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MICROMORPHOLOGY AND ANATOMY OF THE FLORAL ELEMENTS OF *Tradescantia* x *andersoniana* W. LUDW. ROHWEDER

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Abstract

The genus *Tradescantia* comprises about 70 species. In Poland *Tradescantia x andersoniana* is basically grown as an ornamental plant that is recommended for borders and to be planted around garden ponds. The present study investigated flowering as well as the micromorphological and anatomical features of some floral elements of *Tradescantia x andersoniana* W. Ludw. Rohweder 'Karin'. The macro- and micromorphology of the flowers was examined using stereoscopic, light, and scanning electron microscopy (SEM). Spiderwort produces flowers with a diameter of 4.6 – 5 cm, which open early in the morning and close at noon. Non-glandular and glandular hairs grow on the abaxial surface of the calyx and on the apical part of the ovary. The glandular hairs develop a several-celled stalk and a unicellular spherical or elongated head. The staminal filaments produce chain-shaped trichomes. Striate cuticular ornamentation is found on their surface and on the epidermis covering the perianth. The striae on the cells of the stamen hairs run to the two poles of the cell. The pistil develops a wet stigma with unfused unicellular papillae. The cuticle on their surface is smooth, whereas on the style near the stigma it forms dense folds.

Key words: *Tradescantia x andersoniana*, flowers, anatomy stamen, pistil, trichomes, stomata, micromorphology, SEM

INTRODUCTION

Spiderwort (*Tradescantia*) belongs to the family Commelinaceae (H a r d y and S t e v e n s o n , 2000b). This genus includes about 70 species (F a d e n , 1998) that are native to the southern states of the USA and Mexico (D a h l g r e n et al. 1985). Spiderworts are found in their natural environment in America. Some of them are used as ornamental house and border plants. White spiderwort (*T. albiflora* Kurth.) is one of the most known decorative interior plants (Ś w i d z i ń s k a , 1998). The literature describes taxa of the genus *Tradescantia* which have allergenic effect, inducing allergic reactions in the form of itchy face or throat, reddening of the conjunctiva as well as lip swelling and dyspnoea (W ü t h r i c h and J o h a n s s o n , 1997). In Brazil *T. diuretica* is used in medicine as a diuretic and in the treatment of hemorrhoids. *T. andersoniana* is a hybrid bred by crossing *T. ochinensis* Raf. x *T. subaspera* Ker–Gawl. x *T. virginiana* L. (E r h a r d t et al. 2002). This species reaches a height of 40-70 cm. The colour of the corolla in the flowers of this taxon varies depending on the variety: it can be white, red, or blue (D a h l g r e n et al. 1985; C h m i e l , 2000). Perennials of the genus *Tradescantia* like moist sites and they are recommended to be planted on borders and around garden ponds (C h m i e l , 2000).

The aim of the present study was to conduct observations of the flowering process and to determine the micromorphological and anatomical features of some floral elements of spiderwort.

MATERIALS AND METHODS

The flowers of *Tradescantia x andersoniana* W. Ludw. Rohweder 'Karin' were collected for examination at the Botanical Garden of the Maria Curie-Skłodowska University in Lublin. The macro- and micromorphology of the flowers were examined at two growth stages (bud break and full bloom) using stereoscopic (SM), light (LM), fluorescence (FM) and scanning electron microscopy (SEM). Initial observations of portions of fresh flowers were performed in a stereoscopic microscope equipped with a Nikon Coolpix 4500 camera. The structure of the hairs, perianth and generative elements was investigated in a Nikon Eclipse 400 light microscope.

Semi-permanent glycerol-mounted and semi-thin sections were prepared. Live cell staining was performed with neutral red, Lugol's iodine, and Sudan III. Auramine was also used to view the cross sections under a fluorescence microscope, in which the tissues were also compared under autofluorescence conditions. The micromorphology of the epidermis covering the surface of the perianth, stamens, ovary and stigma was examined in a Tescan VEGA II LMU scanning electron microscope (SEM).

The flowers at the initial flowering stage, sampled for SEM examination, were fixed in 4% glutaraldehyde and in 0.1M phosphate buffer with a pH of 7.0. Next, this material was postfixed in 1% OsO_4 for 1.5 hours and stained in a 0.5% aqueous solution of uranyl acetate. After dehydration in alcohol and acetone series, the fixed sections were critical-point dried in liquid CO_2 and coated with gold.

To make semi-thin cross sections of the perianth, the upper part of the ovary and the stigma of the pistil as well as of the staminal filaments, the plant material that had been fixed and dehydrated in ethanol was embedded in Spurr's low viscosity resin. The semi-thin sections with a thickness of 0.8 - 1 μm were cut on a Reichert Ultracut S microtome. The slides were stained with 1% toluidine blue.

RESULTS

Tradescantia x andersoniana starts flowering in the third decade of May and finishes in September; it produces flowers with a diameter of 4.6-5 cm that are clustered in terminal umbels. The flowers open early in the morning and close at noon, but when the weather is cloudy, the time of blooming is longer. The perianth produces three outer green sepals performing the role of the calyx and three inner coloured corollas (Figs 1A; 2A).

