INTERNATIONAL JOURNAL OF SCIENCE AND NATURE

www.scienceandnature.org

DISTRIBUTION OF CALCIUM OXALATE CRYSTAL CONTAINING IDIOBLASTS IN THE LEAVES OF *AGLAONEMA COMMUTATUM* Schott.

Sk. Md. Abu Imam Saadi & Amal kumar Mondal
Department of Botany and Forestry, Plant taxonomy, Biosystematics and Molecular Taxonomy Laboratory
Vidyasagar University, Midnapore- 721 102, West Bengal, India

ABSTRACT
The leaves of *Aglaonema commutatum* Schott. 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 *Aglaonema commutatum***:** the non defensive raphide idioblasts, which are elongated 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.

KEY WORDS: 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; 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; FrankandJensen, 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 *Aglaonema commutatum*. These determinations have been correlated with an aspect of development.

MATERIALS AND M ETHODS

Leaves were excised at different stages of development from nursery plants of *Aglaonema commutatum*. 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. Then 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. The latter, 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

Druses are compound crystals, composed of many small plate like crystals [Fig.-6]. Which are found throughout the sub epidermal mesophyll, and, rarely, in the aerenchyma, of *Aglaonema commutatum* leaves. The measurement of spherical druse cells contained, approximately from 30-31 um, in diameter [Table 2]. Two forms of raphide idioblasts occur in *Aglaonema commutatum* leaves. Elongated cells, which are typically, 145.22 um long and 39.4 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 [Fig.-1]. Some or all of the crystals may be situated at oblique angles or like tree trunk [Fig.-3] within the cell or the ends of the crystals may interdigitate. These raphide cells will be referred to as the non defensive raphide idioblasts [Fig.-1, 2 & 3]. Other raphide cells, 132.11 um long and 33.01 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 whole surface [Fig.-8 & 9]. This needle like calcium oxalate crystals are aligned parallel with the long axis [Fig.-4 & 5] 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 [Fig.-10]. These idioblasts will be referred to as the defensive raphide idioblasts. 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 & Graph 1]. On the average, there were more druse cells than raphide cells [Fig.-7]. Neither the number of crystal cells nor the 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 90% 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 at the midvein and near other major veins than elsewhere in the leaves.

TABLE-1. Distribution of idoblasts and druses in permicroscopic field (10 x) in the leaf of *Aglaonema commutatum*

NO. of microscopic field	Stages of leaf	NO. of Idioblasts	Mean value	NO. of Druses	Mean value
1.	Young	6	6.33	17	13.66
2.		8		10	
3.		5		14	
1.	Medium	4	5.33	12	14.66
2.		7		14	
3.		5		18	
1.	Mature	4	3	20	17.33
2.		2		17	
3.		3		15	

GRAPH -1. Distribution of idoblasts and druses in per microscopic field (10 x) in the leaf of *Aglaonema commutatum*

