ANATOMY OF STAMINAL HAIRS FROM TRADESCANTIA AS A BACKGROUND FOR TRANSLOCATION STUDIES

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SUMMARY

Longitudinal walls have a cuticle almost impermeable to water and sucrose and no ectodesmata. The interconnecting walls have plasmodesmata scattered over the wall; av. diam. 0.045 μ m, appr. 11 per μ m², covering 1.65% of the wall area. This area, together with the cross-sectional area of the cytoplasmic layer between plasmalemma and tonoplast, gives an impression of capacity for transport through the cytoplasmic part of the symplasm. The two areas were compared.

1. INTRODUCTION

The study of translocation in *Tradescantia* hairs has the advantage that linear transport can be investigated in a row of single cells. Such without the interference of parallell cells or the long-distance transport systems such as xylem or phloem.

When a translocation study in *Tradescantia* hairs was started it became necessary to gather information on anatomy, especially on ultrastructure, number and distribution of the plasmodesmata in the walls between the cells, in comparison with the cross-sectional area of the cytoplasm in the cells.

2. MATERIAL AND METHODS

Staminal hairs were used of the blue Tradescantia virginica L.

- For electron microscopy fixation was 1 h in 5% glutaraldehyde in phosphate buffer 0.1 M, pH 7.2 at 22°C. After prolonged and careful rinsing in phosphate buffer, the hairs were postfixed in a 1% OsO₄ solution for 2.5 h at 22°C. During dehydration, the hairs were poststained with 2% uranylacetate in 70% ethanol.
- 2. Thickness of cytoplasmic layer along the longitudinal walls. Hairs were placed on a sheet of water-repellent plastic and a minute droplet of 0.5% aqueous uranin was placed on the wounds. After keeping for 30 min in a moist atmosphere, the droplets were blotted off and the hairs left to dry for 5 min in the room. They were then observed immersed in paraffin oil with fluorescence microscopy with light of 400 nm. Measurements were taken from micrographs.

- 3. Callose. The presence of callose deposits was tested by a solution of 2% aqueous Wasserblau under a fluorescence microscope.
- 4. The impermeability of the cuticle for water and sucrose was tested by exposing hairs with sealed wounds, either in the room for 1 or 2 h, or for
- 2 h in aqueous sucrose solution 1 M. Hairs with open wounds were used as controls.

3. RESULTS AND DISCUSSION

3.1. Dimensions

Hairs can reach a maximum of 8 mm with over 40 cells. The cells are long at the base and gradually diminish in length until they are almost globular at the tip. The diameter of the crosswalls is smaller, often giving the cells a barrel-shaped appearance. The surface area of the interconnecting walls in relation to a crosssection through the broadest place of the lumen was 36% for basal cells and 11% for tip cells.

Table 1. Dimensions of cells of hairs of *Tradescantia virginica* (light microscope). Averages of 10 cells.

	length	largest diam. lumen in cross- section (μm)	diam. of con- necting walls (µm)	approx. volume 1 mm length of hair (ml)
basal cells	300-510	77–133	39–74	
	av. 405	av. 94	av. 56	7×10^{-6}
tip cells (fifth from	88-147	70–84	21–28	
tip measured)	av. 119	av. 80	av. 26	5×10^{-6}

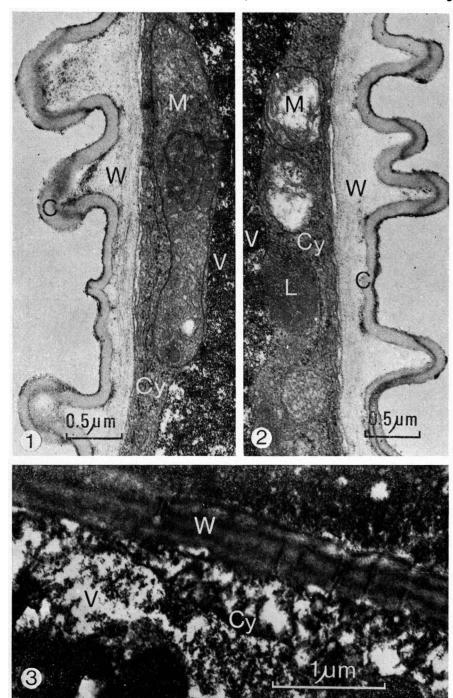
3.2. Thickness of protoplasmic layer along the longitudinal walls Uranin is accumulated by the cytoplasm and not by the vacuole or cell wall. It contrasts sharply with both in fluorescence microscopy (Döring 1935 for *Allium*). It is also translocated through a successive number of cells as found for *Cucurbita* leaf hairs by SCHUMACHER (1936). With fluorescence microscopy the cytoplasm along the longitudinal walls is sharply demarcated. Micrographs indicate an average thickness of approx. 0.9 µm (plate II, fig. 5).

