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The density and distribution of calcium oxalate crystals in reproductive organs of wild taro (Colocasia Esculenta (L.) Schott)

Vo Thi Thanh Phuong
Department of Biology, School of Education, CanTho University, Vietnam

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Abstract
The study's aim was to investigate the density and the distribution of calcium oxalate crystals in reproductive organ of "green" morph type of wild taro (Colocation esculent (L.) Schott). Reproductive organs of "green" wild taro included flowers, fruits, and seeds. Morphological features of wild taro were evaluated based on the physiognomic characteristics of fresh specimens. Reproductive organs of "green" type were sliced thin horizontally and vertically, and double-stained with carmine alum claque–iodine green dye. The results showed that three types of crystals (druse, rawhide bundle, and free needle-liked crystals) were detected at wild taro. Calcium oxalate crystals, depend on types, were endowed very frequently at spandex pith, female flower, male flower, fruit, and seed. Druses were located in parenchyma and collenchyma cells, and present in almost parts of reproductive organs except ovule, placenta, and seed. Druse crystals were founded dense on the top of synandrium. Rawhide bundles were observed in shape of box, before or biforine-liked cells, and often located in parenchyma or collenchyma area. Free needle-liked crystals were dispersed scatteredly or gathered in clusters. It presumed that three types of crystals in reproductive organs played an important role in self-protection against herbivorous animals or insects.

Keywords: colocasia esculenta (L.) Schott, calcium oxalate crystals, reproductive organs

Introduction

Wild taro, a natural type of *Colocassia esculenta* (L.) Schott, has little studied and is poorly known (Matthews *et al*., 2012) [24]. Wild taro thrives in freshwater slough and riparian areas. Wild taro is recorded widespread from India to Southeast Asia and the western Pacific; Eastern Asia; New Guinea, and Northern Australia (Matthew, 1991; Matthew & Naing, 2005) [21, 23]. However, it is considered to be an emerging invasive plant in many countries such as U.S. (Driescher *et al*., 2002; Everitt *et al*., 2007; Moran and Yang, 2012; Annual report, 2017) [7, 9, 25, 1], Australia (Matthew, 2003) [22], Southern Europe (García-de-Lomas *et al*., 2012; Dana *et al*., 2017) [13, 6], and New Zealand (Parshotam, 2018) [30]. According Ivancic and Lebot (1999) [16], based on morphological characteristics, wild taro is separated into three distinct morphotypes: "purple", "green" and "green with purple vein junction on lamina". From ancient times, wild taro has valid as a vegetable and traditional medicine source. The leaf juice of wild taro is useful in cure of internal haemorrhages, otalgia, otorrhoea, adenitis and buboes and the corm juice is used to treat somatalgia, alopecia areata, haemorrhoids and congestion of the portal system (Namrata *et al*., 2011) [28]. In South-east Asia, leaf of wild taro is popularly used as a green fodder for pigs in Philippine (Matthew *et al*., 2012) [24], Myanmar (Matthew & Naing, 2005) [23], and Vietnam (Hang & Preston, 2010). Besides, wild taro corn meal as an energy source for broilers (Samarasinghe and Rajaguru, 1992). Leaf and flower of wild taro are used as wild edible vegetable and medical herb in India (Yesodharan and Sujana, 2007; Kar and Borthakur, 2007; Kar *et al*., 2013) [19, 18] and Malaysia (Sulaini and Sabran, 2002). In Thailand, Vietnam and Campodia, young leaf is picked for vegetable cuisine (Sungkajanttranon *et al*., 2019; Ogle, *et al*., 2003; Buntha, *et al*., 2020) [36, 29]. Especially, peduncle and spathe of inflorescences are popularly used to salting, sour soup, and curry in Malaysia (Matthews & Naing, 2005) [23]; soup, sauce, packet in Philippine (Matthew *et al*., 2012) [24]. However, it is recommended that leaves, stolons and inflorescences need pre-processing to destroy the acridity that cause swelling of lip, mouth and throat before utilization (Bradbury & Nixon, 1998) [2]. The acridity of wild taro results from calcium oxalate crystals that are presented in the different parts such as blade, petiole, corm, and root (Saadi and Mondal, 2012) [33]. For human and animal body, calcium oxalate causes a toxic effect when accumulating in excess levels (Franceschi and Horner, 1980) [11]. The high quantities of dietary oxalate is counterproductive as an antinutrient or as pathogenesis agents of hyperoxaluria (Nakata, 2012) or urolithiasis (Vijaya *et al*., 2013). Five forms of calcium oxalate crystal are discribed at plants as raphide, druse, styloid, prism and sand (Konyar *et al*., 2014) in which raphide and druse are commonly present in Araceae (Keating, 2004). Oxalate crystals are distributed in stem, leaf, root and flower (Franceschi and Horner, 1980; Tilton and Horner, 1980; Horner and Wagner, 1985; Prychid and Rudall, 1999) [33, 31]. There are not many studies on calcium oxalate crystals in flower. Raman *et al*. (2017) [32] described calcium oxalate crystals related to floral anatomy of *Amorphophallus titanum* (Araceae).

Coté and Gibernau (2012) [5] are investigated the distribution of calcium oxalate crystals in floral organs (*e.g.* tepals, female organ, male organ, sterile organ, appendix, and spadix pith) of 21 aroid species but except *C. esculenta*. Coté (2009) [4] studied calcium oxalate crystals on both vegetative and reproductive organs of *Diefenbachia seguine* (Araceae). At *C. esculenta (taro)*, calcium oxalate crystals are identified in leaf (Eco & Belonias, 2017; Sunell & Healey, 1985) [8, 35], and petiole (Ezeabara *et al*., 2015) [10].

There has little studied on calcium oxalate crystals at reproductive organs of wild taro.

