Pollen and Stigma Biology

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2. Pollen and Stigma Biology

- Role of pollen for plant reproductive success and species conservation
- Pollen Development
- Pollen cytology

- Pollen morphology
- Pollen taxonomy

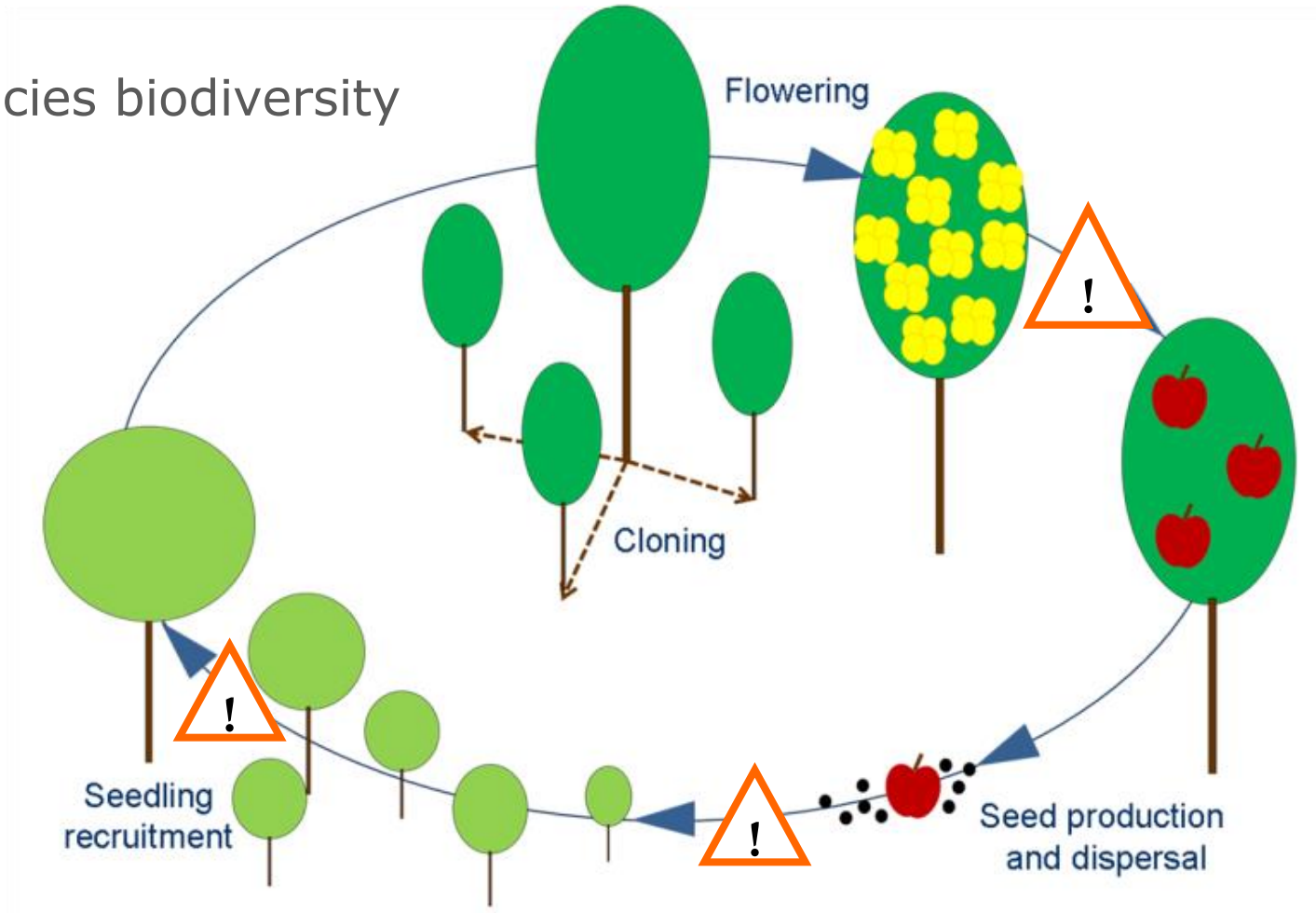
- Pollen functionality
- Methods to assess pollen viability and pollen germination

- Study cases on the effects of environmental factors on pollen functionality

Plant reproductive success and species conservation

Successful reproduction is basic for

- Generation turnover
- Maintaining within-species biodiversity



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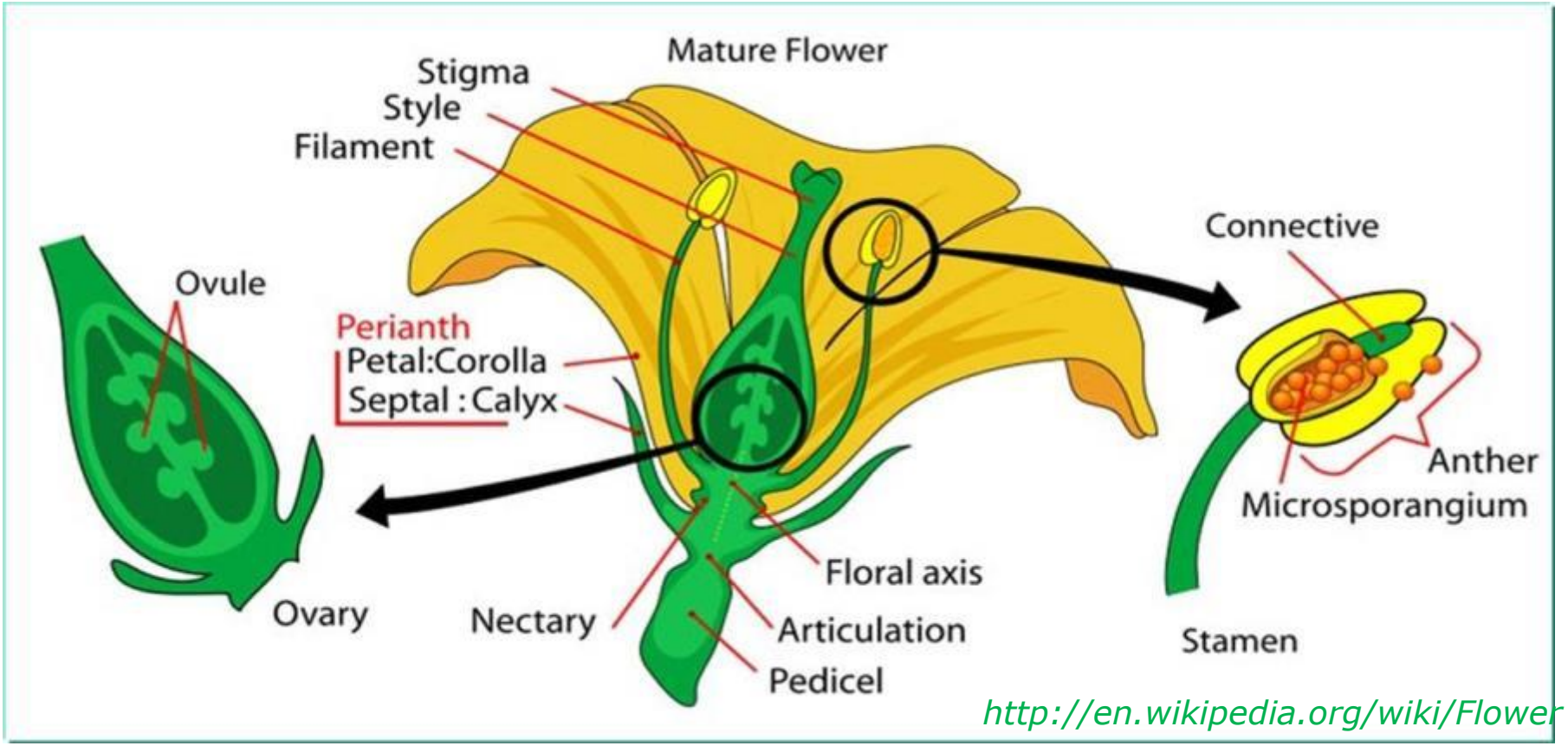
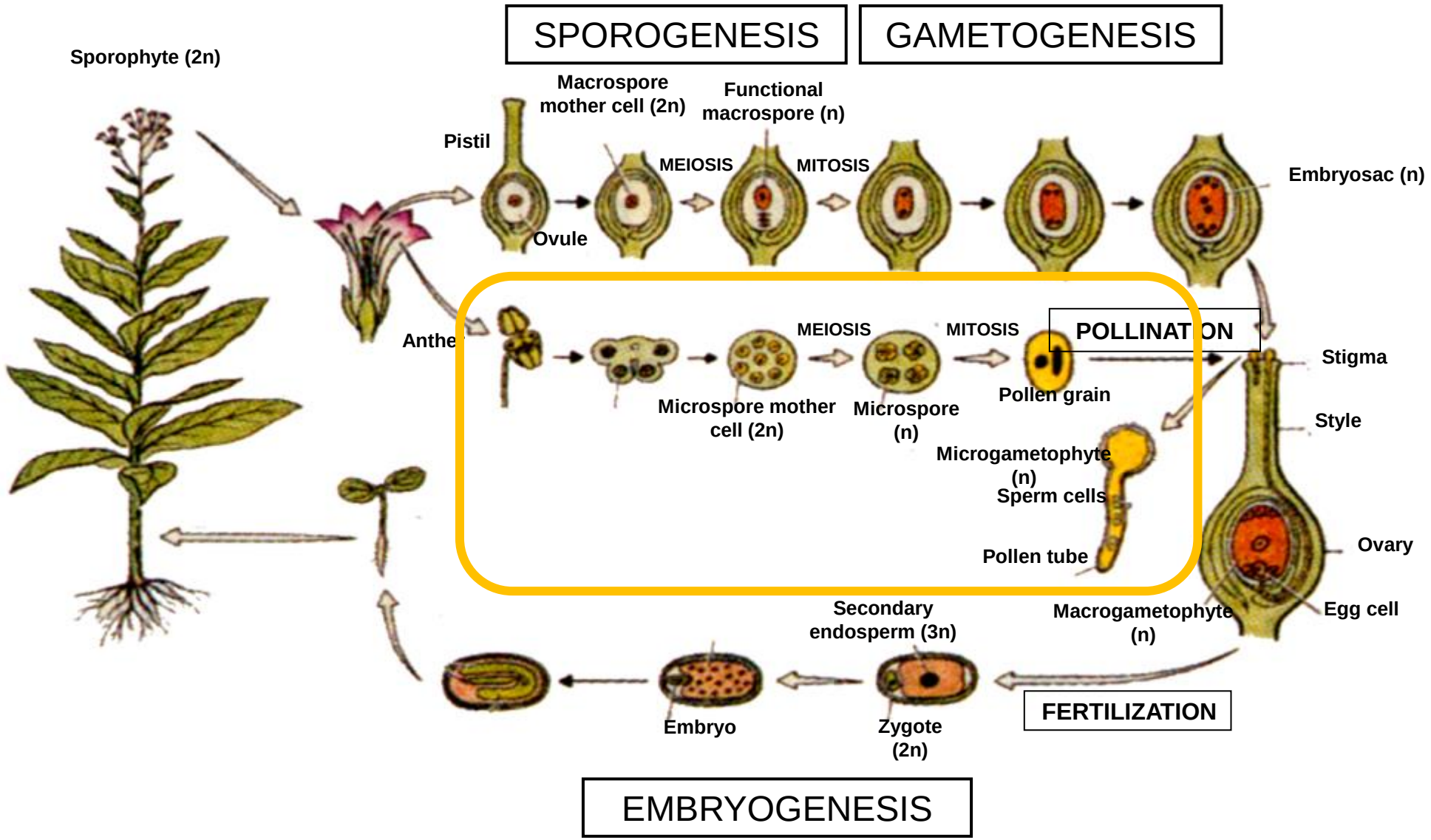
Angiosperm reproductive organs

