

**DIVERSITY OF CHROMOSOME AND BIOLOGY  
OF THE FAMILY ARACEAE IN THAILAND**

**RATTANAVALLEE SENAVONGSE**

**A dissertation submitted in partial fulfillment of the requirements for  
the degree of Doctor of Philosophy in Biodiversity  
at Mahasarakham University**

**March 2018**

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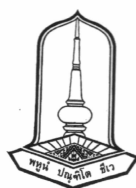
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### ABSTRACT

Diversity of Chromosome and Biology of the Family Araceae in Thailand found that the chromosomes numbers, karyotypes and idiograms of 20 Araceae species in Thailand is studied from roots tip by Feulgen squash technique. The chromosomes number is found  $2n$  (diploid) = 18, 24, 26, 28, 40, 42 and 58, respectively. Karyotype formula symmetrical (10 species) asymmetrical (10 species) are reported. In this study, nine species somatic chromosomes are found satellite at the end of the chromosome. The chromosomes numbers of seven species are recorded for the first time. Pollen grain of 18 Araceae species in Thailand are studied. The characters of all pollen are presented in monad, bilateral and radial symmetry, monoporate, diporate and monosulcate aperture. Three size groups of pollen grains include small, medium and large size are reported. Five shape groups of pollen grains include pheroidal, prolate, oblate spheroidal, subprolate and prolate shapes are reported. Six exine sculpturing groups of pollen grain are reported. The pollen grains of 16 species are recorded for the first time. Leaf epidermal cells are studied and found that like a jigsaw. The characteristic of all species are reported in rectangular to polygonal in form with smooth, undulate and sinuate anticlinal cell walls on adaxial and abaxial surfaces. The stomata on both surfaces are presented paracytic, hexacytic, anomocytic, cyclocytic with 2,3,4 subsidiary cells, 2,3,4,6 subsidiary cells and 2,3,4,5,6 subsidiary cells. Cuticle was presented smooth on both adaxial and abaxial surfaces. Solitary crystal is presented on the adaxial and abaxial surfaces of three species. Trichomes are presented on the adaxial and abaxial surface of four species. Tannin is found on the both of four species. The Anatomy of 15 species is recorded for the first time. Traditional uses of the Araceae family in the Northeastern Thailand are studied. Local herbalists and the elders are interviewed about local names, part of use and how to use. A total of 10 species belonging to 8 genera were recorded in this study. All species are recognized as food, medicine, ornamental, commercial propagation and rituals. The most widely part of use was young stem and young leaves for foods. Micropropagation young leaves culture of *Typhonium glaucum* culture on MS medium supplemented with NAA+BA and activated charcoal and no add activated charcoal for 8 weeks are studied. MS medium supplemented with 2 mg/l NAA combination with 2 mg/l BA and activated charcoal are the best cultured for develop plantlets or shoots media. While cultured MS medium supplemented with 2 mg/l NAA and 2 mg/l BA without activated charcoal are the best cultured for develop callus media. After that transfer the callus induction shoot and root found that the 1 mg/l NAA combined with 1 mg/l BA the best induced shoot and root. And shoot proliferation found that the cultured on MS medium supplemented with 0.1 mg/l NAA and 2 mg/l BA are the best induced shoot media and while cultured MS medium supplemented with 0.5 mg/l NAA continent with 2 mg/l BA are the best induced shoot media.



ชื่อเรื่อง	ความหลากหลายของโครโมโซมและชีววิทยาของพืชวงศ์บูกบอนในประเทศไทย		
ผู้วิจัย	นางสาวรัตนาวลี เสนาวงศ์		
ปริญญา	ปรัชญาดุษฎีบัณฑิต	สาขาวิชา	ความหลากหลายทางชีวภาพ
อาจารย์ที่ปรึกษา	ผู้ช่วยศาสตราจารย์.ดร.สุรพล แสนสุข และผู้ช่วยศาสตราจารย์.ดร.ปิยะพร แสนสุข		
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### บทคัดย่อภาษาไทย

ความหลากหลายของโครโมโซมและชีววิทยาของพืชวงศ์บูกบอนในประเทศไทยพบว่า จำนวนโครโมโซม แคริโอไทป์ และไอดีโอแกรมของพืชวงศ์บูกบอนในประเทศไทย ศึกษาจากปลายราก 20 ชนิด ด้วยเทคนิคบดขยี้เซลล์ มีจำนวนโครโมโซมดิพลอยด์เท่ากับ 18, 24, 26, 28, 40, 42 และ 58 แห่งตามลำดับ แคริโอไทป์แบบสมมาตร 10 ชนิด แบบไม่สมมาตร 10 ชนิด การศึกษานี้พบว่ามี 9 ชนิดโครโมโซมปรากฏเอ็นโออาร์ และรายงานจำนวนโครโมโซมครั้งแรก 7 ชนิด ละอองเรณู 18 ชนิด พบว่าลักษณะสัณฐานวิทยาของเรณูทั้งหมดเป็นเม็ดเดี่ยว เรณูมี 3 ขนาด ได้แก่ เรณูขนาดเล็ก ขนาดกลาง และขนาดใหญ่ สมมาตรแบบรัศมี มีช่องเปิด 2 แบบ คือ 1 ช่องเปิดรี และ 2 ช่องเปิดกลม รูปร่างแบ่งออกเป็น 5 กลุ่ม ประกอบด้วย pheroidal, prolate, oblate spheroidal, subprolate และ prolate ลวดลายบนผนังเรณู 6 แบบ และรายงานละอองเรณูครั้งแรก 16 ชนิด กายวิภาคศาสตร์ของพืช 20 ชนิด พบรูปร่างเซลล์เนื้อเยื่อชั้นผิวของใบพืชมีรูปร่างแบบจิกซอร์ว รูปสี่เหลี่ยมผืนผ้า และรูปหลายเหลี่ยมผนังเซลล์เรียบ หยักโค้ง และเป็นคลื่นทั้งเนื้อเยื่อชั้นผิวใบด้านบนและด้านล่าง เซลล์เนื้อเยื่อชั้นผิวทั้งสองด้านมีปากใบแบบ paracytic, hexacytic, anomocytic, cyclocytic, 2, 3, 4 subsidiary cells, 2, 3, 4, 6 subsidiary cells และ 2, 3, 4, 5 and 6 subsidiary ลวดลายคิวตินแบบเรียบพบในเนื้อเยื่อชั้นผิวทั้งสองด้าน ผลึกรูปดาวในเซลล์ในเนื้อเยื่อชั้นผิวทั้งสองด้าน 3 ชนิด พบขนต่อมในเซลล์ในเนื้อเยื่อชั้นผิวทั้งสองด้าน 4 ชนิด พบสารสะสมในเนื้อเยื่อชั้นผิวใบด้านบนและด้านล่าง 4 ชนิด และรายงานกายวิภาคศาสตร์ของพืชครั้งแรก 15 ชนิด การใช้ประโยชน์พืชวงศ์บูกบอนในภาคตะวันออกเฉียงเหนือของประเทศไทย โดยสอบถามผู้สูงอายุและหมอพื้นบ้านเกี่ยวกับชื่อพืชในภาษาท้องถิ่น สรรพคุณ ส่วนของพืชและวิธีใช้ พบทั้งหมด 10 ชนิด ใน 8 สกุล จำแนกการใช้ประโยชน์ออกเป็น ด้านอาหาร ด้านสมุนไพร ด้านไม้ดอกไม้ประดับ ด้านการค้า และด้านพิธีกรรมความเชื่อ ลำต้นอ่อนและใบอ่อนเป็นส่วนของพืชที่มีการนำมาใช้ประโยชน์ด้านอาหารมากที่สุด การเพิ่มจำนวน *Typhonium glaucum* ด้วยการเพาะเลี้ยงใบอ่อนบนอาหารสังเคราะห์สูตร MS ที่เติม NAA ร่วมกับ BA ร่วมกับการที่เติมและไม่เติมผงถ่าน 8 สัปดาห์ พบว่าใบอ่อนที่เพาะเลี้ยงในอาหารสูตร MS ที่เติม NAA 2 มก./ล. ร่วมกับ BA 2 มก./ล. และผงถ่าน ใบอ่อนเจริญเป็นยอดได้ดีที่สุดในขณะที่อาหารสูตร MS ที่เติม NAA 2 มก./ล. ร่วมกับ BA 2 มก./ล. ที่ไม่เติมผงถ่านเจริญเป็นแคลลัสได้ดีที่สุด ย้ายแคลลัสเพาะเลี้ยงบนอาหารสูตร MS ที่เติม NAA ร่วมกับ BA ที่พบว่าอาหารสูตร MS ที่เติม NAA 1 มก./ล. ร่วมกับ BA 1 มก./ล. แคลลัสเกิดเป็นต้นใหม่และรากได้ดีที่สุด นำต้นอ่อนไปเพาะเลี้ยงบนอาหารสูตร MS ที่เติม NAA ร่วมกับ BA พบว่าเกิดเป็นต้นจำนวนมากได้ดีที่สุดบนอาหารสูตร MS ที่เติม NAA 0.1 มก./ล. ร่วมกับ BA 2 มก./ล. ในขณะที่เพาะเลี้ยงบนอาหารสูตร MS ที่เติม NAA 0.5 มก./ล. ร่วมกับ BA 2 มก./ล. เกิดรากได้ดีที่สุด



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# CHAPTER 1

## INTRODUCTION

### 1.1 Introduction

Thailand is located in the region of tropical forest and the areas with high biodiversity areas in the world. There are a variety of plants and an animal. There is a difference to the environment and landscape. In present, plants and animals are threatened because forest encroachment to be utilized a lot. The plants are valuable to focus on the economy in many aspects, such as herbs, ornamental plants, and food crops or for conservation. It is likely to be threatened and utilization. Conservation will need to know the correct type. In this plant, Araceae is a family that has been used variously versatile (Sangnin, 2007).

Araceae is a large family in monocots plants. Family Araceae is distributed in tropical forests around the world, especially in the North and South America, Asia and throughout tropical western pacific and eastern Australia comprises 120 genera and more than 3,800 published taxa (Boyce *et al.*, 2012). The center diversity is in tropical zones. Many species are not yet to be formally described. Some species of the family have toxic sap such as *Alocasia macrorrhizos*, *Amorphophallus campanulatus*, *Colocasia esculenta*, *Colocasia fallax* and *Typhonium glaucum* etc. In Thailand, there are 30 genera (including the four genera of former Lemnaceae) with 210 species. Seventy eight (>36%) species are endemic to Thailand (Boyce *et al.*, 2012). Araceae can be used for medicines, foods, in rituals and ornamental plants. Commercial propagation plants are found in this family.

The cytological investigation of plants has proven interesting for the study of plant systematics, chromosome numbers can be used as the basis of classification of species, genera and families, and for advanced study, cytological investigation gives basic informative knowledge about the molecular genetics (Soontornchainacksaeng *et al.*, 2001).

Palynology is supporting for plant Identification. Many characteristics of pollen grains i.e. shape, size, symmetry and aperture can be used for classification and identify species of plant, (Erdtman, 1966; Nairs, 1971; Moore *et al.*, 1991 and Theilade *et al.*, 1993).

Plant anatomy, is another supporting data for plant Identification. Plant anatomical data is often extremely useful in solving problems of relationships because they can often suggest with greater confidence the homologies of morphological character states, and they can help in the interpretation of evolutionary directionality (Stuessy, 1990).

Tissue culture techniques have been used widely to increase the number of plants *in vitro* for commercial importance (Rao, 1977; Arditti and Ernst, 1993; Chung *et al.* 1985; Chen and Chang, 2000; Chen and Chang, 2004). In addition, the methods of tissue culture have been used for the conservation and rapid propagation of important plant species (Fay, 1992).

Moreover, the karyotype and idiogram of Araceae never studied before were except some species of the genus *Amorphophallus* (Kongkung, 1999). The palynology and leaf anatomy traditional uses are a very few studies in previously study. So this study is probably finding new information. Therefore, chromosome, palynology,



anatomy, traditional uses and conservation techniques are need to provide for the family Araceae in Thailand.

## **1.2 Objectives of research**

1.2.1 To study the chromosome number, karyotype and idiogram of 20 species Araceae in Thailand.

1.2.2 To study the pollen morphology of Araceae in Thailand.

1.2.3 To study the leaf surface anatomy of Araceae in Thailand.

1.2.4 To study the traditional uses of Araceae in Thailand.

1.2.5 To study the conservation an important species of Araceae in Thailand by tissue culture technique.

1.2.6 Comparison of the anatomical data, pollen morphology, chromosome numbers, idiogram and karyotype in the Araceae, this can be used for identification and classification.

## **1.3 Scope of research**

1.3.1 Twenty species of Araceae will be studied chromosome numbers, karyotype and idiogram by Feulgen Squash Technique and Computer program.

1.3.2 Pollen morphology of the Araceae family will be examined under light and scanning electron microscopies.

1.3.3 Study of the leaf surface anatomy of the Araceae in Thailand will be by investigation of the leaf epidermal peeling and transverse sections from blades method.

1.3.4 Plants traditional uses study by explore consumer inquiries and food processing.

1.3.5 Conservation one an important species of Araceae by tissue culture technique.

## **1.4 Anticipated outcomes**

This study will provide fundamental information on the Araceae family for Thailand. Information on chromosome number, some karyotypes, palynology anatomy, traditional uses and conservation rare species of this family will be useful for other researchers and important for plant genetic resource planning and management in Thailand.

## **1.5 Places of study**

This project will be conducted at:

1.5.1 Walai Rukhavej Botanical Research Institute, Mahasarakham University.

1.5.2 Department of Biology, Faculty of Science, Mahasarakham University.

1.5.3 Field trips in Thailand.

1.5.4 Herbaria: Bangkok Herbarium (BK), Forest Herbarium, Bangkok (BKF), Queen Sirikit Botanic Garden Herbarium (QSBG), Department of Biology Herbarium Chiang Mai University.



## 1.6 Long-term study

Work	2015			2016			2017			2018
	1-4	5-8	9-12	1-4	5-8	9-12	1-4	5-8	9-12	1-4
1. Literature Reviews	←————→									
2. Field trips	←————→									
3. Chromosome study	←————→									
4. Pollen study					←————→					
5. Leaf surface anatomy study					←————→					
6. Traditional use study	←————→									
7. Tissue culture study					←————→					
8. Writing the conclusion					←————→					
9. Preparing manuscript for publication					←————→					
10. Writing the research			←————→							
11. Thesis defense and complete thesis									←————→	

## 1.7 Araceae

### 1.7.1 Classification and general characteristics of Araceae (Daniel, 2009)

Kingdom: Plantae

Subkingdom: Viridaeplantae (Green Plants)

Phylum: Tracheophyta (Vascular Plants)

Subphylum: *Euphyllophytina*

Intraphylum: Radiatopses

Class: Liliopsida

Subclass: Aridae

Superorder: Aranae

Order: Arales

Family: Araceae



### 1.7.2 The morphological characteristics (Boyce *et al.*, 2012)

Araceae is a perennial herbs of diverse habit including epiphytes, climbers, floating, aquatics, helophytes, mesophytes, pachycaul shrubs and geophytes.

### 1.7.3 Habit (Boyce *et al.*, 2012)

Underground stems absent, or present and then rhizomatous or tuberous; aerial stems variously produced or not, often evergreen; bulbils for vegetative reproduction sometimes produced, e.g., on leaf or on special shoots.

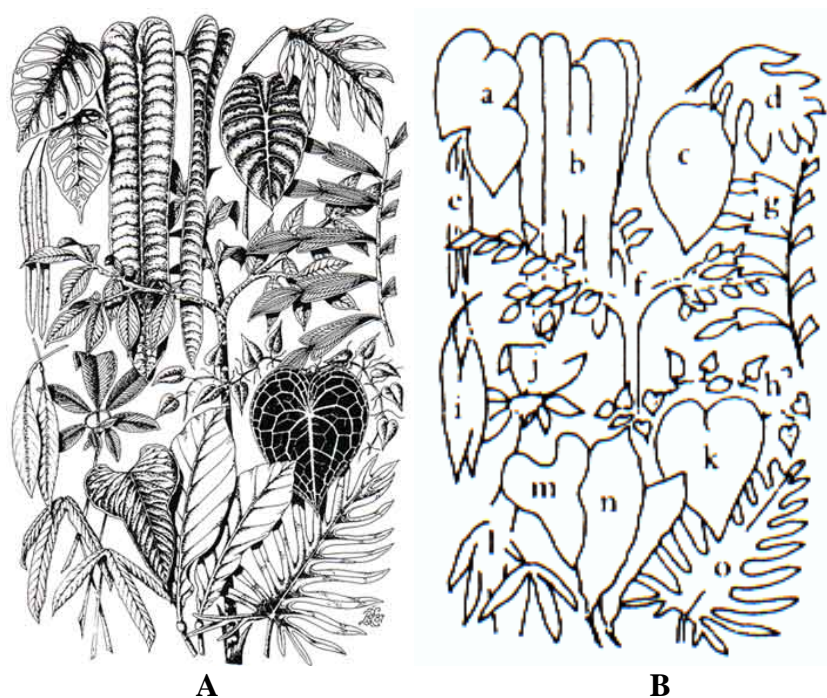
**Leaves** alternate or apparently basal, usually petiolate with sheathing bases, often subtended by prophylls and/or cataphylls; leaf blade various, e.g., linear, simple (base often cordate to sigillate), sometimes peltate or variously compound (e.g., pinnate, radiate, pedate, or decompound), or still more complex and “dracontoid” (elaborate forms of sagittate, hastate, or trisect leaves in which anterior and posterior divisions are highly dissected and subdivided) (Figuer 1.1 and Figuer 1.2).

**Inflorescences** (sometime preconscious) subtended by membranous prophylls and/or cataphylls, consisting of a spadix subtended by a spathe; **spathe** commonly with tube-like base (margins fused or not) fully persistent or with limb deciduous, or spathe entirely deciduous; **spadix** bearing bisexual or unisexual flowers, in latter case plants parodioecious or monoecious (spadix pistillate proximally and staminate distally), very rarely with morphologically bisexual but functionally unisexual flowers; **bisexual flowers**: **tepals** 0,4 or 6; **stamens** 4-6(-22); **filaments** free, anthers with 2 thecae; **gynoecium** usually 3-loculed or more loculed or 1-loculed (pseudomonomerous); unisexual flowers almost always naked [rare exceptions (only 3 genera, these all from Africa, including (Thailand cultivated) *Zamioculcas* with perigyniate flowers)]: **staminate flowers** represented by 1-6 (usually 2-4) free stamens or 2-12 (rarely up to 32) stamens connate into a synandrium overtopped by a common synconnective, anthers often subsessile, usually dehiscing apically by separate or confluent pores, or straight or horseshoe-shaped slits; **pistillate flowers** consisting of a monad gynoecium (sometimes associated with one or more sterile staminodes), commonly 1-loculed by abortion (pseudomonomerous), sometimes with 3 or 4 locules; **ovules** 1 to many per locule, placentation parietal, axile, basal, or apical, often intermediate between two types. Most unisexual-flowered genera with variously elaborate sterile floral structures (staminodes, pistillodes, synadroides, etc.) separating the pistillate and staminate floral zones, and/or forming a terminal appendix (then sometimes naked and comprising the largest and most conspicuous portion of the spadix, e.g., *Amorphophallus*) (Figuer 1.3 and Figuer 1.4).

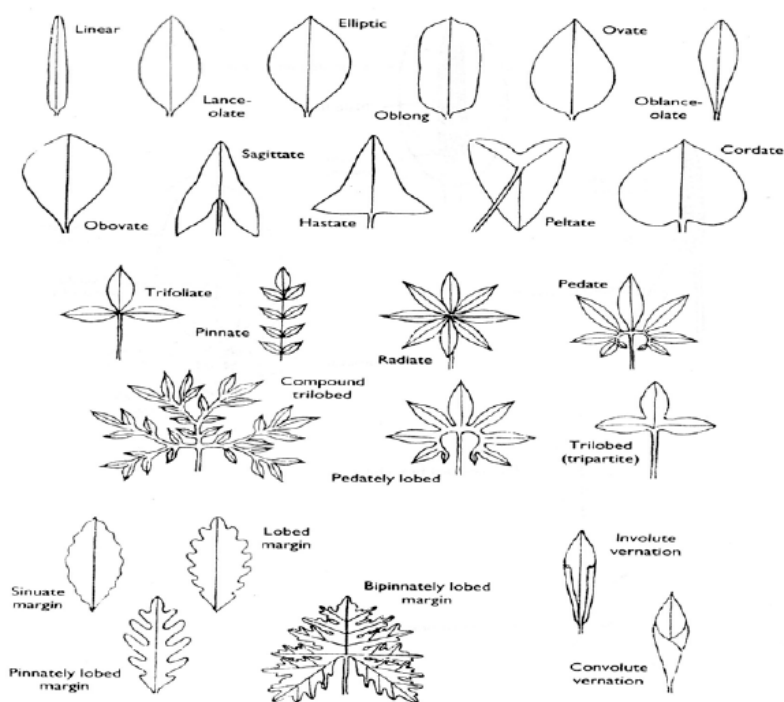
**Inflorescences** usually a head of 1- to many-seeded indehiscent separate berries, or fruits dehiscent via shedding stylar plate (‘monstercarp’- Monstereae excluding *Amydrium*), or syncarpous and apically dehiscent (*Cryptocoryne*), or syncarpous and indehiscent (*Syngonium*, cultivated in Thailand), commonly red, green, white, or yellow, rarely blue. (Boyce *et al.*, 2012)







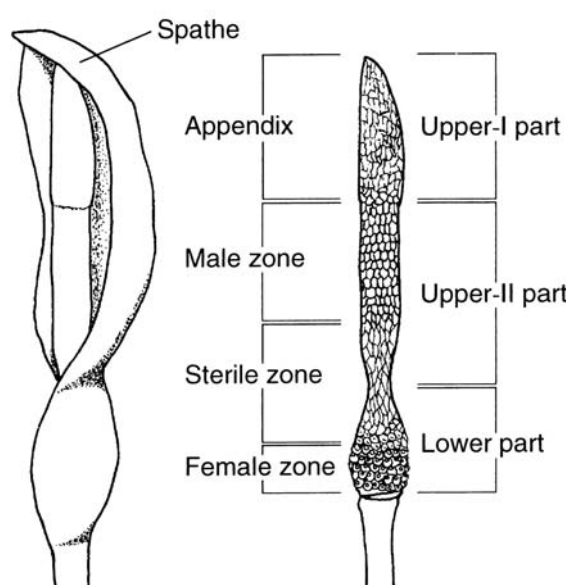
**Figure 1.1** A. The characteristic shape of Araceae. B. Drawings show the leaves of plants in the genus. A) *Monstera* B) E) K) L) M) and N) *Anthurium* C) *Alocasia* D) *Alloschemone* F) *Amorphophallus* G) *Pothos* H) J) and O) *Philodendron* I) *Holochlamys* (Bown, 2000; Napiroon, 2013)



**Figure 1.2** Drawings the leaf shapes of family Araceae (Bown, 2000; Napiroon, 2013)



**Figure 1.3** A. The inflorescence flora family Araceae. B. Drawings the inflorescence of plants in the genus. A) *Colocasia* B) *Spathiphyllum* C) and H) *Anthurium* D) G) *Dracontium* E) *Biarum* F) *Zantedeschia* I) M) and P) *Amorphophallus* J) K) *Arisaema* L) *Arum* N) *Pothos* O) *Spathicarpa* Q) *Arisarum* and R) *Cryptocoryne* (Bown, 2000; Napiroon, 2013)



**Figure 1.4** Drawings show the characteristics of the inflorescence flora family Araceae (Miyake and Yafuso, 2003; Napiroon, 2013)



The families Araceae in Thailand are 30 genera (including the four genera of former Lemnaceae (Table 1.1). Currently ca 210 species are recorded in Thailand. Seventy eight (>36%) species are endemic to Thailand (Boyce *et al.*, 2012).

**Table 1.1** Distribution of diversity Araceae in Thailand (Boyce *et al.*, 2012)

No.	Genera	No. species
1	<i>Aglaodorum</i>	1 species
2	<i>Aglaonema</i> (Hora or Khiao-Muen-Pi)	9 species
3	<i>Alocasia</i> (Bon)	12 species
4	<i>Amorphophallus</i> (Bok)	56 species
5	<i>Amydrium</i> (Khod-Nok-Kut)	1 species
6	<i>Anadendrum</i>	4 species
7	<i>Apoballis</i>	2 species
8	<i>Ariopsis</i> (Bon-Hin)	1 species
9	<i>Arisaema</i> (Wan or Buk)	17 species
10	<i>Colocasia</i> (Phueak)	4 species
11	<i>Cryptocoryne</i>	8 species
12	<i>Epipremnum</i> (Phlu-Dang)	3 species
13	<i>Hapaline</i> (Bon)	4 species
14	<i>Homalomena</i> (Tao-Kiat)	7 species
15	<i>Lasia</i> (Phuk-Nam)	1 species
16	<i>Pipthospatha</i>	1 species
17	<i>Pistia</i> (Chok)	1 species
18	<i>Pothos</i> (Thao-Phan-Dong)	8 species
19	<i>Pycnospatha</i> (Uttaphit or Hora)	2 species
20	<i>Remusatia</i> (Bon-Pha)	2 species
21	<i>Rhaphidophora</i> (Ta-Khap)	17 species
22	<i>Sauromatum</i> (Taphit)	3 species
23	<i>Schismatoglottis</i> (Bon-Khiao)	3 species
24	<i>Scindapsus</i> (Phlu-Chang)	4 species
25	<i>Steudnera</i>	3 species
26	<i>Typhonium</i> (Uttapid or Ta-Phit)	32 species
27	<i>Landoltia</i> *	1 species
28	<i>Lemna</i> *	1 species
29	<i>Spirodela</i> *	1 species
30	<i>Wolffia</i> *	1 species
<b>Total species</b>		210 species

\* Remarks are genera formerly in family Lemnaceae

## CHAPTER 2

### CHROMOSOME STUDIES

#### 2.1 Introduction

Scientific method of plant taxonomy, genetic diversity, plant breeding, phylogeny and evolution can identify chromosome number (Somatic chromosome number =  $2n$ ) and a type of cell. Plants in the same species or which are related morphologically usually have the same chromosome number and karyotype (Stebbins, 1971; De Robertis and De Robertis, 1980).

Karyotype study is the basic information of the chromosomes of an organism species by studying the number, size and type of chromosomes, which are specific in many organisms (Appels *et al.*, 1998). The karyotype of plants the seeds of each species within the same genus may be similar or different. The normal karyotype, of each species is constant. But may be during evolutionary changed such as the found B chromosome or sex chromosome etc. In addition, the variation of the karyotype caused by abnormalities of chromosomes structural (Chaiyasut, 1989). So, Araceae family found the karyotype and idiogram studied is not much.

A study on karyotypes of plants can help identify the cause of the morphological changes that occurred from the environment or from a change involving chromosomes. Either the structural changes to chromosomes or chromosome number change. It is also used as evidence in the study of the relationships of organisms in evolutionary (Chaiyasut, 1989).

The chromosome number, karyotype and idiogram of plant Araceae family in Thailand have not been studies yet. The aim of this study is to examine somatic chromosome number, karyotype and idiogram from the root tip of plant Araceae family in Thailand.

#### 2.2 Chromosome

##### 2.2.1 Definition and scope of Chromosome

The fact that chromosomes are the carriers of genetic information provided an impetus for their studies since the establishment of the chromosome theory of inheritance in the second decade of the century. The steady increased amount of information on chromosomes and the development of cytogenetic concepts and methods demonstrated the importance of chromosome in several fields of plant biology. In the field of plant systematics and flora, the spectacular rise in the publication of reliable chromosome data in the past 25 years began to make clear the cytogenetic mechanisms involved in the evolution and delimitation of plant taxa and the variety of way these mechanisms are expressed in the great diversity of plant comprising the World flora. Chromosomal features are being regarded as decision making characters in the study of phylogenetic affinities and evolutionary development, and as indicators of appropriate classifications of several plant groups (Jones, 1978).



Chromosomal information has also been used in the study of floras particularly those regarding chromosome number because it is the easiest to assess. Chromosome numbers indicate the occurrence of polyploidy and reflect difference in the basic chromosome numbers among plants which may be reflected in their treatments in floras. It is estimated that over 3,000 chromosome counts are annually recorded every year. However, chromosome numbers are only known for 15-20% of angiosperm species (Moore, 1981). Other karyotype features, such as chromosome size, position of centromere and the presence of satellites in the chromosomes of the karyotype are recorded in less than 1% of angiosperm. More detailed karyotype characters, such as the disposition of heterochromatic segments, revealed by banding techniques, are very poorly known (Badr and Gasim, 1992).

**Chromosomes** (King and Stansfield, 1990);

1. In prokaryotes, the circular DNA molecule containing the entire set of genetic instructions essential for life of the cell.

2. In the eukaryotic nucleus, one of the threadlike structures consisting of chromatin and carrying genetic information arranged in a linear sequence.

**Karyotype** is the number, size, and morphology of the chromosome set of a cell, individual, or species. The term is often used for photomicrographs of chromosome preparations; e.g., the metaphase chromosome arranged in a standard sequence; an ideogram (King and Stansfield, 1990).

**Idiogram** is a diagrammatic representation of the karyotype of an organism (King and Stansfield, 1990).

The chromosome number is one of the primary basic of taxonomy study. It is the general rule that successful crosses are easier to achieve between individual plants under it the same species rather than between different species under the same genus. However, it is not the diploid number of chromosomes alone that determines. If to be more precise, it is the karyotype study, referring to the number, as well as the size and shape of the chromosomes of an individual and other consideration are the types and shape of chromosomes.

### 2.2.2 Literature reviews of chromosome

The documents are studied found that a chromosome of family Araceae in Thailand and overseas countries are studied the follow in Table 2.1, which many species in Thailand has not yet been studied in chromosome, karyotype and idiogram.

**Table 2.1** The literature reviews of chromosome family Araceae.

No.	Species	Chromosome number (2n)	Reference
1	<i>Aglaodorum griffithii</i> (Schott) Schott	40	Petersen (1989)
2	<i>Aglaonema commutatum</i> Schott	14	Subramanian and Munian (1988)
3	<i>A. modestum</i> Schott ex Engl.	60	Chen <i>et al.</i> (2003)
4	<i>A. pictum</i> (Roxb.) Kunth	40	Okada (1982)



**Table 2.1** The literature reviews of chromosome family Araceae (Continued).

No.	Species	Chromosome number (2n)	Reference
5	<i>Alocasia acuminata</i> Schott	28	Mehra and Sachdeva (1979); Bhattacharya (1974); Sachdeva (1977); Sharma (1970)
6	<i>Alocasia argyrea</i> Sander	56	Sharma (1970)
7	<i>Alocasia crassifolia</i> Engl.	28	Okada (1982)
8	<i>Alocasia cucullata</i> (Lour.) G.Don	28	Ankei (1987); Ishida (2001)
9	<i>Alocasia cucullata</i> (Lour.) Schott	28	Bhattacharya (1974)
10	<i>Alocasia cuprea</i> K.Koch	28	Ishida (2001)
11	<i>Alocasia decipiens</i> Schott	24 (32)	Bhattacharya (1974)
12	<i>Alocasia fornicata</i> (Roxb.) Schott	42	Ramachandran (1978)
13	<i>Alocasia gageana</i> Engl. & K.Krause	28	Ishida (2001)
14	<i>Alocasia indica</i> (Lour.) Schott	28, 42	Chaudhuri & Sharma (1979); Ramachandran (1978); Bhattacharya (1974)
15	<i>Alocasia indica</i> var. <i>variegata</i> (K.Koch & Bouché) Engl.	28	Bhattacharya (1974)
16	<i>Alocasia lindenii</i> Rodigas	40, 56	Bhattacharya (1974); Sharma (1970)
17	<i>Alocasia longiloba</i> Miq.	56	Bhattacharya (1974); Sharma (1970)
18	<i>Alocasia lowii</i> Hook.f.	28, 70, 40	Ishida (2001); Bhattacharya (1974)
19	<i>Alocasia macrorrhiza</i> (L.) Schott	28 (18)	Bhattacharya (1974); Sharma (1970)
20	<i>Alocasia macrorrhizos</i> (L.) G.Don	28	Ishida (2001)
21	<i>Alocasia micholitziana</i> Sander	28	Ishida (2001)
22	<i>Alocasia montana</i> (Roxb.) Schott	28	Ramachandran (1978)
23	<i>Alocasia navicularis</i> (K.Koch & C.D.Bouché) K.Koch & C.D.Bouché	68	Sharma (1970)



**Table 2.1** The literature reviews of chromosome family Araceae (Continued).

No.	Species	Chromosome number (2n)	Reference
24	<i>Alocasia odora</i> (Roxb.) K.Koch	28 (29), 56	Ankei (1987); Nguyen <i>et al.</i> (1998); Bhattacharya (1974)
25	<i>Alocasia odora</i> (Lodd.) Spach	28	Ishida (2001)
26	<i>Alocasia plumbea</i> Van Houtte	28	Ishida (2001)
27	<i>Alocasia porphyroneura</i> Hallier f.	28	Ishida (2001)
28	<i>Alocasia portei</i> Schott	28	Ishida (2001)
29	<i>Alocasia putii</i> Gagnep.	28	Sharma (1970)
30	<i>Alocasia regina</i> N.E.Br.	28	Bhattacharya (1974); Sharma (1970)
31	<i>Alocasia sanderiana</i> W.Bull	28	Ishida (2001)
32	<i>Alocasia thibautiana</i> Mast.	28	Bhattacharya (1974); Sharma (1970)
33	<i>Alocasia wentii</i> Engl. & K.Krause	42	Ishida (2001)
34	<i>Alocasia zebrina</i> K.Koch & Veitch	28, 42	Ishida, (2001); Sharma, (1970); Bhattacharya (1974)
35	<i>Amorphophallus abyssinicus</i> (A.Rich.) N.E.Br.	26	Chauhan and Brandham (1985)
36	<i>Amorphophallus albus</i> P.Y.Liu & J.F.Chen	26	Liu <i>et al.</i> (1985); Zheng and Liu (1989)
37	<i>Amorphophallus ankarana</i> Hett., Ittenb. & Bogner	26	Hetterscheid <i>et al.</i> (1999)
38	<i>Amorphophallus asterostigmatus</i> Bogner & Hett.	26	Petersen (1992)
39	<i>Amorphophallus bannanensis</i> H.Li	26	Gu <i>et al.</i> (1992)
40	<i>Amorphophallus bulbifer</i> (Roxb.) Blume	26, 39	Kuruvilla <i>et al.</i> (1989); Chauhan and Brandham (1985); Ishida (2004)



**Table 2.1** The literature reviews of chromosome family Araceae (Continued).

No.	Species	Chromosome number (2n)	Reference
41	<i>Amorphophallus campanulatus</i> Blume ex Decne.	26, 28	Chaudhuri and Sharma (1979); Subramanian and Munian (1988); Sharma (1970)
42	<i>Amorphophallus commutatus</i> (Schott) Engl.	26	Chauhan and Brandham (1985)
43	<i>Amorphophallus dixenii</i> K.Larsen & S.S.Larsen	28	Larsen and Larsen (1974)
44	<i>Amorphophallus dracontoides</i> (Engl.) N.E.Br.	26	Chauhan and Brandham (1985)
45	<i>Amorphophallus dubius</i> Blume	28	Chauhan and Brandham (1985)
46	<i>Amorphophallus dunnii</i> Tutchener	26	Zheng and Liu (1989)
47	<i>Amorphophallus goetzii</i> (Engl.) N.E.Br.	26	Chauhan and Brandham (1985)
48	<i>Amorphophallus henryi</i> N.E.Br.	26	Ishida (2004)
49	<i>Amorphophallus hildebrandtii</i> (Engl.) Engl. & Gehrm.	26	Chauhan and Brandham (1985)
50	<i>Amorphophallus hirtus</i> N.E.Br.	26	Ishida (2004)
51	<i>Amorphophallus johnsonii</i> N.E.Br.	26	Chauhan and Brandham (1985)
52	<i>Amorphophallus kerrii</i> N.E.Br.	26	Chauhan and Brandham (1985)
53	<i>Amorphophallus kiusiana</i> (Makino) Makino	26	Ishida (2004)
54	<i>Amorphophallus konjac</i> K.Koch	26	Chauhan and Brandham (1985); Cheng <i>et al.</i> (1991)
55	<i>Amorphophallus konjac</i> K.Koch ex Matsum. & Hayata	26	Ishida (2004)



**Table 2.1** The literature reviews of chromosome family Araceae (Continued).

No.	Species	Chromosome number (2n)	Reference
56	<i>Amorphophallus konjac</i> K.Koch ex Matsum. & Hayata misc. cultivars	26	Ishida and Akagi (2000)
57	<i>Amorphophallus konkanensis</i> Hett., S.R.Yadav & Patil	26	Hetterscheid <i>et al.</i> (1994); Patil and Dixit (1995)
58	<i>Amorphophallus krausei</i> Engl.	26	Ishida (2004)
59	<i>Amorphophallus lambii</i> Mayo & Widjaja	26	Chauhan and Brandham (1985)
60	<i>Amorphophallus laxiflorus</i> N.E.Br.	26	Chauhan and Brandham (1985)
61	<i>Amorphophallus mairei</i> H.Lév.	26	Zheng and Liu (1989)
62	<i>Amorphophallus margaritifer</i> (Roxb.) Kunth	26	De Sarker and Hetterscheid (1997)
63	<i>Amorphophallus muelleri</i> Blume	39	Ishida (2004)
64	<i>Amorphophallus oncophyllus</i> Prain ex Hook.f.	39	Chauhan and Brandham (1985)
65	<i>Amorphophallus paeoniifolius</i> (Dennst.) Nicolson	28	Chauhan and Brandham (1985); Ishida (2004)
66	<i>Amorphophallus palawanensis</i> Bogner & Hett.	26	Petersen (1992)
67	<i>Amorphophallus pingbianensis</i> H.Li & C.L.Long	26	Ishida (2004)
68	<i>Amorphophallus prainii</i> Hook.f.	28	Chauhan and Brandham (1985)
69	<i>Amorphophallus rivieri</i> Durieu ex Carrière	26	Liu <i>et al.</i> (1985); Zheng and Liu (1989)
70	<i>Amorphophallus sinensis</i> Belval	26	Sun (1999)



**Table 2.1** The literature reviews of chromosome family Araceae (Continued).

No.	Species	Chromosome number (2n)	Reference
71	<i>Amorphophallus sutepensis</i> Gagnep.	26	Chauhan and Brandham (1985)
72	<i>Amorphophallus taurostigma</i> Ittenb., Hett. & Bogner	26	Hetterscheid <i>et al.</i> (1999)
73	<i>Amorphophallus titanum</i> (Becc.) Becc.	26	Giordano (1999); Ishida (2004)
74	<i>Amorphophallus variabilis</i> Blume	26	Chauhan and Brandham (1985)
75	<i>Amorphophallus yunnanensis</i> Engl.	26	Ishida (2004)
76	<i>Amydrium humile</i> Schott	60	Petersen (1989)
77	<i>Amydrium medium</i> (Zoll. & Moritzi) Nicolson	60	Petersen (1989)
78	<i>Anadendrum microstachyum</i> (de Vries & Miq.) Backer & Alderw.	60	Petersen (1989)
79	<i>Arisaema consanguineum</i> Schott	28, 48, 56	Kuruvilla <i>et al.</i> (1989); Sarkar <i>et al.</i> (1978); Sarkar and Chatterjee, (1978); Sarkar and Datta (1978); Wang (1996); Murata and Iijima (1983)
80	<i>Arisaema prazeri</i> Hook.f.	26	Murata (2006)
81	<i>Arisaema yunnanense</i> Buchet	24, 48	Murata and Iijima (1983) ; Murata (2006)
82	<i>Colocasia antiquorum</i> Schott	26, 28, 30, 36, 38, 42, 44, 48, 52, 58, 116	Chaudhuri and Sharma (1979); Subramanian and Munian (1988); Subramanian (1979); Chakraborty and Bhattacharya (1984); Sarkar (1991); Ramachandran (1978); Sharma (1970); Kawahara (1978)
83	<i>Colocasia bicolor</i> C.L.Long & L.M.Cao	28	Cao (2003); Cao and Long (2004); Cao and Long (2006 )





**Table 2.1** The literature reviews of chromosome family Araceae (Continued).

No.	Species	Chromosome number (2n)	Reference
84	<i>Colocasia esculenta</i> (L.) Schott	28, 36, 38, 42, 84	Coates <i>et al.</i> (1988); Subramanian and Munian (1988); Li (1989); Zhang and Yang (1984); Tanimoto and Matsumoto (1986); Huang <i>et al.</i> (1989); Sreekumari and Mathew (1989); Okada and Hambali (1989); Kuruvilla <i>et al.</i> (1989); Sreekumari and Mathew (1991a); Sreekumari and Mathew (1991b); Sreekumari and Mathew (1991c); Sreekumari and Mathew (1995d); Ivancic and Lebot (1999); Zhang (1998); Kuruvilla and Singh (1981); Ramachandran (1978)
85	<i>Colocasia fallax</i> Schott.	28	Begum and Alam (2009)
86	<i>Colocasia gaoligongensis</i> H.Li & C.L.Long	28	Yang <i>et al.</i> (2003); Cao and Long, (2004)
87	<i>Colocasia gigantea</i> (Blume) Hook.f.	2n=28	Tanimoto and Matsumoto (1986); Okada and Hambali (1989); Yang <i>et al.</i> (2003); Cao and Long (2004)
88	<i>Colocasia gongii</i> C.L.Long & H.Li	2n=28	Yang <i>et al.</i> (2003); Cao and Long (2004)
89	<i>Colocasia heterochroma</i> H.Li & Z.X.Wei	2n=28	Cao and Long (2004); Cao and Long, (2006)
90	<i>Colocasia indica</i> (Lour.) Hassk.	2n=28	Ankei (1987)
91	<i>Colocasia konishii</i> Hayata	2n=28	Cao and Long (2004)
92	<i>Colocasia lihengiae</i> C.L.Long & K.M.Liu	2n=28	Long & Liu (2001); Cao and Long (2004)
93	<i>Cryptocoryne affinis</i> N.E.Br. ex Hook.f.	2n=34	Arends <i>et al.</i> (1982); Jacobsen (1977)
94	<i>Cryptocoryne albida</i> R.Parker	2n=36	Arends <i>et al.</i> (1982); Jacobsen (1977)



**Table 2.1** The literature reviews of chromosome family Araceae (Continued).

No.	Species	Chromosome number (2n)	Reference
95	<i>Cryptocoryne ciliata</i> (Roxb.) Fisch. ex Wydl.	2n=22, 33	Arends <i>et al.</i> (1982); Sarkar <i>et al.</i> (1978); Sarkar (1991); Sarkar and Chatterjee (1978); Sarkar <i>et al.</i> (1979); Sarkar <i>et al.</i> (1976)
96	<i>Cryptocoryne cordata</i> Griff.	2n= 34, 68, 85, 102	Arends <i>et al.</i> (1982)
97	<i>Cryptocoryne crispatula</i> Engl.	2n=36, 54	Arends <i>et al.</i> (1982); Jacobsen (1977)
98	<i>Epipremnum mirabile</i> Schott.	2n=70	Sharma (1970)
99	<i>Hapaline brownii</i> Hook.f.	2n=28	Petersen (1989)
100	<i>Homalomena consobrina</i> Engl.	2n=40	Okada (2000)
101	<i>Homalomena gadutensis</i> M.Hotta	38	Okada (1985); Hotta (1985)
102	<i>Homalomena griffithii</i> (Schott) Hook.f.	40	Okada (1982); Okada (2000)
103	<i>Homalomena hastata</i> M.Hotta	40	Okada (1985); Hotta (1985)
104	<i>Homalomena lancifolia</i> Hook.f.	40	Okada (2000)
105	<i>Homalomena megalophylla</i> M.Hotta	40	Okada (1985); Hotta (1985)
106	<i>Homalomena rubescens</i> var. <i>latifolia</i> Engl.	40	Okada (1982)
107	<i>Homalomena pygmaea</i> (Hassk.) Engl.	40	Okada (1982)
108	<i>Homalomena rusdii</i> M.Hotta	40	Okada (2000)
109	<i>Homalomena sagittifolia</i> Jungh. ex Schott.	40	Okada (1982)
110	<i>Homalomena sagittifolia</i> var. <i>sumatrana</i> Alderw.	80	Okada (1985); Hotta (1985)
111	<i>Homalomena speariae</i> Bogner & Moffler	42	Petersen (1989); Moffler and Bogner (1984)



**Table 2.1** The literature reviews of chromosome family Araceae (Continued).

No.	Species	Chromosome number (2n)	Reference
112	<i>Homalomena</i> cf. <i>sulcata</i> Engl.	40	Okada (2000)
113	<i>Lasia heterophylla</i> (Roxb.) Schott.	26	Sharma (1970)
114	<i>Lasia spinosa</i> (L.) Thwaites	26, 27	Ramachandran (1978); Sultana <i>et al.</i> (2006)
115	<i>Pistia stratiotes</i> L.	28	Ramachandran (1978); Sarkar and Chatterjee (1978); Sarkar <i>et al.</i> (1978); Subramanian and Munian (1988); Sarkar (1991), Selvaraj (1993)
116	<i>Pothos longipes</i> Schott	26	Briggs <i>et al.</i> (2002)
117	<i>Pothos scandens</i> D.Don	24	Sarkar and Chatterjee (1978)
118	<i>Pothos scandens</i> L.	24	Sarkar <i>et al.</i> (1978); Sarkar (1991)
119	<i>Pothos viridis</i> Parment. ex Steudel	60	Sharma (1970)
120	<i>Remusatia hookeriana</i> Schott	28, 42	Long <i>et al.</i> , (1989); Sarkar <i>et al.</i> (1978); Li and Hay (1992); Gu <i>et al.</i> (1992); Sarkar (1991); Sarkar <i>et al.</i> (1976)
121	<i>Ramasetia hookeriana</i> Schott	28	Sarkar <i>et al.</i> (1978)
122	<i>Remusatia ornata</i> (Schott) Heng Li & Q.F.Guo	42	Long <i>et al.</i> (1989)
123	<i>Remusatia pumila</i> (D.Don)	20	Li & Hay (1992)
124	<i>Remusatia vivipara</i> (Roxb.) Schott	42, 28 (42), 28	Long <i>et al.</i> , (1989); Li & Hay (1992); Ramachandran (1978)
125	<i>Rhaphidophora beccarii</i> (Engl.) Engl.	2n=60	Okada (2000)
126	<i>Rhaphidophora decursiva</i> (Roxb.) Schott	26,56	Sarkar <i>et al.</i> (1978); Chaudhuri and Sharma (1979)
127	<i>Rhaphidophora glauca</i> (Wall.) Schott	56	Chaudhuri and Sharma (1979)
128	<i>Rhaphidophora lancifolia</i> Schott	56	Sharma (1970)



**Table 2.1** The literature reviews of chromosome family Araceae (Continued).

No.	Species	Chromosome number (2n)	Reference
129	<i>Rhaphidophora peepla</i> (Roxb.) Schott	42	Sharma (1970)
130	<i>Sauromatum gaoligongense</i> Z.L.Wang & H.Li	26	Bian <i>et al.</i> (2001)
131	<i>Sauromatum guttatum</i> (Wall.) Schott	26	Chaudhuri and Sharma (1979)
132	<i>Sauromatum venosum</i> (Dryand. ex Aiton) Kunth	26	Sarkar <i>et al.</i> (1978); Sarkar (1991) Sarkar and Chatterjee (1978); Bian <i>et al.</i> (2001); Mehra and Sachdeva (1976); Sarkar <i>et al.</i> (1976)
133	<i>Schismatoglottis batoeensis</i> Engl.	26	Okada (1982)
134	<i>Schismatoglottis brevipes</i> Hook.f.	26	Okada (2000)
135	<i>Schismatoglottis bulbifera</i> H.Okada, Tsukaya & Y.Mori	26	Okada (2000)
136	<i>Schismatoglottis calyptrata</i> (Roxb.) Zoll. & Moritzi	26	Okada (1982)
137	<i>Schismatoglottis celebica</i> Engl.	26	Okada (2000)
138	<i>Schismatoglottis erecta</i> M.Hotta	26	Okada (2000)
139	<i>Schismatoglottis homalomenoidea</i> M.Hotta	26	Okada (2000)
140	<i>Schismatoglottis irrorata</i> Engl.	52	Okada (1992); Okada (2000)
141	<i>Schismatoglottis lancifolia</i> Hallier f. & Engl.	26	Okada (1982); Hotta (1982); Hotta <i>et al.</i> (1984); Hotta <i>et al.</i> (1985); Okada (2000)
142	<i>Schismatoglottis multiflora</i> Ridl.	26	Okada (2000)



**Table 2.1** The literature reviews of chromosome family Araceae (Continued).

No.	Species	Chromosome number (2n)	Reference
143	<i>Schismatoglottis okadae</i> M.Hotta	26	Okada (2000)
144	<i>Schismatoglottis parvifolia</i> Alderw.	26	Okada (2000)
145	<i>Schismatoglottis picta</i> Schott	30	Sharma (1970)
146	<i>Schismatoglottis roseospatha</i> Bogner	26	Petersen (1989)
147	<i>Schismatoglottis rupestris</i> Zoll. & Moritzi ex Zoll.	26	Okada (1982); Okada (2000)
148	<i>Scindapsus aureus</i> (Linden & André) Engl. & K.Krause	48, 56	Subramanian and Munian (1988); Chen <i>et al.</i> (2003)
149	<i>Scindapsus hederaceus</i> Schott	64	Okad (1982)
150	<i>Scindapsus lucens</i> Bogner & P.C.Boyce	60	Petersen (1993)
151	<i>Scindapsus megaphyllus</i> Merr.	56	Huang <i>et al.</i> (1989)
152	<i>Scindapsus officinalis</i> (Roxb.) Schott	56	Chaudhuri and Sharma (1979)
153	<i>Scindapsus pictus</i> Hassk.	70	Sharma (1970)
154	<i>Steudnera colocasiifolia</i> K.Koch	36	Sharma (1970)
155	<i>Steudnera colocasioides</i> Hook.f.	28	Kuruvilla <i>et al.</i> (1989)
156	<i>Steudnera discolor</i> W.Bull	16	Jos <i>et al.</i> (1971)
157	<i>Steudnera henryana</i> Engl.	28	Petersen (1993)
158	<i>Typhonium blumei</i> Nicolson & Sivad.	52	Wang and Yang (1996); Bian <i>et al.</i> (2002)
159	<i>Typhonium bulbiferum</i> Dalzell	20	Ramachandran (1978)
160	<i>Typhonium calcicola</i> C.Y.Wu	52	Bian <i>et al.</i> (2002)



**Table 2.1** The literature reviews of chromosome family Araceae (Continued).

No.	Species	Chromosome number (2n)	Reference
161	<i>Typhonium cuspidatum</i> (Blume) Decne.	16	Subramanian and Munian (1988); Jos and Magoon (1971); Sarkar, (1991); Ramachandran (1978); Sharma (1970)
162	<i>Typhonium divaricatum</i> (L.) Decne.	16, 52, 65	Jos and Magoon (1971); Ramachandran (1978)
163	<i>Typhonium diversifolium</i> Wall.	52	Mehra and Sachdeva (1976)
164	<i>Typhonium diversifolium</i> Wall. ex Schott	52	Das <i>et al.</i> (2006)
165	<i>Typhonium flagelliforme</i> (Lodd.) Blume	24 (32), 16	Bian <i>et al.</i> (2002); Das <i>et al.</i> (2006)
166	<i>Typhonium giganteum</i> Engl.	56	Bian <i>et al.</i> (2002)
167	<i>Typhonium jinpingense</i> Z.T.Wang, H.Li & F.H.Bian	10	Wang <i>et al.</i> (2002)
168	<i>Typhonium roxburghii</i> Schott	18, 26	Bian <i>et al.</i> (2002); Das <i>et al.</i> (2006)
169	<i>T. trilobatum</i> (L.) Schott	18, 18 (26), 36	Subramanian and Munian (1988); Zaman, and Podder, (1981); Bian <i>et al.</i> (2002); Das <i>et al.</i> (2006); Chaudhuri and Sharma (1979); Ramachandran, (1978)
170	<i>T. venosum</i> (Dryand. ex Aiton) Hett. & P.C.Boyce	18	Das <i>et al.</i> (2006)

A study chromosome number of family Araceae from table 2.1 can be summarized as follows; the chromosome number of genus *Aglaodorum*  $2n=2x=40$ , *Aglaonema*  $2n=2x=28$  and  $3x=60$ , *Alocasia*  $2n=4x=68$  and  $5x=70$ , *Amorphophallus*  $2n=2x=26$  and  $28$ , *Amydrium*  $2n=2x=60$ , *Anadendrum*  $2n=2x=60$ , *Arisaema*  $2n=2x=26$ ,  $4x=48$  and  $56$ , *Colocasia*  $2n=6x=84$ , *Cryptocoryne*  $2n=6x=102$ , *Epipremnum*  $2n=2x=70$ , *Hapaline*  $2n=2x=28$ , *Homalomena*  $2n=4x=80$ , *Lasia*  $2n=2x=26$ , *Pothos*  $2n=5x=60$ , *Remusatia*  $2n=2x=20$  and  $3x=42$ , *Rhaphidophora*  $2n=4x=56$ , *Sauromatum*  $2n=2x=26$ , *Schismatoglottis*  $2n=2x=30$  and  $4x=52$ , *Scindapsus*  $2n=2x=48$ ,  $56$ ,  $60$ ,  $64$



and 70, *Steudnera*  $2n=2x=16$ , 28 and 36, and  $2n=2x=10$ , 16, 18, 20, 24, 26 and  $3x=32$ , 36 and 52 of genus *Typhonium*.

### 2.2.3 The chromosome studied in Thailand

Kongkung (1999) studied the karyotypic studies on 14 species of elephant yams of the genus *Amorphophallus* (Araceae) and one species of the genus *Tacca* (Taccaceae) from root tips to compare between the Hematoxylin staining and Feulgen squash methods. The Feulgen squash method proved to be easier, more convenient and revealed metaphase chromosomes more clearly than the Hematoxylin staining method. Eight species had the chromosome number ( $2n=26$ ) and formula karyotypes as: *Amorphophallus blumei* Schott. ( $2n=26$ ) =  $L^{m_{14}} + L^{sm_2} + M^{sm_4} + M^a_6$ , *A. corrugatus* N.E.Br. ( $2n=26$ ) =  $L^{m_{18}} + L^{sm_2} + M^m_2 + M^{sm_2} + M^a_2$ , *A. bangkokensis* Gagnep. ( $2n=26$ ) =  $L^m_4 + L^{sm_4} + M^m_6 + M^{sm_6} + M^a_6$ , *A. oncophyllus* Prain. ( $2n=26$ ) =  $L^m_4 + L^{sm_6} + L^{sm_2} + M^{sm_4} + M^a_{10}$ , *A. putii* Gagnep. ( $2n=26$ ) =  $L^m_8 + M^m_{10} + M^a_8$ , *A. saraburiensis* Gagnep. ( $2n=26$ ) =  $L^m_8 + M^m_6 + M^{sm_8} + M^a_4$ , *A. variabilis* Bl. ( $2n=26$ ) =  $L^m_6 + M^m_{10} + M^{sm_6} + S^{sm_2} + S^a_2$  and *Amorphophallus* sp. ( $2n=26$ ) =  $L^m_2 + L^{sm_4} + M^m_6 + M^{sm_{12}} + M^a_2$ . The basic chromosome number of elephant yams plant is  $x = 13$ . Therefore, this group are presented in diploid ( $2n$ ) plant. Seven species had the chromosome number of  $2n=28$  and formula karyotypes as: *Amorphophallus* sp. ( $2n=28$ ) =  $L^{m_{12}} + M^m_6 + M^{sm_8} + M^a_2$ , *A. campanulatus* Bl. ex Decne. ( $2n=28$ ) =  $L^m_2 + L^{sm_4} + M^m_8 + M^{sm_{10}} + M^a_2 + S^{sm_2}$ , *A. kerrii* Gagnep. ( $2n=28$ ) =  $L^{m_{10}} + L^a_4 + M^m_6 + M^a_6$ , *A. koratensis* Gagnep. ( $2n=28$ ) =  $L^m_8 + L^{sm_4} + M^m_4 + M^{sm_6} + M^a_4 + S^{sm_2}$ , *A. longituberosus* ( $2n=28$ ) =  $L^{m_{14}} + M^m_2 + M^{sm_2} + M^a_{10}$  and *Amorphophallus* sp. ( $2n=28$ ) =  $L^{m_{12}} + L^m_2 + M^m_4 + M^{sm_2} + M^a_8$ . This group had a basic number ( $x$ ) = 14 and are diploid plant. All of the elephant yams had an asymmetrical karyotype that consisted metacentric, submetacentric, and acrocentric chromosomes but no telocentric ones. The chromosome number of two species: *Amorphophallus campanulatus* Bl. ex Decne. and *Tacca leontopelaloides* Ktze. had previously been reported. This was the first report on the other 13 species of elephant yams.

Oonsalung *et al.* (2011) reported chromosome number of *Homalomena lindenii*, *Homalomena rubercens* and *Homalomena* sp. of those three species were  $2n=40$ .

Karyotype and idiogram of some Araceae species are studied by few scientists. Therefore, many species in this study will study for the first time.

### 2.3 Plants material

All species of Araceae were collected from Thai forests and cultivated plant and transplanted in the nursery of Walai Rukhavej Botanical Research Institute, Mahasarakham University. Voucher specimens were deposited at the Mahasarakham University Herbarium (Table 2.2).



**Table 2.2** Plants used in the study

Species	Common name	Location	Collector number
1. <i>Agloanema modestum</i>	Khiao muen pi	Loei	R. Senavongse 001/2016
2. <i>A. simplex</i>	Wan ngot hin	Kalasin	R. Senavongse 006/2016
3. <i>Alocacia macrorrhizos</i>	Wan nang-kwak	Mukdahan	R. Senavongse 011/2016
4. <i>A. cucullata</i>	Kacho nok	Songkhla	R. Senavongse 016/2016
5. <i>A. longiloba</i>	Kradat	Maharakham	R. Senavongse 021/2016
6. <i>A. sp.</i>	-	Changmai	R. Senavongse 026/2016
7. <i>Amorphophallus serrulatus</i>	-	Ubon Ratchathani	R. Senavongse 031/2016
8. <i>Arisaema maxwellii</i>	-	Changmai	R. Senavongse 036/2016
9. <i>Colocasia esculenta</i>	Phueak or Bon	Chaiyaphum	R. Senavongse 016/2015
10. <i>C. fallax</i>	Bon-Doi	Changmai	R. Senavongse 041/2016
11. <i>C. gigantea</i>	Khun	Lampang	R. Senavongse 046/2016
12. <i>C. lihengiae</i>	Bon-Yunnan	Tak	R. Senavongse 051/2016
13. <i>Hapaline benthamiana</i>	Bon tao	Chaiyaphum	R. Senavongse 056/2016
14. <i>Homalonema griffithii</i>	Bon khiao	Songkhla	R. Senavongse 061/2016
15. <i>Lasia spinosa</i>	Phuk-Nam	Roi-Et	R. Senavongse 066/2016
16. <i>Pistia stratiotes</i>	Jok	Maharakham	R. Senavongse 071/2016
17. <i>Pycnosphata palmata</i>	-	Ubon Ratchathani	R. Senavongse 076/2016
18. <i>Scittomaglottis calyptrata</i>	Bon khiao	Trang	R. Senavongse 081/2016
19. <i>Typhonium glaucum</i>	-	Ubon Ratchathani	R. Senavongse 086/2016
20. <i>T. trilobatum</i>	Uttapit	Chumphon	R. Senavongse 091/2016

Not: sp. = species

## 2.4 Materials

1. Pot
2. Planting soil
3. Bottles for sampling roots
4. Forceps
5. Slide Warmers
6. Slide and cover glass
7. Beaker
8. Petri dish
9. Dropper
10. Pencils with erasers for knocking chromosome
11. Permanent pen
12. Blotting paper
13. Bottles for soak color
14. Transparent Color Nail Polish
15. Light microscope (Zeiss: Axiostar plus)
16. Distilled water
17. 8-hydroxyquinoline
18. Glacial acetic acid
19. 95% ethyl alcohol
20. 70% ethyl alcohol
21. 1 Normal Hydrochloric acid
22. Aceto-orcin
23. Oil Immersion





## 2.5 Chromosome study

Chromosome studies were made from the root-tips, pre-treated with 8-hydroxyquinoline (2 mM) at 4 °C for 24 hour, and then fixed in a fresh solution of ethanol and glacial acetic acid (3:1) for half-hour at room temperature. Root tips were washed in 95% and 70% ethanol at 5 minutes each and stored in 70% ethanol at 4 °C. After that the storage roots were hydrolyzed in 1 N HCl at 60 °C for 5 minutes. They were washed three times for 5 minutes each in distilled water. They were cut off at about 5 mm from the tip, stained with aceto-orcein about 5 minutes and squashed (Dalington and La Cour, 1966; Sharma and Sharma, 1980). Chromosome numbers were examined under a Light Microscope. Counts were made from 20 cells of each taxon at well-spread metaphase stage. Photographs were taken using light microscope (Zeiss Axiostar plus) at 100x magnification.

## 2.6 Karyotype Analysis

The karyotype studies, chromosome number counting was performed on mitotic metaphase cells under a Light Microscope. Twenty clearly observable and well-spread metaphase chromosome cells were selected and photographed. The length of the short arm chromosome (Ls) and long arm chromosome (Ll) were measured to calculate the length of the total arm chromosome (LT,  $LT = Ls + Ll$ ). The relative length (RL), the centromeric index (CI) and standard deviation (SD) of the RL and CI were estimated (Chaiyasut, 1989).

$$RL = \frac{\text{Length of each bars chromosome (LT = Ll+Ls)}}{\text{Length of total arm chromosome of all pairs (ΣLT)}}$$

$$CI = \frac{\text{Length of each bars chromosome (Ll)}}{\text{Length of total arm chromosome of all pairs (LT)}}$$

Chromosomes are value CI between 0.500-0.599 is metacentric chromosomes type

Chromosomes are value CI between 0.600-0.699 is submetacentric chromosomes type

Chromosomes are value CI between 0.700-0.899 is acrocentric chromosomes type

Chromosomes are value CI between 0.900-1.000 is telocentric chromosomes type

To determine the size of the chromosome was Large (L) size chromosome smaller than half the length of the largest chromosome combined with the smallest chromosome.

$$L > (LT_{\text{largest}} + LT_{\text{smallest}}) / 2$$

Medium (M) size chromosome was chromosomes smaller than half the length of the largest chromosome combined with the smallest chromosome. But, more than half of the average length of the largest chromosome.

$$(LT_{\text{largest}} + LT_{\text{smallest}}) / 2 > M > LT_{\text{largest}} / 2$$

Small (S) size chromosome was chromosomes smaller than half the length of the largest chromosome.



$$S < LT_{\text{largest}} / 2$$

The fundamental number (number of chromosome arm, NF) was obtained by assigning a value of two to metacentric, submetacentric and acrocentric chromosomes and one to telocentric chromosomes. All parameters were used in karyotyping and idiogramming.

## 2.7 Idiogram and karyotype formula

Well spread and clearly twenty cells from root tip were performed on mitotic metaphase cells and photographs under light microscope (Zeiss: Axiostar plus). The length of short arm chromosomes (Ls) and the lengths of long arm chromosomes (Ll) were measured and total arm length of the chromosomes were calculated (LT,  $LT = Ls + Ll$ ). The relative length (RL) and the centromeric index (CI) were estimated. CI was also computed to classify the types of chromosomes according to Chaivasut (1989). All parameters were used in karyotyping and idiogramming.

## 2.8 Results

The chromosome number, karyotype and idiogram are 12 genus 20 species of Araceae in Thailand (Table 3.7.1, 3.8.1) were studied from root tip by Feulgen squash Technique (modification Sharma and Sharma, 1980). Chromosome counting was performed on mitotic metaphase cells under light microscope. The software Adobe photoshop and Microsoft office Excel was used for karyotype and idiogram study. The results of chromosome number, karyotype and idiogram of each of species are detailed as follows Table 2.3, 2.24.