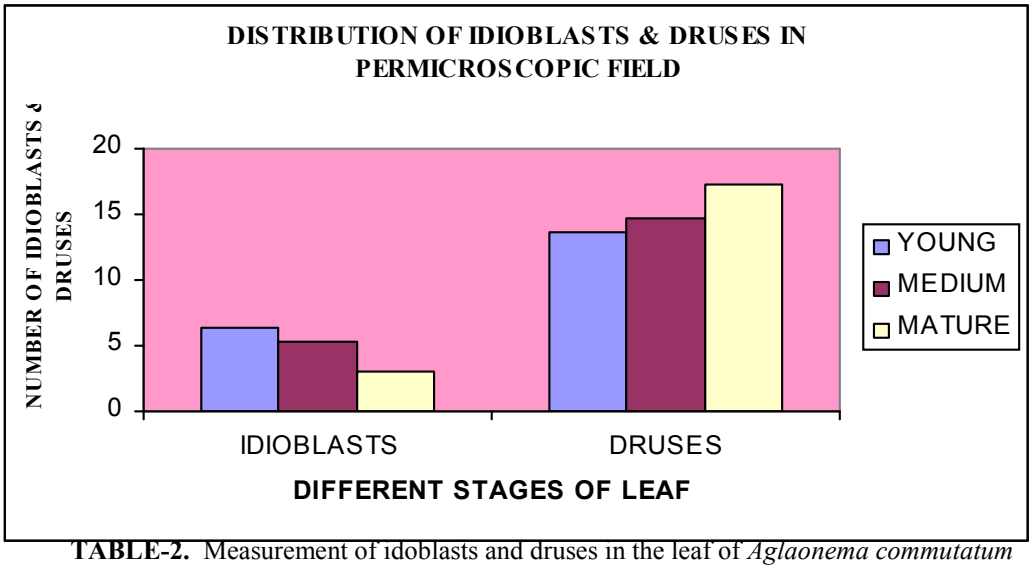

TABLE-2. Measurement of idoblasts and druses in the leaf of *Aglaonema commutatum*

SL.NO.	Non defensive idioblasts				Defensive idioblasts				Druses			
	L. (um)		B. (um)		L. (um)		B. (um)		L.(um)		B.(um)	
1.	144.65		35.20		125.16		28.43		29.96		28.36	
2.	146.16	145.22	37.95	39.4	131.24	132.11	33.17	33.01	27.24	30.38	27.92	30.2
3.	144.87		45.05		139.95		37.45		33.96		34.32	

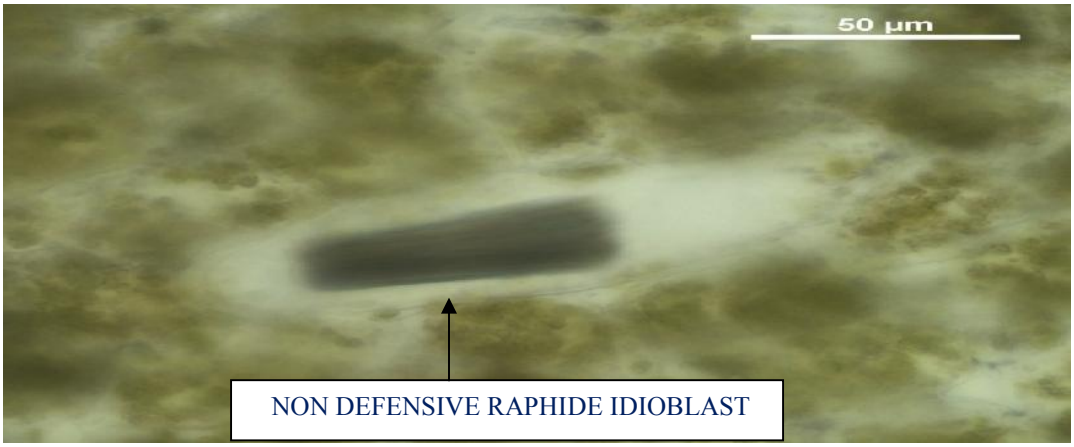

FIGURE 1. Non defensive idioblast in the leaf of *Aglaonema commutatum* observed in L.M.

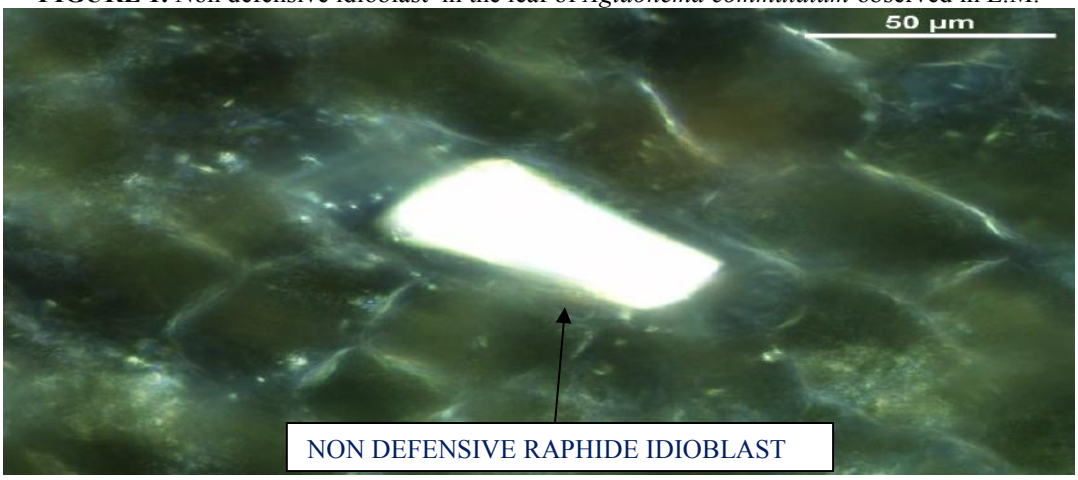

FIGURE 2. Non defensive idioblast in the leaf of *Aglaonema commutatum* observed in P.L.