According to Ivancic and Lebot (1999) [16], the flowering of "green" morphology population of wild taro is low. Approximately 0.5 - 4% of plants are flowering. Each cluster develop two to four inflorescences. Each inflorescence of green wild taro is made up of peduncle, spathe and spadix. The longest spathe was 44.20 cm and the longest spadix was 28.50 cm. Four parts of the spadix were female portion, neutral region, male portion and sterile appendix. The corresponding average length of female portion, male portion and sterile appendix were 5.25 cm, 1.5 cm and 9.95 cm. The average ratio between the length of the sterile appendix and the length of the male portion was 1.50. Ivancic *et al.* (2011) [17] was described female flowers of taro *C. esculent* as sessile and green. The male part of the spandex consists of sessile staminate flowers that are connoted into a synandrium. The apex of the synandrium is mostly flat and hexagonal. The pollen grains are spherical

The objects of study were the density and distribution of calcium oxalate crystals in the reproductive organs of wild taro (*Colocasia esculenta* (L.) Schott).

Materials and methods

Collection of plant materials

Wild taro in this research was "green" morphotype (*Colocasia esculenta* (L.) Schott) that were collected at Can Tho province, Mekong Delta, Viet Nam. Reproductive organs of "green" wild taro included flowers, fruits, and seeds. All specimens were washed to remove dust and dried under room temperature.

Morphological and anatomical study

Morphological study

Morphological features were evaluated by physical observation and description about the physiognomic characteristics of fresh "green"-typed specimens.

Anatomical study Reproductive organs of "green" type were sliced thin horizontally and vertically as following discribed. For peduncle, segment three regions (the top, the midlle and bottom), split vertically to the edge 5 - 8 mm and cut thin slices horizontally. For spadix pith, after removing female and male flowers, spadix pith were cut the same manner as peduncle. Flower spathe and fruit spathe were cut cross thinly into slices with each 5 - 8 mm long. Female flower and male flower (synandrium) were cut into horizontal and vertical slices. Fruit and seed were cut lengthwise into thin slices. All slices were double-stained with carmine alum laque–iodin green dye. Carmin stains pink cellulose cell wall and green iodine stains green carpentry cell wall. A Olympus light microscope was used to view the slices. Microphotographs were obtained by a digital camera attached on the microscope eyepiece.

Method of investigating the density and distribution of crystals

Measure 1 mm^2 of the specimen area with microscope micrometers. The density of crystals was investigated by counting the number of each type of crystals per 1 mm^2 unit of specimen area.

Table 1: Conventions of calcium oxalate crystal density

Number of crystals/ 1 mm^2	Density	Symbol
0	none	–
1 - 10	Rarely/ Scatteredly	+
10 - 30	Sometimes/ Occationally	++
30 – 60	Often/ Frequently	+++
> 60	Always/ Very frequently	++++

Results and discussion

Disciption of wild taro's reproductive organs

The morphological type of wild taro in this research had the whole blade and petiole green, called "green" morphotype. There were one to a few inflorescences in each wild taro bush (Figure A). Each inflorescence consisted of peduncle, spathe and spadix (Figure B). The spathe was covered spadix and included two parts: the below part was green, short, soft, and covered the female portion; the top part was yellow, thin, head-tapered, and covered the remain of spadix. Four portions of spadix were a female portion (lowest), a sterile region, a male portion and an appendix (highest) (Figure C). Female portion consisted of many green and sessile female flowers which were attached around the flower-bearing axis or female spadix pith. Male portion consisted of many male flowers attached around male spadix pith. Each male flower was made up of 6 filaments that were connated into synandrium. The connated synandrium was called the stout connectives. The sterile region was a narrow part that separate the female portion and the male portion. The appendix had no flowers attached on. Fruits were attached on fruit-bearing axis or fruit spadix pith (Figure D). The fruit is watery, spherical shape and contains many seeds. Seeds was yellow-brown in color and had longitudinally slots.

Fig 1: Reproductive organs of "green" morphotype. A. Wild taro with flowers; B. Flower (1. Young, 2. Mature, 3. Flower with peeled spathe); C. Spadix and peeled spathe; D. Fruit (1. Young, 2. Fruit with peeled spathe). *Abbreviations: pe - peduncle, spat - spathe, spad - spadix, fe - female portion, ste - sterile region, ma - male portion, ap - appendix, fru - fruit, fru spat - fruit spathe. Scale: 20 cm.*

Distribution of calcium oxalate crystals in the flower of wild taro

Three types of calcium oxalate crystals investigated in the flower of "green" morphotype were druse, raphide bundle, and free needle-liked crystals. The density of crystals in flower varied depend on each portion and their own anatomical features. Some cases, the different slices of the same reproductive portion could showed differences in terms density. In generally, the crystal density was summarized in table 2.

Druses were found sparcely in peduncle and spadix pith; occasionally in female portion and sterile region; frequently in spathe and male flower; very frequently in appendix.

Raphide bundles were present scattered in spathe; occasionally in peduncle; frequently in sterile region, male flower, and appendix; very frequently spadix pith and female flower. Free needle-liked crystals were scatteredly detected in peduncle and spathe; occasionally in spadix pith and appendix; very frequently in female flower, sterile region, and male flower.

Table 2. The density of calcium oxalate crystals in flower

Crystals	Peduncle	Spadix Pith	Green Spathe	Yellow Spathe	Female Flower	Sterile Region	Male Flower	Appendix
Druse	+	+	+++	+++	++	++	+++	++++
Raphide bundle	++	++++	+	+	++++	+++	++	+++
Free needle-liked	+	++	+	+	++++	++++	++++	++

Peduncle: Druse crystals were scatteredly presented in parenchyma cells with various sizes. They distributed individually or sometimes in pairs. Numerous raphide bundles were also present in parenchyma region and almost located at the centre of peduncle section. Raphide bundles were different in size and shape, and tended to be invasive into air space. Some idioblasts containing raphide bundle were oval in shape and larger than surrounding parenchyma cells. Free needle-liked crystals were dispersed throughout the peduncle section including the epidermis and parenchyma region (but not seen at low magnification). They were mostly observed near by protruded or broken raphide bundles (Figure 2).