The flowers are borne on long (1.7-2.1 mm) anthocyanin-coloured pedicles, 1.3 mm in diameter. In the cross-sectional view of the flower stalk, two layers of parenchymal cells can be seen under the epidermis; in some of these cells calcium oxalate crystals are found in the form of raphides or single needles. The chlorenchyma that is composed of 4-6 layers is located deeper. Collateral vascular bundles, surrounded by the parenchyma (Fig. 1B), are situated in the central part.

Calyx. The sepals, 1-1.2 wide and 1.3-1.5 cm long, reach the half length of the petals. Numerous, long (0.9-3.5 mm), sharp-pointed, non-glandular hairs grow on the abaxial surface of the epidermis. They are composed of 2-7 single, elongated, living cells arranged in rows and narrowed at the apex, which show a high degree of vacuolization (Fig. 1C, D, G).

The glandular trichomes are found less frequently in the abaxial epidermis and they consist of a several-celled (2-5) stalk and a unicellular head of different shapes: spherical (Fig. 1H), cylindrically elongated (Fig. 1I), or elongated with a slight narrowing at the base (Fig. 1J, K). A thick cytoplasm and a large cell nucleus are visible in the glandular cell, as well as oil in the subcuticular space (Fig. 1H-K). The stalk cell adjacent to the base of the hair has a thicker cell wall compared to the higher located cells, which is their reinforcement (Fig. 1H, I). The elongated cells of the stalk were characterized by a large nucleus and vacuolization of the protoplast, which decreased in the direction of the head (Fig. 1H-K). The height of these trichomes was within the range of 198-236 μm.

Amaryllidaceae-type tetracytic stomata occur between the trichomes. The pores are located at the level of other epidermal cells (Fig. 1C-F) or slightly below (Fig. 1O). A striated cuticle or wax with irregular fluffy structures is visible on the surface of the epidermis (Fig. 1F).

The cells of the sepals at the apical part (cross section) form 4-6 layers (Fig. 1L-N) with a thickness ranging from 128 to 236 μm. The walls of the adaxial epidermis exhibit autofluorescence (Fig. 1M). The outer wall is thicker (6.5 μm) than the other walls (3.5 μm). Numerous plastids are present in the mesophyll cells (Fig. 1L, M); some of them are red coloured in the light of a UV – 2A filter.

The corolla forms slightly elongated, pink-violet petals that are twice larger than the sepals. Their width and length are within the range of 2.1-2.4/2.5 -2.8 cm, respectively. The adaxial epidermis is composed of elongated cells (Fig. 2B, C) that are covered by striate cuticular ornamentation. The densely packed striae, which run along the longer cell axis, have a corrugated contour (Fig. 2D, E). When viewed in cross section, the striae are visible in the form of teeth (Fig. 2F, H). Conical cuticular projections are found at some places on both surfaces of the epidermis; an odorous substance probably accumulates underneath them (Fig. 2F, G).

3 layers of cells: the lower and upper epidermis as well as one layer of parenchyma (Fig. 2G, H), can be seen in the apical part of the petal (cross section). The thickness of these tissues is 47-52 μm, but the petal is about 2.5 times thicker at the place where the vascular bundles run (Fig. 2I). The radial walls of the epidermal cells form narrow, tubular or irregular, horizontal ingrowths (Fig. 2F-I). Their protoplast is vacuolized and has a large cell nucleus.

