Reconstructing the Evolutionary History of Reproductive Characters in *Anthurium* (Araceae)

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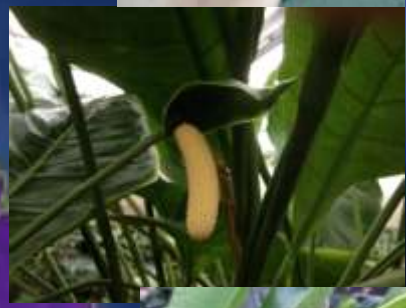
Overview

I. Introduction
II. Research Goals
III. Methods
IV. Results & Discussion
V. Conclusion
VI. Future Research

Anthurium (Araceae)

- 912 species (and counting)
- Large Neotropical distribution
- Present in a variety of ecosystems
- Variable vegetative and reproductive morphology
- Usually bee, beetle or fly-pollinated
- Epiphytic, lithophytic, or terrestrial
- Some members economically important

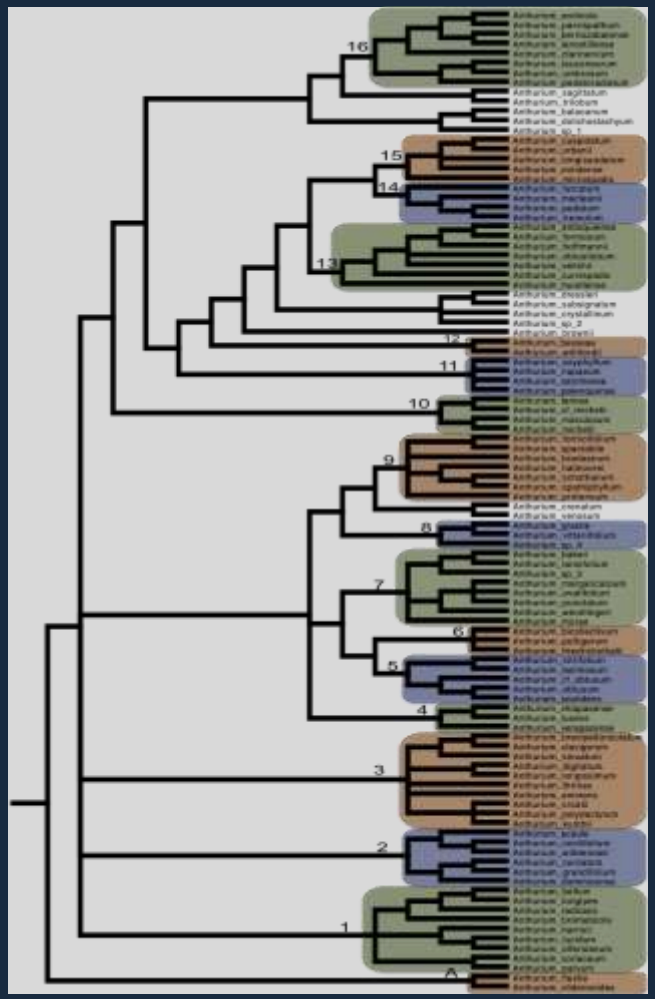





https://earth.google.com/

Classification

- Croat & Sheffer (1983) proposed 18 sections and 2 series
- Carlsen & Croat (2013) generated molecular phylogeny with 17 highly supported clades
- Series *Multinervia,* series *Pachyneurium,* and sections *Calomystrium*, *Leptanthurium, Tetraspermium, Dactylophyllium, and Polyphyllium* supported by Carlsen & Croat



Carlsen & Croat (2013)

Characters and Classification

Grayum (1990)

- For classification, reproductive morphology is generally more useful than vegetative morphology
- Reproductive morphological changes → reproductive isolation → speciation

Carlsen & Croat (2013)

- "Reproductive characters…seem to be more reliable to characterize clades within *Anthurium*"

Research Goals

1. Use reproductive morphology to characterize clades proposed by Carlsen & Croat (2013)
2. Evaluate the importance of reproductive character shifts in the evolution of *Anthurium*