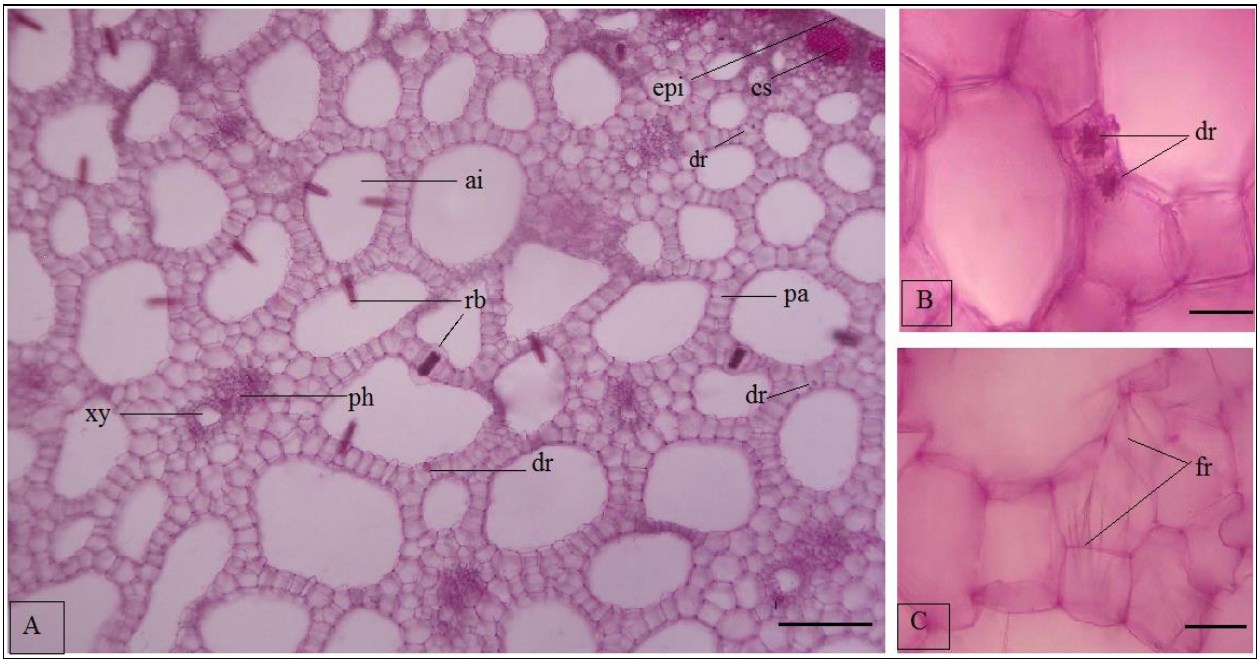

Fig 2: Distribution of calcium oxalate crystals in cross section of peduncle at different magnification. *Abbreviations: epi – epidermis, cs - colerenchyma strand, pa – parenchyma, ai – air space, xy – xylem, ph – phloem, rb – raphide bundle; dr – druse, fr – free needle-liked crystal; Bar: A = 100 µm; B and C = 20 µm.*

Spadix: Spadix pith of all three female, sterile, and male portions were presented calcium oxalate crystals. Druse crystals were scattered at the collenchyma area but with low density at the central part of section.

The raphide bundles were densely endowed throughout the parenchyma cells. Mostly raphide bundles appeared to be biforine or biforine-liked cells.

Biforine were identified as spindle-shaped idioblasts with terminal nipples, capable of expelling their crystals out. However, their density at the spadix pith of female region was higher than that of the spadix pith of both male and sterile region.

Free needle-liked crystals were rare and distributed nearby the broken raphide bundles (Figure 3).

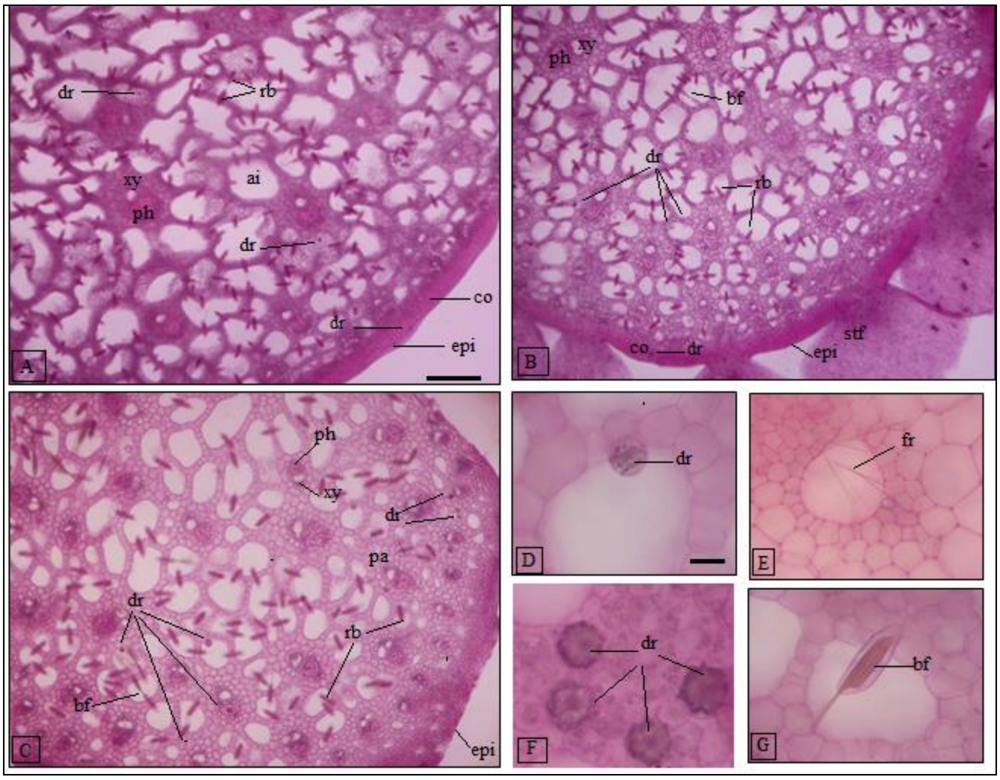

Fig 3: Distribution of calcium oxalate in spadix pith. A. Spadix pith of female portion; B. Spadix pith of sterile region; C. Spadix pith of male portion. Crystals in spadix pith of female spadix pith (D), sterile spadix pith (E), and male spadix pith (F, G). *Abbreviations: bf – biforine/biforine-liked cells, stf – sterile flower; others as in Figure 2. Bar: A = 100 µm, D = 10 µm.*

Spathe: At both green and yellow parts of flower spathe, many druse crystals were present in the parenchyma cell and collenchyma strand nearby the upper epidermis in one by one or pairs.