**Table 2.3** Chromosome numbers (diploid) and Fundamental Number (NF) of 20 species Araceae in Thailand.

Species	Chromosome numbers (2n)	NF	Basic Chromosome numbers (x)
1. <i>Agloanema modestum</i>	40	80	2x
2. <i>A. simplex</i>	42	84	2x
3. <i>Alocasia cucullata</i>	28	56	2x
4. <i>A. macrorrhizos</i>	28	56	2x
5. <i>A. longiloba</i>	58	112	2x
6. <i>A. sp.</i>	28	56	2x
7. <i>Amorphophallus serrulatus</i>	26	52	2x
8. <i>Arisaema maxwellii</i>	24	48	2x
9. <i>Colocasia esculenta</i>	28	56	2x
10. <i>C. fallax</i>	28	56	2x
11. <i>C. gigantea</i>	28	56	2x
12. <i>C. lihengiae</i>	28	56	2x
13. <i>Hapaline benthamiana</i>	26	52	2x
14. <i>Homalonema griffithii</i>	40	80	2x
15. <i>Lasia spinosa</i>	26	52	2x
16. <i>Pistia stratiotes</i>	28	56	2x



**Table 2.3** Chromosome numbers (diploid) and Fundamental Number (NF) of 20 species Araceae in Thailand (Continued).

Species	Chromosome numbers (2n)	NF	Basic Chromosome numbers (x)
17. <i>Pycnosphata palmata</i>	26	52	2x
18. <i>Scittomaglottis calyptrata</i>	24	52	2x
19. <i>Typhonium glaucum</i>	24	48	2x
20. <i>T. trilobatum</i>	18	36	2x

Notes: sp. = species, 2n = diploid chromosome number and NF = fundamental number (number of chromosome arm).

A study chromosome number of 20 species of Araceae in Thailand from table 2.3 can be summarized as follows; the chromosome number of *Agloanema modestum*  $2n=2x=40$ , *A. simplex*  $2n=2x=42$ , *Alocasia cucullata*  $2n=2x=28$ , *A. macrorrhizos*  $2n=2x=28$ , *A. longiloba*  $2n=2x=58$ , *A. sp.*  $2n=2x=28$ , *Amorphophallus serrulatus*  $2n=2x=26$ , *Arisaema maxwellii*  $2n=2x=24$ , *Colocasia esculenta*  $2n=2x=28$ , *C. fallax*  $2n=2x=28$ , *C. gigantea*  $2n=2x=28$ , *C. lihengiae*  $2n=2x=28$ , *Hapaline benthamiana*  $2n=2x=26$ , *Homalonema griffithii*  $2n=2x=40$ , *Lasia spinosa*  $2n=2x=26$ , *Pistia stratiotes*  $2n=2x=28$ , *Pycnosphata palmata*  $2n=2x=26$ , *Scittomaglottis calyptrata*  $2n=2x=24$ , *Typhonium glaucum*  $2n=2x=24$  and *T. trilobatum*  $2n=2x=18$ , respectively.



### 2.8.1 *Aglaonema modestum* Schott ex Engl.

Counting somatic metaphase chromosome from 20 cells of *Aglaonema modestum* and then the arrangement of karyotypes and diogram by average length of the short arm (Ls) average length of the long arm (Ll) are measured by the length of each chromosome (LT). The average measurement of Relative Length (RL), Chromosome Index (CI), standard deviation (SD) of RL and CI from metaphase chromosomes in 20 cells and summarizes the size and type of chromosomes are shown in Table 2.4.



**Figure 2.1** *Aglaonema modestum* Schott ex Engl.

- A. Plants in habitat
- B. Leaves and fruits
- C. Inflorescence

**Table 2.4** Mean length of short arm chromosome (Ls), long arm chromosome (Ll), total arm chromosome (LT), relative length (RL), centromeric index (CI) and standard deviation (SD) of RL, CI from 20 metaphases of *Aglaonema modestum* (diploid,  $2n=40$ ).

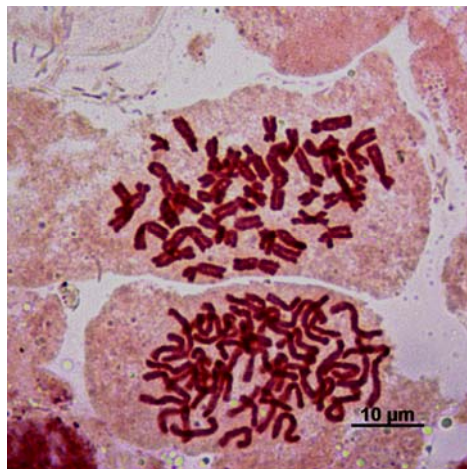
Chromosome pair	Ls	Ll	LT	RL $\pm$ SD	CI $\pm$ SD	Chromosome size	Chromosome type
1	78.119	87.783	165.902	0.076 $\pm$ 0.002	0.529 $\pm$ 0.000	Large	Metacentric
2	63.939	86.358	150.297	0.069 $\pm$ 0.001	0.575 $\pm$ 0.000	Large	Metacentric
3	59.284	76.141	135.425	0.061 $\pm$ 0.000	0.556 $\pm$ 0.000	Large	Metacentric
4	30.608	103.631	134.239	0.061 $\pm$ 0.002	0.772 $\pm$ 0.000	Large	Acrocentric
5	54.364	68.087	122.451	0.054 $\pm$ 0.003	0.686 $\pm$ 0.000	Large	Submetacentric
6	52.695	69.725	122.42	0.053 $\pm$ 0.000	0.535 $\pm$ 0.000	Large	Metacentric
7	37.447	81.897	119.344	0.048 $\pm$ 0.000	0.580 $\pm$ 0.000	Large	Metacentric
8	44.765	70.876	115.641	0.056 $\pm$ 0.000	0.556 $\pm$ 0.000	Large	Metacentric
9	53.688	61.704	115.392	0.054 $\pm$ 0.004	0.613 $\pm$ 0.000	Large	Metacentric
10	32.557	82.286	114.843	0.056 $\pm$ 0.000	0.570 $\pm$ 0.000	Large	Metacentric
11	44.370	65.647	110.017	0.052 $\pm$ 0.000	0.717 $\pm$ 0.000	Large	Acrocentric
12	44.304	61.260	105.564	0.048 $\pm$ 0.005	0.740 $\pm$ 0.000	Large	Acrocentric
13	28.146	77.069	105.215	0.050 $\pm$ 0.000	0.597 $\pm$ 0.000	Large	Metacentric
14	39.690	65.371	105.061	0.043 $\pm$ 0.000	0.620 $\pm$ 0.000	Large	Submetacentric
15	36.263	59.065	95.328	0.048 $\pm$ 0.006	0.622 $\pm$ 0.000	Medium	Submetacentric
16	33.636	56.594	90.230	0.041 $\pm$ 0.000	0.634 $\pm$ 0.000	Medium	Submetacentric
17	35.217	54.306	89.523	0.041 $\pm$ 0.000	0.607 $\pm$ 0.000	Medium	Submetacentric
18	32.390	49.357	81.747	0.037 $\pm$ 0.000	0.604 $\pm$ 0.000	Medium	Submetacentric
19	26.170	41.020	67.190	0.031 $\pm$ 0.003	0.611 $\pm$ 0.000	Small	Submetacentric
20	20.913	29.698	50.611	0.023 $\pm$ 0.002	0.587 $\pm$ 0.000	Small	Metacentric



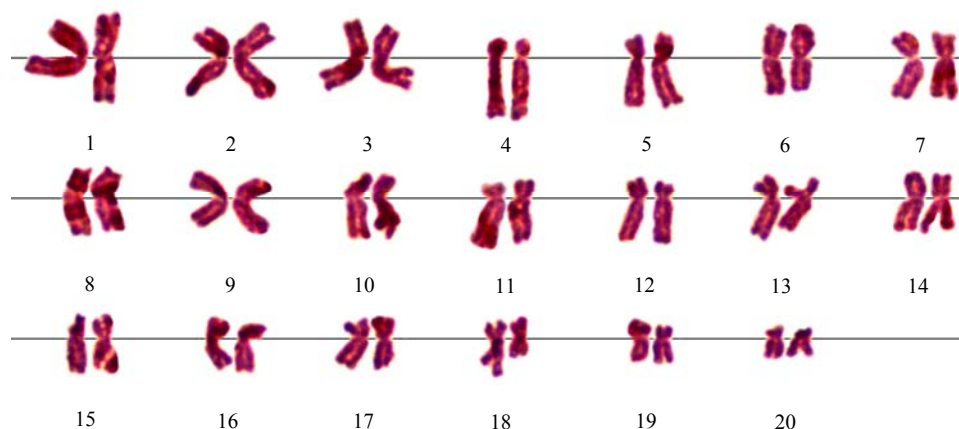
Karyotype formula is  $L_{18}^m + L_4^{sm} + L_6^a + M_8^{sm} + S_2^m + S_2^{sm}$

#### Study on number, type and structure of the Chromosome

Chromosome number of *Aglaonema modestum* is diploid chromosome  $2n=40$  contains the chromosomes 20 pairs or 40 bars. The karyotype formula of this species was found to be asymmetrical,  $L_{18}^m + L_4^{sm} + L_6^a + M_8^{sm} + S_2^m + S_2^{sm}$ , include 9 pair a large metacentric chromosome, 2 pair a large submetacentric chromosome, 3 pair a large acrocentric chromosome, 4 pair a medium submetacentric chromosome, 1 pair a small metacentric chromosome and 1 pair a small submetacentric chromosome. The relative length is a value between  $0.023 \pm 0.002$  to  $0.076 \pm 0.002$  (Table 2.4 and Figures 2.2-2.4).

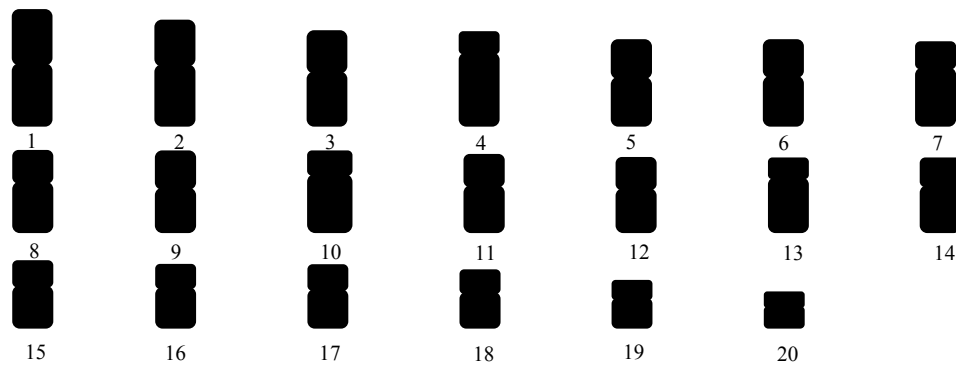


**Figure 2.2** Somatic metaphase chromosome plate of *Aglaonema modestum* (diploid,  $2n=40$ ).



**Figure 2.3** Karyotype of *Aglaonema modestum* by conventional staining.





**Figure 2.4** Idiogram of *Aglaonema modestum* by conventional staining.



### 2.8.2 *Aglaonema simplex* (Blume) Blume

Counting somatic metaphase chromosome from 20 cells of *Aglaonema simplex* and then the arrangement of karyotypes and diogram by average length of the short arm (Ls) average length of the long arm (Ll) are measured by the length of each chromosome (LT). The average measurement of Relative Length (RL), Chromosome Index (CI), standard deviation (SD) of RL and CI from metaphase chromosomes in 20 cells and summarizes the size and type of chromosomes are shown in Table 2.5.



**Figure 2.5** *Aglaonema simplex* (Blume) Blume

**Table 2.5** Mean length of short arm chromosome (Ls), long arm chromosome (Ll), total arm chromosome (LT), relative length (RL), centromeric index (CI) and standard deviation (SD) of RL, CI from 20 metaphases of *Aglaonema simplex* (diploid,  $2n=42$ ).

Chromosome pair	Ls	Ll	LT	RL $\pm$ SD	CI $\pm$ SD	Chromosome size	Chromosome type
1	60.281	127.598	187.879	0.065 $\pm$ 0.003	0.640 $\pm$ 0.000	Large	Submetacentric
2	49.923	128.280	178.203	0.063 $\pm$ 0.002	0.660 $\pm$ 0.000	Large	Submetacentric
3	51.401	118.985	170.386	0.060 $\pm$ 0.000	0.650 $\pm$ 0.000	Large	Submetacentric
4	35.374	125.951	161.325	0.057 $\pm$ 0.000	0.747 $\pm$ 0.000	Large	Acrocentric
5	46.460	111.740	158.200	0.056 $\pm$ 0.003	0.688 $\pm$ 0.003	Large	Submetacentric
6	42.615	110.670	153.285	0.054 $\pm$ 0.000	0.661 $\pm$ 0.000	Large	Submetacentric
7	46.220	106.280	152.500	0.053 $\pm$ 0.000	0.751 $\pm$ 0.000	Large	Acrocentric
8	27.301	125.046	152.347	0.053 $\pm$ 0.002	0.601 $\pm$ 0.000	Large	Submetacentric
9	60.427	91.054	151.481	0.053 $\pm$ 0.000	0.508 $\pm$ 0.000	Large	Metacentric
10	73.874	76.001	149.875	0.052 $\pm$ 0.000	0.643 $\pm$ 0.002	Large	Submetacentric
11	46.962	102.700	149.662	0.051 $\pm$ 0.001	0.545 $\pm$ 0.000	Large	Metacentric
12	55.502	85.045	140.547	0.049 $\pm$ 0.000	0.703 $\pm$ 0.000	Large	Acrocentric
13	32.972	106.919	139.891	0.049 $\pm$ 0.000	0.682 $\pm$ 0.000	Large	Submetacentric
14	45.021	84.752	129.773	0.046 $\pm$ 0.001	0.627 $\pm$ 0.001	Large	Submetacentric
15	40.219	80.343	120.562	0.042 $\pm$ 0.000	0.733 $\pm$ 0.000	Medium	Acrocentric
16	36.100	77.843	113.943	0.039 $\pm$ 0.000	0.748 $\pm$ 0.000	Medium	Acrocentric
17	42.116	71.027	113.143	0.039 $\pm$ 0.001	0.695 $\pm$ 0.000	Medium	Submetacentric
18	46.160	66.360	112.520	0.036 $\pm$ 0.000	0.523 $\pm$ 0.000	Medium	Metacentric
19	39.263	52.702	91.965	0.032 $\pm$ 0.002	0.687 $\pm$ 0.000	Small	Submetacentric
20	33.916	45.340	79.256	0.028 $\pm$ 0.000	0.686 $\pm$ 0.000	Small	Submetacentric
21	25.094	40.707	65.801	0.023 $\pm$ 0.002	0.609 $\pm$ 0.000	Small	Submetacentric



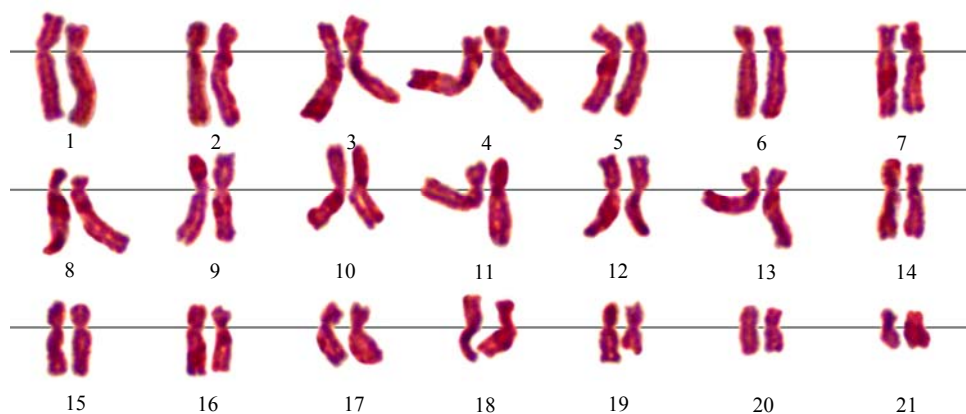
Karyotype formula is  $L^m_4 + L^{sm}_{18} + L^a_6 + M^m_2 + M^{sm}_2 + M^a_4 + S^{sm}_6$

Study on number, type and structure of the Chromosome

Chromosome number of *Aglaonema simplex* is diploid chromosome  $2n=42$  contains the chromosomes 21 pairs or 42 bars. The karyotype formula of this species was found to be asymmetrical,  $L^m_4 + L^{sm}_{18} + L^a_6 + M^m_2 + M^{sm}_2 + M^a_4 + S^{sm}_6$ , include 2 pair a large metacentric chromosome, 9 pair a large submetacentric chromosome, 3 pair a large acrocentric chromosome, 1 pair a medium metacentric chromosome, 1 pair a medium submetacentric chromosome, 2 pair a medium acrocentric chromosome and 3 pair a small submetacentric chromosome. The relative length is a value between  $0.023 \pm 0.002$  to  $0.065 \pm 0.003$  (Table 2.5 and Figures 2.6-2.8).



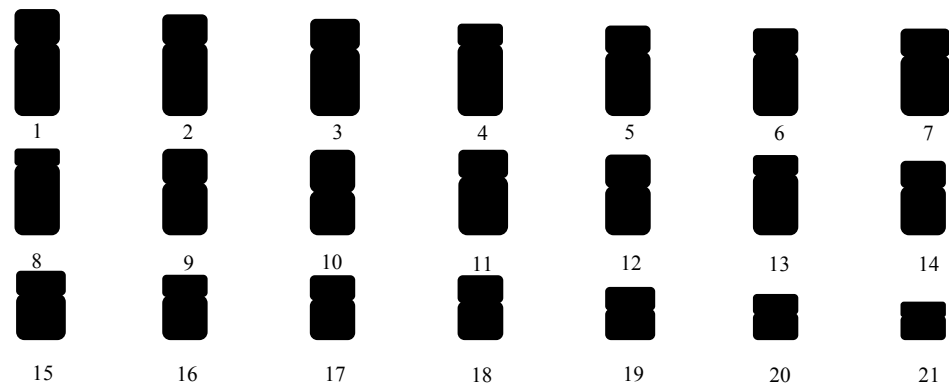
**Figure 2.6** Somatic metaphase chromosome plate of *Aglaonema simplex* (diploid,  $2n=42$ ).



**Figure 2.7** Karyotype of *Aglaonema simplex* by conventional staining.







**Figure 2.8** Idiogram of *Aglaonema simplex* by conventional staining.



### 2.8.3 *Alocasia cucullata* (Lour.) G.Don

Counting somatic metaphase chromosome from 20 cells of *Alocasia cucullata* and then the arrangement of karyotypes and diogram by average length of the short arm (Ls) average length of the long arm (Ll) are measured by the length of each chromosome (LT). The average measurement of Relative Length (RL), Chromosome Index (CI), standard deviation (SD) of RL and CI from metaphase chromosomes in 20 cells and summarizes the size and type of chromosomes are shown in Table 2.6.



**Figure 2.9** *Alocasia cucullata* (Lour.) G.Don

- A. Plants in habitat  
B. Inflorescence

**Table 2.6** Mean length of short arm chromosome (Ls), long arm chromosome (Ll), total arm chromosome (LT), relative length (RL), centromeric index (CI) and standard deviation (SD) of RL, CI from 20 metaphases of *Alocasia cucullata* (diploid,  $2n=28$ ).

Chromosome pair	Ls	Ll	LT	RL $\pm$ SD	CI $\pm$ SD	Chromosome size	Chromosome type
*1	54.944	59.056	114.000	0.091 $\pm$ 0.000	0.518 $\pm$ 0.000	Large	Metacentric
*2	56.943	44.426	101.369	0.081 $\pm$ 0.000	0.500 $\pm$ 0.000	Large	Metacentric
3	49.600	49.671	99.271	0.079 $\pm$ 0.000	0.500 $\pm$ 0.000	Large	Metacentric
4	39.346	56.617	95.963	0.077 $\pm$ 0.002	0.590 $\pm$ 0.000	Large	Metacentric
5	42.593	53.213	95.806	0.076 $\pm$ 0.000	0.555 $\pm$ 0.000	Large	Metacentric
6	44.343	49.772	94.115	0.075 $\pm$ 0.000	0.529 $\pm$ 0.000	Large	Metacentric
7	42.830	51.170	94.000	0.075 $\pm$ 0.003	0.544 $\pm$ 0.000	Large	Metacentric
8	43.056	50.701	93.757	0.075 $\pm$ 0.001	0.541 $\pm$ 0.000	Large	Metacentric
9	40.050	51.451	91.501	0.073 $\pm$ 0.000	0.562 $\pm$ 0.000	Large	Metacentric
10	39.795	43.451	83.246	0.066 $\pm$ 0.001	0.522 $\pm$ 0.000	Medium	Metacentric
11	26.539	56.254	82.793	0.066 $\pm$ 0.000	0.679 $\pm$ 0.000	Medium	Submetacentric
12	23.803	46.472	70.275	0.056 $\pm$ 0.001	0.661 $\pm$ 0.000	Medium	Submetacentric
13	27.049	43.201	70.250	0.056 $\pm$ 0.000	0.615 $\pm$ 0.000	Medium	Submetacentric
14	29.549	37.966	67.515	0.054 $\pm$ 0.000	0.562 $\pm$ 0.000	Medium	Metacentric

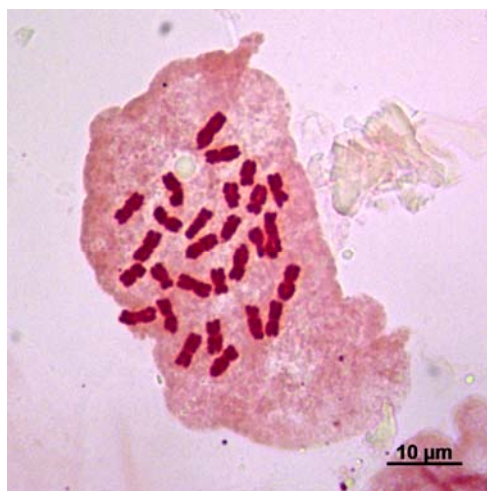
Remarks: \*=satellite chromosome (NORs).

Karyotype formula is  $L_{18}^m + M_4^m + M_6^{sm}$

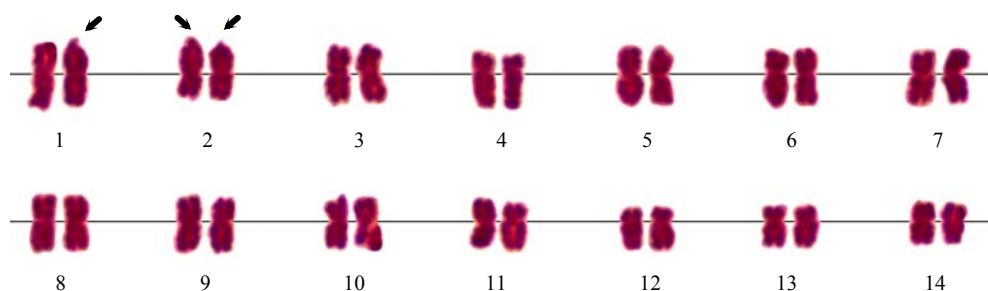


### Study on number, type and structure of the Chromosome

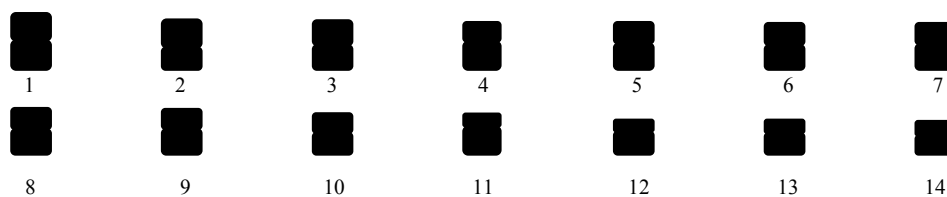
Chromosome number of *Alocasia cucullata* is diploid chromosome  $2n=28$  contains the chromosomes 14 pairs or 28 bars. The karyotype formula of this species was found to be symmetrical,  $L_{18}^m + M_4^m + M_6^{sm}$ , include 9 pair a large metacentric chromosome, 2 pair a medium metacentric chromosome and 3 pair a medium submetacentric chromosome. The relative length is a value between  $0.054 \pm 0.000$  to  $0.091 \pm 0.000$  and three visible satellites (Table 2.6 and Figures 2.10-2.12).



**Figure 2.10** Somatic metaphase chromosome plate of *Alocasia cucullata* (diploid,  $2n=28$ ).



**Figure 2.11** Karyotype of *Alocasia cucullata* (Arrows indicate satellite chromosomes.) by conventional staining.

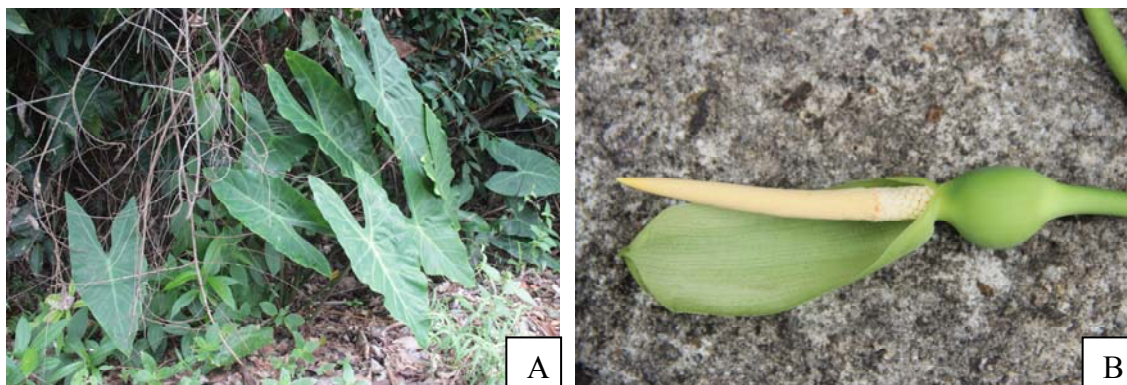


**Figure 2.12** Idiogram of *Alocasia cucullata* by conventional staining.



#### 2.8.4 *Alocasia longiloba* Miq.

Counting somatic metaphase chromosome from 20 cells of *Alocasia longiloba* and then the arrangement of karyotypes and diogram by average length of the short arm (Ls) average length of the long arm (Ll) are measured by the length of each chromosome (LT). The average measurement of Relative Length (RL), Chromosome Index (CI), standard deviation (SD) of RL and CI from metaphase chromosomes in 20 cells and summarizes the size and type of chromosomes are shown in Table 2.7.



**Figure 2.13** *Alocasia longiloba* Miq.

A. Plants in habitat

B. Inflorescence

**Table 2.5** Mean length of short arm chromosome (Ls), long arm chromosome (Ll), total arm chromosome (LT), relative length (RL), centromeric index (CI) and standard deviation (SD) of RL, CI from 20 metaphases of *Alocasia longiloba* (diploid,  $2n=58$ ).

Chromosome pair	Ls	Ll	LT	RL $\pm$ SD	CI $\pm$ SD	Chromosome size	Chromosome type
1	47.471	61.453	108.924	0.048 $\pm$ 0.003	0.564 $\pm$ 0.000	Large	Metacentric
2	46.516	55.938	102.454	0.046 $\pm$ 0.000	0.546 $\pm$ 0.000	Large	Metacentric
3	41.828	54.049	95.877	0.043 $\pm$ 0.000	0.564 $\pm$ 0.000	Large	Metacentric
4	43.075	50.666	93.741	0.042 $\pm$ 0.000	0.540 $\pm$ 0.000	Large	Metacentric
5	38.105	54.392	92.497	0.041 $\pm$ 0.001	0.588 $\pm$ 0.000	Large	Metacentric
6	38.234	51.999	90.233	0.040 $\pm$ 0.001	0.576 $\pm$ 0.000	Large	Metacentric
7	38.014	49.306	87.320	0.039 $\pm$ 0.000	0.565 $\pm$ 0.000	Large	Metacentric
8	39.087	45.293	84.380	0.038 $\pm$ 0.001	0.537 $\pm$ 0.000	Large	Metacentric
9	35.530	48.625	84.155	0.037 $\pm$ 0.002	0.578 $\pm$ 0.000	Large	Metacentric
10	38.909	44.719	83.628	0.037 $\pm$ 0.000	0.535 $\pm$ 0.000	Large	Metacentric
11	27.813	54.669	82.482	0.037 $\pm$ 0.000	0.663 $\pm$ 0.000	Large	Metacentric
12	35.506	46.210	81.716	0.036 $\pm$ 0.002	0.566 $\pm$ 0.000	Large	Metacentric
13	34.165	45.780	79.945	0.036 $\pm$ 0.000	0.577 $\pm$ 0.000	Medium	Metacentric
14	37.482	42.396	79.878	0.036 $\pm$ 0.002	0.531 $\pm$ 0.000	Medium	Metacentric
15	31.636	44.840	76.476	0.034 $\pm$ 0.003	0.586 $\pm$ 0.000	Medium	Metacentric
16	29.730	44.701	74.431	0.033 $\pm$ 0.000	0.601 $\pm$ 0.000	Medium	Submetacentric
17	36.221	37.602	73.823	0.033 $\pm$ 0.003	0.509 $\pm$ 0.000	Medium	Metacentric
18	28.660	43.096	71.756	0.032 $\pm$ 0.000	0.601 $\pm$ 0.000	Medium	Submetacentric
19	30.129	40.190	70.319	0.031 $\pm$ 0.000	0.572 $\pm$ 0.000	Medium	Metacentric
20	24.866	44.430	69.296	0.031 $\pm$ 0.000	0.641 $\pm$ 0.000	Medium	Submetacentric



Chromosome pair	Ls	LI	LT	RL±SD	CI±SD	Chromosome size	Chromosome type
21	19.975	48.533	68.508	0.030±0.000	0.708±0.000	Medium	Acrocentric
22	23.665	43.846	67.511	0.030±0.000	0.650±0.000	Medium	Submetacentric
23	24.882	42.174	67.056	0.030±0.000	0.629±0.000	Medium	Submetacentric
24	20.413	45.410	65.823	0.029±0.000	0.690±0.000	Medium	Submetacentric
25	28.700	35.090	63.790	0.028±0.002	0.550±0.000	Medium	Metacentric
26	21.417	42.122	63.539	0.028±0.000	0.663±0.000	Medium	Submetacentric
27	19.423	40.510	59.933	0.027±0.003	0.676±0.000	Medium	Submetacentric
28	23.633	34.389	58.022	0.026±0.000	0.593±0.000	Medium	Metacentric
29	17.916	33.368	51.284	0.023±0.001	0.651±0.000	Small	Submetacentric

Karyotype formula is  $L_{24}^m + M_{14}^m + M_{16}^{sm} + M_2^a + S_2^{sm}$

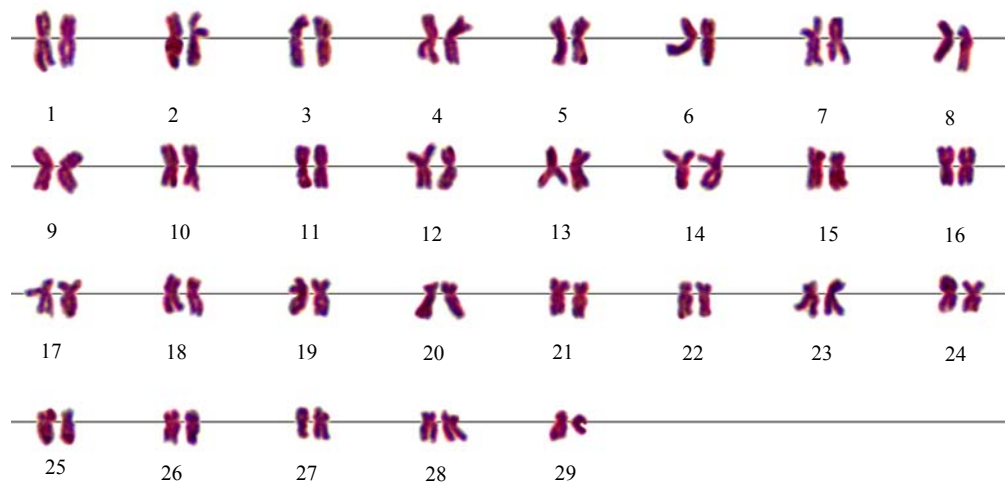
#### Study on number, type and structure of the Chromosome

Chromosome number of *Alocasia longiloba* is diploid chromosome  $2n=58$  contains the chromosomes 29 pairs or 58 bars. The karyotype formula of this species was found to be asymmetrical,  $L_{24}^m + M_{14}^m + M_{16}^{sm} + M_2^a + S_2^{sm}$ , include 12 pair a large metacentric chromosome, 7 pair a medium metacentric chromosome, 8 pair a medium submetacentric chromosome, 1 pair a medium acrocentric chromosome and 1 pair a small submetacentric chromosome. The relative length is a value between  $0.023\pm0.001$  to  $0.048\pm0.003$  (Table 2.7 and Figures 2.14-2.16).

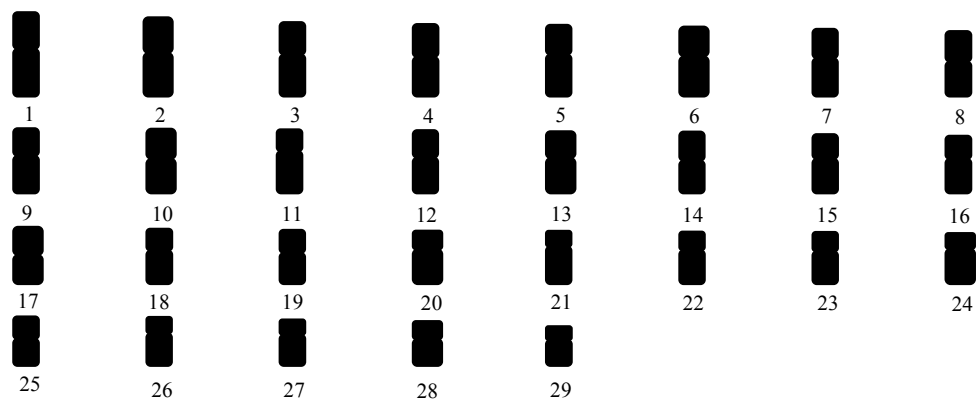


**Figure 2.14** Somatic metaphase chromosome plate of *Alocasia longiloba* (diploid,  $2n=58$ ).





**Figure 2.15** Karyotype of *Alocasia longiloba* by conventional staining.



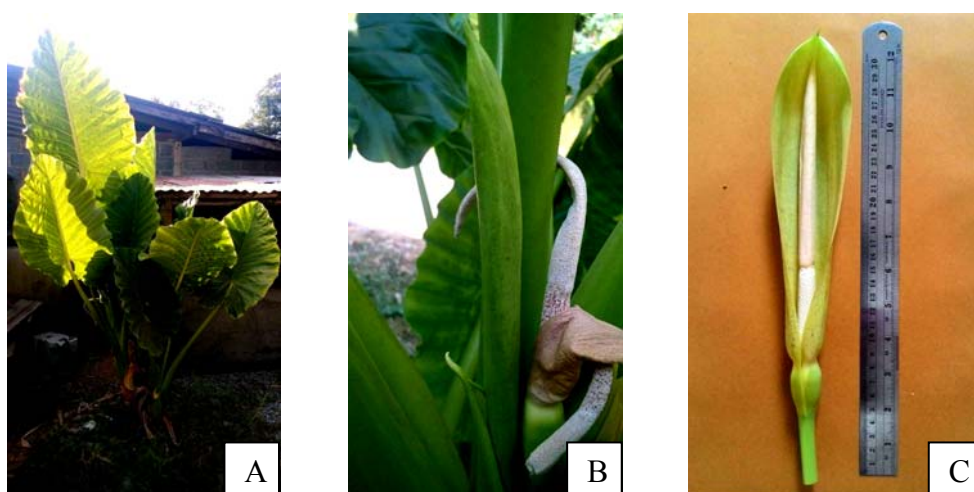
**Figure 2.16** Idiogram of *Alocasia longiloba* by conventional staining.





### 2.8.5 *Alocasia macrorrhizos* (L.) G.Don

Counting somatic metaphase chromosome from 20 cells of *Alocasia macrorrhizos* and then the arrangement of karyotypes and diagram by average length of the short arm (Ls) average length of the long arm (Ll) are measured by the length of each chromosome (LT). The average measurement of Relative Length (RL), Chromosome Index (CI), standard deviation (SD) of RL and CI from metaphase chromosomes in 20 cells and summarizes the size and type of chromosomes are shown in Table 2.8.



**Figure 2.17** *Alocasia macrorrhizos* (L.) G.Don

A. Plants in habitat

B-C. Inflorescence

**Table 2.8** Mean length of short arm chromosome (Ls), long arm chromosome (Ll), total arm chromosome (LT), relative length (RL), centromeric index (CI) and standard deviation (SD) of RL, CI from 20 metaphases of *Alocasia macrorrhizos* (diploid,  $2n=28$ ).

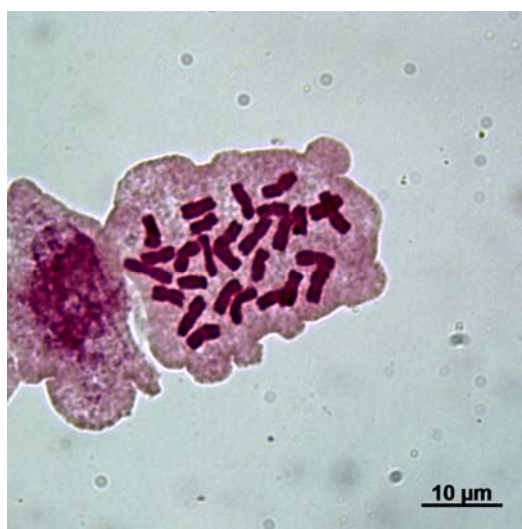
Chromosome pair	Ls	Ll	LT	RL $\pm$ SD	CI $\pm$ SD	Chromosome size	Chromosome type
1	55.193	56.863	112.056	0.093 $\pm$ 0.000	0.507 $\pm$ 0.000	Large	Metacentric
2	48.335	53.171	101.506	0.084 $\pm$ 0.000	0.524 $\pm$ 0.000	Large	Metacentric
3	43.221	55.887	99.108	0.082 $\pm$ 0.000	0.564 $\pm$ 0.000	Large	Metacentric
4	45.585	45.103	90.688	0.075 $\pm$ 0.000	0.497 $\pm$ 0.000	Large	Metacentric
5	39.76	47.753	87.513	0.073 $\pm$ 0.000	0.546 $\pm$ 0.000	Medium	Metacentric
6	35.332	51.819	87.151	0.072 $\pm$ 0.000	0.595 $\pm$ 0.000	Medium	Metacentric
7	33.287	51.422	84.709	0.070 $\pm$ 0.000	0.607 $\pm$ 0.000	Medium	Submetacentric
8	31.543	52.578	84.121	0.070 $\pm$ 0.000	0.625 $\pm$ 0.000	Medium	Submetacentric
9	37.895	43.95	81.845	0.068 $\pm$ 0.000	0.537 $\pm$ 0.000	Medium	Metacentric
10	26.836	51.431	78.267	0.065 $\pm$ 0.000	0.657 $\pm$ 0.000	Medium	Submetacentric
11	35.307	42.361	77.668	0.065 $\pm$ 0.000	0.545 $\pm$ 0.000	Medium	Metacentric
12	32.938	43.956	76.894	0.064 $\pm$ 0.000	0.572 $\pm$ 0.000	Medium	Metacentric
13	32.34	41.212	73.552	0.061 $\pm$ 0.000	0.560 $\pm$ 0.000	Medium	Metacentric
14	22.739	45.706	68.445	0.057 $\pm$ 0.000	0.668 $\pm$ 0.000	Medium	Submetacentric



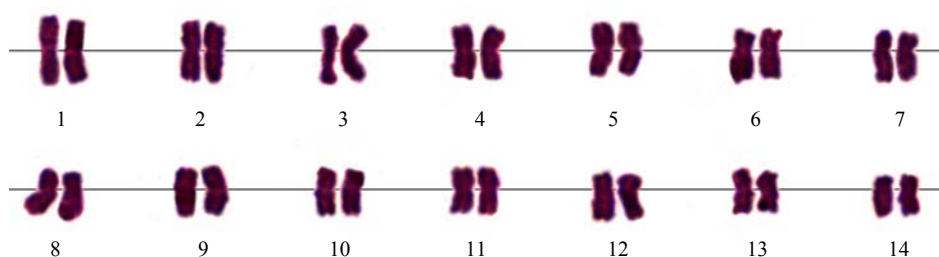
Karyotype formula is  $L^m_8 + M^m_{12} + M^{sm}_8$

Study on number, type and structure of the Chromosome

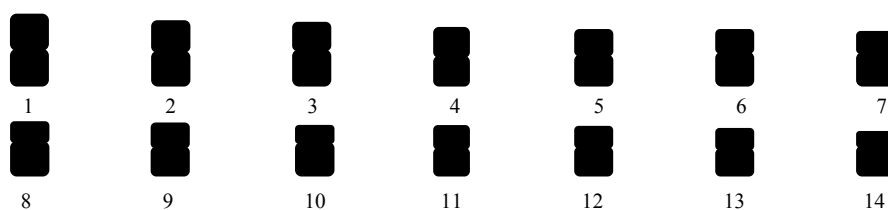
Chromosome number of *Alocasia macrorrhizos* is diploid chromosome  $2n=28$  contains the chromosomes 14 pairs or 28 bars. The karyotype formula of this species was found to be symmetrical,  $L^m_8 + M^m_{12} + M^{sm}_8$ , include 4 pair a large metacentric chromosome, 6 pair a medium metacentric chromosome and 4 pair a medium submetacentric chromosome. The relative length is a value between  $0.057 \pm 0.000$  to  $0.093 \pm 0.000$  (Table 2.8 and Figures 2.18-2.20).



**Figure 2.18** Somatic metaphase chromosome plate of *Alocasia macrorrhizos* (diploid,  $2n=28$ ).



**Figure 2.19** Karyotype of *Alocasia macrorrhizos* by conventional staining.



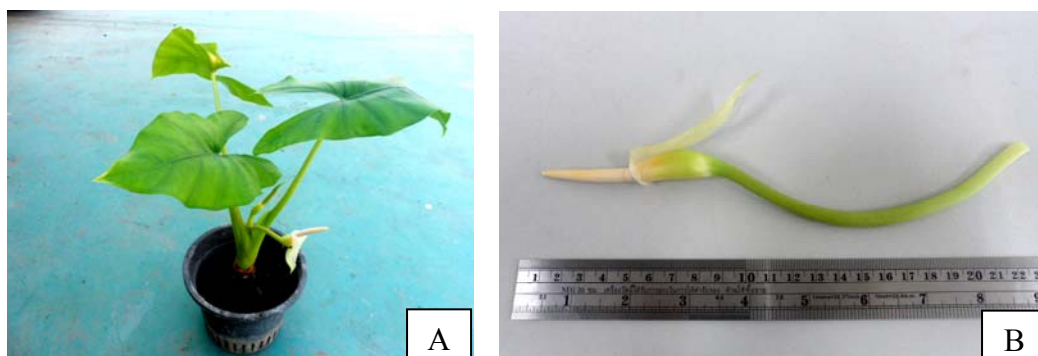
**Figure 2.20** Idiogram of *Alocasia macrorrhizos* by conventional staining.





### 2.8.6 *Alocasia* sp.

Counting somatic metaphase chromosome from 20 cells of *Alocasia* sp. and then the arrangement of karyotypes and diagram by average length of the short arm (Ls) average length of the long arm (Ll) are measured by the length of each chromosome (LT). The average measurement of Relative Length (RL), Chromosome Index (CI), standard deviation (SD) of RL and CI from metaphase chromosomes in 20 cells and summarizes the size and type of chromosomes are shown in Table 2.9.



**Figure 2.21** *Alocasia* sp.  
A. Plants in habitat  
B. Inflorescence

**Table 2.9** Mean length of short arm chromosome (Ls), long arm chromosome (Ll), total arm chromosome (LT), relative length (RL), centromeric index (CI) and standard deviation (SD) of RL, CI from 20 metaphases of *Alocasia* sp. (diploid,  $2n=28$ ).

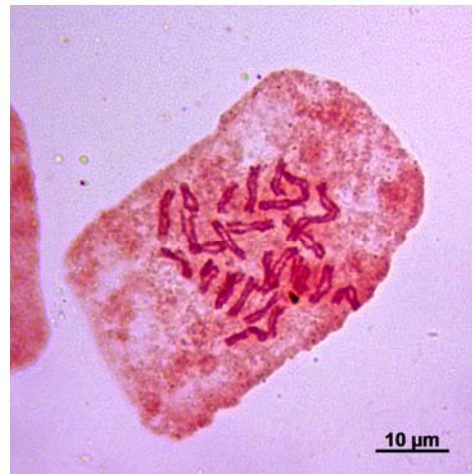
Chromosome pair	Ls	Ll	LT	RL $\pm$ SD	CI $\pm$ SD	Chromosome size	Chromosome type
1	41.120	109.440	150.560	0.096 $\pm$ 0.005	0.727 $\pm$ 0.000	Large	Acrocentric
2	69.090	74.838	143.928	0.092 $\pm$ 0.000	0.520 $\pm$ 0.000	Large	Metacentric
3	62.248	67.698	129.945	0.083 $\pm$ 0.005	0.521 $\pm$ 0.001	Large	Metacentric
4	37.538	88.251	125.789	0.080 $\pm$ 0.000	0.702 $\pm$ 0.000	Large	Acrocentric
5	56.786	68.424	125.210	0.080 $\pm$ 0.000	0.547 $\pm$ 0.000	Large	Metacentric
6	30.262	89.146	119.408	0.076 $\pm$ 0.003	0.747 $\pm$ 0.000	Large	Acrocentric
7	37.727	74.636	112.363	0.072 $\pm$ 0.000	0.664 $\pm$ 0.000	Medium	Submetacentric
8	32.175	73.196	105.371	0.067 $\pm$ 0.000	0.695 $\pm$ 0.000	Medium	Submetacentric
9	45.555	55.611	101.166	0.065 $\pm$ 0.001	0.550 $\pm$ 0.000	Medium	Submetacentric
10	46.960	54.024	100.984	0.064 $\pm$ 0.000	0.535 $\pm$ 0.000	Medium	Metacentric
11	31.556	66.840	98.396	0.063 $\pm$ 0.002	0.679 $\pm$ 0.000	Medium	Submetacentric
12	31.881	61.546	93.427	0.060 $\pm$ 0.000	0.659 $\pm$ 0.000	Medium	Submetacentric
13	31.541	56.529	88.067	0.056 $\pm$ 0.002	0.642 $\pm$ 0.001	Medium	Submetacentric
14	30.015	45.125	75.134	0.048 $\pm$ 0.002	0.601 $\pm$ 0.000	Small	Submetacentric

Karyotype formula is  $L_6^m + L_6^a + M_2^m + M_{12}^{sm} + S_2^{sm}$

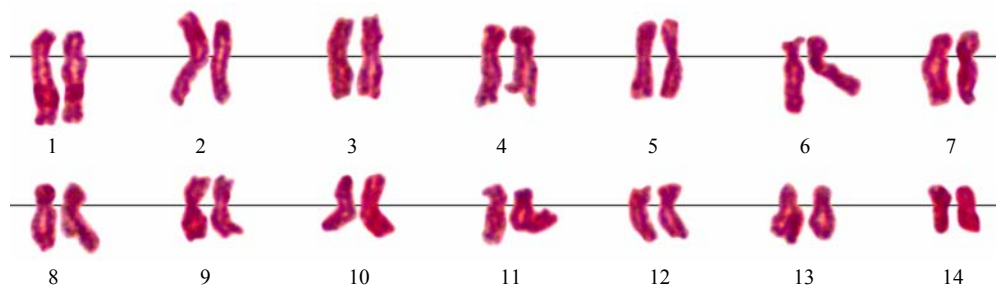


### Study on number, type and structure of the Chromosome

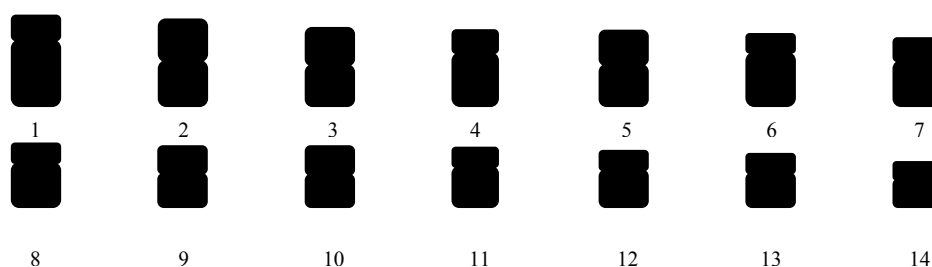
Chromosome number of *Alocasia* sp. is diploid chromosome  $2n=28$  contains the chromosomes 14 pairs or 28 bars. The karyotype formula of this species was found to be asymmetrical,  $L^m_6 + L^a_6 + M^m_2 + M^{sm}_{12} + S^{sm}_2$ , include 3 pair a large metacentric chromosome, 3 pair a large acrocentric chromosome, 1 pair a medium metacentric chromosome, 6 pair a medium submetacentric chromosome and 1 pair a small submetacentric chromosome. The relative length is a value between  $0.048 \pm 0.002$  to  $0.096 \pm 0.005$  (Table 2.9 and Figures 2.22-2.24).



**Figure 2.22** Somatic metaphase chromosome plate of *Alocasia* sp. (diploid,  $2n=28$ ).



**Figure 2.23** Karyotype of *Alocasia* sp. by conventional staining.

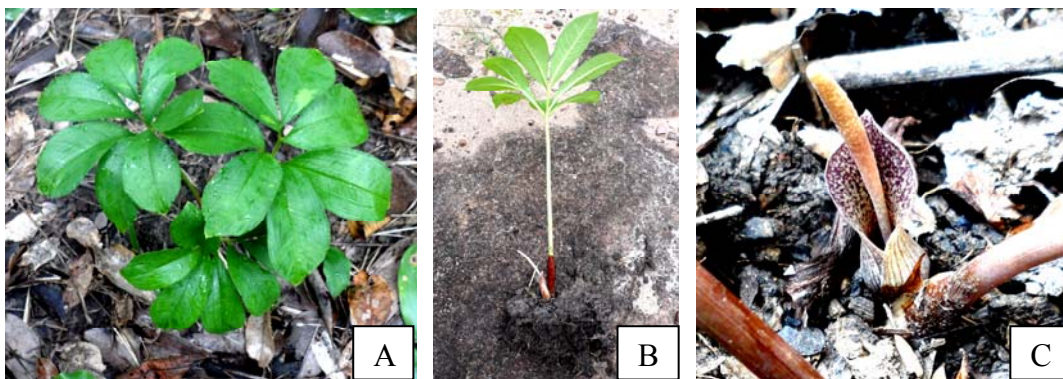


**Figure 2.24** Idiogram of *Alocasia* sp. by conventional staining.



### 2.8.7 *Amorphophallus serrulatus* Hett. & A.Galloway

Counting somatic metaphase chromosome from 20 cells of *Amorphophallus serrulatus* and then the arrangement of karyotypes and diogram by average length of the short arm (Ls) average length of the long arm (Ll) are measured by the length of each chromosome (LT). The average measurement of Relative Length (RL), Chromosome Index (CI), standard deviation (SD) of RL and CI from metaphase chromosomes in 20 cells and summarizes the size and type of chromosomes are shown in Table 2.10.



**Figure 2.25** *Amorphophallus serrulatus* Hett. & A.Galloway

A. Plants in habitat

B-C. Inflorescence

**Table 2.10** Mean length of short arm chromosome (Ls), long arm chromosome (Ll), total arm chromosome (LT), relative length (RL), centromeric index (CI) and standard deviation (SD) of RL, CI from 20 metaphases of *Amorphophallus serrulatus* (diploid,  $2n=26$ ).

Chromosome pair	Ls	Ll	LT	RL $\pm$ SD	CI $\pm$ SD	Chromosome size	Chromosome type
*1	93.842	86.614	180.456	0.112 $\pm$ 0.000	0.580 $\pm$ 0.000	Large	Metacentric
2	81.506	82.429	163.935	0.102 $\pm$ 0.000	0.597 $\pm$ 0.000	Large	Metacentric
3	65.573	85.832	151.405	0.094 $\pm$ 0.002	0.567 $\pm$ 0.000	Large	Metacentric
4	60.454	76.172	136.626	0.085 $\pm$ 0.000	0.558 $\pm$ 0.000	Large	Metacentric
5	37.608	96.046	133.654	0.083 $\pm$ 0.001	0.719 $\pm$ 0.000	Large	Acrocentric
6	57.521	69.838	127.359	0.079 $\pm$ 0.000	0.548 $\pm$ 0.000	Medium	Metacentric
7	38.470	85.552	124.022	0.077 $\pm$ 0.000	0.690 $\pm$ 0.000	Medium	Submetacentric
8	29.265	84.180	113.445	0.070 $\pm$ 0.003	0.742 $\pm$ 0.000	Medium	Acrocentric
9	31.530	76.516	108.046	0.067 $\pm$ 0.000	0.708 $\pm$ 0.000	Medium	Acrocentric
10	43.022	63.519	106.541	0.066 $\pm$ 0.001	0.596 $\pm$ 0.000	Medium	Metacentric
11	30.068	62.854	92.922	0.058 $\pm$ 0.000	0.676 $\pm$ 0.000	Medium	Submetacentric
12	29.540	63.104	92.644	0.057 $\pm$ 0.000	0.681 $\pm$ 0.000	Medium	Submetacentric
13	23.533	60.015	83.548	0.052 $\pm$ 0.001	0.718 $\pm$ 0.000	Small	Acrocentric

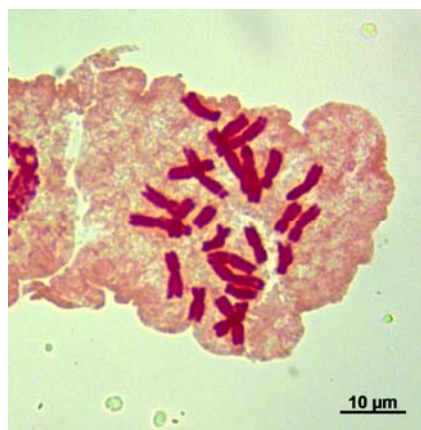
Remarks: \*=satellite chromosome (NORs).

Karyotype formula is  $L^m_8 + L^a_2 + M^m_4 + M^{sm}_6 + M^a_4 + S^a_2$

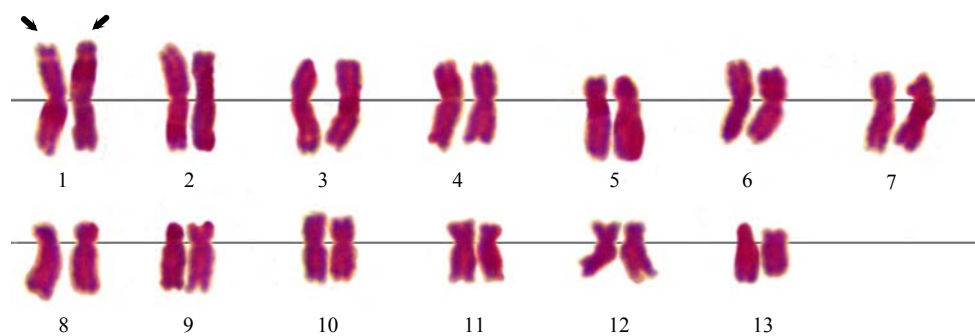


### Study on number, type and structure of the Chromosome

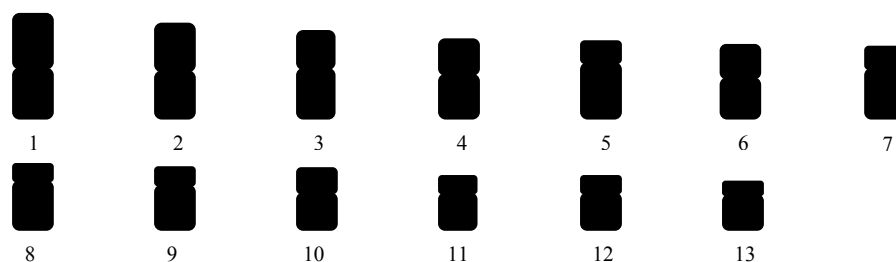
Chromosome number of *Amorphophallus serrulatus* is diploid chromosome  $2n=26$  contains the chromosomes 13 pairs or 26 bars. The karyotype formula of this species was found to be asymmetrical,  $L^m_8 + L^a_2 + M^m_4 + M^{sm}_6 + M^a_4 + S^a_2$ , include 4 pair a large metacentric chromosome, 1 pair a large acrocentric chromosome, 2 pair a medium metacentric chromosome, 3 pair a medium submetacentric chromosome, 2 pair a medium acrocentric chromosome and 1 pair a medium acrocentric chromosome. The relative length is a value between  $0.052 \pm 0.001$  to  $0.112 \pm 0.000$  and two visible satellites (Table 2.10 and Figures 2.26-2.28).



**Figure 2.26** Somatic metaphase chromosome plate of *Amorphophallus serrulatus* (diploid,  $2n=26$ ).



**Figure 2.27** Karyotype of *Amorphophallus serrulatus* (Arrows indicate satellite chromosomes) by conventional staining.



**Figure 2.28** Idiogram of *Amorphophallus serrulatus* by conventional staining.



### 2.8.8 *Arisaema maxwellii* Hett. & Gusman

Counting somatic metaphase chromosome from 20 cells of *Arisaema maxwellii* and then the arrangement of karyotypes and diagram by average length of the short arm (Ls) average length of the long arm (Ll) are measured by the length of each chromosome (LT). The average measurement of Relative Length (RL), Chromosome Index (CI), standard deviation (SD) of RL and CI from metaphase chromosomes in 20 cells and summarizes the size and type of chromosomes are shown in Table 2.11.



**Figure 2.29** *Arisaema maxwellii* Hett. & Gusman

A. Plants in habitat  
B. Inflorescence

**Table 2.11** Mean length of short arm chromosome (Ls), long arm chromosome (Ll), total arm chromosome (LT), relative length (RL), centromeric index (CI) and standard deviation (SD) of RL, CI from 20 metaphases of *Arisaema maxwellii* (diploid,  $2n=24$ ).

Chromosome pair	Ls	Ll	LT	RL $\pm$ SD	CI $\pm$ SD	Chromosome size	Chromosome type
1	45.973	50.04	96.013	0.133 $\pm$ 0.000	0.521 $\pm$ 0.000	Large	Metacentric
2	36.970	39.808	76.778	0.106 $\pm$ 0.000	0.518 $\pm$ 0.000	Large	Metacentric
3	27.378	38.090	65.468	0.091 $\pm$ 0.000	0.582 $\pm$ 0.000	Medium	Metacentric
4	33.471	31.806	65.277	0.090 $\pm$ 0.000	0.487 $\pm$ 0.000	Medium	Metacentric
5	27.376	36.841	64.217	0.089 $\pm$ 0.000	0.574 $\pm$ 0.000	Medium	Metacentric
*6	36.296	25.245	61.541	0.085 $\pm$ 0.000	0.410 $\pm$ 0.000	Medium	Metacentric
7	24.134	35.814	59.948	0.083 $\pm$ 0.000	0.597 $\pm$ 0.000	Medium	Metacentric
8	25.940	26.547	52.487	0.073 $\pm$ 0.000	0.506 $\pm$ 0.000	Medium	Metacentric
9	17.443	30.217	47.660	0.066 $\pm$ 0.000	0.634 $\pm$ 0.000	Small	Submetacentric
10	21.954	24.323	46.277	0.064 $\pm$ 0.000	0.526 $\pm$ 0.000	Small	Metacentric
11	20.943	23.857	44.800	0.062 $\pm$ 0.000	0.533 $\pm$ 0.000	Small	Metacentric
12	18.459	23.331	41.790	0.058 $\pm$ 0.000	0.558 $\pm$ 0.000	Small	Metacentric

Remarks: \*=satellite chromosome (NORs).

Karyotype formula is  $L^m_4 + M^m_{12} + S^m_6 + S^{sm}_2$



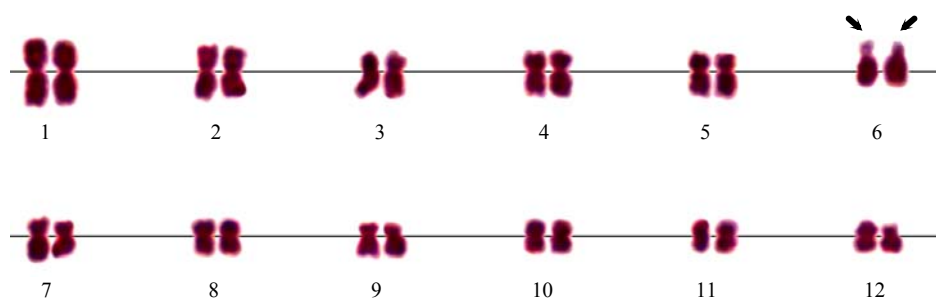


### Study on number, type and structure of the Chromosome

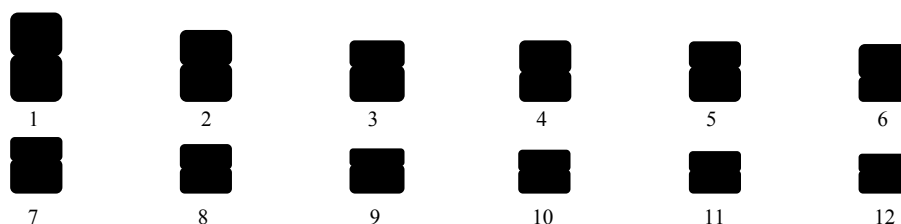
Chromosome number of *Arisaema maxwellii* is diploid chromosome  $2n=24$  contain the chromosomes 12 pairs or 24 bars. The karyotype formula of this species was found to be symmetrical,  $L^m_4 + M^m_{12} + S^m_6 + S^{sm}_2$ , include 2 pair a large metacentric chromosome, 6 pair a medium metacentric chromosome, 3 pair a small metacentric chromosome and 1 pair a small submetacentric chromosome. The relative length is a value between  $0.058 \pm 0.000$  to  $0.133 \pm 0.000$  and two visible satellites (Table 2.11 and Figures 2.30-2.32).



**Figure 2.30** Somatic metaphase chromosome plate of *Arisaema maxwellii* (diploid,  $2n=24$ ).



**Figure 2.31** Karyotype of *Arisaema maxwellii* (Arrows indicate satellite chromosomes.) by conventional staining.



**Figure 2.32** Idiogram of *Arisaema maxwellii* by conventional staining.



### 2.8.9 *Colocasia esculenta* (L.) Schott

Counting somatic metaphase chromosome from 20 cells of *Colocasia esculenta* and then the arrangement of karyotypes and diagram by average length of the short arm (Ls) average length of the long arm (Ll) are measured by the length of each chromosome (LT). The average measurement of Relative Length (RL), Chromosome Index (CI), standard deviation (SD) of RL and CI from metaphase chromosomes in 20 cells and summarizes the size and type of chromosomes are shown in Table 2.12.



**Figure 2.33** *Colocasia esculenta* (L.) Schott

A. Plants in habitat

B-C. Inflorescence

**Table 2.12** Mean length of short arm chromosome (Ls), long arm chromosome (Ll), total arm chromosome (LT), relative length (RL), centromeric index (CI) and standard deviation (SD) of RL, CI from 20 metaphases of *Colocasia esculenta* (diploid,  $2n=28$ ).