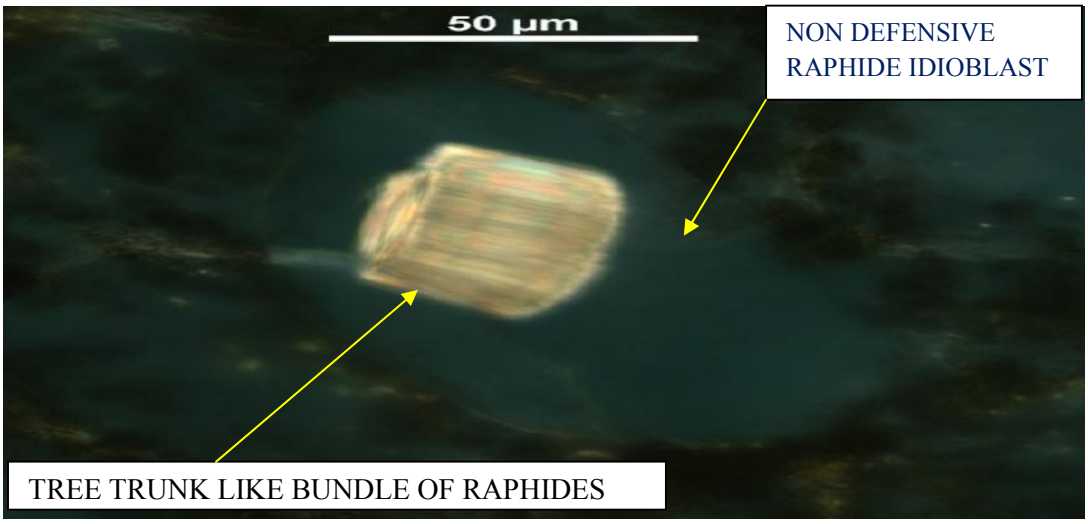

FIGURE 3. Tree trunk like Non defensive idioblast in the leaf of *Aglaonema commutatum* observed in P.L.

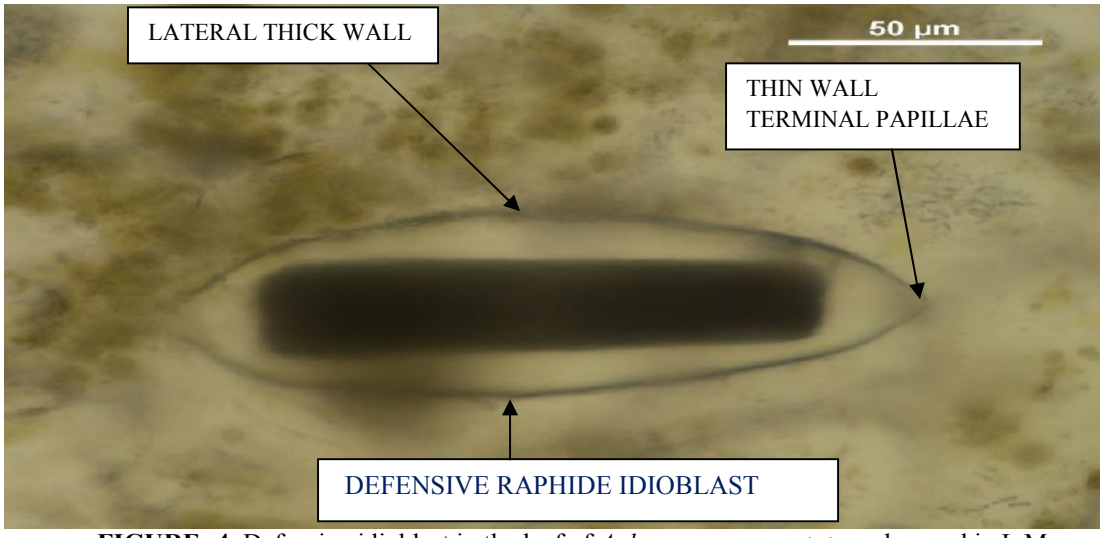

FIGURE -4. Defensive idioblast in the leaf of *Aglaonema commutatum* observed in L.M.