From the central region to the lower epidermis, the druse crystals were rarely observed. Raphide bundles in shape of biforine or biforine-liked cells were scatteredly distributed, but mostly concentrated at the central region. Some large idioblasts containing rectangular-shaped raphide bundle were found. Free needle-liked crystals were rarely found at the central parenchyma region (but not seen at low magnification) (Figure 4).

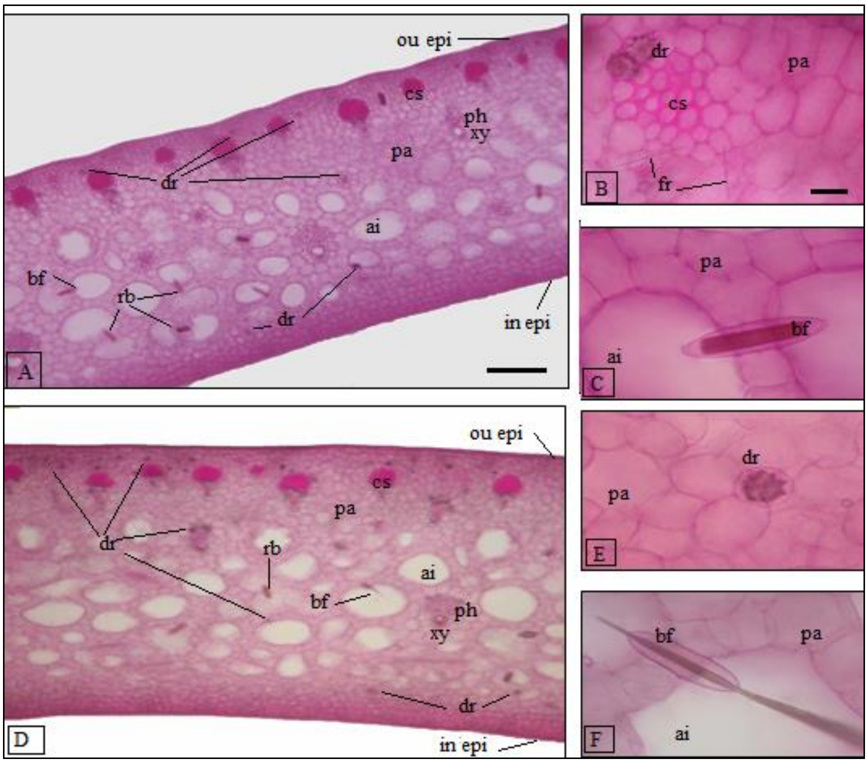

Fig 4: Distribution of calcium oxalate crystals in spathe. A. Green spathe, D. Yellow spathe. Calcium oxalate crystals in green spathe (B, C) and in yellow spathe (E, F). *Abbreviations: ou epi - outer epidermis, in epi - inner epidermis; others as in Figure 2, bf – biforine/biforine-liked cell. Bar: A = 100 µm; B = 20 µm.*

Appendix: At the appendix, many druse crystals were arranged rows below the epidermic layers, distributed abundantly at the inner parenchyma region but scatteredly at the central colenchyma. Free needle-liked crystals are scattered throughout the section. The deeper into the central colenchyma, the less density the free needle-liked crystals was. At the outer area of appendix, raphide bundles were box-shaped contained inside the large idioblasts. In the center of appendix, biforine or biforine-liked cells were found and invasive in the air space (Figure 5).

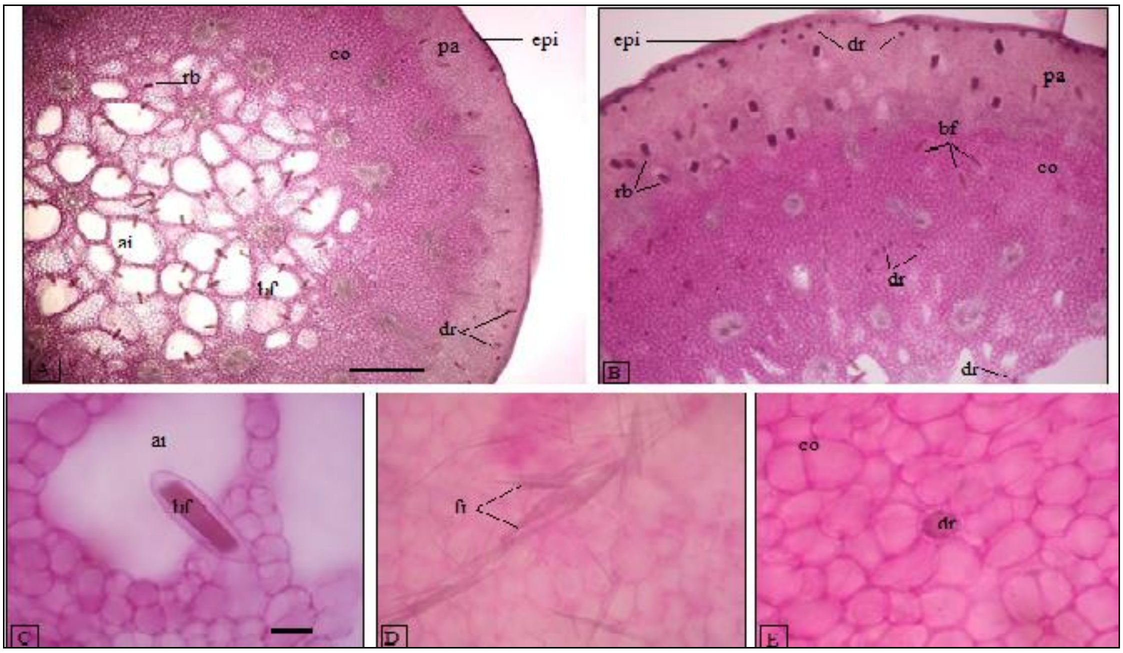

Fig 5: Distribution of calcium oxalate crystals in appendix (A, B). Calcium oxalate crystals in appendix (C, D, E). *Abbreviations: co – colenchyma; others as in Figure 2. Bar: A = 100 µm, C = 20 µm*

Female flower: Longitudinal and cross section of female flower showed three type of crystals (druse, raphide bundle and free needle-liked crystals) that were different in density and distribution. At the stigma and style, there were scattered druse crystals and no raphide bundle observed. A large number of raphide bundles were attached on ovary wall. Free needle-like crystals were also dense on ovary wall (but not seen at low magnification). On the oval ovules and placenta, raphide bundles were scatteredly attached. Howerver, there were no druses were detected on these two parts (Figure 6).