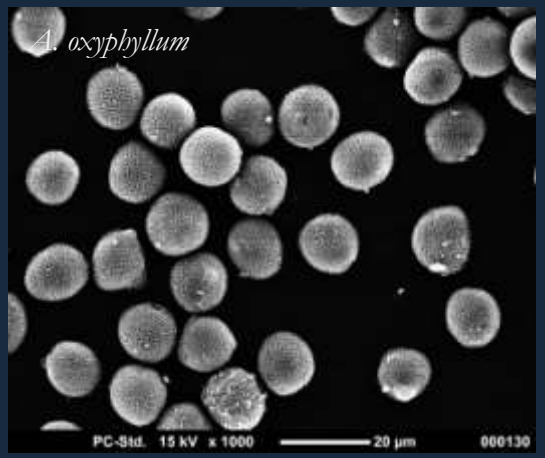
Materials and Methods: Data Collection

Reproductive morphology:

- 32 characters
- 98 species

Pollen:

- 15 characters
- 60 species



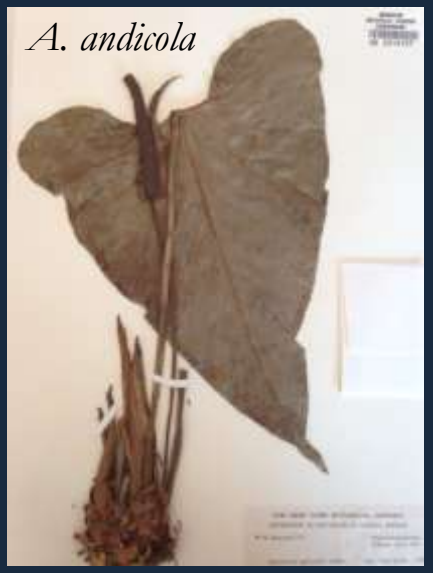
Inflorescence Data Collection

Observation of specimen under dissecting scope

Berries

Spadix

Epiphytic bryophyte



III. Methods

Reproductive characters (32)

Inflorescence (18)

Quantitative (6)

- Peduncle length
- Spathe length
- Spathe width
- Stipe length
- Spadix length
- Spadix diameter

Qualitative (12)

- Inflorescence length/leaf length
- Peduncle thickness /petiole thickness
- Peduncle length/petiole length
- Spathe shape
- Spathe 3D-shape
- Spathe disposition at anthesis
- Spathe color
- Presence of punctations on spathe
- Presence of stipe
- Spadix shape
- Spadix position
- Spadix color

Fruit (6)

Quantitative (2)

- Berry length
- Berry width

Qualitative (4)

- Berry color
- Shape of berry tip
- Berry seed abundance
- Spathe persistence when plant is in fruit

Flower (8)

Quantitative (6)

- Flowers/ right spiral
- Flowers/ left spiral
- Flowers/ principle spiral
- Flowers/secondary spiral
- Flower length
- Flower width

Qualitative (2)

- Stamens habit
- Presence of tepal protuber-ances


A. moodeanum


A. muyunense


A. buganum

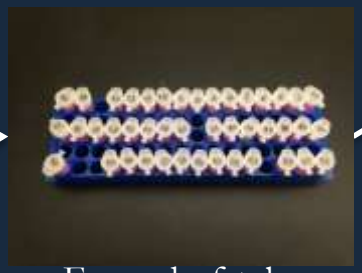

A. wagenurianum


A. crenatum

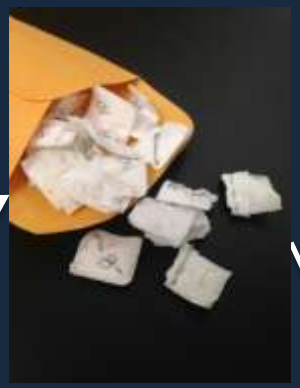
Collecting Pollen Data

Pollen from herbarium specimens

Removal of anthers



Eppendorf tubes



Kimwipes™ packets

Dehydration series

Critical-point dryer (Emitech K850)

Sputter-coater (Denton Vacuum Desk V)

Scanning electron microscope (JCM 5000 Neoscope)

Pollen from pickled collections

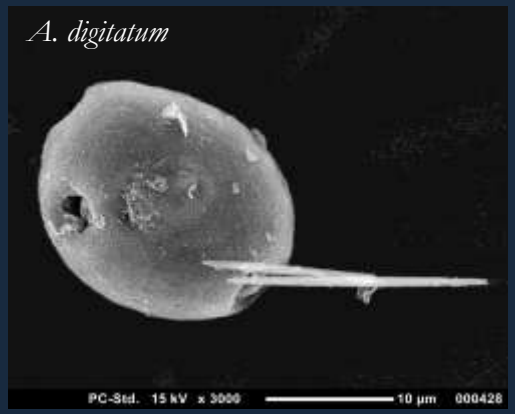
Pollen Characters (15)