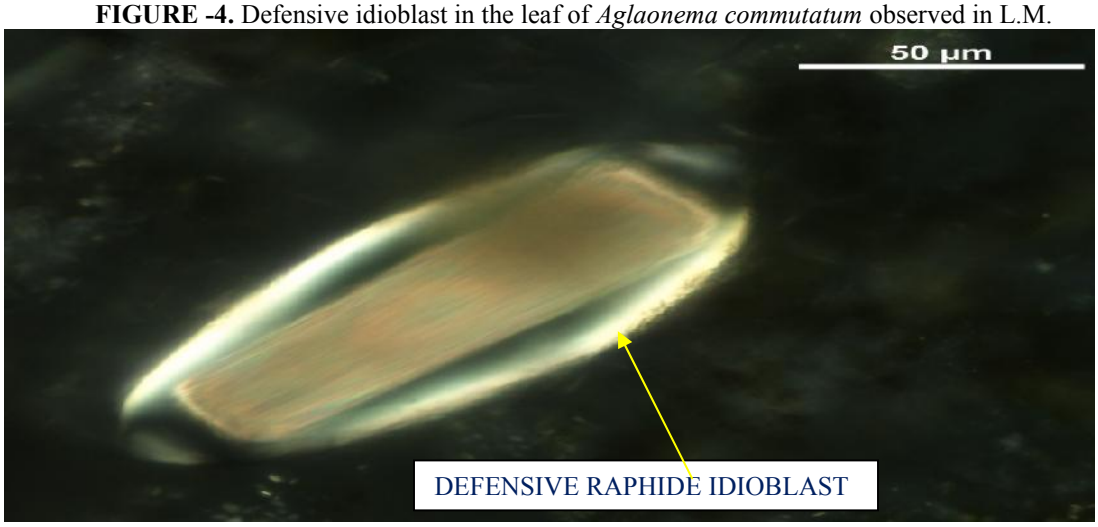

FIGURE 5. Defensive idioblast in the leaf of *Aglaonema commutatum* observed in P.L.

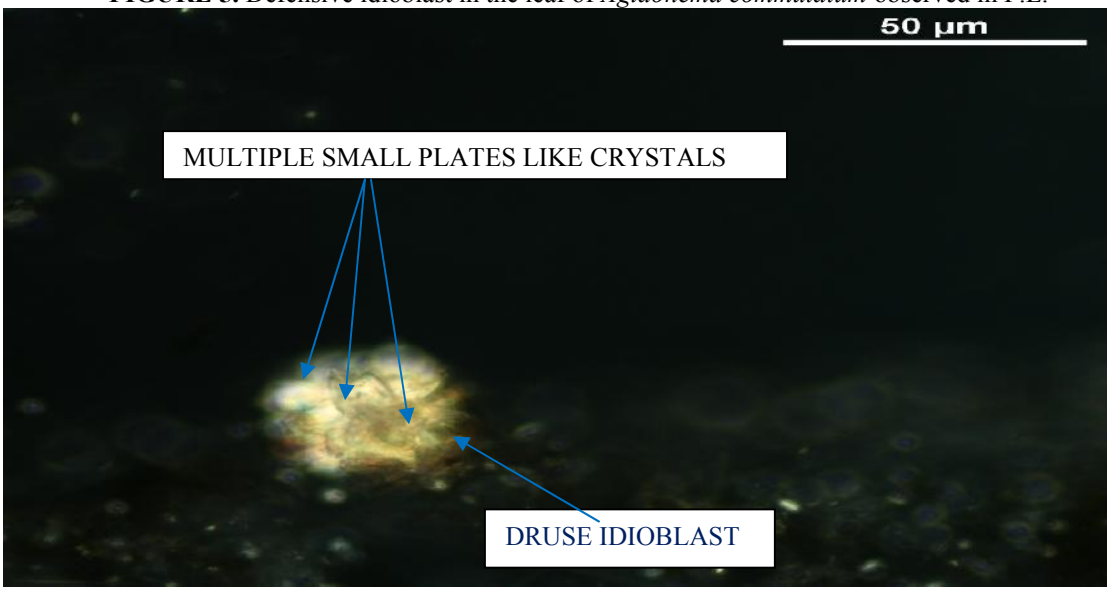

FIGURE 6. Druse in the leaf of *Aglaonema commutatum* observed in P.L.