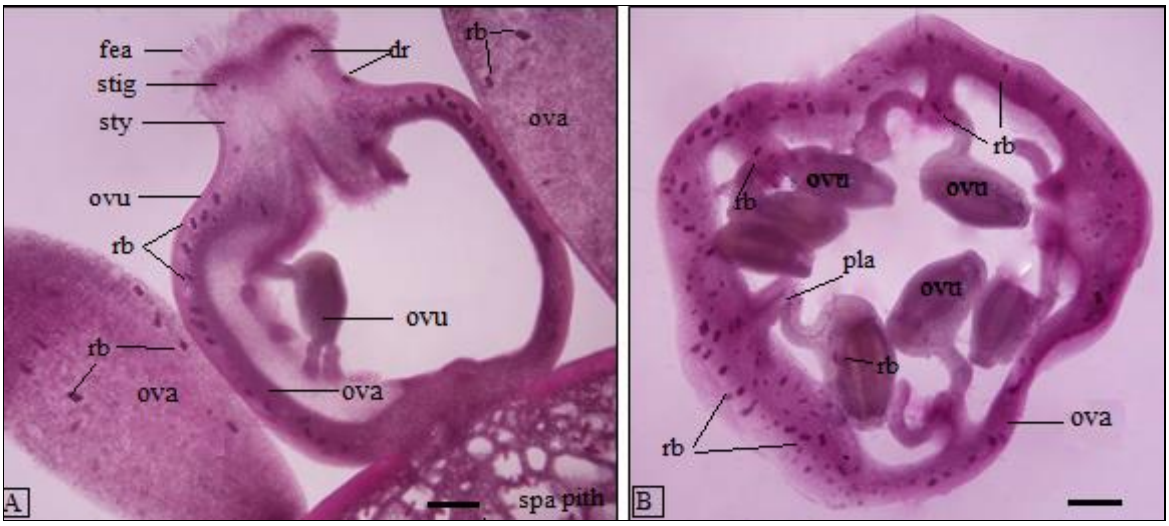

Fig 6: Distribution of calcium oxalate crystals at female flower. A. Longitudinal section; B. Cross section. *Abbreviations: fea – feather, stig - stigma, sty - style, ovu - ovule, ova - ovary, spa pith - spadix pith of female portion; pla – placenta; others as in Figure 2. Bar: A, B =100 µm.*

Sterile flower: At the sterile flowers or staminodia, a few druse crystals were investigated in the outer and centre of parenchyma area. Free needle-liked crystals presented dense throughout parenchyma cells. At some sections, free needle-liked crystals clustered into a dense group. Large box-shaped raphide bundles containing in idioblasts were concentrated more at the outer parenchyma regions than the bottom of flower (Figure 7).

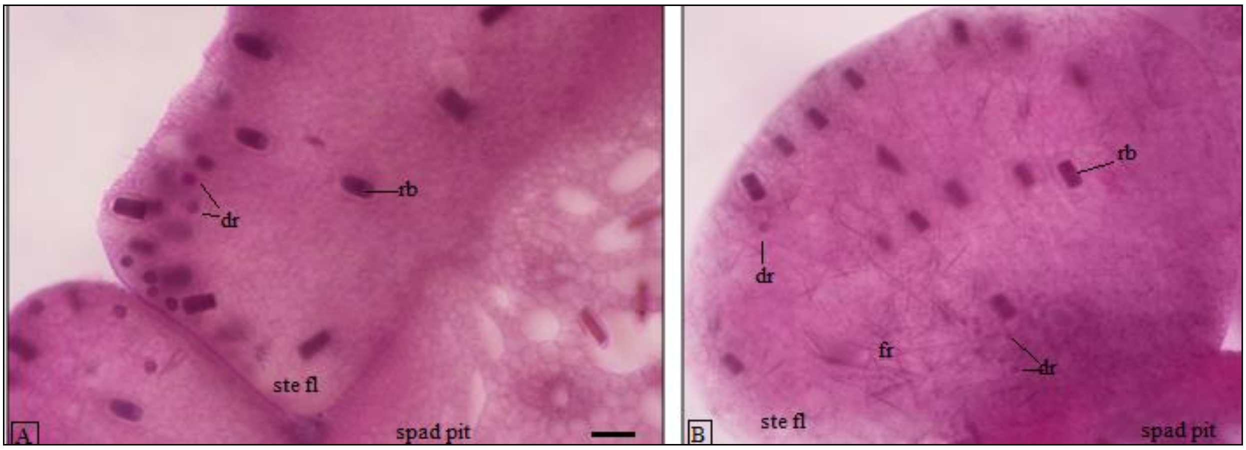

Fig 7: Distribution of calcium oxalate crystals in the longitudinal section of sterile flower. *Abbreviations: ste fl - sterile flower, spad pith - spadix pith of sterile region, others as in Figure 2. Bar: A = 50 µm.*

Male flower: At "green" morphotype, male flower had no petals and sepals. Filaments of stamens in flower fused into synandrium. Calcium crystals mostly were concentrated at the stout connectives and anther walls. The longitudinal section of male flower showed that druses were abundance in the top of synadrium, and sparcely in the anther wall (Figure 10A). Different cross sections of synandrium showed diferent distribution of calcium oxalate. The top of synandrous cross section, the druse crystals were dense and raphide bundles were scattered (figure 10B). At the lower cross section of the synandrium, druse crystals were converged at the center, free needle-liked crystals were seen throughout the section, and the raphide bundles were occationally at the peripheral section. Raphide bundles were located in large idioblast with horizontal increase in size (Figure 10C).