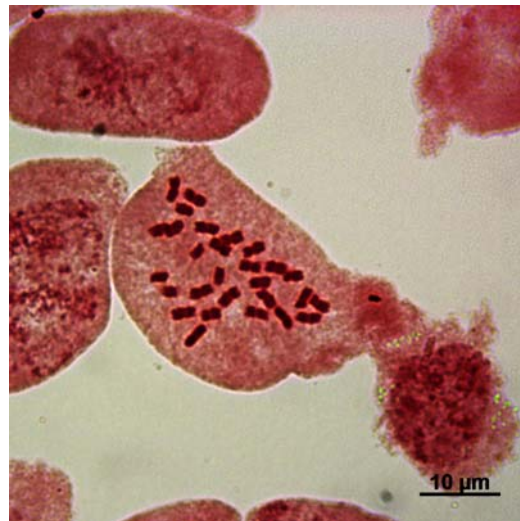
Chromosome pair	Ls	Ll	LT	RL $\pm$ SD	CI $\pm$ SD	Chromosome size	Chromosome type
1	29.552	33.177	62.729	0.087 $\pm$ 0.000	0.529 $\pm$ 0.000	Large	Metacentric
2	26.002	32.705	58.707	0.081 $\pm$ 0.000	0.557 $\pm$ 0.000	Large	Metacentric
3	23.169	33.671	56.840	0.078 $\pm$ 0.000	0.592 $\pm$ 0.000	Large	Metacentric
4	25.673	29.537	55.210	0.076 $\pm$ 0.000	0.535 $\pm$ 0.000	Large	Metacentric
5	25.508	29.535	55.043	0.076 $\pm$ 0.000	0.537 $\pm$ 0.000	Large	Metacentric
6	22.685	31.106	53.791	0.074 $\pm$ 0.000	0.578 $\pm$ 0.000	Large	Metacentric
7	22.166	31.101	53.267	0.073 $\pm$ 0.000	0.584 $\pm$ 0.000	Large	Metacentric
8	23.393	29.529	52.922	0.073 $\pm$ 0.000	0.558 $\pm$ 0.000	Large	Metacentric
9	22.404	27.994	50.398	0.070 $\pm$ 0.000	0.555 $\pm$ 0.000	Medium	Metacentric
10	20.395	29.528	49.923	0.069 $\pm$ 0.000	0.591 $\pm$ 0.000	Medium	Metacentric
11	19.220	29.532	48.752	0.067 $\pm$ 0.000	0.606 $\pm$ 0.000	Medium	Submetacentric
12	16.224	29.536	45.760	0.063 $\pm$ 0.000	0.645 $\pm$ 0.000	Medium	Submetacentric
13	11.728	31.333	43.061	0.059 $\pm$ 0.000	0.728 $\pm$ 0.000	Medium	Acrocentric
14	12.069	26.288	38.357	0.053 $\pm$ 0.000	0.685 $\pm$ 0.000	Medium	Submetacentric

Karyotype formula is  $L_{16}^m + M_4^m + M_6^{sm} + M_2^a$

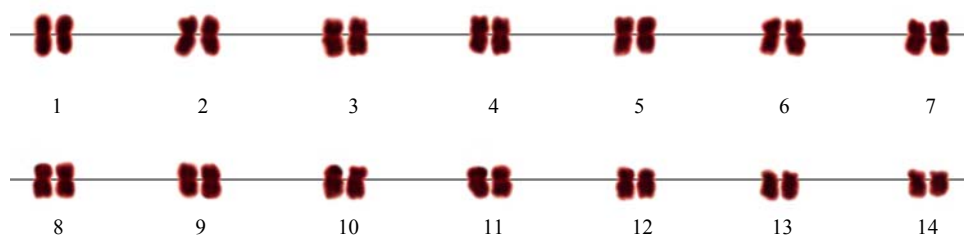


### Study on number, type and structure of the Chromosome

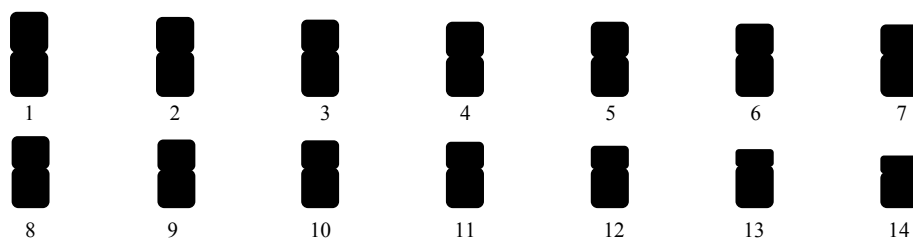
Chromosome number of *Colocasia esculenta* is diploid chromosome  $2n=28$  contains the chromosomes 14 pairs or 28 bars. The karyotype formula of this species was found to be asymmetrical,  $L_{16}^m + M_4^m + M_6^{sm} + M_2^a$ , include 8 pair a large metacentric chromosome, 2 pair a medium metacentric chromosome, 3 pair a medium submetacentric chromosome and 1 pair a medium acrocentric chromosome. The relative length is a value between  $0.053 \pm 0.000$  to  $0.087 \pm 0.000$  (Table 2.12 and Figures 2.34-2.36).



**Figure 2.34** Somatic metaphase chromosome plate of *Colocasia esculenta* (diploid,  $2n=28$ ).



**Figure 2.35** Karyotype of *Colocasia esculenta* by conventional staining.



**Figure 2.36** Idiogram of *Colocasia esculenta* by conventional staining.



### 2.8.10 *Colocasia fallax* Schott

Counting somatic metaphase chromosome from 20 cells of *Colocasia fallax* and then the arrangement of karyotypes and diagram by average length of the short arm (Ls) average length of the long arm (Ll) are measured by the length of each chromosome (LT). The average measurement of Relative Length (RL), Chromosome Index (CI), standard deviation (SD) of RL and CI from metaphase chromosomes in 20 cells and summarizes the size and type of chromosomes are shown in Table 2.13.



**Figure 2.37** *Colocasia fallax* Schott

A. Plants in habitat

B. Inflorescence

**Table 2.13** Mean length of short arm chromosome (Ls), long arm chromosome (Ll), total arm chromosome (LT), relative length (RL), centromeric index (CI) and standard deviation (SD) of RL, CI from 20 metaphases of *Colocasia fallax* (diploid,  $2n=28$ ).

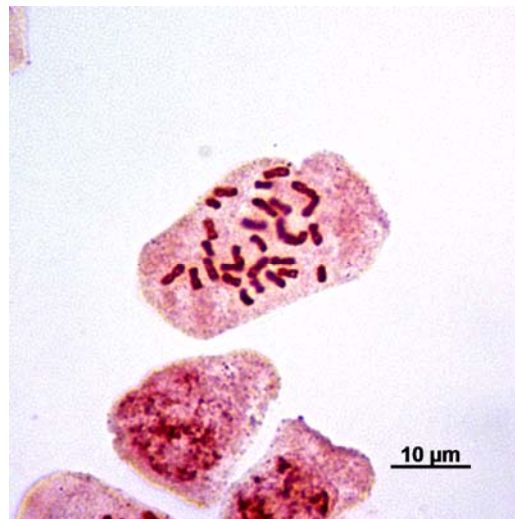
Chromosome pair	Ls	Ll	LT	RL $\pm$ SD	CI $\pm$ SD	Chromosome size	Chromosome type
1	35.362	36.149	71.511	0.098 $\pm$ 0.000	0.506 $\pm$ 0.000	Large	Metacentric
2	27.338	37.162	64.500	0.088 $\pm$ 0.000	0.576 $\pm$ 0.000	Large	Metacentric
3	27.319	32.218	59.537	0.081 $\pm$ 0.000	0.541 $\pm$ 0.000	Large	Metacentric
4	27.331	30.005	57.336	0.078 $\pm$ 0.000	0.523 $\pm$ 0.000	Large	Metacentric
5	26.426	29.537	55.963	0.076 $\pm$ 0.000	0.528 $\pm$ 0.000	Large	Metacentric
6	25.522	29.544	55.066	0.075 $\pm$ 0.000	0.537 $\pm$ 0.000	Large	Metacentric
7	24.615	29.538	54.153	0.074 $\pm$ 0.000	0.545 $\pm$ 0.000	Medium	Metacentric
8	23.830	29.530	53.360	0.073 $\pm$ 0.000	0.553 $\pm$ 0.000	Medium	Metacentric
9	23.093	29.536	52.629	0.072 $\pm$ 0.000	0.561 $\pm$ 0.000	Medium	Metacentric
10	21.845	24.675	46.520	0.063 $\pm$ 0.000	0.530 $\pm$ 0.000	Medium	Metacentric
11	20.833	24.423	45.256	0.062 $\pm$ 0.000	0.540 $\pm$ 0.000	Medium	Metacentric
12	17.084	24.194	41.278	0.056 $\pm$ 0.000	0.586 $\pm$ 0.000	Medium	Metacentric
13	14.584	24.420	39.004	0.053 $\pm$ 0.000	0.626 $\pm$ 0.000	Medium	Submetacentric
14	13.330	23.925	37.255	0.051 $\pm$ 0.000	0.642 $\pm$ 0.000	Medium	Submetacentric

Karyotype formula is  $L_{12}^m + M_{12}^m + M_4^{sm}$

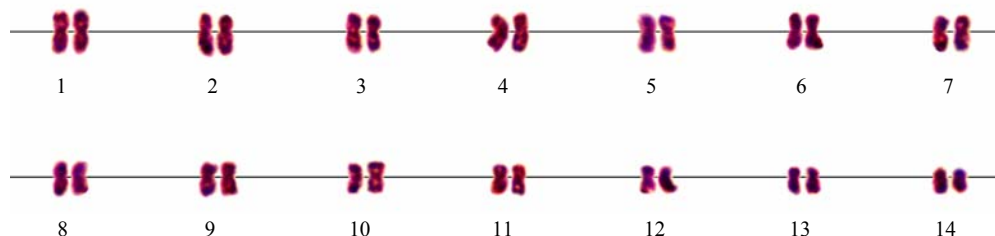


### Study on number, type and structure of the Chromosome

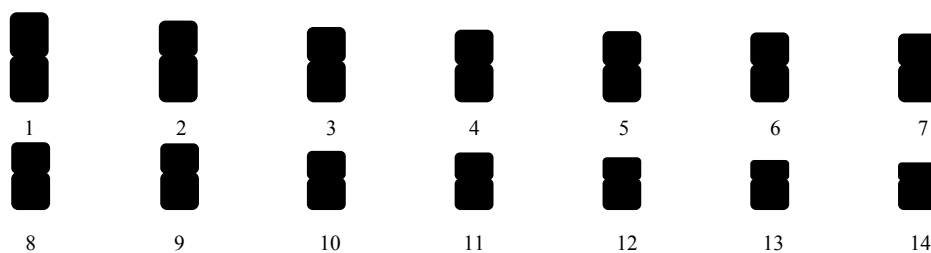
Chromosome number of *Colocasia fallax* is diploid chromosome  $2n=28$  contains the chromosomes 14 pairs or 28 bars. The karyotype formula of this species was found to be symmetrical,  $L_{12}^m + M_{12}^m + M_4^{sm}$ , include 6 pair a large metacentric chromosome, 6 pair a medium metacentric chromosome and 2 pair a medium submetacentric chromosome. The relative length is a value between  $0.051 \pm 0.000$  to  $0.098 \pm 0.000$  (Table 2.13 and Figures 2.38-2.40).



**Figure 2.38** Somatic metaphase chromosome plate of *Colocasia fallax* (diploid,  $2n=28$ ).



**Figure 2.39** Karyotype of *Colocasia fallax* by conventional staining.

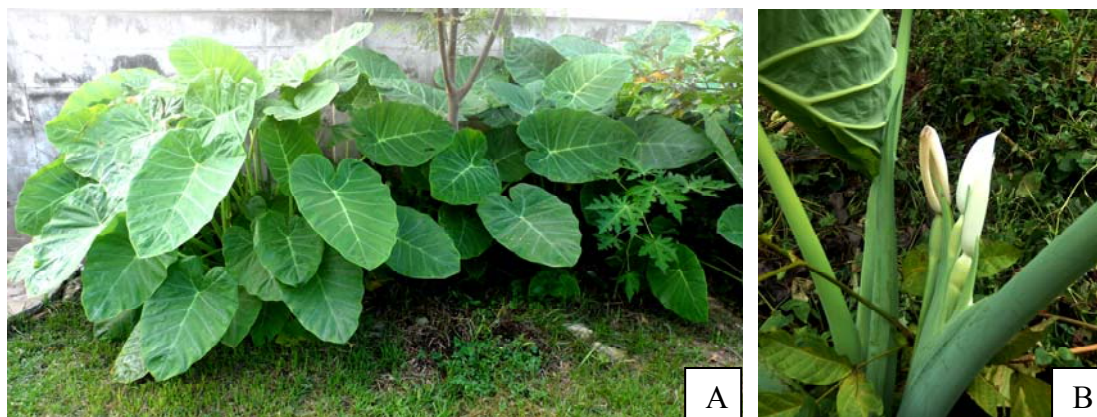


**Figure 2.40** Idiogram of *Colocasia fallax* by conventional staining.



### 2.8.11 *Colocasia gigantea* (Blume) Hook.f.

Counting somatic metaphase chromosome from 20 cells of *Colocasia gigantea* and then the arrangement of karyotypes and diagram by average length of the short arm (Ls) average length of the long arm (Ll) are measured by the length of each chromosome (LT). The average measurement of Relative Length (RL), Chromosome Index (CI), standard deviation (SD) of RL and CI from metaphase chromosomes in 20 cells and summarizes the size and type of chromosomes are shown in Table 2.14.



**Figure 2.41** *Colocasia gigantea* (Blume) Hook.f.

A. Plants in habitat  
B. Inflorescence

**Table 2.14** Mean length of short arm chromosome (Ls), long arm chromosome (Ll), total arm chromosome (LT), relative length (RL), centromeric index (CI) and standard deviation (SD) of RL, CI from 20 metaphases of *Colocasia gigantea* (diploid,  $2n=28$ ).

Chromosome pair	Ls	Ll	LT	RL $\pm$ SD	CI $\pm$ SD	Chromosome size	Chromosome type
1	34.572	38.907	73.479	0.086 $\pm$ 0.000	0.530 $\pm$ 0.000	Large	Metacentric
2	27.272	43.280	70.552	0.083 $\pm$ 0.000	0.613 $\pm$ 0.000	Large	Submetacentric
3	31.381	38.659	70.040	0.082 $\pm$ 0.002	0.552 $\pm$ 0.000	Large	Metacentric
4	33.404	34.288	67.692	0.079 $\pm$ 0.000	0.507 $\pm$ 0.000	Large	Metacentric
5	29.542	35.531	65.073	0.076 $\pm$ 0.001	0.546 $\pm$ 0.000	Large	Metacentric
6	31.633	32.907	64.540	0.075 $\pm$ 0.000	0.510 $\pm$ 0.000	Large	Metacentric
7	28.931	35.160	64.091	0.075 $\pm$ 0.001	0.549 $\pm$ 0.000	Large	Metacentric
8	26.065	35.560	61.625	0.072 $\pm$ 0.000	0.577 $\pm$ 0.000	Large	Metacentric
9	28.236	32.685	60.921	0.071 $\pm$ 0.000	0.537 $\pm$ 0.000	Large	Metacentric
*10	36.073	22.061	58.134	0.068 $\pm$ 0.001	0.579 $\pm$ 0.000	Large	Metacentric
11	22.575	32.686	56.260	0.066 $\pm$ 0.000	0.609 $\pm$ 0.000	Medium	Submetacentric
12	25.071	29.536	54.607	0.064 $\pm$ 0.001	0.541 $\pm$ 0.000	Medium	Metacentric
13	13.567	35.06	48.627	0.057 $\pm$ 0.000	0.721 $\pm$ 0.000	Medium	Acrocentric
14	14.315	26.686	41.001	0.048 $\pm$ 0.000	0.651 $\pm$ 0.000	Medium	Submetacentric

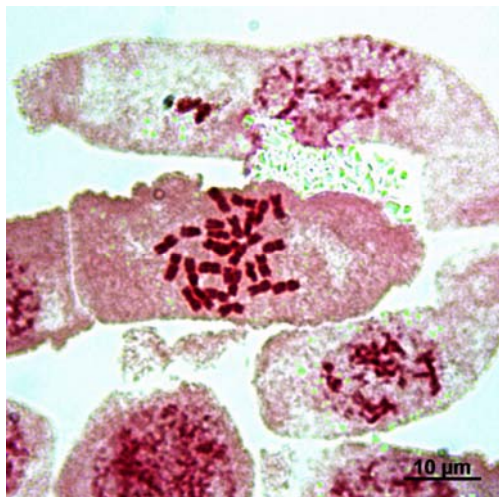
Remarks: \*=satellite chromosome (NORs).

Karyotype formula is  $L^{m}_{18} + L^{sm}_{2} + M^{m}_{2} + M^{sm}_{4} + M^{a}_{2}$

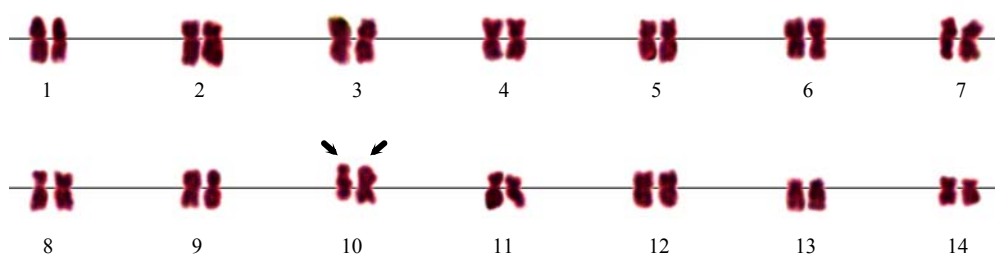


### Study on number, type and structure of the Chromosome

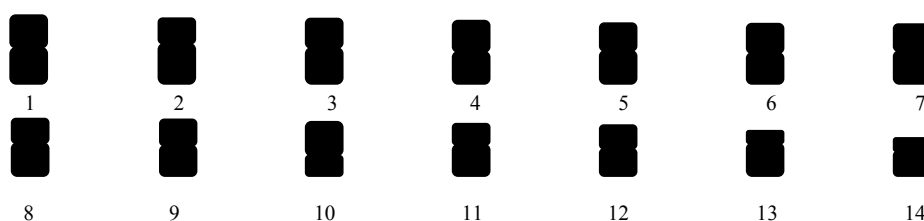
Chromosome number of *Colocasia gigantea* is diploid chromosome  $2n=28$  contains the chromosomes 14 pairs or 28 bars. The karyotype formula of this species was found to be asymmetrical,  $L^m_{18} + L^{sm}_2 + M^m_2 + M^{sm}_4 + M^a_2$ , include 9 pair a large metacentric chromosome, 1 pair a large submetacentric chromosome, 1 pair a medium metacentric chromosome, 2 pair a medium submetacentric chromosome and 1 pair a medium acrocentric chromosome. The relative length is a value between  $0.048 \pm 0.000$  to  $0.861 \pm 0.000$  and two visible satellites (Table 4.14 and Figures 2.42-2.44).



**Figure 2.42** Somatic metaphase chromosome plate of *Colocasia gigantea* (diploid,  $2n=28$ ).



**Figure 2.43** Karyotype of *Colocasia gigantea* (Arrows indicate satellite chromosomes) by conventional staining.



**Figure 2.44** Idiogram of *Colocasia gigantea* by conventional staining.





### 2.8.12 *Colocasia lihengiae* C.L.Long & K.M.Liu

Counting somatic metaphase chromosome from 20 cells of *Colocasia lihengiae* and then the arrangement of karyotypes and diogram by average length of the short arm (Ls) average length of the long arm (Ll) are measured by the length of each chromosome (LT). The average measurement of Relative Length (RL), Chromosome Index (CI), standard deviation (SD) of RL and CI from metaphase chromosomes in 20 cells and summarizes the size and type of chromosomes are shown in Table 2.15.



**Figure 2.45** *Colocasia lihengiae* C.L.Long & K.M.Liu

**Table 2.15** Mean length of short arm chromosome (Ls), long arm chromosome (Ll), total arm chromosome (LT), relative length (RL), centromeric index (CI) and standard deviation (SD) of RL, CI from 20 metaphases of *Colocasia lihengiae* (diploid,  $2n=28$ ).

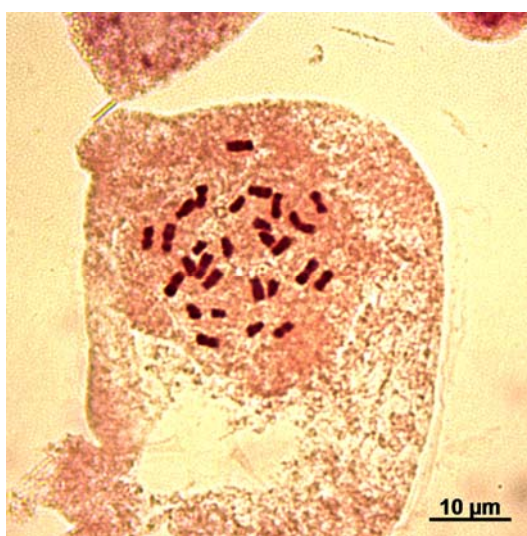
Chromosome pair	Ls	Ll	LT	RL $\pm$ SD	CI $\pm$ SD	Chromosome size	Chromosome type
1	34.997	36.18	71.177	0.097 $\pm$ 0.000	0.508 $\pm$ 0.000	Large	Metacentric
2	29.604	34.341	63.945	0.087 $\pm$ 0.000	0.537 $\pm$ 0.000	Large	Metacentric
3	21.010	42.045	63.055	0.086 $\pm$ 0.000	0.667 $\pm$ 0.000	Large	Submetacentric
4	27.002	32.010	59.012	0.080 $\pm$ 0.000	0.542 $\pm$ 0.000	Large	Metacentric
5	25.832	29.540	55.372	0.075 $\pm$ 0.000	0.733 $\pm$ 0.000	Large	Acrocentric
6	17.500	37.006	54.506	0.074 $\pm$ 0.000	0.779 $\pm$ 0.000	Large	Acrocentric
7	24.898	29.532	54.430	0.074 $\pm$ 0.000	0.543 $\pm$ 0.000	Large	Metacentric
8	23.830	29.864	53.694	0.073 $\pm$ 0.000	0.556 $\pm$ 0.000	Large	Metacentric
9	17.218	35.303	52.521	0.071 $\pm$ 0.000	0.772 $\pm$ 0.000	Medium	Acrocentric
10	16.057	30.693	46.750	0.063 $\pm$ 0.000	0.757 $\pm$ 0.000	Medium	Acrocentric
11	19.052	24.959	44.011	0.060 $\pm$ 0.000	0.567 $\pm$ 0.000	Medium	Metacentric
12	19.385	23.621	43.006	0.058 $\pm$ 0.000	0.549 $\pm$ 0.000	Medium	Metacentric
13	17.553	22.952	40.505	0.055 $\pm$ 0.000	0.567 $\pm$ 0.000	Medium	Metacentric
14	15.219	20.128	35.347	0.048 $\pm$ 0.000	0.569 $\pm$ 0.000	Small	Metacentric

Karyotype formula is  $L_{10}^m + L_4^{sm} + L_2^a + M_6^m + M_4^a + S_2^m$

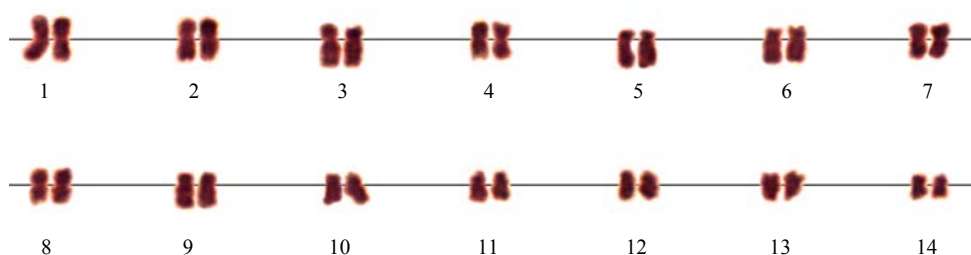


### Study on number, type and structure of the Chromosome

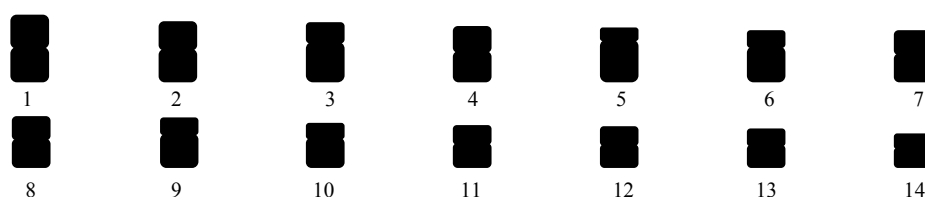
Chromosome number of *Colocasia lihengiae* is diploid chromosome  $2n=28$  contains the chromosomes 14 pairs or 28 bars. The karyotype formula of this species was found to be asymmetrical,  $L^m_{10} + L^{sm}_4 + L^a_2 + M^m_6 + M^a_4 + S^m_2$ , include 5 pair a large metacentric chromosome, 2 pair a large submetacentric chromosome, 1 pair a large acrocentric chromosome, 3 pair a medium metacentric chromosome, 2 pair a medium acrocentric chromosome and 1 pair a small metacentric chromosome. The relative length is a value between  $0.048 \pm 0.000$  to  $0.097 \pm 0.000$  (Table 2.15 and Figures 2.46-2.48).



**Figure 2.46** Somatic metaphase chromosome plate of *Colocasia lihengiae* (diploid,  $2n=28$ ).



**Figure 2.47** Karyotype of *Colocasia lihengiae* by conventional staining.



**Figure 2.48** Idiogram of *Colocasia lihengiae* by conventional staining.

### 2.8.13 *Hapaline benthamiana* Schott

Counting somatic metaphase chromosome from 20 cells of *Hapaline benthamiana* and then the arrangement of karyotypes and diogram by average length of the short arm (Ls) average length of the long arm (Ll) are measured by the length of each chromosome (LT). The average measurement of Relative Length (RL), Chromosome Index (CI), standard deviation (SD) of RL and CI from metaphase chromosomes in 20 cells and summarizes the size and type of chromosomes are shown in Table 2.16.



**Figure 2.49** *Hapaline benthamiana* Schott

**Table 2.16** Mean length of short arm chromosome (Ls), long arm chromosome (Ll), total arm chromosome (LT), relative length (RL), centromeric index (CI) and standard deviation (SD) of RL, CI from 20 metaphases of *Hapaline benthamiana* (diploid,  $2n=26$ ).

Chromosome pair	Ls	Ll	LT	RL $\pm$ SD	CI $\pm$ SD	Chromosome size	Chromosome type
1	37.486	39.280	76.766	0.104 $\pm$ 0.000	0.512 $\pm$ 0.000	Large	Metacentric
2	34.229	37.828	72.058	0.098 $\pm$ 0.000	0.525 $\pm$ 0.000	Large	Metacentric
3	31.884	37.047	68.931	0.094 $\pm$ 0.000	0.537 $\pm$ 0.000	Large	Metacentric
4	31.876	29.580	61.456	0.084 $\pm$ 0.000	0.481 $\pm$ 0.000	Large	Metacentric
5	29.202	27.044	56.246	0.077 $\pm$ 0.000	0.481 $\pm$ 0.000	Medium	Metacentric
6	25.190	29.532	54.722	0.074 $\pm$ 0.000	0.540 $\pm$ 0.000	Medium	Metacentric
7	24.983	29.536	54.519	0.074 $\pm$ 0.000	0.542 $\pm$ 0.000	Medium	Metacentric
8	24.473	29.528	54.000	0.073 $\pm$ 0.000	0.547 $\pm$ 0.000	Medium	Metacentric
*9	24.223	29.549	53.771	0.073 $\pm$ 0.000	0.550 $\pm$ 0.000	Medium	Metacentric
10	23.945	25.062	49.008	0.067 $\pm$ 0.000	0.511 $\pm$ 0.000	Medium	Metacentric
11	21.454	24.812	46.267	0.063 $\pm$ 0.000	0.536 $\pm$ 0.000	Medium	Metacentric
12	20.196	24.313	44.509	0.061 $\pm$ 0.000	0.546 $\pm$ 0.000	Medium	Metacentric
13	19.443	23.315	42.758	0.058 $\pm$ 0.000	0.545 $\pm$ 0.000	Medium	Metacentric

Remarks: \*=satellite chromosome (NORs).

Karyotype formula is  $L^m_8 + M^m_{18}$



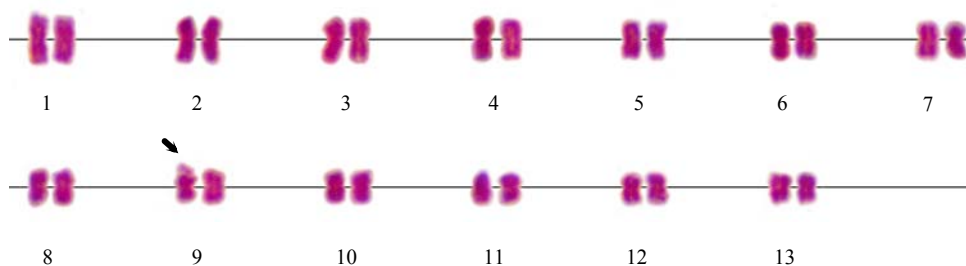


### Study on number, type and structure of the Chromosome

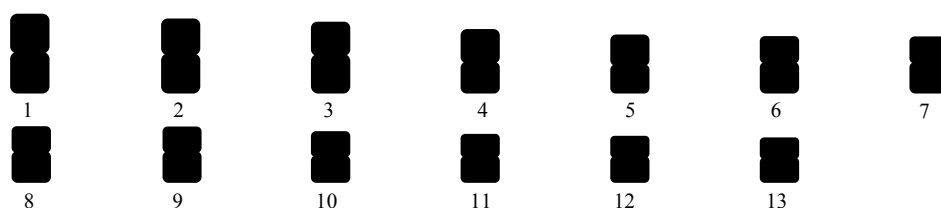
Chromosome number of *Hapaline benthamiana* is diploid chromosome  $2n=26$  contains the chromosomes 13 pairs or 26 bars. The karyotype formula of this species was found to be symmetrical,  $L^m_8 + M^m_{18}$ , include 4 pair a large metacentric chromosome and 9 pair a medium metacentric chromosome. The relative length is a value between  $0.058 \pm 0.000$  to  $0.104 \pm 0.000$  and one visible satellite (Table 2.16 and Figures 2.50-2.52).



**Figure 2.50** Somatic metaphase chromosome plate of *Hapaline benthamiana* (diploid,  $2n=26$ ).



**Figure 2.51** Karyotype of *Hapaline benthamiana* (Arrows indicate satellite chromosomes) by conventional staining.



**Figure 2.52** Idiogram of *Hapaline benthamiana* by conventional staining.



### 2.8.14 *Homalomena griffithii* (Schott) Hook.f.

Counting somatic metaphase chromosome from 20 cells of *Homalomena griffithii* and then the arrangement of karyotypes and diagram by average length of the short arm (Ls) average length of the long arm (Ll) are measured by the length of each chromosome (LT). The average measurement of Relative Length (RL), Chromosome Index (CI), standard deviation (SD) of RL and CI from metaphase chromosomes in 20 cells and summarizes the size and type of chromosomes are shown in Table 2.17.



**Figure 2.53** *Homalomena griffithii* (Schott) Hook.f.

**Table 2.17** Mean length of short arm chromosome (Ls), long arm chromosome (Ll), total arm chromosome (LT), relative length (RL), centromeric index (CI) and standard deviation (SD) of RL, CI from 20 metaphases of *Homalomena griffithii* (diploid, 2n=40).

Chromosome pair	Ls	Ll	LT	RL±SD	CI±SD	Chromosome size	Chromosome type
1	27.859	33.813	61.672	0.080±0.002	0.548±0.000	Large	Metacentric
2	22.928	31.245	54.174	0.070±0.000	0.577±0.000	Large	Metacentric
*3	36.488	16.171	52.659	0.068±0.000	0.507±0.000	Large	Metacentric
4	17.474	29.746	47.220	0.061±0.000	0.630±0.000	Large	Submetacentric
5	16.974	27.959	44.933	0.058±0.001	0.622±0.000	Large	Submetacentric
6	17.278	27.439	44.717	0.058±0.000	0.614±0.000	Large	Submetacentric
7	17.477	24.786	42.264	0.055±0.000	0.586±0.000	Large	Metacentric
8	14.963	24.353	39.315	0.051±0.003	0.619±0.000	Large	Submetacentric
9	15.719	23.313	39.032	0.051±0.001	0.597±0.000	Large	Metacentric
10	15.958	22.784	38.742	0.050±0.000	0.588±0.000	Large	Metacentric
11	16.738	20.025	36.763	0.048±0.002	0.545±0.000	Medium	Metacentric
12	15.945	20.313	36.258	0.047±0.000	0.560±0.000	Medium	Metacentric
13	11.469	24.303	35.772	0.047±0.000	0.679±0.000	Medium	Submetacentric
14	16.544	18.854	35.398	0.046±0.003	0.533±0.000	Medium	Metacentric
15	16.414	14.844	31.258	0.041±0.000	0.575±0.000	Medium	Metacentric
16	14.432	16.348	30.780	0.040±0.000	0.531±0.000	Medium	Metacentric
17	13.432	16.121	29.553	0.038±0.000	0.545±0.000	Medium	Metacentric
18	12.557	15.405	27.962	0.036±0.000	0.551±0.000	Medium	Metacentric
19	13.415	12.590	26.005	0.034±0.001	0.584±0.000	Medium	Metacentric
20	5.922	8.596	14.518	0.019±0.000	0.593±0.000	Medium	Metacentric

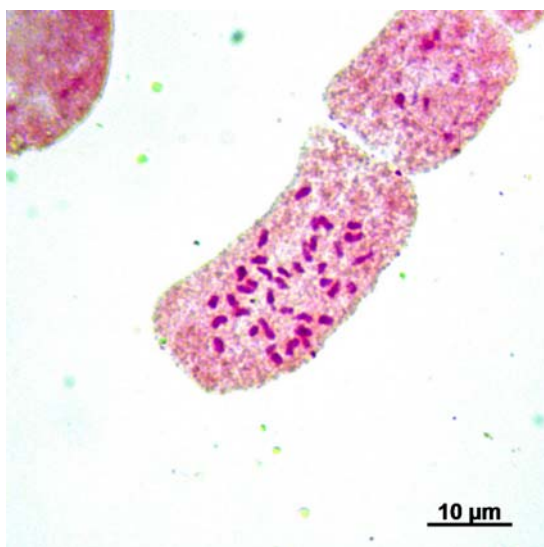
Remarks: \*=satellite chromosome (NORs).



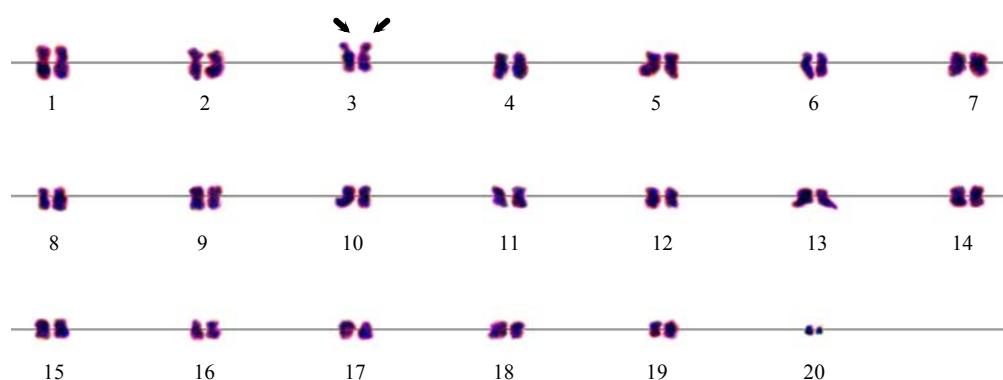
Karyotype formula is  $L_{12}^m + L_8^{sm} + M_{18}^m + M_2^{sm}$

Study on number, type and structure of the Chromosome

Chromosome number of *Homalomena griffithii* is diploid chromosome  $2n=40$  contain the chromosomes 20 pairs or 40 bars. The karyotype formula of this species was found to be symmetrical,  $L_{12}^m + L_8^{sm} + M_{18}^m + M_2^{sm}$ , include 6 pair a large metacentric chromosome, 4 pair a large submetacentric chromosome, 9 pair a medium metacentric chromosome and 1 pair a medium submetacentric chromosome. The relative length is a value between  $0.019 \pm 0.000$  to  $0.080 \pm 0.002$  and two visible satellites (Table 2.17 and Figures 2.54-2.56).

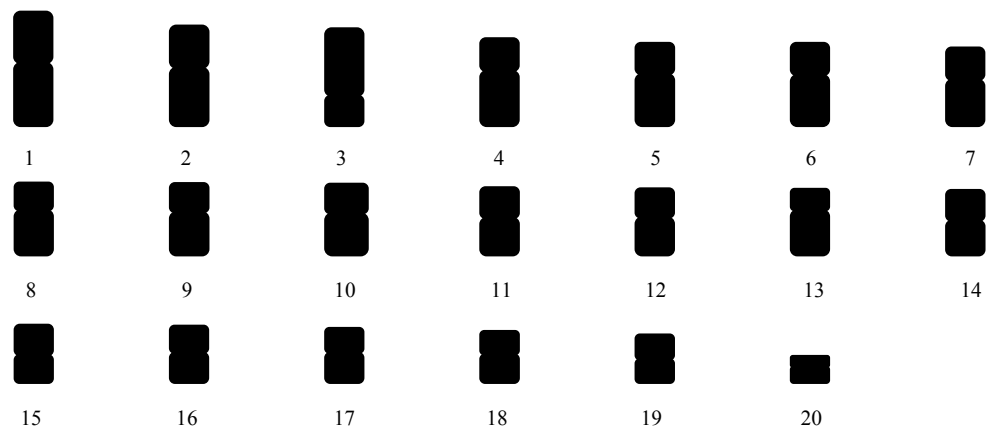


**Figure 2.54** Somatic metaphase chromosome plate of *Homalomena griffithii* (diploid,  $2n=40$ ).



**Figure 2.55** Karyotype of *Homalomena griffithii* (Arrows indicate satellite chromosomes) by conventional staining.





**Figure 2.56** Idiogram of *Homalomena griffithii* by conventional staining.



### 2.8.15 *Lasia spinosa* (L.) Thwaites

Counting somatic metaphase chromosome from 20 cells of *Lasia spinosa* and then the arrangement of karyotypes and diagram by average length of the short arm (Ls) average length of the long arm (Ll) are measured by the length of each chromosome (LT). The average measurement of Relative Length (RL), Chromosome Index (CI), standard deviation (SD) of RL and CI from metaphase chromosomes in 20 cells and summarizes the size and type of chromosomes are shown in Table 2.18.



**Figure 2.57** *Lasia spinosa* (L.) Thwaites  
 A. Plants in habitat  
 B. leaves, inflorescence and fruits  
 C. Fruits and seed

**Table 2.18** Mean length of short arm chromosome (Ls), long arm chromosome (Ll), total arm chromosome (LT), relative length (RL), centromeric index (CI) and standard deviation (SD) of RL, CI from 20 metaphases of *Lasia spinosa* (diploid,  $2n=26$ ).

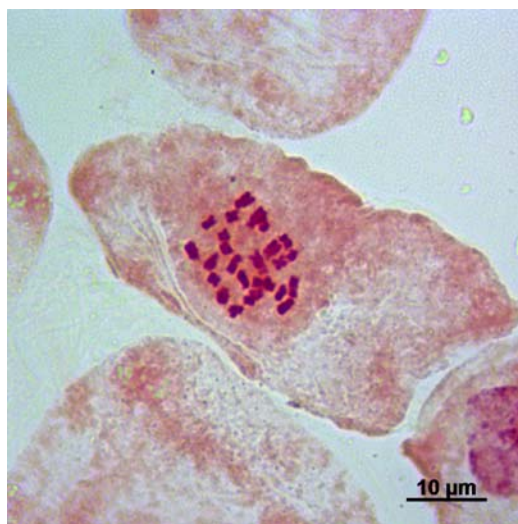
Chromosome pair	Ls	Ll	LT	RL $\pm$ SD	CI $\pm$ SD	Chromosome size	Chromosome type
1	17.587	43.923	61.510	0.124 $\pm$ 0.000	0.714 $\pm$ 0.000	Large	Acrocentric
2	18.115	31.853	49.968	0.101 $\pm$ 0.000	0.637 $\pm$ 0.000	Large	Submetacentric
3	16.849	29.555	46.404	0.094 $\pm$ 0.000	0.637 $\pm$ 0.000	Large	Submetacentric
4	15.088	29.555	44.643	0.090 $\pm$ 0.000	0.662 $\pm$ 0.000	Large	Submetacentric
5	17.345	23.675	41.021	0.083 $\pm$ 0.000	0.577 $\pm$ 0.000	Medium	Metacentric
6	13.830	24.673	38.503	0.078 $\pm$ 0.000	0.641 $\pm$ 0.000	Medium	Submetacentric
7	17.054	19.207	36.260	0.073 $\pm$ 0.000	0.530 $\pm$ 0.000	Medium	Metacentric
8	14.050	19.203	33.253	0.067 $\pm$ 0.000	0.577 $\pm$ 0.000	Medium	Metacentric
9	15.428	17.080	32.508	0.066 $\pm$ 0.000	0.575 $\pm$ 0.000	Medium	Metacentric
10	13.305	16.681	29.986	0.061 $\pm$ 0.000	0.556 $\pm$ 0.000	Medium	Metacentric
11	12.574	15.704	28.279	0.057 $\pm$ 0.000	0.555 $\pm$ 0.000	Medium	Metacentric
12	9.050	18.454	27.504	0.056 $\pm$ 0.000	0.671 $\pm$ 0.000	Medium	Submetacentric
13	11.074	13.238	24.312	0.049 $\pm$ 0.000	0.545 $\pm$ 0.000	Medium	Metacentric

Karyotype formula is  $L^{sm}_6 + L^a_2 + M^{sm}_{14} + M^{sm}_4$

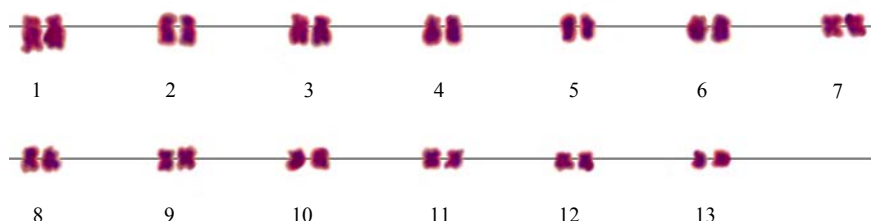


### Study on number, type and structure of the Chromosome

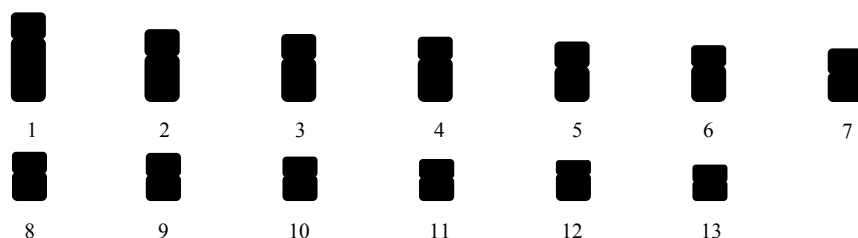
Chromosome number of *Lasia spinosa* is diploid chromosome  $2n=26$  contains the chromosomes 13 pairs or 26 bars. The karyotype formula of this species was found to be asymmetrical,  $L^{\text{sm}}_6 + L^{\text{a}}_2 + M^{\text{m}}_{14} + M^{\text{sm}}_4$ , include 3 pair a large submetacentric chromosome, 1 pair a large acrocentric chromosome, 7 pair a medium metacentric chromosome and 2 pair a medium submetacentric chromosome. The relative length is a value between  $0.049 \pm 0.000$  to  $0.124 \pm 0.000$  (Table 2.18 and Figures 2.58-2.60).



**Figure 2.58** Somatic metaphase chromosome plate of *Lasia spinosa* (diploid,  $2n=26$ ).



**Figure 2.59** Karyotype of *Lasia spinosa* by conventional staining.



**Figure 2.60** Idiogram of *Lasia spinosa* by conventional staining.



### 2.8.16 *Pistia stratiotes* L.

Counting somatic metaphase chromosome from 20 cells of *Pistia stratiotes* and then the arrangement of karyotypes and diagram by average length of the short arm (Ls) average length of the long arm (Ll) are measured by the length of each chromosome (LT). The average measurement of Relative Length (RL), Chromosome Index (CI), standard deviation (SD) of RL and CI from metaphase chromosomes in 20 cells and summarizes the size and type of chromosomes are shown in Table 2.19.



**Figure 2.61** *Pistia stratiotes* L.

A. Plants in habitat  
B. Inflorescence

**Table 2.19** Mean length of short arm chromosome (Ls), long arm chromosome (Ll), total arm chromosome (LT), relative length (RL), centromeric index (CI) and standard deviation (SD) of RL, CI from 20 metaphases of *Pistia stratiotes* (diploid,  $2n=28$ ).

Chromosome pair	Ls	Ll	LT	RL $\pm$ SD	CI $\pm$ SD	Chromosome size	Chromosome type
1	9.514	10.305	19.819	0.094 $\pm$ 0.000	0.520 $\pm$ 0.000	Large	Metacentric
2	8.279	9.286	17.565	0.083 $\pm$ 0.000	0.529 $\pm$ 0.000	Large	Metacentric
3	7.945	8.644	16.589	0.079 $\pm$ 0.000	0.521 $\pm$ 0.000	Large	Metacentric
4	7.508	8.467	15.975	0.076 $\pm$ 0.000	0.530 $\pm$ 0.000	Large	Metacentric
5	6.959	8.902	15.861	0.075 $\pm$ 0.000	0.561 $\pm$ 0.000	Large	Metacentric
6	7.733	7.784	15.517	0.074 $\pm$ 0.000	0.502 $\pm$ 0.000	Medium	Metacentric
7	7.278	8.120	15.398	0.073 $\pm$ 0.000	0.527 $\pm$ 0.000	Medium	Metacentric
8	7.108	7.954	15.062	0.071 $\pm$ 0.000	0.528 $\pm$ 0.000	Medium	Metacentric
9	5.958	8.576	14.535	0.069 $\pm$ 0.000	0.590 $\pm$ 0.000	Medium	Metacentric
10	6.473	7.900	14.374	0.068 $\pm$ 0.000	0.550 $\pm$ 0.000	Medium	Metacentric
11	6.950	7.237	14.187	0.067 $\pm$ 0.000	0.510 $\pm$ 0.000	Medium	Metacentric
12	6.307	6.842	13.148	0.062 $\pm$ 0.000	0.520 $\pm$ 0.000	Medium	Metacentric
13	4.957	6.730	11.687	0.055 $\pm$ 0.000	0.576 $\pm$ 0.000	Medium	Metacentric
14	4.442	6.704	11.147	0.053 $\pm$ 0.000	0.596 $\pm$ 0.000	Medium	Metacentric

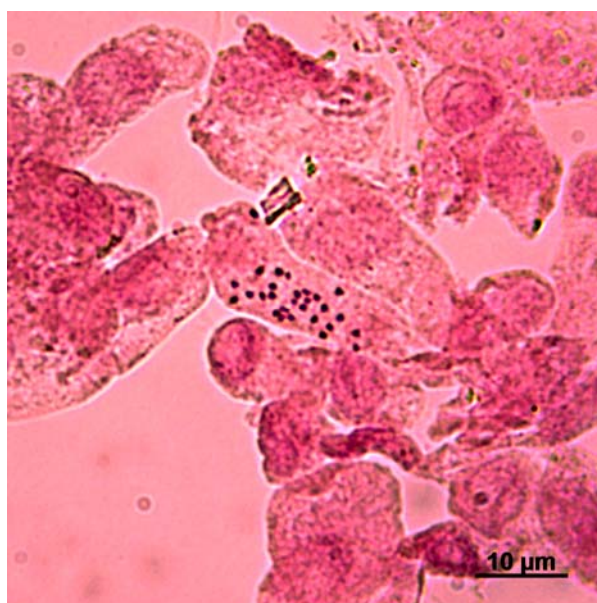
Karyotype formula is  $L^m_{10} + M^m_{18}$



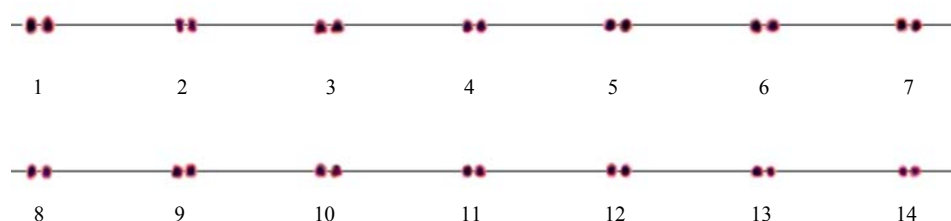


### Study on number, type and structure of the Chromosome

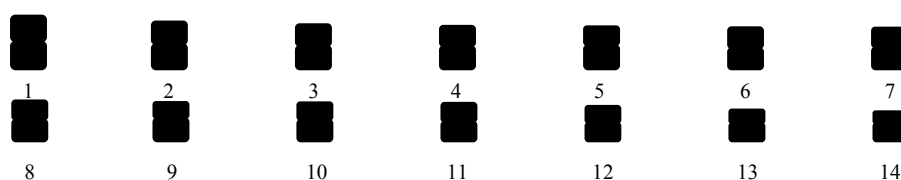
Chromosome number of *Pistia stratiotes* is diploid chromosome  $2n=28$  contains the chromosomes 14 pairs or 28 bars. The karyotype formula of this species was found to be symmetrical,  $L_{10}^m + M_{18}^m$ , include 5 pair a large metacentric chromosome and 9 pair a medium metacentric chromosome. The relative length is a value between  $0.053 \pm 0.000$  to  $0.094 \pm 0.000$  (Table 2.19 and Figures 2.62-2.64).



**Figure 2.62** Somatic metaphase chromosome plate of *Pistia stratiotes* (diploid,  $2n=28$ ).



**Figure 2.63** Karyotype of *Pistia stratiotes* by conventional staining.

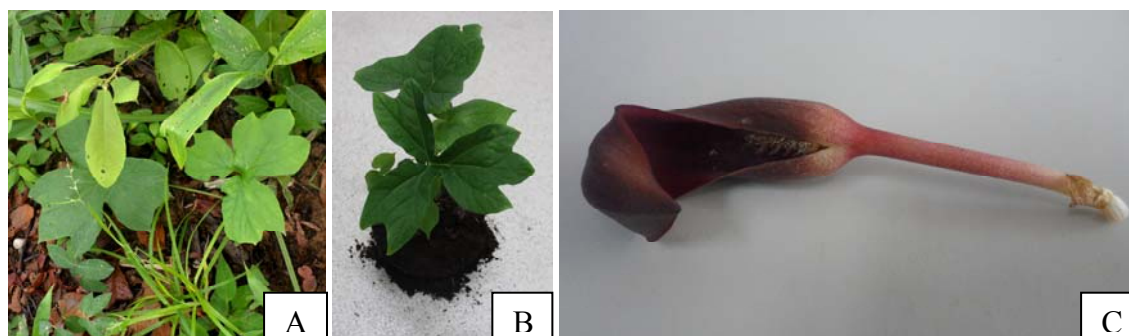


**Figure 2.64** Idiogram of *Pistia stratiotes* by conventional staining.



### 2.8.17 *Pycnospatha palmata* Gagnep.

Counting somatic metaphase chromosome from 20 cells of *Pycnospatha palmata* and then the arrangement of karyotypes and diagram by average length of the short arm (Ls) average length of the long arm (Ll) are measured by the length of each chromosome (LT). The average measurement of Relative Length (RL), Chromosome Index (CI), standard deviation (SD) of RL and CI from metaphase chromosomes in 20 cells and summarizes the size and type of chromosomes are shown in Table 2.20.



**Figure 2.65** *Pycnospatha palmata* Gagnep.

A. Plants in habitat  
B-C. Inflorescence

**Table 2.20** Mean length of short arm chromosome (Ls), long arm chromosome (Ll), total arm chromosome (LT), relative length (RL), centromeric index (CI) and standard deviation (SD) of RL, CI from 20 metaphases of *Pycnospatha palmata* (diploid, 2n=26).

Chromosome pair	Ls	Ll	LT	RL±SD	CI±SD	Chromosome size	Chromosome type
1	47.198	52.208	99.407	0.098±0.001	0.525±0.000	Large	Metacentric
2	44.382	50.673	95.055	0.093±0.000	0.533±0.000	Large	Metacentric
3	42.336	46.331	88.668	0.087±0.001	0.523±0.000	Large	Metacentric
4	40.795	47.234	88.029	0.087±0.002	0.537±0.000	Large	Metacentric
5	38.838	44.707	83.545	0.082±0.000	0.535±0.000	Large	Metacentric
6	30.374	52.788	83.162	0.082±0.002	0.635±0.000	Large	Submetacentric
7	27.509	52.725	80.233	0.079±0.002	0.657±0.000	Large	Submetacentric
8	36.535	39.057	75.592	0.074±0.000	0.517±0.000	Large	Metacentric
9	20.352	48.821	69.173	0.068±0.003	0.706±0.000	Medium	Acrocentric
10	29.142	39.529	68.671	0.067±0.000	0.576±0.000	Medium	Metacentric
*11	34.345	34.025	68.370	0.067±0.003	0.502±0.000	Medium	Metacentric
12	21.629	44.713	66.343	0.065±0.001	0.674±0.000	Medium	Submetacentric
13	23.422	27.769	51.191	0.050±0.000	0.542±0.000	Medium	Metacentric

Remarks: \*=satellite chromosome (NORs).

Karyotype formula is  $L_{12}^m + L_4^{sm} + M_6^m + M_2^{sm} + M_2^a$

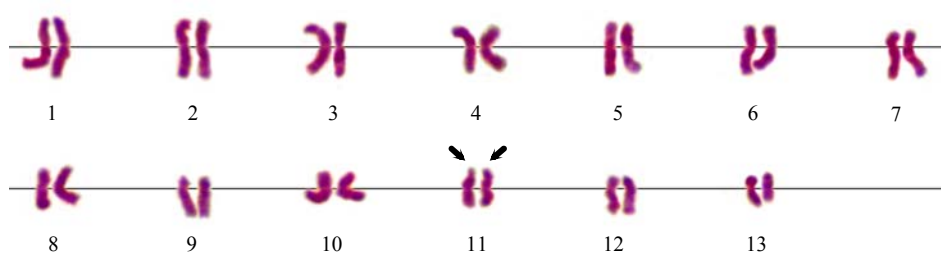


### Study on number, type and structure of the Chromosome

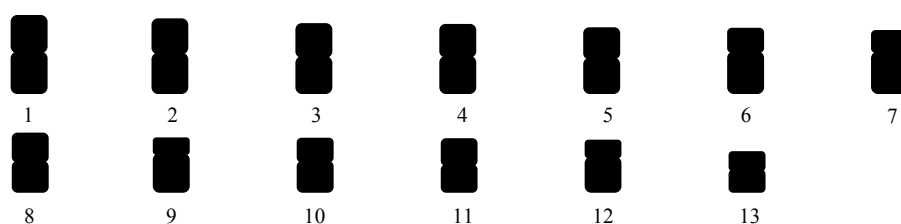
Chromosome number of *Pycnospatha palmata* is diploid chromosome  $2n=26$  contains the chromosomes 13 pairs or 26 bars. The karyotype formula of this species was found to be asymmetrical,  $L^{m}_{12} + L^{sm}_{4} + M^{m}_{6} + M^{sm}_{2} + M^a_{2}$ , include 6 pair a large metacentric chromosome, 2 pair a large submetacentric chromosome, 3 pair a medium metacentric chromosome, 1 pair a medium submetacentric chromosome and 1 pair a medium acrocentric chromosome. The relative length is a value between  $0.050 \pm 0.000$  to  $0.098 \pm 0.000$  and two visible satellites (Table 2.20 and Figures 2.66-2.68).



**Figure 2.66** Somatic metaphase chromosome plate of *Pycnospatha palmata* (diploid,  $2n=26$ ).



**Figure 2.67** Karyotype of *Pycnospatha palmata* (Arrows indicate satellite chromosomes) by conventional staining.

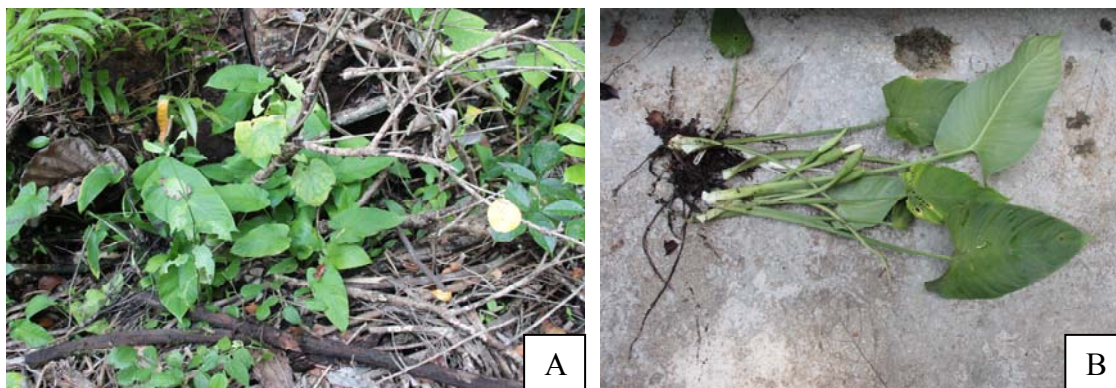


**Figure 2.68** Idiogram of *Pycnospatha palmate* by conventional staining.



### 2.8.18 *Schismatoglottis calyptrata* (Roxb.) Zoll. & Moritzi

Counting somatic metaphase chromosome from 20 cells of *Schismatoglottis calyptrata* and then the arrangement of karyotypes and diogram by average length of the short arm (Ls) average length of the long arm (Ll) are measured by the length of each chromosome (LT). The average measurement of Relative Length (RL), Chromosome Index (CI), standard deviation (SD) of RL and CI from metaphase chromosomes in 20 cells and summarizes the size and type of chromosomes are shown in Table 2.21.



**Figure 2.69** *Schismatoglottis calyptrata* (Roxb.) Zoll. & Moritzi

A. Plants in habitat

B. Inflorescence

**Table 2.21** Mean length of short arm chromosome (Ls), long arm chromosome (Ll), total arm chromosome (LT), relative length (RL), centromeric index (CI) and standard deviation (SD) of RL, CI from 20 metaphases of *Schismatoglottis calyptrata* (diploid,  $2n=26$ ).

Chromosome pair	Ls	Ll	LT	RL $\pm$ SD	CI $\pm$ SD	Chromosome size	Chromosome type
1	30.232	35.721	65.953	0.103 $\pm$ 0.000	0.542 $\pm$ 0.000	Large	Metacentric
*2	35.097	23.900	58.998	0.093 $\pm$ 0.003	0.505 $\pm$ 0.000	Large	Metacentric
3	18.098	37.433	55.531	0.087 $\pm$ 0.000	0.674 $\pm$ 0.000	Large	Submetacentric
4	22.615	29.566	52.181	0.082 $\pm$ 0.000	0.567 $\pm$ 0.000	Large	Metacentric
5	17.080	33.957	51.037	0.080 $\pm$ 0.000	0.605 $\pm$ 0.000	Medium	Submetacentric
6	25.580	24.940	50.520	0.079 $\pm$ 0.002	0.594 $\pm$ 0.000	Medium	Metacentric
*7	29.342	20.705	50.047	0.079 $\pm$ 0.002	0.514 $\pm$ 0.000	Medium	Metacentric
8	15.315	32.302	47.617	0.075 $\pm$ 0.000	0.678 $\pm$ 0.000	Medium	Submetacentric
9	16.050	29.549	45.599	0.072 $\pm$ 0.001	0.648 $\pm$ 0.000	Medium	Submetacentric
10	17.050	25.125	42.175	0.066 $\pm$ 0.000	0.601 $\pm$ 0.000	Medium	Submetacentric
11	16.315	24.963	41.277	0.065 $\pm$ 0.000	0.605 $\pm$ 0.000	Medium	Submetacentric
12	12.554	27.555	40.109	0.063 $\pm$ 0.000	0.687 $\pm$ 0.000	Medium	Submetacentric
13	15.839	20.440	36.279	0.057 $\pm$ 0.000	0.563 $\pm$ 0.000	Medium	Metacentric

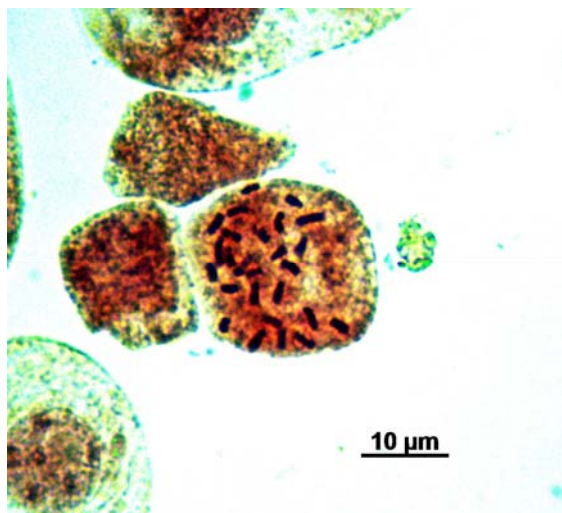
Remarks: \*=satellite chromosome (NORs).

Karyotype formula is  $L^m_6 + L^{sm}_2 + M^m_6 + M^{sm}_{12}$

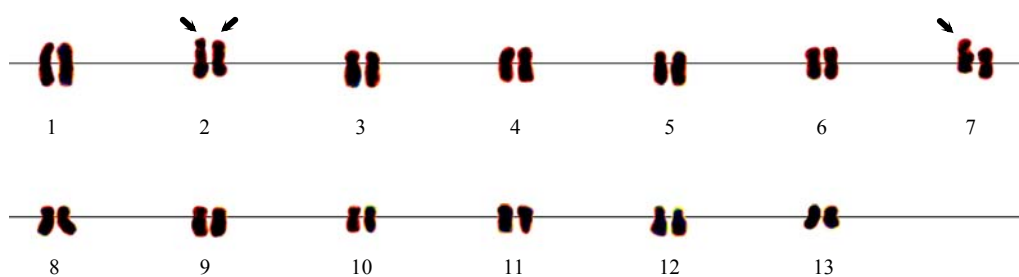


### Study on number, type and structure of the Chromosome

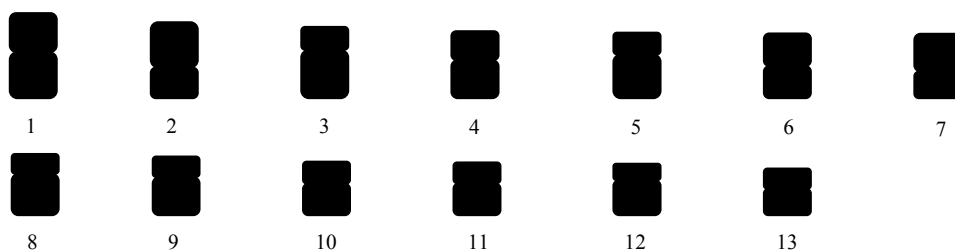
Chromosome number of *Schismatoglottis calyprata* is diploid chromosome  $2n=26$  contains the chromosomes 13 pairs or 26 bars. The karyotype formula of this species was found to be symmetrical,  $L^m_6 + L^{sm}_2 + M^m_6 + M^{sm}_{12}$ , include 3 pair a large metacentric chromosome, 1 pair a large submetacentric chromosome, 3 pair a medium metacentric chromosome and 6 pair a medium submetacentric chromosome. The relative length is a value between  $0.057 \pm 0.000$  to  $0.103 \pm 0.000$  and three visible satellites (Table 2.21 and Figures 2.70-2.72).



**Figure 2.70** Somatic metaphase chromosome plate of *Schismatoglottis calyprata* (diploid,  $2n=26$ ).



**Figure 2.71** Karyotype of *Schismatoglottis calyprate* (Arrows indicate satellite chromosomes) by conventional staining.



**Figure 2.72** Idiogram of *Schismatoglottis calyprate* by conventional staining.





### 2.8.19 *Typhonium glaucum* Hett. & Sookchaloem

Counting somatic metaphase chromosome from 20 cells of *Typhonium glaucum* and then the arrangement of karyotypes and diagram by average length of the short arm (Ls) average length of the long arm (Ll) are measured by the length of each chromosome (LT). The average measurement of Relative Length (RL), Chromosome Index (CI), standard deviation (SD) of RL and CI from metaphase chromosomes in 20 cells and summarizes the size and type of chromosomes are shown in Table 2.22.



**Figure 2.73** *Typhonium glaucum* Hett. & Sookchaloem

**Table 2.22** Mean length of short arm chromosome (Ls), long arm chromosome (Ll), total arm chromosome (LT), relative length (RL), centromeric index (CI) and standard deviation (SD) of RL, CI from 20 metaphases of *Typhonium glaucum* (diploid, 2n=24).

Chromosome pair	Ls	Ll	LT	RL±SD	CI±SD	Chromosome size	Chromosome type
*1	43.865	46.173	90.037	0.137±0.000	0.513±0.000	Large	Metacentric
2	36.594	42.449	79.043	0.120±0.000	0.537±0.000	Large	Metacentric
3	29.538	40.176	69.714	0.106±0.000	0.576±0.000	Large	Metacentric
4	23.092	34.317	57.409	0.087±0.000	0.598±0.000	Medium	Metacentric
5	20.580	30.298	50.878	0.077±0.000	0.596±0.000	Medium	Metacentric
6	19.112	30.334	49.446	0.075±0.000	0.613±0.000	Medium	Submetacentric
7	18.844	29.549	48.393	0.074±0.000	0.611±0.000	Medium	Submetacentric
8	16.816	29.555	46.371	0.071±0.000	0.637±0.000	Medium	Submetacentric
9	22.306	22.703	45.009	0.068±0.000	0.504±0.000	Small	Metacentric
10	16.580	24.982	41.563	0.063±0.000	0.601±0.000	Small	Submetacentric
11	17.068	23.740	40.808	0.062±0.000	0.582±0.000	Small	Metacentric
12	14.066	24.733	38.799	0.059±0.000	0.637±0.000	Small	Submetacentric

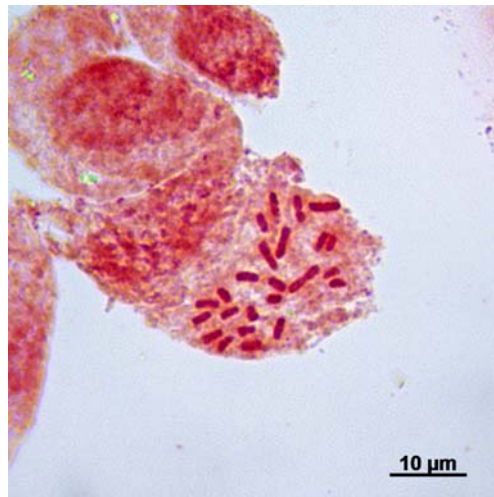
Remarks: \*=satellite chromosome (NORs).

