



Research Paper

DISTRIBUTION OF CALCIUM OXALATE CRYSTAL CONTAINING IDIOBLASTS IN THE LEAVES OF SYNGONIUM PODOPHYLLUM SCHOTT

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The leaves of *Syngonium podophyllum* were examined microscopically to determine the distribution of both druse and raphide idioblasts. Druse crystal idioblasts are small spherical cells found throughout the lamina mostly in sub-epidermal areas. Two types of raphide idioblasts were observed in the leaves of *Syngonium podophyllum*: the non defensive raphide idioblasts, which are elongated or more or less egg shaped cells usually found embedded in tissues of the leaf margins; and the defensive raphide idioblasts, also elongated cells, but usually found suspended between mesophyll cells in leaf airspaces. The average densities of raphide cells were highest in young leaf than the mature leaf and average densities of druse cells were highest in mature leaf than the young leaf. The raphide and druse cells showed a bilaterally symmetrical distribution during all stages from young to mature leaves but, generally contain more druses than raphides.

Keywords: Druse, Defensive and Non defensive, Raphide idioblasts

INTRODUCTION

The specialized plant cells and structures occur in apparently nonrandom arrangements; these include guard cells, silica cells, trichomes, resin ducts, sclereids, and crystal idioblasts, which by their nature are ideal subjects for the study of histogenetic patterning in plants. Though idioblasts comprise a small minority of all cells in a tissue, their numbers and densities are variable. Their occurrence may be sensitive to

environmental factors and may be subject to experimental manipulations (Al-Talib and Torrey, 1961; Paupardin, 1965; Knecht and O'Leary, 1972; Sharma, Chandler and Salemi, 1980). The most extensively examined structures, in terms of distribution and patterns, have been the epidermal derivatives: guard cells and trichomes. Actual numbers of stomata, for example, have been recorded for many species (Morren, 1863; Eckerton, 1900; Timmerman, 1927; Gupta, 1961;

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Knecht and O'Leary, 1972; Sharma *et al.*, 1980). The more ordered pattern of stomata's on leaves of *Crinium* has been assessed by Sachs (1974). Less has been done with the patterns of trichomes, despite the considerable interest in their morphology and ultrastructure. Both trichomes and stomates are external features of plant organs; as such, they are more readily accessible to the quantitative microscopist than are idioblasts of the interior, such as oil cells, laticifers, sclereids, and calcium oxalate crystal cells. Conspicuous patterns have been noted for some of these internal idioblasts, particularly crystal-containing cells (Frank, 1967; Frank and Jensen, 1970; Arnott, 1973; Sunell and Healey, 1979). The variation in density and local distributions of these cells is considerable, and fluctuations may be correlated with development or growth rate, as with resin canals in pine needles (White and Beals, 1968) and crystal idioblasts in taro corms and leaves (Sunell and Healey, 1979 and Sunell and Healey, 1985). Generally, however, few authors have carefully examined the spatial or temporal patterns of idioblast distributions in entire organs. In this paper, we report the distribution of the calcium oxalate crystal idioblasts, both raphides and druses; in the leaves of *Syngonium podophyllum*. These determinations have been correlated with an aspect of development.

MATERIALS AND METHODS

Leaves were excised at different stages of development from nursery plants of *Syngonium podophyllum*. Entire leaves were cleared by first placing them in 70% ethanol at 60 °C for several hours, then in 95% ethanol at room temperature for 1 hr. After washing briefly in distilled water, leaves were placed in 5% NaOH for 1 hr at room temperature, and then washed three times in

distilled water. The leaves were cut from different regions of the leaf lamina and they were mounted in glycerol on glass slides. Numbers of cells containing raphide and druse crystals were determined by counting in per microscopic field.