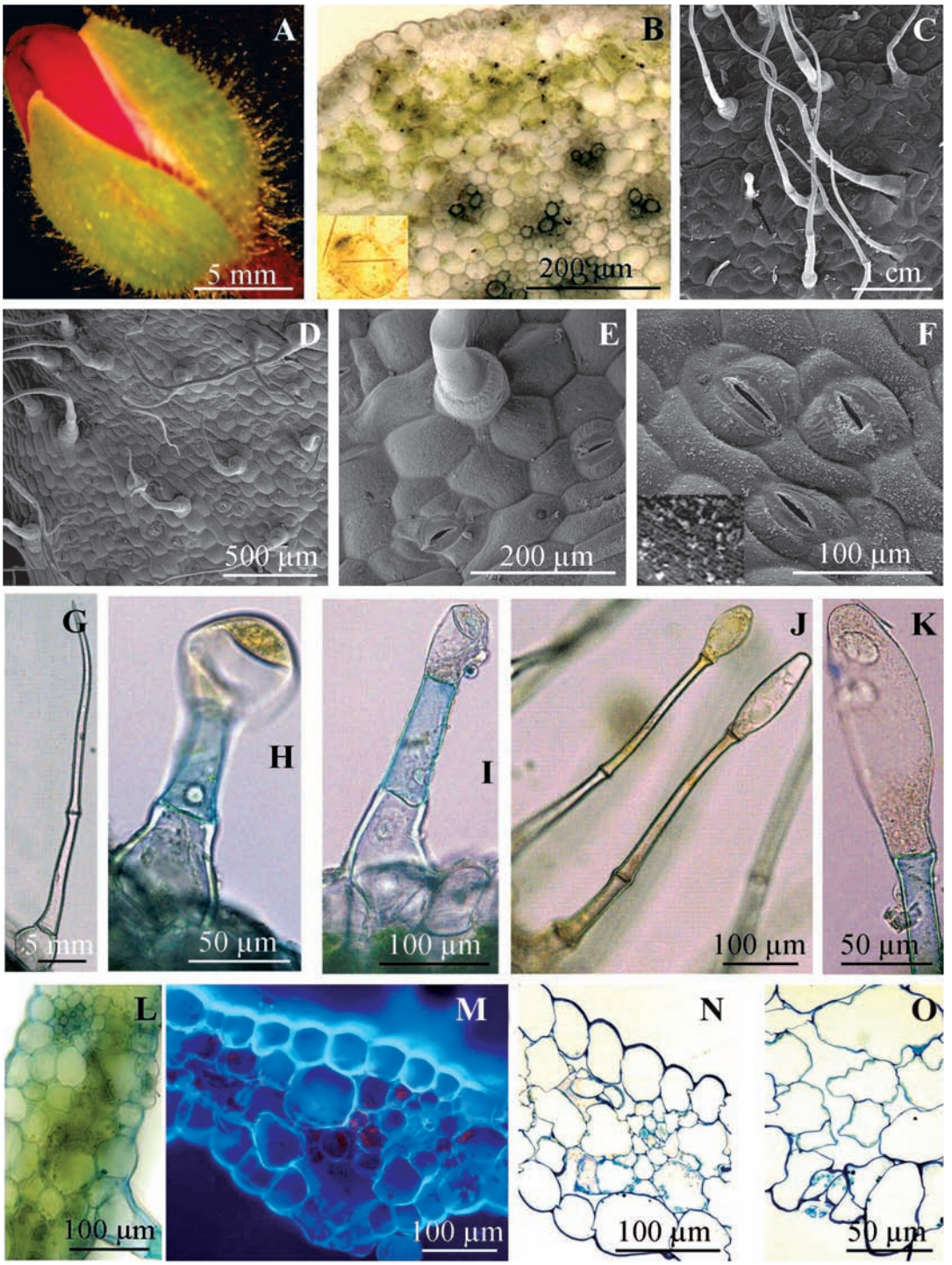

Fig. 1. A-I – Floral elements of *Trandescantia andersoniana*.
A – Bursting flower bud,
B – Cross-sectional view of the flower stalk,
C-F – Abaxial epidermis of the calyx; visible are several-celled, non-glandular and glandular (arrow) trichomes as well as stomata,
G – Non-glandular hair,
H-K – Glandular trichomes on the abaxial surface of the epidermis; there can be seen the varying shape of the glandular cell, a large nucleus and the oil in the subcuticular space,
L-O – Cross-sectional view of the calyx; visible are the thick outer wall of the adaxial epidermis (L - M) and plastids in the mesophyll cells as well as a stoma (N).
A – (SM); B,G-L, N, O – (LM); C-F – (SEM); M – (FM).