DISCUSSIONS

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 *Aglaonema commutatum* 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 *Aglaonema commutatum*, i.e., their thick lateral walls [Fig.-4] and thin-walled terminal papillae [Fig.-4], and the location of these cells in the airspaces from which they are easily dislodged, are probably ecologically significant. The position of non-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 *Aglaonema commutatum*, wide variations in the numbers and densities of idioblasts are observed, among different leaves at the different stages of development [Fig.-7]. Comparing the ranges of raphide cell densities and druse cell densities, it is apparent that druse cell occurrence is especially variable [Fig.-7]. 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 Gingko (Arnott, 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 contain raphide cells. This suggests that raphide idioblasts are determined early on and that druse cell diffierentiation begins later in leaf development. The differentiation of raphide cells, especially the defensive type, is presumably a highly conserved phenomenon in *Aglaonema commutatum* leaves. The pattern of crystal cell distribution in *Aglaonema commutatum* leaves is different for raphide and druse idioblasts [Fig.-11]. 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.

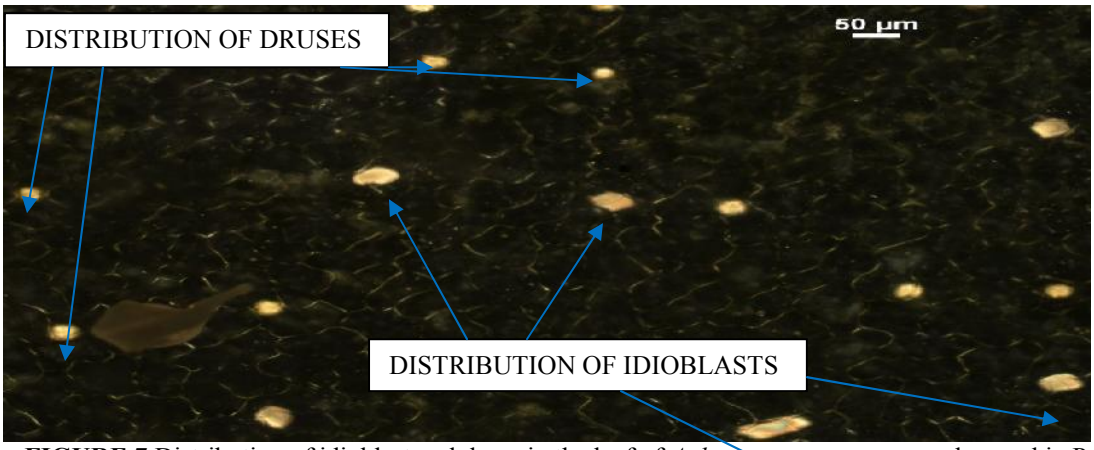

FIGURE 7. Distribution of idioblast and druse in the leaf of *Aglaonema commutatum* observed in P.L.

