Ultrastructure of traumatic resin duct formation in *Cupressus* sempervirens L. in response to the attack of the fungus Seiridium cardinale (Wag.) Sutton & Gibson

<u>ALESSIO PAPINI¹</u>,*, SALVATORE MORICCA², ROBERTO DANTI³, CORRADO TANI¹, ISABELLA POSARELLI², SARA FALSINI¹, GIANNI DELLA ROCCA³

¹ Department of Biology University of Florence, Italy; ² Department of Agrifood Production and Environmental Sciences, Plant Pathology and Entomology Division, University of Florence, Italy; ³ Institute for Sustainable Plant Protection, IPSP-CNR, Sesto Fiorentino, Italy

Cupressus sempervirens L. (cypress), belongs to family Pinaceae and represents an important tree for the typical tuscan and eastern-mediterranean landscape. In the last years this tree is frequently affected by fungal attack by the fungus *Seiridium cardinale* (Wag.) Sutton & Gibson, the cause of the so called cypress canker.

The main mechanism of defense of cypress from the pathogen is the post-infectional development of a well-structured necrophylactic periderm (NP) [1], the increase in polyphenol cells and the ex-novo formation of traumatic resin ducts.

This investigation deals with the microscopical analysis of the development of the traumatic resin ducts and the ultrastructure of the activity of the epithelial cells of the duct itself.

The samples were fixed in 1.25 % glutaraldehyde at 4°C in 0.1 M phosphate buffer (pH 6.8), then post-fixed in 1% OsO₄ in 0.1 M phosphate buffer (pH 6.8) for 1 hr. After ethanol series dehydration and a final propylene oxide step, the samples were embedded in Spurr's epoxy resin. The 80nm thick sections were stained with uranyl acetate and lead citrate. The observations were done with a Philips EM201 TEM.

The formation of the traumatic resin duct starts in the secondary phloem around a layer of sclerenchyma fibers (Fig. 1) as a result of degeneration (PCD) of sieve cells and the reprogramming of albuminous cells into epithelial cells that produce terpens that will enter into the duct to form the resin.

The epithelial cells show plastids specialized in the production of terpens. The plastids are surrounded by ER elements (Fig. 2) and fill themselves with a granular material that eventually will cross the plasmamembrane and enter into the resin duct.

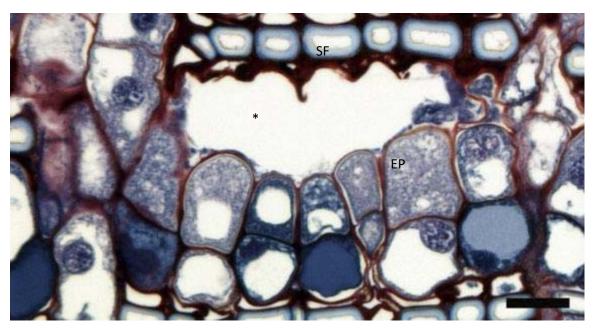


Figure 1. Traumatic resin duct. Light microscope. Toluidine blue staining. The lumen is indicated by an asterisk. EP = epithelial cell. SF = Sclerenchyma fiber. $Bar = 25 \mu m$. $Bar = 1 \mu m$

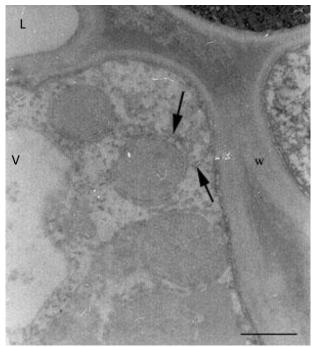


Figure 2. Traumatic resin duct. Transmission Electron Microscope. Detail of an epithelial cell. ER elements (arrows) are surrounding the plastids. $W = cell \ wall; \ V = vacuole; \ L = lumen. \ Bar = 1 \mu m.$

References

[1] Danti R, Rotordam MG, Emiliani G, Giovannelli A, Papini A, Tani C, Barberini S, Della Rocca G (2018) Different clonal responses to cypress canker disease based on transcription of suberinrelated genes and bark carbohydrates' content. Trees 32(6): 1707–1722