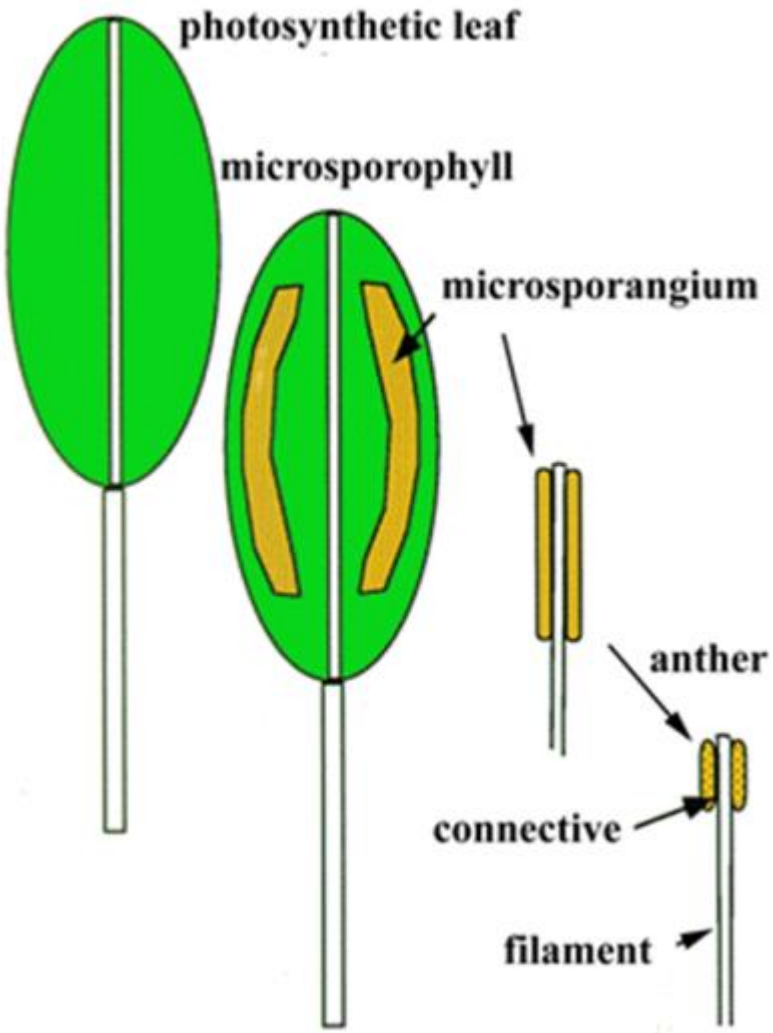


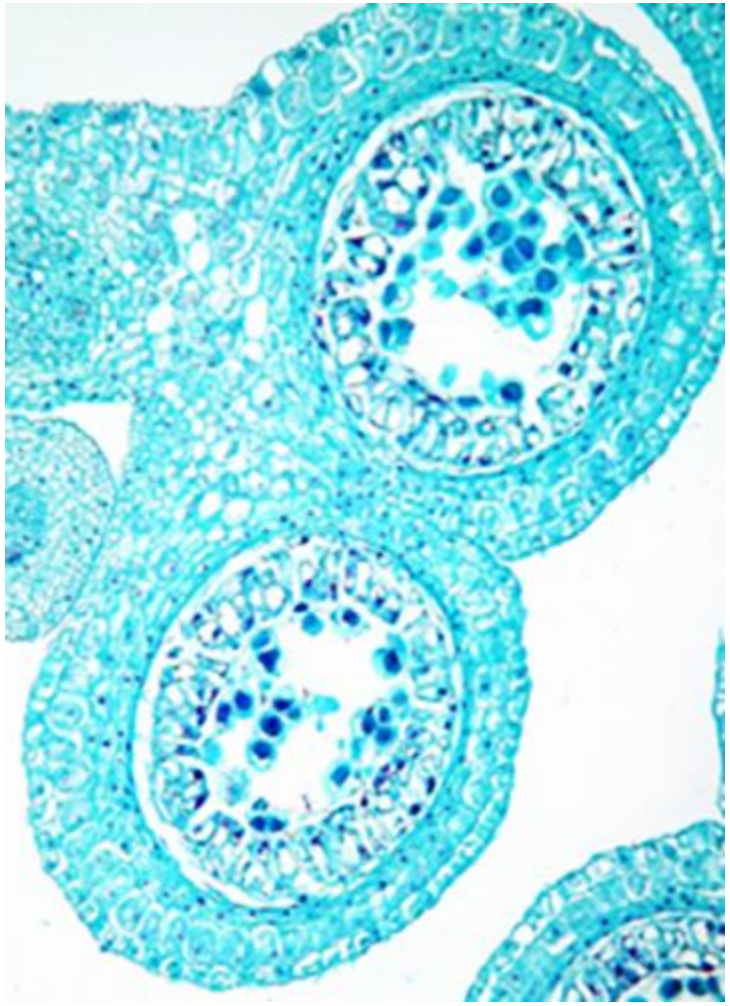
Flowers are modified shoots a) to produce spores and gametes, b) to attract pollen vectors and c) to finally produce and disperse the embryos