Karyotype formula is  $L^m_6 + M^m_4 + M^{sm}_6 + S^m_4 + S^{sm}_4$

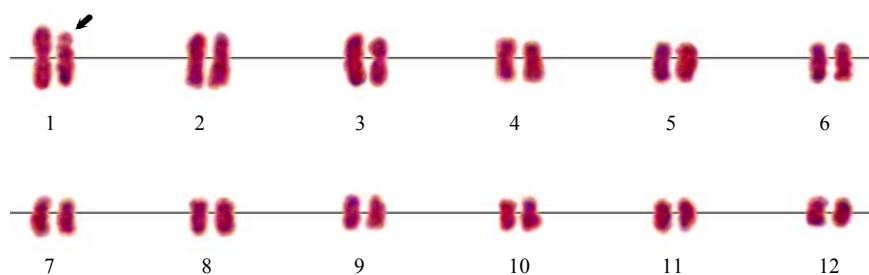


### Study on number, type and structure of the Chromosome

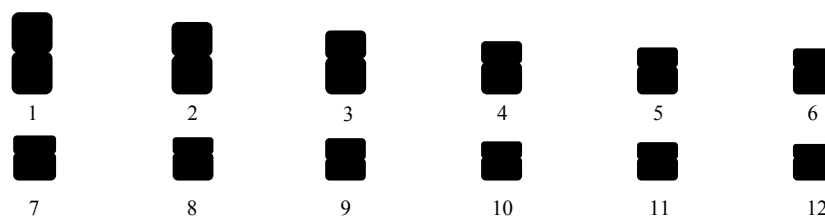
Chromosome number of *Typhonium glaucum* is diploid chromosome  $2n=24$  contains the chromosomes 12 pairs or 24 bars. The karyotype formula of this species was found to be symmetrical,  $L^m_6 + M^m_4 + M^{sm}_6 + S^m_4 + S^{sm}_4$ , include 3 pair a large metacentric chromosome, 2 pair a medium metacentric chromosome, 3 pair a medium submetacentric chromosome, 2 pair a small metacentric chromosome and 2 pair a small submetacentric chromosome. The relative length is a value between  $0.059 \pm 0.000$  to  $0.137 \pm 0.000$  and one visible satellite (Table 2.22 and Figures 2.74-2.76).



**Figure 2.74** Somatic metaphase chromosome plate of *Typhonium glaucum* (diploid,  $2n=24$ ).



**Figure 2.75** Karyotype of *Typhonium glaucum* (Arrows indicate satellite chromosomes) by conventional staining.



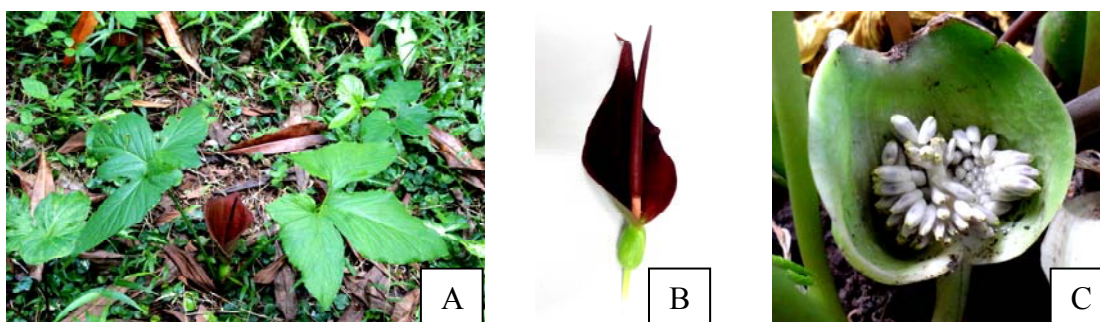
**Figure 2.76** Idiogram of *Typhonium glaucum* by conventional staining.





### 2.8.20 *Typhonium trilobatum* (L.) Schott

Counting somatic metaphase chromosome from 20 cells of *Typhonium trilobatum* and then the arrangement of karyotypes and diagram by average length of the short arm (Ls) average length of the long arm (Ll) are measured by the length of each chromosome (LT). The average measurement of Relative Length (RL), Chromosome Index (CI), standard deviation (SD) of RL and CI from metaphase chromosomes in 20 cells and summarizes the size and type of chromosomes are shown in Table 2.23.



**Figure 2.77** *Typhonium trilobatum* (L.) Schott

- A. Plants in habitat
- B. Inflorescence
- C. Seeds

**Table 2.23** Mean length of short arm chromosome (Ls), long arm chromosome (Ll), total arm chromosome (LT), relative length (RL), centromeric index (CI) and standard deviation (SD) of RL, CI from 20 metaphases of *Typhonium trilobatum* (diploid,  $2n=18$ ).

Chromosome pair	Ls	Ll	LT	RL $\pm$ SD	CI $\pm$ SD	Chromosome size	Chromosome type
1	26.329	63.609	89.938	0.171 $\pm$ 0.003	0.707 $\pm$ 0.000	Large	Acrocentric
2	24.002	57.508	81.510	0.155 $\pm$ 0.001	0.706 $\pm$ 0.000	Large	Acrocentric
3	22.464	51.803	74.266	0.141 $\pm$ 0.001	0.600 $\pm$ 0.000	Large	Acrocentric
4	25.794	40.534	66.327	0.126 $\pm$ 0.002	0.611 $\pm$ 0.000	Large	Submetacentric
5	25.086	32.301	57.387	0.109 $\pm$ 0.000	0.563 $\pm$ 0.000	Medium	Metacentric
6	17.891	26.769	44.660	0.085 $\pm$ 0.000	0.599 $\pm$ 0.000	Small	Metacentric
7	16.404	23.466	39.870	0.076 $\pm$ 0.001	0.589 $\pm$ 0.000	Small	Metacentric
8	15.079	22.980	38.060	0.072 $\pm$ 0.001	0.599 $\pm$ 0.000	Small	Metacentric
9	14.214	20.579	34.792	0.066 $\pm$ 0.002	0.591 $\pm$ 0.000	Small	Metacentric

Karyotype formula is  $L^{sm}_2 + L^a_6 + M^m_2 + S^m_8$

Study on number, type and structure of the Chromosome

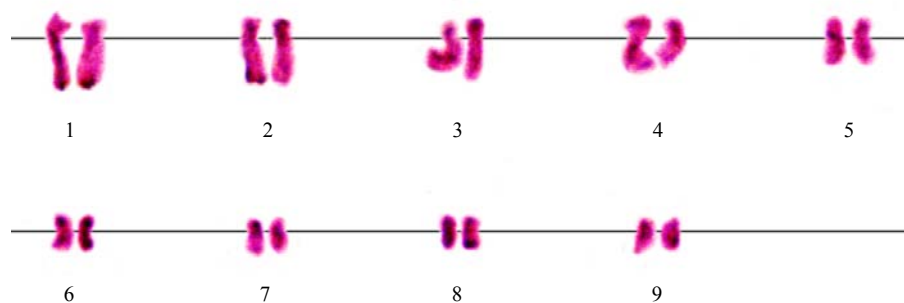
Chromosome number of *Typhonium trilobatum* is diploid chromosome  $2n=18$  contains the chromosomes 9 pairs or 18 bars. The karyotype formula of this species was found to be asymmetrical,  $L^{sm}_2 + L^a_6 + M^m_2 + S^m_8$ , include 1 pair a large submetacentric chromosome, 3 pair a large acrocentric chromosome, 1 pair a medium metacentric chromosome and 4 pair a small metacentric chromosome. The relative length is a value



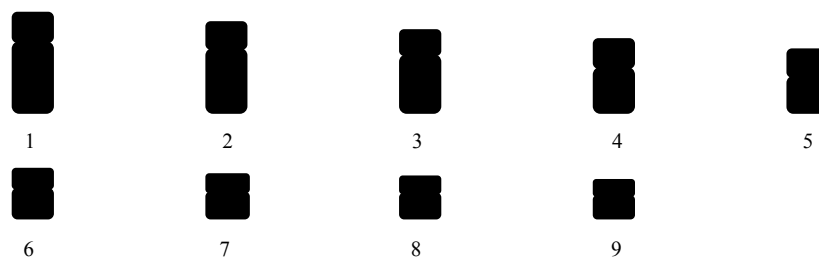
between  $0.066 \pm 0.002$  to  $0.171 \pm 0.003$  and three visible satellites (Table 2.23 and Figures 2.78-2.80).



**Figure 2.78** Somatic metaphase chromosome plate of *Typhonium trilobatum* (diploid,  $2n=18$ ).



**Figure 2.79** Karyotype of *Typhonium trilobatum* by conventional staining.



**Figure 2.80** Idiogram of *Typhonium trilobatum* by conventional staining.

## 2.9 Conclusion and Discussion

Results of this study the chromosomes number, karyotype and idiogram from root tip of 20 species Araceae in Thailand are studied with the Feulgen squash technique under Light Microscope and Computer Program (Tables 2.3). The chromosomes number and the karyotypes formula are shown in Table 2.24 and 2.25.

**Table 2.24** The chromosomes numbers and the karyotypes formula of 20 species plant Araceae family (Thailand) in this study including the data by other authors.

Species	Chromosome numbers (2n)	karyotype formulas	NOR
1. <i>Agloanema modestum</i>	40	$L_{18}^m + L_{4}^{sm} + L_6^a + Ms_8^m + S_2^m + S_2^{sm}$	-
2. <i>A. simplex</i>	42	$L_4^m + L_{18}^{sm} + L_6^a + M_2^m + M_2^{sm} + M_4^a + S_6^{sm}$	-
3. <i>Alocacia cucullata</i>	28	$L_{18}^m + M_4^m + M_6^{sm}$	3(STR)
4. <i>A. longiloba</i>	58	$L_{24}^m + M_{14}^m + M_{16}^{sm} + M_2^a + S_2^{sm}$	-
5. <i>A. macrorrhizos</i>	28	$L_8^m + M_{12}^m + M_8^{sm}$	-
6. <i>A. sp.</i>	28	$L_6^m + L_6^a + M_2^m + M_{12}^{sm} + S_2^{sm}$	-
7. <i>Amorphophallus serrulatus</i>	26	$L_8^m + M_{18}^m$	2(STR)
8. <i>Arisaema maxwellii</i>	24	$L_4^m + M_{12}^m + S_6^m + S_2^{sm}$	2(STR)
9. <i>Colocasia esculenta</i>	28	$L_{16}^m + M_4^m + M_6^{sm} + M_2^a$	
10. <i>C. fallax</i>	28	$L_{12}^m + M_{12}^m + M_4^{sm}$	2(STR)
11. <i>C. gigantea</i>	28	$L_{18}^m + L_2^{sm} + M_2^m + M_4^{sm} + M_2^a$	-
12. <i>C. lihengiae</i>	28	$L_{10}^m + L_4^{sm} + L_2^a + M_6^m + M_4^a + S_2^m$	-
13. <i>Hapaline benthamiana</i>	26	$L_8^m + M_{18}^m$	1(STR)
14. <i>Homalonema griffithii</i>	40	$L_{12}^m + L_8^{sm} + M_{18}^m + M_2^{sm}$	2(SCR)
15. <i>Lasia spinosa</i>	26	$L_6^{sm} + L_2^a + M_{14}^m + M_4^{sm}$	-
16. <i>Pistia stratiotes</i>	28	$L_{10}^m + M_{18}^m$	-
17. <i>Pycnosphata palmata</i>	26	$L_{12}^m + L_4^{sm} + M_6^m + M_2^{sm} + M_2^a$	2(STR)
18. <i>Scittomaglottis calyptrata</i>	26	$L_2^m + L_8^{sm} + M_8^m + M_6^{sm}$	3(STR)
19. <i>Typhonium glaucum</i>	24	$L_6^m + M_4^m + M_6^{sm} + S_4^m + S_4^{sm}$	1(STR)
20. <i>T. trilobatum</i>	18	$L_2^{sm} + L_6^a + M_2^m + S_8^m$	-

Notes: 2n = diploid chromosome number, m = metacentric chromosome, sm = submetacentric chromosome, a = acrocentric chromosome, t = telocentric chromosome, st = subtelocentric chromosome, NOR = number organism regions, STR = subtelomeric region, SCR = subcentromeric regions, and - = not available.



**Table 2.25** The chromosomes numbers of 20 species plant Araceae family (Thailand) in this study including the data by other authors. New records are marked with an asterisk.

Species (Specimens investigated)	Present study (2n)	Previous recorded		
		n	2n	Author
1. <i>Agloanema modestum</i>	40	-	60	Chen <i>et al.</i> (2003)
2. <i>A. simplex</i>	42*	-	-	-
3. <i>Alocacia cucullata</i>	28	-	28	Darlington and Wylie (1945); Ankei (1987) and Ishida (2001)
4. <i>A. longiloba</i>	58	-	56	Bhattacharya (1974); Sharma (1970)
5. <i>A. macrorrhizos</i>	28	-	28	Bhattacharya (1974); Ishida (2001); Sharma (1970)
6. <i>A. sp.</i>	28*	-	-	-
7. <i>Amorphophallus serrulatus</i>	26*	-	-	-
8. <i>Arisaema maxwellii</i>	24*	-	-	-
9. <i>Colocasia esculenta</i>	28	-	28	Li (1989); Sreekumari and Mathew (1989); Okada and Hambali (1989); Kuruvilla <i>et al.</i> (1989); Sreekumari and Mathew (1991a); Sreekumari and Mathew (1991b); Ivancic and Lebot (1999); Parvin <i>et al.</i> (2008)
			28 (42)	Coates <i>et al.</i> (1988); Zhang and Yang (1984); Sreekumari and Mathew (1995d); Kuruvilla and Singh (1981); Ramachandran (1978)
			36	Huang <i>et al.</i> (1989)
			38 (42)	Tanimoto and Matsumoto (1986)
			42	Subramanian and Munian (1988); Zhang (1998)
			42 (84)	Sreekumari and Mathew (1991c)
10. <i>C. fallax</i>	28	-	28	Begum and Alam (2009)
11. <i>C. gigantea</i>	28	-	28	Nakajima (1936); Moore (1973); Tanimoto and Matsumoto (1986); Okada and Hambali (1989); Yang <i>et al.</i> (2003); Cao and Long (2004)
12. <i>C. lihengiae</i>	28		28	Long and Liu (2001); Cao and Long (2004)
13. <i>Hapaline benthamiana</i>	26*	-	-	-
14. <i>Homalonema griffithii</i>	40	-	40	Okada (1982); Okada (2000)
15. <i>Lasia spinosa</i>	26	-	26	Ramachandran (1978)
			26 (27)	Sultana <i>et al.</i> (2006)



**Table 2.25** The chromosomes numbers of 20 species plant Araceae family (Thailand) in this study including the data by other authors. New records are marked with an asterisk (Continued).

Species (Specimens investigated)	Present study (2n)	Previous recorded		
		n	2n	Author
16. <i>Pistia stratiotes</i>	28	-	28	Ramachandran (1978); Sarkar and Chatterjee (1978); Sarkar <i>et al.</i> (1978); Subramanian and Munian (1988), Sarkar (1991), Selvaraj (1993)
17. <i>Pycnosphata palmata</i>	26*	-	-	-
18. <i>Scittomaglottis calyptrata</i>	26	-	26	Okada (1982)
19. <i>Typhonium glaucum</i>	24*	-	-	-
20. <i>T. trilobatum</i>	18	-	18	Subramanian and Munian (1988); Zaman and Podder (1981); Bian <i>et al.</i> (2002); Das <i>et al.</i> (2006);
			18 (26)	Chaudhuri and Sharma (1979);
			36	Ramachandran, (1978)

### *Agloanema*

The somatic chromosome of *A. modestum* is chromosome number 2n (diploid) = 40 and NF = 80 which is differs with Chen *et al.* (2003), somatic chromosome number of 2n = 60. The karyotype formula composes of  $L_{18}^m + L_4^{sm} + L_6^a + M_8^{sm} + S_2^m + S_2^{sm}$  is asymmetrical karyotype.

The somatic chromosome of *A. simplex* is chromosome number 2n (diploid) = 42 and NF = 84. The karyotype formula is  $L_4^m + L_{18}^{sm} + L_6^a + M_2^m + M_2^{sm} + M_4^a + S_6^{sm}$ , which is asymmetrical karyotype. The chromosome number, karyotype and idiogram of this species are the first time study.

### *Alocacia*

The somatic chromosome number of *A. cucullata* is 2n (diploid) = 28 corroborated Ishida (2001) which is consistent with Darlington and Wylie (1945) has somatic chromosome number of 2n = 28 this consistent with Ankei (1987) and it NF = 56. The karyotype formula is  $L_{18}^m + M_4^m + M_6^{sm}$  with symmetrical karyotype and three visible satellites

The somatic chromosome number of *A. longiloba* is 2n (diploid) = 58 NF = 116 which is differs with Bhattacharya (1974) and Sharma (1970) reported 2n = 56. The karyotype formula is  $L_{24}^m + M_{14}^m + M_{16}^{sm} + M_2^a + S_2^{sm}$  with asymmetrical karyotype.

The somatic chromosome number of *A. macrorrhizos* is 2n (diploid) = 28 which is consistent with Ishida (2001) and it has NF= 56. The karyotype formula is  $L_8^m + M_{12}^m + M_8^{sm}$  and symmetrical karyotype.



The somatic chromosome number of *Alocacia* sp. is  $2n$  (diploid) = 28 with NF = 56. The karyotype formula is  $L^m_6 + L^a_6 + M^m_2 + M^{sm}_{12} + S^{sm}_2$  with asymmetrical karyotype. This is the first time reported of chromosome number, karyotype and idiogram of this species.

### ***Amorphophallus***

The somatic chromosome number of *A. serrulatus* is  $2n$  (diploid) = 26 and has NF = 52. The karyotype formula composes of  $L^m_8 + M^{sm}_{18}$  is symmetrical karyotype and two visible satellites. The chromosome number, karyotype and idiogram of this species are the first time study.

### ***Arisaema***

The somatic chromosome number of *A. maxwellii* is  $2n$  (diploid) = 24 with NF = 48. The karyotype formula is  $L^m_4 + M^{sm}_{12} + S^{sm}_6 + S^{sm}_2$  and symmetrical karyotype with two visible satellites. This is the first time reported of chromosome number, karyotype and idiogram of this species.

### ***Colocasia***

The somatic chromosome number of *C. esculenta* is  $2n$  (diploid) = 28 and it has NF = 56 which is consistent with Larsen (1969)  $2n=28$ . While Marchant (1971) reported basic chromosome number of *C. esculenta*  $x = 7$ , chromosome small size and somatic chromosome number  $2n=28$ . In addition, Moore (1973) reported differs somatic chromosome number  $2n=28$  but indiffers with Sharma and Sarkar (1963) reported somatic chromosome number  $2n=22, 26, 38$  and Yen and Wheeler (1968) reported somatic chromosome number of *C. esculenta*  $2n=42$ . The karyotype formula  $L^{m_{16}} + M^{m_4} + M^{sm_6} + M^a_2$  with asymmetrical karyotype which is consistent with Sreekumari and Mathew (1991a)  $2n = 28$  karyotype formula is (II)  $20m + 6sm + 2a$  but inconsistent with Sreekumari and Mathew (1991a) reported  $2n=28$  but the karyotype formula are different (I)  $20m + 8sm$  (III)  $18m + 8sm + 2a$  (IV)  $22m + 2sm + 6a$  (V)  $6m + 12sm + 10a$  there are present karyotype formula symmetrical and asymmetrical. Parvin *et al.* (2008) reported somatic chromosome number of *C. esculenta*  $2n=28$  and karyotype formula (Cytotype 1)  $18m + 6sm + 4a$ ,  $2n=28$  (Cytotype 2)  $20m + 4sm + 4a$  has somatic chromosome number  $2n=28$  (Cytotype 3)  $38m + 4sm$  chromosome number  $2n=42$  (Cytotype 4)  $12m+2sm$ ,  $2n=21$  (Cytotype 5)  $24m + 4sm$ ,  $2n=28$  (Cytotype 6)  $28m$ ,  $2n=28$  (Cytotype 7)  $22m + 6sm$ ,  $2n=28$ , respectively.

The somatic chromosome number of *C. fallex* is  $2n$  (diploid) = 28 and NF = 56 karyotype formula is  $L^{m_{12}} + M^{m_{12}} + M^{sm_4}$  with symmetrical karyotype which is consistent with Begum and Alam (2009) reported somatic chromosome number of *C. fallex* three morphological form including Deep purple petiole form  $2n = 28$  NF = 56 karyotype formula is  $24^m + 4^{sm}$  while two form indiffers in Green petiole form  $2n = 28$  karyotype formula is  $20^m + 8^{sm}$  and Light purple petiole form  $2n = 30$  karyotype formula is  $26^m + 2^{sm} + 2^{ac}$ , respectively.

The somatic chromosome number of *C. gigantea* is  $2n$  (diploid) = 28 with NF = 56 which is consistent with Moore (1973), Tanimoto & Matsumoto (1986), Okada & Hambali (1989), Yang *et al.* (2003) and Cao & Long (2004) reported somatic





chromosome number of *C. gigantea*  $2n = 28$  but it differs with Nakajima (1936) reported that there somatic chromosome number of *C. gigantea*  $2n = 42$ . The karyotype formula is  $L_{18}^m + L_2^{sm} + M_2^m + M_4^{sm} + M_2^a$  is asymmetrical karyotype and two visible satellites.

The somatic chromosome number of *C. lihengiae* is  $2n$  (diploid) = 28 and has  $NF = 56$  which is inconsistent with Long & Liu (2001) reported that there somatic chromosome number of *C. lihengiae*  $2n = 28$ . In addition, Cao and Long (2004) reported that there somatic chromosome number of *C. lihengiae*  $2n = 28$  but  $NF = 52$  karyotype formula composes  $18^m + 6^{sm} + 4^{st} 1(SCR)$  and has one visible satellite that are different somatic chromosome number  $2n = 28$   $NF = 56$  and the karyotype formula composes of  $L_{10}^m + L_4^{sm} + L_2^a + M_6^m + M_4^a + S_2^m$  is asymmetrical karyotype in this study.

### ***Hapaline***

The somatic chromosome number of *H. benthamiana* is  $2n$  (diploid) = 26 and has  $NF = 52$ . The karyotype formula is  $L_8^m + M_{18}^m$  and symmetrical karyotype with one visible satellite. This species is recorded somatic chromosome number for the first time.

### ***Homalonema***

The somatic chromosome number of *H. griffithii* is  $2n$  (diploid) = 38 with  $NF = 76$  which is consistent with Okada (1982) and Okada (2000) that are reported somatic chromosome number of *H. griffithii*  $2n = 40$ . The karyotype formula is  $L_{12}^m + L_8^{sm} + M_{18}^m + M_2^{sm}$  with symmetrical karyotype and two visible satellites.

### ***Lasia***

The somatic chromosome number of *L. spinosa* is  $2n$  (diploid) = 26 and it has  $NF = 52$ . The karyotype formula is  $L_6^{sm} + L_2^a + M_{14}^m + M_4^{sm}$  with asymmetrical karyotype. While, Ramachandran (1978) reported  $2n = 26$ , Sultana *et al.* (2006) reported  $2n = 26-27$

### ***Pistia***

The somatic chromosome number of *P. stratiotes* is  $2n$  (diploid) = 28 and it has  $NF = 56$  which is consistent with Ramachandran (1978), Sarkar & Chatterjee (1978), Sarkar *et al.* (1978), Subramanian & Munian (1988), Sarkar (1991) and Selvaraj (1993) reported somatic chromosome number of *P. stratiotes*  $2n = 18$ . The karyotype formula is  $L_{10}^m + M_{18}^m$  with symmetrical karyotype.

### ***Pycnosphata***

The somatic chromosome number of *P. palmata* is  $2n$  (diploid) = 26 and it has  $NF = 52$ . The karyotype formula is  $L_{12}^m + L_4^{sm} + M_6^m + M_2^{sm} + M_2^a$  with asymmetrical karyotype and two visible satellites. This species is recorded somatic chromosome number for the first time.





### *Scittomaglottis*

The somatic chromosome number of *S. calyptrate* is  $2n$  (diploid) = 26 which is consistent with Okada (1982) has somatic chromosome number of  $2n = 26$  and it has  $NF = 52$ . The karyotype formula composes of  $L^m_6 + L^{sm}_2 + M^m_6 + M^{sm}_{12}$  with symmetrical karyotype and three visible satellites.

### *Typhonium*

The somatic chromosome number of *T. glaucum* is  $2n$  (diploid) = 24 and it has  $NF = 48$ . The karyotype formula is  $L^m_6 + M^m_4 + M^{sm}_6 + S^m_4 + S^{sm}_4$  with symmetrical karyotype and one visible satellite. This species is recorded somatic chromosome number for the first time.

The somatic chromosome number of *T. trilobatum* is  $2n$  (diploid) = 18 and it has  $NF = 36$  which is consistent with Subramanian & Munian (1988), Zaman, & Podder, (1981), Bian *et al.* (2002) and Das *et al.* (2006) has somatic chromosome number of  $2n = 18$ , moreover, it consistent with Ramachandran, (1978) reported somatic chromosome number of *T. trilobatum* is  $2n = 36$ . In addition, Chaudhuri & Sharma (1979) reported that there somatic chromosome number  $2n = 18$  and 26. The karyotype formula is  $L^{sm}_2 + L^a_6 + M^m_2 + S^m_8$  with asymmetrical karyotype.

## 2.10 Summary

Chromosome counts of 20 species Araceae in Thailand divided into seven groups as follows;

1. The first group is chromosome number  $2n = 18$  i.e. *Typhonium trilobatum* (L.) Schott.
2. The second group is chromosome number  $2n = 24$  including *Arisaema maxwellii* Hett. & Gusman, and *Typhonium glaucum* Hett. & Sookchaloem.
3. The third group is chromosome number  $2n = 26$  including *Amorphophallus serrulatus* Hett. & A.Galloway, *Hapaline benthamiana* Schott, *Lasia spinosa* (L.) Thwaites, *Pycnospatha palmata* Gagnep. and *Schismatoglottis calyptrata* (Roxb.) Zoll. & Moritzi.
4. The fourth group is chromosome number  $2n = 28$  including *Alocasia cucullata* (Lour.) G.Don, *Alocasia macrorrhizos* (L.) G.Don, *Alocasia* sp., *Colocasia esculenta* (L.) Schott, *Colocasia fallax* Schott, *Colocasia gigantea* (Blume) Hook.f., *Colocasia lihengiae* C.L.Long & K.M.Liu and *Pistia stratiotes* L..
5. The fifth group is chromosome number  $2n = 40$  i.e. *Aglaonema simplex* (Blume) Blume and *Homalomena griffithii* (Schott) Hook.f..
6. The sixth group is chromosome number  $2n = 42$  such as *Aglaonema modestum* Schott ex Engl..
7. The seventh group is chromosome number  $2n = 58$  i.e. *Alocasia longiloba* Miq..



Ten species has karyotype formula is symmetrical including *Alocasia cucullata* (Lour.) G.Don, *Alocasia macrorrhizos* (L.) G.Don, *Amorphophallus serrulatus* Hett. & A.Galloway, *Arisaema maxwellii* Hett. & Gusman, *Colocasia fallax* Schott, *Hapaline benthamiana* Schott, *Homalomena griffithii* (Schott) Hook.f., *Pistia stratiotes* L., *Schismatoglottis calyptrata* (Roxb.) Zoll. & Moritzi and *Typhonium glaucum* Hett. & Sookchaloem.

Ten species has karyotype formula is asymmetrical including *Aglaonema modestum* Schott ex Engl., *Aglaonema simplex* (Blume) Blume, *Alocasia longiloba* Miq., *Alocasia* sp., *Colocasia esculenta* (L.) Schott, *Colocasia gigantea* (Blume) Hook.f., *Colocasia lihengiae* C.L.Long & K.M.Liu, *Lasia spinosa* (L.) Thwaites, *Pycnospatha palmata* Gagnep. and *Typhonium trilobatum* (L.) Schott.

In this study, nine species somatic chromosomes were also found to be satellite chromosome, found that the end of the chromosome. It is assumed that during the evolution of plants it will change position. There is an increase or decrease in the number and size of the satellite, which is correlated with the position and the number of nucleolus (Tanomtong, 2011).

The chromosomes number of seven species has been recorded for the first time as follows; *Aglaonema simplex* (Blume) Blume, *Alocasia* sp., *Amorphophallus serrulatus* Hett. & A.Galloway, *Arisaema maxwellii* Hett. & Gusman, *Hapaline benthamiana* Schott, *Pycnospatha palmata* Gagnep. and *Typhonium glaucum* Hett. & Sookchaloem.

Chromosome numbers of *Aglaonema modestum* Schott ex Engl., *Alocasia cucullata* (Lour.) G.Don, *Alocasia longiloba* Miq., *Alocasia macrorrhizos* (L.) G.Don, *Colocasia esculenta* (L.) Schott, *Colocasia fallax* Schott, *Colocasia gigantea* (Blume) Hook.f., *Colocasia lihengiae* C.L.Long & K.M.Liu, *Homalomena griffithii* (Schott) Hook.f., *Lasia spinosa* (L.) Thwaites, *Pistia stratiotes* L., *Schismatoglottis calyptrata* (Roxb.) Zoll. & Moritzi and *Typhonium trilobatum* (L.) Schott. were reported the first time. However, the chromosome number data could not supported for classification in this study.

Stebbins (1950) suggested that the karyotype of organisms is involved to the size of chromosome and chromosome types which have similar metacentric and submetacentric chromosome belonging to only symmetrical karyotypes, but asymmetrical karyotypes contain very different chromosomes, including metacentric, submetacentric, acrocentric and telocentric chromosome. An asymmetrical karyotype has evolution higher more than symmetrical karyotype (Stebbins, 1971; Chaiyasut, 1989; Sheidai, 2000; Vargas *et al.*, 2007). The changes karyotype from symmetrical karyotype to asymmetrical karyotype involves changing the shape or structure of the chromosome. It may be caused by unequal translocation or pericentric inversion or is missing some of the chromatin (Stebbins, 1971; Chaiyasut, 1989; Sheidai, 2000; Vargas *et al.*, 2007; Brooker, 2009). In the evolution of plants there will be changes in chromosome size, both reduce the size and increase the size of chromosome. Evolution to reduce the size of chromosome, found in plants such as *Muscari* (Liliaceae) and *Crepis*, *Youngia* (Asteraceae or Compositae) etc. and evolution to increase the size of chromosome found in plants such as Graminae for example genus *Hordeum* etc. (Stebbins, 1971).

The different plants of genera, but the same chromosome number can be found in many family such as Zingiberaceae genus *Alpinia*, *Etlingera* and *Amomum* had  $2n = 48$  (Eksomtrame *et al.*, 2002) and Asteraceae or Compositae in the genus



*Argyranthemum*, *Tanacetum*, *Opisthopappus*, and *Crossostephium* had  $2n = 18$  (Jian *et al.*, 2008) etc. Also, plants in the same genus, but different species found that the same number of chromosomes such as Zingiberaceae genus *Zingiber* 5 species  $2n = 22$  equal (Mahanty, 1970) or Fabaceae genus *Lathyrus* 10 species number of chromosomes had  $2n = 14$  equal (Seijo and Fernandez, 2003) etc. So, the information of chromosome number to help support the classification organisms in the case of the same chromosome number. But, it may not be enough and need to study the karyotype of plant. Furthermore, the characteristics of satellite chromosome and chromosome length can be used for the identification of plants. However, in the same genus may find a species, with satellite chromosome and some species may not have satellite chromosome (Yu *et al.*, 2009). Therefore, a study on Karyotype of plants can help indicate the cause of the morphological changes that occurred from the environment or from changes associated with chromosome as the chromosome structure or changes in chromosome number. It is also used as evidence in the study of the relationships of organisms in evolution (Chaiyasut, 1989).



## CHAPTER 3

### PALYNOLOGY STUDIES

#### 3.1 Introduction

Palynology is an important the science of pollen and spore morphology in understanding for studying plant Taxonomy. The knowledge of palynology can be used as an instrument of multiple researches for systematic botany, paleobotany, paleoecology, pollen analysis, aeropalynology, criminology, allergy, stratigraphic correlation of oil bearing rocks and coalfields, drugs and improvement of honey. It is used as a means of tracing the history of cultivated cereals (Erdtman, 1954). Many characteristic of the pollen grain can be used identify to species of plants i.e. shape, polarity, symmetry, size and aperture (Erdtman, 1966; Nairs, 1971; Moore *et al.*, 1991).

#### 3.2 Palynology

##### 3.2.1 Definition and scope of palynology

Palynology is subject to study pollen of flowering plant and spores of non-flowering plant. In a restricted sense we consider palynology to be the study of fossil spore's pollen algae fungi and similar sized entities principally of plant origin and their significance in time life space and energy. The role of palynology in the exploration for oil is essentially com parable to that of any other branch of paleontology. The application of palynology to problems in stratigraphy correlation paleoecology and other aspects of geology and to various problems in botany archeology and meteorology has expanded rapidly since 1935 (Cross, 1964).

##### 3.2.2 Literature reviews of palynology

Erdtman (1971) studied the palynology of *Colocasia antiquorum* var. *esculenta* found that pollen are having 1-sulcate and 20x25  $\mu\text{M}$  sizes.

Huang (1972) studied the palynology of *Alocasia macrorrhiza* (L.) Schott found that 33x40-46x29  $\mu\text{M}$  size. The pollen wall: exine about 1  $\mu\text{M}$  thick, psilate or granulate.

Huang (1972) reported the palynology of *Colocasia formosana* Hay. found that 20-29x23-30x17-23  $\mu\text{M}$  size and about 4  $\mu\text{M}$  thick of pollen wall which the structure tectum layer inside equatorial is echinate 2  $\mu\text{M}$  long but at the polar aperture is presented a small spinose.

Hay and Wise (1991) studied the pollen of genus *Alocasia* found that spheroidal or subspheroidal, inaperture, apolar, radiosymmetric, an exine sculpturing is spinose are presented.

Few Araceae species is studied by few botanists. However, many species in this study will be study for the first time.



### 3.3 Plants material

All species of Araceae were collected from different parts of Thai forests or cultivated plant and transplanted in the nursery of Walai Rukhavej Botanical Research Institute, Mahasarakham University. Voucher specimens were deposited at the Mahasarakham University Herbarium (Table 3.1).

**Table 3.1** Plants used in the study

Species	Common name	Location	Collector number
1. <i>Agloanema modestum</i>	Khiao muen pi	Loei	R. Senavongse 001/2016
2. <i>A. simplex</i>	Wan ngot hin	Kalasin	R. Senavongse 006/2016
3. <i>Alocacia macrorrhizos</i>	Wan nang-kwak	Mukdahan	R. Senavongse 011/2016
4. <i>A. cucullata</i>	Kacho nok	Songkhla	R. Senavongse 016/2016
5. <i>A. longiloba</i>	Kradat	Mahasarakham	R. Senavongse 021/2016
6. <i>A. sp.</i>	-	Changmai	R. Senavongse 026/2016
7. <i>Amorphophallus serrulatus</i>	-	Ubon Ratchathani	R. Senavongse 031/2016
8. <i>Arisaema maxwellii</i>	-	Changmai	R. Senavongse 036/2016
9. <i>Colocasia esculenta</i>	Phueak or Bon	Chaiyaphum	R. Senavongse 016/2015
10. <i>C. fallax</i>	Bon-Doi	Changmai	R. Senavongse 041/2016
11. <i>C. gigantea</i>	Khun	Lampang	R. Senavongse 046/2016
12. <i>Hapaline benthamiana</i>	Bon tao	Chaiyaphum	R. Senavongse 056/2016
13. <i>Lasia spinosa</i>	Phuk-Nam	Roi-Et	R. Senavongse 066/2016
14. <i>Pistia stratiotes</i>	Jok	Mahasarakham	R. Senavongse 071/2016
15. <i>Pycnosphata palmata</i>	-	Ubon Ratchathani	R. Senavongse 076/2016
16. <i>Scittomaglottis calyptrata</i>	Bon khiao	Trang	R. Senavongse 081/2016
17. <i>Typhonium glaucum</i>	-	Ubon Ratchathani	R. Senavongse 086/2016
18. <i>T. trilobatum</i>	Uttapit	Chumphon	R. Senavongse 091/2016

### 3.4 Materials

1. Bottles for sampling roots
2. Forceps
3. Slide and cover glass
4. Beaker
5. Petri dish
6. Dropper
7. Permanent pen
8. Blotting paper
9. Bottles for soak color
10. Transparent Color Nail Polish
11. Distilled water
12. Absolute alcohol
13. 95% ethyl alcohol
14. 80% ethyl alcohol
15. 70% ethyl alcohol
16. Silicone oil
17. Light microscope (Zeiss: AxioStar plus)



### 3.5 Palynology study

Pollen grains of the family Araceae in Thailand were examined under light microscopy (LM) and scanning electron microscopy (SEM). Pollen samples were obtained from the spirit materials. Samples were dehydrated using an alcohol series of 70%, 80%, 95% and 100%. For LM studies, pollen grains were mounted in silicone oil and sealed with paraffin. At least 20 pollen grains per sample were measured for the diameter ( $\mu\text{m}$ ), exine sculpturing characters, shape and size.

### 3.6 Palynology Analysis

Data analyses will use the means and standard error. Shapes were described according to P/E ratio (the length of the polar axis to the equatorial diameter) (Erdtman, 1966). They are grouped as follows: oblate (oblate spherical = 0.88-0.99; suboblate = 0.76-0.87; oblate = 0.51-0.75; peroblate = 0.50 or lower) and spherical (=1); Prolate (perprolate = 2 or over; prolate = 1.34-1.99; subprolate = 1.15-1.33; prolate spherical = 1.01-1.14). For SEM studies, pollen grains in absolute alcohol were dried and affixed to aluminum stubs with double-sided cellophane tape. Samples were sputter-coated with a gold-palladium, examined and then photographs were taken with a LEO: LEO 1450VP SEM.

### 3.7 Results and Discussion

Pollen of 18 species of Araceae family (Table 3.2 and Figures 3.1-3.18) are monads, spheroidal and inaperturate, with thin exine and thick intine and indistinguishable under light microscope. Exine sculpture echinate with psilate or rugulate between the spines.

#### *Aglaonema modestum* Schott ex Engl.

Pollen grain is monad,  $11.40 \pm 1.22 \mu\text{m}$  in diam. Radial symmetry, heteropolar, monoporate aperture and spheroidal shape. Exine sculpturing is granulate (Table 3.2, Figure 3.1).

#### *Aglaonema simplex* (Blume) Blume

Pollen grain is a monad and  $50.27 \pm 4.84 \mu\text{m}$  in diam. Radial symmetry, apolar polarity, spheroidal shape. Exine sculpturing is echinate. The spine  $3.31 \pm 0.36 \mu\text{m}$  in length. Exine sculpturing echinate with granulate between the spines. The wall thickness  $7.07 \pm 1.74 \mu\text{m}$  (Table 3.2, Figure 3.2).

#### *Alocasia cucullata* (Lour.) G. Don

Pollen grain is a monad and  $22.31 \pm 2.18 \mu\text{m}$  (P)  $24.64 \pm 2.79 \mu\text{m}$  (E) in diam. Radial symmetry, apolar polarity, prolate spheroidal shape. Exine sculpturing is echinate. The spine  $1.36 \pm 0.90 \mu\text{m}$  in length. Exine sculpturing echinate with granulate between the spines. The wall thickness  $2.35 \pm 0.25 \mu\text{m}$  similar to Hay and Wise (1991)





found that pollen are spheroidal or subspheroidal, inaperture, apolar, radiosymmetric, an exine sculpturing is spinose of genus *Alocasia* (Table 3.2, Figure 3.3).

***Alocasia longiloba* Miq.**

Pollen grain is a monad and  $35.87 \pm 2.27 \mu\text{m}$  (P)  $37.02 \pm 7.28 \mu\text{m}$  (E) in diam. Radial symmetry, apolar polarlity, oblate spheroidal shape. Exine sculpturing is echinate. The spine  $3.02 \pm 0.33 \mu\text{m}$  in length. Exine sculpturing echinate with psilate between the spines. The wall thickness  $2.78 \pm 0.53 \mu\text{m}$ . similar to Hay and Wise (1991) found that pollen are spheroidal or subspheroidal, inaperture, apolar, radiosymmetric, an exine sculpturing is spinose of genus *Alocasia* (Table 3.2, Figure 3.4).

***Alocasia macrorrhizos* (L.) G.Don**

Pollen grain is a monad and  $36.85 \pm 3.72 \mu\text{m}$  (P)  $39.20 \pm 3.96 \mu\text{m}$  (E) in diam. Radial symmetry, apolar polarlity, prolate spheroidal shape. Exine sculpturing is echinate. The spine  $2.47 \pm 0.13 \mu\text{m}$  in length. Exine sculpturing echinate with regulate between the spines. The wall thickness  $2.81 \pm 1.11 \mu\text{m}$ . similar to Huang (1972) found that a pollen are has  $33 \times 40 - 46 \times 29 \mu\text{m}$  size. The pollen wall: exine about  $1 \mu\text{m}$  thick, psilate or granulate of *Alocasia macrorrhiza* (L.) Schott (Table 3.2, Figure 3.5).

***Alocasia* sp.**

Pollen grain is a monad and  $51.53 \pm 1.26 \mu\text{m}$  in diam. Radial symmetry, apolar polarlity, spheroidal shape. Exine sculpturing is echinate. The spine  $3.20 \pm 0.38 \mu\text{m}$  in length. Exine sculpturing echinate with regulate between the spines. The wall thickness  $2.59 \pm 1.56 \mu\text{m}$ . similar to Hay and Wise (1991) found that pollen are spheroidal or subspheroidal, inaperture, apolar, radiosymmetric, an exine sculpturing is spinose of genus *Alocasia* sp. (Table 3.2, Figure 3.6).

***Amorphophallus serrulatus* Hett. & A.Galloway**

Pollen grain is a monad and  $35.03 \pm 1.92 \mu\text{m}$  (P)  $38.81 \pm 2.61 \mu\text{m}$  (E) in diam. Bilateral symmetry, isopolar polarlity, diporate aperture, subprolate shape. Exine sculpturing is rugulate. The wall thickness  $5.48 \pm 1.69 \mu\text{m}$  (Table 3.2, Figure 3.7).

***Arisaema maxwellii* Hett. & Gusman**

Pollen grain is a monad and  $28.53 \pm 2.63 \mu\text{m}$  in diam. Radial symmetry, apolar polarlity, spheroidal shape. Exine sculpturing is echinate. The spine  $1.32 \pm 0.90 \mu\text{m}$  in length. Exine sculpturing echinate with regulate between the spines. The wall thickness  $1.56 \pm 1.01 \mu\text{m}$  (Table 3.2, Figure 3.8).

***Colocasia esculenta* (L.) Schott**

Pollen grain is a monad and  $29.06 \pm 1.74 \mu\text{m}$  in diam. Radial symmetry, apolar polarlity, spheroidal shape. Exine sculpturing is echinate. The spine  $2.69 \pm 0.36 \mu\text{m}$  in length. Exine sculpturing echinate with regulate between the spines. The wall thickness



2.63±0.32 µm similarity Erdtman (1971) found that pollen are having 1-sulcate and 20x25 µm sizes the pollen grain is a medium size of *Colocasia antiquorum* var. *esculenta* (Table 3.2, Figure 3.9).

***Colocasia fallax* Schott**

Pollen grain is a monad and 21.42±1.15 µm in diam. Radial symmetry, apolar polarity, spheroidal shape. Exine sculpturing is echinate. The spine 1.43±0.29 µm in length. Exine sculpturing echinate with regulate between the spines. The wall thickness 2.01±0.41 µm (Table 3.2, Figure 3.10).

***Colocasia gigantea* (Blume) Hook.f.**

Pollen grain is a monad and 26.90±1.11 µm (P) 28.46±1.26 µm (E) in diam. Radial symmetry, apolar polarity, prolate spheroidal shape. Exine sculpturing is echinate. The spine 3.66±0.68 µm in length. Exine sculpturing echinate with regulate between the spines. The wall thickness 2.57±0.20 µm (Table 3.2, Figure 3.11).

***Hapaline benthamiana* Schott**

Pollen grain is a monad and 41.93±2.05 µm in diam. Radial symmetry, apolar polarity, spheroidal shape. Exine sculpturing is echinate. The spine 1.99±0.14 µm in length. Exine sculpturing echinate with regulate between the spines. The wall thickness 2.41±0.29 µm (Table 3.2, Figure 3.12).

***Lasia spinosa* (L.) Thwaites**

Pollen grain is a monad and 17.31±1.26 µm (P) 21.71±0.87 µm (E) in diam. Bilateral symmetry, apolar polarity, subprolate shape. Exine sculpturing is rugulate. The wall thickness 1.37±0.29 µm (Table 3.2, Figure 3.13).

***Pistia stratiotes* L.**

Pollen grain is a monad and 26.79±1.76 µm (P) 45.05±4.49 µm (E) in diam. Bilateral symmetry, apolar polarity, prolate shape. Exine sculpturing is psilate. The wall thickness 1.93±0.45 µm (Table 3.2, Figure 3.14).

***Pycnospatha palmata* Gagnep.**

Pollen grain is a monad and 16.14±2.91 µm (P) 24.05±1.98 µm (E) in diam. Bilateral symmetry, heteropolar polarity, monosulcate aperture, prolate shape. Exine sculpturing is reticulate-perifoliate. The wall thickness 3.53±0.99 µm (Table 3.2, Figure 3.15).

***Schismatoglottis calyptrata* (Roxb.) Zoll. & Moritzi**

Pollen grain is a monad and 12.72±1.83 µm (P) 13.57±0.85 µm (E) in diam. Radial symmetry, apolar polarity, prolate spheroidal shape. Exine sculpturing is psilate. The wall thickness 1.18±1.60 µm (Table 3.2, Figure 3.16).



### ***Typhonium glaucum* Hett. & Sookchaloem**

Pollen grain is a monad and  $42.39 \pm 1.43$   $\mu\text{m}$  in diam. Radial symmetry, apolar polarity, spheroidal shape. Exine sculpturing is echinate. The spine  $1.84 \pm 0.20$   $\mu\text{m}$  in length. Exine sculpturing echinate with regulate between the spines. The wall thickness  $1.35 \pm 0.20$   $\mu\text{m}$  (Table 3.2, Figure 3.17).

### ***Typhonium trilobatum* (L.) Schott**

Pollen grain is a monad and  $36.54 \pm 1.24$   $\mu\text{m}$  in diam. Radial symmetry, apolar polarity, spheroidal shape. Exine sculpturing is scabrate. The spine  $0.93 \pm 0.90$   $\mu\text{m}$  in length. Exine sculpturing echinate with granulate between the spines. The wall thickness  $3.90 \pm 0.96$   $\mu\text{m}$  (Table 3.2, Figure 3.18).

## **3.8 Conclusion**

The result of pollen morphology show the 18 species plants Araceae family in Thailand by acetolysis applied method and study under light microscope and a scanning electron microscope.

Summary characters of 18 Thai Araceae species pollens: They are a monad and polarity is apolar. The different characteristics of the 18 species plants are a radial symmetry, aperture, shape, size, exine sculpturing, wall thickness and length of spine (Table 3.2). The above to classify pollen grain characteristics into the following groups.

#### 1. Thai Araceae pollen can be divided into tree groups based on pollen size.

1.1 The first group is small sized pollen including *Aglaonema modestum*, *Alocasia cucullata*, *Colocasia fallax*, *Lasia spinosa*, *Pycnosphata palmate* and *Scittomaglottis calyptrate*.

1.2 The second group is medium sized pollen including *Aglaonema simplex*, *Alocasia longiloba*, *Alocasia macrorrhizos*, *Amorphophallus serrulatus*, *Arisaema maxwellii*, *Colocasia esculenta*, *Colocasia gigantea*, *Hapaline benthamiana*, *Pistia stratiotes*, *Typhonium glaucum* and *Typhonium trilobatum*.

1.3 The third group is large sized is a *Alocasia* sp..

#### 2. Two pollen groups based on symmetry.

2.1 Bilateral symmetry includes *Amorphophallus serrulatus*, *Lasia spinosa*, *Pistia stratiotes* and *Pycnosphata palmata*.

2.2 Radial symmetry includes *Aglaonema modestum*, *Aglaonema simplex*, *Alocasia cucullata*, *Alocasia longiloba*, *Alocasia macrorrhizos*, *Alocasia* sp., *Arisaema maxwellii*, *Colocasia esculenta*, *Colocasia fallax*, *Colocasia gigantea*, *Hapaline benthamiana*, *Scittomaglottis calyptrate*, *Typhonium glaucum* and *Typhonium trilobatum*.

#### 3. Three groups of pollen by aperture.

3.1 Monoporate aperture group are an *Aglaonema modestum*.

3.2 Monosulcate aperture group is a *Pycnosphata palmata*.

3.3 Diporate aperture group is an *Amorphophallus serrulatus*.



4. Pollen of this study can be divided into five groups by shape.

4.1 The first group is spheroidal shape pollen including *Aglaonema modestum*, *Aglaonema simplex*, *Alocasia* sp., *Arisaema maxwellii*, *Colocasia esculenta*, *Colocasia fallax*, *Hapaline benthamiana*, *Typhonium glaucum* and *T. trilobatum*.

4.2 The second group is prolate shape pollen including *Alocacia cucullata*, *Colocasia gigantea* and *Scittomaglottis calyptrate*.

4.3 The third group is oblate spheroidal shape pollen is an *Alocacia longiloba*.

4.4 The fourth group is subprolate shape pollen including an *Amorphophallus serrulatus*, *Lasia spinosa*.

4.5 The fifth group is prolate shape pollen including *Pistia stratiotes* and *Pycnosphata palmata*.

5. Pollen of this study can be divided into six groups by exine sculpturing.

5.1 The first group is granulate exine sculpturing pollen is an *Aglaonema modestum*.

5.2 The second group is echinate exine sculpturing pollen including *Aglaonema simplex*, *Alocacia cucullata*, *Alocacia longiloba*, *Alocacia macrorrhizos*, *Alocasia* sp., *Arisaema maxwellii*, *Colocasia esculenta*, *C. fallax*, *C. gigantea*, *Hapaline benthamiana* and *Typhonium trilobatum*.

5.3 The third group is rugulate exine sculpturing pollen including *Amorphophallus serrulatus* and *Lasia spinosa*.

5.4 The fourth group is psilate exine sculpturing pollen including *Pistia stratiotes* and *Scittomaglottis calyptrate*.

5.5 The fifth group is reticulate-perifolate exine sculpturing pollen is a *Pycnosphata palmata*.

5.6 The sixth group is scabrate exine sculpturing pollen is a *Typhonium trilobatum*.

6. Pollen of this study found that the seventeen species with wall thickness pollen including *Aglaonema simplex*, *Alocacia cucullata*, *Alocacia longiloba*, *Alocacia macrorrhizos*, *Alocasia* sp., *Amorphophallus serrulatus*, *Arisaema maxwellii*, *Colocasia esculenta*, *Colocasia fallax*, *Colocasia gigantea*, *Hapaline benthamiana*, *Lasia spinosa*, *Pistia stratiotes*, *Pycnosphata palmata*, *Scittomaglottis calyptrata*, *Typhonium glaucum* and *T. trilobatum*.

7. Pollen of this study found that the twelve species with of spine pollen including *Aglaonema simplex*, *Alocacia cucullata*, *A. longiloba*, *A. sp.*, *Amorphophallus serrulatus*, *Arisaema maxwellii*, *Colocasia esculenta*, *C. fallax*, *C. gigantea*, *Hapaline benthamiana*, *Typhonium glaucum* and *T. trilobatum*.

The pollen grains of 16 species have been recorded for the first time as follows; *Aglaonema simplex*, *Alocacia cucullata*, *Alocacia longiloba*, *Alocasia* sp., *Amorphophallus serrulatus*, *Arisaema maxwellii*, *Colocasia fallax*, *Colocasia gigantea*, *Hapaline benthamiana*, *Lasia spinosa*, *Pistia stratiotes*, *Pycnosphata palmata*, *Scittomaglottis calyptrata*, *Typhonium glaucum* and *T. trilobatum*.

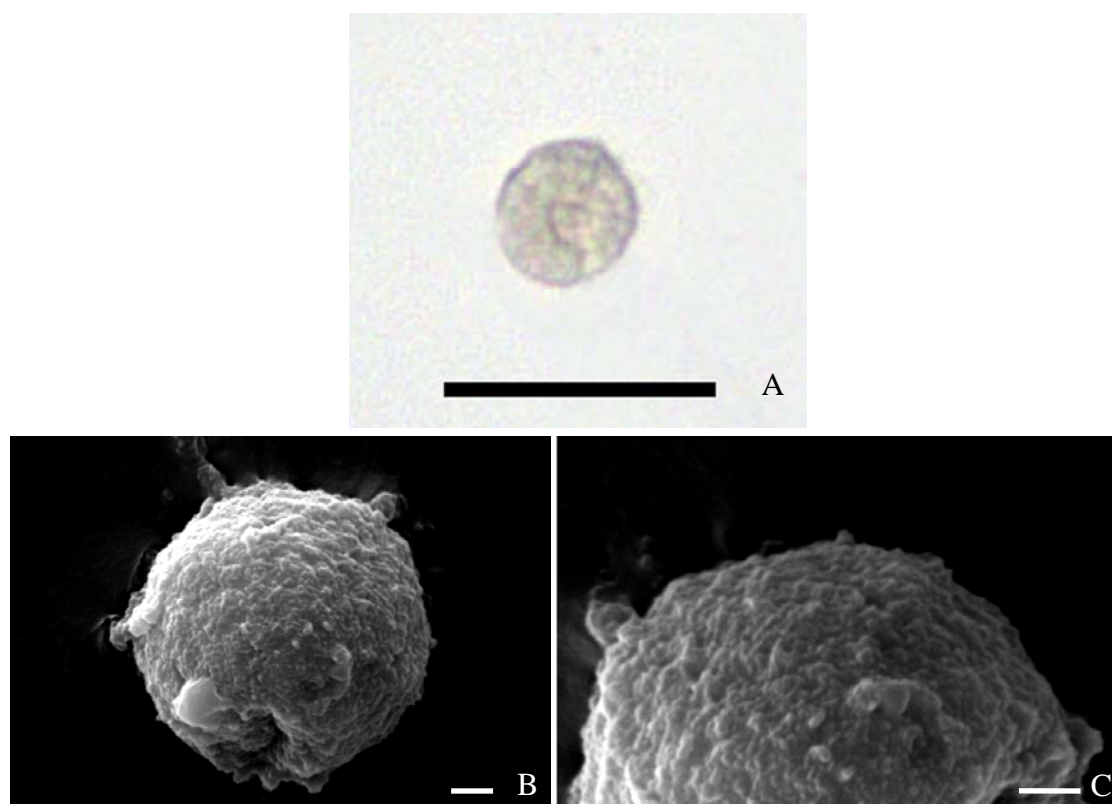


**Table 3.2** Pollen morphology of 18 species of Araceae in Thailand

Species	Symmetry	Polarity	Aperture	Shape (P/E)	Size ( $\mu\text{m}$ ) (M $\pm$ SD)		diameter ( $\mu\text{m}$ ) (M $\pm$ SD)	Size	Exine sculpturing	Wall thickness ( $\mu\text{m}$ ) (M $\pm$ SD)	Length of spine ( $\mu\text{m}$ ) (M $\pm$ SD)
					P	E					
1. <i>Agloanema modestum</i>	radial	hetero-polar	mp	spheroidal	-	-	11.40 $\pm$ 1.22	S	granulate	-	-
2. <i>A. simplex</i>	radial	apolar	n	spheroidal	-	-	50.27 $\pm$ 4.84	M	echinate	7.07 $\pm$ 1.74	3.31 $\pm$ 0.36
3. <i>Alocacia macrorrhizos</i>	radial	apolar	n	prolate spheroidal	22.31 $\pm$ 2.18	24.64 $\pm$ 2.79	-	S	echinate	2.35 $\pm$ 0.25	1.36 $\pm$ 0.90
4. <i>A. cucullata</i>	radial	apolar	n	oblate spheroidal	35.87 $\pm$ 2.27	37.02 $\pm$ 7.28	-	M	echinate	2.78 $\pm$ 0.53	3.02 $\pm$ 0.33
5. <i>A. longiloba</i>	radial	apolar	n	prolate spheroidal	36.85 $\pm$ 3.72	39.20 $\pm$ 3.96	-	M	echinate	2.81 $\pm$ 1.11	2.47 $\pm$ 0.13
6. <i>A. sp.</i>	radial	apolar	n	spheroidal	-	-	51.53 $\pm$ 1.26	L	echinate	2.59 $\pm$ 1.56	3.20 $\pm$ 0.38
7. <i>Amorphophallus serrulatus</i>	bilateral	isopolar	dp	subprolate	35.03 $\pm$ 1.92	38.81 $\pm$ 2.61	-	M	rugulate	5.48 $\pm$ 1.69	-
8. <i>Arisaema maxwellii</i>	radial	apolar	n	spheroidal	-	-	28.53 $\pm$ 2.63	M	echinate	1.56 $\pm$ 1.01	1.32 $\pm$ 0.90
9. <i>Colocasia esculenta</i>	radial	apolar	n	spheroidal	-	-	29.06 $\pm$ 1.74	M	echinate	2.63 $\pm$ 0.32	2.69 $\pm$ 0.36
10. <i>C. fallax</i>	radial	apolar	n	spheroidal	-	-	21.42 $\pm$ 1.15	S	echinate	2.01 $\pm$ 0.41	1.43 $\pm$ 0.29
11. <i>C. gigantea</i>	radial	apolar	n	prolate spheroidal	26.90 $\pm$ 1.11	28.46 $\pm$ 1.26	-	M	echinate	2.57 $\pm$ 0.20	3.66 $\pm$ 0.68
12. <i>Hapaline benthamiana</i>	radial	apolar	n	spheroidal	-	-	41.93 $\pm$ 2.05	M	echinate	2.41 $\pm$ 0.29	1.99 $\pm$ 0.14
13. <i>Lasia spinosa</i>	bilateral	apolar	n	subprolate	17.31 $\pm$ 1.26	21.71 $\pm$ 0.87	-	S	rugulate	1.37 $\pm$ 0.29	-
14. <i>Pistia stratiotes</i>	bilateral	apolar	n	prolate	26.79 $\pm$ 1.76	45.05 $\pm$ 4.49	-	M	psilate	1.93 $\pm$ 0.45	-
15. <i>Pycnosphata palmata</i>	bilateral	hetero-polar	ms	prolate	16.14 $\pm$ 2.91	24.05 $\pm$ 1.98	-	S	reticulate-perifoliate	3.53 $\pm$ 0.99	-
16. <i>Scittomaglottis calyptrata</i>	radial	apolar	n	prolate spheroidal	12.72 $\pm$ 1.83	13.57 $\pm$ 0.85	-	S	psilate	1.18 $\pm$ 1.60	-
17. <i>Typhonium glaucum</i>	radial	apolar	n	spheroidal	-	-	42.39 $\pm$ 1.43	M	echinate	1.35 $\pm$ 0.20	1.84 $\pm$ 0.20
18. <i>T. trilobatum</i>	radial	apolar	n	spheroidal	-	-	36.54 $\pm$ 1.24	M	scabrate	3.90 $\pm$ 0.96	0.93 $\pm$ 0.90

Note: E= Equatorial view, M= Medium, L = Large, P= Polar view, S = Small, dp = diporate, ms = monosulcate, mp = monoporate, n = non-aperture, - = non





**Figure 3.1** Light micrograph and scanning electron micrograph of pollen morphology of *Aglaonema modestum* Schott ex Engl.