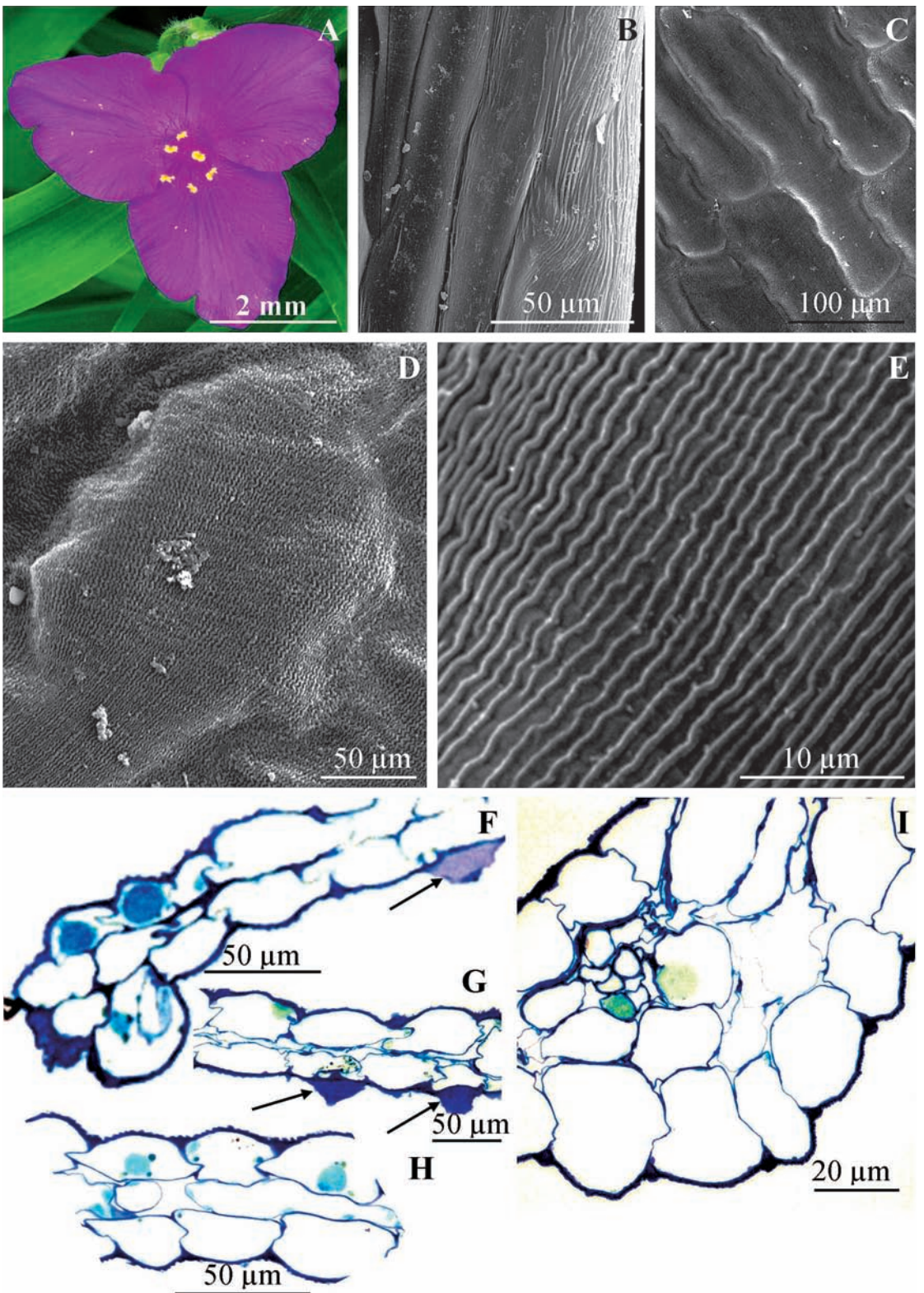

Fig. 2. A – Flower of *Trandescantia andersoniana,*
B-I – Parts of the corolla,
B-E – Adaxial surface of the epidermis; the cuticular striae arranged along the longer cell axis can be seen, (SEM),
F-I – Cross-sectional views of the petal; the following can be seen: the edge (F); the cells between the veins (G, H) and at the place of the vein (I); the contour of the cuticular striae (H) and the thicker strands of cuticle at the place where the cells meet; a conical projection of the cuticle on the surface of the epidermal cells (arrow), (LM).