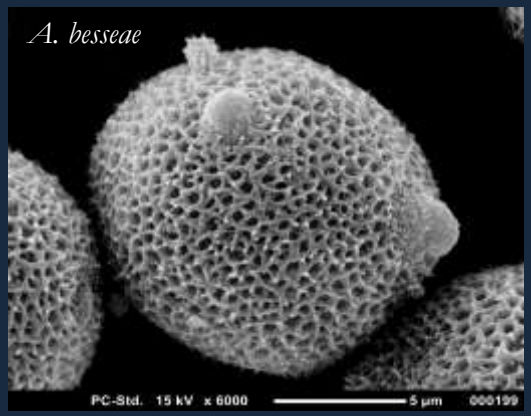
Quantitative (5)

- Lumen size
- Pollen range of sizes
- Pollen size
- Pollen range (variability)
- Aperture number

Qualitative (10)

- Pollen unit
- Pollen shape
- Pollen symmetry
- Pollen polarity
- Pollen aperture type
- Pollen aperture shape
- Exine ornamentation
- Muri type
- Presence of crystals
- Other characters

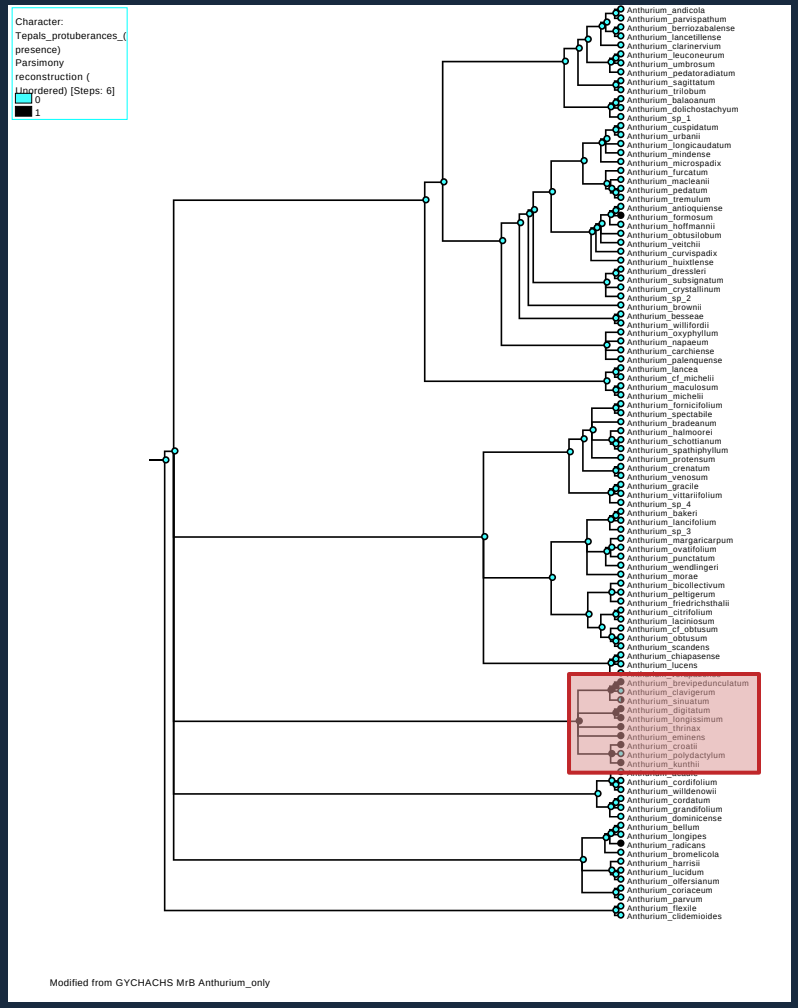




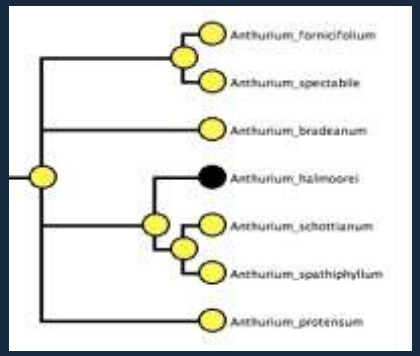
Materials and Methods: Data Analysis

- Mesquite software (Maddison & Maddison 2011) used to map character states onto molecular phylogeny (Carlsen & Croat 2013), for 47 different characters
- Trace Character History function used under Maximum Parsimony
- Synapomorphies identified for each clade