Slides were examined with bright field and polarization microscopy and later, viewing the slides through polarizing sunglasses. The filters on the light source were rotated to achieve extinction of background illumination when viewed through the sunglasses. Digital micrographs were taken with a Nikon [NIKON ECLIPSE (LV100 POL)] digital camera mounted on a Nikon Eclipse 50i microscope (Nikon, Tokyo, Japan). Some low magnification digital micrographs were taken with an light microscope (10X x 40X) [Olimpus], Phase Contrast Microscope (Leica DM- 1000)

RESULTS

Druse was most common crystal type in leaves, and occasionally very small druses were also seen. Which are compound crystals, composed of many small plates like crystals (Figure 5 and 6) which are found throughout the sub epidermal mesophyll, and, rarely, in the aerenchyma, of *Syngonium podophyllum* leaves. The measurement of spherical druse cells contained, approximately from 22-30 um, in diameter (Table 2). Two forms of raphide idioblasts occur in *Syngonium podophyllum* leaves. Elongated or more or less egg shaped cells, which are typically, 75.64 um long and 46.53 um in diameter (Table 2), occur mainly along the leaf margins and occasionally along major veins and in the mesophyll. The crystals occupy a central position in the idioblast. Generally, they are aligned parallel to the long axis of the cell but are not tightly compacted into a bundle (Figure 7). Some or all of the crystals may be situated at oblique angles

(Figures 7 and 8) within the cell or the ends of the crystals may interdigitate. These raphide cells will be referred to as the non defensive raphide idioblasts (Figures 7 and 8). Other raphide cells, 128.02 um long and 27.55 um in diameter (Table 2), are suspended in the airspaces of the mesophyll. They are positioned approximately parallel to the surfaces of the leaf lamina. Most of the raphide idioblast rupture very easily due to the pressure of the adjoining cells and distribute the needle shaped calcium oxalate crystals throughout the adjoining cells (Figure 9). This needle like calcium oxalate crystals are aligned

parallel with the long axis (Figure 4) of the idioblast and fill up nearly the entire cell. Crystals are ejected through one or both thin-walled papillae at the poles of the cell (Figures 2 and 3). These idioblasts will be referred to as the defensive raphide idioblasts (Figures 2, 3 and 4). The numbers and densities of druse idioblasts were more variable than the numbers and densities of raphide crystal cells in all leaves (young, medium and mature) examined in per microscopic field in different position (Table 1 and Graph 1). On the average, there were more druse cells than raphide cells. Neither the number of crystal cells nor the

Table 1: Distribution of Idoblasts and Druses in Per Microscopic Field (10 X) in the Leaf of *Syngonium Podophyllum*

No. of Microscopic Field	Stages of Leaf	No. Ofidioblasts	Mean Value	No. of Druses	Mean Value
1.2.3.	Young	191513	15.66	605070	60
1.2.3.	Medium	12149	11.66	806560	68.33
1.2.3.	Mature	9811	9.33	907970	79.66

Table 2: Mesurement of Idoblasts and Druses in the Leaf of *Syngonium Podophyllum*

S. No.	Non Defensive Idioblasts				Defensive Idioblasts				Druses			
	L. (um)		B. (um)		L. (um)		B. (um)		L.(um)		B.(um)	
1.	79.49		51.86		116.75		25.90		28.38		29.09	
2.	76.89	75.64	47.47	46.53	134.10	128.02	29.09	27.55	28.36	26.80	27.05	26.08
3.	70.54		40.25		133.20		27.68		23.66		22.10	

[30-01-2012 19:37 "/Data1" (2455956)]

One population t-Test on Data1 col(B):

Data	Mean	Variance	N
B	3.4665	0.24345	3

$t = 12.16885$

$p = 0.00669$

At the 0.05 level,

the two means are significantly different.

[30-01-2012 19:37 "/Data1" (2455956)]