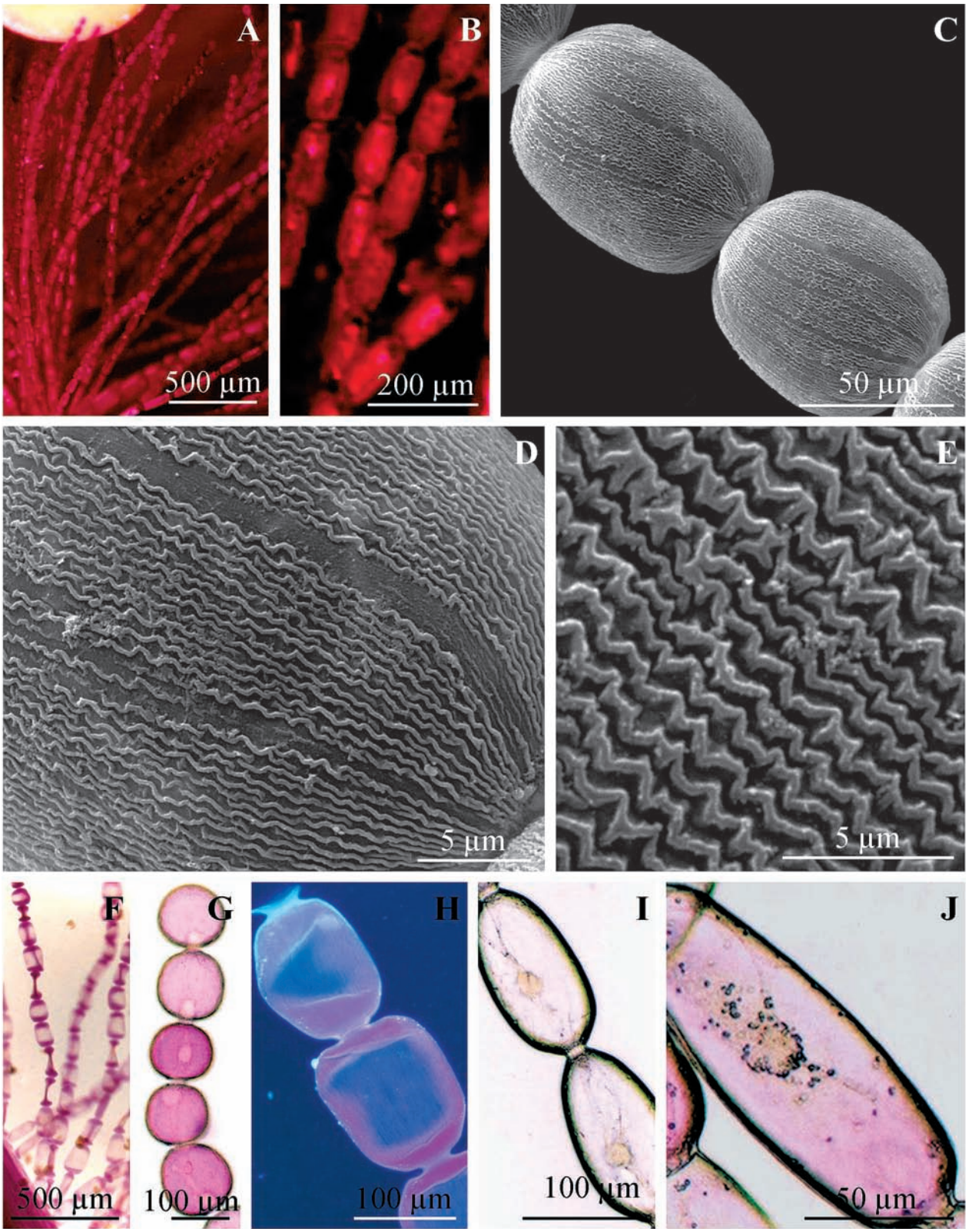

Fig. 3. A-L – Hairs growing out of the epidermis of the stamen filaments of *Trandescantia andersoniana*:
A-C – Cells of the chain-shaped hairs,
D, E – Surface portion of the hair cells; visible are the cuticular striae with a wavy arrangement of the striae,
F-J – Different-shaped hair cells with a thick layer of cytoplasm (H) and a large cell nucleus (G, I, J).
A, B, F, G – (SM); C-E – (SEM); H – (FM); I, J – (LM).