Angiosperm life cycle

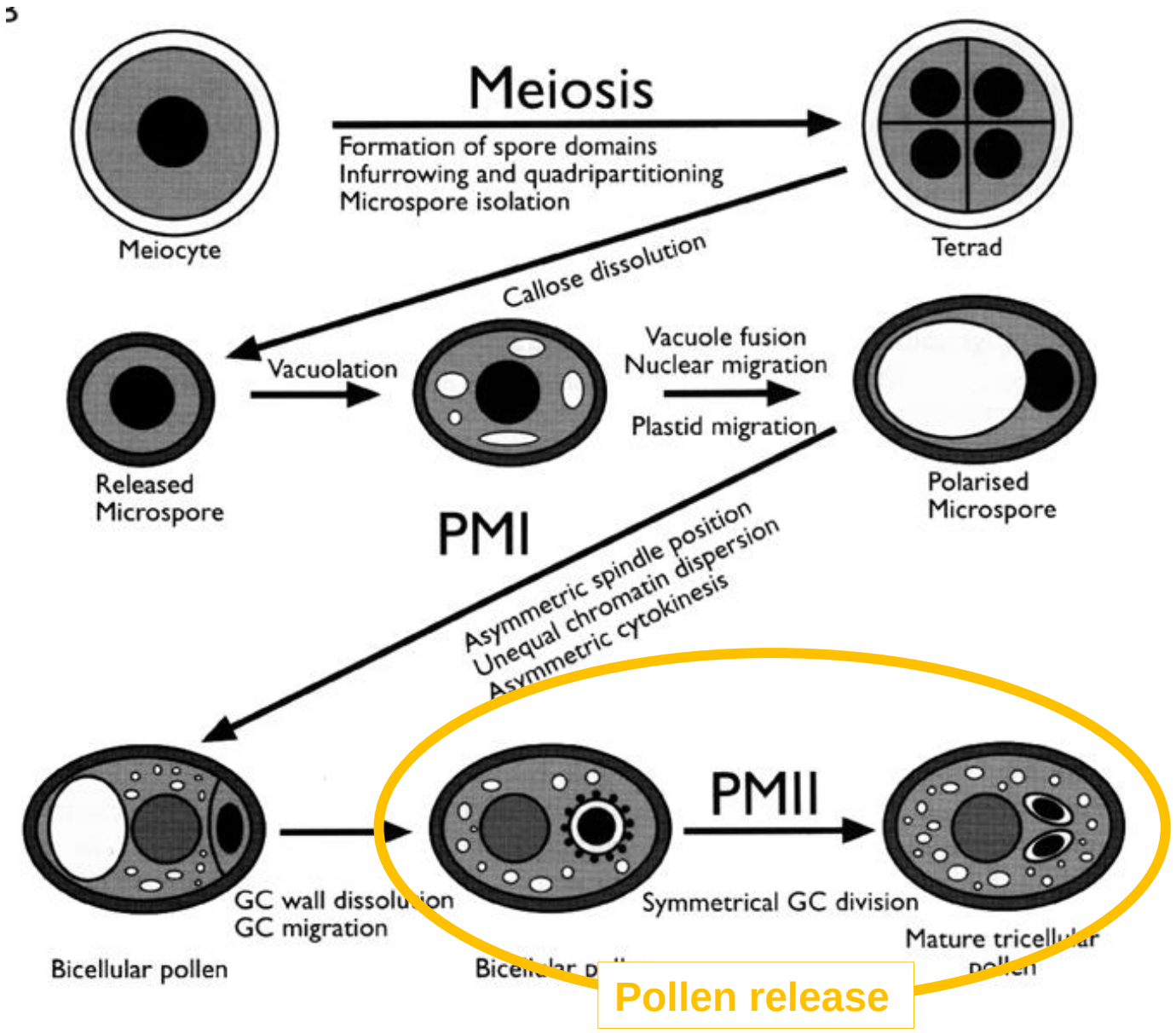


Anther structure and pollen development

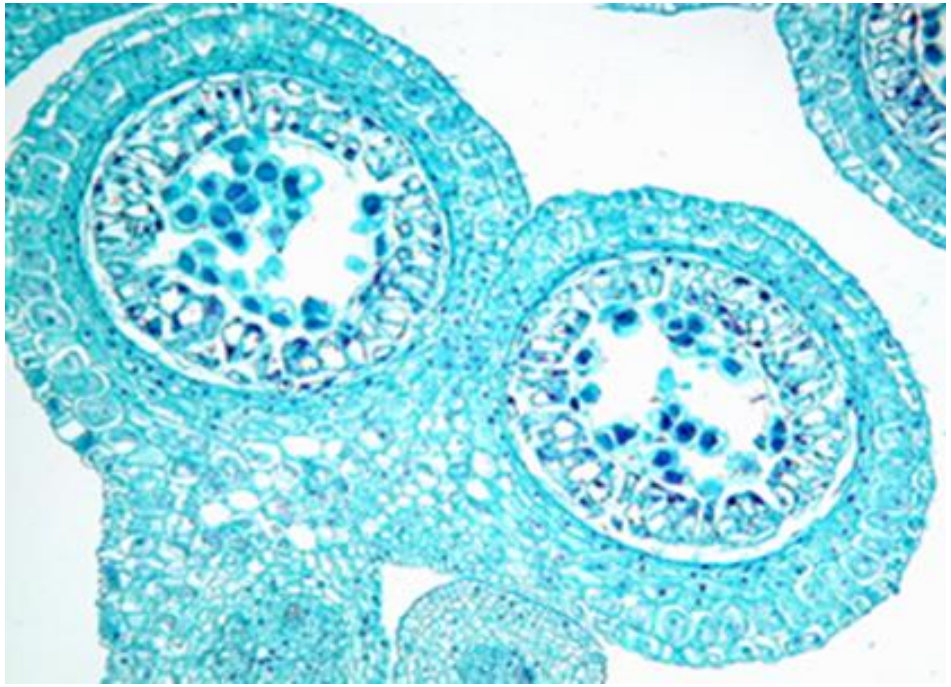




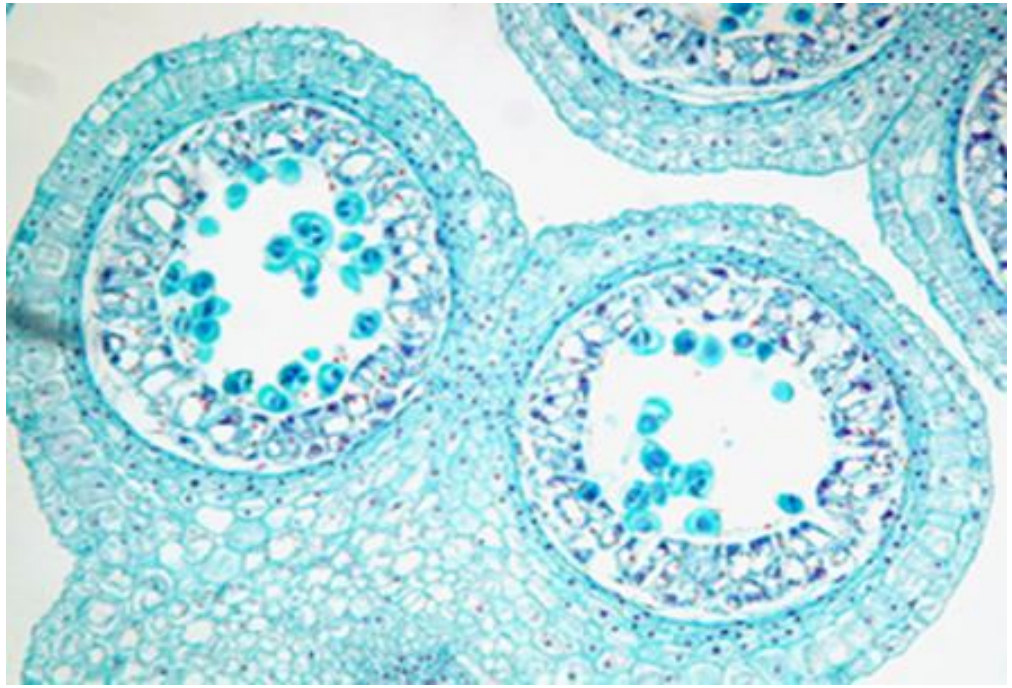
Microsporogenesis

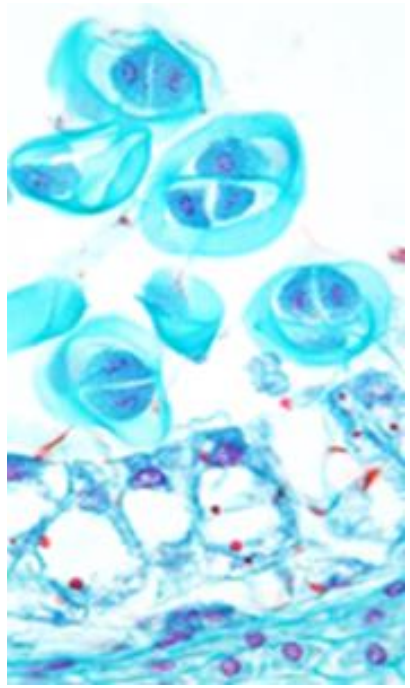


Pollen development



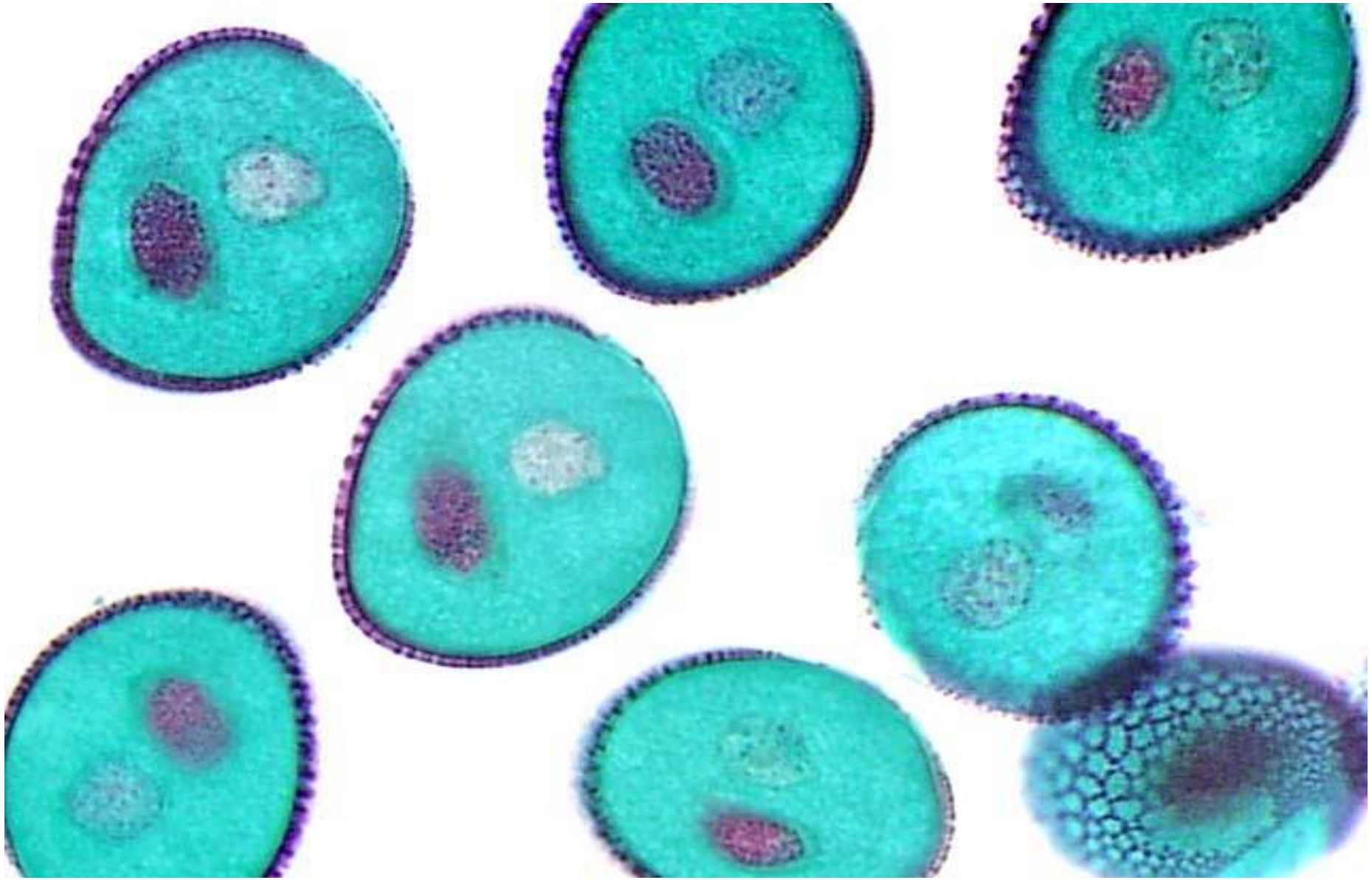
Meiosis I



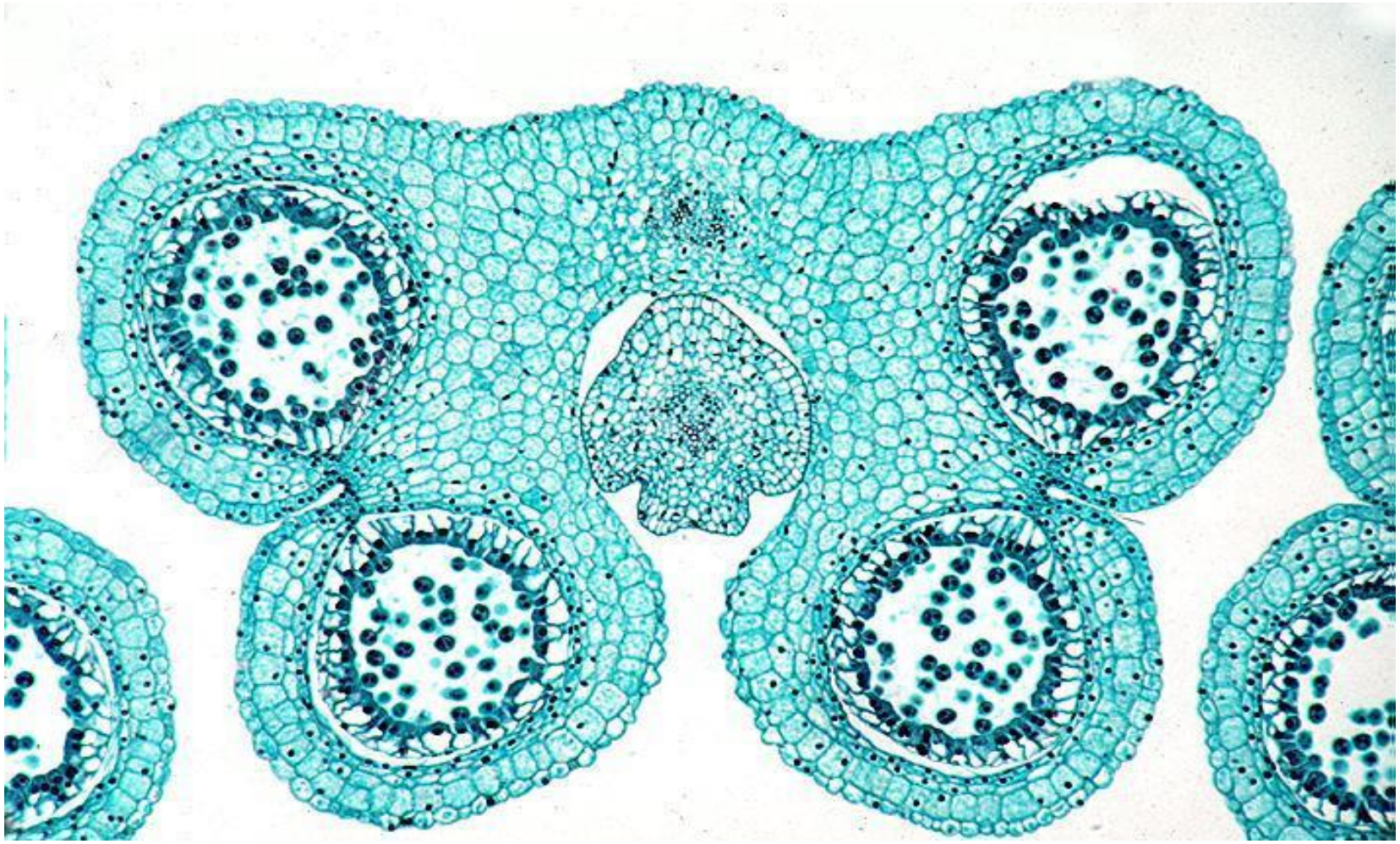


Tetrads

Mature pollen

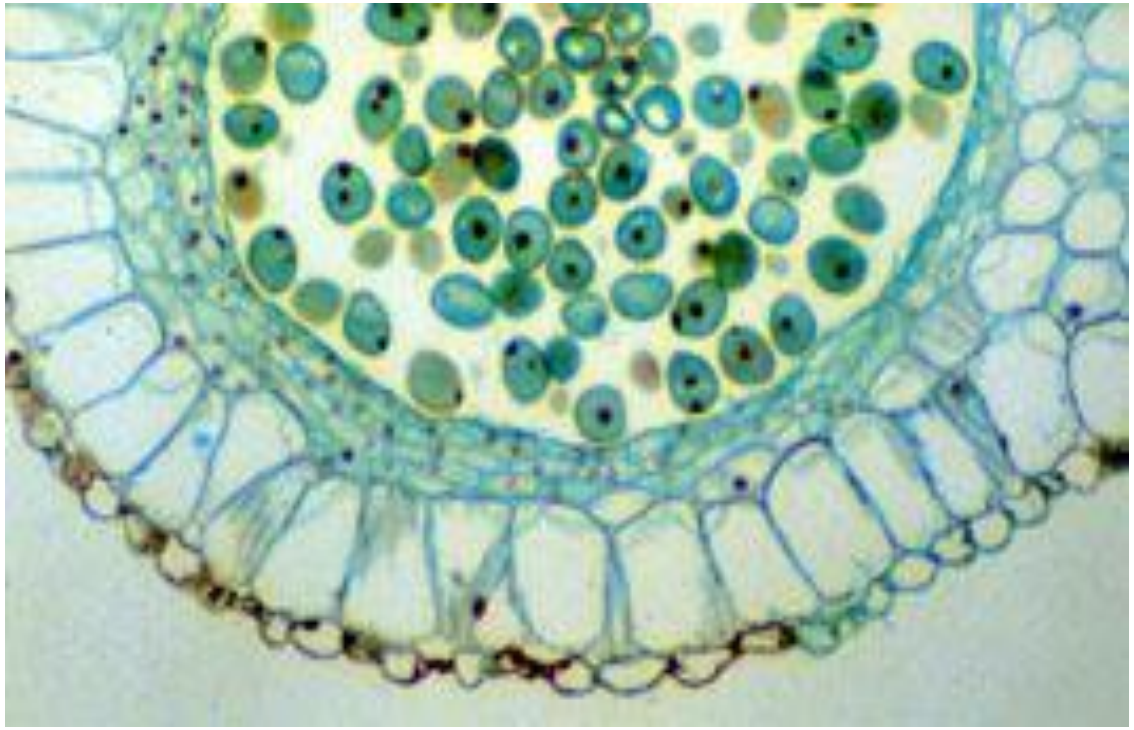


Tapetum



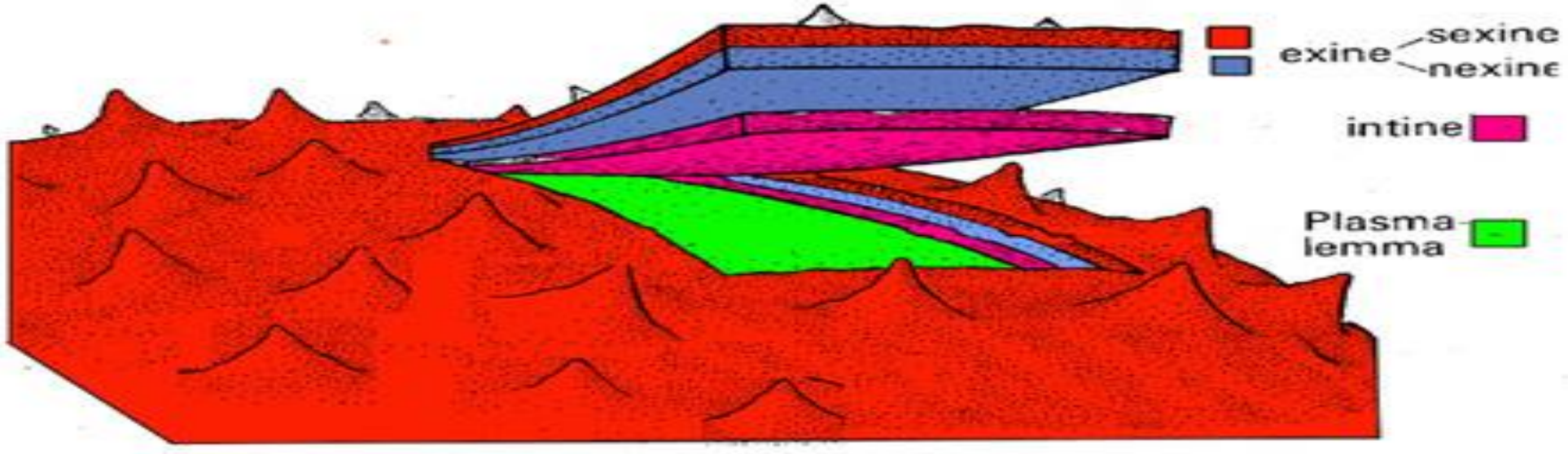
- Plasmodial tapetum
- Secretory tapetum

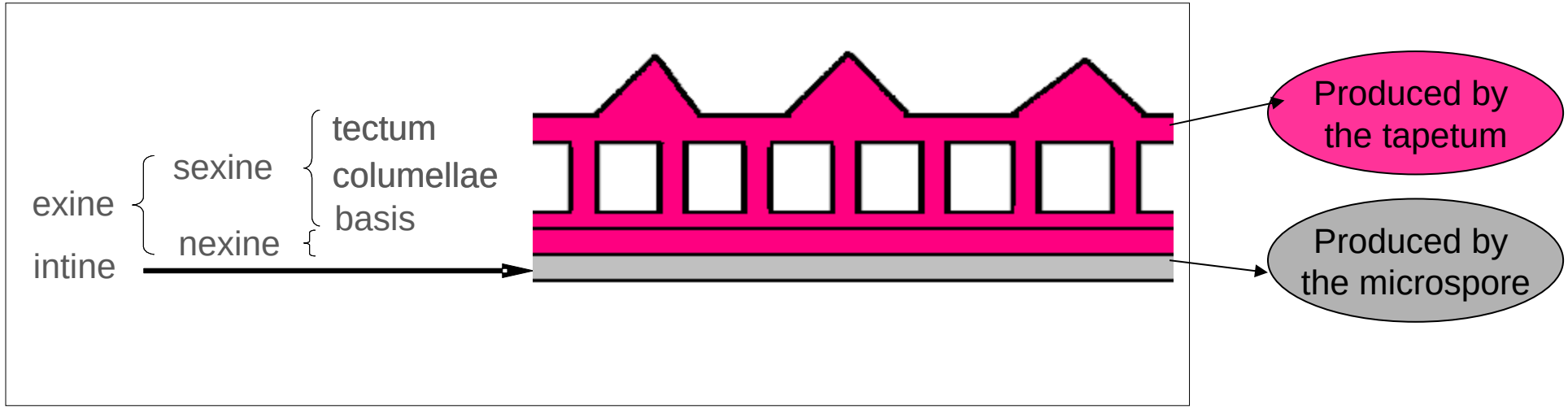
Tapetum functions



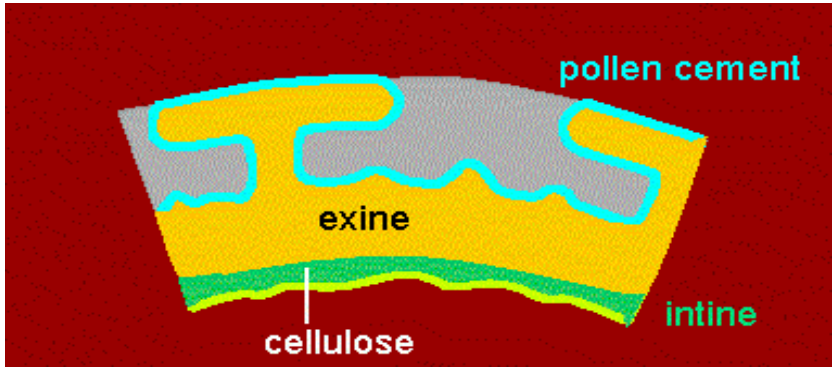
Functions:
-To co-operate in exine development
-To nourish the microspores
-To take part in triphyne and pollenkitt formation