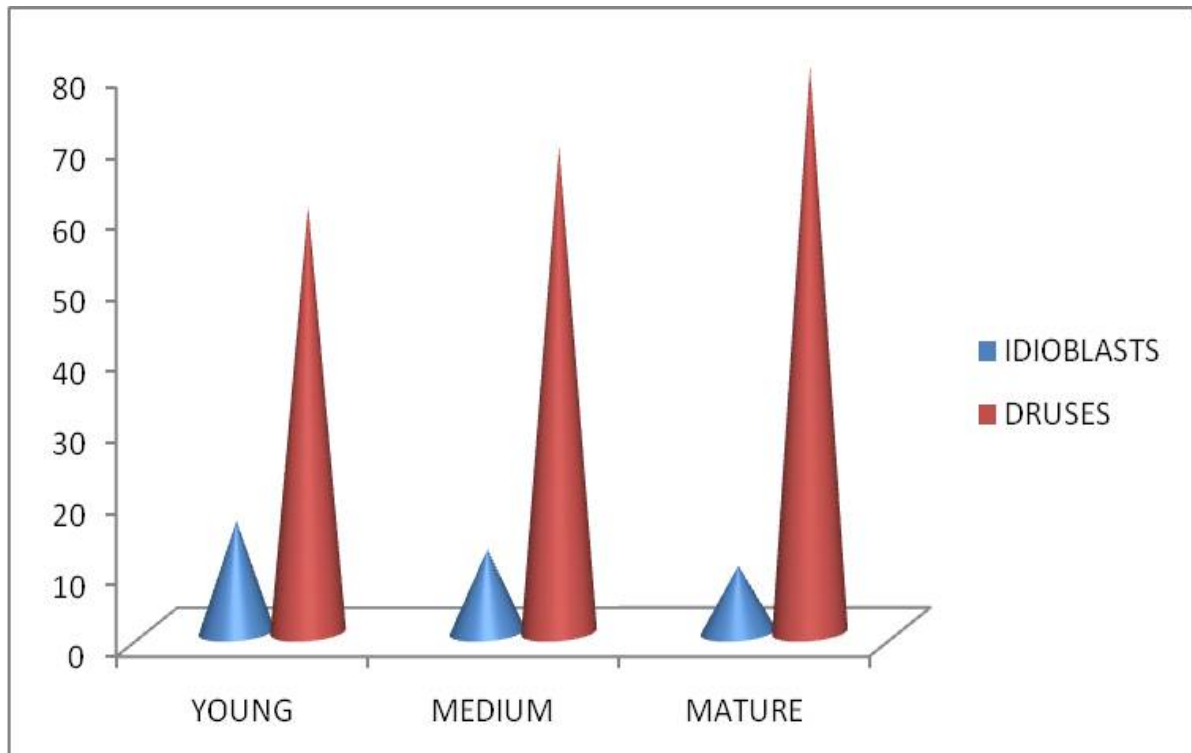
One population t-Test on Data1 col(B):

Data	Mean	Variance	N
B	69.33	97.3789	3
$t = 12.16885$ $p = 0.00669$ At the 0.05 level, the two means are significantly different. [30-01-2012 19:38 "/Graph3" (2455956)] Linear Regression for Data1_B: $Y = A + B * X$			
Parameter	Value	Error	
A	49.67	1.87083	
B	9.83	0.86603	
R	SD	N	P
0.99614	1.22474	3	0.05594
[30-01-2012 19:40 "/Graph3" (2455956)] Linear Regression for Data1_B: $Y = A + B * X$			
Parameter	Value	Error	
A	49.67	1.87083	
B	9.83	0.86603	
R	SD	N	P
0.99614	1.22474	3	0.05594
[30-01-2012 20:18 "/Graph2" (2455956)] Linear Regression for Data1_B: $Y = A + B * X$			
Parameter	Value	Error	
A	49.67	1.87083	
B	9.83	0.86603	
R	SD	N	P
0.99614	1.22474	3	0.05594

density was strongly correlated with leaf size. The average density of raphide cells was greatest in younger leaves. These data confirm that the average density of crystal cells, especially the defensive raphide cells, was lowest in the oldest leaves of a plant. The average density of druse crystals also was lowest in the most mature

leaves. Up to nearly 95% of the raphide and druse crystal cells counted were located in nonvein areas of the leaves. Raphide idioblasts were most dense at the leaf margins and least dense near the midvein. The density of druse idioblasts was consistently greatest throughout the leaf lamina mainly marginal areas of the leaves (Figure 1).

Graph 1: Graphical Representation of Idoblasts and Druses in Per Microscopic Field (10 X) in the Leaf of *Syngonium Podophyllum*



DISCUSSION

The presence of two types of raphide idioblasts has been reported in another aroid, *Diefenbachiam aculata* (Sakai and Nagao, 1980). These are similar to the two types identified in *Syngonium podophyllum* leaves. The defensive raphide idioblasts, which eject their calcium oxalate needles, have been implicated in the irritation produced when aroids are eaten or handled fresh (Saadi and Mondal, 2011). Both the specialized morphology of the cells in *Syngonium podophyllum*, i.e., their thick lateral walls (Figure 4) and thin-walled terminal papillae (Figure 4), and the location of these cells in the airspaces from which they were easily dislodged, are probably ecologically significant. The position of non-

Figure 1: Distribution of the Druse Idioblast in the Leaf of *Syngonium podophyllum* Observed in PM