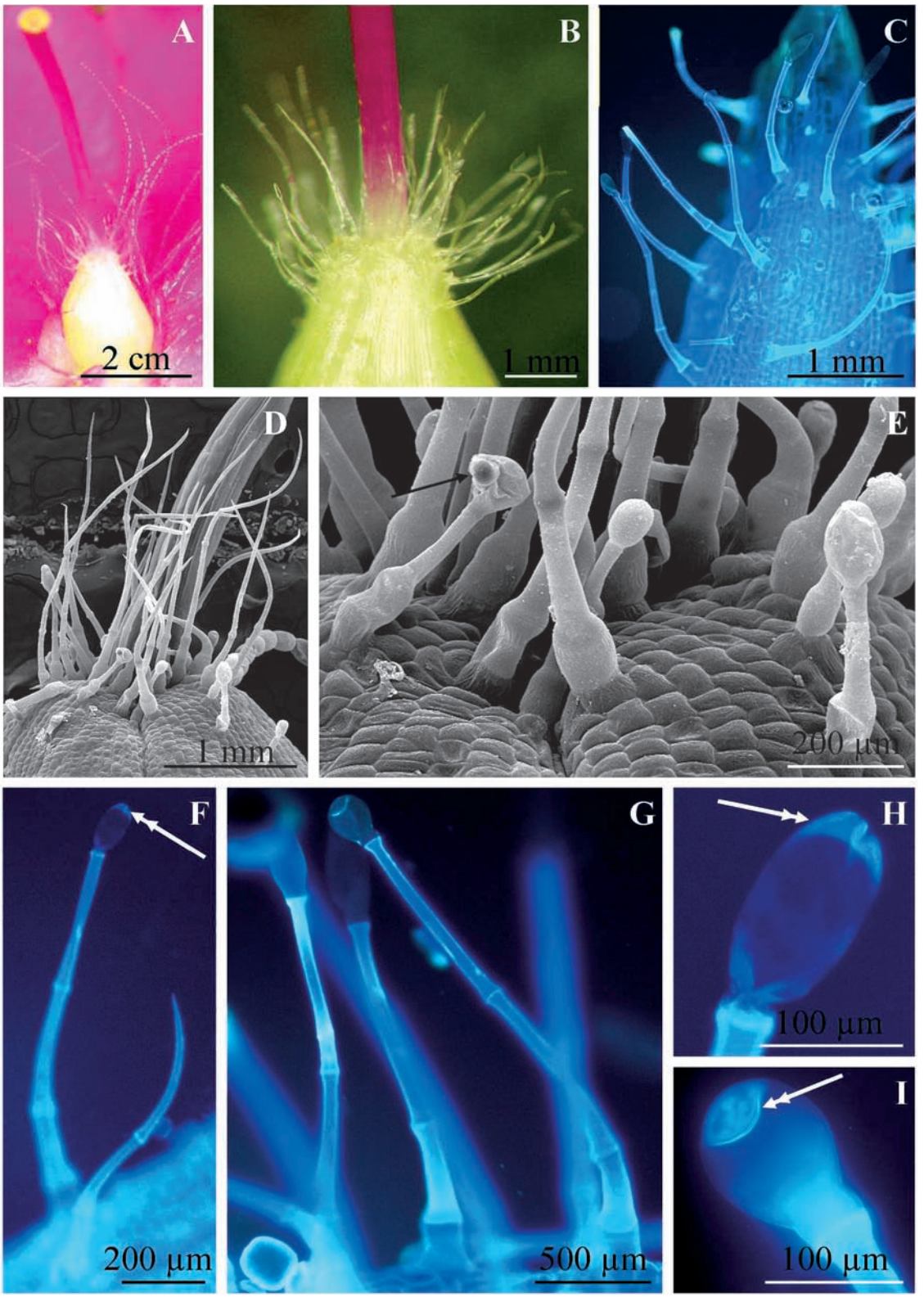

Fig. 4A – Part of the flower of *Trandescantia andersoniana*; visible is an elongated ovary with a long style and a circular stigma.
B-E – Fragments of the apical part of the ovary of the pistil; visible are non-glandular and glandular trichomes, a raised cuticle on the surface of the glandular head (arrow) (D) as well as a striated cuticle on the cell adjacent to the hair base and on other epidermal cells of the ovary (D, E),
F-I – Glandular trichomes; there can be seen the subcuticular space that accumulates the oil (F, H, I) (double-headed arrow). A, B – (SM); C, F-I – (FM); D, E – (SEM).

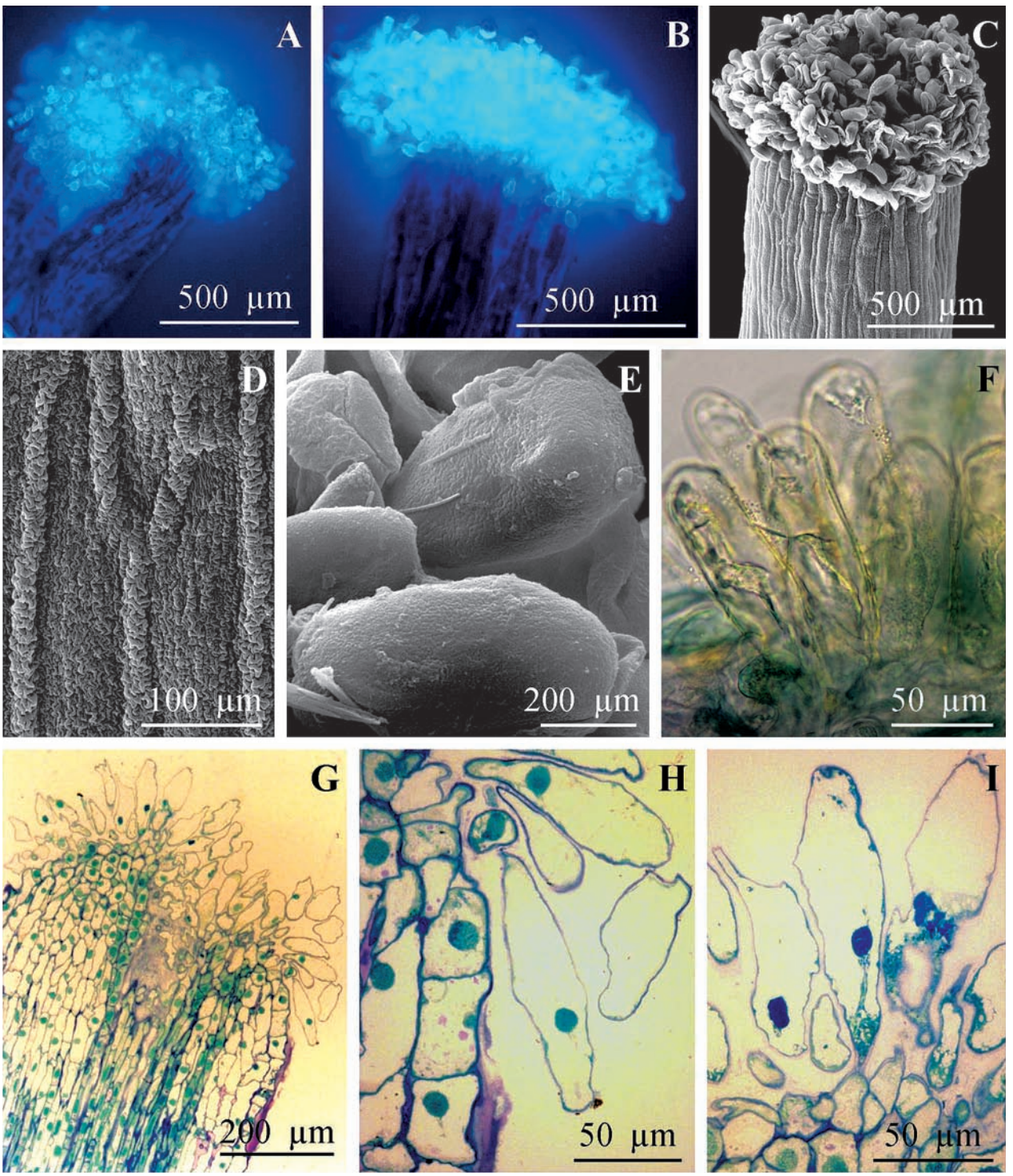

Fig. 5 A-I – Parts of the stigma and of the style of the pistil:
A-C – Stigma of the pistil with tightly packed papillae,
D – Part of the style near the stigma; a folded cuticle can be seen,
E – Apical part of the papilla; a smooth cuticle can be seen on their surface,
F-I – Longitudinal sections of the stigma; visible are unfused club-shaped papillae with a large nucleus.
A-B – (FM); C-E – (SEM); F-I – (LM).