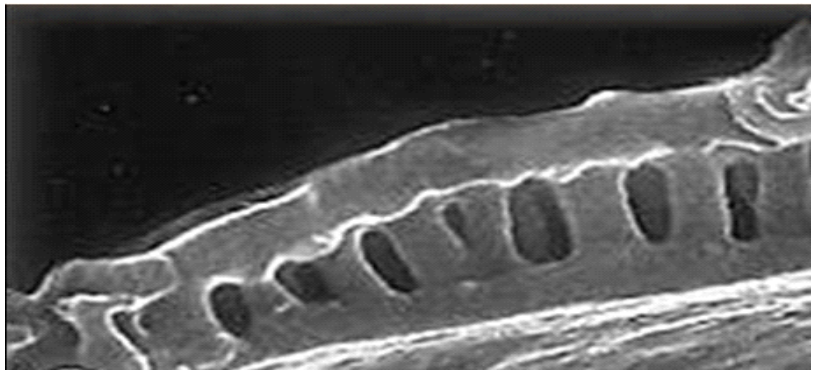
Pollen wall structure





Pollen wall composition

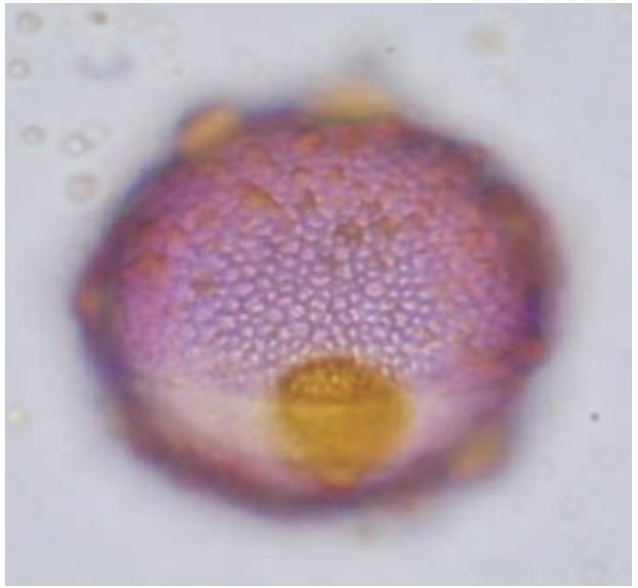




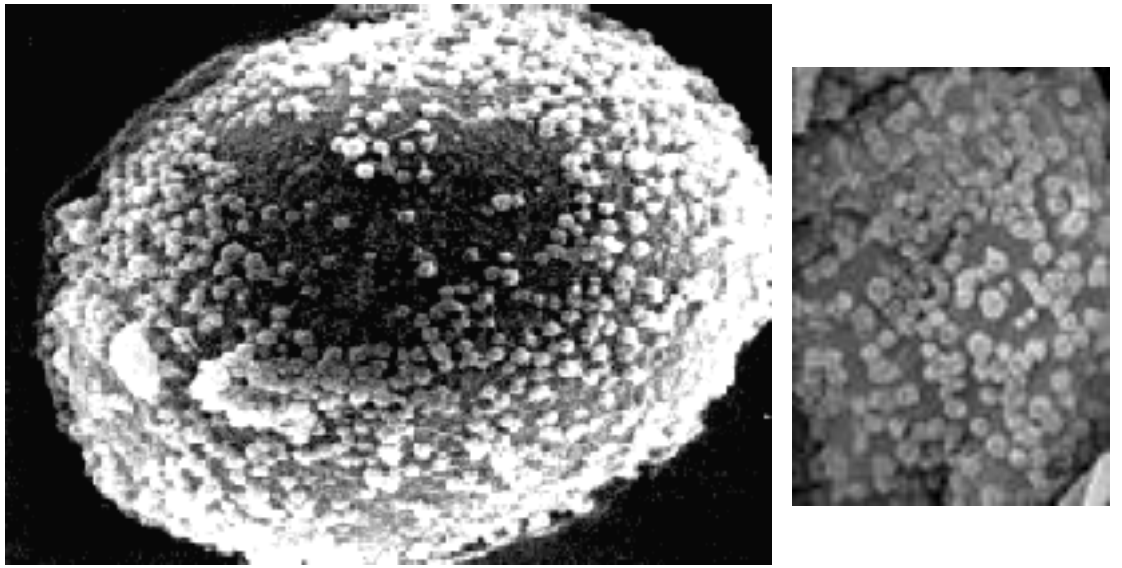
INTINE: **Cellulose and pectin.**

EXINE: **Sporopollenin.**
It is a complex substance and its exact composition is not completely defined.
There are biopolymers with long chain fatty acids, phenylpropanoid, phenols, carotenoids. The phenylalanine may be its precursor.

Elements outside the exine

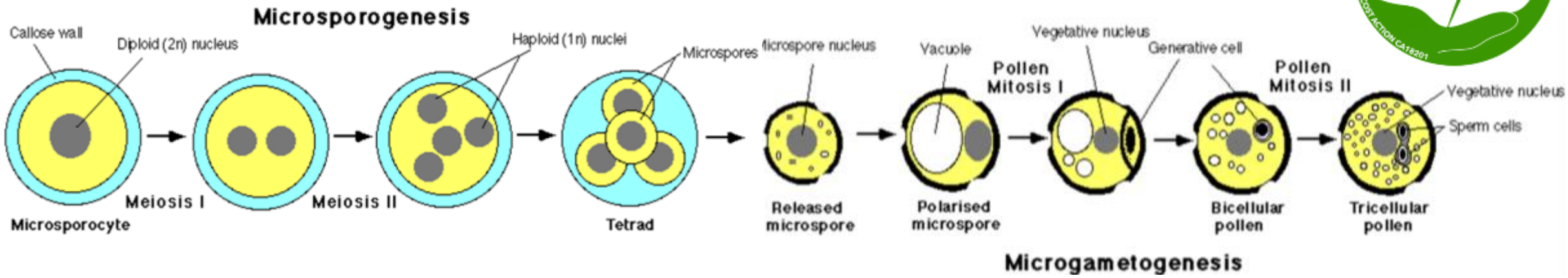


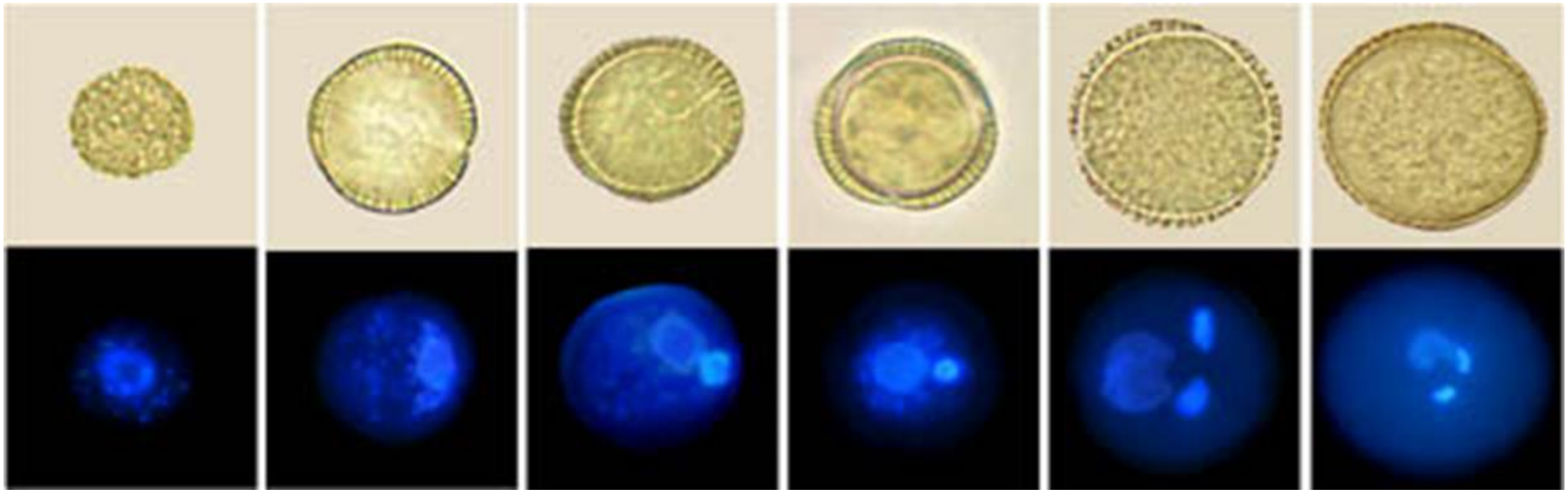
Pollenkitt



Orbicoles – Ubish bodies

Pollen cytology

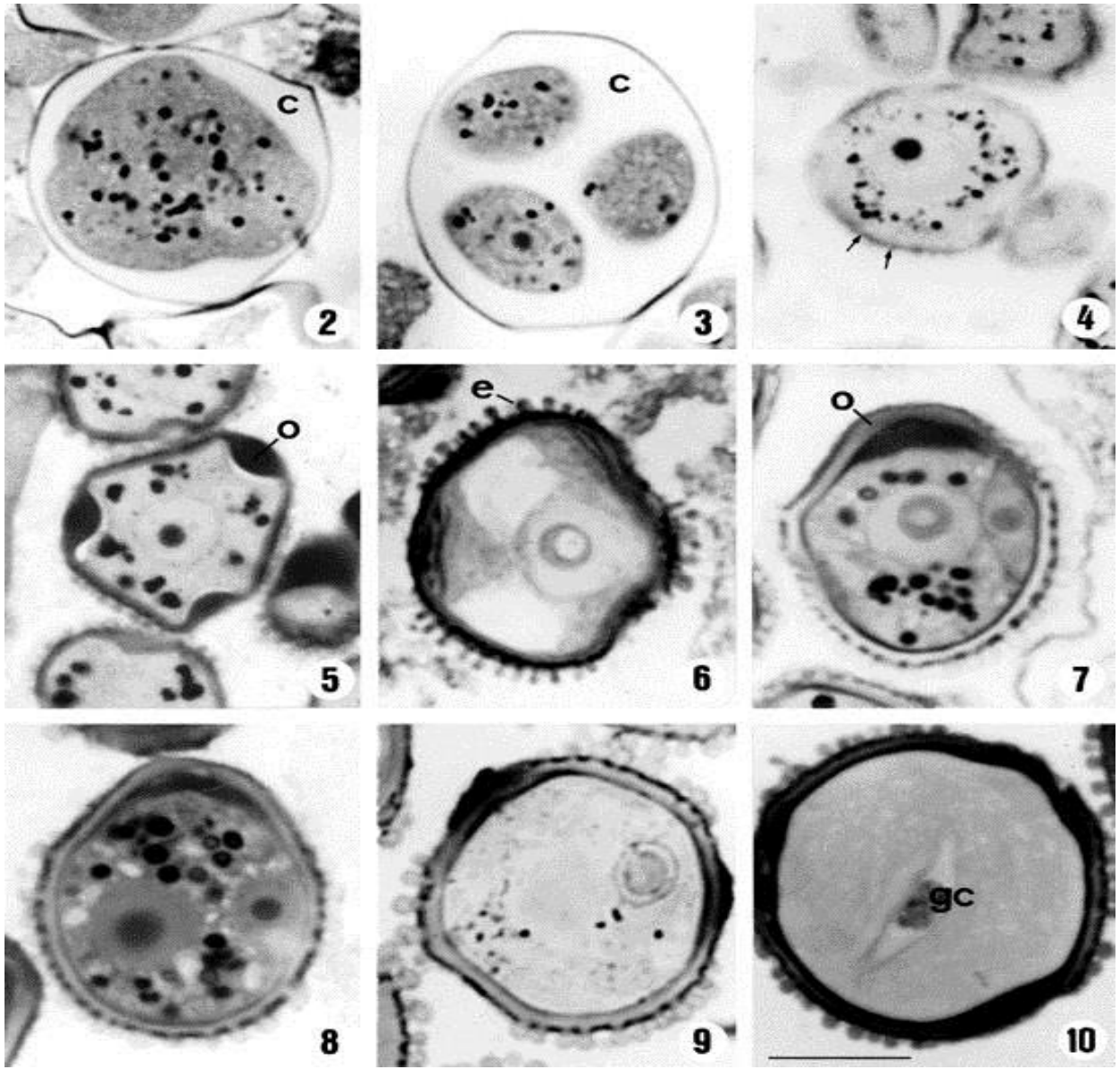


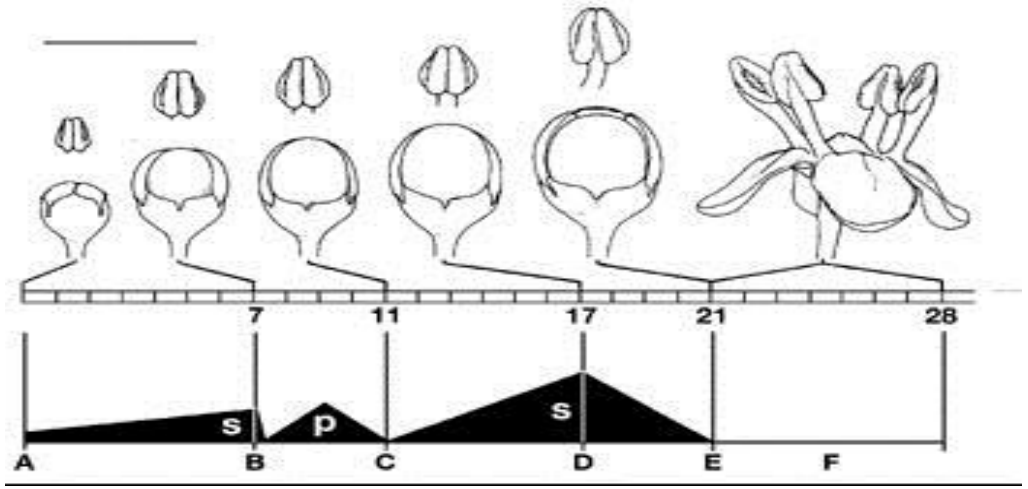


LM

FM
DAPI

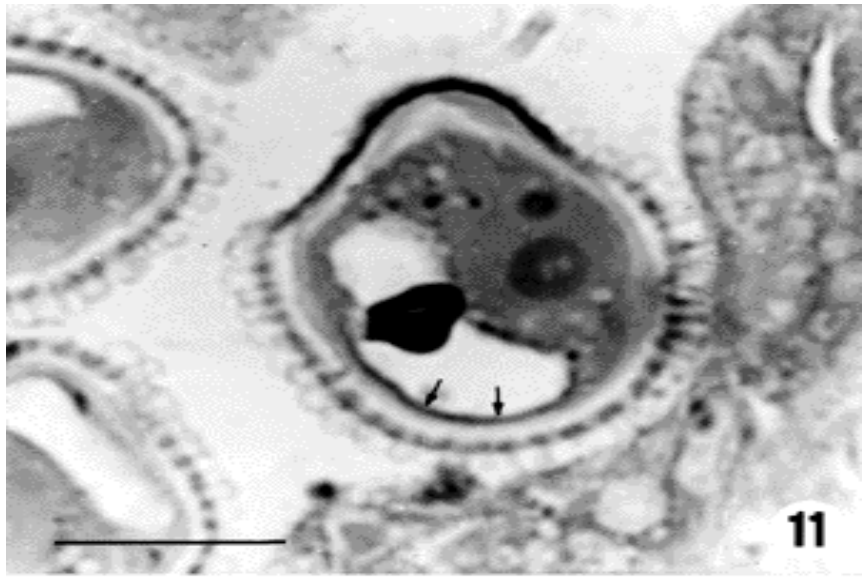
Cytoplasmic content

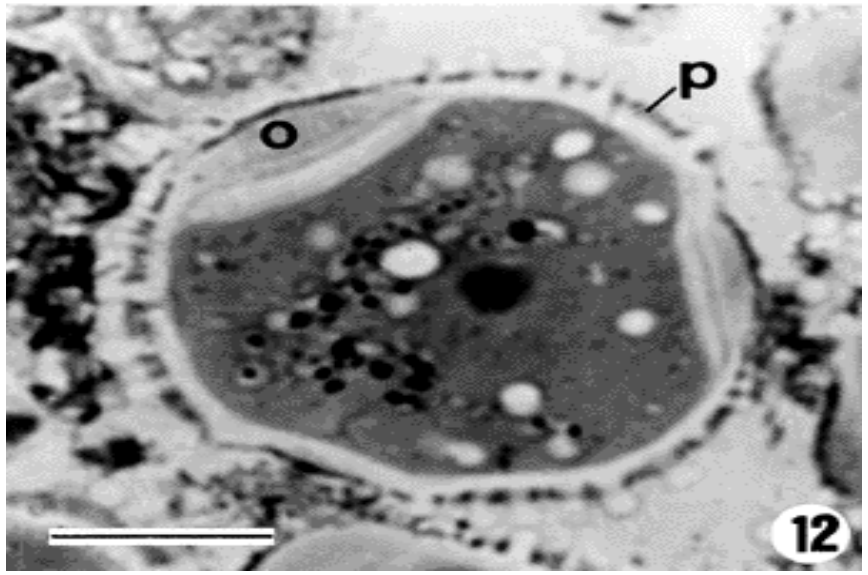




Dynamic of starch (s) and protein (p) storage in *Ilex paraguariensis*

Cytoplasmic content





Presence of protein, in the vacuole on the tonoplast (11) and exine (11-12) of *Ilex paraguariensis* pollen