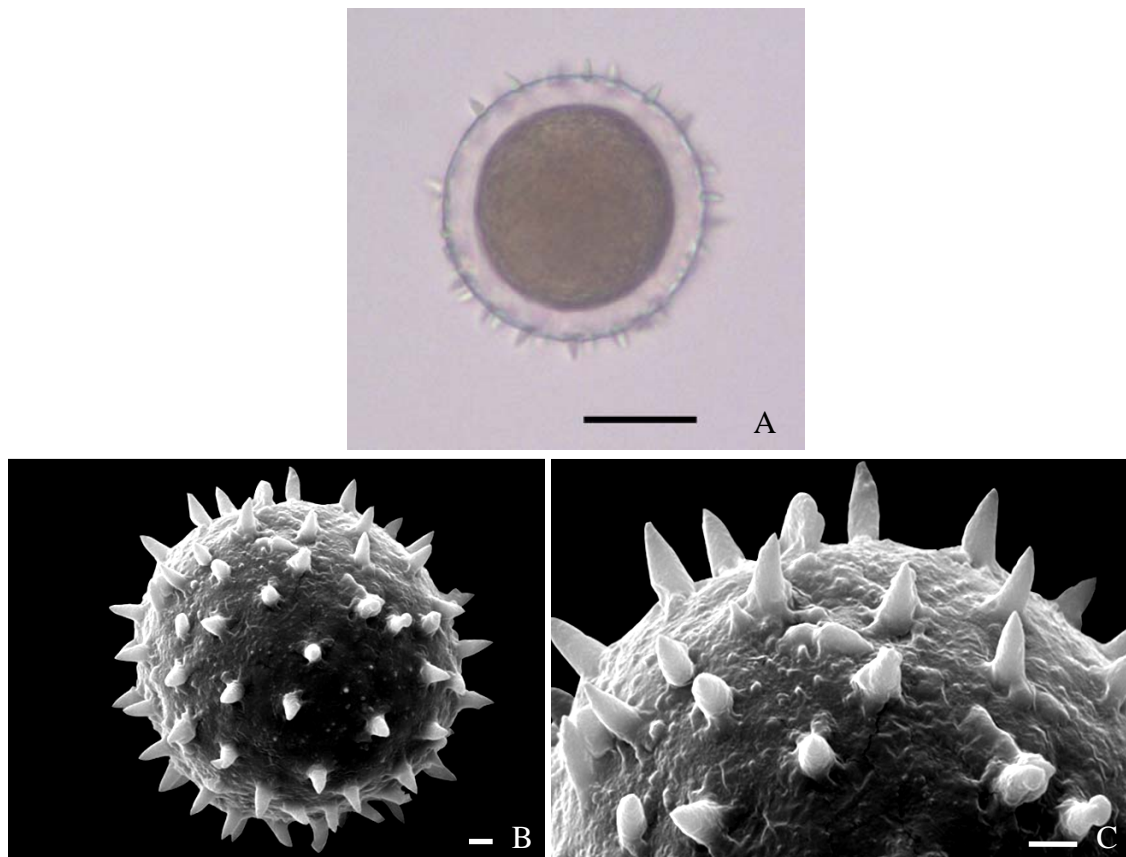
A. Pollen grain (LM) scale bars = 20  $\mu\text{m}$

B. Pollen grain (SEM) scale bars = 1  $\mu\text{m}$

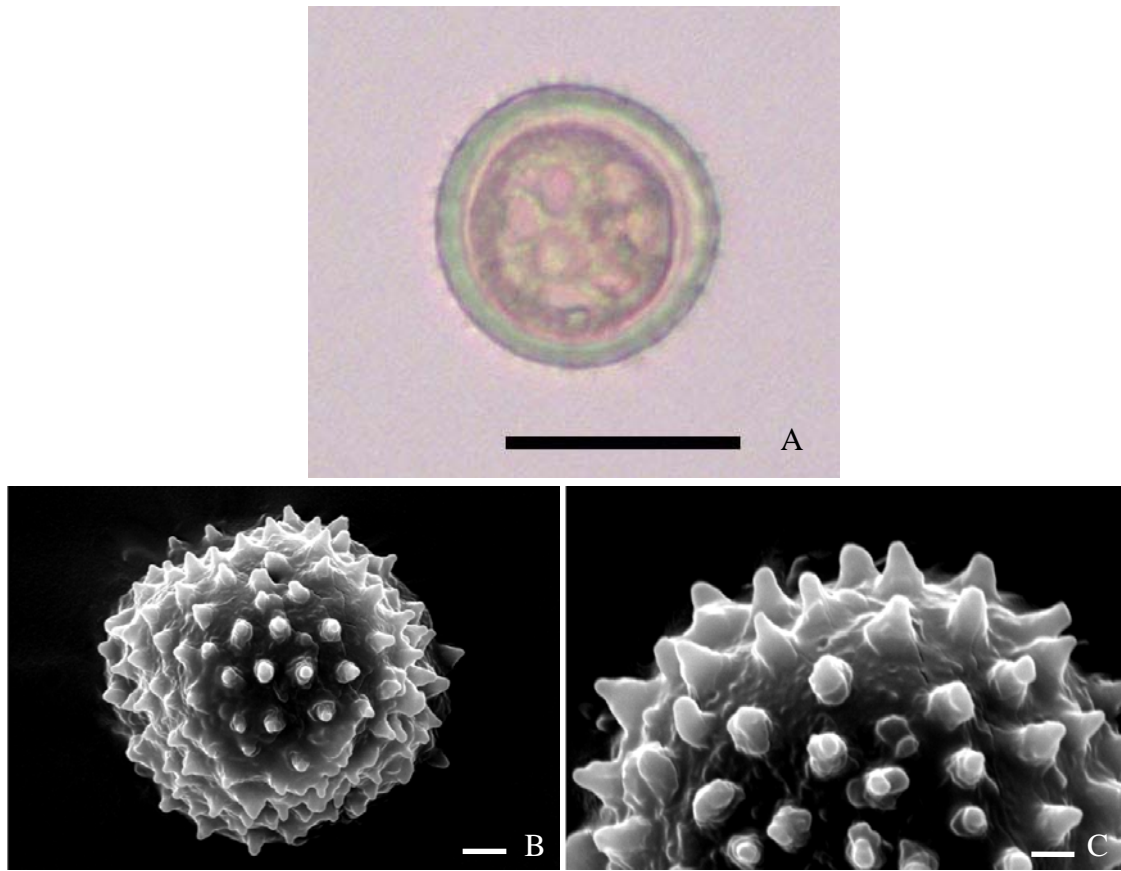
C. Exine sculpturing (SEM) scale bars = 1  $\mu\text{m}$



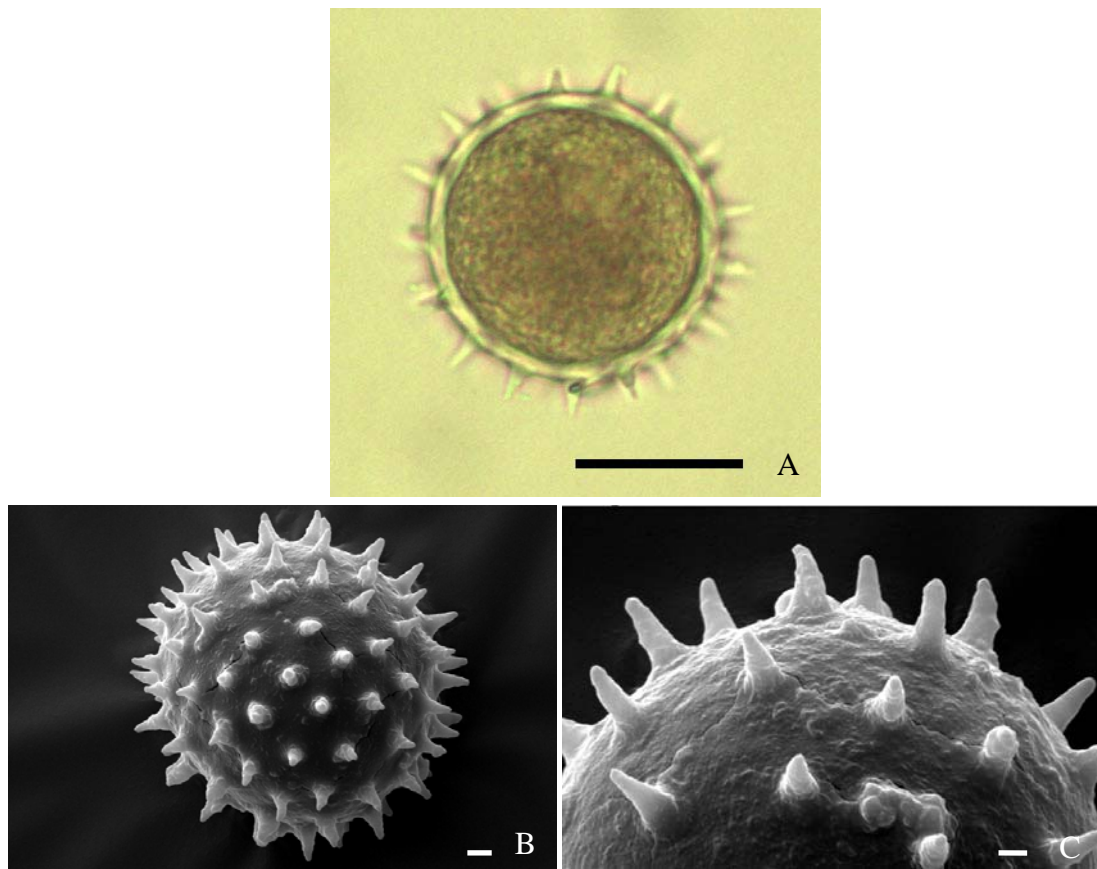




**Figure 3.2** Light micrograph and scanning electron micrograph of pollen morphology of *Aglaonema simplex* (Blume) Blume  
A. Pollen grain (LM) scale bars = 20  $\mu\text{m}$   
B. Pollen grain (SEM) scale bar = 2  $\mu\text{m}$   
C. Exine sculpturing (SEM) scale bar = 2  $\mu\text{m}$



**Figure 3.3** Light micrograph and scanning electron micrograph of pollen morphology of *Alocasia cucullata* (Lour.) G.Don  
 A. Pollen grain (LM) scale bar = 20  $\mu\text{m}$   
 B. Pollen grain (SEM) scale bar = 2  $\mu\text{m}$   
 C. Exine sculpturing (SEM) scale bar = 1  $\mu\text{m}$

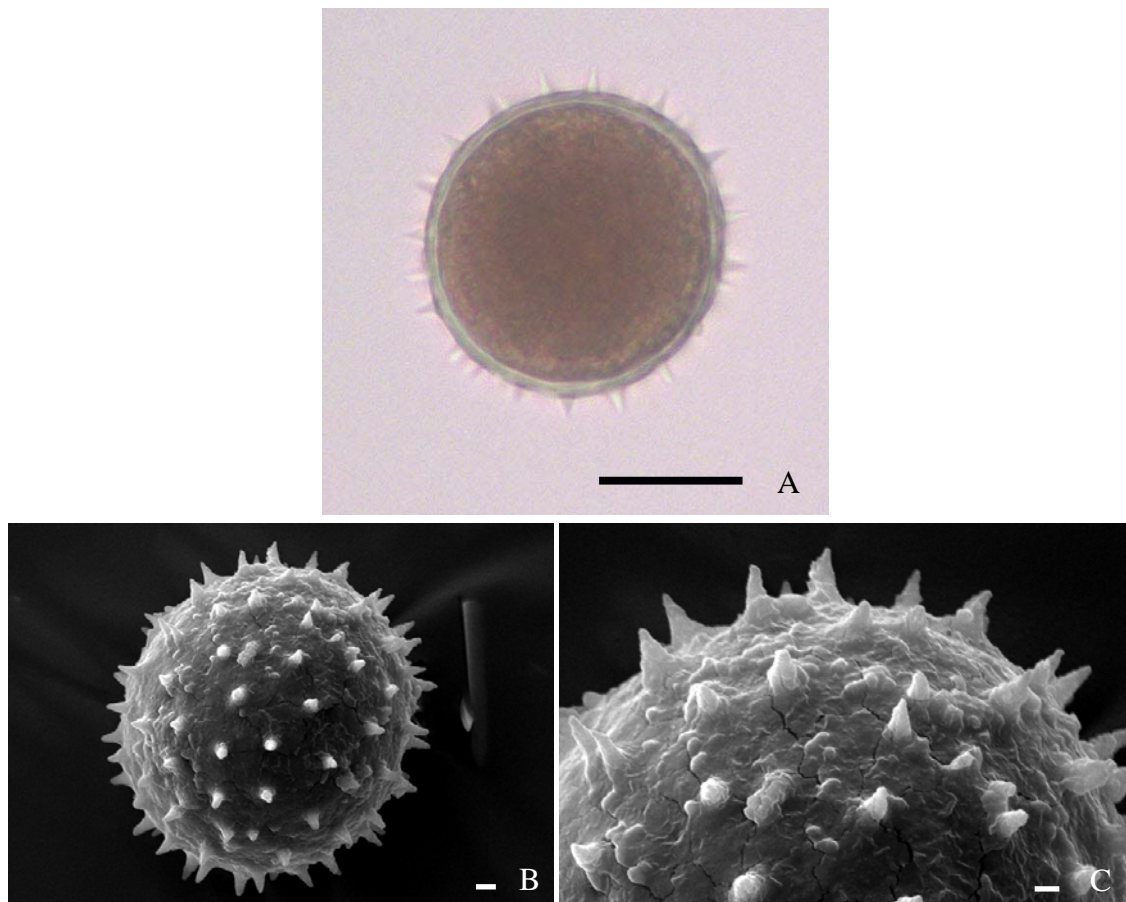


**Figure 3.4** Light micrograph and scanning electron micrograph of pollen morphology of *Alocasia longiloba* Miq.

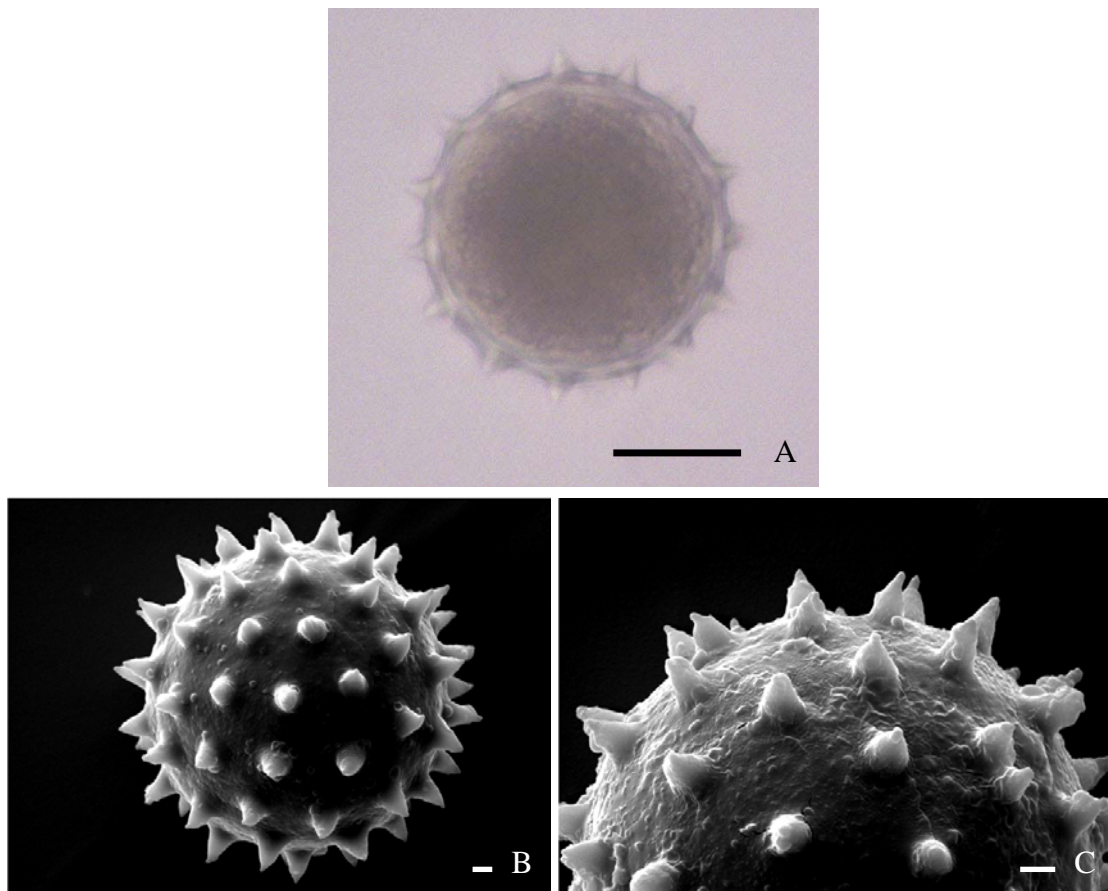
A. Pollen grain (LM) scale bar = 20  $\mu\text{m}$

B. Pollen grain (SEM) scale bar = 2  $\mu\text{m}$

C. Exine sculpturing (SEM) scale bar = 1  $\mu\text{m}$



**Figure 3.5** Light micrograph and scanning electron micrograph of pollen morphology of *Alocasia macrorrhizos* (L.) G. Don  
A. Pollen grain (LM) scale bar = 20  $\mu\text{m}$   
B. Pollen grain (SEM) scale bar = 2  $\mu\text{m}$   
C. Exine sculpturing (SEM) scale bar = 1  $\mu\text{m}$



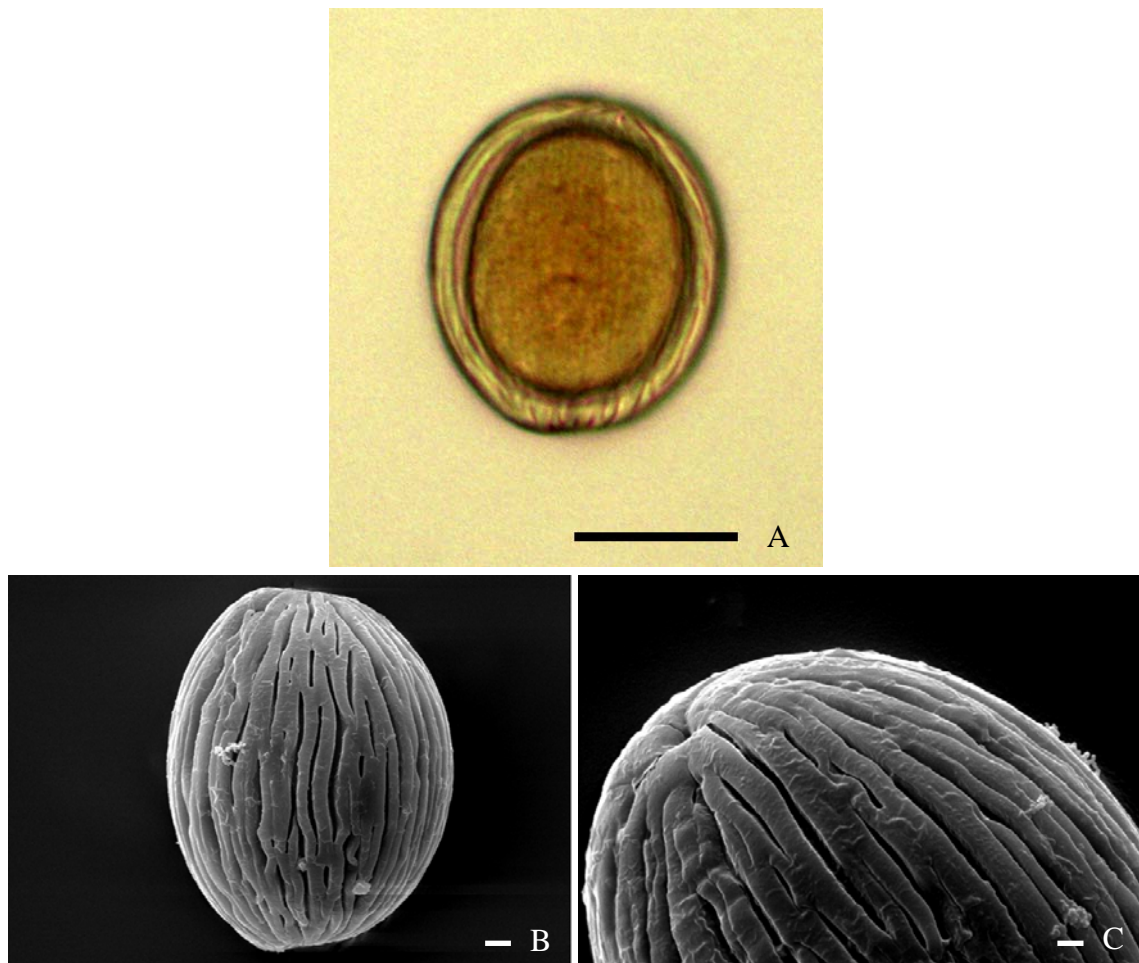
**Figure 3.6** Light micrograph and scanning electron micrograph of pollen morphology of *Alocasia* sp.

A. Pollen grain (LM) scale bar = 20  $\mu\text{m}$

B. Pollen grain (SEM) scale bar = 2  $\mu\text{m}$

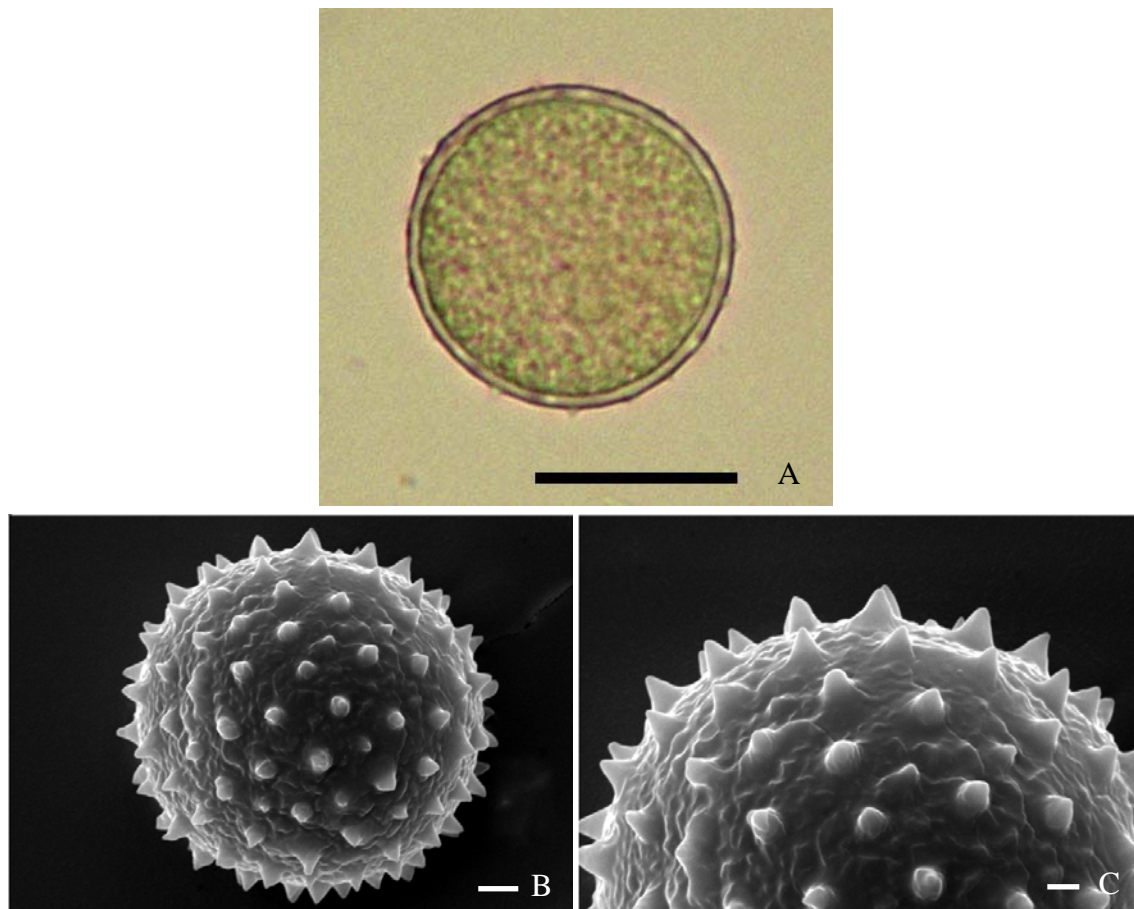
C. Exine sculpturing (SEM) scale bar = 2  $\mu\text{m}$



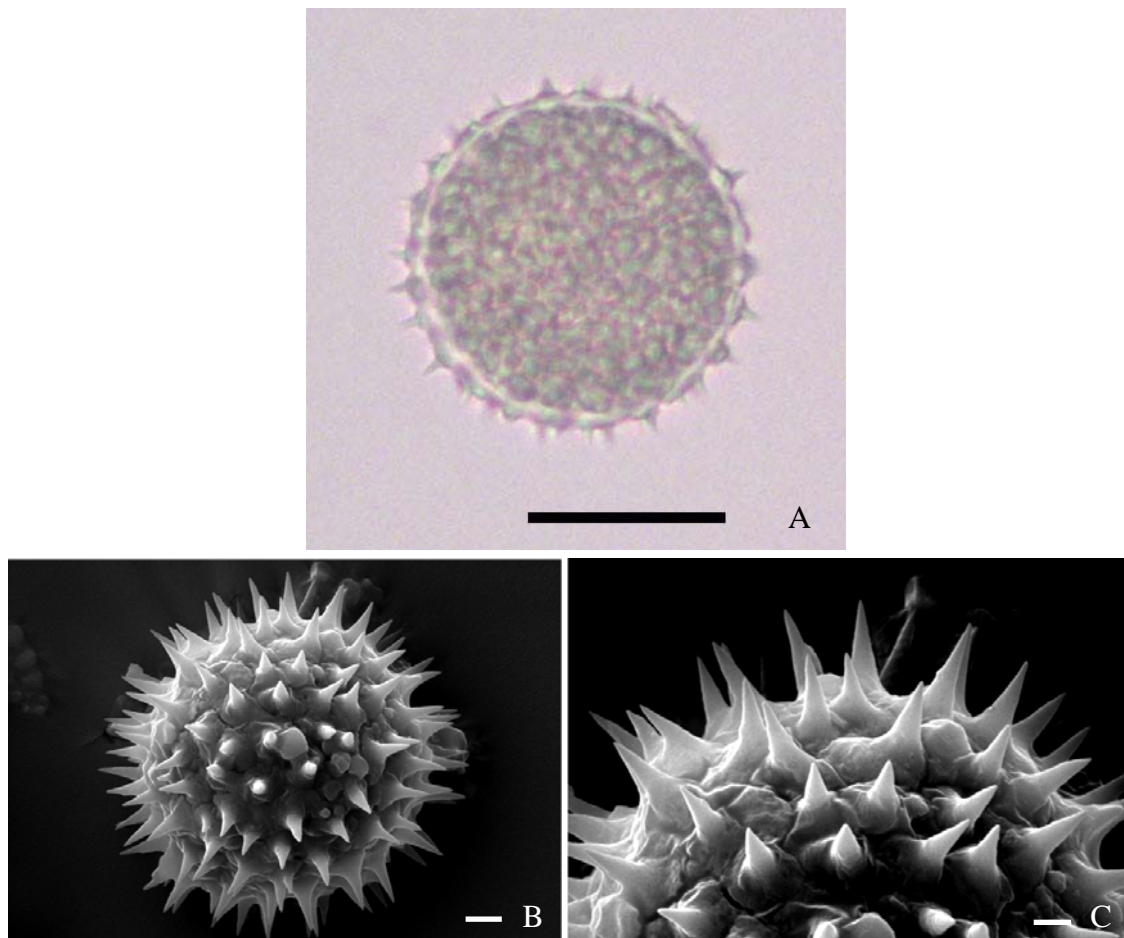


**Figure 3.7** Light micrograph and scanning electron micrograph of pollen morphology of *Amorphophallus serrulatus* Hett. & A.Galloway  
 A. Pollen grain (LM) scale bar = 20  $\mu\text{m}$   
 B. Pollen grain (SEM) scale bar = 2  $\mu\text{m}$   
 C. Exine sculpturing (SEM) scale bar = 1  $\mu\text{m}$

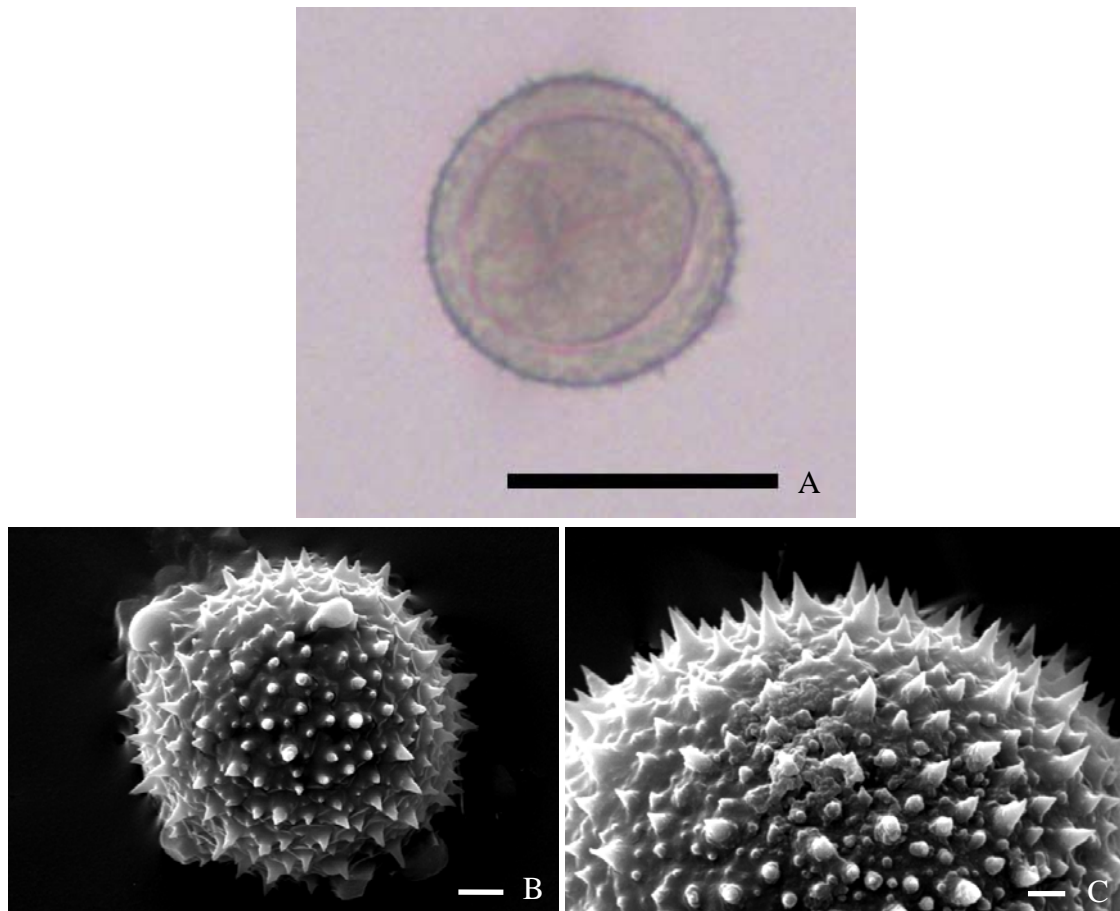




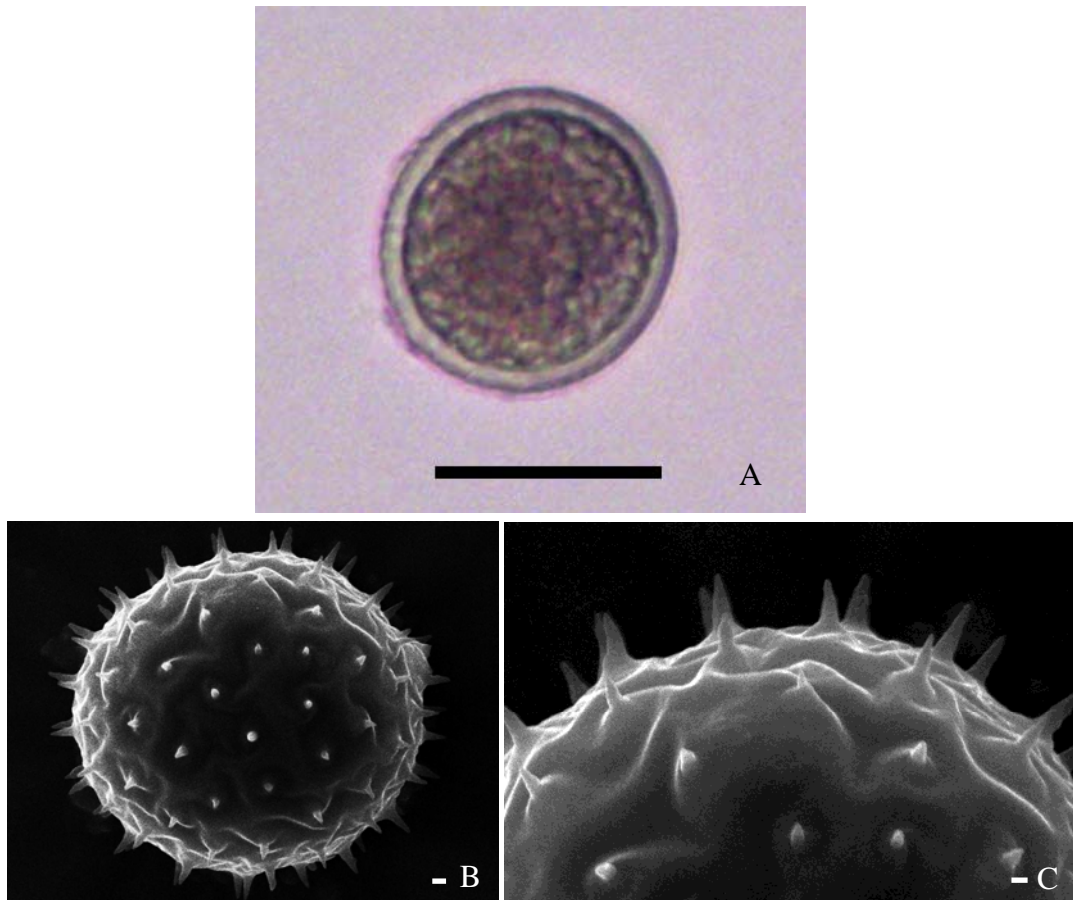
**Figure 3.8** Light micrograph and scanning electron micrograph of pollen morphology of *Arisaema maxwellii* Hett. & Gusman  
A. Pollen grain (LM) scale bar = 20  $\mu\text{m}$   
B. Pollen grain (SEM) scale bar = 2  $\mu\text{m}$   
C. Exine sculpturing (SEM) scale bar = 1  $\mu\text{m}$



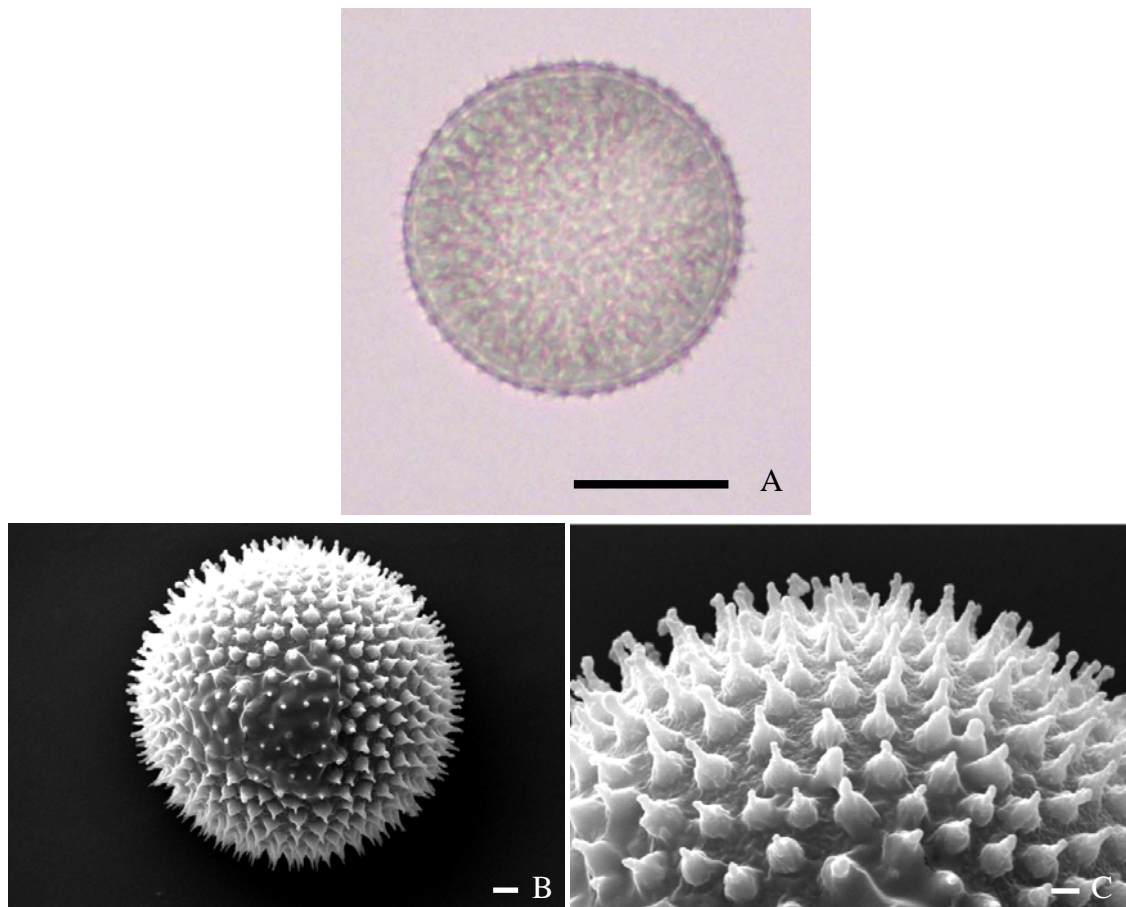
**Figure 3.9** Light micrograph and scanning electron micrograph of pollen morphology of *Colocasia esculenta* (L.) Schott  
A. Pollen grain (LM) scale bar = 20  $\mu\text{m}$   
B. Pollen grain (SEM) scale bar = 2  $\mu\text{m}$   
C. Exine sculpturing (SEM) scale bar = 1  $\mu\text{m}$



**Figure 3.10** Light micrograph and scanning electron micrograph of pollen morphology of *Colocasia fallax* Schott  
A. Pollen grain (LM) scale bar = 20  $\mu\text{m}$   
B. Pollen grain (SEM) scale bar = 2  $\mu\text{m}$   
C. Exine sculpturing (SEM) scale bar = 1  $\mu\text{m}$

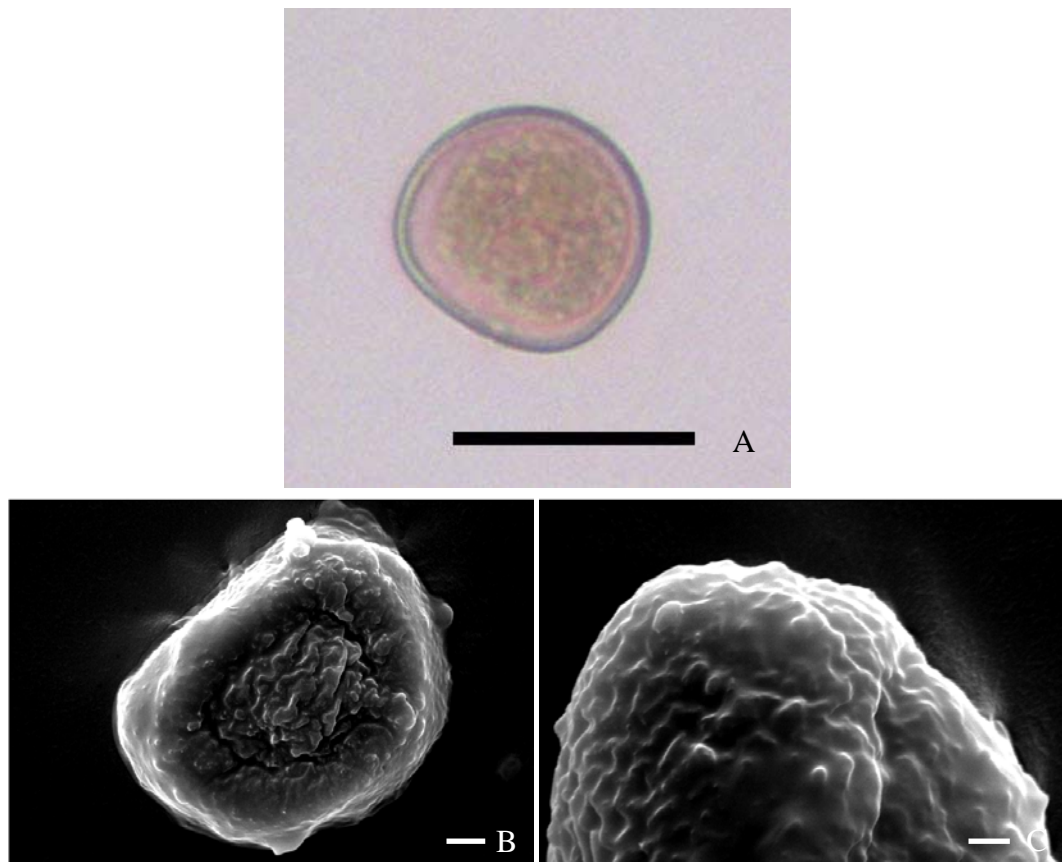


**Figure 3.11** Light micrograph and scanning electron micrograph of pollen morphology of *Colocasia gigantea* (Blume) Hook.f.  
A. Pollen grain (LM) scale bar = 20  $\mu\text{m}$   
B. Pollen grain (SEM) scale bar = 2  $\mu\text{m}$   
C. Exine sculpturing (SEM) scale bar = 1  $\mu\text{m}$



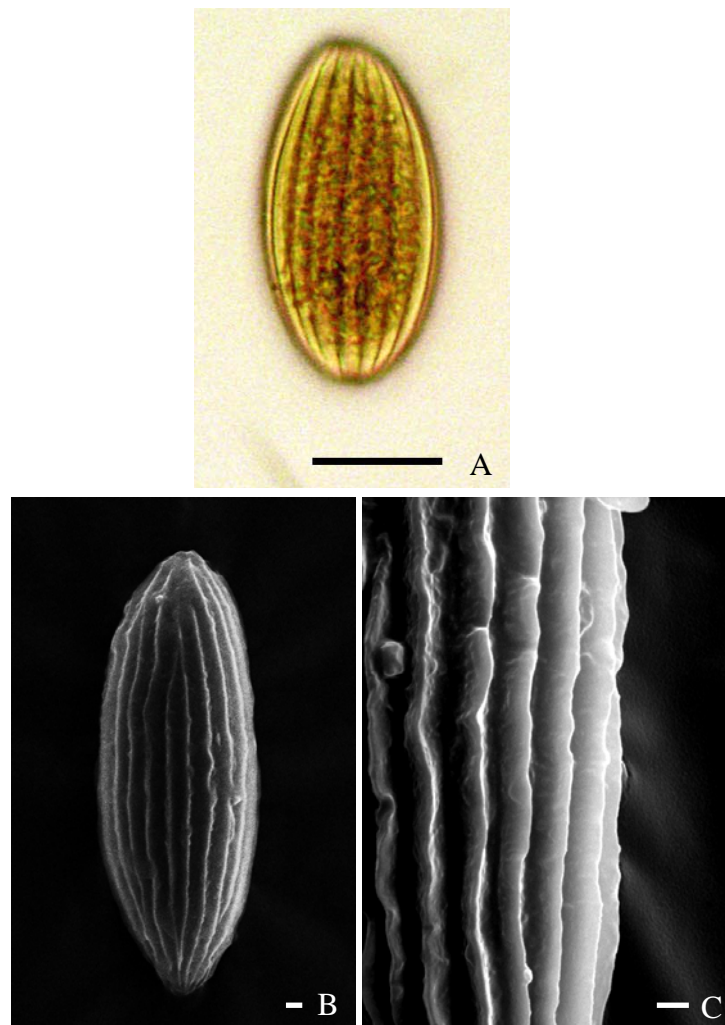
**Figure 3.12** Light micrograph and scanning electron micrograph of pollen morphology of *Hapaline benthamiana* Schott  
A. Pollen grain (LM) scale bar = 20  $\mu\text{m}$   
B. Pollen grain (SEM) scale bar = 2  $\mu\text{m}$   
C. Exine sculpturing (SEM) scale bar = 1  $\mu\text{m}$





**Figure 3.13** Light micrograph and scanning electron micrograph of pollen morphology of *Lasia spinosa* (L.) Thwaites  
A. Pollen grain (LM) scale bar = 20  $\mu\text{m}$   
B. Pollen grain (SEM) scale bar = 2  $\mu\text{m}$   
C. Exine sculpturing (SEM) scale bar = 1  $\mu\text{m}$





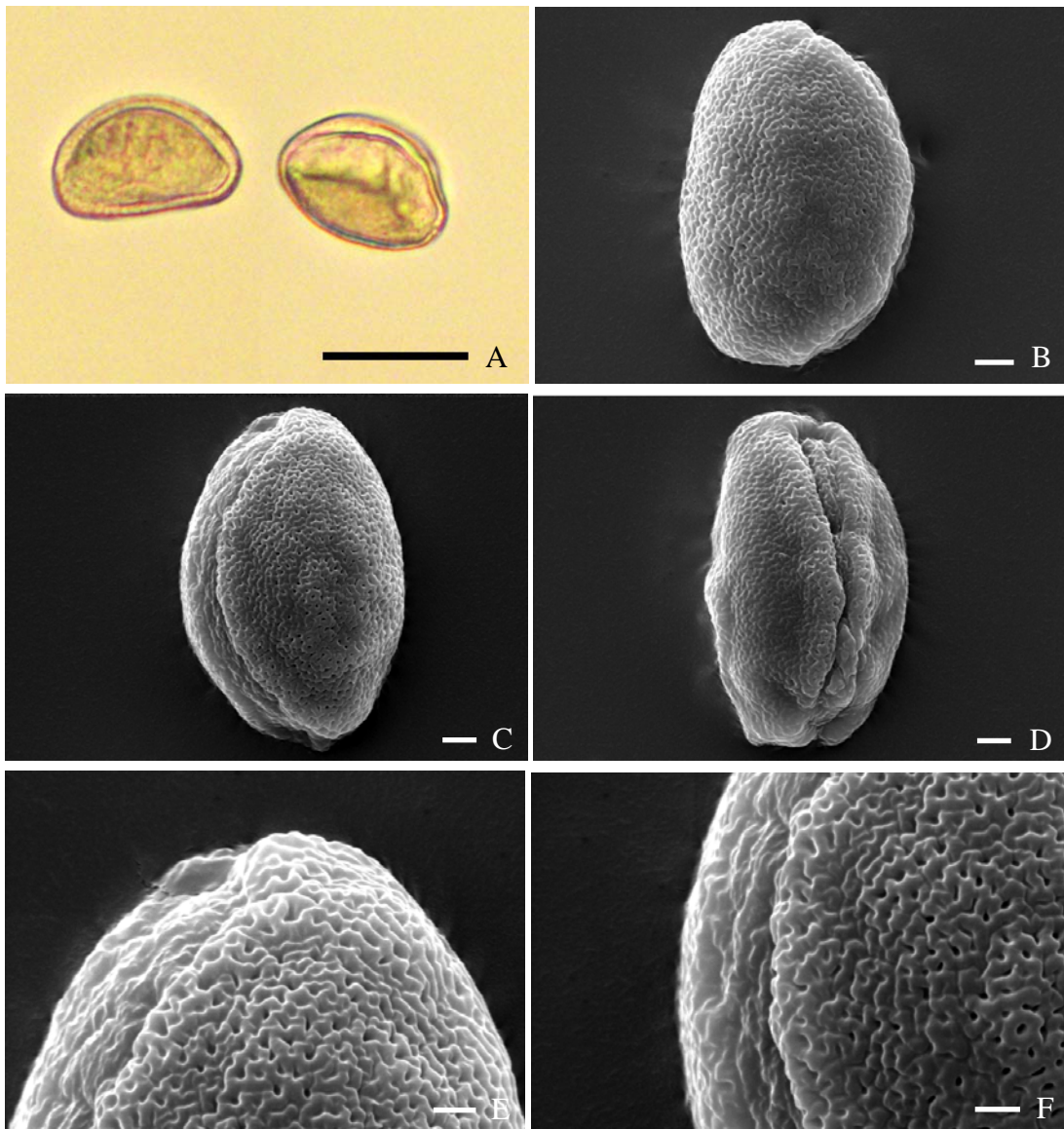
**Figure 3.14** Light micrograph and scanning electron micrograph of pollen morphology of *Pistia stratiotes* L.

A. Pollen grain (LM) scale bar = 20  $\mu\text{m}$

B. Pollen grain (SEM) scale bar = 2  $\mu\text{m}$

C. Exine sculpturing (SEM) scale bar = 1  $\mu\text{m}$





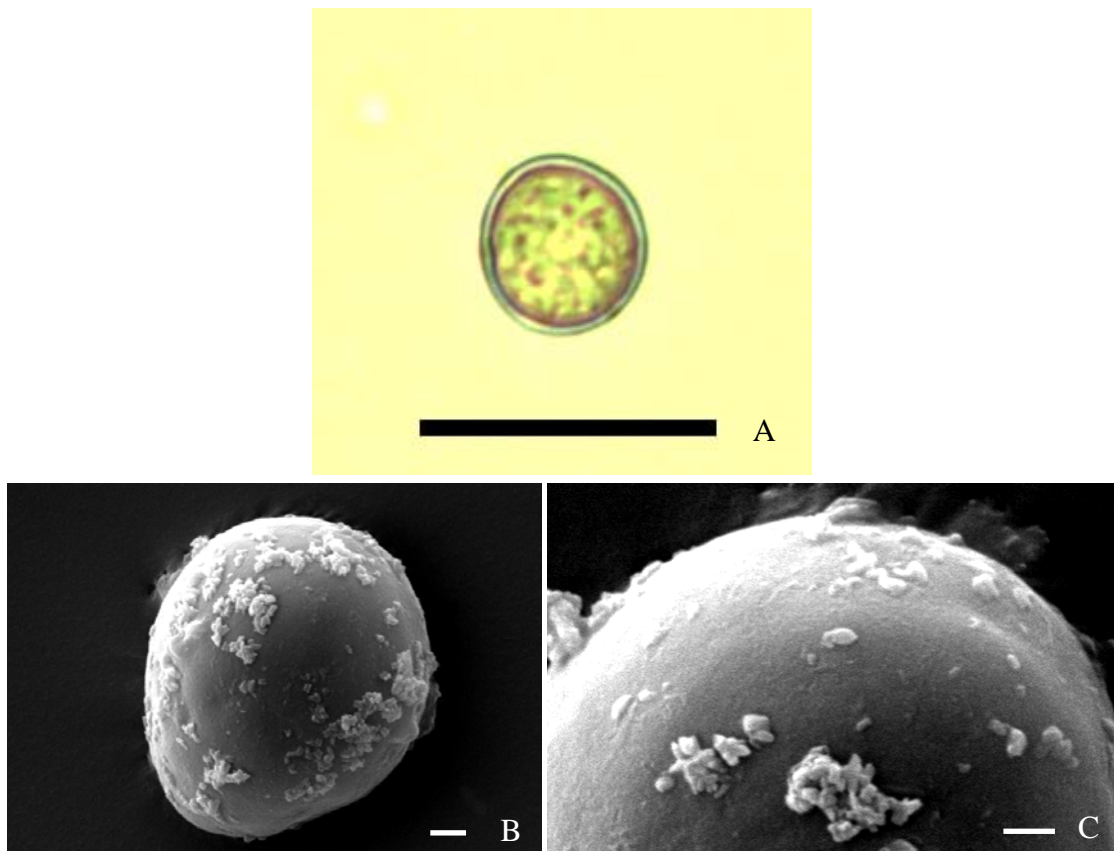
**Figure 3.15** Light micrograph and scanning electron micrograph of pollen morphology of *Pycnospatha palmata* Gagnep.

A. Pollen grain (LM) scale bar = 20  $\mu\text{m}$

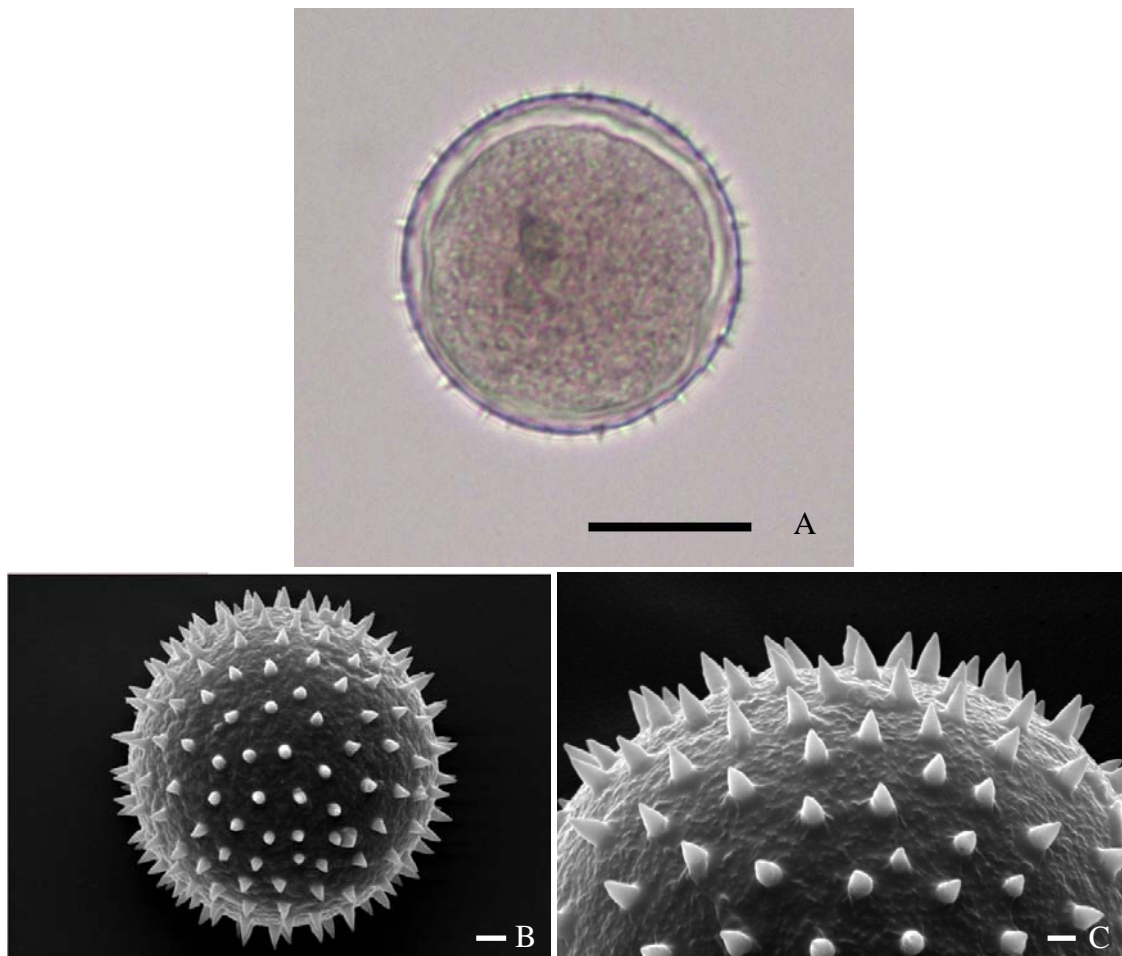
B-D. Pollen grain (SEM) scale bars = 2  $\mu\text{m}$

E-F. Exine sculpturing (SEM) scale bars = 1  $\mu\text{m}$

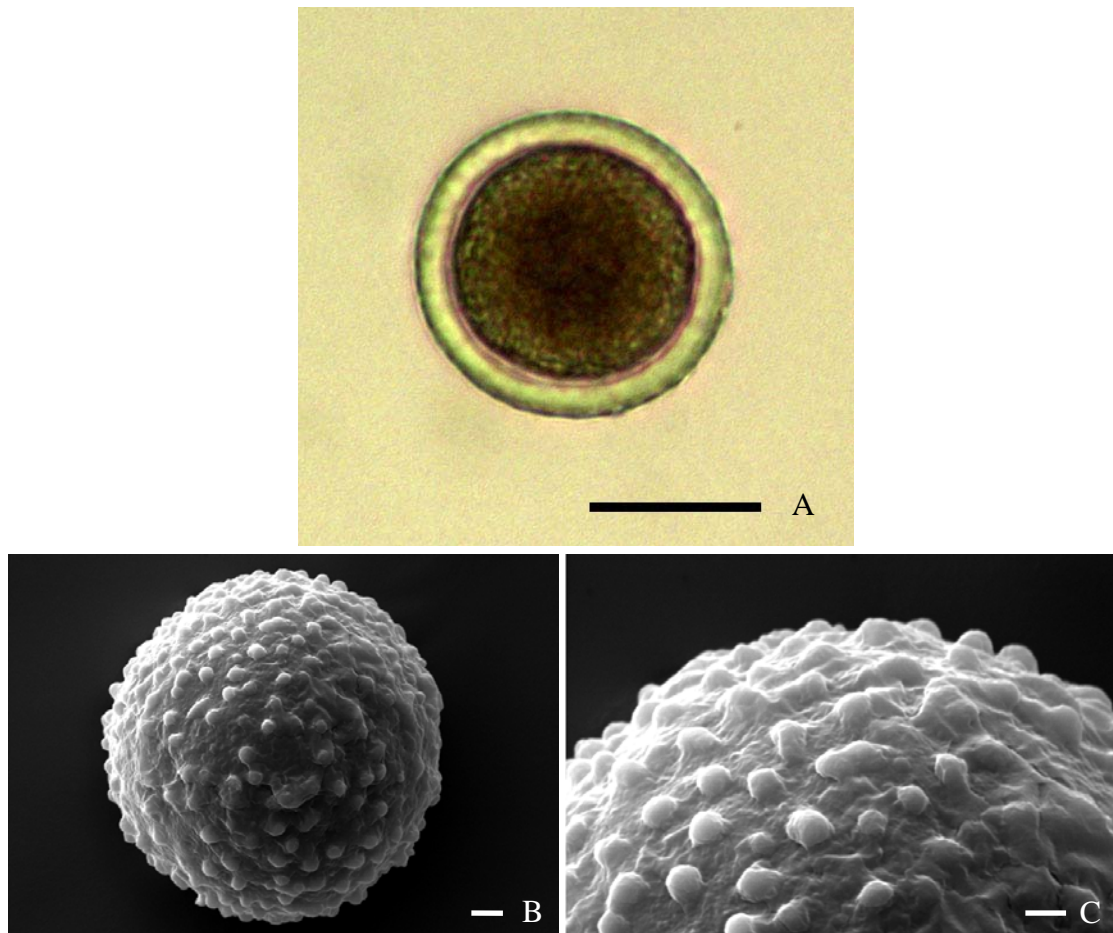




**Figure 3.16** Light micrograph and scanning electron micrograph of pollen morphology of *Schismatoglottis calyptrata* (Roxb.) Zoll & Moritzi  
A. Pollen grain (LM) scale bar = 20  $\mu\text{m}$   
B. Pollen grain (SEM) scale bar = 1  $\mu\text{m}$   
C. Exine sculpturing (SEM) scale bar = 1  $\mu\text{m}$



**Figure 3.17** Light micrograph and scanning electron micrograph of pollen morphology of *Typhonium glaucum* Hett. & Sookchaloem  
A. Pollen grain (LM) scale bar = 20  $\mu\text{m}$   
B. Pollen grain (SEM) scale bar = 2  $\mu\text{m}$   
C. Exine sculpturing (SEM) scale bar = 1  $\mu\text{m}$



**Figure 3.18** Light micrograph and scanning electron micrograph of pollen morphology of *Typhonium trilobatum* (L.) Schott  
A. Pollen grain (LM) scale bar = 20  $\mu\text{m}$   
B. Pollen grain (SEM) scale bar = 2  $\mu\text{m}$   
C. Exine sculpturing (SEM) scale bar = 1  $\mu\text{m}$



## CHAPTER 4

### ANATOMY STUDIES

#### 4.1 Introduction

Anatomy, or the internal form and structure of plant organs, is once techniques supported in plant classification and identification. Anatomical data are often extremely useful in solving problems of relationships because they can often suggest with greater confidence the homologies of morphological character states, and they can help in the interpretation of evolutionary directionality (Stuessy, 1990). In addition, this applies in confirming the identification of economic plant products ranging from timbers to food-stuffs as well as crude drugs of vegetable origin and natural plant fiber. Moreover, there are numerous problems in forensic science that can be solved by the histological methods (Metcalf, 1954).

#### 4.2 Leaf anatomy

Correct botanical identity based on the external morphology is possible when a complete plant specimen is available. Anatomical characters can also help the identification when morphological features are indistinct (David *et al.*, 2008).

##### 4.2.1 Literature reviews of anatomy

Sungkajanttranon *et al.* (2010) studied on anatomical characteristics of 10 genera 15 species in family Araceae which were surveyed in Chaleamrattanakosin National Park, Kanchanaburi province during November 27-28, 2008 and May 15-16, 2009 such as; *Aglaonema simplex* Blume, *Amorphophallus cruddasianus* Prain ex Engl., *Amorphophallus muelleri* Blume, *Amorphophallus myosuroides* Barnes & Fischer, *Amorphophallus paeoniifolius* (Dennst.) Nicolson, *Amorphophallus saraburiensis* Gagnep., *Alocasia lowii* Hook.f., *Arisaema putii* Gagnep., *Colocasia esculenta* (L.) Schott, *Colocasia gigantea* (Blume) Hook.f., *Cyrtosperma johnstoni* N.E.Br., *Hapaline kerrii* Gagnep., *Homalomena* sp., *Lasia spinosa* L. and *Philodendron* sp. The results found that root vascular bundles were polyarch and stem were atactostele which were monocotyledon type. Cell types and shapes in tissues, raphid and druse crystal in idioblast and laticifer, pigments in cell and tissues were different from species to species.

Boontang *et al.* (2011) studied anatomy of leaf epidermis of 10 species in eight genera of Araceae in Thailand: *Aglaonema modestum* Schott, *Caladium bicolor* Vent., *Dieffenbachia seguine* (Jacq.) Schott, *Homalomena rubescens* Kunth., *Philodendron domesticum* G. S. Bunting, *P. erubescens* K. Koch & Augustin, *P. selloum* Schott, *Scindapsus aureus* Engl., *Spathiphyllum clevelandii* Schott and *Syngonium podophyllum* Schott were studied by using epidermal peeling and leaf clearing techniques. The results indicated that the anatomical characters of epidermal surface such as epidermal cell shapes, anticlinal cell walls, and the presence of serulate cutin, stomatal types and distributions as well as the presence of druse crystals.

Sookchaloem *et al.* (2017) studied 23 species of *Amorphophallus* Blume ex Decne. in Thailand. The results showed the different anatomical characteristics of each





species. The upper and lower epidermal cell walls had three subtypes straight-sided, undulate or sinuous anticlinal. Both sides of the epidermal cell wall can be similar or can vary in each species. There were 1, 2, 3, 4 or 6 subsidiary cells along both sides of paired guard cells and the stomatal type was paracytic and stomatal subtypes varied from species to species, being brachyparacytic, hemiparacytic, amphibrachyparacytic, paratetracytic or parahexacytic. The stomatal number was 16-104/mm<sup>2</sup> of leaf area and varied with the leaf gloss and leaf texture of each species.

Therefore, many species will be study for the first time.

### 4.3 Plants material

All species of Araceae were collected from different parts of Thai forests or cultivated plant and transplanted in the nursery of Walai Rukhavej Botanical Research Institute, Mahasarakham University. Voucher specimens were deposited at the Mahasarakham University Herbarium (Table 4.1).

**Table 4.1** Plants used in the study

Species	Common name	Location	Collector number
1. <i>Agloanema modestum</i>	Khiao muen pi	Loei	R. Senavongse 001/2016
2. <i>A. simplex</i>	Wan ngot hin	Kalasin	R. Senavongse 006/2016
3. <i>Alocacia macrorrhizos</i>	Wan nang-kwak	Mukdahan	R. Senavongse 011/2016
4. <i>A. cucullata</i>	Kacho nok	Songkhla	R. Senavongse 016/2016
5. <i>A. longiloba</i>	Kradat	Mahasarakham	R. Senavongse 021/2016
6. <i>A. sp.</i>	-	Changmai	R. Senavongse 026/2016
7. <i>Amorphophallus serrulatus</i>	-	Ubon Ratchathani	R. Senavongse 031/2016
8. <i>Arisaema maxwellii</i>	-	Changmai	R. Senavongse 036/2016
9. <i>Colocasia esculenta</i>	Phueak or Bon	Chaiyaphum	R. Senavongse 016/2015
10. <i>C. fallax</i>	Bon-Doi	Changmai	R. Senavongse 041/2016
11. <i>C. gigantea</i>	Khun	Lampang	R. Senavongse 046/2016
12. <i>C. lihengiae</i>	Bon-Yunnan	Tak	R. Senavongse 051/2016
13. <i>Hapaline benthamiana</i>	Bon tao	Chaiyaphum	R. Senavongse 056/2016
14. <i>Homalonema griffithii</i>	Bon khiao	Songkhla	R. Senavongse 061/2016
15. <i>Lasia spinosa</i>	Phuk-Nam	Roi-Et	R. Senavongse 066/2016
16. <i>Pistia stratiotes</i>	Jok	Mahasarakham	R. Senavongse 071/2016
17. <i>Pycnosphata palmata</i>	-	Ubon Ratchathani	R. Senavongse 076/2016
18. <i>Scittomaglottis calyptrata</i>	Bon khiao	Trang	R. Senavongse 081/2016
19. <i>Typhonium glaucum</i>	-	Ubon Ratchathani	R. Senavongse 086/2016
20. <i>T. trilobatum</i>	Uttapit	Chumphon	R. Senavongse 091/2016

Note: sp. = species

### 4.4 Materials

1. Pot
2. Planting soil
3. Bottles for sampling roots
4. Forceps
5. Slide Warmers
6. Slide and cover glass
7. Beaker
8. Petri dish



9. Dropper
10. Razor blade
11. Permanent pen
12. Blotting paper
13. Bottles for soak color
14. Transparent Color Nail Polish
15. Light microscope (Zeiss: Axiostar plus)
16. Distilled water
17. Glacial acetic acid
18. Absolute alcohol
19. 15% ethyl alcohol
20. 30% ethyl alcohol
21. 50% ethyl alcohol
22. 70% ethyl alcohol
23. 80% ethyl alcohol
24. 95% ethyl alcohol
25. DePeX

#### 4.5 Anatomical study

##### Epidermal peeling

Mature leaves of family Araceae were collected from Thailand. Voucher specimens were deposited at the Mahasarakham University herbarium. Epidermal peeling was prepared by peeling method (Thammathaworn, 1996) between midrib and margin of the lamina and staining with 1% safranin in water. The specimens were then dehydrated using an alcohol series: 15%, 30%, 50%, 70%, 80%, 95% and 100%, mixture of absolute alcohol and xylene (1:1) and pure xylene. Then specimens were mounted in DePeX. The samples are studied under light microscopic study.

#### 4.6 Anatomical Analysis

Data analyses will use to determine the shape of the epidermal cell, type of stomata, length of guard cell, density of stomata, length of trichome, type of trichome, density of trichome and secretion in epidermal cell. Photographs were taken under light microscope (Zeiss: Axiostar plus).

#### 4.7 Results

Descriptions of epidermal peeling twenty Araceae species in Thailand.

##### *Aglaonema modestum* Schott ex Engl.

Leaf epidermal cells shapes exhibit a rectangular to polygonal in form with undulate anticlinal cell walls on both surfaces on the adaxial surface and on the abaxial surface. The epidermis cells on both surfaces of stomata were present in both adaxial and abaxial surfaces, and they possessed hexacytic stomatal type. Length of guard cell on the adaxial and the abaxial surface are  $57.05 \pm 0.59$  and  $61.35 \pm 0.67$   $\mu\text{m}$  respectively. The stomata density on the adaxial surface was  $1.31 \pm 0.21$   $\text{mm}^{-2}$  and on the abaxial



surface was  $14.36 \pm 2.17 \text{ mm}^{-2}$ . Sinuate cuticle sculpturing was present in both adaxial and abaxial surfaces (Table 4.2-4.3, Figure 4.1).

#### ***Aglaonema simplex* (Blume) Blume**

Leaf epidermal cells shapes exhibit a jigsaw in form with sinuate anticlinal cell walls on both surfaces on the adaxial surface and on the abaxial surface. The epidermis cells on both surfaces of stomata were present in both adaxial and abaxial surfaces, and they possessed hexacytic stomatal type. Length of guard cell on the adaxial and the abaxial surface are  $53.77 \pm 0.80$  and  $59.97 \pm 0.97 \text{ }\mu\text{m}$  respectively. The stomata density on the adaxial surface was  $3.65 \pm 0.32 \text{ mm}^{-2}$  and on the abaxial surface was  $21.30 \pm 3.42 \text{ mm}^{-2}$ . Undulate cuticle sculpturing was present in both adaxial and abaxial surfaces (Table 4.2-4.3, Figure 4.2).

