosporus tetrahedral tetrads (Fig. 1a–e), so the present pollen should not be the pseudomonad.

Warner and Chinnappa (1986) postulated that the evolutionary trend in pollen dispersal unit within Ericales is from tetrad to monad. The pollen tetrad without septa is probably another evolutionary trend from standard pollen tetrads (with septa). The tetrads without septa of these two *Ceratostema* species are probably the most derived pollen character state within Vaccinioideae. The small tetrad size of *Ceratostema* pollen may be the necessary, but not the sufficient conditions for the absence of the septum as we have found relatively smaller pollen tetrads with well developed septa in some other genera within Ericaceae (Sarwar and Takahashi, unpublished data).

The mature pollen tetrads without septa are the first report for any angiosperm families as well as the family Ericaceae. A detailed ontogenetic study is obviously needed for better understanding and clarification of this special palynological feature of the pollen *Ceratostema* tetrads in Ericaceae.

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Legends

Fig. 1. Pollen grains of *Ceratostema* species united in tetrads. **a, c–d, f–g** *C. lanigerum*, **b, e & h** *C. loranthiflorum*. **a**–**b** Three grains and three apertures noticed in each pollen tetrad. LM. **c**–**e** Pollen tetrad. SEM. **f & h** Apocolpial exine sculpture. SEM. **g** Mesocolpial exine sculpture. SEM. **c, e–f & h** Polar view. **d & g** Equatorial view.

Fig. 2. Palynological features of *Ceratostema loranthiflorum* by TEM. **a**–**b** Pollen tetrad showing aperture region (arrow) and aborted septal exine (asterisk). **c**–**e** Detail of aborted and convoluted septal exine. Exine composed of thick ektexine; tectum (*T*), columellae (*C*), and foot layer (*F*), and thin endexine (*En*); *I* intine.