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- Pollen functionality
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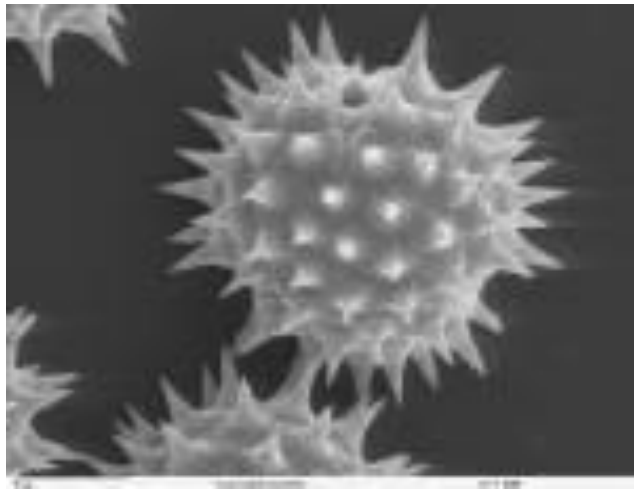
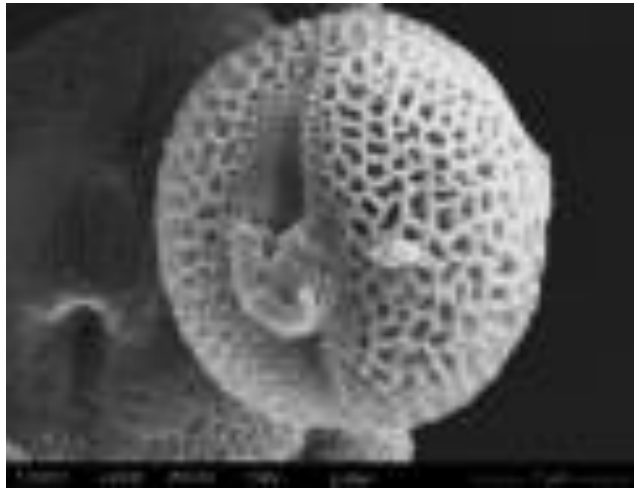
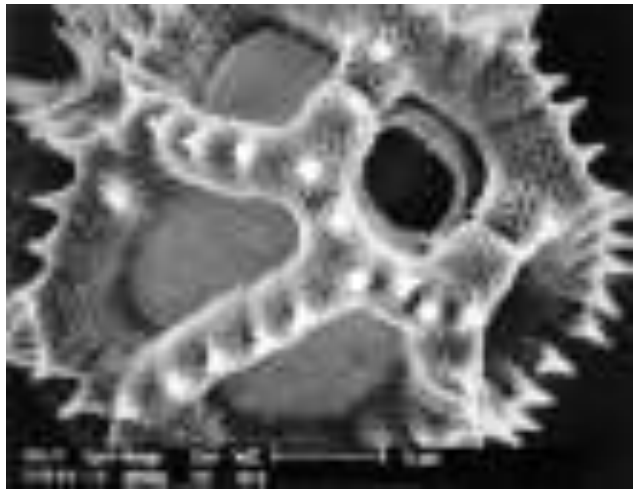
Pollen morphology and palynology

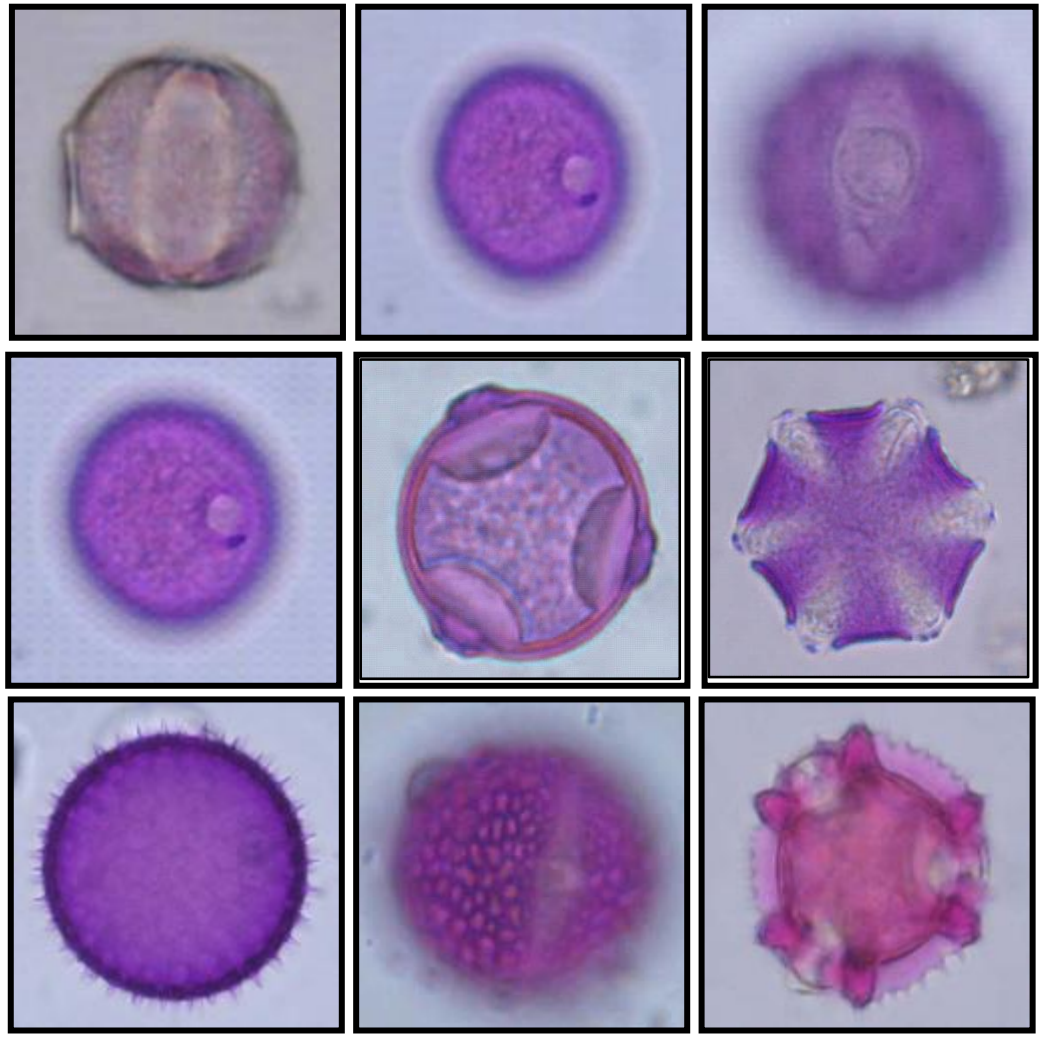






Palynology: the study of spores and pollen morphology, often used to identify a particular plant taxon (paleopalynology, forensic palinology, melissopalynology, …)

Pollen taxonomy

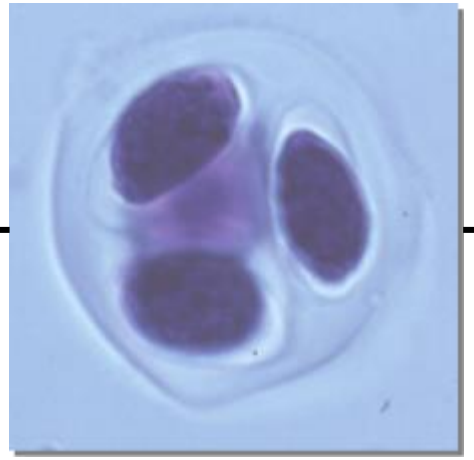


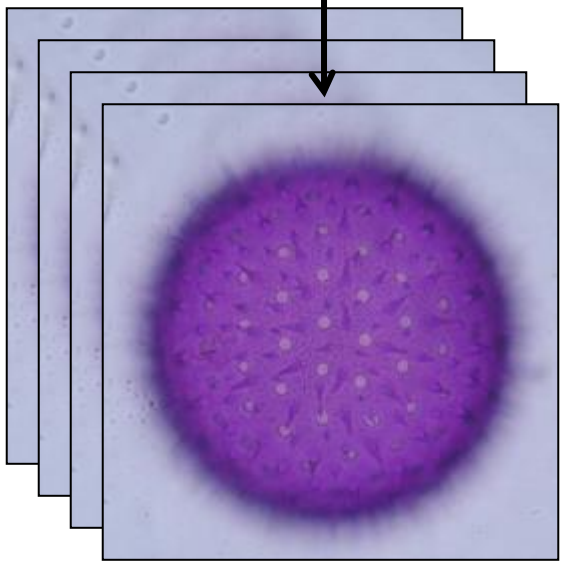
Pollen taxonomy is based on pollen grain morphology and exine sculpture.

It requires highly skilled experts but builds on a few simple criteria:

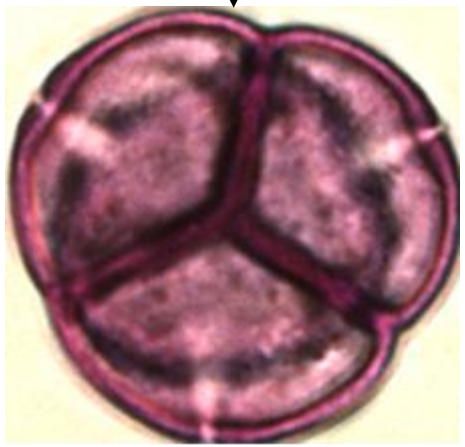
- Aggregation
- Polarity
- Shape
- Apertures
- Ornamentation