#### ***Alocasia cucullata* (Lour.) G.Don**

Leaf epidermal cells shapes exhibit a rectangular to polygonal in form with smooth anticlinal cell walls on both surfaces on the adaxial surface and on the abaxial surface. The epidermis cells on both surfaces of stomata were present in both adaxial and abaxial surfaces, and they possessed cyclocytic stomatal type. Length of guard cell on the adaxial and the abaxial surface are  $33.65 \pm 0.77$  and  $35.88 \pm 0.47 \text{ }\mu\text{m}$  respectively. The stomata density on the adaxial surface was  $35.61 \pm 1.02 \text{ mm}^{-2}$  and on the abaxial surface was  $61.14 \pm 2.36 \text{ mm}^{-2}$ . Smooth cuticle sculpturing was present in both adaxial and abaxial surfaces (Table 4.2-4.3, Figure 4.3).

#### ***Alocasia longiloba* Miq.**

Leaf epidermal cells shapes exhibit a jigsaw in form with undulate anticlinal cell walls on both surfaces on the adaxial surface and on the abaxial surface. The epidermis cells on both surfaces of stomata were present in both adaxial and abaxial surfaces, and they possessed hexacytic stomatal type. Length of guard cell on the adaxial and the abaxial surface are  $37.68 \pm 0.53$  and  $39.98 \pm 0.59 \text{ }\mu\text{m}$  respectively. The stomata density on the adaxial surface was  $13.47 \pm 1.35 \text{ mm}^{-2}$  and on the abaxial surface was  $40.57 \pm 3.14 \text{ mm}^{-2}$ . Smooth cuticle sculpturing was present in both adaxial and abaxial surfaces (Table 4.2-4.3, Figure 4.4).

#### ***Alocasia macrorrhizos* (L.) G.Don**

Leaf epidermal cells shapes exhibit a rectangular to polygonal in form with smooth anticlinal cell walls on both surfaces on the adaxial surface and on the abaxial surface. The epidermis cells on both surfaces of stomata were present in both adaxial and abaxial surfaces, and they possessed cyclocytic stomatal type. Length of guard cell on the adaxial and the abaxial surface are  $29.74 \pm 0.54$  and  $26.85 \pm 0.30 \text{ }\mu\text{m}$  respectively. The stomata density on the adaxial surface was  $54.15 \pm 1.33 \text{ mm}^{-2}$  and on the abaxial surface was  $69.29 \pm 1.80 \text{ mm}^{-2}$ . Smooth cuticle sculpturing was present in both adaxial and abaxial surfaces (Table 4.2-4.3, Figure 4.5).



### ***Alocasia* sp.**

Leaf epidermal cells shapes exhibit a jigsaw in form with undulate anticlinal cell walls on both surfaces on the adaxial surface and on the abaxial surface. The epidermis cells on both surfaces of stomata were present in both adaxial and abaxial surfaces, and they possessed hexacytic stomatal type. Length of guard cell on the adaxial and the abaxial surface are  $30.17 \pm 0.35$  and  $34.55 \pm 0.44$   $\mu\text{m}$  respectively. The stomata density on the adaxial surface was  $17.07 \pm 0.33$   $\text{mm}^{-2}$  and on the abaxial surface was  $48.77 \pm 3.64$   $\text{mm}^{-2}$ . Smooth cuticle sculpturing was present in both adaxial and abaxial surfaces (Table 4.2-4.3, Figure 4.6).

### ***Amorphophallus serrulatus* Hett. & A.Galloway**

Leaf epidermal cells shapes exhibit a rectangular to polygonal on the adaxial surface and a jigsaw on the abaxial surface in form with smooth anticlinal cell walls on both surfaces on the adaxial surface and on the abaxial surface. The epidermis cells only surfaces of stomata were present in abaxial surfaces, and it is possessed paracytic stomatal type. Length of guard cell on the abaxial  $50.62 \pm 0.74$   $\mu\text{m}$  so, stomata only on the lower leaf surface. The stomata density on the abaxial surface was  $63.34 \pm 4.92$   $\text{mm}^{-2}$ . Smooth cuticle sculpturing was present in both adaxial and abaxial surfaces. Tannin is presented in epidermal cells on the both adaxial and abaxial surfaces (Table 4.2-4.3, Figure 4.7).

### ***Arisaema maxwellii* Hett. & Gusman**

Leaf epidermal cells shapes exhibit a rectangular to polygonal on the adaxial surface and a jigsaw on the abaxial surface in form with smooth anticlinal cell walls on both surfaces on the adaxial surface and on the abaxial surface. The epidermis cells only surfaces of stomata were present in abaxial surfaces, and it is possessed hexacytic stomatal type. Length of guard cell on the abaxial  $37.19 \pm 0.83$   $\mu\text{m}$  so, stomata only on the lower leaf surface. The stomata density on the abaxial surface was  $48.19 \pm 3.50$   $\text{mm}^{-2}$ . Smooth cuticle sculpturing was present in both adaxial and abaxial surfaces (Table 4.2-4.3, Figure 4.8).

### ***Colocasia esculenta* (L.) Schott**

Leaf epidermal cells shapes exhibit a rectangular to polygonal in form with smooth anticlinal cell walls on both surfaces on the adaxial surface and on the abaxial surface. The epidermis cells on both surfaces of stomata were present in both adaxial and abaxial surfaces, and they possessed anomocytic stomatal type. Length of guard cell on the adaxial and the abaxial surface are  $25.30 \pm 0.32$  and  $28.72 \pm 0.14$   $\mu\text{m}$  respectively. The stomata density on the adaxial surface was  $143.07 \pm 1.25$   $\text{mm}^{-2}$  and on the abaxial surface was  $193.03 \pm 2.37$   $\text{mm}^{-2}$ . Tannin is presented in epidermal cells on the both adaxial and abaxial surfaces (Table 4.2-4.3, Figure 4.9).



### ***Colocasia fallax* Schott**

Leaf epidermal cells shapes exhibit a rectangular to polygonal in form with smooth anticlinal cell walls on both surfaces on the adaxial surface and on the abaxial surface. The epidermis cells on both surfaces of stomata were present in both adaxial and abaxial surfaces, and they possessed anomocytic stomatal type. Length of guard cell on the adaxial and the abaxial surface are  $28.92 \pm 0.33$  and  $33.80 \pm 0.46$   $\mu\text{m}$  respectively. The stomata density on the adaxial surface was  $17.34 \pm 1.03$   $\text{mm}^{-2}$  and on the abaxial surface was  $84.48 \pm 2.98$   $\text{mm}^{-2}$ . Tannin is presented in epidermal cells on the both adaxial and abaxial surfaces. Glandular trichomes are presented in epidermal cell on the abaxial surface (Table 4.2-4.3, Figure 4.10).

### ***Colocasia gigantea* (Blume) Hook.f.**

Leaf epidermal cells shapes exhibit a rectangular to polygonal in form with smooth anticlinal cell walls on both surfaces on the adaxial surface and on the abaxial surface. The epidermis cells only surfaces of stomata were present in abaxial surfaces, and it is possessed were 2, 3, 4, 5 and 6 subsidiary cells and five subtypes of stomatal. Length of guard cell on the adaxial and the abaxial surface are  $26.41 \pm 0.40$  and  $28.56 \pm 0.32$   $\mu\text{m}$  respectively. The stomata density on the adaxial surface was  $49.97 \pm 1.51$   $\text{mm}^{-2}$  and on the abaxial surface was  $83.02 \pm 3.66$   $\text{mm}^{-2}$ . Smooth cuticle sculpturing was present in both adaxial and abaxial surfaces (Table 4.2-4.3, Figure 4.11).

### ***Colocasia lihengiae* C.L.Long & K.M.Liu**

Leaf epidermal cells shapes exhibit a rectangular to polygonal on the adaxial surface and a jigsaw on the abaxial surface in form with undulate anticlinal cell walls on both surfaces on the adaxial surface and on the abaxial surface. The epidermis cells on both surfaces of stomata were present in both adaxial and abaxial surfaces, and they possessed paracytic stomatal type. Length of guard cell on the adaxial and the abaxial surface are  $26.25 \pm 0.37$  and  $30.91 \pm 0.32$   $\mu\text{m}$  respectively. The stomata density on the adaxial surface was  $15.30 \pm 0.74$   $\text{mm}^{-2}$  and on the abaxial surface was  $72.21 \pm 0.88$   $\text{mm}^{-2}$ . Smooth cuticle sculpturing was present in both adaxial and abaxial surfaces (Table 4.2-4.3, Figure 4.12).

### ***Hapaline benthamiana* Schott**

Leaf epidermal cells shapes exhibit a rectangular to polygonal on the adaxial surface and a jigsaw on the abaxial surface in form with smooth anticlinal cell walls on both surfaces on the adaxial surface and on the abaxial surface. The epidermis cells on both surfaces of stomata were present in both adaxial and abaxial surfaces, and they possessed hexacytic stomatal type. Length of guard cell on the adaxial and the abaxial surface are  $43.25 \pm 0.61$  and  $43.35 \pm 0.49$   $\mu\text{m}$  respectively. The stomata density on the adaxial surface was  $2.98 \pm 0.20$   $\text{mm}^{-2}$  and on the abaxial surface was  $39.63 \pm 2.96$   $\text{mm}^{-2}$ . Smooth cuticle sculpturing was present in both adaxial and abaxial surfaces (Table 4.2-4.3, Figure 4.13).



***Homalomena griffithii* (Schott) Hook.f.**

Leaf epidermal cells shapes exhibit a rectangular to polygonal in form with smooth anticlinal cell walls on both surfaces on the adaxial surface and on the abaxial surface. The epidermis cells on both surfaces of stomata were present in both adaxial and abaxial surfaces, and they possessed hexacytic stomatal type. Length of guard cell on the adaxial and the abaxial surface are  $32.46 \pm 0.48$  and  $31.42 \pm 0.38$   $\mu\text{m}$  respectively. The stomata density on the adaxial surface was  $66.36 \pm 1.03$   $\text{mm}^{-2}$  and on the abaxial surface was  $114.92 \pm 1.40$   $\text{mm}^{-2}$ . Undulate cuticle sculpturing was present in both adaxial and abaxial surfaces. A solitary crystal and raphides are presented in epidermal cell on the adaxial surface and on the abaxial surface. Tannin is presented in epidermal cells on the both adaxial and abaxial surfaces (Table 4.2-4.3, Figure 4.14).

***Lasia spinosa* (L.) Thwaites**

Leaf epidermal cells shapes exhibit a rectangular to polygonal on the adaxial surface and a jigsaw on the abaxial surface in form with smooth and undulate anticlinal cell walls on both surfaces on the adaxial surface and on the abaxial surface. The epidermis cells on both surfaces of stomata were present in both adaxial and abaxial surfaces, and they possessed paracytic stomatal type. Length of guard cell on the adaxial and the abaxial surface are  $40.93 \pm 0.15$  and  $37.03 \pm 0.45$   $\mu\text{m}$  respectively. The stomata density on the adaxial surface was  $37.65 \pm 1.14$   $\text{mm}^{-2}$  and on the abaxial surface was  $77.85 \pm 2.63$   $\text{mm}^{-2}$ . Smooth cuticle sculpturing was present in both adaxial and abaxial surfaces (Table 4.2-4.3, Figure 4.15).

***Pistia stratiotes* L.**

Leaf epidermal cells shapes exhibit a rectangular to polygonal in form with smooth anticlinal cell walls on both surfaces on the adaxial surface and on the abaxial surface. The epidermis cells on both surfaces of stomata were present in both adaxial and abaxial surfaces, and they possessed anomocytic stomatal type. Length of guard cell on the adaxial and the abaxial surface are  $16.60 \pm 0.41$  and  $18.73 \pm 0.46$   $\mu\text{m}$  respectively. The stomata density on the adaxial surface was  $129.65 \pm 1.59$   $\text{mm}^{-2}$  and on the abaxial surface was  $134.29 \pm 1.77$   $\text{mm}^{-2}$ . Smooth cuticle sculpturing was present in both adaxial and abaxial surfaces. A solitary crystal is presented in epidermal cell on the adaxial surface and on the abaxial surface. Multi-cellular trichomes are presented in epidermal cell on the adaxial surface and on the abaxial surface (Table 4.2-4.3, Figure 4.16).

***Pycnospatha palmata* Gagnep.**

Leaf epidermal cells shapes exhibit a rectangular to polygonal on the adaxial surface and a jigsaw on the abaxial surface in form with smooth and undulate anticlinal cell walls on both surfaces on the adaxial surface and on the abaxial surface. The epidermis cells on both surfaces of stomata were present in both adaxial and abaxial surfaces, and they possessed paracytic stomatal type. Length of guard cell on the adaxial and the abaxial surface are  $35.86 \pm 0.81$  and  $37.97 \pm 0.84$   $\mu\text{m}$  respectively. The stomata density on the adaxial surface was  $7.73 \pm 0.35$   $\text{mm}^{-2}$  and on the abaxial surface  $121.14 \pm 0.85$   $\text{mm}^{-2}$ . Smooth cuticle sculpturing was present in both adaxial and abaxial surfaces (Table 4.2-4.3, Figure 4.17).





### ***Schismatoglottis calyptrata* (Roxb.) Zoll. & Moritzi**

Leaf epidermal cells shapes exhibit a rectangular to polygonal in form with smooth anticlinal cell walls on both surfaces on the adaxial surface and on the abaxial surface. The epidermis cells on both surfaces of stomata were present in both adaxial and abaxial surfaces, and they possessed hexacytic stomatal type. Length of guard cell on the adaxial and the abaxial surface are  $35.87 \pm 0.40$  and  $34.60 \pm 0.35$   $\mu\text{m}$  respectively. The stomata density on the adaxial surface was  $22.50 \pm 0.41$   $\text{mm}^{-2}$  and on the abaxial surface was  $80.51 \pm 1.71$   $\text{mm}^{-2}$ . A solitary crystal is presented in epidermal cell on the adaxial surface and on the abaxial surface. Undulate cuticle sculpturing was present in both adaxial and abaxial surfaces. Glandular trichomes are presented in epidermal cell on the adaxial surface and on the abaxial surface (Table 4.2-4.3, Figure 4.18).

### ***Typhonium glaucum* Hett. & Sookchaloem**

Leaf epidermal cells shapes exhibit a rectangular to polygonal in form with smooth anticlinal cell walls on both surfaces on the adaxial surface and on the abaxial surface. The epidermis cells on both surfaces of stomata were present in both adaxial and abaxial surfaces, and they possessed 2, 3, 4 subsidiary cells type. Length of guard cell on the adaxial and the abaxial surface are  $49.79 \pm 0.73$  and  $45.27 \pm 0.50$   $\mu\text{m}$  respectively. The stomata density on the adaxial surface was  $29.92 \pm 1.16$   $\text{mm}^{-2}$  and on the abaxial surface was  $30.81 \pm 1.73$   $\text{mm}^{-2}$ . Smooth cuticle sculpturing was present in both adaxial and abaxial surfaces. Glandular trichomes are presented in epidermal cell on the adaxial surface and on the abaxial surface (Table 4.2-4.3, Figure 4.19).

### ***Typhonium trilobatum* (L.) Schott**

Leaf epidermal cells shapes exhibit a rectangular to polygonal on the adaxial surface and a jigsaw on the abaxial surface in form with smooth and undulate anticlinal cell walls on both surfaces on the adaxial surface and on the abaxial surface. The epidermis cells on both surfaces of stomata were present in both adaxial and abaxial surfaces, and they possessed 2, 3, 4, 5 and 6 subsidiary cells and five subtypes of stomatal. Length of guard cell on the adaxial and the abaxial surface are  $28.65 \pm 0.38$  and  $29.38 \pm 0.61$   $\mu\text{m}$  respectively. The stomata density on the adaxial surface was  $36.55 \pm 1.87$   $\text{mm}^{-2}$  and on the abaxial surface was  $169.02 \pm 1.95$   $\text{mm}^{-2}$ . Smooth cuticle sculpturing was present in both adaxial and abaxial surfaces (Table 4.2-4.3, Figure 4.20).

## **4.8 Conclusion and Discussion**

The result of anatomical characteristics show the 20 species plants Araceae in Thailand by peeling method study under light microscope and concluded of leaf epidermis as Table 4.2 and 4.3. The characteristics of anatomy found that can be divided into groups as follows.

1. Leaf surface anatomy can be divided into four groups based on epidermal cell shapes.

1.1 The first group is a rectangular to polygonal on the adaxial surface including *Aglaonema modestum*, *Alocasia cucullata*, *A. macrorrhizos*, *Amorphophallus serrulatus*, *Arisaema maxwellii*, *Colocasia esculenta*, *C. fallax*, *C. gigantea*, *C. lihengiae*, *Hapaline benthamiana*, *Homalomena griffithii*, *Lasia spinosa*, *Pistia*



*stratiotes*, *Pycnosphata palmata*, *Scittomaglottis calyptrate*, *Typhonium glaucum* and *Typhonium trilobatum*.

1.2 The second group is a jigsaw on the adaxial surface including *Aglaonema simplex*, *Alocacia longiloba* and *A. sp.*.

1.3 The third group is rectangular to polygonal on the abaxial surface including *Aglaonema modestum*, *Alocacia cucullata*, *A. macrorrhizos*, *Colocasia esculenta*, *C. fallax*, *C. gigantea*, *Homalomena griffithii*, *Pistia stratiotes*, *Scittomaglottis calyptrate* and *Typhonium glaucum*.

1.4 The fourth group is a jigsaw on the abaxial surface including *Aglaonema simplex*, *Alocacia longiloba*, *A. sp.*, *Amorphophallus serrulatus*, *Arisaema maxwellii*, *Colocasia lihengiae*, *Hapaline benthamiana*, *Lasia spinosa*, *Pycnosphata palmata* and *Typhonium trilobatum*.

## 2. Seven groups anatomical characteristics based on stomatal types.

2.1 The first group is anomocytic stomatal types including *Colocasia esculenta*, *C. fallax* and *Pistia stratiotes*.

2.2 The second group is paracytic stomatal types including *Amorphophallus serrulatus*, *Colocasia lihengiae*, *Lasia spinosa* and *Pycnosphata palmata*.

2.3 The third group is hexacytic stomatal types including *Aglaonema modestum*, *A. simplex*, *Alocacia longiloba*, *A. macrorrhizos*, *A.sp.*, *Arisaema maxwellii*, *Hapaline benthamiana*, *Homalomena griffithii* and *Scittomaglottis calyptrate*.

2.4 The fourth group is cyclocytic cells stomatal types is an *Alocacia cucullata*.

2.5 The fifth group is 2, 3, 4 subsidiary cells stomatal types including *Typhonium glaucum*.

2.6 The sixth group is 2, 3, 4, 5, 6 subsidiary cells stomatal types including *Typhonium trilobatum*.

2.7 The seventh group is 2, 3, 4, 6 subsidiary cells stomatal types including *Colocasia gigantea*.

3. The group has surfaces of stomata were present only in abaxial surfaces including *Amorphophallus serrulatus* and *Arisaema maxwellii*.

## 4. Four groups of anatomical characteristics of cutin sculpturing.

4.1 The first group is smooth cutin sculpturing of this study found that the thirteen species including *Alocacia cucullata*, *A. longiloba*, *A. macrorrhizos*, *A. sp.*, *Amorphophallus serrulatus*, *Arisaema maxwellii*, *Colocasia gigantea*, *C. lihengiae*, *Hapaline benthamiana*, *Lasia spinosa*, *Pistia stratiotes*, *Pycnosphata palmata*, *Typhonium glaucum* and *Typhonium trilobatum*.

4.2 The second group is sinuate cutin sculpturing of this study found that the only one species is an *Aglaonema modestum*.

4.3 The third group is undulate cutin sculpturing of this study found that the two species including *Aglaonema simplex*, *Homalomena griffithii* and *Scittomaglottis calyptrate*.

4.4 The fourth group absents the cutin sculpturing of this study found that the four species including *Colocasia esculenta* and *C. fallax*.



5. Three groups it the same of anatomical characteristics on both surfaces on the adaxial surface and on the abaxial surface of anticlinal cell wall and one group it is not the same on both surfaces.

5.1 The first group is smooth anticlinal cell wall of this study found that the thirteen species including, *Alocacia cucullata*, *A. macrorrhizos*, *Amorphophallus serrulatus*, *Arisaema maxwellii*, *Colocasia esculenta*, *C. fallax*, *C. gigantea*, *Hapaline benthamiana*, *Homalomena griffithii*, *Pistia stratiotes*, *Scittomaglottis calyptrate*, *Typhonium glaucum* and *Typhonium trilobatum*.

5.2 The second group is sinuate anticlinal cell wall of this study found that the only one species an *Aglaonema simplex*.

5.3 The third group is undulate anticlinal cell wall of this study found that the forth species including *Aglaonema modestum*, *Alocacia longiloba*, *A. sp.* and *Colocasia lihengiae*.

5.4 The forth group is anticlinal cell wall it is not the same on both surfaces of this study found that the two species including *Lasia spinosa* (upper=smooth, lower=undulate) and *Pycnosphata palmata* (upper=smooth, lower= undulate).

6. Anatomical characteristics of this study found that the three species with of inclusions/distributions including *Homalomena griffithii* (druse, raphides), *Pistia stratiotes* (druse) and *Scittomaglottis calyptrate* (druse).

7. Anatomical characteristics of this study found that the four species with of trichomes including *Colocasia fallax* (glandular trichome), *Pistia stratiotes* (multi-cellular), *Scittomaglottis calyptrate* (glandular trichome) and *Typhonium glaucum* (glandular trichome).

8. Anatomical characteristics of this study found that the four species with of tannin including *Amorphophallus serrulatus*, *Colocasia esculenta*, *C. fallax* and *Homalomena griffithii*.

These results are anatomical characteristics of *Aglaonema modestum* Schott is in agreement with Boontang *et al.* (2011) reported anatomical characteristics found that the leaf epidermal cells shapes exhibit a jigsaw in form with undulate anticlinal cell walls on both surfaces on the adaxial surface and on the abaxial surface. The epidermis cells on both surfaces of stomata were present in both adaxial and abaxial surfaces, and they possessed hexacytic stomatal type. And smooth cuticle sculpturing was present in both adaxial and abaxial surfaces. The only difference epidermal cells shapes exhibit a rectangular to polygonal.

The results are anatomical characteristics of *Aglaonema simplex* (Blume) Blume is the epidermis cells on surfaces of stomata were present hexacytic stomatal type. This result is in disagreement with Sungkajanttranon *et al.* (2010) reported anatomical characteristics found that the number of 4 subsidiary cell on leaf surfaces.

The results are anatomical characteristics of *Colocasia esculenta* (L.) Schott agreement with Sungkajanttranon *et al.* (2010) and Ezeabara (2015) reported the leaves possessed anomocytic stomatal type with the stomata present in both adaxial and abaxial surfaces.

The results are anatomical characteristics of *Colocasia gigantea* (Blume) Hook.f. is the epidermis cells only surfaces of stomata were present in abaxial surfaces,



and it is possessed were 2, 3, 4, 5 and 6 subsidiary cells and five subtypes of stomatal is in disagreement with Sungkajanttranon *et al.* (2010) reported anatomical characteristics found that the number of 2 subsidiary cell on leaf surfaces.

The results are anatomical characteristics of *Lasia spinosa* (L.) Thwaites. show that the epidermis cells on both surfaces of stomata were present in both adaxial and abaxial surfaces, and they possessed paracytic stomatal type is in disagreement with Sungkajanttranon *et al.* (2010) reported anatomical characteristics found that the number of 4-5 subsidiary cell on leaf surfaces.

Moreover, in leaves, stomata may occur on both sides or only on one side, usually the lower side. Although, occasionally some species exist which have several types of stomata on one leaf, most have one type only (Cutler, 1978). Esau (1977) stated that stomata occur on all aerial parts of the plant, but they are most abundant on leaves. Moreover, Sharma (1993) noted that leaf anatomy provides various characters of taxonomic importance. It has been a critical tool in the hand of taxonomists in the classification and separation of species (Illoh *et al.*, 2011).

The Anatomy of 15 species have been recorded for the first time as follows; *Alocacia cucullata*, *Alocacia longiloba*, *Alocasia macrorrhizos*, *Alocasia* sp., *Amorphophallus serrulatus*, *Arisaema maxwellii*, *Colocasia fallax*, *Hapaline benthamiana*, *Lasia spinosa*, *Pistia stratiotes*, *Pycnosphata palmata*, *Scittomaglottis calyptrata*, *Typhonium glaucum* and *Typhonium trilobatum*.

However, there should be more study of the anatomy character, such as cutting the transverse leaf, petioles, stems and roots and to characteristics that help to identify more clearly the kinds of plants. Also, should be a study of samples of plants in each genus further. To get more like the anatomy of plants in this family provides the most complete. This will result in the classification and identification of plant species to be accurate, complete and up the next.



**Table 4.2** Comparisons of some character of leaf surface anatomy (adaxial side) of the 20 species Araceae in Thailand

Species	Epidermal cell shapes	Stomatal types	Length of guard cell M±SD (µm)	Density of stomata M±SD (mm <sup>-2</sup> )	Cutin sculpturing	Anticlinal cell wall	Inclusions/ Distributions	Trichomes	T
1. <i>Aglaonema modestum</i>	rectangular to polygonal	hexacytic	57.05±0.59	1.31±0.21	sinuate	undulate	-	-	-
2. <i>Aglaonema simplex</i>	Jigsaw	hexacytic	53.77±0.80	3.65±0.32	undulate	sinuate	-	-	-
3. <i>Alocacia cucullata</i>	rectangular to polygonal	cyclocytic	33.65±0.77	35.61±1.02	smooth	smooth	-	-	-
4. <i>Alocacia longiloba</i>	jigsaw	hexacytic	37.68±0.53	13.47±1.35	smooth	undulate	-	-	-
5. <i>Alocacia macrorrhizos</i>	rectangular to polygonal	hexacytic	29.74±0.54	54.15±1.33	smooth	smooth	-	-	-
6. <i>Alocacia</i> sp.	jigsaw	hexacytic	30.17±0.35	17.07±0.33	smooth	undulate	-	-	-
7. <i>Amorphophallus serrulatus</i>	rectangular to polygonal	absent	absent	absent	smooth	smooth	-	-	✓
8. <i>Arisaema maxwellii</i>	rectangular to polygonal	absent	absent	absent	smooth	smooth	-	-	-
9. <i>Colocasia esculenta</i>	rectangular to polygonal	anomocytic	25.30±0.32	143.07±1.25	-	smooth	-	-	✓
10. <i>Colocasia fallax</i>	rectangular to polygonal	anomocytic	28.92±0.33	17.34±1.03	-	smooth	-	-	✓
11. <i>Colocasia gigantea</i>	rectangular to polygonal	2,3,4,6 subsidiary cells	26.41±0.40	49.97±1.51	smooth	smooth	-	-	-
12. <i>Colocasia lihengiae</i>	rectangular to polygonal	paracytic	26.25±0.37	15.30±0.74	smooth	undulate	-	-	-
13. <i>Hapaline benthamiana</i>	rectangular to polygonal	hexacytic	43.25±0.61	2.98±0.20	smooth	smooth	-	-	-

**Table 4.2** Comparisons of some character of leaf surface anatomy (adaxial side) of the 20 species Araceae in Thailand (Continued).

Species	Epidermal cell shapes	Stomatal types	Length of guard cell M $\pm$ SD ( $\mu$ m)	Density of stomata M $\pm$ SD (mm <sup>-2</sup> )	Cutin sculpturing	Anticlinal cell wall	Inclusions/ Distributions	Trichomes	T
14. <i>Homalomena griffithii</i>	rectangular to polygonal	hexacytic	32.46 $\pm$ 0.48	66.36 $\pm$ 1.03	undulate	smooth	druse, raphides	-	✓
15. <i>Lasia spinosa</i>	rectangular to polygonal	paracytic	40.93 $\pm$ 0.15	37.65 $\pm$ 1.14	smooth	smooth/ undulate	-	-	-
16. <i>Pistia stratiotes</i>	rectangular to polygonal	anomocytic	16.60 $\pm$ 0.41	129.65 $\pm$ 1.59	smooth	smooth	druse	multi-cellular hair	-
17. <i>Pycnosphata palmata</i>	rectangular to polygonal	paracytic	35.86 $\pm$ 0.81	7.73 $\pm$ 0.35	smooth	smooth/ undulate	-	-	-
18. <i>Scittomaglottis calyptrata</i>	rectangular to polygonal	hexacytic	35.87 $\pm$ 0.40	22.50 $\pm$ 0.41	undulate	smooth	druse	glandular trichome	-
19. <i>Typhonium glaucum</i>	rectangular to polygonal	2,3,4 subsidiary cells	49.79 $\pm$ 0.73	29.92 $\pm$ 1.16	smooth	smooth	-	glandular trichome	-
20. <i>Typhonium trilobatum</i>	rectangular to polygonal	2,3,4,5,6 subsidiary cells	28.65 $\pm$ 0.38	36.55 $\pm$ 1.87	smooth	smooth	-	-	-

Note: T=tannin, “-” = absence, “✓” = present



**Table 4.3** Comparisons of some character of leaf surface anatomy (abaxial side) of the 20 species Araceae in Thailand

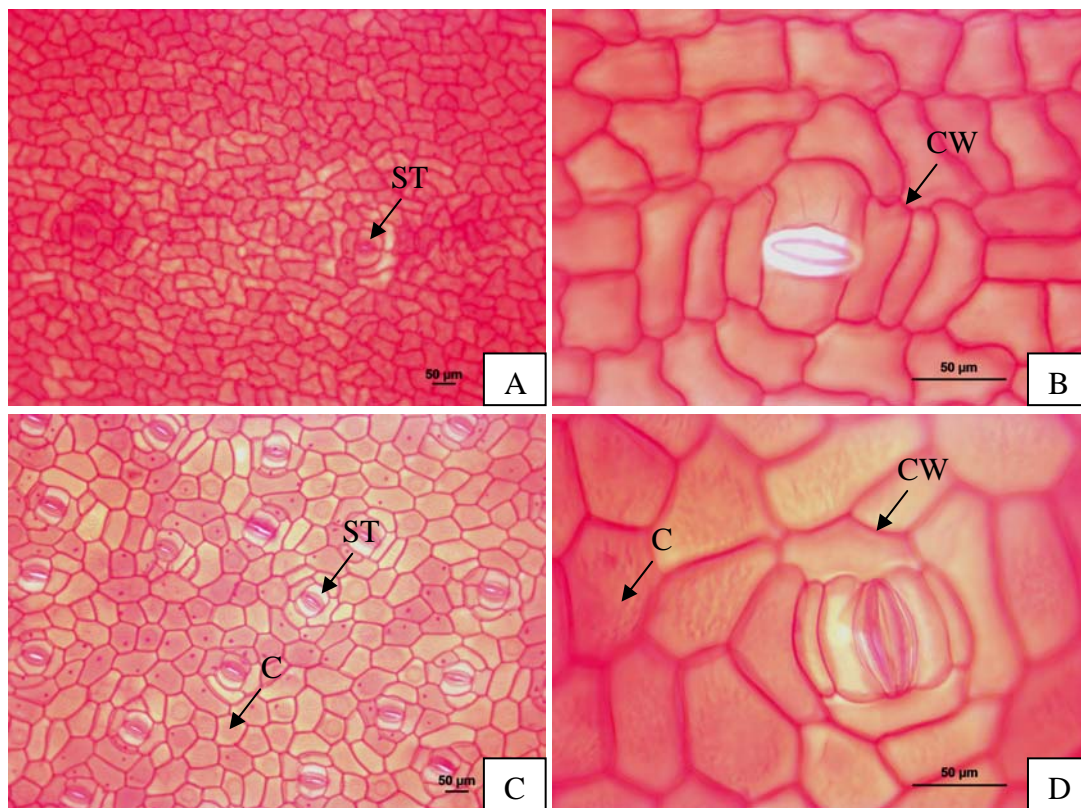
Species	Epidermal cell shapes	Stomatal types	Length of guard cell M $\pm$ SD ( $\mu$ m)	Density of stomata M $\pm$ SD (mm <sup>-2</sup> )	Cutin sculpturing	Anticlinal cell wall	Inclusions/ Distributions	Trichomes	T
1. <i>Aglaonema modestum</i>	rectangular to polygonal	hexacytic	61.35 $\pm$ 0.67	14.36 $\pm$ 2.17	sinuate	undulate	-	-	-
2. <i>Aglaonema simplex</i>	jigsaw	hexacytic	59.97 $\pm$ 0.97	21.30 $\pm$ 3.42	undulate	sinuate	-	-	-
3. <i>Alocacia cucullata</i>	rectangular to polygonal	cyclocytic	35.88 $\pm$ 0.47	61.14 $\pm$ 2.36	smooth	smooth	-	-	-
4. <i>Alocacia longiloba</i>	jigsaw	hexacytic	39.98 $\pm$ 0.59	40.57 $\pm$ 3.14	smooth	undulate	-	-	-
5. <i>Alocacia macrorrhizos</i>	rectangular to polygonal	hexacytic	26.85 $\pm$ 0.30	69.29 $\pm$ 1.80	smooth	smooth	-	-	-
6. <i>Alocacia</i> sp.	jigsaw	hexacytic	34.55 $\pm$ 0.44	48.77 $\pm$ 3.64	smooth	undulate	-	-	-
7. <i>Amorphophallus serrulatus</i>	jigsaw	paracytic	50.62 $\pm$ 0.35	63.34 $\pm$ 4.92	smooth	smooth	-	-	✓
8. <i>Arisaema maxwellii</i>	jigsaw	hexacytic	37.19 $\pm$ 0.31	48.19 $\pm$ 3.50	smooth	smooth	-	-	-
9. <i>Colocasia esculenta</i>	rectangular to polygonal	anomocytic	28.72 $\pm$ 0.14	193.03 $\pm$ 2.37	-	smooth	-	-	✓
10. <i>Colocasia fallax</i>	rectangular to polygonal	anomocytic	33.80 $\pm$ 0.46	84.48 $\pm$ 2.98	-	smooth	-	glandular trichome	✓
11. <i>Colocasia gigantea</i>	rectangular to polygonal	2,3,4,6 subsidiary cells	28.56 $\pm$ 0.32	83.02 $\pm$ 3.66	smooth	smooth	-	-	-
12. <i>Colocasia lihengiae</i>	jigsaw	paracytic	30.91 $\pm$ 0.32	72.21 $\pm$ 0.88	smooth	undulate	-	-	-
13. <i>Hapaline benthamiana</i>	jigsaw	hexacytic	43.35 $\pm$ 0.49	39.63 $\pm$ 2.96	smooth	smooth	-	-	-

**Table 4.3** Comparisons of some character of leaf surface anatomy (abaxial side) of the 20 species Araceae in Thailand (Continued).

Species	Epidermal cell shapes	Stomatal types	Length of guard cell M $\pm$ SD ( $\mu$ m)	Density of stomata M $\pm$ SD (mm <sup>-2</sup> )	Cutin sculpturing	Anticlinal cell wall	Inclusions/ Distributions	Trichomes	T
14. <i>Homalomena griffithii</i>	rectangular to polygonal	hexacytic	31.42 $\pm$ 0.38	144.92 $\pm$ 1.40	smooth	smooth	druse, raphides	-	✓
15. <i>Lasia spinosa</i>	jigsaw	paracytic	37.03 $\pm$ 0.45	77.85 $\pm$ 2.63	undulate	smooth/ undulate	-	-	-
16. <i>Pistia stratiotes</i>	rectangular to polygonal	anomocytic	18.73 $\pm$ 0.46	134.29 $\pm$ 1.77	smooth	smooth	druse	multi-cellular	-
17. <i>Pycnosphata palmata</i>	jigsaw	paracytic	37.97 $\pm$ 0.84	121.14 $\pm$ 0.85	smooth	smooth/ undulate	-	-	-
18. <i>Scittomaglottis calyptrata</i>	rectangular to polygonal	hexacytic	34.60 $\pm$ 0.35	80.51 $\pm$ 1.71	undulate	smooth	druse	glandular trichome	-
19. <i>Typhonium glaucum</i>	rectangular to polygonal	2,3,4 subsidiary cells	45.27 $\pm$ 0.05	30.81 $\pm$ 1.73	smooth	smooth	-	glandular trichome	-
20. <i>Typhonium trilobatum</i>	jigsaw	2,3,4,5,6 subsidiary cells	29.38 $\pm$ 0.61	169.02 $\pm$ 1.95	smooth	smooth	-	-	-

Note: T=tannin, “-” = absence, “✓” = present

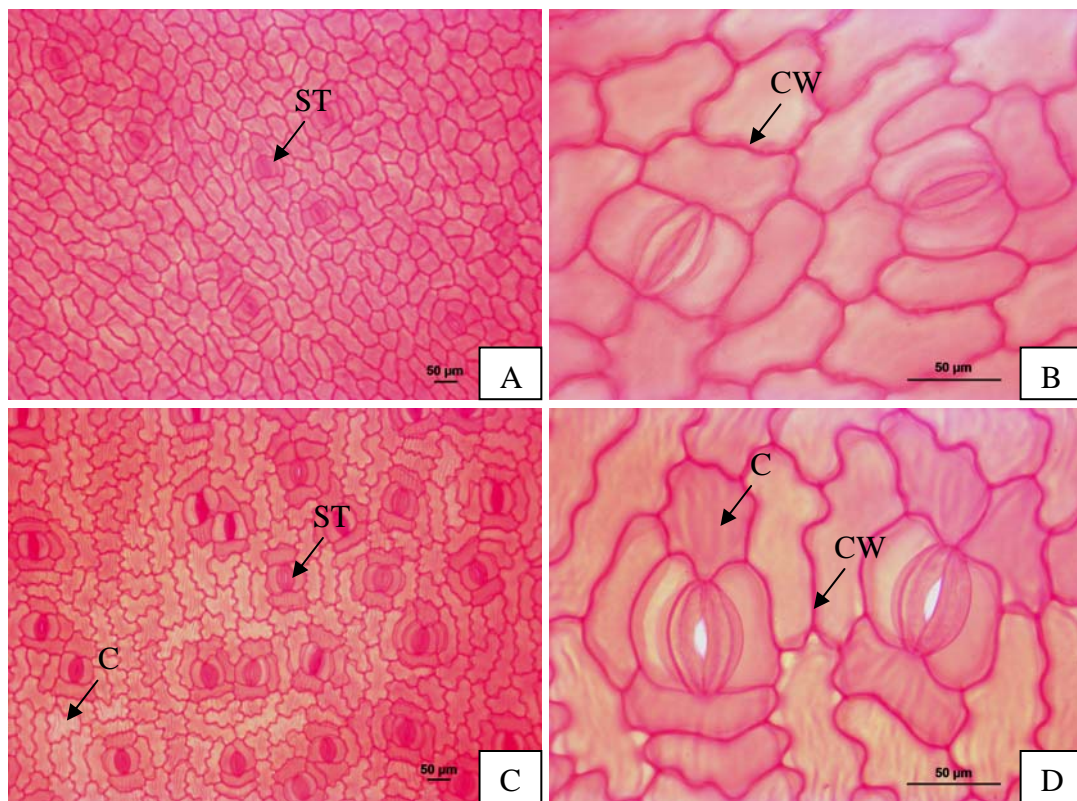




**Figure 4.1** Leaf surfaces anatomy of *Aglaonema modestum* Schott ex Engl. (Scale bars = 50 µm) C = cuticle sculpturing, CW = cell walls, ST = stomata.

- A. An upper (adaxial) epidermis, 10x magnification light microscope
- B. An upper (adaxial) epidermis, 40x magnification light microscope
- C. A lower (abaxial) epidermis, 10x magnification light microscope
- D. A lower (abaxial) epidermis, 40x magnification light microscope

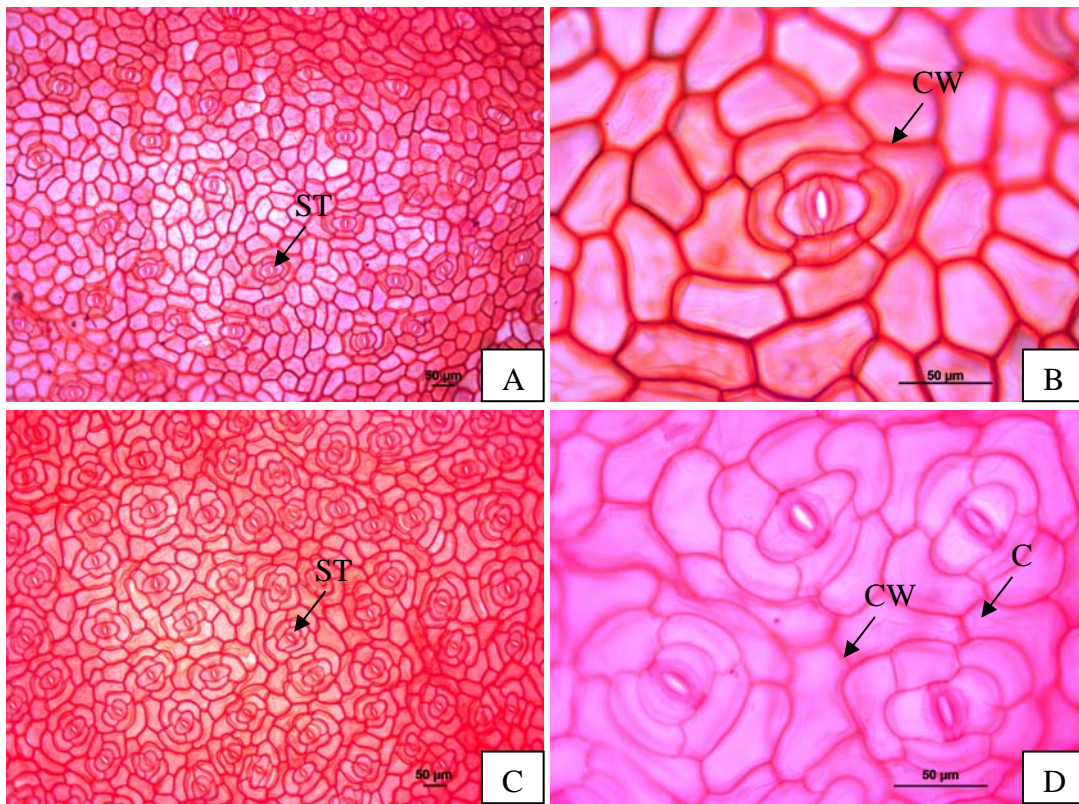




**Figure 4.2** Leaf surfaces anatomy of *Aglaonema simplex* (Blume) Blume (Scale bars = 50 µm) C = cuticle sculpturing, CW = cell walls, ST = stomata.

- A. An upper (adaxial) epidermis, 10x magnification light microscope
- B. An upper (adaxial) epidermis, 40x magnification light microscope
- C. A lower (abaxial) epidermis, 10x magnification light microscope
- D. A lower (abaxial) epidermis, 40x magnification light microscope

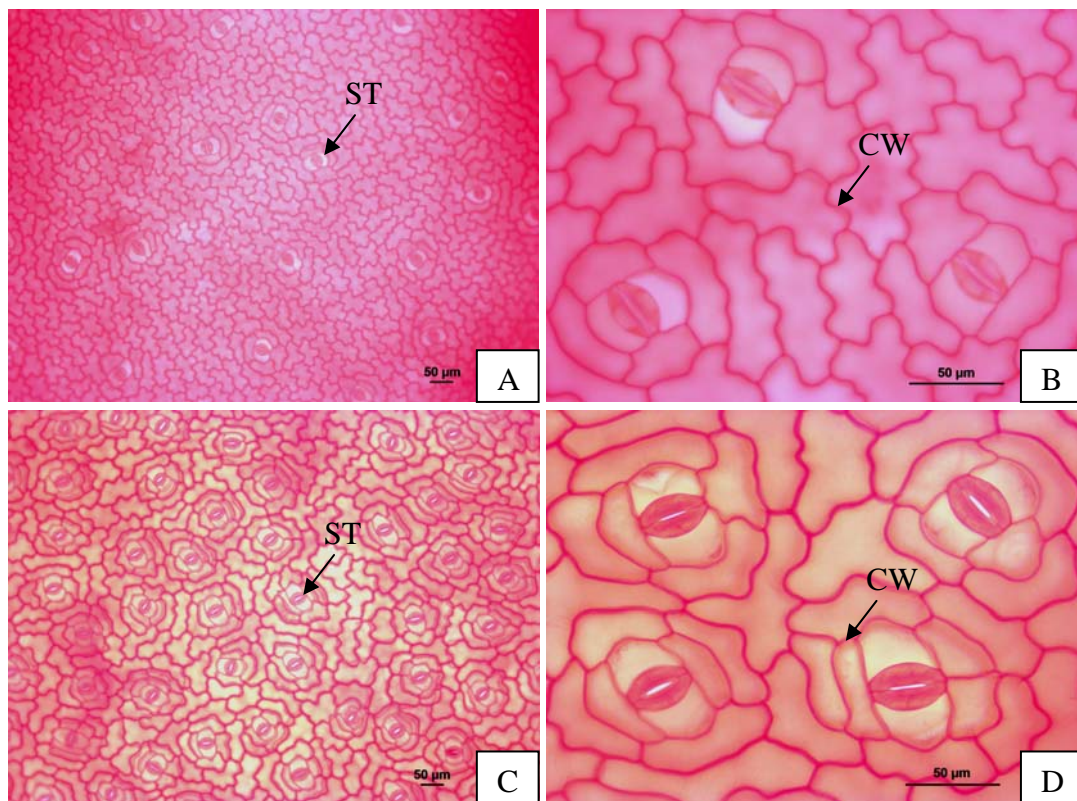




**Figure 4.3** Leaf surfaces anatomy of *Alocasia cucullata* (Lour.) G.Don (Scale bars = 50 µm) C = cuticle sculpturing, CW = cell walls, ST = stomata.

- A. An upper (adaxial) epidermis, 10x magnification light microscope
- B. An upper (adaxial) epidermis, 40x magnification light microscope
- C. A lower (abaxial) epidermis, 10x magnification light microscope
- D. A lower (abaxial) epidermis, 40x magnification light microscope

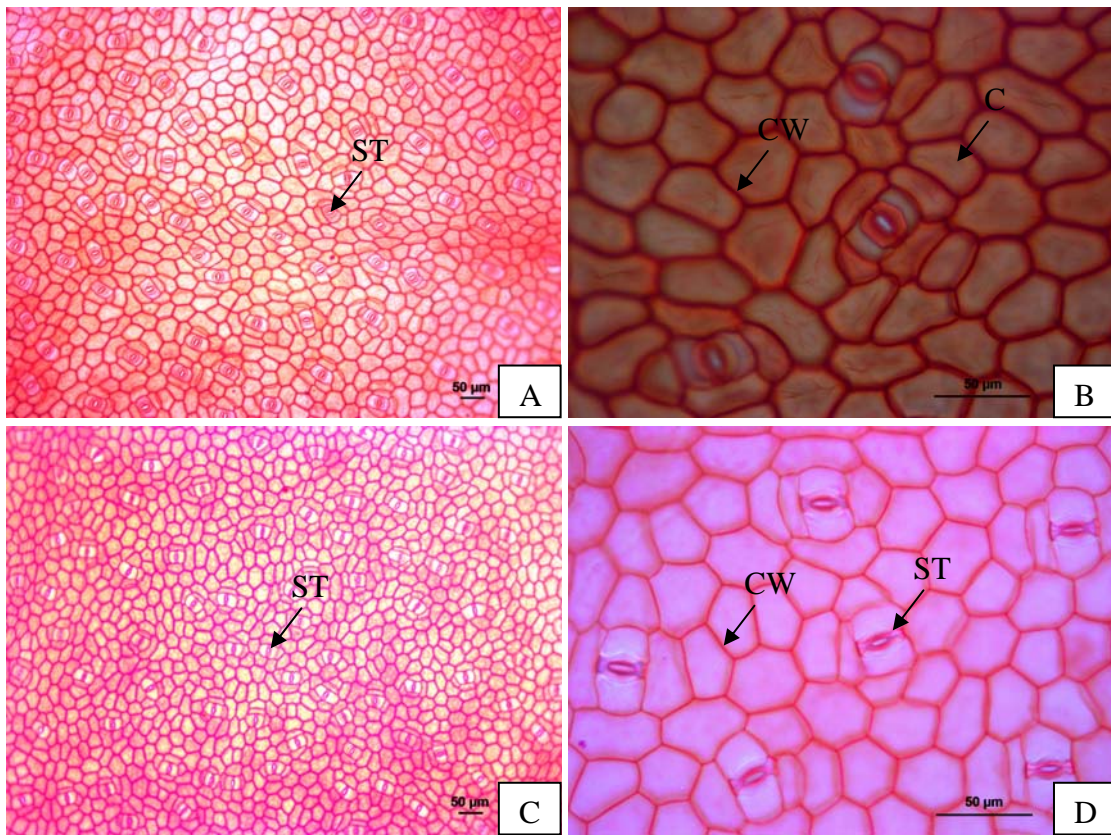




**Figure 4.4** Leaf surfaces anatomy of *Alocasia longiloba* Miq. (Scale bars = 50 µm)  
C = cuticle sculpturing, CW = cell walls, ST = stomata.

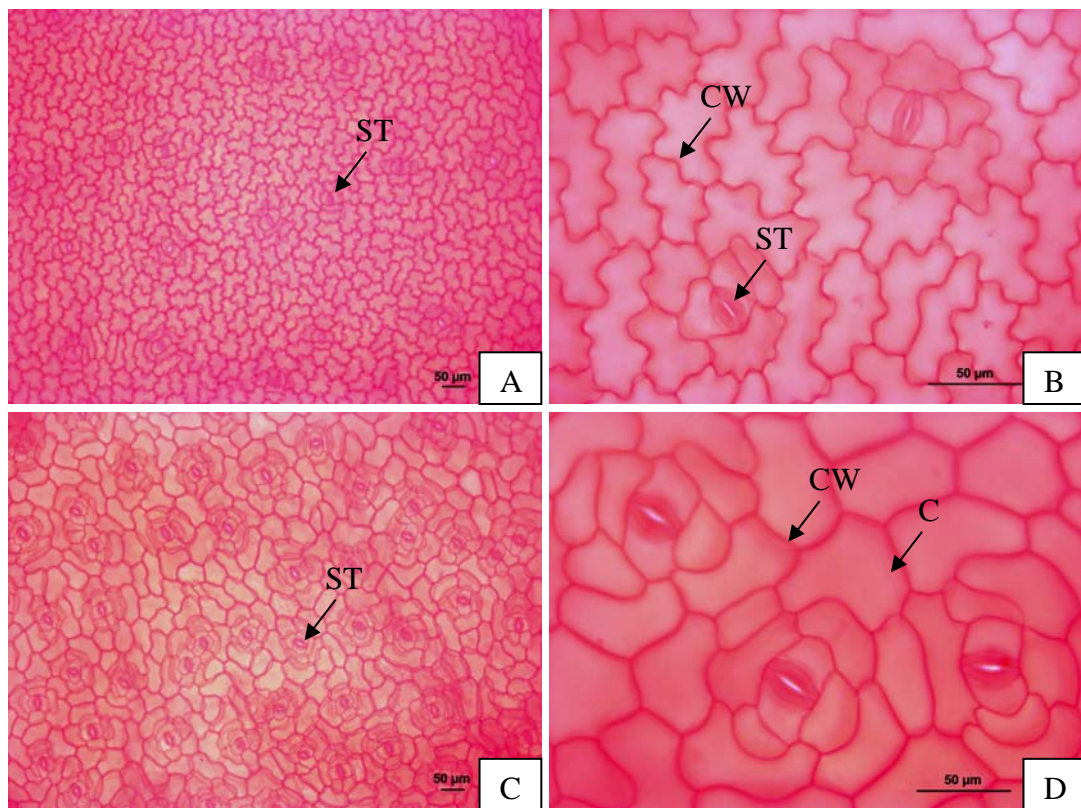
- A. An upper (adaxial) epidermis, 10x magnification light microscope
- B. An upper (adaxial) epidermis, 40x magnification light microscope
- C. A lower (abaxial) epidermis, 10x magnification light microscope
- D. A lower (abaxial) epidermis, 40x magnification light microscope





**Figure 4.5** Leaf surfaces anatomy of *Alocasia macrorrhizos* (L.) G. Don (Scale bars = 50  $\mu$ m) C = cuticle sculpturing, CW = cell walls, ST = stomata.

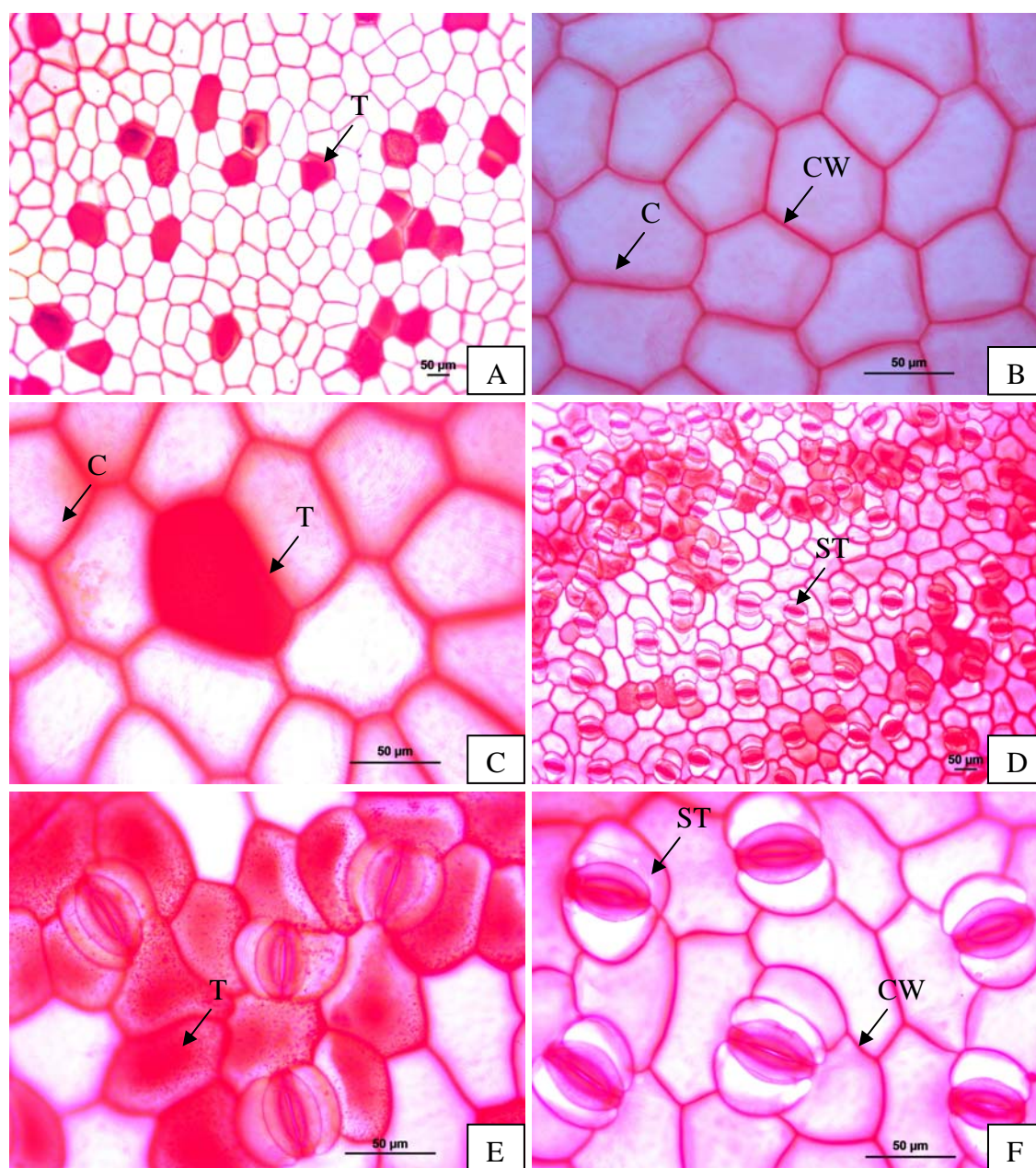
- A. An upper (adaxial) epidermis, 10x magnification light microscope
- B. An upper (adaxial) epidermis, 40x magnification light microscope
- C. A lower (abaxial) epidermis, 10x magnification light microscope
- D. A lower (abaxial) epidermis, 40x magnification light microscope



**Figure 4.6** Leaf surfaces anatomy of *Alocasia* sp. (Scale bars = 50 μm) C = cuticle sculpturing, CW = cell walls, ST = stomata.

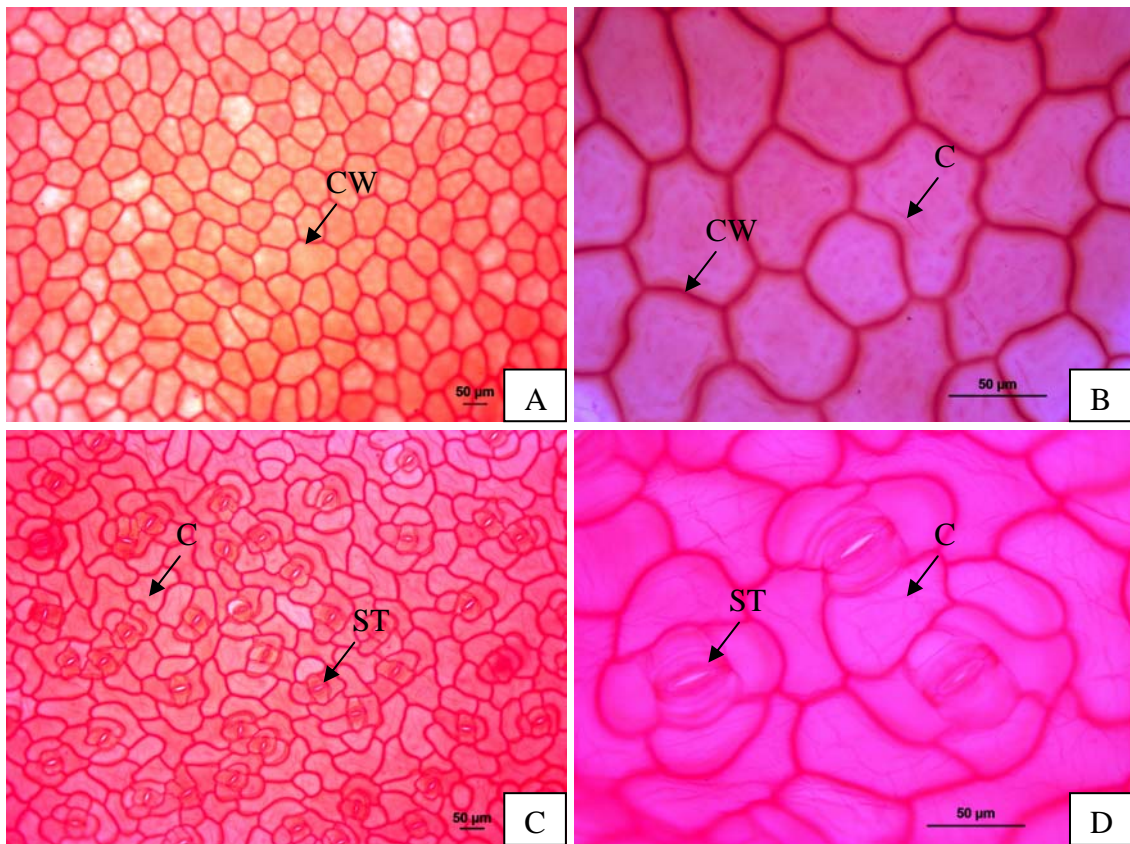
- A. An upper (adaxial) epidermis, 10x magnification light microscope
- B. An upper (adaxial) epidermis, 40x magnification light microscope
- C. A lower (abaxial) epidermis, 10x magnification light microscope
- D. A lower (abaxial) epidermis, 40x magnification light microscope





**Figure 4.7** Leaf surfaces anatomy of *Amorphophallus serrulatus* Hett. & A.Galloway (Scale bars = 50 µm) C = cuticle sculpturing, CW = cell walls, ST = stomata, T= tannin.

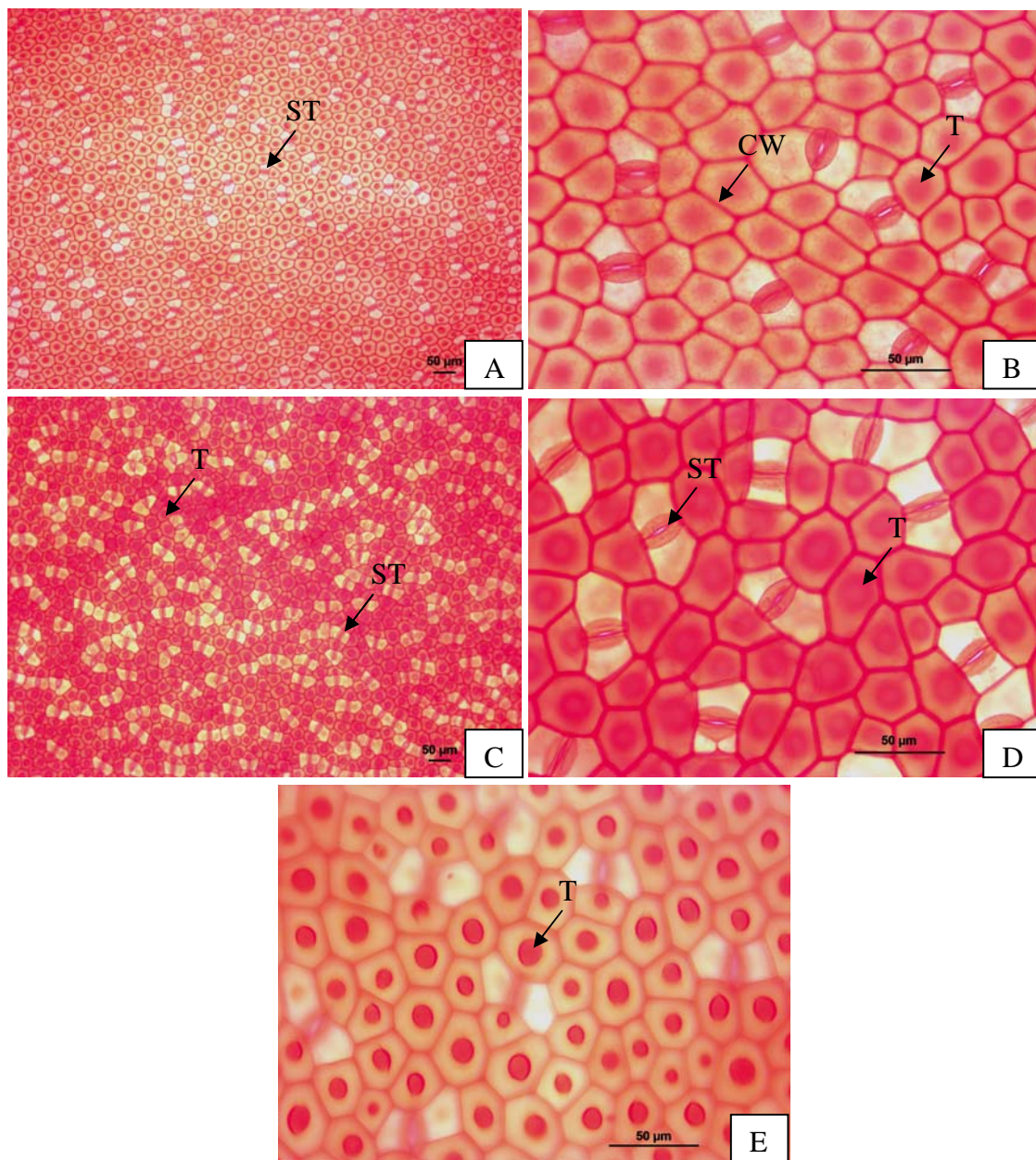
- A. An upper (adaxial) epidermis, 10x magnification light microscope
- B-C. An upper (adaxial) epidermis, 40x magnification light microscope
- D. A lower (abaxial) epidermis, 10x magnification light microscope
- E-F. A lower (abaxial) epidermis, 40x magnification light microscope



**Figure 4.8** Leaf surfaces anatomy of *Arisaema maxwellii* Hett. & Gusman (Scale bars = 50 µm) C = cuticle sculpturing, CW = cell walls, ST = stomata.