Figure 2: Defensive idioblast in which the raphides ejected through its terminal papillae in the leaf of *Syngonium podophyllum* observed in LM

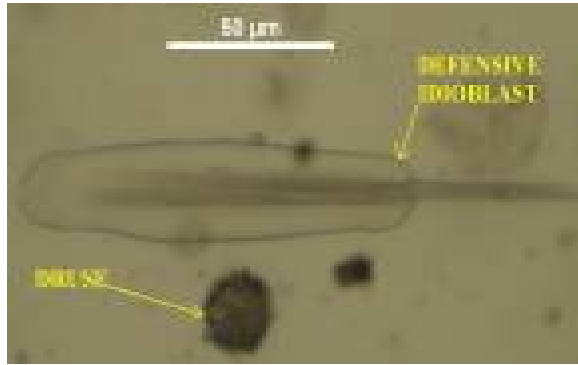


Figure 5: Druse idioblast in the leaf of *Syngonium podophyllum* observed in LM

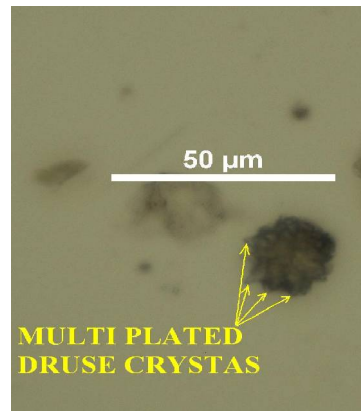


Figure 3: Defensive idioblast in which the raphides ejected through its terminal papillae in the leaf of *Syngonium podophyllum* observed in PM

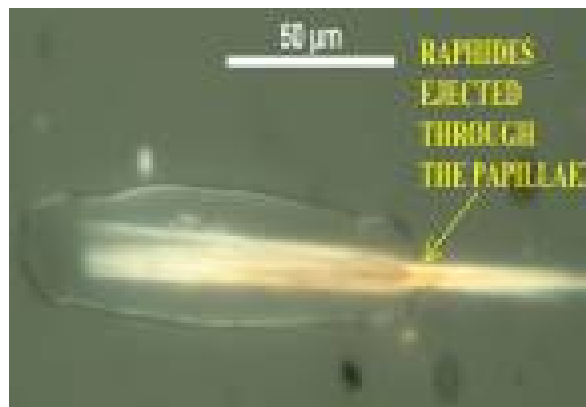


Figure 6: Druse idioblast in the leaf of *Syngonium podophyllum* observed in PM

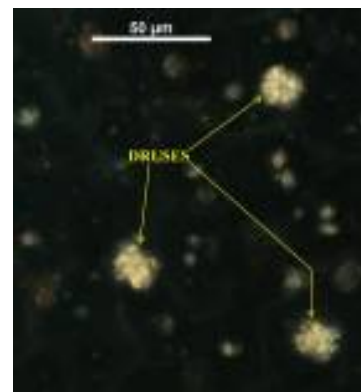


Figure 4: Defensive idioblast in the leaf of *Syngonium podophyllum* observed in PM

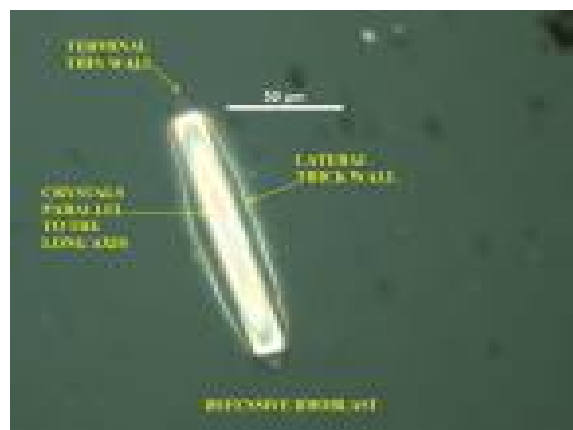
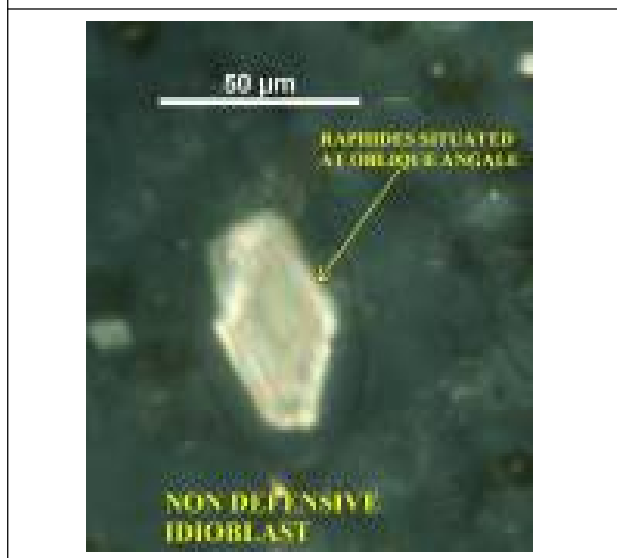