Criteria for pollen taxonomy: Aggregation

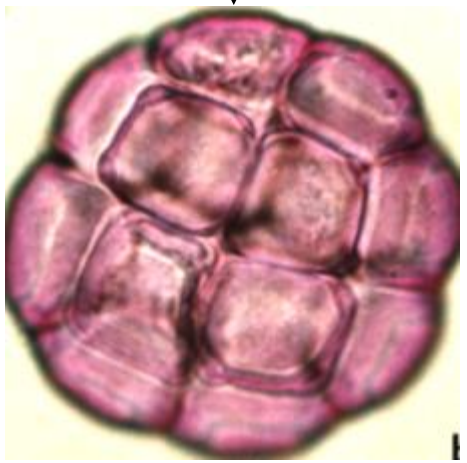




Monad

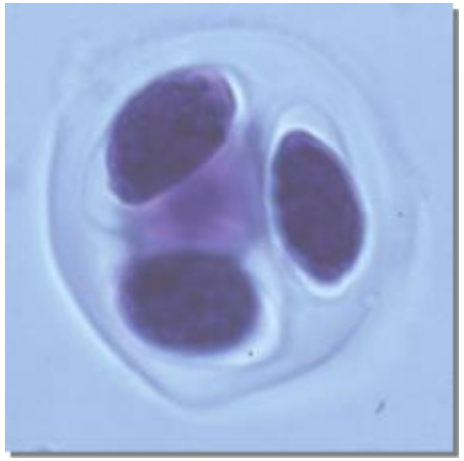


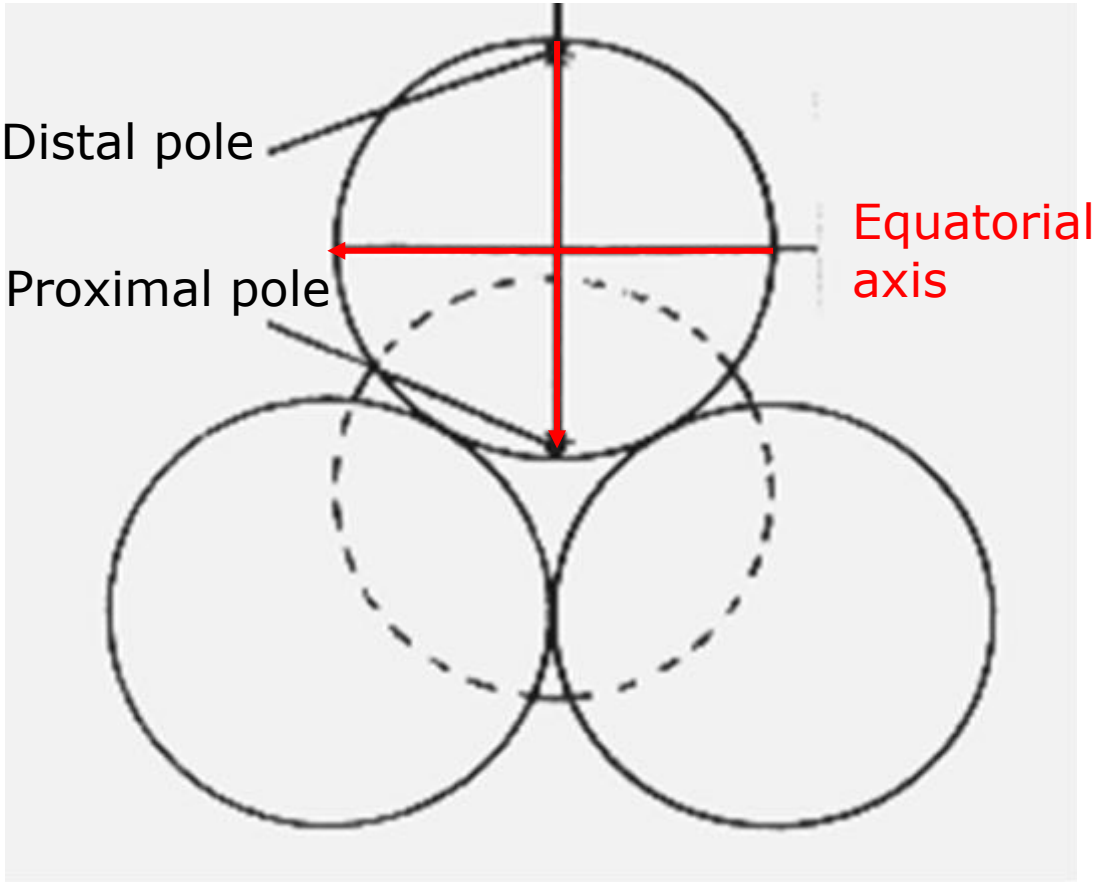
Tetrad



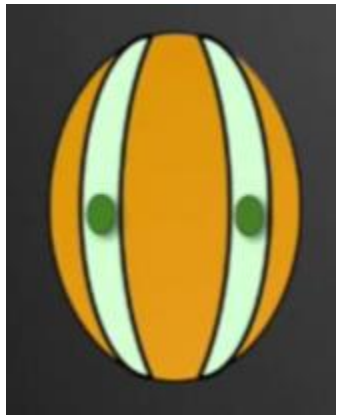
Polyad

Criteria for pollen taxonomy: Polarity





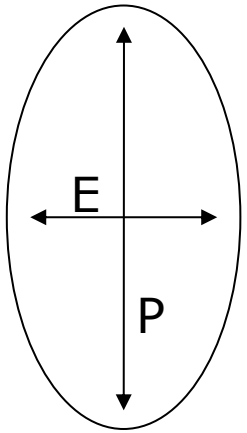
Pollen tetrad



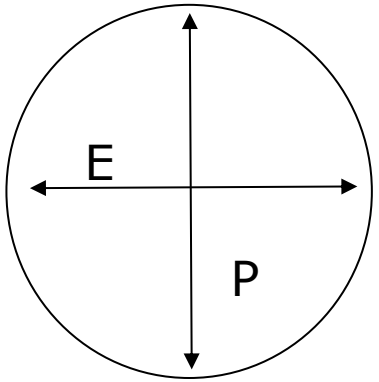
Isopolar

Heteropolar

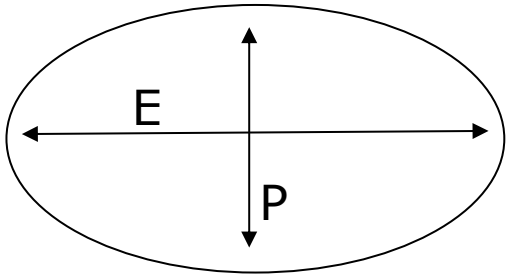
Criteria for pollen taxonomy: Shape



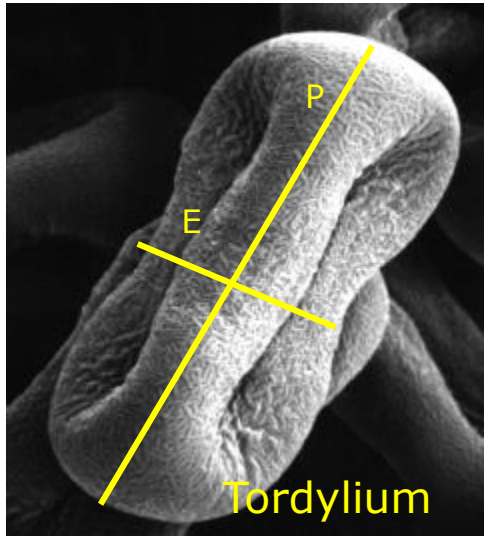
Prolate

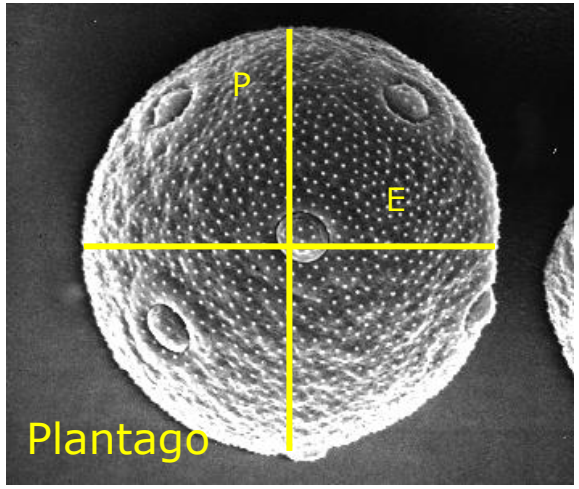


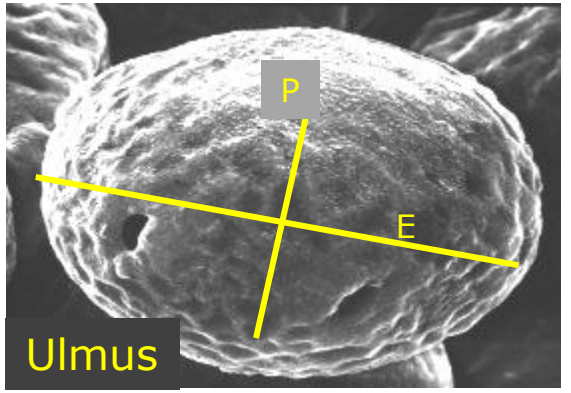
Spherical



Oblate

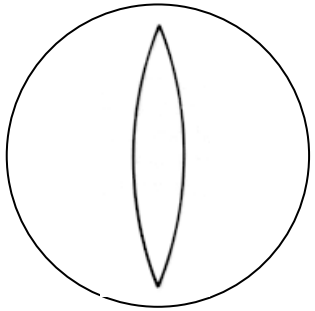




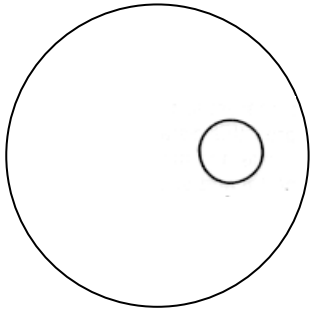


Criteria for pollen taxonomy: Apertures

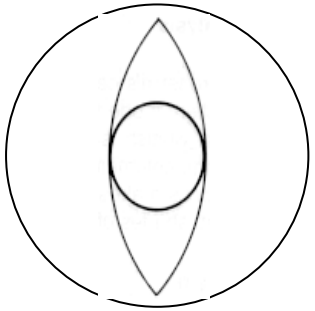
Aperture: an area of the pollen wall with a thin or missing part of the exine



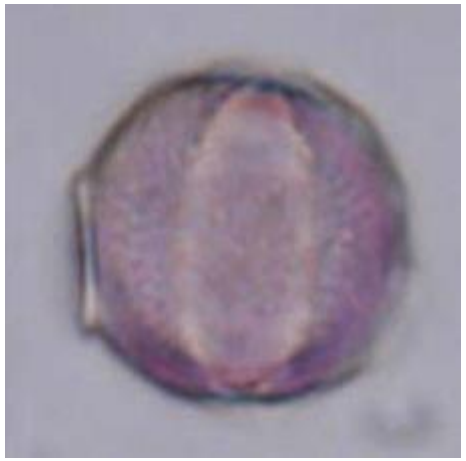
Colpus



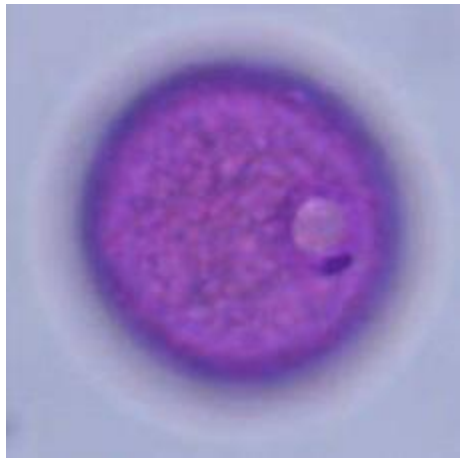
Porus



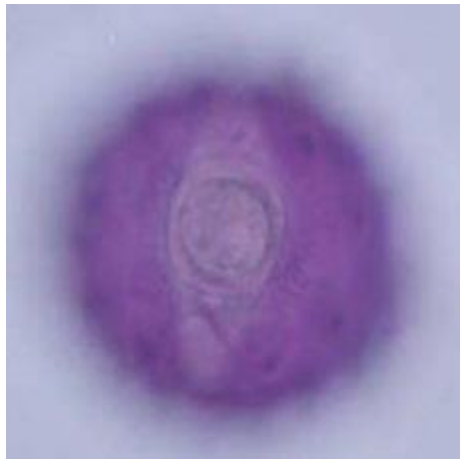
Colpus + Porus



Colpate

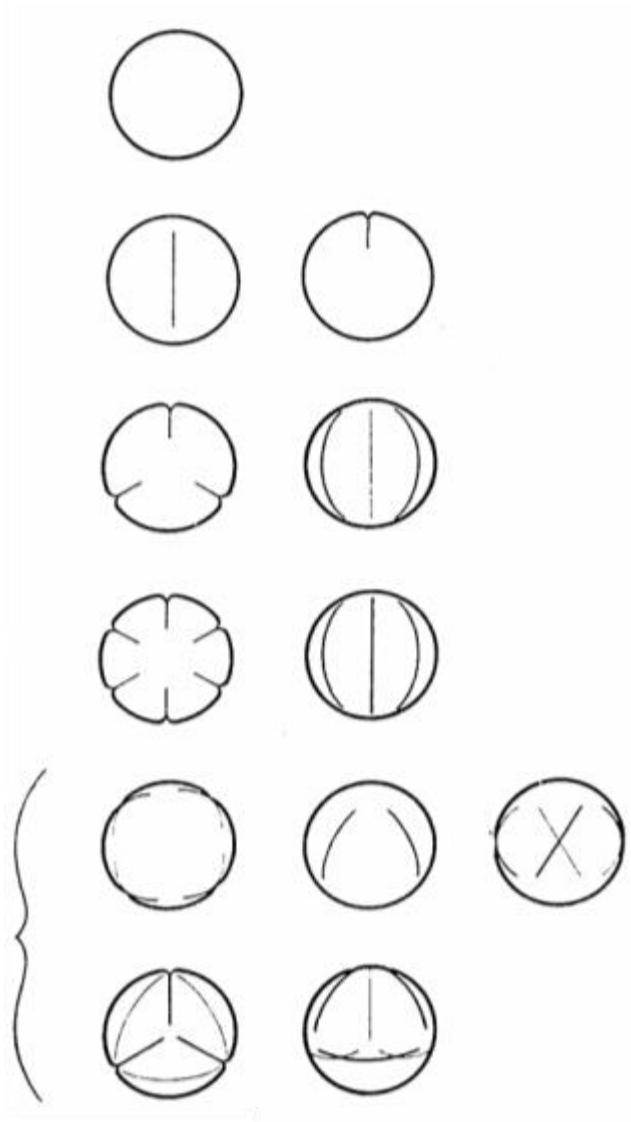


Porate

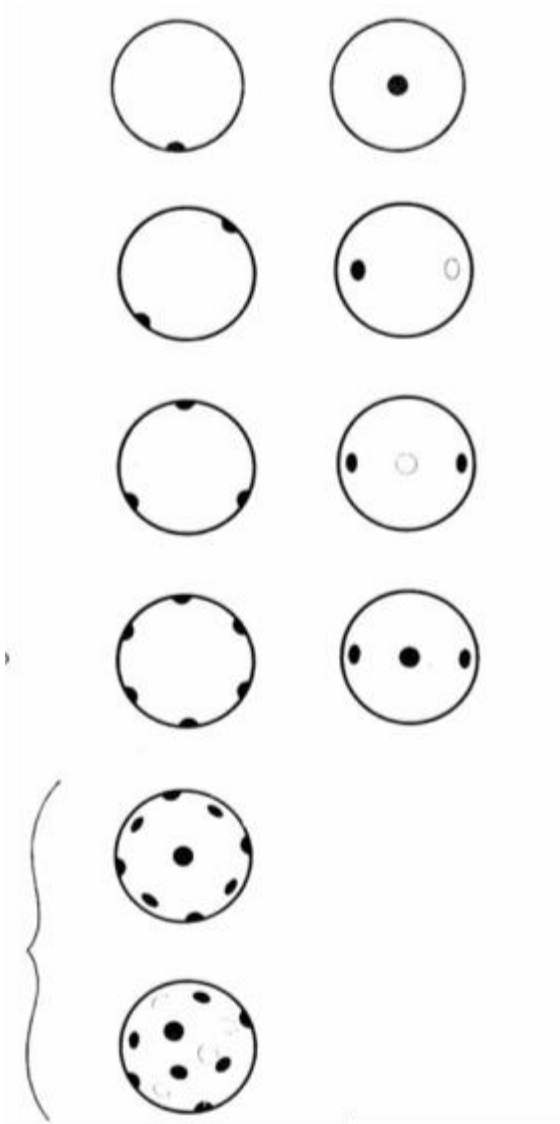


Colporate

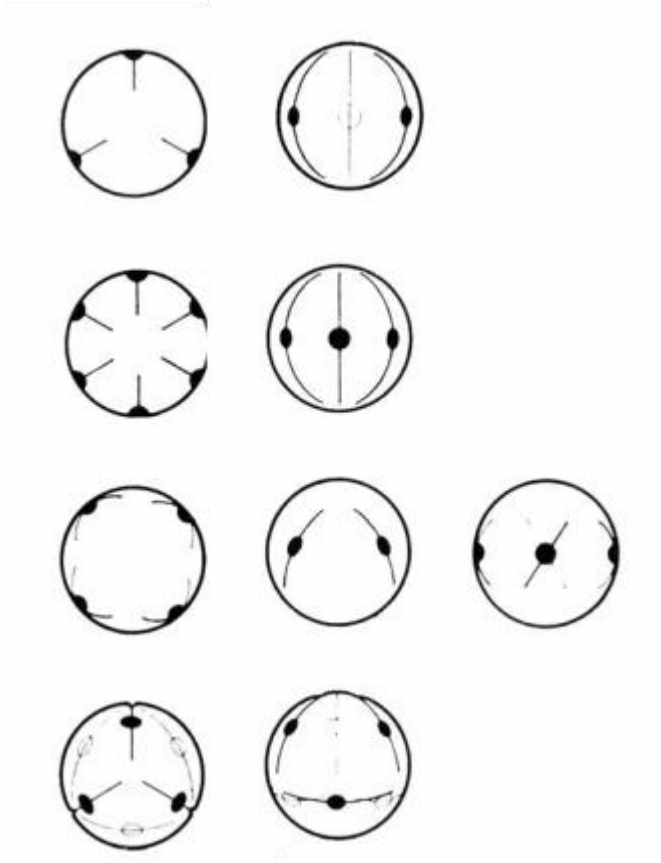
Criteria for pollen taxonomy: Number of apertures