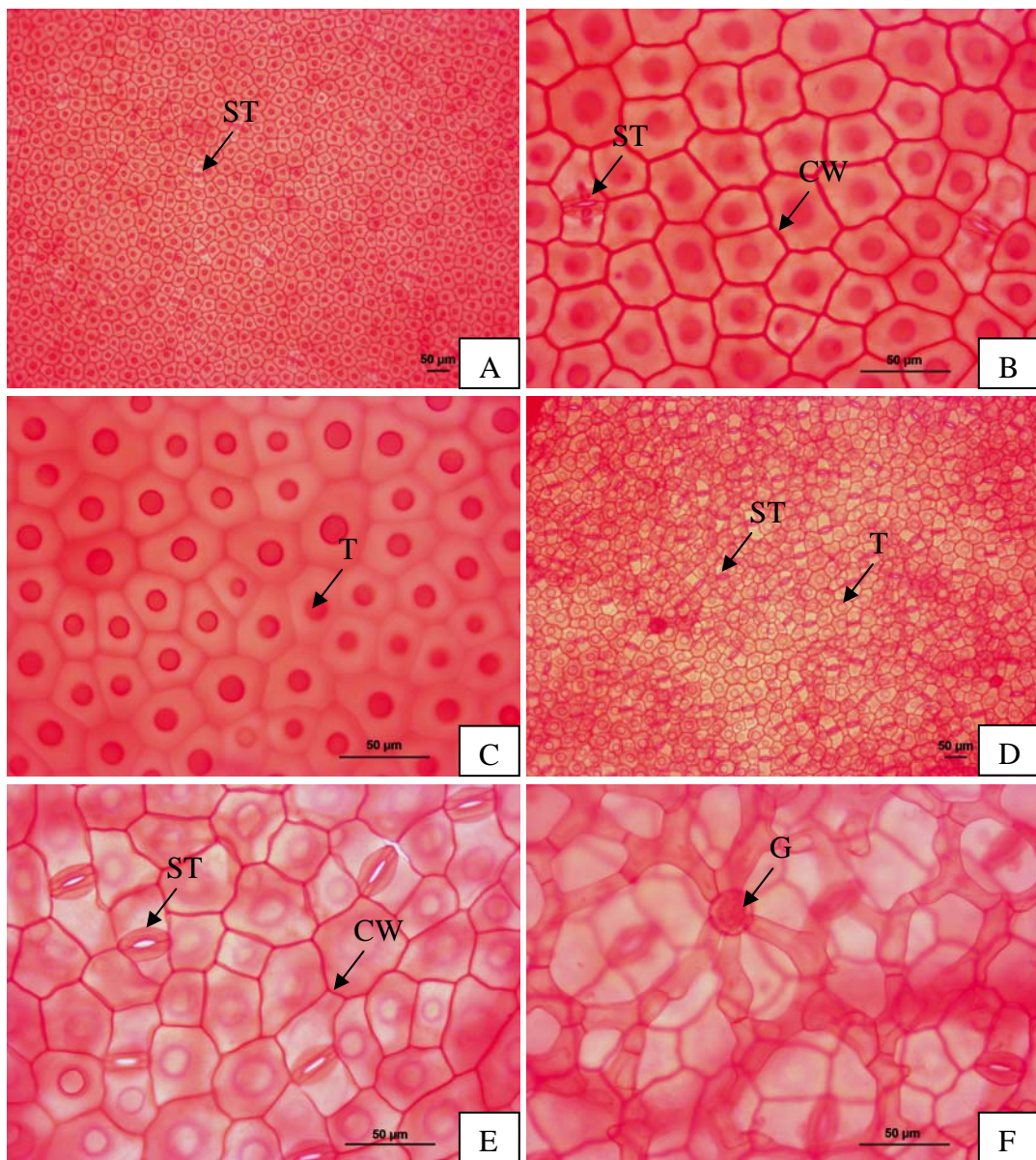
- A. An upper (adaxial) epidermis, 10x magnification light microscope
- B. An upper (adaxial) epidermis, 40x magnification light microscope
- C. A lower (abaxial) epidermis, 10x magnification light microscope
- D. A lower (abaxial) epidermis, 40x magnification light microscope





**Figure 4.9** Leaf surfaces anatomy of *Colocasia esculenta* (L.) Schott (Scale bars = 50 µm) CW = cell walls, ST = stomata, T = tannin.

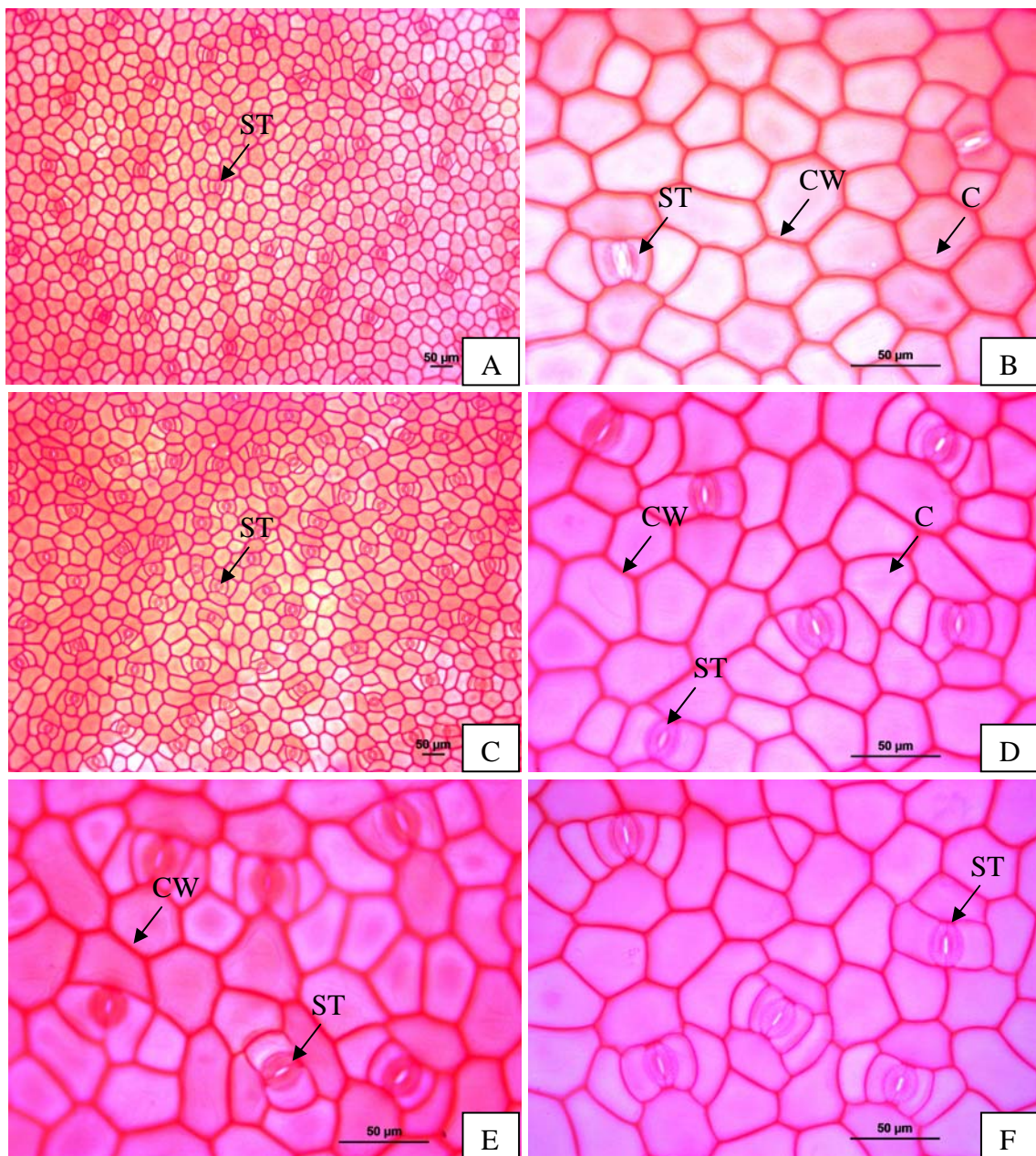
- A. An upper (adaxial) epidermis, 10x magnification light microscope
- B. An upper (adaxial) epidermis, 40x magnification light microscope
- C. A lower (abaxial) epidermis, 10x magnification light microscope
- D-E. A lower (abaxial) epidermis, 40x magnification light microscope



**Figure 4.10** Leaf surfaces anatomy of *Colocasia fallax* Schott (Scale bars = 50 μm)  
 CW = cell walls, G = glandular trichome, ST = stomata, T = tannin.

- A. An upper (adaxial) epidermis, 10x magnification light microscope
- B-C. An upper (adaxial) epidermis, 40x magnification light microscope
- D. A lower (abaxial) epidermis, 10x magnification light microscope
- E-F. A lower (abaxial) epidermis, 40x magnification light microscope

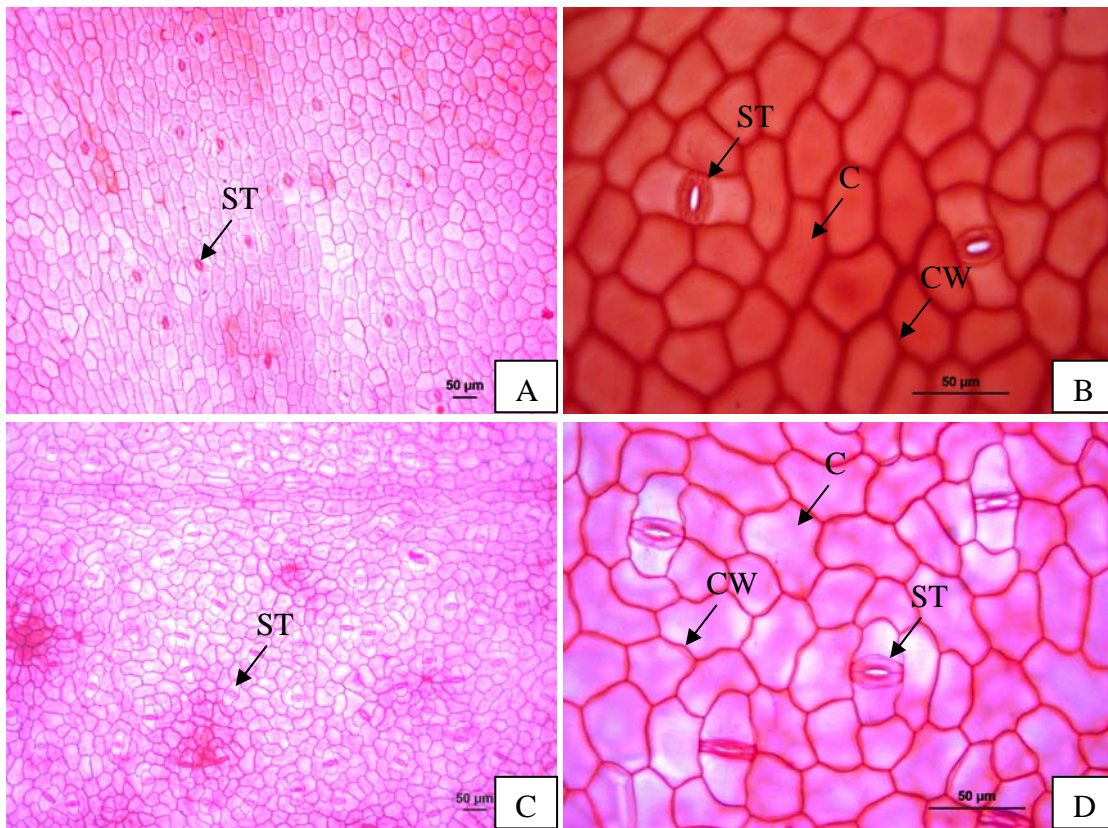




**Figure 4.11** Leaf surfaces anatomy of *Colocasia gigantea* (Blume) Hook.f. (Scale bars = 50 µm) C = cuticle sculpturing, CW = cell walls, ST = stomata.

- A. An upper (adaxial) epidermis, 10x magnification light microscope
- B. An upper (adaxial) epidermis, 40x magnification light microscope
- C. A lower (abaxial) epidermis, 10x magnification light microscope
- D-F. A lower (abaxial) epidermis, 40x magnification light microscope

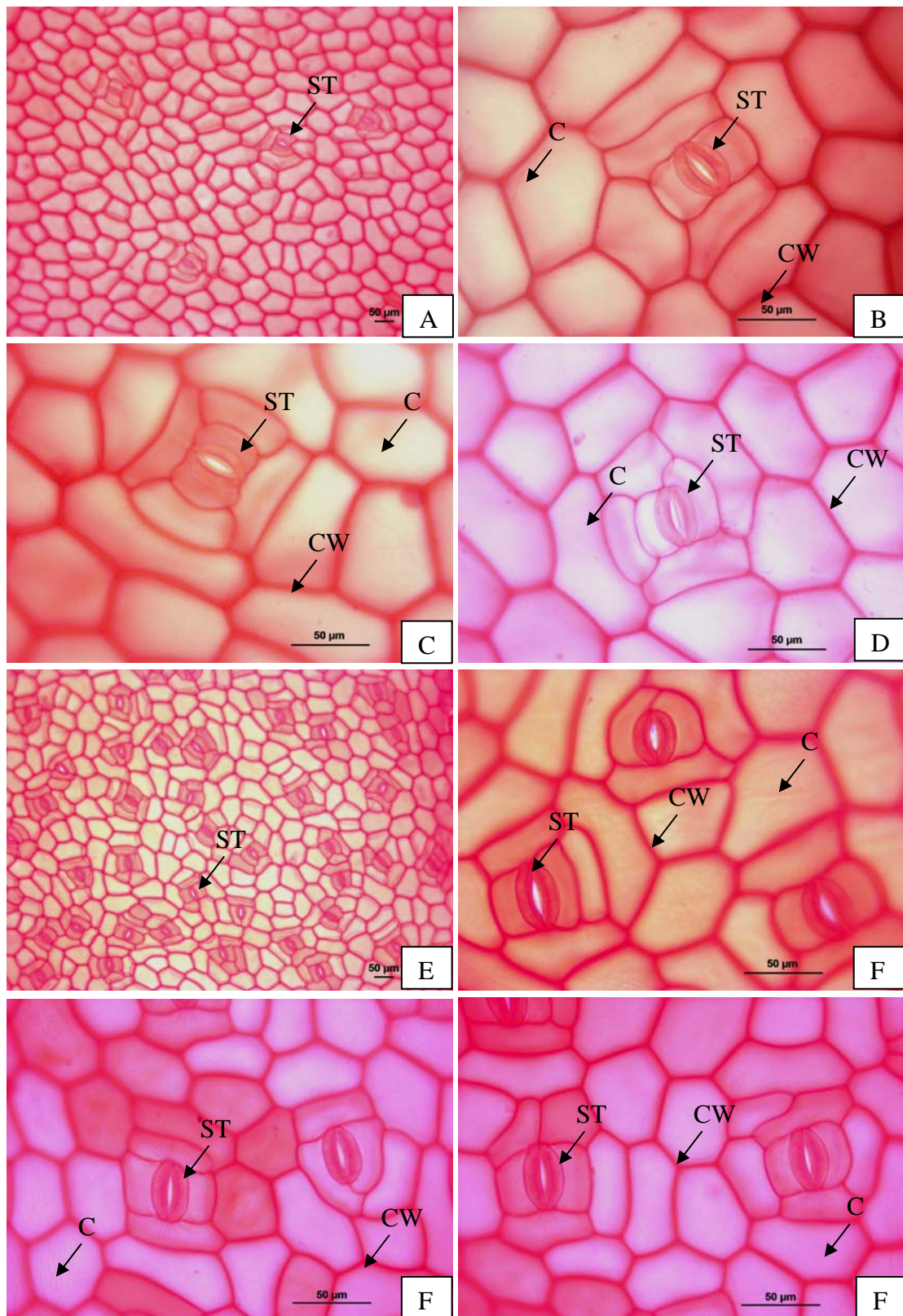




**Figure 4.12** Leaf surfaces anatomy of *Colocasia lihengiae* C.L.Long & K.M.Liu (Scale bars = 50  $\mu$ m) C = cuticle sculpturing, CW = cell walls, ST = stomata.

- A. An upper (adaxial) epidermis, 10x magnification light microscope
- B. An upper (adaxial) epidermis, 40x magnification light microscope
- C. A lower (abaxial) epidermis, 10x magnification light microscope
- D. A lower (abaxial) epidermis, 40x magnification light microscope

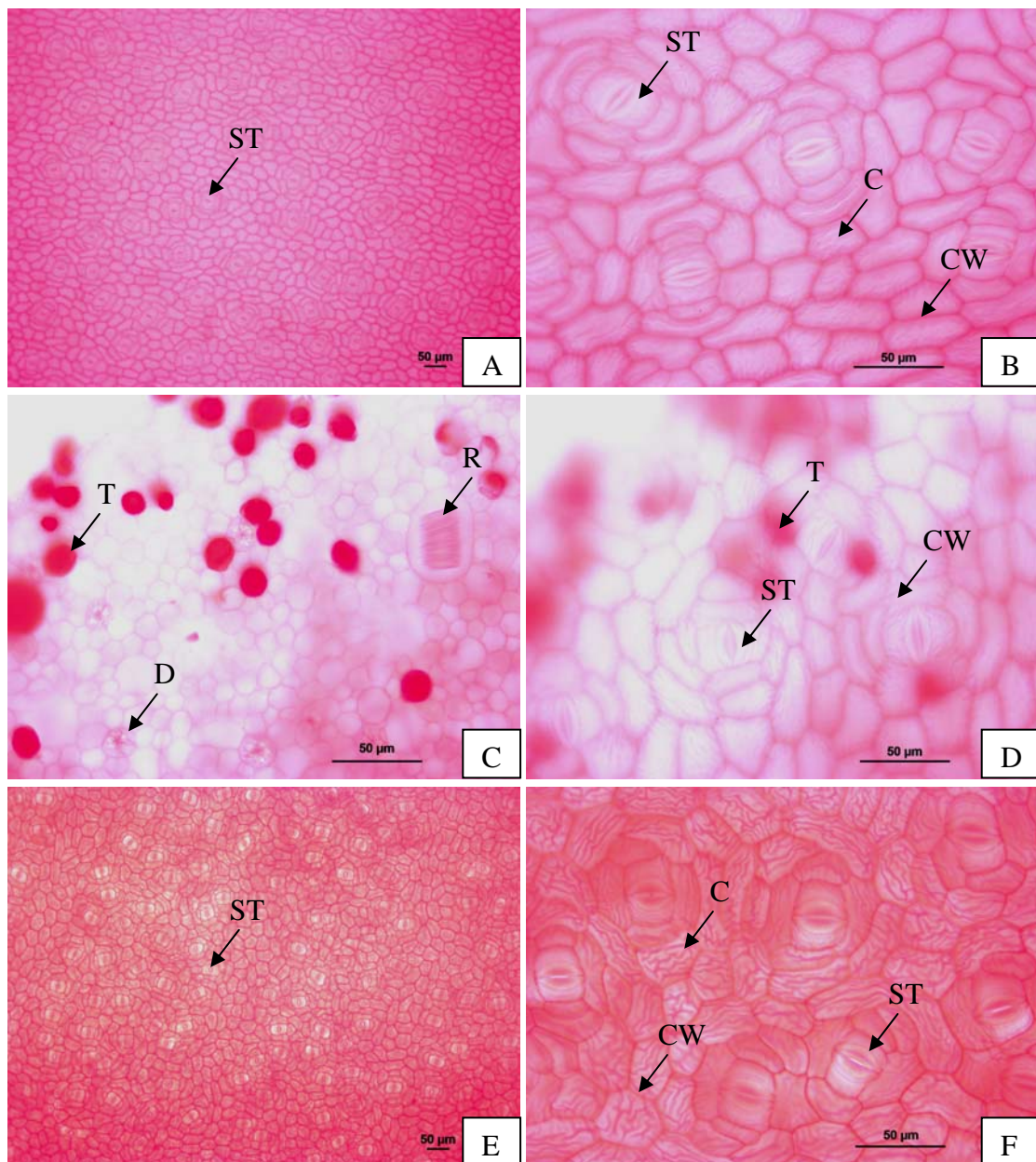




**Figure 4.13** Leaf surfaces anatomy of *Hapaline benthamiana* Schott (Scale bars = 50 µm) C = cuticle sculpturing, CW = cell walls, ST = stomata.

- A. An upper (adaxial) epidermis, 10x magnification light microscope
- B. An upper (adaxial) epidermis, 40x magnification light microscope
- C. A lower (abaxial) epidermis, 10x magnification light microscope
- D-E. A lower (abaxial) epidermis, 40x magnification light microscope



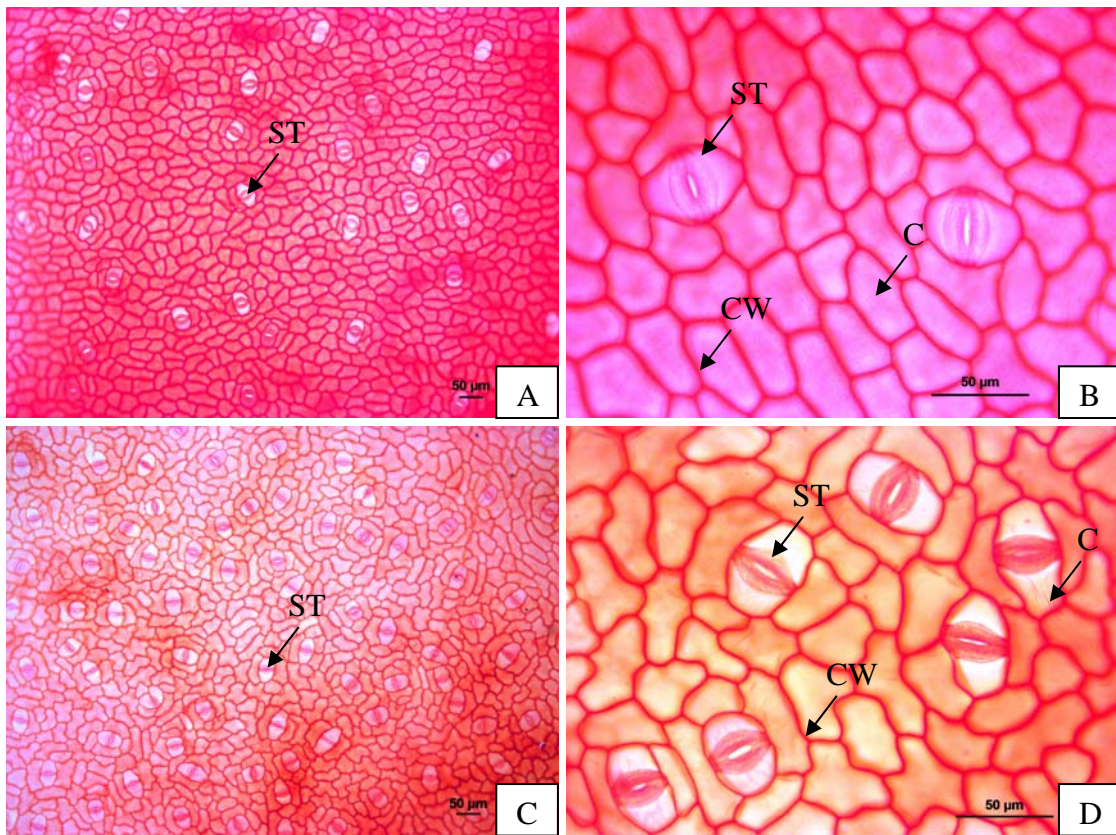


**Figure 4.14** Leaf surfaces anatomy of *Homalomena griffithii* (Schott) Hook.f. (Scale bars = 50 µm) C = cuticle sculpturing, CW = cell walls, D = druse crystal, R = raphides crystal, ST = stomata, T = tannin.

- A. An upper (adaxial) epidermis, 10x magnification light microscope
- B-D. An upper (adaxial) epidermis, 40x magnification light microscope
- E. A lower (abaxial) epidermis, 10x magnification light microscope
- F. A lower (abaxial) epidermis, 40x magnification light microscope



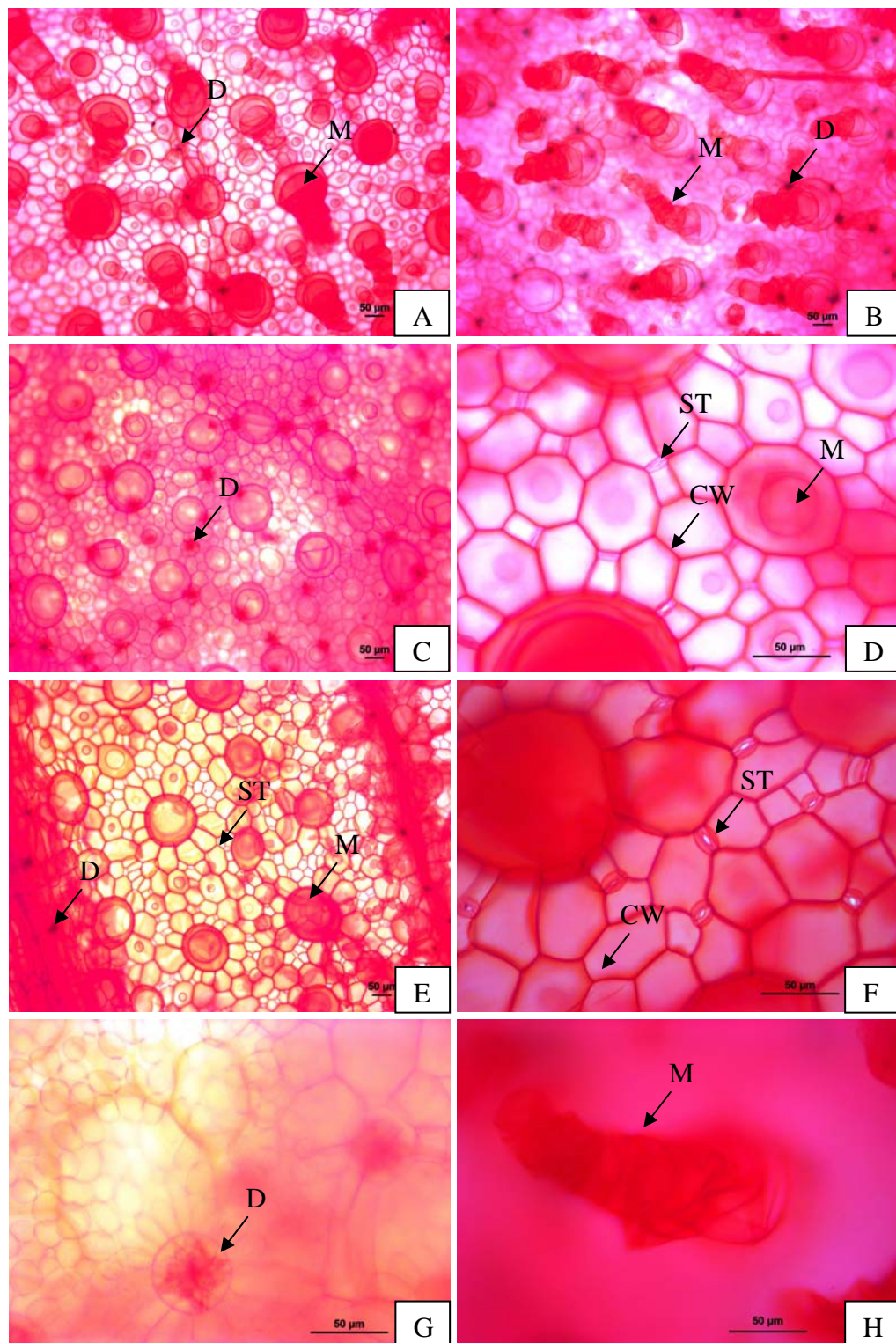




**Figure 4.15** Leaf surfaces anatomy of *Lasia spinosa* (L.) Thwaites (Scale bars = 50 µm)  
C = cuticle sculpturing, CW = cell walls, ST = stomata.

- A. An upper (adaxial) epidermis, 10x magnification light microscope
- B. An upper (adaxial) epidermis, 40x magnification light microscope
- C. A lower (abaxial) epidermis, 10x magnification light microscope
- D. A lower (abaxial) epidermis, 40x magnification light microscope



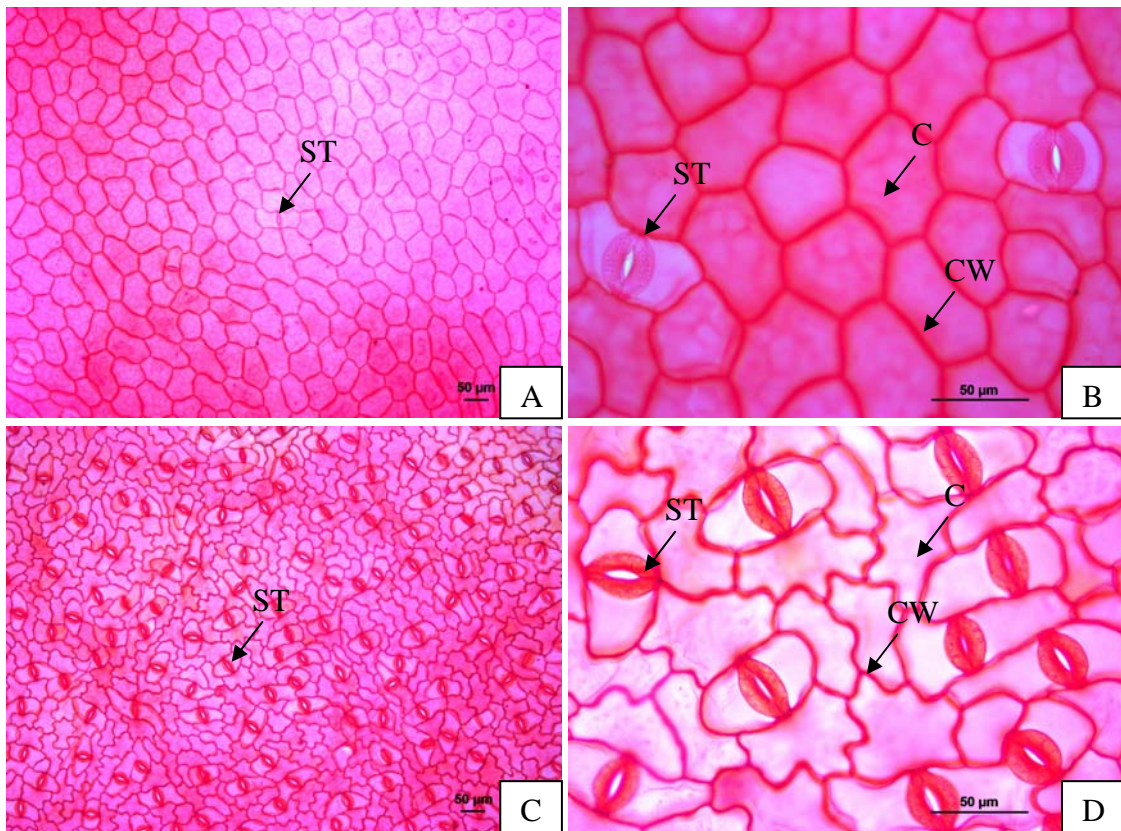


**Figure 4.16** Leaf surfaces anatomy of *Pistia stratiotes* L. (Scale bars = 50 µm) C = cuticle sculpturing, CW = cell walls, D = druse crystal, M = multi-cellular trichomes, ST = stomata.



- A-C. An upper (adaxial) epidermis, 10x magnification light microscope
- D. An upper (adaxial) epidermis, 40x magnification light microscope
- E. A lower (abaxial) epidermis, 10x magnification light microscope
- F-H. A lower (abaxial) epidermis, 40x magnification light microscope

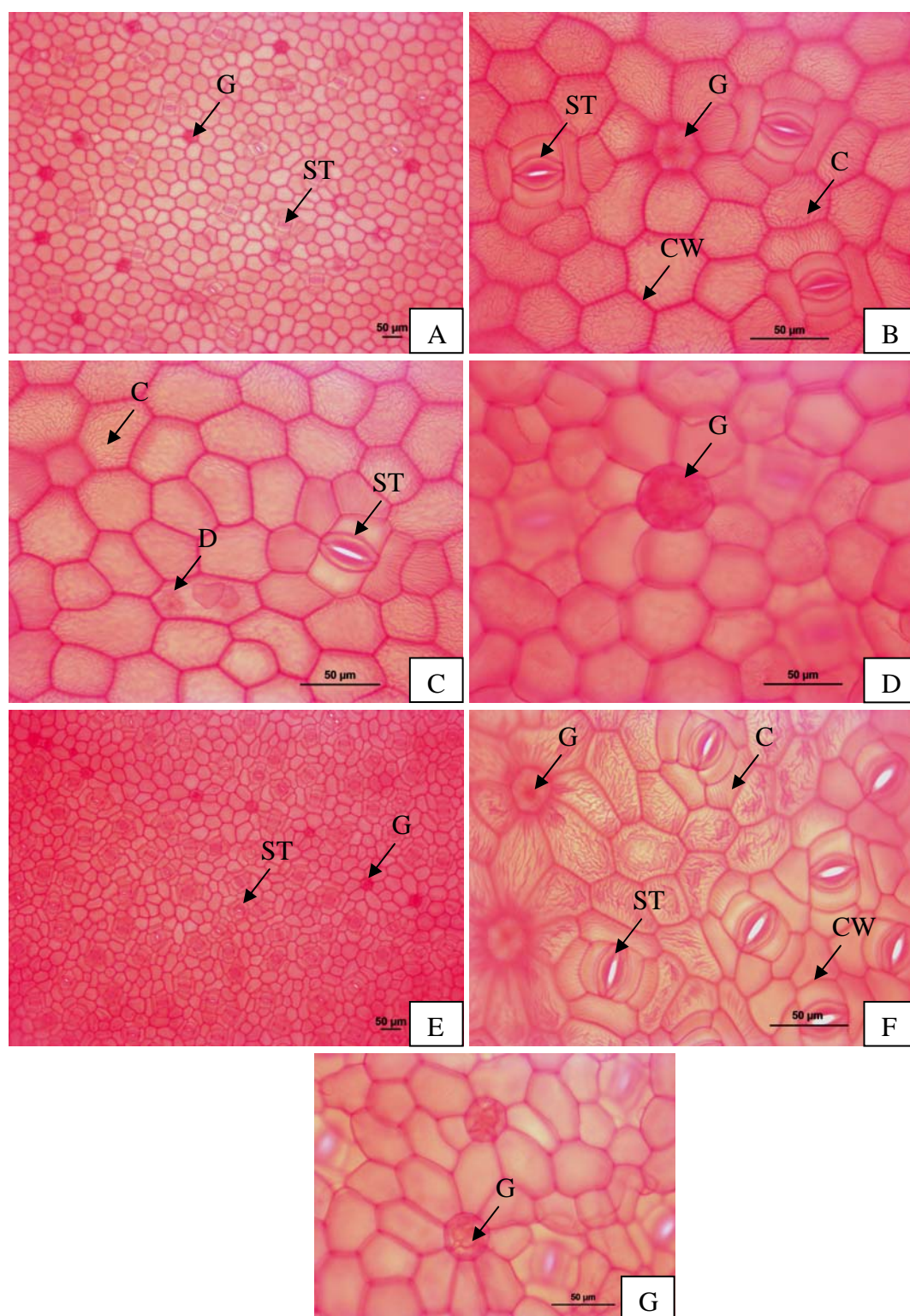




**Figure 4.17** Leaf surfaces anatomy of *Pycnospatha palmata* Gagnep. (Scale bars = 50 μm) C = cuticle sculpturing, CW = cell walls, ST = stomata.

- A. An upper (adaxial) epidermis, 10x magnification light microscope
- B. An upper (adaxial) epidermis, 40x magnification light microscope
- C. A lower (abaxial) epidermis, 10x magnification light microscope
- D. A lower (abaxial) epidermis, 40x magnification light microscope



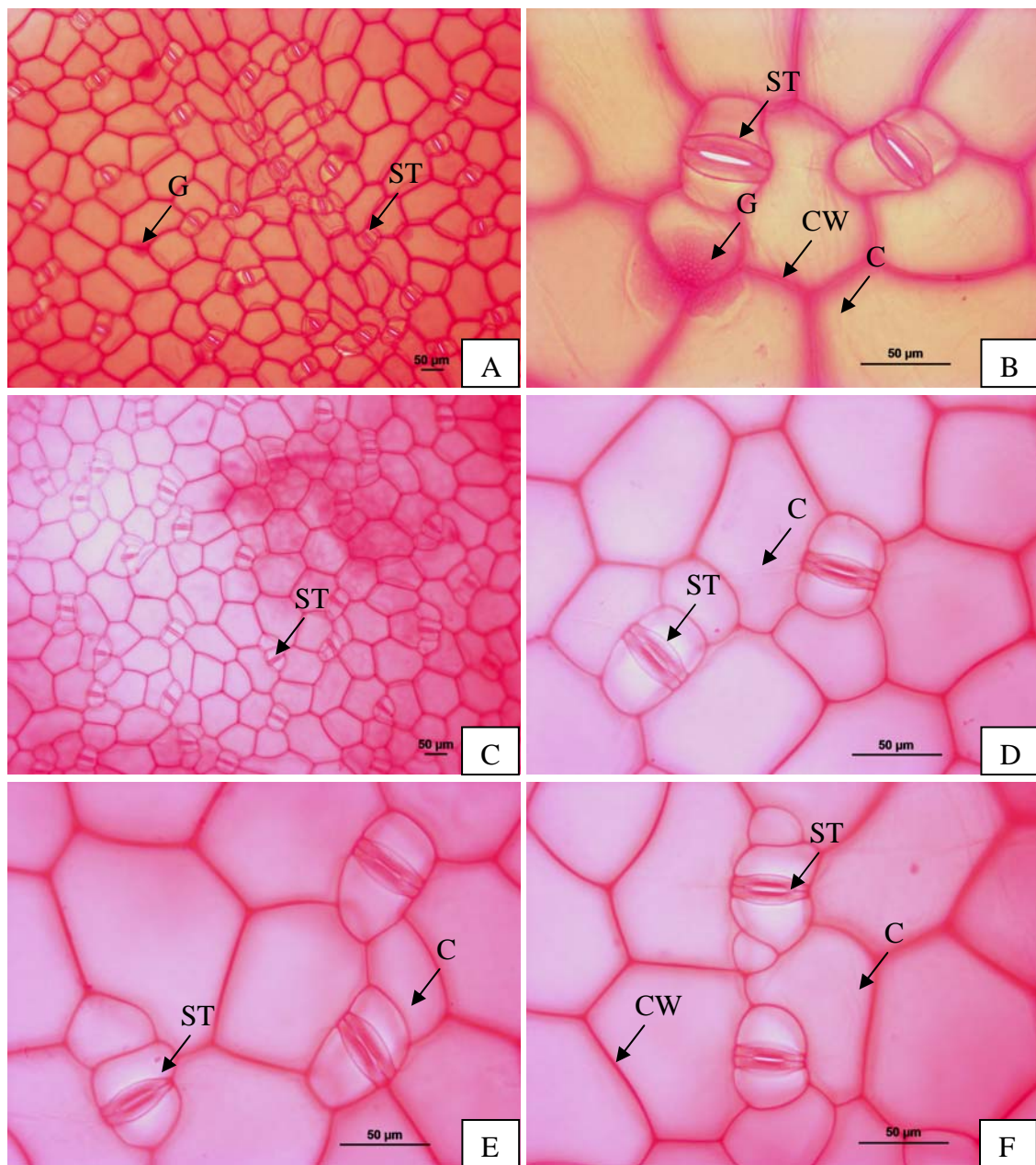


**Figure 4.18** Leaf surfaces anatomy of *Schismatoglottis calyptrata* (Roxb.) Zoll. & Moritzi (Scale bars = 50 µm) C = cuticle sculpturing, CW = cell walls, D = druse crystal, G = glandular trichomes, ST = stomata.

- A. An upper (adaxial) epidermis, 10x magnification light microscope
- B-D. An upper (adaxial) epidermis, 40x magnification light microscope
- E. A lower (abaxial) epidermis, 10x magnification light microscope
- F-G. A lower (abaxial) epidermis, 40x magnification light microscope



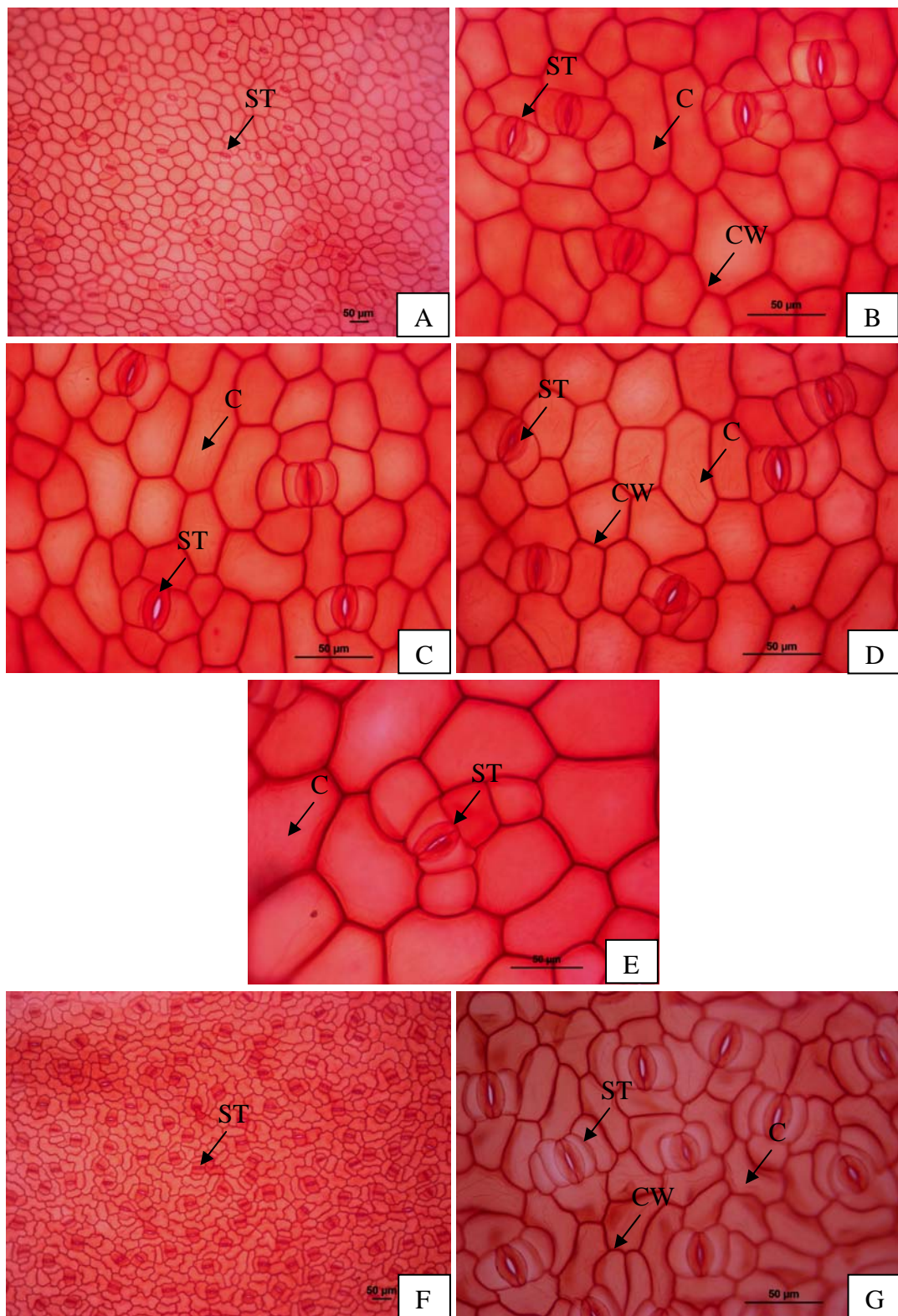




**Figure 4.19** Leaf surfaces anatomy of *Typhonium glaucum* Hett. & Sookchaloem (Scale bars = 50 μm) C = cuticle sculpturing, CW = cell walls, G = glandular trichomes, ST = stomata.

- A. An upper (adaxial) epidermis, 10x magnification light microscope
- B. An upper (adaxial) epidermis, 40x magnification light microscope
- C. A lower (abaxial) epidermis, 10x magnification light microscope
- D-F. A lower (abaxial) epidermis, 40x magnification light microscope





**Figure 4.20** Leaf surfaces anatomy of *Typhonium trilobatum* (L.) Schott (Scale bars = 50 µm) C = cuticle sculpturing, CW = cell walls, ST = stomata.

- A. An upper (adaxial) epidermis, 10x magnification light microscope
- B-D. An upper (adaxial) epidermis, 40x magnification light microscope
- E. A lower (abaxial) epidermis, 10x magnification light microscope
- F-G. A lower (abaxial) epidermis, 40x magnification light microscope



## CHAPTER 5

### TRADITIONAL USES STUDIES

#### 5.1 Introduction

Ethnobotany is the study of how and for what reasons people use plants. Usage usually relates to people's conceptualization of the importance of plants, medicinally and otherwise, and their experience of plants occurring in their local environment. The use of plants for medicinal purposes originated from the beginning of civilization, as evidenced by the earliest recorded uses found in Babylon and in ancient Egypt. Ancient Egyptians believed that medicinal plants were even effective in the afterlife of their Pharaohs, as indicated by the plants recovered from the Giza pyramids (Veilleux & King 1996).

If one considers the questions how and why people are using plants, the ethnobotanist approaches this problem by gathering data from living people. In this manner, an understanding is created not only of the present uses of plants, but also of the importance of plants for food, medicine, construction, etc. in their past existence. It also gives an indication of people's traditional ecological knowledge specifically related to plants and the influence of this knowledge on the research and methods used in ethnobotany. The concept of ethnobotany started to develop in 1985 after a lecture in Philadelphia by Dr. John Harshberger, where he used the term "ethno-botany" to describe his field of study, namely: "the study of plants used by primitive and aboriginal people" (Robbins *et al.*, 1916).

Traditional knowledge on biodiversity concerns the names, uses and management of plants and animals as perceived by the local or indigenous people in a given area. Folk names of plants and animals are the roots of traditional biodiversity knowledge (Khasbagan and Soyolt, 2008). Besides, folk systems of naming and classification of plants and animals are transmitted from generation to generation is constantly created by communities and groups in response to their environment (Tantiado, 2012).

#### 5.2 Traditional uses

##### 5.2.1 Definition and scope of traditional used

Utilization of plants for medicinal and the consumption, management and valuation of wild plants are central aspects of the traditional knowledge in many human populations. Thus, plants gathering, the diffusion and conservation of knowledge within the community are traditional practices that have contribution to the subsistence of many cultures. In most of the societies the medical system coexists with several traditional systems. These traditional medical systems are generally based on the uses of natural and local products which are commonly related to the people's perspective on the world and life (Toledo *et al.*, 2009).





## 5.2.2 Literature reviews of Traditional Uses

### *Alocasia*

Burkill (1966) studied the traditional use of *Alocasia indica* (Roxb.) Schott for a trunk curry in India, a an important food must be cooked in boiling water, then pour about two times before eating In Bengal.

Burkill (1966) reported the stem of *Alocasia macrorrhiza* (L.) Schott. use as food, but must be boiled early.

Burkill (1966) used tuber of *Alocasia cucullata* (Lour.) Schott. for foods and used herb in Chinese.

Burkill (1966) used of *Alocasia denudata* Engl. which found that to have grown as a medicinal plant and toxic latex used to make arrow poison.

Perry (1980) studied the traditional used of *Alocasia indica* (Roxb.) Schott by used rub the oil from the treatment of skin diseases, latex mixed with ashes from the trunk when applied at leg swelling in pregnant women, and fresh leaves applied by the suspension snake bite poisoning caused.

Perry (1980) reported ethnobotany of *Alocasia macrorrhiza* (L.) Schott by eaten tuber for diuretic, solving wind has because of capillary in the brain broken. Arthritis treating Stalk of leaves burned to relieve toothache.

Perry (1980) used the stems of *Alocasia longilola* Miq. for the treatment of festering wounds with animals.

Kala (2005) used root of *Alocasia forniculata* (Roxb.) Schott. for crack of heels.

### *Arisaema*

Haq *et al.* (2011) studied the ethnobotany of *Arisaema flavum* Forssk. by used fruits for cough and cold.

### *Amorphophallus*

Kala (2005) used *Amorphophallus paeoniifolius* (Dennst.) Nicolson for piles.

### *Colocasia*

Burkill (1966) studied the traditional use of *Colocasia esculenta* (L.) Schott found that plants which has grown in Southeast Asia. Tuber has a high carbohydrate food, young leaves and stalk soft and edible and the stems wound healing snake bites.

Burkill (1966) studied the traditional use of *Colocasia gigantea* (Blume.) Hook.f. by used seed for eaten as a condiment.

Perry (1980) studied the traditional use of *Colocasia esculenta* (L.) Schott by used seed, leaf, stalk as a medicinal herb in China. Used as a tonic for pregnant women after delivery in Philippines. Used the leaves as a vegetable eaten the Solomon Islands.

Nicolson (1987) studied the traditional use of *Colocasia esculenta* (L.) Schott found that the growing popularity as food in tropical Asia zone.

Kala (2005) studied the traditional use of *Colocasia affinis* Schott by used leaf for fever, respiratory disorder.





### *Lasia*

Mir *et al.* (2014) studied the *Lasia spinosa* (L.) Thwaites by use the decoction of rhizomes mixed with sugar is consumed orally for poisoning

### *Zantedeschia*

Mir *et al.* (2014) studied the *Zantedeschia aethiopica* (L.) Spreng by use leaf juice applied to cuts, injuries and to relieve uterine contraction.

## 5.2.3 The traditional used studied in Thailand

Pongphangan and Bhuprasert (1991) studied the traditional use of *Alocasia indica* (Roxb.) Schott by used young leaves and leaf stalk, when the outer bark off a fresh vegetable dip and do eaten curry.

Pongphangan and Bhuprasert (1991) studied the traditional use of *Alocasia odora* (Roxb.) C. Koch by used leaf stalk for the food with the fire cooked first and then peels off the outer skin to cook. Bring to a boil or change the water 2-3 times before eating.

Vamanon and Subcharoen (1997) studied the traditional use of *Colocasia gigantea* (Blume.) Hook.f. by used tuber for kindle fire is the fever, toxic heat, lethargic, toxic tan, a disease of children, tuber fresh to relieve etadan the stomach, heal wounds, bites, freckles, pus bite, Stalk and young leaves eaten as a vegetable.

Boonyawatprapatson (1998) studied the traditional use of *Colocasia esculenta* (L.) Schott by used tuber for as a laxative, milk, hemostasis, water from the stems the poisonous scorpions, fever, stems mask for wound healing. Water from the bleeding used rubber wart removal

Werukamkul and Ampornpan (2013) studied the *Lasia spinosa* (L.) Thwaites of Loei provinces in Thailand. Therefore, local people are usually eaten the local vegetable for is the food.

Senavongse *et al.* (2015) studied the *Lasia spinosa* (L.) Thwaites in the Northeast region in Thailand. Local people are usually eaten the local vegetable *L. spinosa* is a one of local vegetable. That is used as in gradient in food as well. It has local name is Puk-Nam. People in the community popular with young shoots, leaves and flowers, soft boiled, steamed, boiled, cooked before being eaten with chili. Because, if eaten fresh to toxin of the *L. spinosa* and sprigs fresh containing hydrocyanic acid. So, before eating it must to be cooked and the *L. spinosa* is characteristically to the sweet taste freshly.



## 5.3 Material and Methods

### 5.3.1 Material

1. Notebook
2. Pen
3. Pencil
4. Eraser
5. Plastic bag
6. Elastic
7. Camera

### 5.3.2 Methods

#### 5.3.2.1 Traditional uses study

##### Field survey and data collection

During field trip and survey collect data for traditional uses of Araceae. Traditional used data was collected by interviewing the informants such as leaders, folk healers, matriarchs, and villagers. Scopes of interview were local name, food plants, medicinal plants, ornamental plants, rituals plants and also parts of plants used.

## 5.4 Results

Traditional used of the Araceae family in the Northeastern Thailand from the field trips was conducted by sample collection and interviewing villagers. The study area research was conducted in 20 provinces in Northeast, Thailand as follows; Amnatcharoen, Buengkan, Buriram, Chaiyaphum, Kalasin, Khonkaen, Loei, Mahasarakham, Mukdahan, Nakhonphanom, Nakhonrajchasi, Nongbualamphu, Nongkhai, Roi-Et, Sakhonakorn, Sisaket, Surin, Ubonratchathani, Udonthani and Yasothorn (January, 2015 to April, 2017). Found that the 10 species 8 genera traditional used of plants 5 aspects. Araceae can be used for foods, medicines, ornamental plants, commercial propagation plants and also in rituals. (Table 5.1, Figure 5.1).

### 5.4.1 Traditional used food

#### Food for humans

*Amorphophallus* (L.) Schott by used the young flower, rhizome, young leaves and young stem for foods (Table 5.1, Figure 5.1).

*Colocasia esculenta* (L.) Schott leaves, young leaves and young stem for foods (Table 5.1, Figure 5.2).

*C. gigantea* (Blume) Hook.f used the leaves, rhizome, stem, young leaves, young stem for foods (Table 5.1, Figure 5.3).

*Lasia spinosa* L. used the young leaves and young stem for foods (Table 5.1, Figure 5.4).

### Food for animals

*Colocasia esculenta* (L.) Schott used stem, young leaves and young stem food for animals (Table 5.1, Figure 5.5).

#### 5.4.2 Traditional used for medicines

*Colocasia esculenta* (L.) Schott used leaves, stem, young leaves and young stem (Table 5.1, Figure 5.6).

*C. gigantea* (Blume) Hook.f used the rhizome (Table 5.1, Figure 5.7).

*Scindapsus officinalis* (Roxb.) Schott used the leaves and stem (Table 5.1).

#### 5.4.3 Traditional used ornamental plants

*Aglaonema modestum* Schott ex Engl. (Table 5.1) use plant as ornamental plants.

*Alocasia cucullata* (Lour.) G.Don (Table 5.1, Figure 5.8) use plant as ornamental plants.

*A. macrorrhizos* (Lour.) G.Don (Table 5.1, Figure 5.9) use plant as ornamental plants.

*Lasia spinosa* L. (Table 5.1, Figure 5.10) use plant as ornamental plants.

*Pistia stratiotes* L. (Table 5.1, Figure 5.11) use plant as ornamental plants.

#### 5.4.3 Traditional used commercial propagation plants

*Amorphophallus* (L.) Schott by used young stem (Table 5.1, Figure 5.1A).

*Aglaonema modestum* Schott ex Engl. used stem (Table 5.1).

*Alocasia cucullata* (Lour.) G.Don used the stem (Table 5.1).

*A. macrorrhizos* (Lour.) G.Don used stem (Table 5.1).

*Colocasia gigantea* (Blume) Hook.f used young stem (Table 5.1, Figure 5.7).

#### 5.4.5 Traditional used in rituals

*Alocasia cucullata* (Lour.) G.Don (Table 5.1, Figure 5.12) use plant as front of the house to make money.

*Typhonium trilobatum* (L.) Schott (Table 5.1, Figure 5.13) use plant as in front of the house to chase ghosts.

### 5.5 Conclusion and Discussion

Traditional used of the Araceae family in the Northeastern Thailand from the filed trips was conducted by sample collection and interviewing villagers. Found that the 10 species 8 genus traditional used of plants 5 aspects.

Surveys of traditional used of *Aglaonema modestum* are in agreement with Hutapat (2010) reported used as ornamental plants.

Surveys of traditional used of *Pistia stratiotes* are in agreement with Chayamarit (2008) reported used as ornamental plants.



The common traditional used were *Colocasia esculenta* (3 aspects), *Colocasia gigantea* (3 aspects), *Alocasia cucullata* (3 aspects), *Aglaonema modestum* (2 aspects), *Amorphophallus brevispathus* (2 aspects), *Alocasia macrorrhizos* (2 aspects), *Lasia spinosa* (2 aspects), *Pistia stratiotes* (1 aspects), *Scindapsus officinalis* (1 aspects), *Typhonium trilobatum* (1 aspects), respectively. The most widely used part was young stem and young leaves for foods and medicines



**Table 5.1** Comparisons of some traditional used of the 10 species 8 genera of Araceae in Thailand

Traditional used		Species	Parst of uses									Thai local name
			P	R	Rz	YS	S	YL	L	Fl	Fr	
Foods	Humans	<i>Amorphophallus brevispathus</i>	-	-	✓	✓	-	✓	-	✓	-	Buk i rok khao
		<i>Colocasia esculenta</i>	-	-	-	✓	-	✓	✓	-	-	Phueak or Bon
		<i>Colocasia gigantea</i>	-	-	✓	✓	✓	✓	✓	-	-	Khun
		<i>Lasia spinosa</i>	-	-	-	✓	-	✓	-	-	-	Phuk-Nam
	Animals	<i>Colocasia esculenta</i>	-	-	-	✓	✓	✓	✓	-	-	Phueak or Bon
Medicines	Humans	<i>Colocasia esculenta</i>	-	-	-	✓	✓	✓	✓	-	-	Phueak or Bon
		<i>Colocasia gigantea</i>	-	-	✓	✓	✓	✓	✓	-	-	Khun
		<i>Scindapsus officinalis</i>	-	-	-	-	✓	-	✓	-	✓	Hi khwai
Ornamental plants		<i>Aglaonema modestum</i>	✓	-	-	-	-	-	-	-	-	Khiao muen pee
		<i>Alocasia cucullata</i>	✓	-	-	-	-	-	-	-	-	Wan nang-kwak
		<i>A. macrorrhizos</i>	✓	-	-	-	-	-	-	-	-	Kradat
		<i>Lasia spinosa</i>	✓	-	-	-	-	-	-	-	-	Phuk-Nam
		<i>Pistia stratiotes</i>	✓	-	-	-	-	-	-	-	-	Jok
Commercial propagation plants		<i>Amorphophallus brevispathus</i>	-	-	-	✓	-	✓	-	-	-	Buk i rok khao
		<i>Aglaonema modestum</i>	✓	-	-	-	-	-	-	-	-	Khiao muen pee
		<i>Alocasia cucullata</i>	✓	-	-	-	-	-	-	-	-	Wan nang-kwak
		<i>A. macrorrhizos</i>	✓	-	-	-	-	-	-	-	-	Kradat
		<i>Colocasia gigantea</i>	✓	-	✓	✓	✓	-	-	-	-	Khun
Rituals		<i>Alocasia cucullata</i>	✓	-	-	-	-	-	-	-	-	Wan nang-kwak
		<i>Typhonium trilobatum</i>	✓	-	-	-	-	-	-	-	-	Wan khee

Note: Fl = flower, Fr = fruit, L = leaves P = plant, R = root, Rz = rhizome, S = stem, YL = young leaves, YS = young stem, “-” = absence, “✓” = presence



### Traditional used food for humans



**Figure 5.1** Food for humans of *Amorphophallus* (L.) Schott

- A. Food by village
- B. Curry or Kaeng i lok



**Figure 5.2** Foods for humans of *Colocasia esculenta* (L.) Schott

- A. Curry or Kaeng Bon-Jeud
- B. Curry or Kaeng Bon-Hwan
- C. Curry or Kaeng Som Bon-Hwan
- D. Taro in coconut milk
- E. The leaf blade wraps yeast rice
- F. The leaf blade wraps shrimp
- G. Curry or Kaeng i lok



**Figure 5.3** Foods for humans of *Colocasia gigantea* (Blume) Hook.f.

- A, C. Prepare to cook of Kaeng Toon
- B. Food by village market of Toon
- D. Curry or Kaeng Toon in coconut milk
- E. Curry or Kaeng Toon
- F, I. Petiole of Toon eat with green papaya salad
- G. Cooking of Kaeng Toon in fermented fish
- H. Kaeng Toon in fermented fish





**Figure 5.4** Foods for humans of *Lasia spinosa* L.

- A, C. Species soft boiled of Phuk-Nam
- B, F. Steamed of Phuk-Nam
- D. Young shoots of Phuk-Nam eat with green papaya salad
- E. Curry or Kaeng Som of Phuk-Nam
- G. Fried with oyster sauce Phuk-Nam

### Traditional used food for animals



**Figure 5.5** Food for animals of *Colocasia esculenta* (L.) Schott

- A. Prepare to cook for animals
- B. Boiled Petiole of Bon
- C. Feeding animals
- D. The animal eats its food

### Traditional used for medicines



**Figure 5.6** Foods for medicines of *Colocasia esculenta* (L.) Schott

- A. Prepare cook for Mok Bon
- B. Fire grill of Mok Bon





**Figure 5.7** Foods for medicines of *Colocasia gigantea* (Blume) Hook.f .

- A. Meiying hmi of Toon
- B-C. Pound of Toon
- D. Prepare cook for Ya Tum
- E. Rhizome of Toon
- F. Materials of Ya Tum
- G. Ya Tum of Toon
- H. The villagers eat Ya Tum

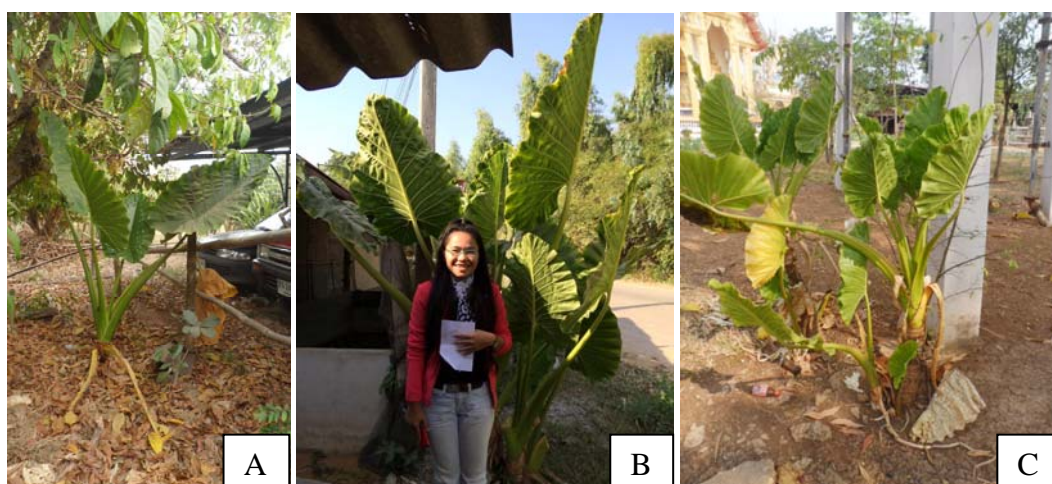
### Traditional used ornamental plants



**Figure 5.8** Ornamental plants of *Alocasia cucullata* (Lour.) G.Don

A-C. Ornamental plant according on houses

D-E. Ornamental plant according on roadside restaurant



**Figure 5.9** Ornamental plants of *Alocasia macrorrhizos* (Lour.) G.Don

A-B. Ornamental plant according on houses

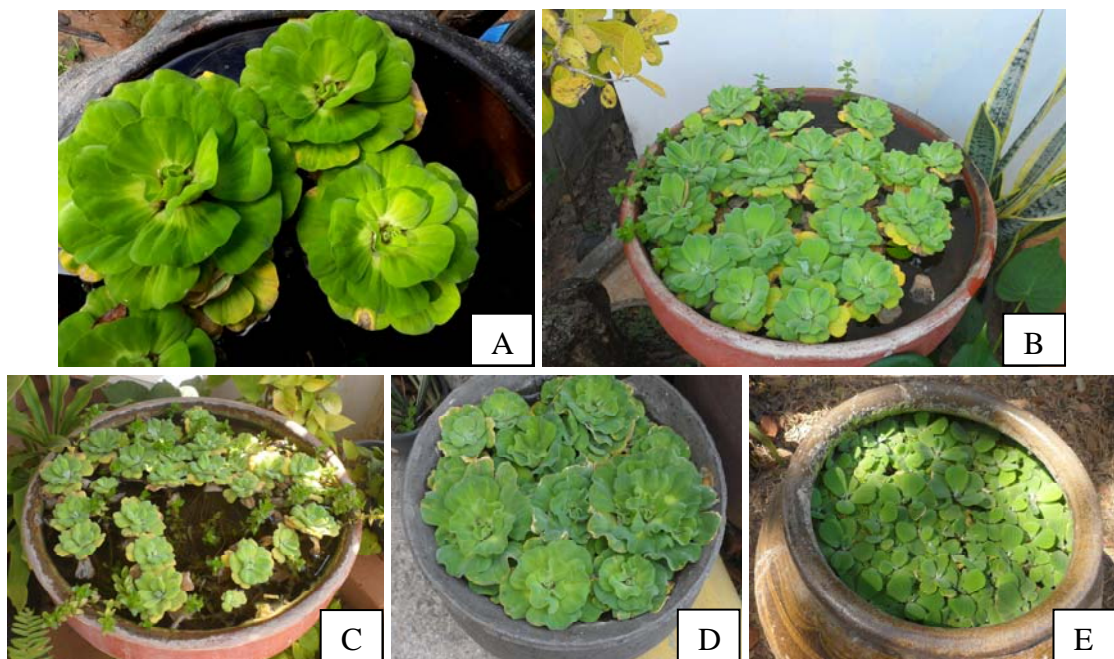
C. Ornamental plant according on temple





**Figure 5.10** Ornamental plants of *Lasia spinosa* L.

A-B. Ornamental plant according on hotle  
 C. Ornamental plant according on houses



**Figure 5.11** Ornamental plants of *Pistia stratiotes* L.

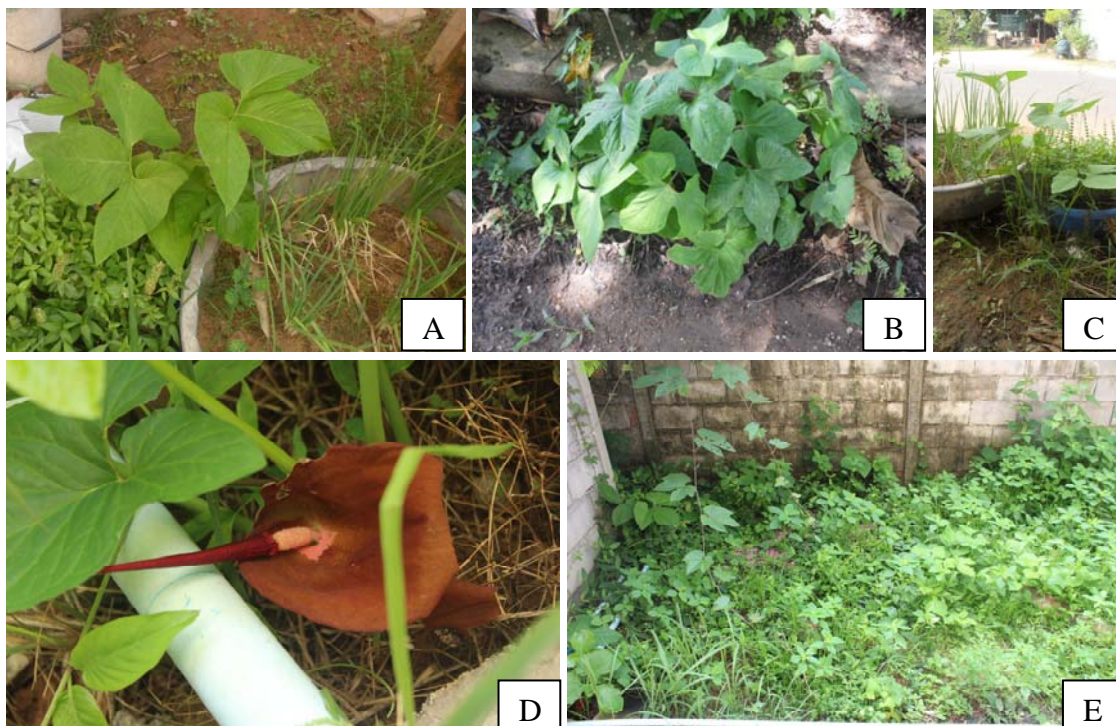
A-E. Ornamental plant according on houses

### Traditional used in rituals



**Figure 5.12** Traditional used in rituals of *Alocasia cucullata* (Lour.) G.Don

A-C. Planting the *Alocasia cucullata* or Wan nang-kwak in front of the house to make money.