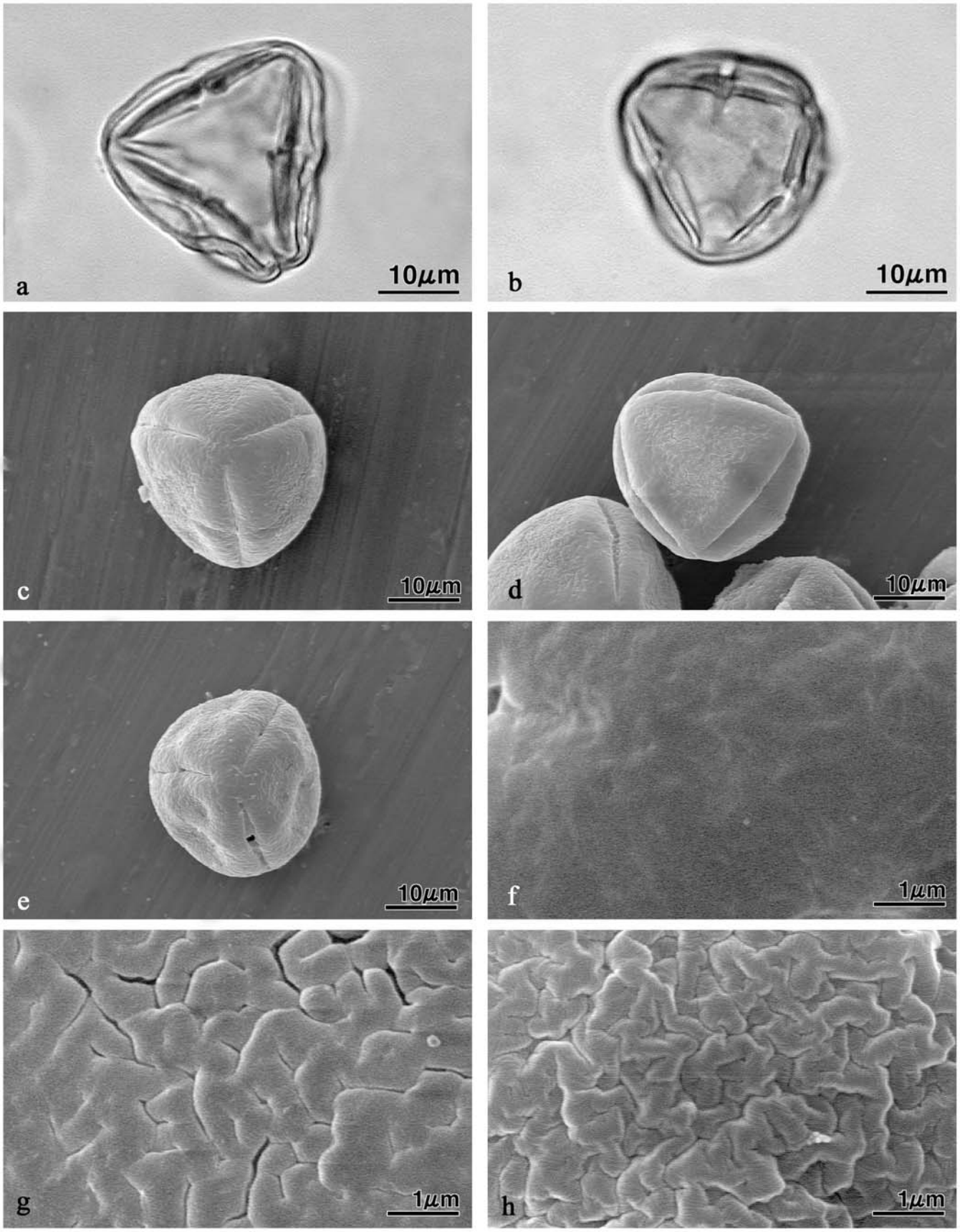

Fig. 1.

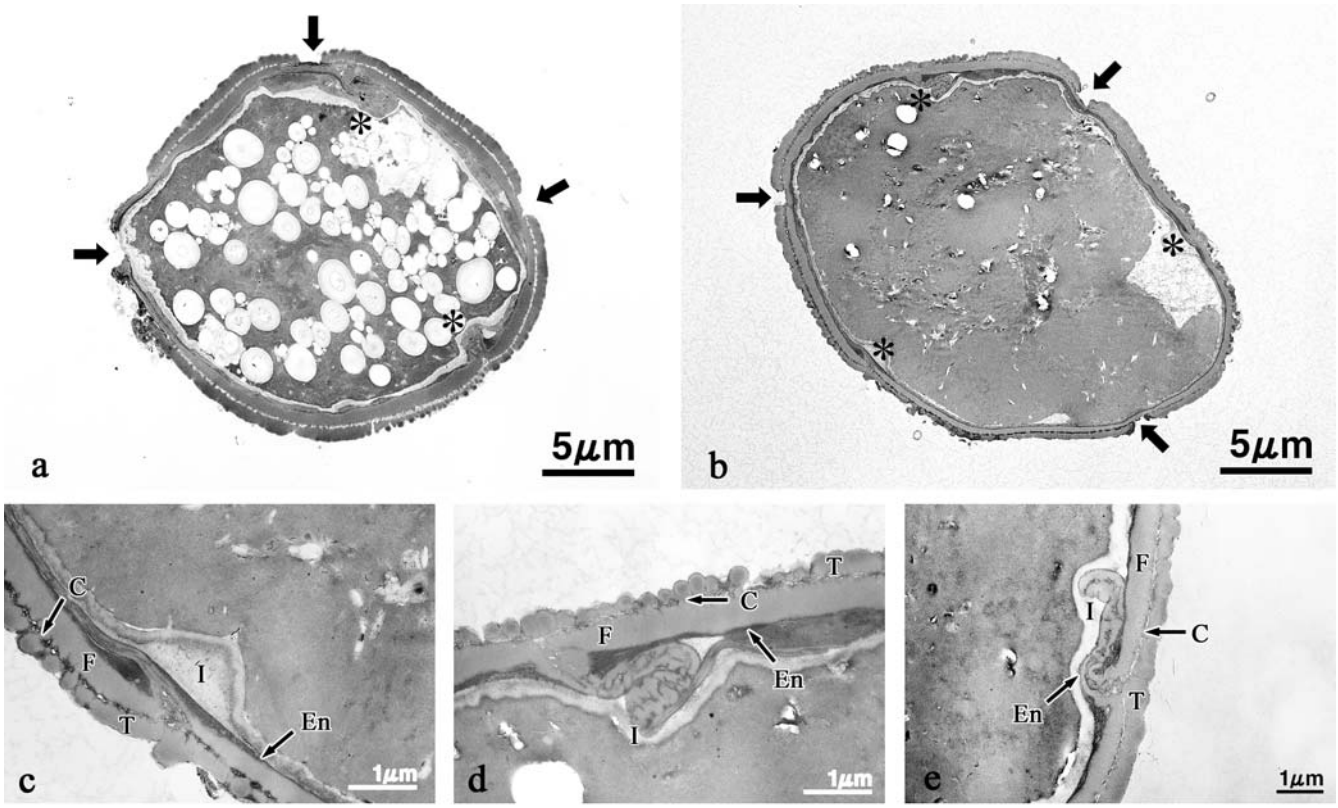

Fig. 2.