Figure 7: Non Defensive idioblast in the leaf of *Syngonium podophyllum* observed in LM

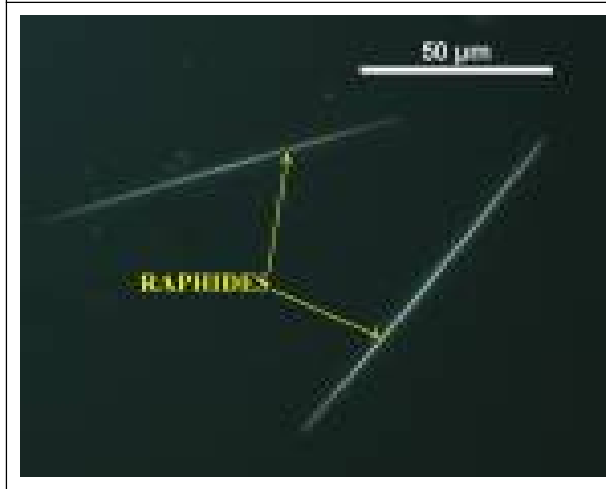


Figure 8: Non Defensive idioblast in the leaf of *Syngonium podophyllum* observed in PM



defensive raphide idioblasts in the leaf may be random, as in the parenchyma, or oriented in longitudinal arrays parallel with the margins and, occasionally, with major veins. The leaves generally contain more druses than raphides. Both types are located primarily in non-vein areas of the lamina. In *Syngonium podophyllum*, wide variations in the numbers and densities of idioblasts are observed, among different leaves at the different stages of development. Comparing the ranges of raphide cell densities and druse cell densities, it is apparent that druse cell occurrence is especially variable. It is known that druse crystals are not permanently deposited. Crystals may be influenced by physiological or environmental factors, though this possibility was not directly investigated in this study. In many cases, druse cells are particularly dense near vascular tissue. This localization has been reported for many other plants, including *Ginkgo* (Amott, 1973) and *Canavalia* (Frank, 1967). In the same study, it was noted that rapidly growing leaves newly emerged from a plant that had been cut back often contain very few druses, but always

Figure 9: Calcium oxalate crystals in the leaf of *Syngonium podophyllum* observed in PM [PM-Polarized Microscope; LM-Light Microscope]



contain raphide cells. This suggests that raphide idioblasts are determined early on and that druse cell differentiation begins later in leaf development. The differentiation of raphide cells, especially the defensive type, is presumably a highly conserved phenomenon in *Syngonium podophyllum* leaves. The pattern of crystal cell distribution in *Syngonium podophyllum* leaves is different for raphide and druse idioblasts. In all leaves examined, most druse cells in mature leaves were located in the side of the leaf blade. Asymmetric distribution is gradually restored as the entire mature leaf. This indicates that the deposition of druse crystals is closely coupled to the later stages of leaf development. Asymmetric patterns of crystal formation have been noted in other plants. Crystal cell differentiation is greatly influenced by the organization of the tissue around the idioblasts.

CONCLUSION

Different parts of the leaf seemed to contain the same kind of crystals idioblasts, in similar densities. The densities of both raphides and

druses idioblast is depend up on the different stages (young, medium and mature) of the leaf development, in which raphide idioblasts are determined early on and that druse cell differentiation begins later in leaf development The uneven distribution of druse cells in the mature leaf, and the subsequent restoration of symmetry as the leaf mature, indicate that druse crystal formation is somehow related to local leaf growth. The defensive raphide idioblasts, with their characteristic shape and location, are probably intimately associated with the formation of the airspace mesophyll.

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