Mono-, Tri-, …., Zono-colpate

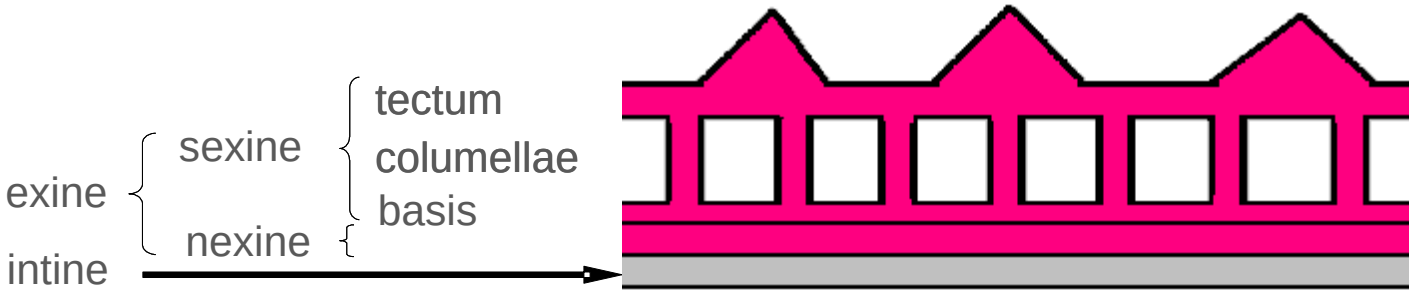


Mono-, Tri-, …., Panto-porate

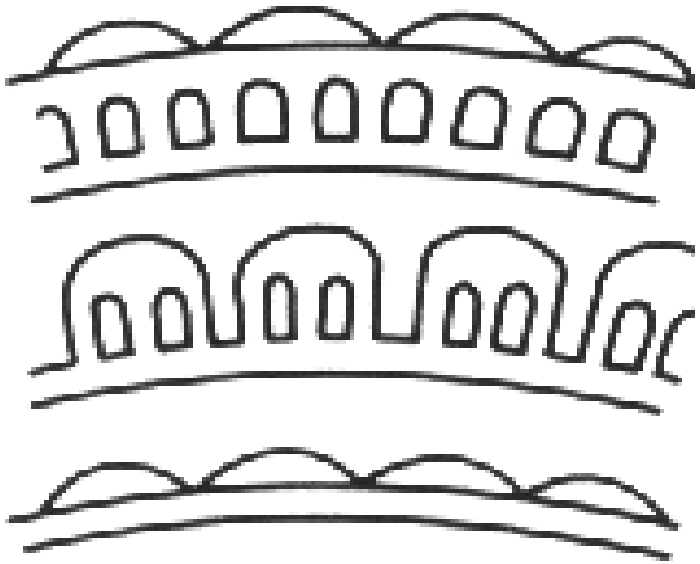


Mono-, Tri-, …., Zono-colporate

Criteria for pollen taxonomy: Ornamentation



Sexine sculpturing

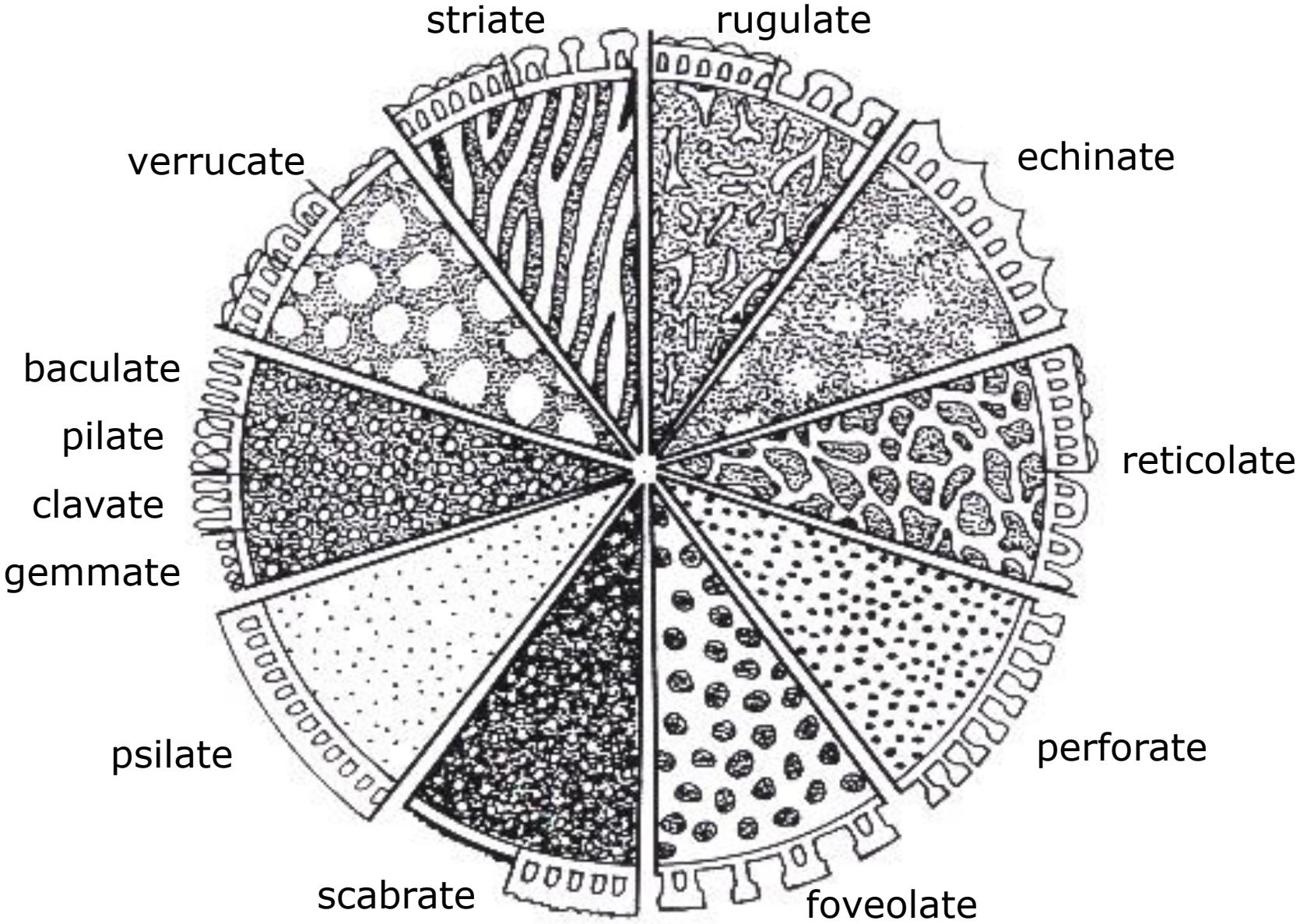


Tectum: complete (tectate grain)

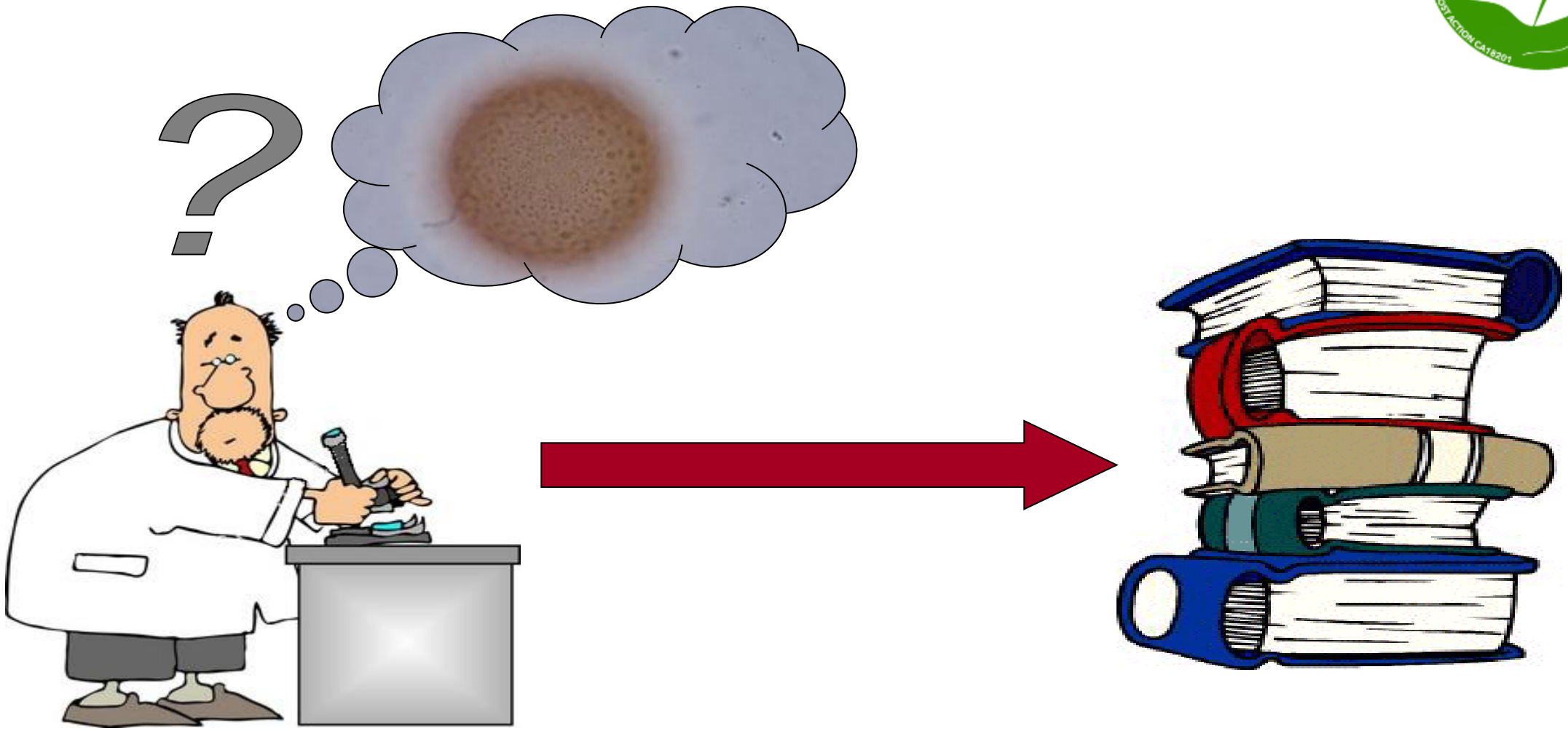
Tectum: partially dissolved (semitectate grain)

Tectum: completely absent (intectate grain)