The base of synadrium was rarely showed the three types of crystals.

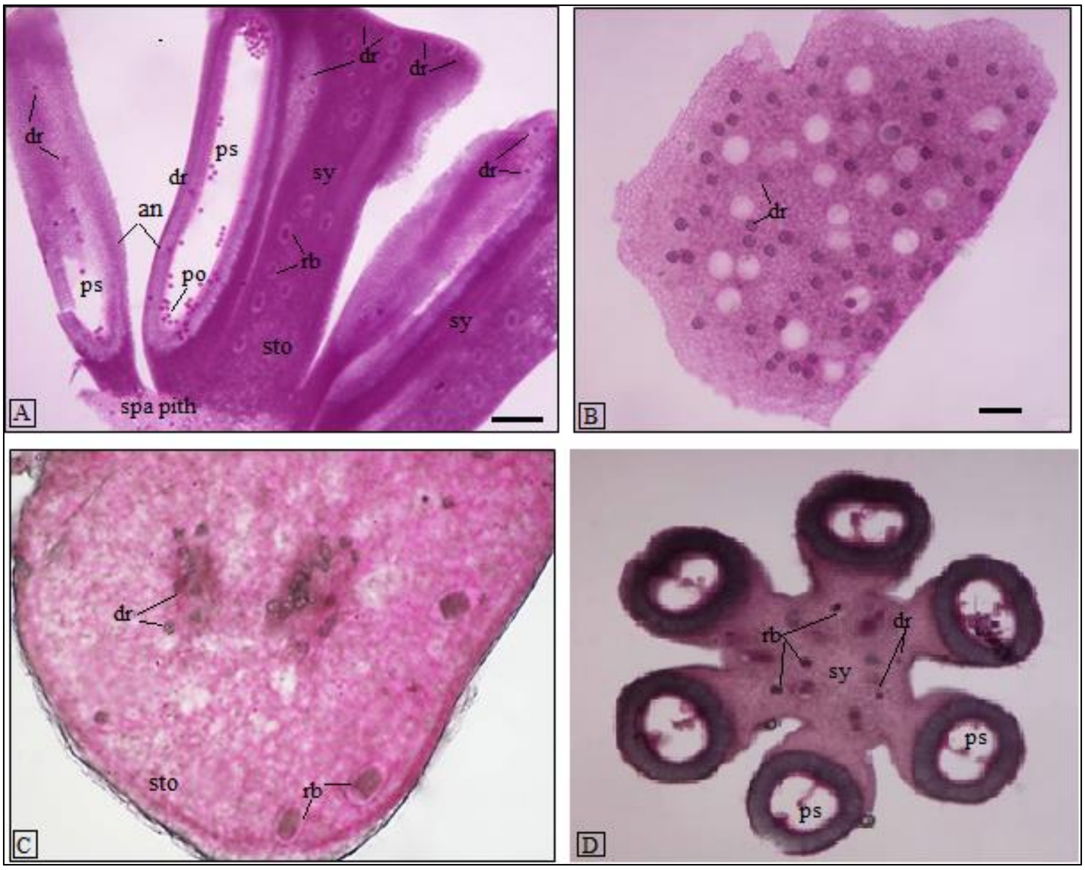

Fig 8 : Distribution of calcium oxalate crystals in male flower. A. Longitudinal section of synandrium, B. Cross section of the top of synandrium, C. Cross section of the stout connectives, D. Cross section of the base of synandrium. *Abbreviations: po - pollen, sto – stout, ps: pollen sac, others as in Figure 2. Bar: A = 100 µm, B = 50 µm.*

3.2. Calcium oxalate crystal distribution in fruits and seeds

The density of druse, raphide bundle and free needle-liked crystals in fruit and seed was showed on table 3. Druses were not detected in seed, but presented scatteredly in spadix pith of fruit, occasionally in fruit wall, and frequently in fruit spathe. Raphide bundles were present occasionally in fruit spathe; very frequently spadix pith of fruit, fruit wall, and seed.

Free needle-liked crystals were distributed occasionally in fruit spathe; frequently in spadix pith of fruit; very frequently in fruit wall, and seeds.

Table 3: Distribution of calcium oxalate crystals in fruits and seeds

	Spadix pith of fruit	Fruit spathe	Fruit wall	Seed
Druse	+	+++	++	–
Raphide bundle	++++	++	++++	++++
Free-needle	+++	++	++++	++++

Spadix pith of fruit: After fertilization, the flower spadix pith of sterile region, male portion, and appendicular portion were atrophy and degeneration. The spadix pith of flower female portion continued to grow into spadix pith of fruit. The cross section of spadix pith of fruit showed that druse crystals were rare in parenchyma cells, free needle-liked crystals were frequent in parenchyma region under epidermis or near broken/protruded raphide bundles. Raphide bundles were presented in abundance in shape of biforine and biforine-liked cells. Many raphide bundles were protruded either side into air space. The number of protruded raphide bundles in fruit spadix pith were more than that of flower spadix pith.