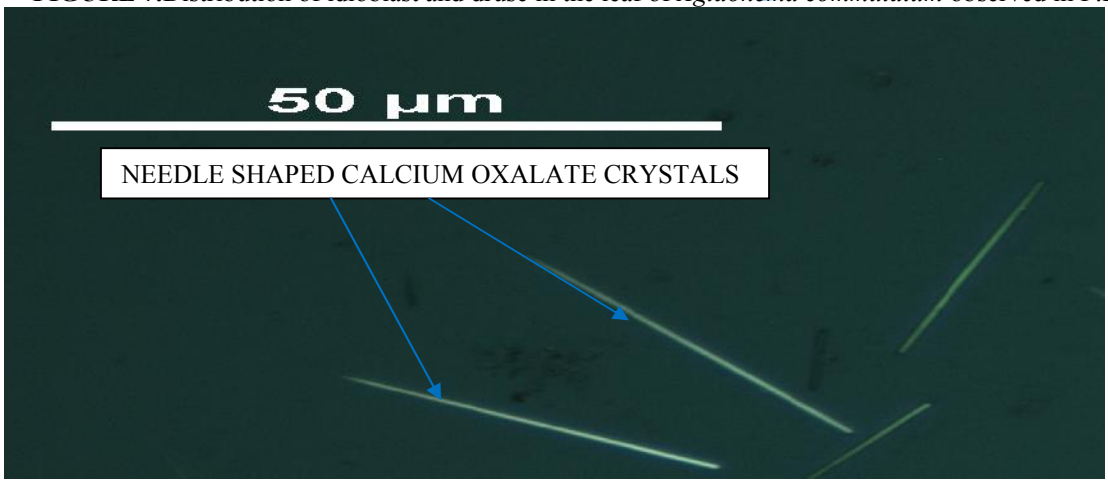

FIGURE 8. Calcium oxalate crystals in the leaf of *Aglaonema commutatum* observed in P.L.

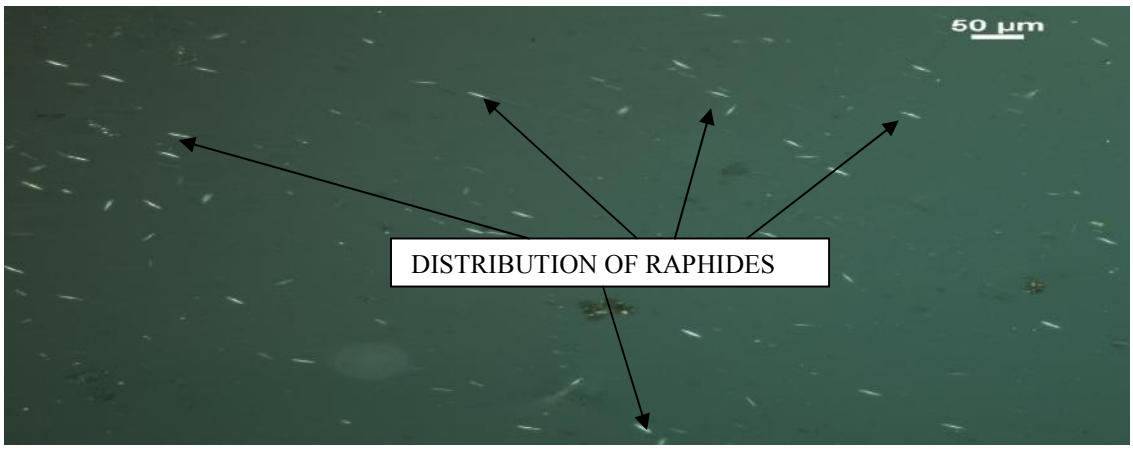

FIGURE 9. Distribution of Calcium oxalate crystals in the leaf of *Aglaonema commutatum* observed in P.L.