Stamens. Hairs grow out of the filaments of six stamens arranged in two whorls (Fig. 3A, B, F). Their region covers the filament from its base up to a height of 4.7 mm; the epidermis is smooth above this area along a section of 3.5 mm reaching the yellow anthers. The long, densely growing, anthocyanin-coloured trichomes are composed, in the basal and central part, of slightly elongated cells, with their width and length ranging respectively 85-115 μm and 111-179 μm, whereas at the tip they have a spherical shape. The cells arranged in one row form chain-shaped trichomes (Fig. 3A-C; F-I). Their number in a hair growing at the basal part of the filaments is from 20 to 38. Striate cuticular ornamentation occurs on their surface. The regularly arranged striae with an undulating contour run to the two poles of the cell (Fig. 3D, E). At the place where the cells are joined together, a thicker strand of cuticle can be seen; it exhibits autofluorescence together with the cell wall (Fig. 3H). A wider strand of cytoplasm, with a large centrally located nucleus around which leucoplasts are gathered, is visible in the cell protoplast (Fig. 3H-J).

Pistil. The spiderwort flowers develop a pistil with a white 3-chambered ovary and a long (3.6-5.5 mm) pink style as well as a spherical stigma (Fig. 4A). Non-glandular and glandular trichomes occur by the style at the tip of the ovary along a section of 1.7-2.1 mm (Fig. 4A-G). The long (1.1-1.9 mm), sharp-pointed, non-glandular trichomes are uniseriate and several-celled (4-5) (Fig. 4D, F).

The glandular trichomes grow between them (Fig. 4C-G); they consist of a 2-5-celled stalk and a unicellular, spherical or slightly elongated, glandular head. The cuticle on the cell surface at the base of the hair is striated (Fig. 4E), while on the higher located cells it is smooth (Fig. 4D-E). A raising of the cuticular layer is visible on the glandular head (Fig. 4D) and the oil accumulated in the so formed subcuticular space can be seen; the cells adjacent to the head show autofluorescence (Fig. 4F-I).

The wet stigma, 0.9-1.1 mm in diameter, develops densely packed (Fig. 5A-C, F-I), unfused, club-shaped, unicellular papillae (Fig. 5F-I). Their diameter at the apical part is 42-52 μm, whereas their length ranges 42-52 μm. The cuticle is smooth on the surface of the papillae (Fig. 5E), while on the style near the stigma it forms dense folds (Fig. 5D).

DISCUSSION

Numerous, several-celled, non-glandular and glandular trichomes, which occur with the greatest density on the sepals, grow on the abaxial surface of the calyx epidermis and in the terminal part of the ovary in the investigated flowers. The glandular trichomes differ structurally in the shape of their head; it can be spherical, cylindrically elongated, or with a narrowing at the base. The characters of their protoplast: a thick cytoplasm, a large cell nucleus, and oil accumulated in the subcuticular space with distinct fluorescence, are evidence of their high secretory activity.

Research on the trichomes in the family Commelinaceae has been conducted in earlier studies by different researchers (Tomlinson, 1969; Hardy and Stevenson, 2000a, b). On the surface of the floral elements of these plants, Tomlinson (1969) distinguished non-glandular, several-celled, sharp-pointed and hooked trichomes as well as glandular micro and macro hairs. Living non-glandular hairs support transpiration (Kopcewicz and Lewak, 2002) and are an attractant for insects (Wojtaszek et al. 2006), while hooked trichomes (Thaler et al. 2001) impede their movement on the surface and reduce foraging (Wojtaszek et al. 2006). They also perform an important function in plant taxonomy in the identification of relevant systematic units (Esau, 1973).

The micro hairs in *T. zebrina* are responsible for mucus secretion (Thaler et al. 2001), while the macro hairs with a unicellular head, which are present on the surface of the perianth of many plant species (Weryszko-Chmielewska and Chwil, 2008; Dmitruk and Weryszko-Chmielewska, 2010), are responsible for flower scent (Esau, 1973; Harborne, 1997).

The epidermal cells are probably responsible for scent emission in the petals of the corolla in the species under study. Conical projections of the cuticle, under which the odorous substance may accumulate, are found at some places on their outer cell wall. The papillae on the staminal filaments in *Asphodelus* (Weryszko-Chmielewska et al. 2007) and on the ovary of the pistil in *Allium* (Żuraw et al. 2010) perform a similar function of the emission layer as in the case of the described secretory structures.