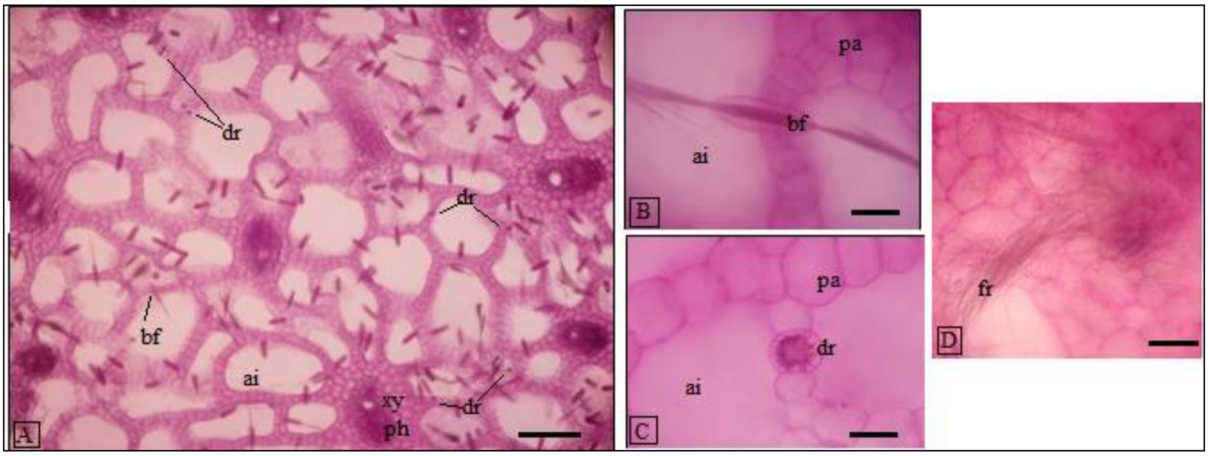

Fig 9: Distribution of calcium oxalate crystals in the spadix pith of fruit. A. Cross section of the spadix pith of fruit; B. Biforine, C. Druse, D. Free needle-liked crystals. *Abbreviations*: as in Figure 2. Bar: A = 100 µm; B, C, D = 20 µm.

Fruit spathe: The fruit spathe were remained developing from the green flower spathe, while the yellow spathe was dried and shrinked. The cross section of the fruit spathe represented all three types of druse, raphide bundle and free needle-liked crystal, which their distribution was the same manner in the green flower spathe. Druse crystals were abundance at region under the outer epidermis, and rarely in the parenchyma region below. At the central parenchyma region, raphide bundles, in shaped of biforin or biforine-liked cells, and free needle-liked crystals were presented scattered (Figure 12).

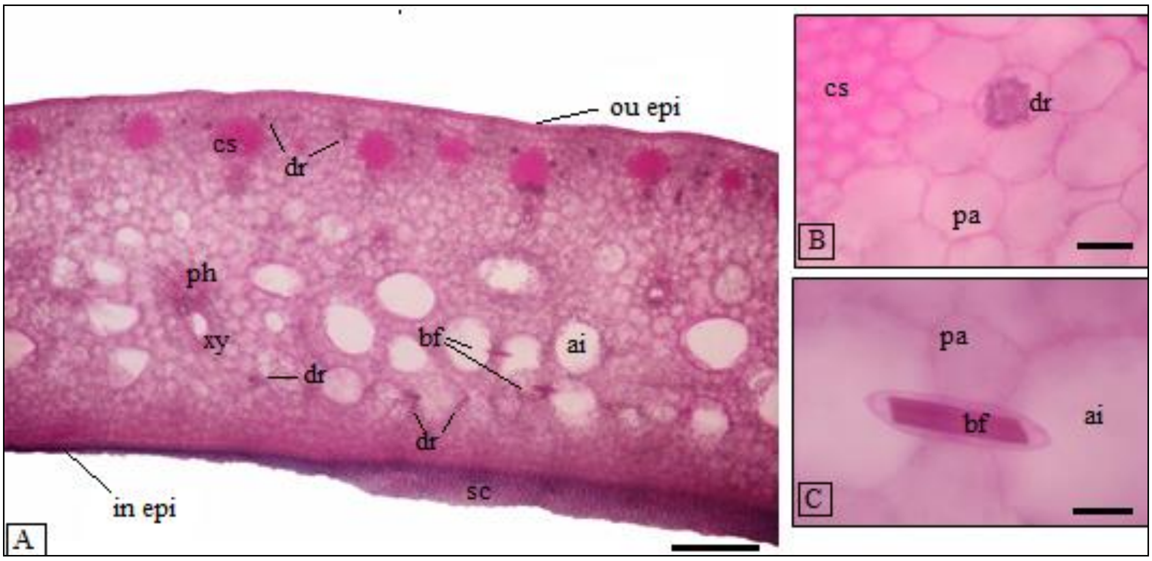

Fig 10: Distribution of calcium oxalate crystals in fruit spathe. A. Cross section of fruit spathe; B. Druse, C. Biforine-liked cell. *Abbreviations: ou epi - outer epidermis, in epi - inner epidermis, sc - sclerenchyma, others as in Figure 2. Bar: A = 100 µm; B, C = 20 µm.*

Fruit: At the longitudinal section of fruit, the druses were rarely observed in the persistent stigma and style. At mesocarp, raphide bundles and free needle-liked crystals were distributed as dense as at ovary wall (free needle-liked crystals not seen in low magnification). Raphide bundles were mostly present in large idioblasts (Figure 13 A)

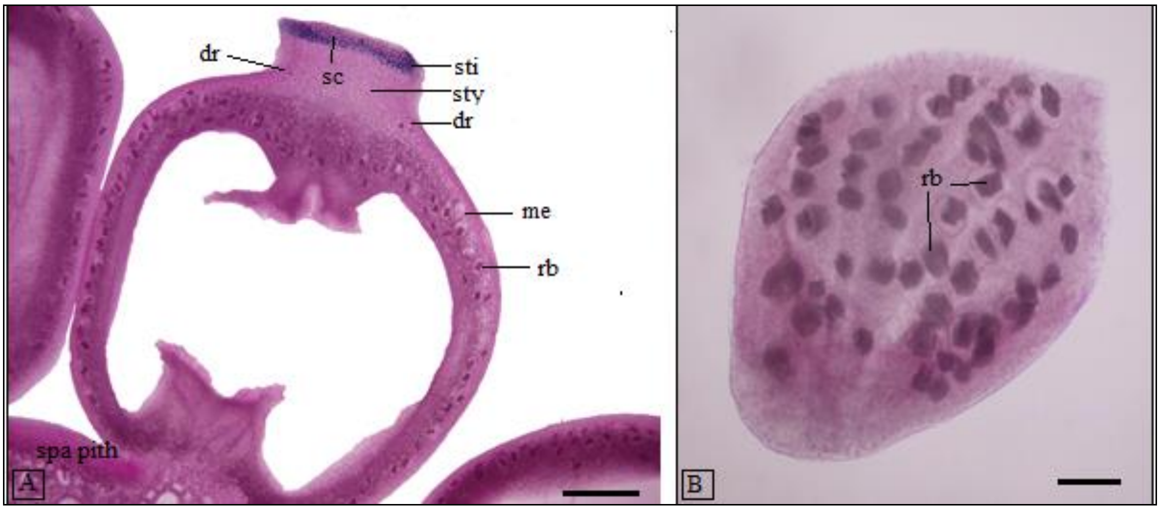

Fig 11: Distribution of calcium oxalate crystals in fruit and seed. A. Longitudinal section of fruit, B. Longitudinal section of seed. *Abbreviations: sc – sclerenchyma, me – mesocarp, others as in Figure 2. Bar: A, B = 100 µm; B = 50 µm*