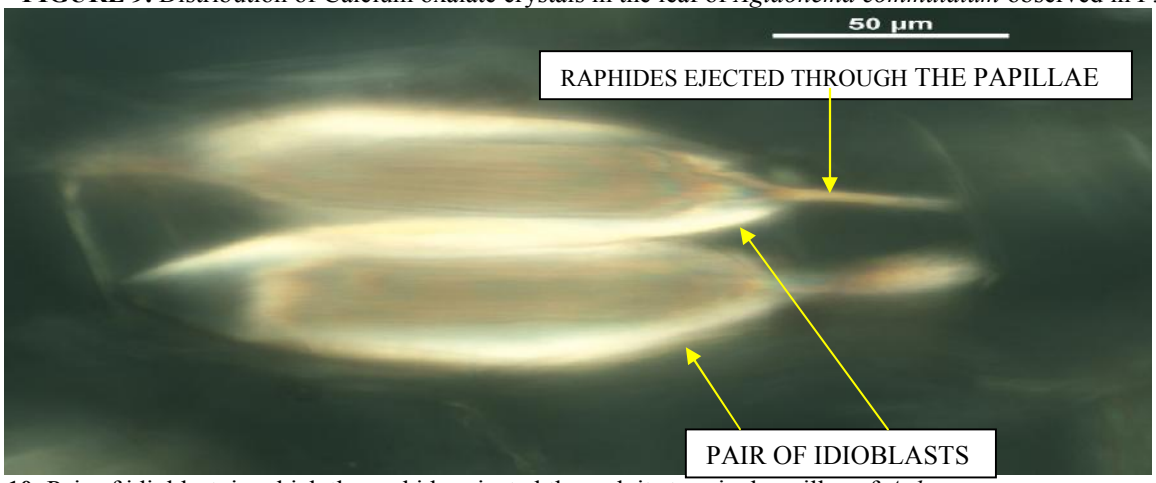

FIGURE 10. Pair of idioblast in which the raphides ejected through its terminal papillae of *Aglaonema commutatum* observed in P.L.

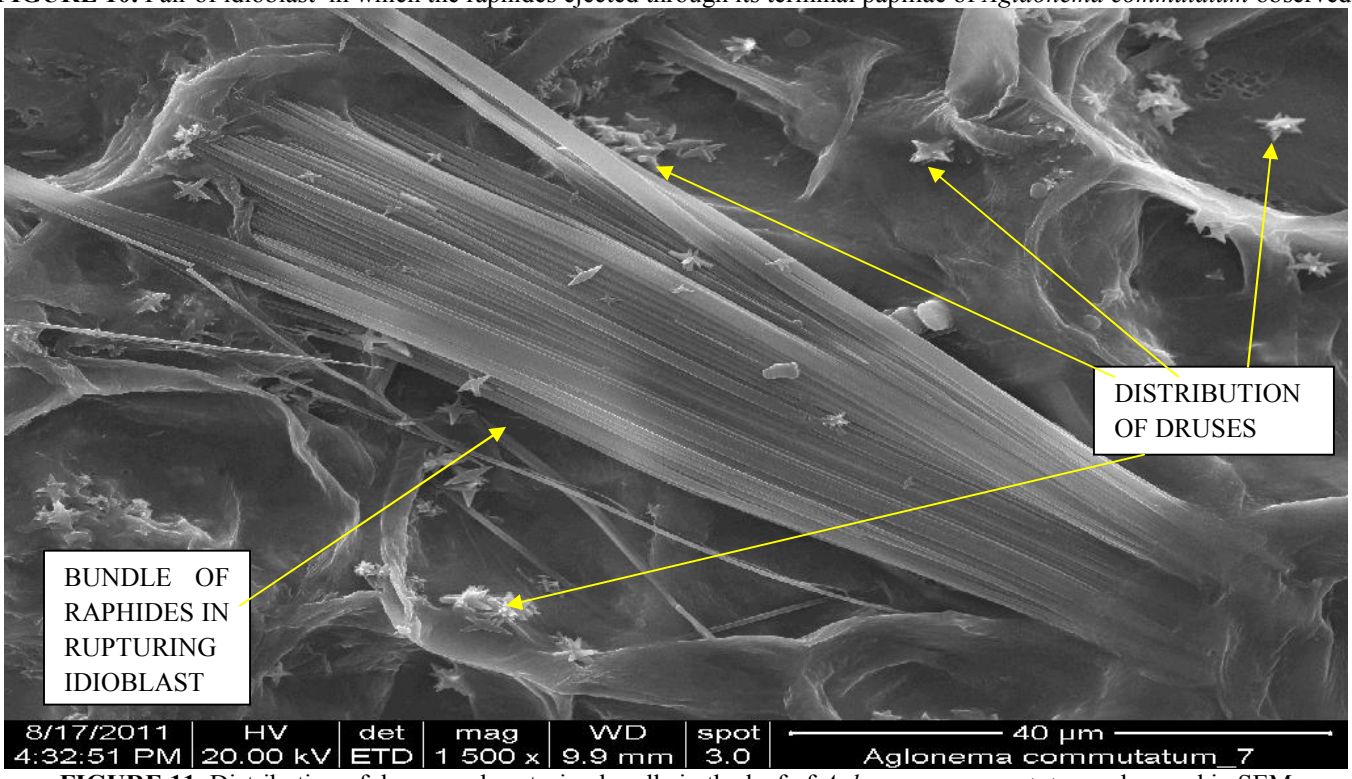

FIGURE 11. Distribution of druses and rupturing bundle in the leaf of *Aglaonema commutatum* observed in SEM

CONCLUSION

The densities of both raphides and druses idioblast is depend up on the different stages (young, medium & 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 and 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.