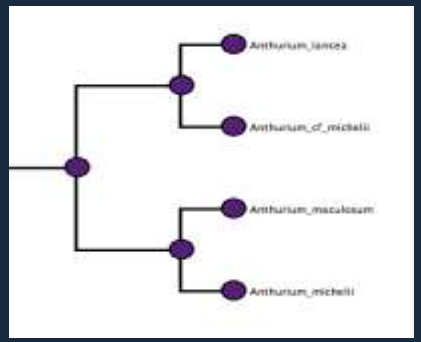
Results: Clade synapomorphies



Tepal protuberances: present or absent



Clade 9: pointed berries

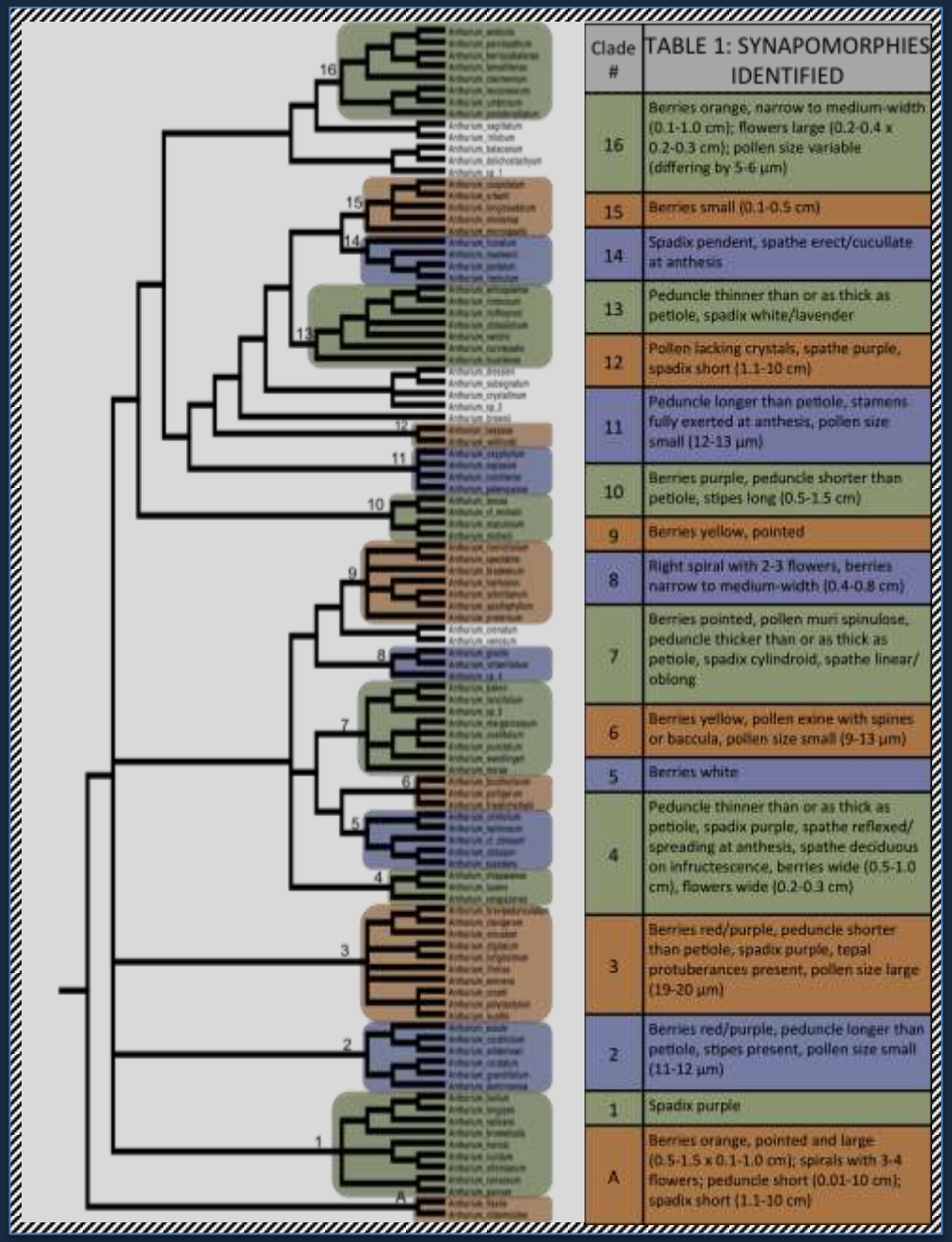


Clade 10: purple berries

Results: Best characters

- Fifty-eight synapomorphic character states identified
- Twenty-nine (62%) useful characters
- 100% fruit characters, 67% general inflorescence characters, 63% flower characters, and 40% pollen characters yielded synapomorphies
- Berry color, spadix color, peduncle/petiole length ratio, and berry width were most important

IV. Results



Clade #	TABLE 1: SYNAPOMORPHIES IDENTIFIED
16	Berries orange, narrow to medium-width (0.1-1.0 cm); flowers large (0.2-0.4 x 0.2-0.3 cm); pollen size variable (differing by 5-6 µm)
15	Berries small (0.1-0.5 cm)
14	Spadix pendent, spathe erect/cucullate at anthesis
13	Peduncle thinner than or as thick as petiole, spadix white/lavender
12	Pollen lacking crystals, spathe purple, spadix short (1.1-10 cm)
11	Peduncle longer than petiole, stamens fully exerted at anthesis, pollen size small (12-13 µm)
10	Berries purple, peduncle shorter than petiole, stipes long (0.5-1.5 cm)
9	Berries yellow, pointed
8	Right spiral with 2-3 flowers, berries narrow to medium-width (0.4-0.8 cm)
7	Berries pointed, pollen muri spinulose, peduncle thicker than or as thick as petiole, spadix cylindroid, spathe linear/oblong
6	Berries yellow, pollen exine with spines or baccula, pollen size small (9-13 µm)
5	Berries white