**Figure 5.13** Traditional used in rituals of *Typhonium trilobatum* (L.) Schott

A-C. Planting the *Typhonium trilobatum* or Wan-khee in front of the house to chase ghosts.

D-E. Planting the *Typhonium trilobatum* or Wan-khee in area around the house



## CHAPTER 6

### TISSUE CULTURE STUDIES

#### 6.1 Introduction

Plant tissue culture (PTC) broadly refers to cultivation of plant cells, tissues, organs, and plantlets on artificial medium under aseptic and controlled environmental conditions. It is the art of growing experimental plants, selecting a suitable plant organ or tissue to initiate cultures, cleaning, sterilization and trimming it to a suitable size, and planting it on a culture medium in right orientation while maintaining complete asepsis. It also requires an experienced and vigilant eye to select healthy and normal tissues for subculture. PTC involves a scientific approach to systematically optimize physical (nature of the substrate, pH, light, temperature and humidity), chemical (composition of the culture medium, particularly nutrients and growth regulators), biological (source, physiological status and size of the explant), and environmental (gaseous environment inside the culture vial) parameters to achieve the desired growth rate, cellular metabolism, and differentiation (Bhojwani and Dantu, 2013).

The most important of PTC is the demonstration of the unique capacity of plant cells to regenerate full plants, via organogenesis or embryogenesis, irrespective of their source (root, leaf, stem, floral parts, pollen, endosperm) and ploidy level (haploid, diploid, triploid). PTC is also the best technique to exploit the cellular totipotency of plant cells for numerous practical applications, and offers technologies for crop improvement (haploid and triploid production, *in vitro* fertilization, hybrid embryo rescue, variant selection), clonal propagation (Micropropagation), virus elimination (shoot tip culture), germplasm conservation, production of industrial phytochemicals, and regeneration of plants from genetically manipulated cells by recombinant DNA technology (genetic engineering) or cell fusion (somatic hybridization). PTC has been extensively employed for basic studies related to plant physiology (photosynthesis, nutrition of plant cells, and embryos), biochemistry, cellular metabolism, morphogenesis (organogenesis, embryogenesis), phytopathology (plant microbe interaction), histology (cytodifferentiation), cytology (cell cycle), etc. Indeed the discovery of first cytokinin is based on PTC studies (Bhojwani and Dantu, 2012).

Therefore, PTC is an exciting area of basic and applied sciences with considerable scope for further research. Considerable work is being done to understand the physiology and genetics of embryogenesis and organogenesis using PTC systems, especially *Arabidopsis* and carrot, which are likely to enhance the efficiency of *in vitro* regeneration protocols. Thus, PTC forms a part of most of the courses on plant sciences (Developmental Botany, Embryology, Physiology, Genetics, Plant Breeding, Horticulture, Sylviculture, Phytopathology, etc.) and is an essential component of Plant Biotechnology (Bhojwani and Dantu, 2012).

Plant tissue culture is a technique of culturing plant cells, tissues and organs on synthetic media under aseptic environment and controlled conditions of light, temperature, and humidity. The development of plant tissue culture as a fundamental science is closely linked with the discovery and characterization of plant hormones, and has facilitated our understanding of plant growth and development. Furthermore, the ability to grow plant cells and tissues in culture and to control their development forms





the basis of many practical applications in agriculture, horticulture industrial chemistry and is a prerequisite for plant genetic engineering (Mineo, 1990).

## 6.2 Literature reviews

Malamug *et al.* (1992) induced callus of Taro (*Colocasia esculenta*) from shoot tip explants on a modified Nitsch medium supplemented with 2,4-D and BA at 1 mg/l each. The effect of agar concentrations (0, 0.2, 0.4, 0.6, 0.8%, w/v) in a modified Murashige and Skoog (MS) (1962) medium on the regeneration of shoots from taro callus was investigated. Shoot differentiation was observed only in 0.2, 0.4 and 0.6% agar concentrations. A high level of shoot differentiation at 64% was measured on 0.6% agar medium. However, root differentiation was observed regardless of agar concentrations. Shoot differentiation was observed on the medium supplemented with 1 mg/l BA as early as 10 weeks of culture. Addition of 0.1% (w/v) charcoal did not directly induce the regeneration of shoots from the callus, but lengthened the longevity of the calli since no necrosis was observed after 20 weeks of culture. Proliferation of regenerated shoots was enhanced with the addition of  $\alpha$ -naphthalene acetic acid (NAA) and BA at 1 mg/l. The plantlets can be readily potted in a vermiculite medium for acclimatization before planting in the field.

Sai *et al.* (2000) studied an *in vitro* culture system was developed for *Typhonium flagelliforme* using buds from the rhizomes. The mineral salts of four media were tested. These were Murashige and Skoog (MS), Nitsch and Nitsch (NN), Gamborg B5 (GB5) and White (W) of which MS medium was found to be the best medium for *in vitro* culture of *T. flagelliforme*. The addition of as low as 0.1 mg/l (0.54 mM)  $\alpha$ -naphthalene acetic acid (NAA) with the presence or absence of N<sub>6</sub>-benzyladenine (BA) in the MS medium caused abnormal shoot formation. The best medium for maximizing shoot number combined with normal complete plantlets from each bud was MS medium supplemented with 0.3 mg/l (1.33 mM) BA and 0.5 mg/l (2.46 mM) indole-3-butyric acid (IBA). The best acclimatization process was to transfer the normal plantlets, with all the leaves removed, into sand plus coconut husks substrate (1:1) and placed in intermittent water mists house or shaded plant house with 50% light exclusion. Ninety two percent of the plantlets survived using this acclimatization method.

Bhuiyan *et al.* (2011) studied an *in vitro* root initiation in Mukhikachu (*Colocasia esculenta* var. *globulifera*) was assessed in a factorial experiment using three levels of IAA (0.5, 1.0, and 2.0 mg/l), three levels of NAA (0.5, 1.0, and 2.0 mg/l) and control. Fifty percent intact shoots were used as usual, which was named as normal cut explant and the rest 50 % shoots were cut slantly to expose fresh surface i.e., cambium zone and named as slant cut explant. Low levels of IAA (0.5 mg/l) initiated the roots earliest ( $\approx$ 14 DAC) and gave the highest percentage of root (49.71). This treatment also gave the maximum roots/culture (3.63). Root initiation was higher (61.33 %) with slant cut when cultured on a medium containing 0.5 mg/l IAA. The cultures with slant cut end also produced more number of roots and longest roots whereas, the highest root initiation (45.05 %) was given by the treatment 1.0 mg/l NAA, but 2.0 mg/l NAA gave lower percentage of roots (39.89). The maximum number of roots/culture was also obtained by 1.0 mg/l NAA. Slant cut explant performed better regarding root initiation (%), number of roots/culture and length of roots. In this experiment, slant cut explant



performed better than that of normal cut and either IAA (0.05 mg/l) or NAA (1.0 mg/l) might be used for root initiation in Mukhikachu.

Hossain (2012) studied an *in vitro* organogenesis of an upland species of *Colocasia esculenta* cv. *antiquorum* L. was examined in relation to different explants like meristem and parenchymatous storage tissues with or without anthocyanin layer, four levels of each of Kn, 2,4-D, NAA and BAP and four incubation environments such as: 1) 16 h 3 Kl light intensity + 24°C ± 2°C; 2) 24 h dark + 24°C ± 2°C; 3) 24 h dark + 30°C ± 3°C and 4) 12 h diffuse light + 30°C ± 3°C. Only meristems showed proliferation with various degree of intensity both at 16 h 3 Kl light + 24°C ± 2°C and 24 h dark + 24°C ± 2°C conditions and poor response with different levels of Kn + NAA either in light or in the dark. Cultures with NAA + BAP were proliferated very quickly with very high degree of intensity. The cultures under dark did not proliferate for 20 days which upon transfer to light showed high degree of proliferation. Cultures with NAA + BAP formed calluses more pronouncedly at dark than that occurred in the light. Parenchymatous tissues with or without anthocyanin did not proliferate but the tissues with anthocyanin lost pigmentation after 25 - 30 days and turned to grey colour after 50 days while tissues without anthocyanin turned to green colour with shinny pimples indicating that protocorm may be developed. No culture under high temperature environment (30°C ± 3°C) neither survived nor proliferated. The meristems in culture were died within 15 - 20 days while others within 25 - 30 days. In conclusion, a combination of NAA (0.5 - 3.0 mg/l) and BAP (0.5 - 2.0 mg/l) and an incubation photoperiod of 16 hour coupled with temperature of 24°C ± 2°C were found most suitable for *in vitro* culture of *Colocasia esculenta* cv. *antiquorum* L.

Stanly (2012) studied an *in vitro* shoot cultures of *Homalonema pineodora* were initiated from the rhizomatous buds on MS basal medium. The best conditions for propagating *H. pineodora* was found to be MS medium supplemented with 3% sucrose and 0.5 mg/l BA under 24 h of cool fluorescent light which produced an average of 3.8 shoot per explant. Presence of an auxin was not necessary for plantlet production. Liquid MS medium supplemented with 0.5 mg/l BA, enhanced the shoot production of *H. pineodora* as compared to agar-gelled medium with same composition. All the *in vitro* plantlets of *H. pineodora* were successfully acclimatized with 100% survival rate. Scanning electron microscopy confirmed the similarity of leaf microstructures between the *in vitro* and mother plants of *H. pineodora*.

### 6.3 The plant tissue culture studied in Thailand

Laohavisuti and Mitnoi (2005) studied the micropropagation of *Aglaonema simplex* using sterile apical buds culture on MS medium supplemented with the combinations of NAA at 0, 1, 2 mg/l and BA at 0, 1, 2 mg/l. After 6 weeks, it was found that the shoot proliferation from the apical bud explants were significantly enhanced by the addition of 2 mg/l BA in MS medium ( $p < 0.05$ ). The increment of NAA supplemented in the media tended to decrease shoot proliferation in *A. simplex in vitro* culture ( $p < 0.01$ ).

Sanguansermisri *et al.* (2011) studied an *in vitro* culture of *Caladium bicolor* Vent. using young leaf explants. The leaf explants were surface sterilization with clorox 10 percent for 10 min and followed with clorox 5 percent for 5 minute, which sterilize leaf explants. The sterilized leaf explants were culture on modified MS medium



supplemented with BA at concentrate 0.1, 0.5, 1 mg/l combination with NAA at concentrate 2 mg/l. The result showed that modified MS medium with 0.5 mg/l BA and 2 mg/l NAA could be the best for induced 12 shoot per explants.

## 6.4 Materials and methods

*Typhonium glaucum* Hett. & Sookchaloem transplanted in the nursery of Walai Rukhavej Botanical Research Institute, Mahasarakham University was used for plant material in this study.

### 6.4.1 Materials

1. Pot
2. Planting soil
3. Bottles for sampling roots
4. Forceps
5. Surgical knife
6. Beaker
7. Petri dish
8. Dropper
9. Permanent pen
10. Blotting paper
11. Bottles for soak color
12. Distilled water
13. 70% ethyl alcohol
14. 95% ethyl alcohol
15. Activated charcoal
16. Spatula
17. Refrigerator
18. Aluminium foil
19. Magnetic bar
20. Stirring rod
21. Pipette rubber bulb
22. Paper
23. Label
24. Paint brush
25. Knife cutter
26. Bottle 4 and 8 ounces
27. Pipet
28. Cylinder
29. Flask
30. Balance
31. pH meter
32. Hot plate and magnetic stirrer
33. Hot air oven
34. Autoclave
35. Laminar air-flow cabinet



36. Turnel
37. Surgeon knife
38. Cloth sterilize
39. Metal screen
40. Matches
41. Paper sterilize
42. Air condition
43. Timer
44. UV lamp
45. Fluorescent lamp
46. Stock solution MS medium (Murashige and Skoog, 1962)
47. NAA ( $\alpha$ -naphthalene acetic acid) and BA (N<sup>6</sup>-benzyladenine)
48. Sucrose
49. Agar
50. 1 N NaOH
- 51 N HCl
52. Distilled water sterilize
53. Teepol
54. Sodium hypochlorite, NaOCl<sub>2</sub>
55. Tween 20

#### 6.4.2 Tissue culture study

##### 6.4.2.1 Preparation of plant materials

Young leaves of *Typhonium glaucum* Hett. & Sookchaloem were first washed under running tap water for 20 minutes, rinsed with 70% (v/v) ethyl alcohol for 30 seconds. They were surface-sterilized with 1% sodium hypochlorite enriched with 2 drops of Tween 20 for 15 minutes and again rinsed three times with sterilized distilled water. The young leaf were cut 1 x 1 cm in size and then were culture on MS medium (Murashige and Skoog, 1962) supplemented with NAA and BA, 3% sucrose and 0.7% agar for shoot multiplication. The pH of the medium was adjusted to 5.7-5.8 before autoclaving at 121°C and 1.05 kg cm<sup>-2</sup> pressure for 20 minutes. Each piece of leaves proliferated into multiple shoots within 4 weeks. The multiple shoots were separated into single shoots and were transferred to MS medium without added hormones for 8 weeks, to eliminate the effects of exogenous hormones. After that the individual shoots (about 1 cm) were excised aseptically and subsequently used as explants for all experiments. All experiments were conducted in a culture room at 25±2 °C under white, fluorescent light (2,000 lux) at a 16 hour photoperiod.

##### 6.4.2.2 Callus induction and shoot formation

Young leaves of *Typhonium glaucum* cultured on MS medium supplemented with various combinations of 0, 0.5, 1, 1.5 and 2 mg/l NAA and 0, 0.5, 1, 1.5 and 2 mg/l BA with or without activated charcoal for 8 weeks for callus induction and shoot formation. Observation on the percentage of callus information, size of callus, percentage of shoot formation, average number of shoots per explant, average shoot



length (cm) and average number of leaves per plant were recorded after 8 weeks of incubation.

#### 6.4.2.3 Shoot and root formation

Calluses, approximate 1x1 cm culture on the MS medium supplemented with various combinations of 0, 0.1, 0.5 and 1 mg/l NAA and 0, 0.1, 0.5 and 1 mg/l BA for 8 weeks for shoot and root formation. Observations on the average number of shoots per explant, average number of roots per shoot, average shoot length per explant, average root length per explant and average number of leaves per shoot were recorded after 8 weeks of incubation.

#### 6.4.2.4 Shoot proliferation

Shoots, approximate 1 cm cultured on the MS medium supplemented with 0, 0.1, 0.5 and 1 mg/l NAA and 0, 0.5, 1, 1.5 and 2 mg/l BA for 8 weeks for shoot proliferation. Observations on the average number of shoots per explant, average number of roots per shoot, average shoot length per explant, average root length per explant and average number of leaves per shoot were recorded after 8 weeks of incubation.

#### 6.4.2.5 Analysis

Data analyses will be used the means and deviation. All experiments were conducted using completely randomized design (CRD) with 10 culture tubes per replicate, each with one explant. Data were analyzed for significance using ANOVA and the differences contrasted using a Duncan's multiple range test (DMRT). All statistical analysis was performed at the 5% level using the SPSS program version 16.

### 6.5 Results

#### 6.5.1 Effect of NAA and BA on callus induction and shoot formation of *Typhonium glaucum* Hett. & Sookchaloem

Young leaves of *Typhonium glaucum* were cultured on MS medium supplemented with different concentrations of NAA and BA with or without activated charcoal for 8 weeks for callus induction and shoot formation. The result show that the young leaves were cultured on MS medium supplemented with 2 mg/l NAA plus 2 mg/l BA and activated charcoal is produced the highest average number of shoots per explant ( $8.80 \pm 0.35$ ), the average shoot length ( $2.55 \pm 0.33$  cm) and the average number of leaves per plant ( $12.90 \pm 0.43$ ), respectively. (Table 6.1, Figuer 6.1). Young leaves were cultured on MS medium supplemented with 2 mg/l NAA with 2 mg/l BA without activated charcoal the highest size of callus  $1.91 \pm 0.09$  cm (Table 6.1, Figuer 6.1).

After that transfer the callus and shoots were cultured on MS basal medium without growth regulator for reconditioned plantlet for 8 weeks. And cut 1x1 cm of callus was used for callus induction shoot and root and cut 1 cm of plantlet was used for shoot proliferation induction shoot and root experiments.





### 6.5.2 Effect of NAA and BA on shoot and root formation of *Typhonium glaucum* Hett. & Sookchaloem

After 8 weeks of callus culture on the MS basal medium and transfer cut 1x1 calluses on MS medium supplemented with various combinations of 0, 0.1, 0.5 and 1 mg/l NAA and 0, 0.1, 0.5 and 1 mg/l BA for 8 weeks. Found that the callus induction show much increase in multiple shoots formation on 1 mg/l NAA and 1 mg/l BA. The highest average number of shoots per plant is  $12.10 \pm 0.56$ , the highest average number of roots per plant  $3.20 \pm 0.29$ , the highest average shoot length  $4.46 \pm 0.08$  cm, the highest average root length  $1.41 \pm 0.04$  cm and the highest average number of leaves per plant is  $18.7 \pm 0.63$ , respectively (Table 6.2, Figuer 6.2).

### 6.5.3 Effect of NAA and BA on shoot proliferation of *Typhonium glaucum* Hett. & Sookchaloem

After 8 weeks of shoot culture on the MS basal medium and transfer cut 1 shoot length on MS medium supplemented with various combinations of 0, 0.1, 0.5 and 1 mg/l NAA and 0, 0.5, 1, 1.5 and 2 mg/l BA and culture for 8 weeks. Regeneration of plantlets from shoot found that the shoot proliferation induction shoot and the result show much increase in multiple shoots formation on the most suitable medium 0.1 mg/l NAA and 2 mg/l BA. The highest average number of shoots per plant is  $11.10 \pm 0.48$ , the highest average shoot length  $4.71 \pm 0.07$  cm, and the highest average number of leaves per plant is  $12.90 \pm 0.38$ , respectively. Found that the shoot proliferation induction root show much increase in multiple shoots formation on 0.5 mg/l NAA and 2 mg/l BA. The highest average number of roots per plant  $7.00 \pm 0.39$  and the highest average root length  $5.60 \pm 0.08$  cm, respectively (Table 6.3, Figuer 6.3).

## 6.6 Discussion

### Effect of NAA and BA on callus induction and shoot formation of *Typhonium glaucum* Hett. & Sookchaloem

The study micropropagation on young leaves culture of *Typhonium glaucum* Hett. & Sookchaloem cultured on MS medium supplemented with various combinations of 0, 0.5, 1, 1.5 and 2 mg/l NAA and 0, 0.5, 1, 1.5 and 2 mg/l BA and activated charcoal and non-added activated charcoal and cultured for 8 weeks. Show that the young leaves were cultured in MS supplemented with 2 mg/l NAA combination with 2 mg/l BA and activated charcoal young leaves grow to the plantlet is the highest average number of shoots per explant ( $8.80 \pm 0.35$ ), the highest average shoot length ( $2.55 \pm 0.33$  cm) and average number of leaves per plant ( $12.90 \pm 0.43$ ) these result is in disagreement with Sanguanserm Sri *et al.* (2011) reported that the cultured of *Caladium bicolor* using young leaf explants. And were culture on modified MS medium supplemented with BA at concentrate 0.1, 0.5, 1 mg/l combination with NAA at concentrate 2 mg/l. The result showed that modified MS medium with BA 0.5 mg/l and NAA 2 mg/l could the best for induced shoot  $12.00 \pm 1.28$  shoot per explants. And Sai *et al.* (2000) reported that the cultured of *Typhonium flagelliforme* using buds from the rhizomes showed the best



medium which enabled *T. flagelliforme* buds cultured on MS medium supplemented with 0.5 mg/l NAA and 0.5 mg/l BA to produce most shoots 24.5 shoots per bud.

And young leaves were cultured in MS supplemented with 2 mg/l NAA combination with 2 mg/l BA without activated charcoal young leaves develop to most calluses is the highest of size of callus is  $1.91 \pm 0.09$  these result is in disagreement with Treesaksri (2014) reported that the effect of MS medium supplement with NAA and BA on callus induction from leaf segment of *Schoenorchis fragrans* (Parish & Rehb.f.) Seidenf. & Smitinand were cultured on MS medium supplemented with 0.1 and 5 mg/l NAA combined with 0, 3, 5 and 10 mg/l BA for 16 week. It was found that the highest score 1.34 of callus growth of leaf segment were achieved from MS medium supplemented with 1 mg/l NAA and 3 mg/l BA. The highest percentage 22.22 of explants formed callus were achieved from MS medium supplemented with 5 mg/l BA.

#### **Effect of NAA and BA on shoot and root formation of *Typhonium glaucum* Hett. & Sookchaloem**

After that transfer the shoots were cultured on MS medium without growth regulator for 8 weeks. After that transfer the callus induction shoot and root on the MS medium supplemented with various combinations of 0, 0.1, 0.5 and 1 mg/l NAA and 0, 0.1, 0.5 and 1 mg/l BA cultured for 8 weeks. Found that the highest average number of shoots per plant is  $12.10 \pm 0.56$ , the highest average number of roots per plant  $3.20 \pm 0.29$ , the highest average shoot length  $4.46 \pm 0.08$  cm, the highest average root length  $1.41 \pm 0.04$  cm and the highest average number of leaves per plant is  $18.7 \pm 0.63$  were cultured on MS medium 1 mg/l NAA and 1 mg/l BA. These results are in agreement with Malamug *et al.* (1992) reported that the cultured of *Colocasia esculenta* Schott using callus explants on MS medium supplemented with NAA at concentrate 0, 0.1, 1 mg/l and BA at concentrate 0, 0.1, 1 mg/l combination. The result showed that the number of proliferated shoot 3.10 shoots per explant and the highest average shoot length 2.65 cm were cultured on modified MS medium supplemented 1 mg/l NAA and 1 mg/l BA.

#### **Effect of NAA and BA on shoot proliferation of *Typhonium glaucum* Hett. & Sookchaloem**

After that transfer the shoots were cultured on MS medium without growth regulator for 8 weeks. And 1 cm size of plantlet was used for experiments shoot proliferation induction shoot and root on the MS medium supplemented with different concentrations of NAA and different concentrations of BA various combinations of 0, 0.1, 0.5 and 1 mg/l NAA and 0, 0.5, 1, 1.5 and 2 mg/l BA cultured for 8 weeks. The highest average number of shoots per plant is  $11.10 \pm 0.48$ , the highest average shoot length  $4.71 \pm 0.07$  cm, and the highest average number of leaves per plant is  $12.90 \pm 0.38$ , respectively. Found that the shoot proliferation induction root show much increase in multiple shoots formation on 0.5 mg/l NAA and 2 mg/l BA. The highest average number of roots per plant  $7.00 \pm 0.39$  and the highest average root length  $5.60 \pm 0.08$  cm these results are in agreement with Sanguansermisri *et al.* (2011) reported that the cultured of *Caladium bicolor* using young leaf explants. And were culture on modified MS medium supplemented with BA at concentrate 0.1, 0.5, 1 mg/l combination with NAA at concentrate 2 mg/l. The result showed that modified MS medium with BA 0.5



mg/l and NAA 2 mg/l could be the best for induced shoot  $12.00 \pm 1.28$  shoot per explants and average number of leaves  $8.90 \pm 1.59$ . But this result is in disagreement with Hossain (2012) reported that the cultured of *Colocasia esculenta* cv. *antiquorum* L. using meristem explants cultured on MS medium supplemented with different concentrations of NAA and different concentrations of BAP found that the highest average number of one shoots per plant, the highest average shoot length 4.00 cm, the highest average number of leaves per plant is 4.00, highest average number of roots per plant 4.00 and the highest average root length 3.50 cm on MS medium without growth regulator.

An auxin is required by most plant cells for division and root initiation. At high concentrations, auxin can suppress morphogenesis. The auxin 2,4-D is widely used for callus induction: IAA, IBA and NAA are used for root induction. NAA which is synthetic auxin and more stable than IAA, which is the naturally occurring auxin (Smith, 2000). Cytokinin (BA) promote cell division, shoot proliferation and shoot morphogenesis (Miller & Skoog, 1953 and Miller, 1961). So an auxin and cytokinin were considered to be factors for culture shoot and root multiplication.



**Table 6.1** Comparison of suitable medium for culturing of the *Typhonium glaucum* were recorded after cultured for 8 weeks.

MS Medium			Callus		Shoot			
	NAA (mg/l)	BA (mg/l)	%	Size (cm) (M±SD)	%	Average number of shoots (shoots/plant) (M±SD)	Average shoot length (cm) (M±SD)	Average number of leaves (leaves /plant) (M±SD)
Activated charcoal	0	0	10	1.73±0.06 <sup>c</sup>	0	0.00±0.00 <sup>d</sup>	0.00±0.00 <sup>e</sup>	0.00±0.00 <sup>d</sup>
	0.5	0.5	10	0.00±0.00 <sup>f</sup>	10	4.00±0.25 <sup>c</sup>	0.70±0.17 <sup>d</sup>	0.00±0.00 <sup>d</sup>
	1	1	10	0.00±0.00 <sup>f</sup>	10	5.70±0.21 <sup>b</sup>	0.79±0.12 <sup>d</sup>	0.00±0.00 <sup>d</sup>
	1.5	1.5	10	0.00±0.00 <sup>f</sup>	10	6.00±0.21 <sup>b</sup>	1.70±0.18 <sup>b</sup>	4.20±0.35 <sup>b</sup>
	2	2	10	0.00±0.00 <sup>f</sup>	10	8.80±0.35 <sup>a</sup>	2.55±0.33 <sup>a</sup>	12.90±0.43 <sup>a</sup>
Non activated charcoal	0	0	10	1.69±0.07 <sup>d</sup>	0	0.00±0.00 <sup>d</sup>	0.00±0.00 <sup>e</sup>	0.00±0.00 <sup>d</sup>
	0.5	0.5	10	1.86±0.14 <sup>c</sup>	0	0.00±0.00 <sup>d</sup>	0.00±0.00 <sup>e</sup>	0.00±0.00 <sup>d</sup>
	1	1	10	1.86±0.11 <sup>c</sup>	0	0.00±0.00 <sup>d</sup>	0.00±0.00 <sup>e</sup>	0.00±0.00 <sup>d</sup>
	1.5	1.5	10	1.89±0.08 <sup>b</sup>	10	1.20±0.13 <sup>e</sup>	1.24±0.40 <sup>c</sup>	1.50±0.16 <sup>c</sup>
	2	2	10	1.91±0.09 <sup>a</sup>	10	2.80±0.24 <sup>d</sup>	1.33±0.43 <sup>bc</sup>	2.40±0.22 <sup>c</sup>

Note: Percentage Mean values ± SD (standard deviation) within the same column followed by same letter are not significantly different (Duncan test,  $p \leq 0.05$ ).

**Table 6.2** Effect of NAA and BA on shoot and root formation of *Typhonium glaucum* were recorded after 8 weeks of culture.

MS Medium		Average number of shoots (shoots/plant) (M±SD)	Average shoot length (cm) (M±SD)	Average number of roots (roots/plant) (M±SD)	Average root length (cm) (M±SD)	Average number of leaves (leaves /plant) (M±SD)
NAA (mg/l)	BA (mg/l)					
0	0	3.10±0.17 <sup>d</sup>	2.30±0.08 <sup>d</sup>	1.80±0.29 <sup>c</sup>	0.63±0.08 <sup>c</sup>	5.60±0.26 <sup>c</sup>
0.1	0.1	5.70±0.33 <sup>c</sup>	2.68±0.11 <sup>c</sup>	0.00±0.00 <sup>d</sup>	0.00±0.00 <sup>d</sup>	7.20±0.38 <sup>c</sup>
0.5	0.5	7.50±0.26 <sup>b</sup>	3.51±0.11 <sup>b</sup>	1.90±0.27 <sup>b</sup>	0.80±0.04 <sup>b</sup>	10.70±0.70 <sup>b</sup>
1	1	12.10±0.56 <sup>a</sup>	4.46±0.08 <sup>a</sup>	3.20±0.29 <sup>a</sup>	1.41±0.04 <sup>a</sup>	18.70±0.63 <sup>a</sup>

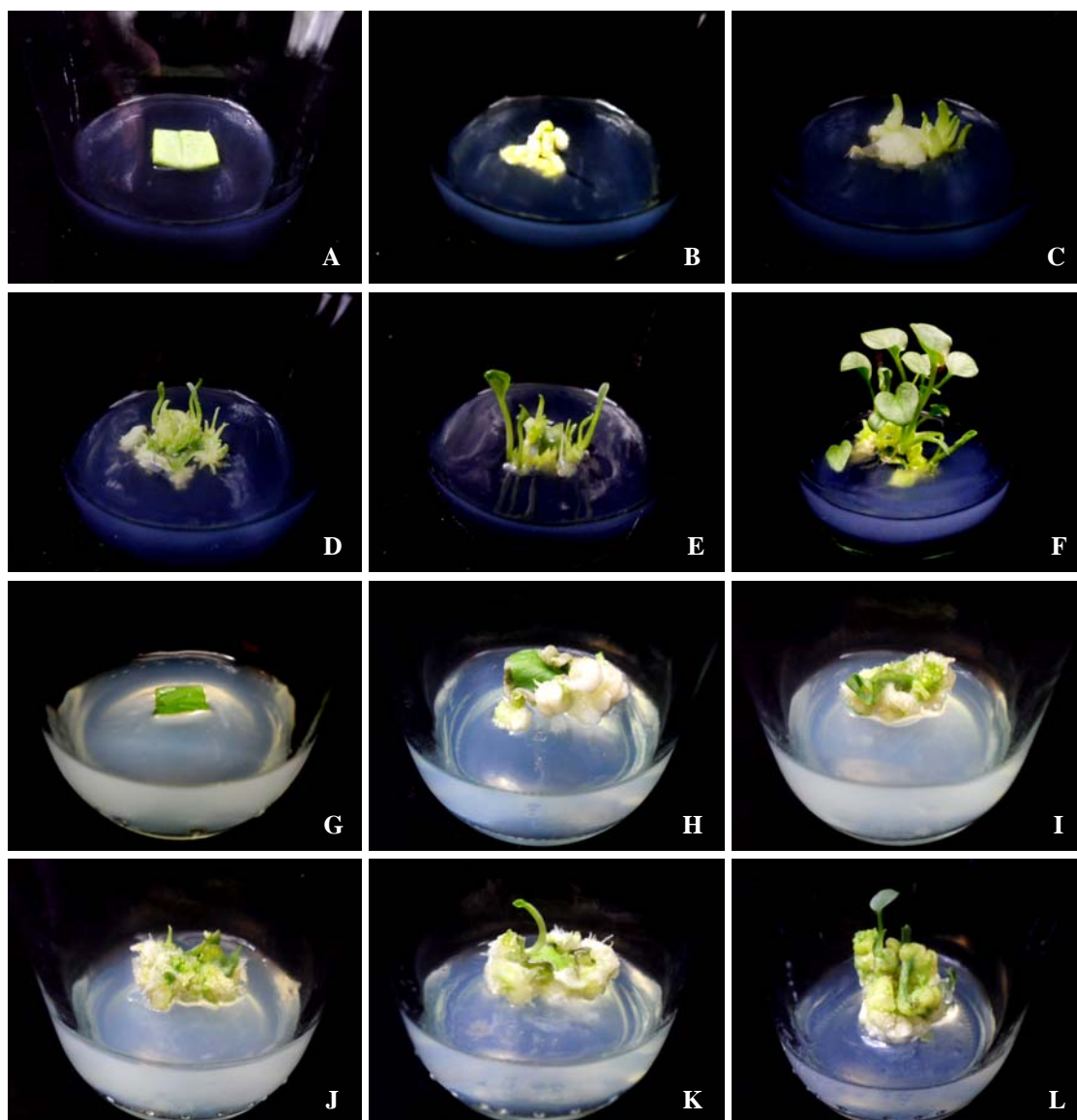
Note: Mean values ± SD (standard deviation) within the same column followed by same letter are not significantly different (Duncan test,  $p \leq 0.05$ ).

**Table 6.3** Effects of NAA and BA on shoot proliferation of *Typhonium glaucum* Hett. & Sookchaloem.

MS Medium		Average number of shoots (shoots/plant) (M±SD)	Average shoot length (cm) (M±SD)	Average number of roots (roots/plant) (M±SD)	Average root length (cm) (M±SD)	Average number of leaves (leaves /plant) (M±SD)
NAA (mg/l)	BA (mg/l)					
0	0	1.40±0.16 <sup>f</sup>	3.17±0.07 <sup>de</sup>	2.90±0.18 <sup>b</sup>	1.48±0.06 <sup>b</sup>	3.70±0.15 <sup>g</sup>
0.1	0.5	4.30±0.21 <sup>cde</sup>	4.15±0.03 <sup>b</sup>	1.80±0.25 <sup>c</sup>	1.23±0.06 <sup>c</sup>	7.40±0.22 <sup>cdef</sup>
0.1	1	4.50±0.27 <sup>cde</sup>	4.17±0.04 <sup>b</sup>	0.00±0.00 <sup>e</sup>	0.00±0.00 <sup>e</sup>	7.70±0.15 <sup>cd</sup>
0.1	1.5	7.30±0.30 <sup>b</sup>	4.61±0.02 <sup>a</sup>	0.00±0.00 <sup>e</sup>	0.00±0.00 <sup>e</sup>	12.10±0.31 <sup>a</sup>
0.1	2	11.10±0.48 <sup>a</sup>	4.71±0.07 <sup>a</sup>	0.00±0.00 <sup>e</sup>	0.00±0.00 <sup>e</sup>	12.90±0.38 <sup>a</sup>
0.5	0.5	4.90±0.31 <sup>c</sup>	4.23±0.03 <sup>b</sup>	1.60±0.22 <sup>c</sup>	0.95±0.08 <sup>d</sup>	9.70±1.17 <sup>b</sup>
0.5	1	4.90±0.23 <sup>c</sup>	4.45±0.04 <sup>b</sup>	1.40±0.16 <sup>c</sup>	1.06±0.05 <sup>cd</sup>	9.30±0.21 <sup>bc</sup>
0.5	1.5	4.60±0.22 <sup>cde</sup>	3.42±0.06 <sup>cd</sup>	1.70±0.15 <sup>c</sup>	1.21±0.07 <sup>c</sup>	8.90±0.35 <sup>bcd</sup>
0.5	2	4.80±0.13 <sup>cd</sup>	3.25±0.05 <sup>cde</sup>	7.00±0.39 <sup>a</sup>	5.60±0.08 <sup>a</sup>	7.50±0.17 <sup>cde</sup>
1	0.5	3.60±0.22 <sup>de</sup>	3.06±0.05 <sup>ef</sup>	1.10±0.13 <sup>d</sup>	0.90±0.00 <sup>d</sup>	5.50±0.17 <sup>fg</sup>
1	1	3.50±0.17 <sup>e</sup>	3.19±0.02 <sup>de</sup>	0.00±0.00 <sup>e</sup>	0.00±0.00 <sup>e</sup>	5.50±0.22 <sup>fg</sup>
1	1.5	3.67±0.29 <sup>cde</sup>	2.78±0.05 <sup>f</sup>	0.00±0.00 <sup>e</sup>	0.00±0.00 <sup>e</sup>	5.67±0.24 <sup>ef</sup>
1	2	4.27±0.19 <sup>cde</sup>	3.51±0.15 <sup>c</sup>	0.00±0.00 <sup>e</sup>	0.00±0.00 <sup>e</sup>	7.09±0.25 <sup>def</sup>

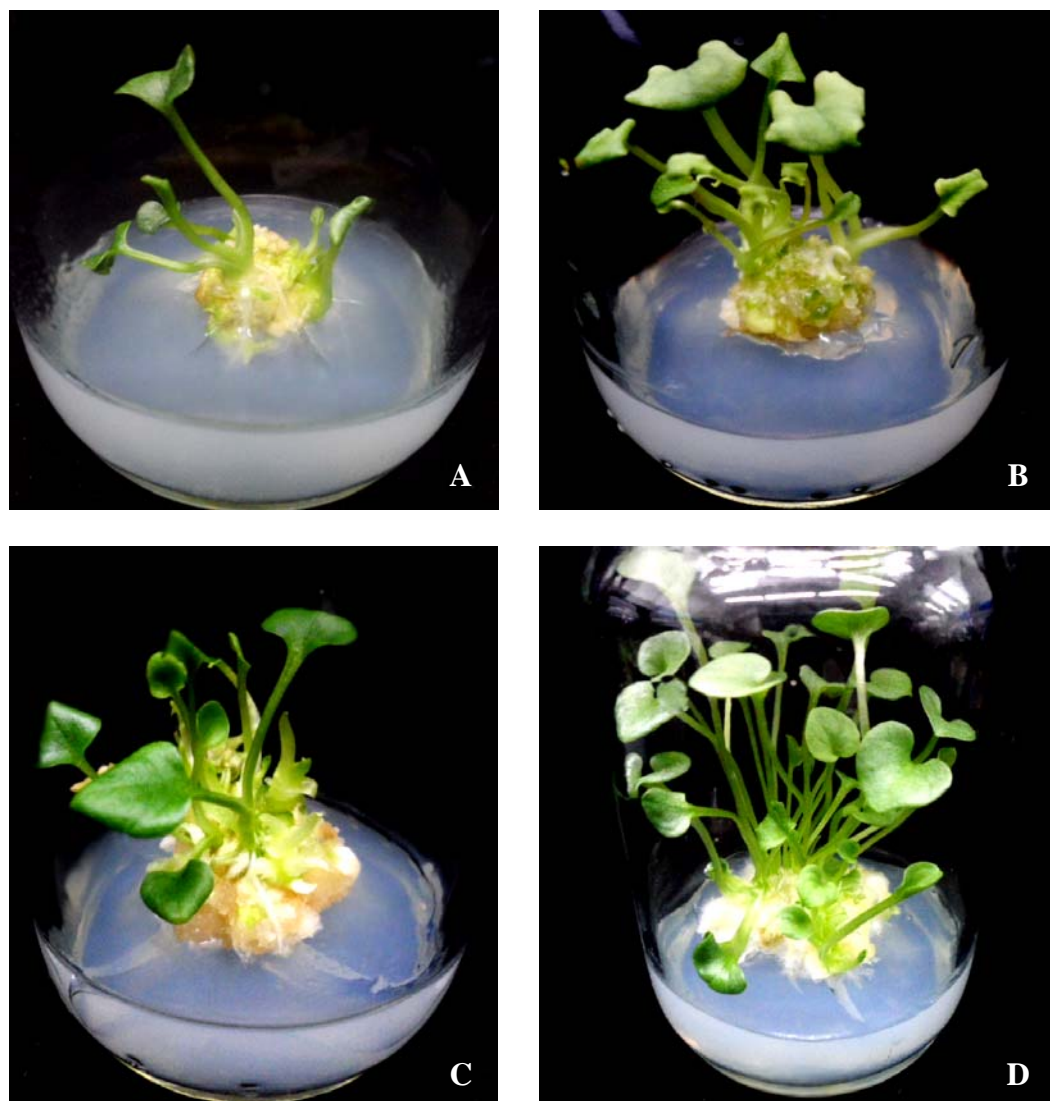
Note: Mean values ± SD (standard deviation) within the same column followed by same letter are not significantly different (Duncan test,  $p \leq 0.05$ ).





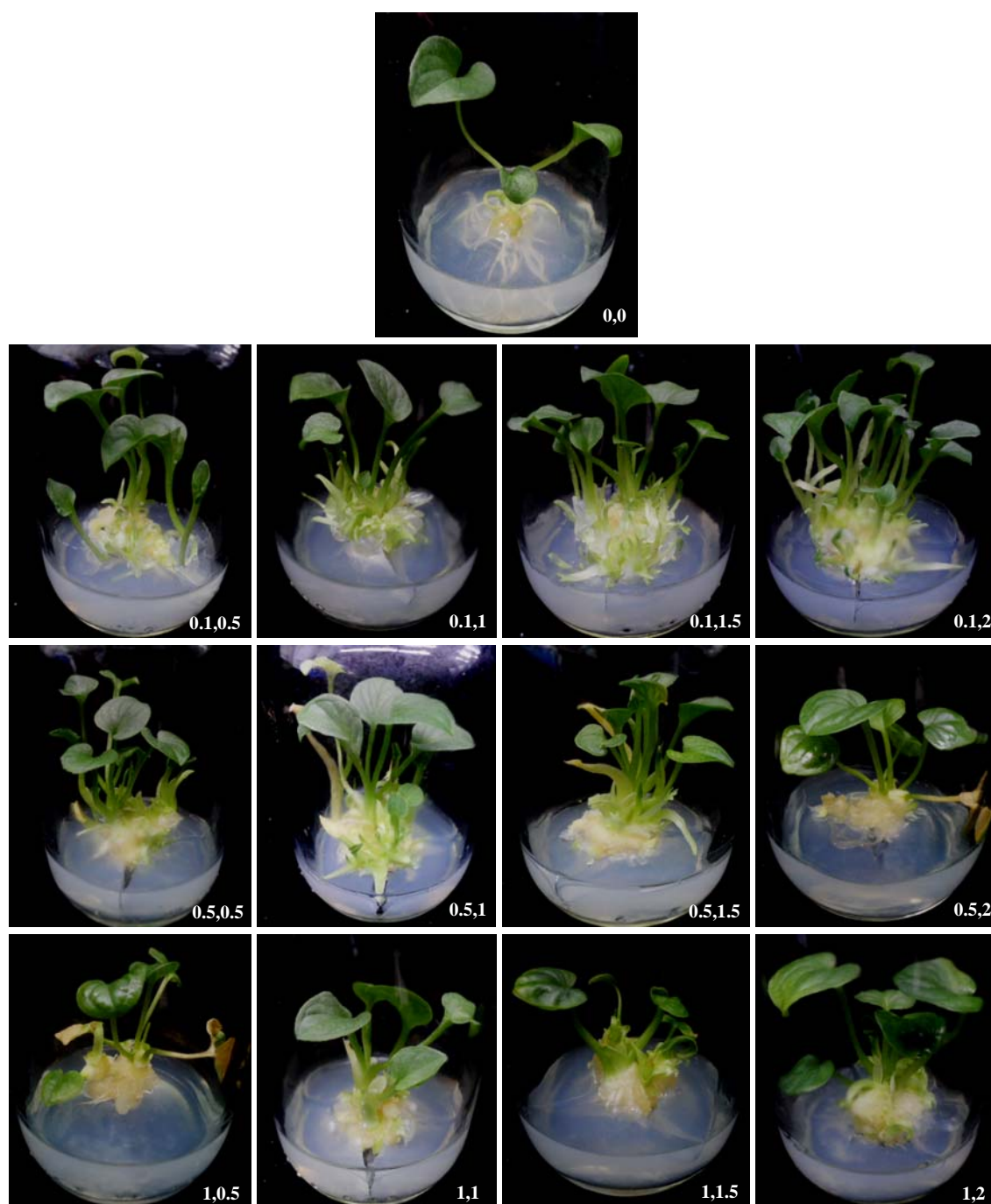
**Figure 6.1** Effects of suitable medium for culturing of the *Typhonium glaucum* Hett. & Sookchaloem on MS medium supplemented with different concentrations of NAA and BA with and without activated charcoal for 8 weeks.

- A. 1x1 young leaves size on MS medium+activated charcoal (AC)
- B. Control+AC
- C. 0.5 mg/l NAA+0.5 mg/l BA+AC
- D. 1 mg/l NAA+1 mg/l BA+AC
- E. 1.5 mg/l NAA+1.5 mg/l BA+AC
- F. 2 mg/l NAA+2 mg/l BA+AC
- G. 1x1 young leaves size on MS medium
- H. Control
- I. 0.5 mg/l NAA+0.5 mg/l BA
- J. 1 mg/l NAA+1 mg/l BA
- K. 1.5 mg/l NAA+1.5 mg/l BA
- L. 2 mg/l NAA+2 mg/l BA



**Figure 6.2** Effect of culturing the *Typhonium glaucum* Hett. & Sookchaloem on MS medium supplemented with different concentrations of NAA and BA on shoot and root for after 8 weeks of callus.

- A. Control
- B. 0.1 mg/l NAA+0.1 mg/l BA
- C. 0.5 mg/l NAA+0.5 mg/l BA
- D. 1 mg/l NAA+1 mg/l BA



**Figure 6.3** Shoots multiplication of *Typhonium glaucum* Hett. & Sookchaloem on MS medium supplemented with NAA and BA after 8 weeks of culture.

## CHAPTER 7

### CONCLUSION AND DISCUSSION

#### 7.1 Chromosome studies

Chromosome studies were made from the roots-tips plant 20 species Araceae in Thailand with the Feulgen squash technique. The karyotype and idiogramming studies of chromosome number counting was performed on mitotic metaphase cells under a Light Microscope. Twenty clearly observable and well-spread cell chromosomes were selected and photographed. The chromosomes number shown is divided into 8 groups as follows  $2n$  (diploid) = 18, 24, 26, 28, 38, 40, 42 and 58. Ten species has karyotype formula is symmetrical and 10 species has karyotype formula is asymmetrical. In this study, 8 species somatic chromosomes were also found to be satellite, found that the end of the chromosome. And the chromosomes number of 7 species has been recorded for the first time.

#### 7.2 Palynology studies

Pollen grains of the 18 species Araceae in Thailand were examined by light microscopy (LM) and scanning electron microscopy (SEM). Pollen grain of all species is monad, three size groups of pollen grains include small, medium and large size of pollen. Two symmetry groups of pollen grains are radial and radial symmetry. Two aperture group's size of pollen grains is monoporate and diporate aperture. Five shape groups of pollen grains include pheroidal, prolate, oblate spheroidal, subprolate and prolate shape. Six exine sculpturing groups of pollen grain. The pollen grains of 16 species have been recorded for the first time.

#### 7.3 Anatomy studies

The study anatomy of 20 species Araceae in Thailand by peeling method. Plant leaf epidermal cells shapes exhibit a jigsaw and rectangular to polygonal in form with smooth, undulate and sinuate anticlinal cell walls on both surfaces on the adaxial surface and on the abaxial surface. The epidermis cells on both surfaces are stomata were present possessed paracytic, hexacytic, anomocytic, cyclocytic, 2,3,4 subsidiary cells, 2,3,4,6 subsidiary cells and 2,3,4,5,6 subsidiary cells stomatal type. Smooth cuticle sculpturing was present in both adaxial and abaxial surfaces and 4 species without cuticle sculpturing. Three species has a solitary crystal is presented in epidermal cell on the adaxial surface and on the abaxial surface. Three species has trichomes are presented in epidermal cell on the adaxial surface and on the abaxial surface. And 4 species has tannin is presented in epidermal cells on the both adaxial and abaxial surfaces. The Anatomy of 15 species has been recorded for the first time.

#### 7.4 Traditional use studies

Surveys of traditional used of the Araceae family in the Northeastern Thailand. Local herbalists and the elders were interviewed about Thai local names, usages, used parts and methods of use. A total of 10 species belonging to 8 genus were recorded in





this study. The common traditional used were *Colocasia esculenta* (3 aspects), *Colocasia gigantea* (3 aspects), *Alocasia cucullata* (3 aspects), *Aglaonema modestum* (2 aspects), *Amorphophallus brevispathus* (2 aspects), *Alocasia macrorrhizos* (2 aspects), *Lasia spinosa* (2 aspects), *Pistia stratiotes* (1 aspects), *Scindapsus officinalis* (1 aspects), *Typhonium trilobatum* (1 aspects), respectively. The most widely used part was young stem and young leaves for foods and medicines.

## 7.5 Tissue culture studies

The study micropropagation on young leaves culture of *Typhonium glaucum* Hett. & Sookchaloem cultured on MS medium (Murashige and Skoog, 1962) supplemented with various combinations of 0, 0.5, 1, 1.5 and 2 mg/l NAA and 0, 0.5, 1, 1.5 and 2 mg/l BA and activated charcoal and non-added activated charcoal and cultured for 8 weeks. Show that the young leaves were cultured in MS supplemented with 2 mg/l NAA combination with 2 mg/l BA and activated charcoal. And young leaves were cultured in MS supplemented with 2 mg/l NAA combination with 2 mg/l BA without activated charcoal young leaves develop to most calluses.

The callus induction shoot and root on the MS medium supplemented with various combinations of 0, 0.1, 0.5 and 1 mg/l NAA and 0, 0.1, 0.5 and 1 mg/l BA cultured for 8 weeks. Found that the highest average number, the highest average number of roots, the highest average shoot length, the highest average root length and the highest average number of leaves per plant were cultured on MS medium 1 mg/l NAA and 1 mg/l BA.

*In vitro* shoot induction and multiplication proliferation shoot and root on the MS medium supplemented with different concentrations of NAA and different concentrations of BA various combinations of 0, 0.1, 0.5 and 1 mg/l NAA and 0, 0.5, 1, 1.5 and 2 mg/l BA cultured for 8 weeks. The highest average number of shoots per plant, the highest average shoots length and the highest average numbers of leaves per plant were culture on the MS medium supplemented with 0.1 mg/l NAA and 2 mg/l BA. And found that the shoot proliferation induction root show much increase in multiple shoots formation on 0.5 mg/l NAA and 2 mg/l BA.

## 7.6 Summary

The data in this study has been information of chromosome, pollen and anatomy foresee the relation evolution closely together in each genus and each species comparison summary as follows. This study found that the chromosome of 12 genus 20 species of Araceae in Thailand. Which the characteristics were compared to see the evolution of relationship the chromosome belongs to symmetrical karyotype and an asymmetry karyotype. Exine sculpturing of pollen can be divided to five in pollen group. While, stomata types used to separate into groups for anatomy. So, evolutionary relationship can be divided in to two groups.

Ten primary evolution species which are karyotype symmetrical of chromosome include; *Alocasia cucullata* (Lour.) G.Don, *Alocasia macrorrhizos* (L.) G.Don, *Amorphophallus serrulatus* Hett. & A.Galloway, *Arisaema maxwellii* Hett. & Gusman, *Colocasia fallax* Schott, *Hapaline benthamiana* Schott, *Homalomena griffithii* (Schott) Hook.f., *Pistia stratiotes* L., *Schismatoglottis calypttrata* (Roxb.) Zoll. &

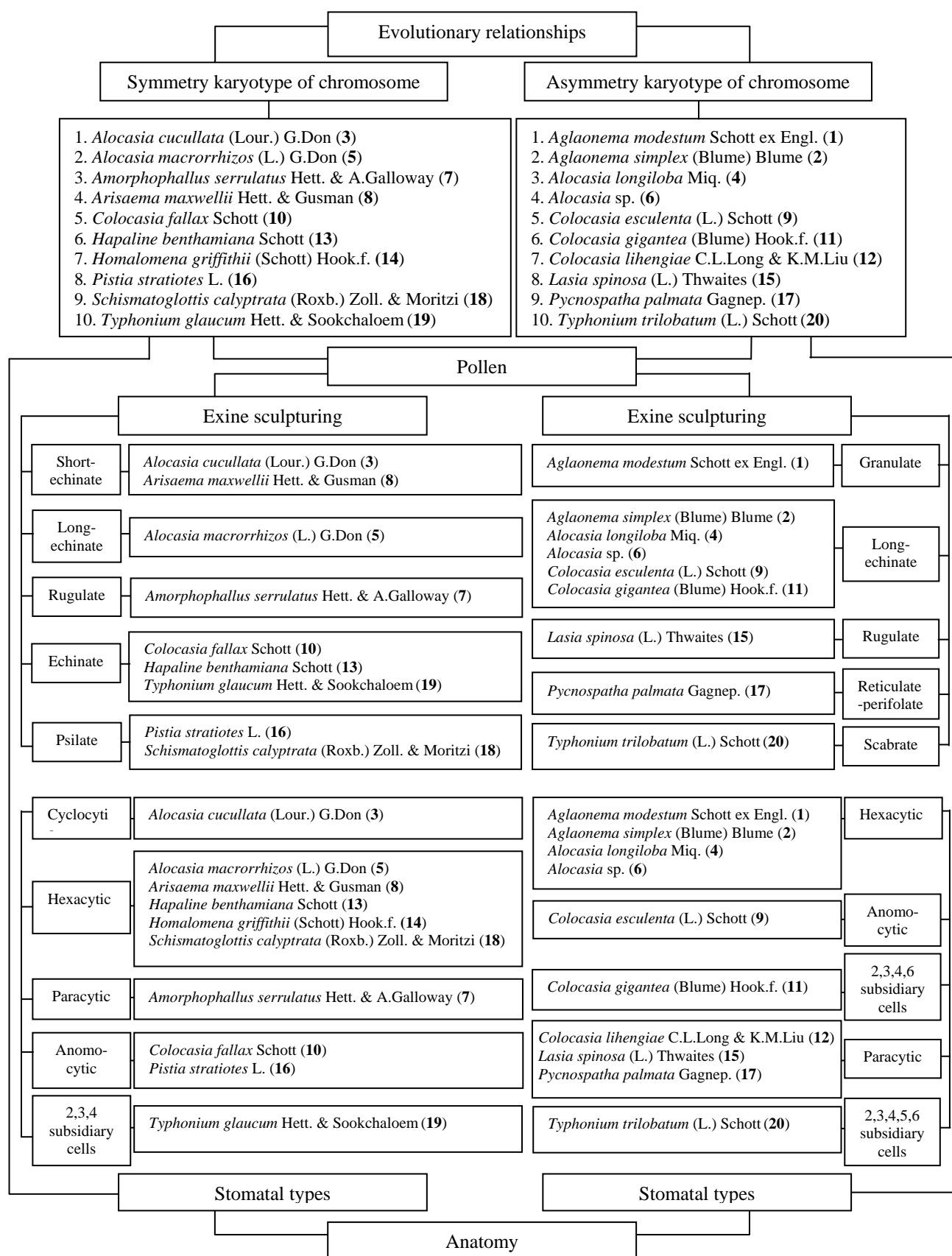




Moritz and *Typhonium glaucum* Hett. & Sookchaloem. Pollen can be divided into five groups by exine sculpturing; the first group is short-echinate exine sculpturing pollen, namely *Alocasia cucullata* (Lour.) G.Don and, *Arisaema maxwellii* Hett.&Gusman; the second group is long-echinate exine sculpturing pollen which is *Alocasia macrorrhizos*(L.) G.Don; third group is regulate exine sculpturing pollen and found only in *Amorphophallus serrulatus*; fourth group is echinate exine sculpturing pollen including *Colocasia esculenta*, *Hapaline benthamiana* and *Typhonium trilobatum* and; fifth group is psilate exine sculpturing pollen including *Pistia stratiotes* and *Scittomaglottis calyptrate*. Anatomical characteristics based on stomatal types has five groups as follows; the first group is cyclocytic cells stomatal types is an *Alocasia cucullata*; the second group is hexacytic stomatal types include *Alocasia macrorrhizos*, *Arisaema maxwellii*, *Hapaline benthamiana*, *Homalomena griffithii* and *Scittomaglottis calyptrate*; the third group is paracytic stomatal types found only in *Amorphophallus serrulatus*; the fourth group is anomocytic stomatal types including *Colocasia fallax* and *Pistia stratiotes* and; the fifth group is 2, 3, 4 subsidiary cells stomatal types found only in *Typhonium glaucum*. From the above information is found that the all species there are close relationship together it is the same chromosome has karyotype formula is asymmetrical. But exine sculpturing of pollen and cells of stomatal types in each species is a different group.

The second group is a high evolved are 10 species which has karyotype asymmetrical of chromosome include; *Aglaonema modestum* Schott ex Engl., *Aglaonema simplex* (Blume) Blume, *Alocasia longiloba* Miq., *Alocasia* sp., *Colocasia esculenta* (L.) Schott, *Colocasia gigantea* (Blume) Hook.f., *Colocasia lihengiae* C.L.Long & K.M.Liu, *Lasia spinosa* (L.) Thwaites, *Pycnospatha palmata* Gagnep. and *Typhonium trilobatum* (L.) Schott. Pollen can be divided into five groups by exine sculpturing; the first group is granulate exine sculpturing pollen, namely *Aglaonema modestum*; the second group is long-echinate exine sculpturing pollen which is *Aglaonema simplex*, *Alocasia longiloba*, *A. sp.*, *Colocasia esculenta* and *C. gigantean*; the third group is rugulate exine sculpturing pollen found only in *Lasia spinosa*; the fourth group is reticulate-perifoliate exine sculpturing pollen is a *Pycnospatha palmata* and; the fifth group is scabrate exine sculpturing pollen is a *Typhonium trilobatum*. Anatomical characteristics based on stomatal types has five groups as follows; the first group is hexacytic cells stomatal types including *Aglaonema modestum*, *A. simplex*, *Alocasia longiloba* and *A. sp.*; the second group is anomocytic stomatal types found only in *Colocasia esculenta*; the third group is 2, 3, 4, 6 subsidiary cells stomatal types found only in *Colocasia gigantean*; the fourth group is paracytic stomatal types including *Colocasia lihengiae*, *Lasia spinosa* and *Pycnospatha palmata* and; the fifth group is 2, 3, 4, 5, 6 subsidiary cells stomatal types found only in *Typhonium trilobatum*. From the above information is found that the *Aglaonema simplex*, *Alocasia longiloba* and *A. sp.* there are close relationship together it is the same chromosome has karyotype formula is asymmetrical, long-echinate exine sculpturing of pollen and hexacytic cells of stomatal types. So, it seems to support that all 3 species an evolutionary relationships closer together. It also found that *Lasia spinosa* and *Typhonium glaucum* found no close ties with any species. The numbers in parentheses is the number species of species studied by sorting the letters A-Z. (Diagram 7.1)





**Figure 7.1** Evolutionary relationships of chromosome, pollen and anatomy.



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## **Appendices**



## **Plant Tissue Culture**





**Table 1** Preparation of MS medium stock solution (Murashige and Skoog, 1962)

Component	Chemical contents (mg/L)	Concentration (mg/L) of stock solution	Concentration (times)	Volume (ml) of stock solution on make 1 liter MS medium
<b>Stock solution 1</b>				50
NH <sub>4</sub> NO <sub>3</sub>	1,650	33,000	20	
KNO <sub>3</sub>	1,900	38,000		
CaCl <sub>2</sub> .2H <sub>2</sub> O	440	8,800		
MgSO <sub>4</sub> .7H <sub>2</sub> O	370	7,400		
KH <sub>2</sub> PO <sub>4</sub>	170	3,400		
<b>Stock solution 2</b>				5
KI	0.83	166	200	
H <sub>3</sub> BO <sub>3</sub>	6.2	1,240		
MnSO <sub>4</sub> .4H <sub>2</sub> O	22.3	4,460		
ZnSO <sub>4</sub> .7H <sub>2</sub> O	8.6	1,720		
Na <sub>2</sub> MoO <sub>4</sub> .2H <sub>2</sub> O	0.25	50		
CuSO <sub>4</sub> .5H <sub>2</sub> O	0.025	5		
CoCl <sub>2</sub> .6H <sub>2</sub> O	0.025	5		
<b>Stock solution 3</b>				5
FeSO <sub>4</sub> .7H <sub>2</sub> O	27.85	5,560	200	
Na <sub>2</sub> .EDTA.2H <sub>2</sub> O	37.3	7,460		
<b>Stock solution 4</b>				5
Myo-inositol	100.50	20,000	200	
Nicotinic acid	0.5	100		
Pyridoxine HCl	0.5	100		
Thiamine HCl	0.5	100		
Glycine	2	400		

One liter of MS medium contains 50 ml of stock solution 1, 5 ml each of stock solution 2, 3 and 4; 30 g sucrose and 7 g Agar. The pH was adjusted to 5.7-5.8 before autoclaving for 20 minutes at 121 °C and 1.05 kgcm<sup>-2</sup> pressures.



## **Biography**



## Biography

<b>Name</b>	Miss Rattanaalee Senavongse
<b>Date of birth</b>	May 13 <sup>st</sup> , 1989
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### Research grants, research publications, presentations & awards

#### Research grants

Scholarship research grant for graduate students (Ph.D.) Fiscal Year 2560, Mahasarakham University, Thailand (This Research was Financially Supported By Mahasarakham University grant year 2017).

#### Research Publications and Presentations

1. Senavongse, R., Saensouk, S. and Saensouk, P. (2015). Diversity and traditional use of the genus *Lasia* (Araceae) in the Northeast of Thailand. In: **The 3<sup>rd</sup> Conference on Botanical Research in Tropical Asia**, 6 -11 December 2015, Lao Plaza Hotel, Vientiane, Lao PDR. (Poster presentation)
2. Senavongse, R., Saensouk, S. and Saensouk, P. (2016). Traditional used of the genus *Colocasia* in the Northeast of Thailand. In: **Thailand's 1<sup>st</sup> Ethnobotany Conference of Thailand (ECT1)**, 24-27 August 2016, UNISERV CMU: University Academic Service Center, Chiang Mai University, Thailand. (Oral presentation)
3. Senavongse, R., Saensouk, S. and Saensouk, P. (2016). Diversity of the genus *Colocasia* (Araceae) in the Northeast of Thailand. In: **The 3<sup>rd</sup> International Postgraduate Symposium on Food, Agriculture and Biotechnology in ASEAN (IPSFAB2016)**, 7-8 September 2016, Faculty of Humanities and Social Sciences , Mahasarakham University, Thailand. (Poster presentation)



4. Senavongse, R., Saensouk, S. and Saensouk, P. (2016). Diversity of the genus *Alocasia* (Araceae) in the Northeast of Thailand. In: **Young Conservation Scientists Conference 2017 (YCS2017)**, 25-27 May 2017, Surasammanakhan Hotel, Suranaree University of Technology, Thailand. (Poster presentation)

### Awards

1. The best oral presentation award for Ph.D. Student at the WRBRI Conference in Biodiversity 2016 (WRBRICB 2016), Walai Rukhavej Botanical Research Institute Mahasarakham University, Maha sarakham, Thailand, 2016.
2. The 2<sup>nd</sup> prize oral presentation award for Ph.D. Student at the 3<sup>rd</sup> WRBRI Conference in Biodiversity 2017 (WRBRICB 2017), Walai Rukhavej Botanical Research Institute Mahasarakham University, Maha sarakham, Thailand, 2017.
3. Received a letter of invitation attend International Training Workshop on Modern Breeding and Cultivation Technology of Vegetables held on September 5<sup>th</sup> to 24<sup>th</sup> 2017, Institute of Vegetables and Flowers Chinese Academy of Agricultural Sciences, in Beijing, China.

### Research output

- Senavongse, R., Saensouk, S. and Saensouk, P. (2018). Comparative Karyotype Analysis in Five Morphological Forms of *Bon Colocasia esculenta* (L.) Schott (Araceae) in Thailand. *Cytologia*, 83(2).