Note: In fluorescence microscopy with light of 400 nm the granular structure of the protoplasm and protoplasmic streaming are no longer visible. When the light is shifted to normal they reappear. SCHUMACHER (1936) concluded a superimposed structure visible in fluorescence

Plate I.

Figs 1 and 2: Section through longitudinal wall: C cuticle, W wall, Cy cytoplasm, M mitochondrion, L lipid, V vacuole, \times 30,000.

Fig. 3: Longitudinal section through interconnecting wall: W wall, Cy cytoplasm, V vacuole. Note plasmodesmata through the wall, \times 30,000.



microscopy after uranin uptake. In our opinion, the observed phenomenon is due to invisible refractory differences and an all-over emittance of fluorescent light appearing in the whole cytoplasm.

Some cytoplasm is accumulated on both sides of the interconnecting walls and around the nucleus. Where strands, that traverse the vacuole, are attached to the cytoplasm along the walls, there is also an accumulation of cytoplasm (plate II, fig. 5).

3.3. Walls

Longitudinal walls have a varying thickness of 0.5-1 μm (electron microscope). They are covered by a cuticle uniformly $0.1~\mu m$ thick. In the longitudinal walls, no ectodesmata were observed (plate I, figs. 1 and 2). Intact hairs with sealed wounds did not show plasmolysis in aqueous sucrose 1 M nor did they change when exposed in the room for 1 or 2 h. Controls without sealed wounds show plasmolysis, proceeding from the open wounds through a number of successive cells. Evaporation through open wounds results in decrease in volume and the formation of fusiform cells. These results, together with the ultrastructural data clearly show the considerable impermeability of the cuticle on the longitudinal walls for water and sucrose.

The interconnecting walls are approx. 0.3 μ m thick (electron microscope) and plasmodesmata can be observed (plate I, fig. 3).

3.4. Plasmodesmata

These plasmodesmata are scattered over the wall area. Often they occur in pairs. Average diam. is $0.045 \,\mu m$ and the frequency is approx. 11 per μm^2 , constituting about 1.65% of the wall area (plate II, figs. 4a and 4b). The combined area of the plasmodesmata is about 15% of the cross-sectional area of the cytoplasm in the middle of the basal cells and about 4% in the tip cells.

Note: Often an electron-translucent ring of $0.09 \,\mu\text{m}$ diam. is observed around the plasmodesmata. Roelofsen & Houwink 1951 published an electron micrograph of a macerated interconnecting wall of a *Tradescantia* hair, showing black holes, often in pairs, with a diam. of $0.09 \,\mu\text{m}$ (plate II, figs. 4a and 4b).

3.5. Callose

Deposits of callose were not found in detached hairs, except rarely a deposit on the first interconnecting wall next to a wound.

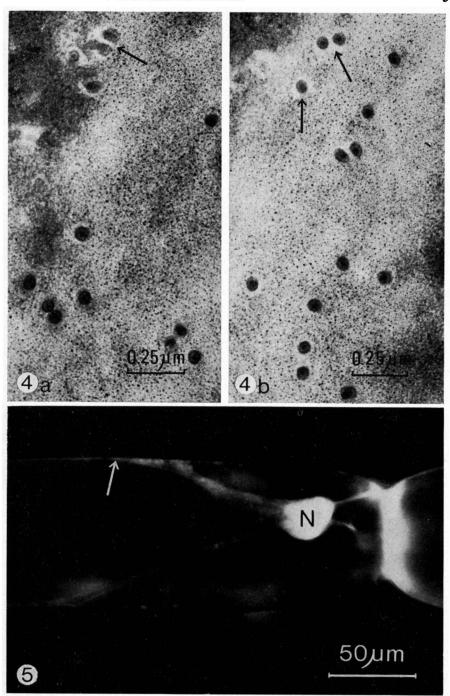
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Plate II.

Figs. 4a and 4b: Parallel section through interconnecting wall with plasmodesmata. The arrows indicate plasmodesmata with translucent rings, \times 60,000.

Fig 5: Uranin translocation in basal cell. Arrow indicates thin cytoplasma layer along wall: N nucleus. Fluorescence microscopy in light of 400 nm, \times 450.



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