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Presence, Distribution and Form of Calcium Oxalate Crystals in Relation to Age of *Actinidia Deliciosa* Leaves and Petioles

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Abstract—

Calcium (Ca^{2+}) is an element essential to plant being involved in plant growth and development. At high concentrations it is toxic and can influence every stage, process and cellular activity of plant life. Given its toxicity, cells implement mechanisms to compartmentalize calcium in vacuole, endoplasmic reticulum, mitochondria, plastids and cell wall. One of the most effective mechanisms to reduce excess of calcium, thus avoiding cellular damage is its complexation with the oxalic acid to form calcium oxalate crystal no longer osmotically or physiologically active. However, the sequestered calcium can be mobilized when the plant needs it. Calcium crystals can be accumulated in the vacuole of specialized sink-cells called idioblasts, with different crystalline forms (druse, raphyde and styloid) of diverse physiological meanings. *Actinidia deliciosa* cv. Hayward, presents raphydes and styloids localized in idioblasts in cells of photosynthetic and non-photosynthetic tissues. The purpose of this work was to understand if there is a relationship between the age of *Actinidia* leaves and the presence, distribution, dimension and shape of oxalate crystal by means of light, fluorescent, polarized and transmission electron microscopy.

Three vines from female plants were chosen at the beginning of the season and used throughout the study. The leaves with petioles were collected at various stages of development from the bottom to the shoot of the plants monthly from April to July. The samples were taken in corresponding areas of central and lateral parts of leaves and of basal portion of petiole.

The results showed that in the leaves the number of raphyd idioblasts decreased with the progress of the growing season, while the styloid idioblasts increased progressively becoming very numerous in the upper nodes of July. In June and in July samples, in the vacuoles of the highest nodes, a portion regular in shape strongly stained with rubeanic acid was present. Moreover, the chlortetracycline (CTC) staining for localization of free calcium, marked the wall of the idioblasts and the wall of the cells near vascular bundles. In April petiole samples, moving towards the youngest nodes, the raphydes idioblast decreased in number and in length of the single raphydes. Besides, crystals stained with rubeanic acid appeared in the vacuoles of some cells. In June samples, numerous raphyde idioblasts oriented parallel to vascular bundles were evident. Under electron microscope, numerous idioblasts presented not homogeneous electrondense aggregates of material, in which a few crystals (styloids) in form of regular holes were scattered. In July samples, an increase in the number of styloid idioblasts in the youngest nodes and little masses stained with CTC near styloids were observed. Peculiar cells stained with rubeanic acid were detected and hypothesized to be involved in the formation of the idioblasts. In conclusion in *Actinidia* leaves and petioles it seems confirmed the hypothesis that the formation of styloid idioblasts can be correlated to increasing calcium levels in growing tissues.