The staminal filaments develop numerous chain-shaped hairs that are composed of 20 to 38 cells at the base of the filament. Their number depends on the time of formation and the location as well as on the origin of the hairs (Mericle and Hazard, 1980). About 800 openings, containing plasmodesmata, were found in the wall joining the cells of these trichomes (Roelofsen and Houwink, 1951). In the ontogeny of another species of the genus *Tradescantia*, the stamen hairs are formed later and within a shorter time than the corolla. Their initiation starts from a small papilla at the base of the filament. The number of trichomes on the surface of a staminal filament is controlled genetically, but it may be modified by environmental factors (Mericle and Hazard,

1980). These hairs are an optical attractant for insects (Faden, 1992). They probably also facilitate the landing of insects and their collection of pollen grains from the stamens with long filaments. Due to their model structure, they are frequently used for experimental studies (Tyree and Tammes, 1975).

On the surface of the stamen hairs and of the perianth in question, there is a striated cuticle with the striae arranged along the longer axis of the cell; the striae meet at the poles of the cell and are less visible. Such ornamentation is in agreement with the reports of Gale and Owens (1983). These authors report that the degree of striation is poorly visible at the base of the cells of each floral element in Commelinaceae plants. Initially, the cuticle is formed uniformly about 10 days before flower formation; then, the striae become visible as a result of the extension the cuticle due to the elongation of the cells (Mericle and Hazard, 1980).

Tetracytic stomata, located at a distance of 2-5 adjacent cells from each other, occur in the abaxial epidermis of the investigated calyx. A similar type and arrangement of pores were found in the leaf epidermis of *T. virginiana* (Tomos et al. 1981). *Tradescantia*, being a plant of moist areas (Chmiel, 2000), develops a large number of stomata in the epidermis, which enables efficient water transport and stomatal transpiration (Kopcewicz and Lewak, 2002). In a water deficit situation, spiderwort effectively copes with adverse environmental conditions; the time needed to close the pores was calculated to be 1-30 s (Tomos et al. 1981).

The stigma of the spiderwort under study develops unicellular unfused papillae covered by a smooth cuticle. These cells are characterized by vacuolization and a large nucleus. A similar type of the stigma has been described in the flowers of different genera of the family Commelinaceae. Vacuolized papillae in several *Tradescantia* species developed a cuticle of varying thickness and contained the main organelles, in addition to chloroplasts (Owens et al. 1984; Hardy and Stevenson, 2000b).

The presence of anthocyanins was observed in the epidermis of the corolla cells and in the stamen hairs. These pigments are finally formed two days before flower opening. According to Tatsuzawa et al. (2010), these are mainly delphinidins and cyanidins and they have been found in the flowers of 12 different taxa of the genus *Tradescantia*. Their colour is more stable compared to anthocyanins from other plants (Baublis et al. 1994); therefore, they can be of importance as food pigments (Lin et al. 1992).

Calcium oxalate crystals are found in the form of needles in the parenchymal cells of the investigated flower stalks. Their presence confirms one of the characters of Commelinaceae plants (Świdzińska, 1998). *T. pallida* accumulates these secretions in different organs, among others, in the parenchyma and collenchyma cells (Brizuela et al. 2007; Chimpan, 2009). The type of the observed crystals in the examined flowers is in agreement with the reports of Brizuela et al. (2007).

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Mikromorfologia i anatomia elementów kwiatowych *Tradescantia* x *andersoniana* W. Ludw. Rohweder

Streszczenie

Rodzaj *Tradescantia* obejmuje około 70 gatunków. W Polsce *Tradescantia x andersoniana* uprawiana jest w gruncie jako roślina ozdobna, zalecana na rabaty i wokół zbiorników wodnych. Przeprowadzono badania obejmujące kwitnienie i cechy mikromorfologiczne oraz anatomiczne wybranych elementów kwiatowych *Tradescantia x andersoniana* W. Ludw. Rohweder 'Karin'. Makro- i mikromorfologię kwiatów analizowano przy użyciu mikroskopii stereoskopowej, świetlnej i skaningowej elektronowej (SEM). Trzykrotka wykształca kwiaty o średnicy 4,6-5 cm, które rozkwitają wczesnym rankiem i zamykają się w południe. Na powierzchni odosiowej epidermy kielicha i wierzchołkowej części zalążni wyrastają włoski ochronne oraz wydzielnicze różniące się strukturalnie. Ich największą liczbę stwierdzono na odosiowej epidermie kielicha. Włoski gruczołowe wykształcają kilkukomórkową nóżkę i jednokomórkową, kulistą lub wydłużoną główkę. Nitki pręcika tworzą trichomy o łańcuszkowatym kształcie. Na ich powierzchni i epidermie okrywającej okwiat występuje prążkowana ornamentacja kutykularna. Prążki na komórkach włosków pręcika zbiegają do dwóch biegunów komórki. Górny słupek wykształca mokre znamię z niezrośniętymi, jednokomórkowymi papillami. Na ich powierzchni kutykula jest gładka, natomiast na szyjce słupka w pobliżu znamienia tworzy gęste pofałdowania.