4	Peduncle thinner than or as thick as petiole, spadix purple, spathe reflexed/spreading at anthesis, spathe deciduous on infructescence, berries wide (0.5-1.0 cm), flowers wide (0.2-0.3 cm)
3	Berries red/purple, peduncle shorter than petiole, spadix purple, tepal protuberances present, pollen size large (19-20 µm)
2	Berries red/purple, peduncle longer than petiole, stipes present, pollen size small (11-12 µm)
1	Spadix purple
A	Berries orange, pointed and large (0.5-1.5 x 0.1-1.0 cm); spirals with 3-4 flowers; peduncle short (0.01-10 cm); spadix short (1.1-10 cm)

Each clade was supported by 1-9 synapomorphies

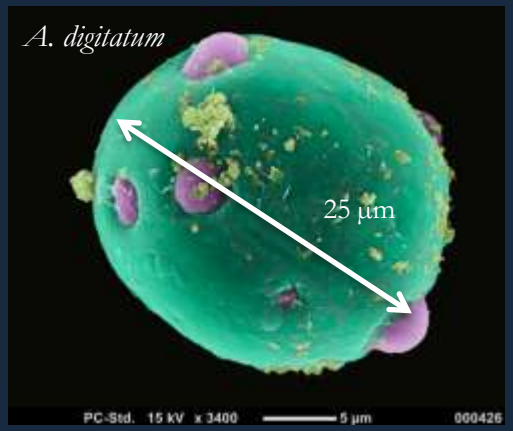
Clades A, 3, 4 and 7 were best supported

Clade A

http://sura.ots.ac.cr/

Synapomorphy: orange, pointed berries

Clade 3



Synapomorphy: large pollen grains

Clade 7

Synapomorphy: spinulose muri

Clade 13

Synapomorphy: white/lavender spadix

Conclusion

Since all 17 clades could be identified with reproductive character shifts, we conclude that reproductive changes have played a major role in the diversification of *Anthurium*, as proposed by Grayum (1990).

Future Studies

- Increase sample size, given that current datasets include only ca. 7-11% of *Anthurium* species (some characters may be more or less important when sampling size is increased)
- Include more characters
- Redefine some character states (e.g. colors)
- Pollination and fruit dispersal could be compared to morphological data

Acknowledgements

- Angie Macias
- Dr. Tom Croat
- Washington University – Microscopy & Digital Imaging Core
- REU Students
- Missouri Botanical Garden and Staff
- National Science Foundation – REU Program