ACKNOWLEDGEMENT

I would like to express my gratitude to my Supervisor Dr. Amal Kumar Mondal, Associate Professor of Botany, Plant Taxonomy, Biosystematics and Molecular Taxonomy Laboratory, Department of Botany & Forestry, Vidyasagar University, Midnapore, West Bengal, India and Dr. Sanjukta Mondal (Parui), Associate Professor, WBES, Department of Zoology, Lady Brabourne College, Kolkata-17, West Bengal for their constant help, encouragement. I would like to acknowledge for the Financial Support, University Grants Commission, in the form of Maulana Azad National Fellowship.

REFERENCES

Al-talib, K. H. and Torrey, J.G. (1961) Sclereid distribution in the leaves of *Pseudotsuga* under natural and experimental conditions. Amer. J. Bot. 48: 71- 79.

Arnott, H. J. (1973) Plant calcification. In I. Zipkin [ed.], Biological mineralization, pp. 609-927.John Wiley & Sons, New York.

Eckerton, S. (1900) The number and size of the stomata. Bot. Gaz. 46: 221-224.

Frank, R. (1967) Zur Bildung des Kristallidioblasten-musters bei *Canavalia ensiformis* DC. I. Z. Pflanzen-physiol. 58: 33-48. ,

And Jensen, W. A. (1970) On the formation of the pattern of crystal idioblasts in *Canavalia ensiformis* DC. IV. The fine structure of the crystal cells. Planta 95: 202-217

Gupta, B. (1961) Correlation of tissues in leaves. 2. Absolute stomatal numbers. Ann. Bot. 25: 71-77.

Knecht, G. N. and O leary, J. W. (1972) The effect of light intensity on stomate number and density of *Phaseolus vulgaris* L. leaves. Bot. Gaz. 133: 132-134.

Morren, E. (1863) Determination d u nombre des stomates chez quelques vegetaux indigenes ou cultives en Belgique. Bull. Acad. R. Sci. Belg., Ser. 2, 16: 489- 509.

Paupardin, C. (1965) Surla morphologie des tissues d'aubepine (*Crataegus monogyra* Jacq.) cultives in vitro et la possibilite pour ces tissues d'ultiliser l'oxalate de calcium comme substance de reserve, pp. 379-389. 90° Congr. Soc. Savantes, Nice.

Saadi, S.M.A.I. and Mondal A.K. (2011) Studies on the calcium oxalate crystals of some seleted aroids (Araceae) in Eastern India. Advances In Bioresearch.Vol.2:134-143

Sachs, T. (1974) The developmentalo rigin of stomate pattern in *Crinium*. Bot. Gaz. 135: 314-318.

Sakai, W. S. and Nagao, M. A. (1980) Raphide structure in Dieffenbachia maculata. J . Amer. Soc. Hort. Sci. 105: 124-126.

Sarma, S. K. and Terpo, A. (1980) The occurrence of different types of calcium oxalate crystals in *Allium cepa* L . and *Allium fistulosum* L . and their importance in taxonomy. Acta. Agron. Acad. Sci. Hung. 29: 25- 37.

Sharma, G. K., Chandler, C. and Salemi, L. (1980) Environmental pollution and leaf cuticular variation in Kudzu (*Pueraria lobata* Willd.) Ann. Bot. 45: 77- 80.

Sunel, L. A. and Healey, P. L. (1979) Distribution of calcium oxalate crystal idioblasts in corms of taro (*Colocasia esculenta*). Amer. J. Bot. 66: 1029-1032

Sunel, L. A. and Healey, P. L. (1982) Distribution of calcium oxalate crystal idioblasts in leaves of taro (*Colocasia esculenta*). Amer. J. Bot. 72: 1854-1860

Timmerman, H. A. (1927) Stomatal numbers: their value for distinguishings pecies. Pharm.J. 118: 241-243.

White, J. B. and Beals, H. O. (1968) Variation in number of resin canals per needle in ponderosa pine. Bot. Gaz.124:251-253.

Wheeler, G. E. (1979) Raphide files in vegetative organs of *Zebrina*. Bot. Gaz. 140: 189-198.