Seed: Both free needle-liked crystals and raphide bundles were observed dense throughout the longitudinal section. Almost raphide bundles had large size. Some raphide bundles possessed width greater than the length, and contained in large idioblasts. No druse crystals were detected in this organ (Figure 13B). According to Meric (2009), mature seeds are rich in nutrients. The presence of calcium oxalate crystals in this organ plays an important role in protecting against herbivore attack or increasing the metabolism of tissues in seed maturation. According to Konyar *et al*. (2014), five forms of calcium oxalate crystal detected in plants were raphide, druse, styloid, prism and sand. Raphide, druse and styloid were three of these crystals recorded in Araceae (Prychid & Rudall, 1999). They were detected at stem, leaf, root and flower. Druses and raphides were investigated in flower organs of *Amorphophallus titanum* (Raman *et al.*, 2017) [32], *Dieffenbacchia seguine* (Coté, 2009) [4] and 21 aroid species with various pollination strategies (Coté & Gibernau, 2012) [5]. In this research, the reproductive organs of "green" morphotype of wild taro (*C. esculenta*) possessed druse, raphide bundle, and free-needled crystals that vary in frequency and distribution. Druses were present in all structural parts of reproductive organs except ovules, placenta, and seeds. At the top of synandrium, druse crystals were founded dense. Druses were accumulated in parenchyma or collenchyma cell under epidermis to the central section. Raphide bundles in shape of box, biforine or biforine-liked cell were dense at the spathe, spadix pith, female flower, male flower, fruit, and seed. Idioblasts containing raphide bundles were larger than surrounding parenchyma cells. Several bundles had been ejecting needle crystals at either side into intercellular space. Free needle-liked crystals were distributed very frequently in female flower, sterile portion, male flower, spadix pith of fruit, fruit wall, and seed. Free needle-liked crystals dispersed individually or gathered in horde might have derived from the protruded raphide bundles or the rupture condition of raphide bundles by anatomically sliced manipulation. Free needle-liked crystals in this research was similar to raphide crystals that were described in *Amorphophallus titanum* (Prychid & Rudall, 1999) and in *Dieffenbacchia seguine* (Coté, 2009) [4]. Needle-shaped crystals were considered as an important indicator of aroids (Saadi and Mondal, 2012). At ovules, studies in other aroid species showed no crystals detected (Coté, 2009; Coté & Gibzernau, 2012; Raman *et al.*, 2017) [4, 32]. However, in this research, a few raphide bundles were scatteredly observed in ovules and placenta. After fertilization, ovules developed into seeds but many large size raphide bundles attached on. This may showed the process of accumulation of crystals occurred for reinforce protection. In anther wall, according to Coté & Gibernau (2012) [4, 5], the anther walls of 21 aroids species seemed to lack crystals of any kind. Some crystals assigned to the connective may have actually been in the wall of an adjacent anther. In this research, some druses were scatteredly observed on anther walls. The functions of crystals on anther made it possible for cracking of anthers (Horner and Wagner, 1985). Another function of calcium oxalate crystals in plants was protected themselves against herbivorous animals or insects (Coté and Gibernau, 2012; Franceschi and Nakata, 2005; Molano, 2001 and Ward *et al.*, 1997) [4, 5, 12, 26, 38]. Raphide bundles were a bunch of narrow, elongated needle-shaped crystals that caused itchy mouth if eaten. Druses have a similar defensive function to that of raphides because of their sharp points resulting in considerable irritation (Prychid & Rudall, 1999) [31]. The spathe were endowed with crystals to protect the inflorescence before anthesis and the developing inflorescence following. Biforines in spadix pith migh eject crystals to hurt herbivore mouth. Spadix pith of female portion denser than spadix of other portions by the need for more protection. The abundance of crystals on the wall of ovary, the top of synandrium, the connectives were to protected ovules, pollens and embryos that resulted in irritating animal's oral tissues when eaten. Prychid & Rudall (1999) [31] was illustrated that aquatic plants often had calcium oxalate crystals associated with aerenchyma tissue and sometimes projecting into air spaces. This research found that raphide bundles and druse crystals were invasive development into aerenchyma tissue of peduncle, spathe, spadix pith of flower, appendix, fruit spathe, and fruit spadix pith. Several biforines were observed expelling from either side into aerenchyma tissue.

Conclusion

Three forms of calcium oxalate crystals (druse, raphide bundle, and free needle-liked crystals) were found in the reproductive organs of "green" morphotype of wild taro

(*Colocassia esculenta* (L.) Schott). These crystals were almostly located in parenchyma and collenchyma region with various densities. Druse crystals were detected all portions of reproductive organs except ovules, placenta, and seed. At the top of synandrium, druse crystals were founded dense. Raphide bundles were observed differently in size and shape. At spadix pith, female flower, fruid, and seed, raphide bundles were detected very frequently. Many of crystals were found in forms of biforine that had been protruding crystals into air space. "Green" morphotype, aquatic plant, possessed aerenchyma tissue which raphide bundles and druses were observed invaded into. Free needle-liked crystals were dispersed individually or gathered in horde with different density throughout the sliced sections. Free needle-liked crystals could derived from the protruded raphide bundles or the rupture condition of raphide bundles by anatomically sliced manipulation. The exist of calcium oxalate calcium in reproductives organs of wild taro might related to self-protection mechanism.

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