Criteria for pollen taxonomy: Most common exine sculpturing



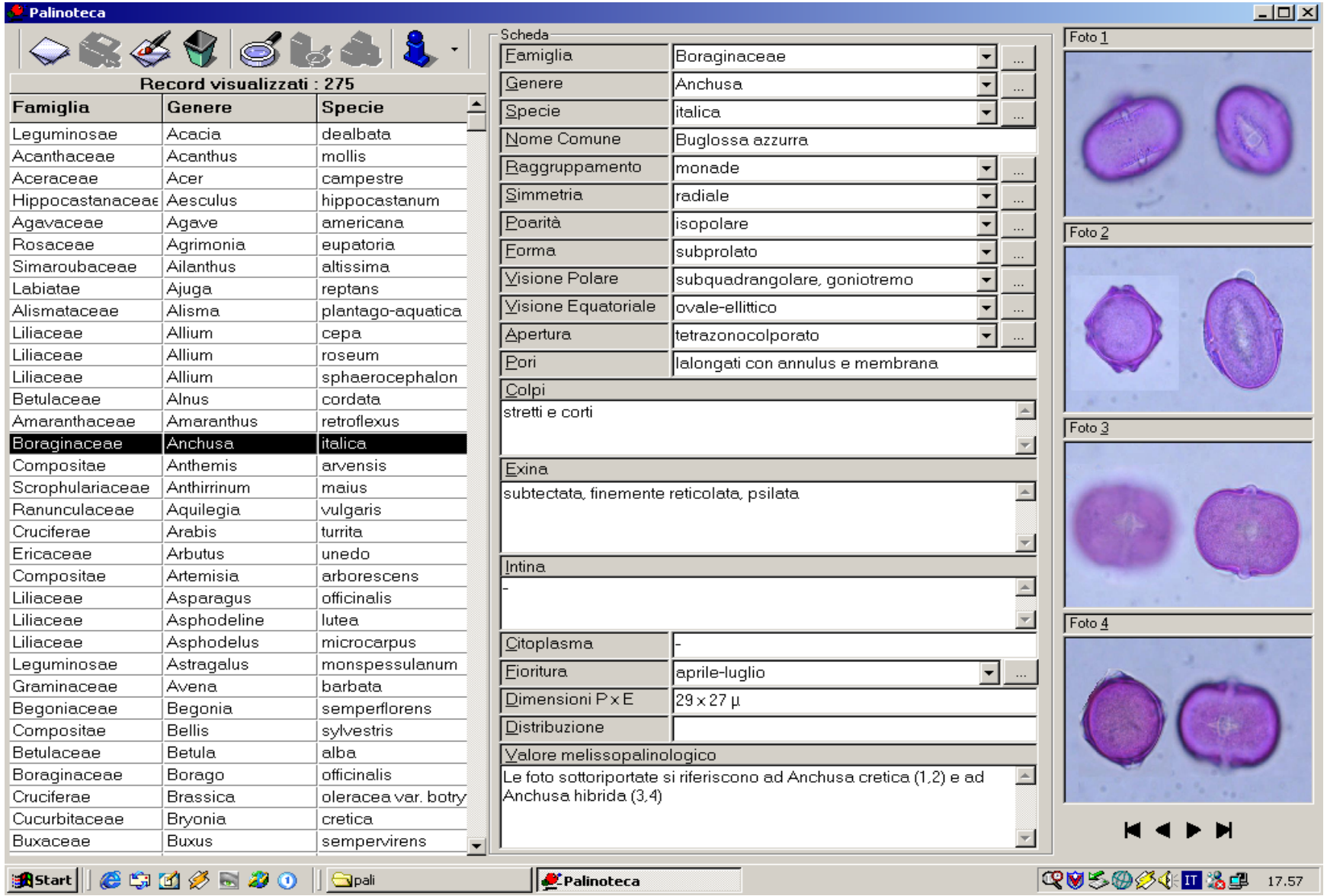
Pollen taxonomy: Traditional approach



Pollen atlas are in taxonomical or alphabetic order

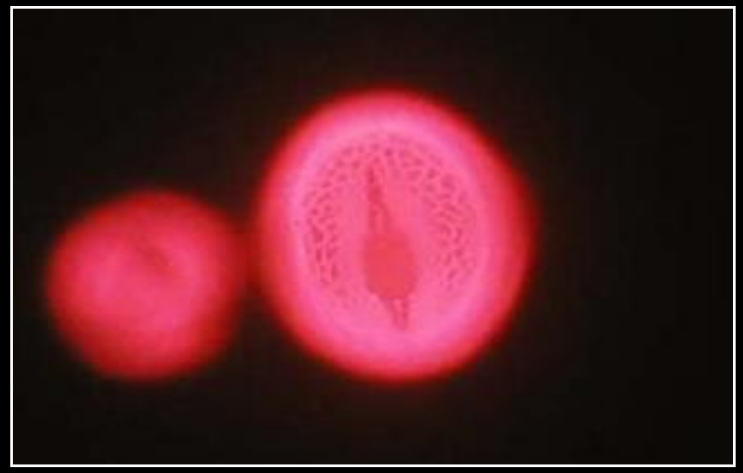
Pollen taxonomy: Current approach

Pollen database

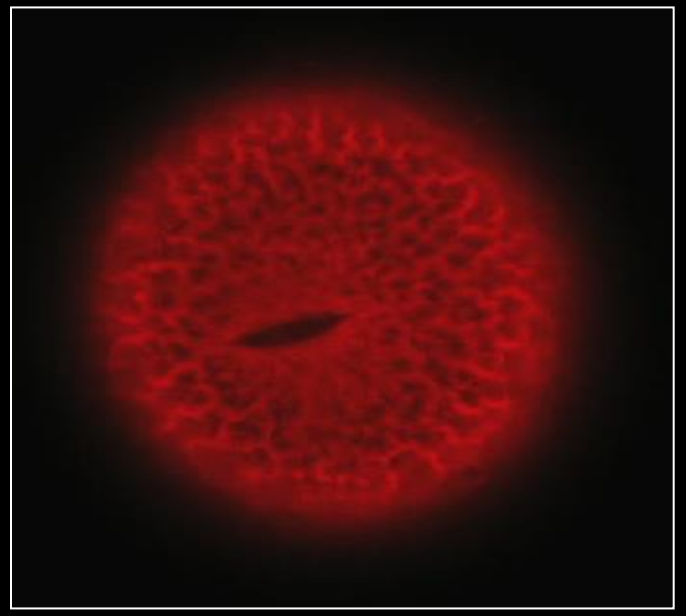


Pollen taxonomy: Future approach

Trifolium incarnatum L.



Chorisia sp.



- Image analysis
- 3D reconstruction
- Machine intelligence
- …

2. Pollen and Stigma Biology

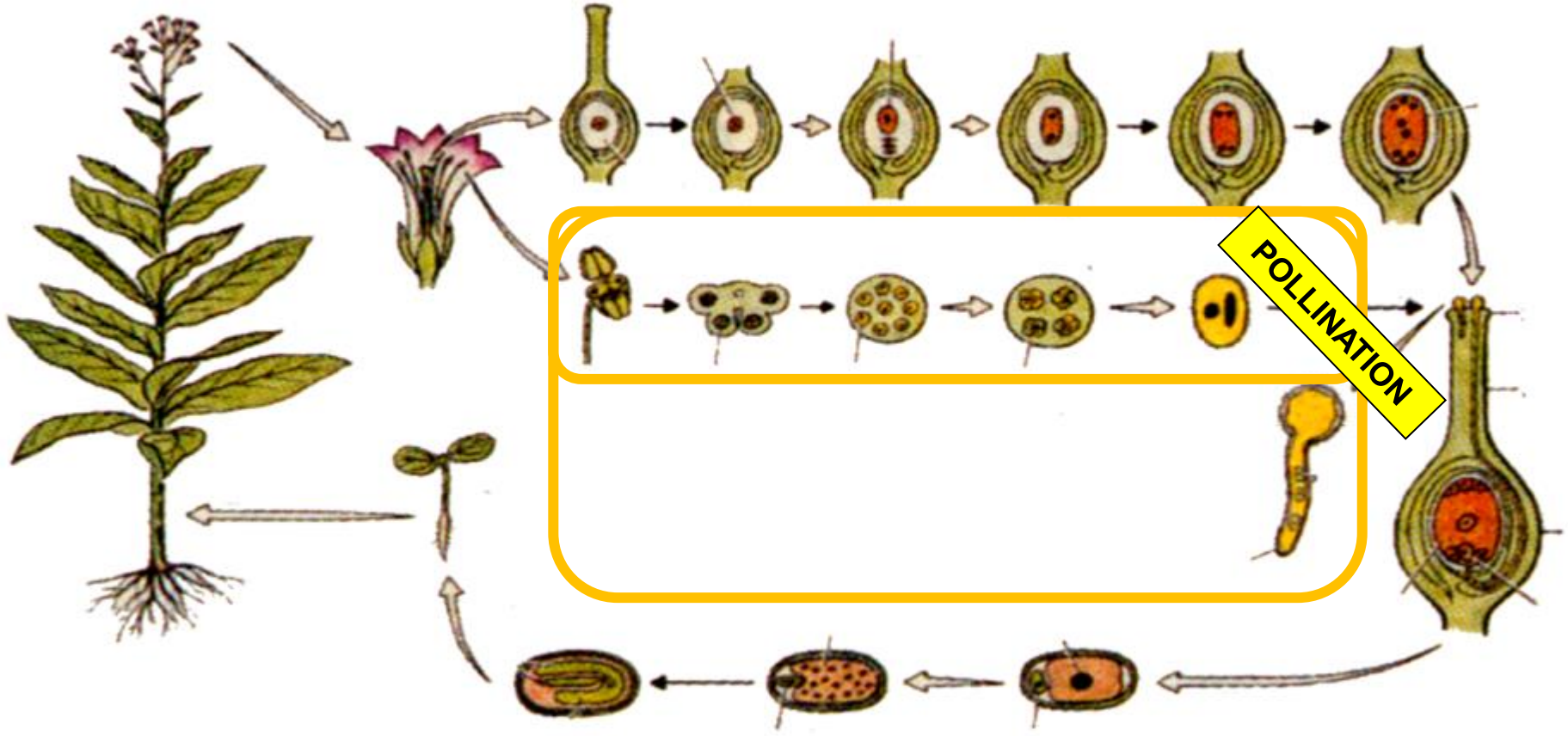
- Role of pollen for plant reproductive success and species conservation
- Pollen Development
- Pollen cytology

- Pollen morphology
- Pollen taxonomy

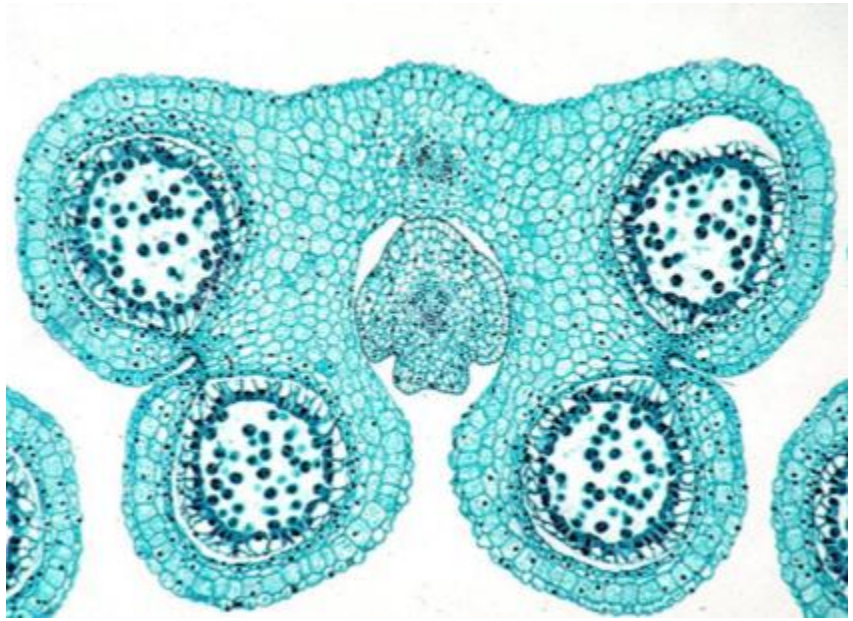
- Pollen functionality
- Methods to assess pollen viability and pollen germination

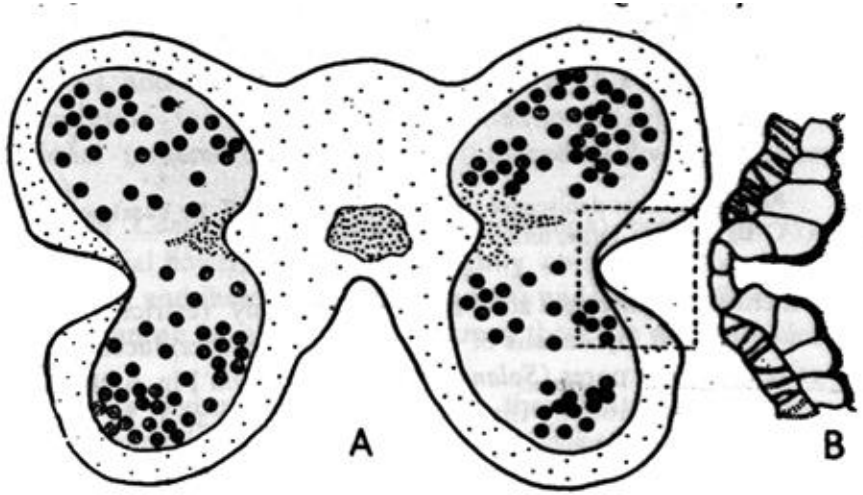
- Study cases on the effects of environmental factors on pollen functionality

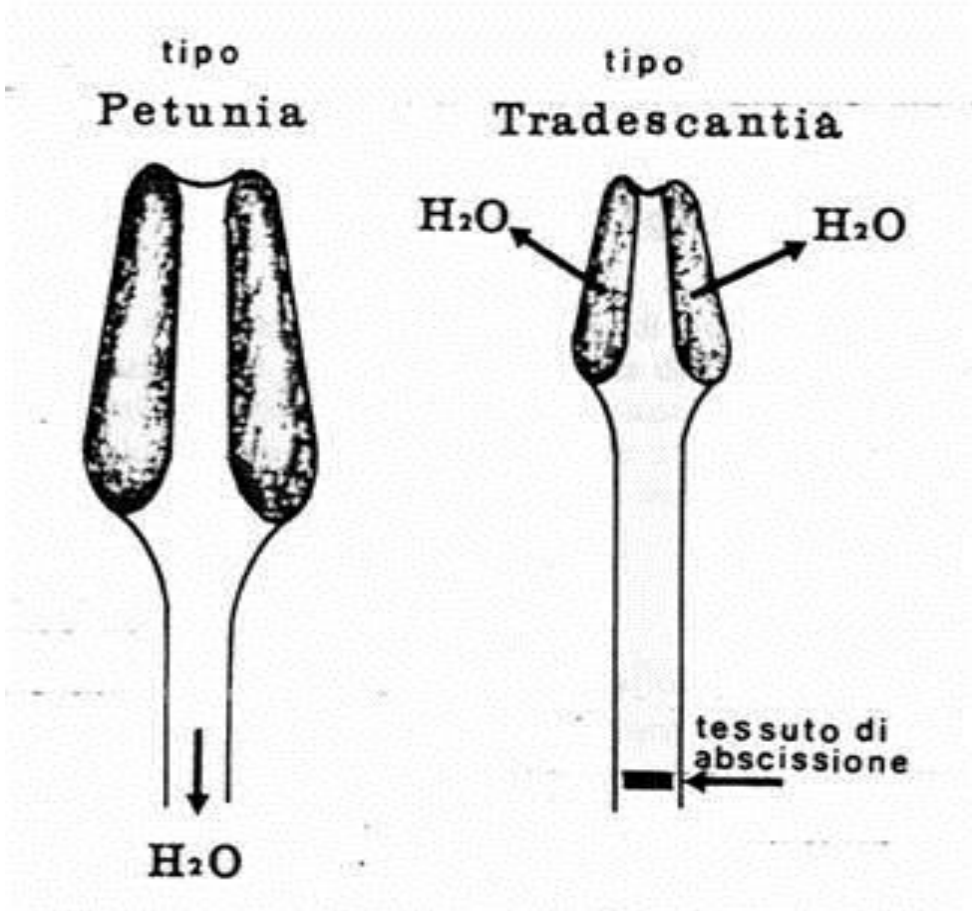
Angiosperm life cycle



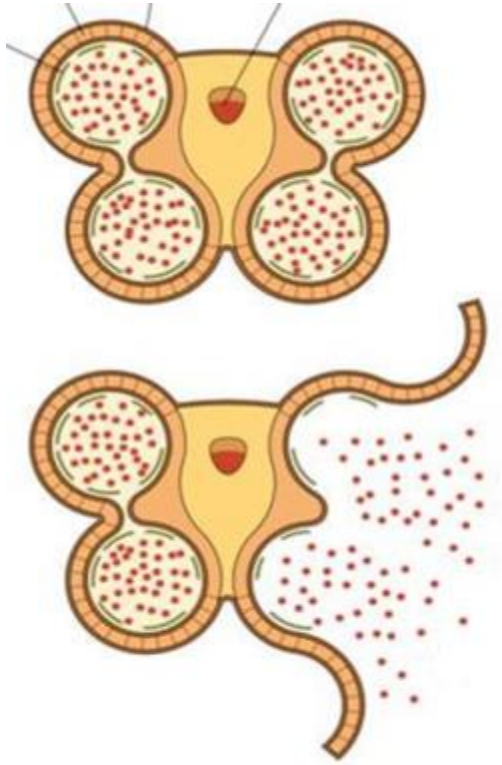
Endothecium dehydration and anther dehiscence



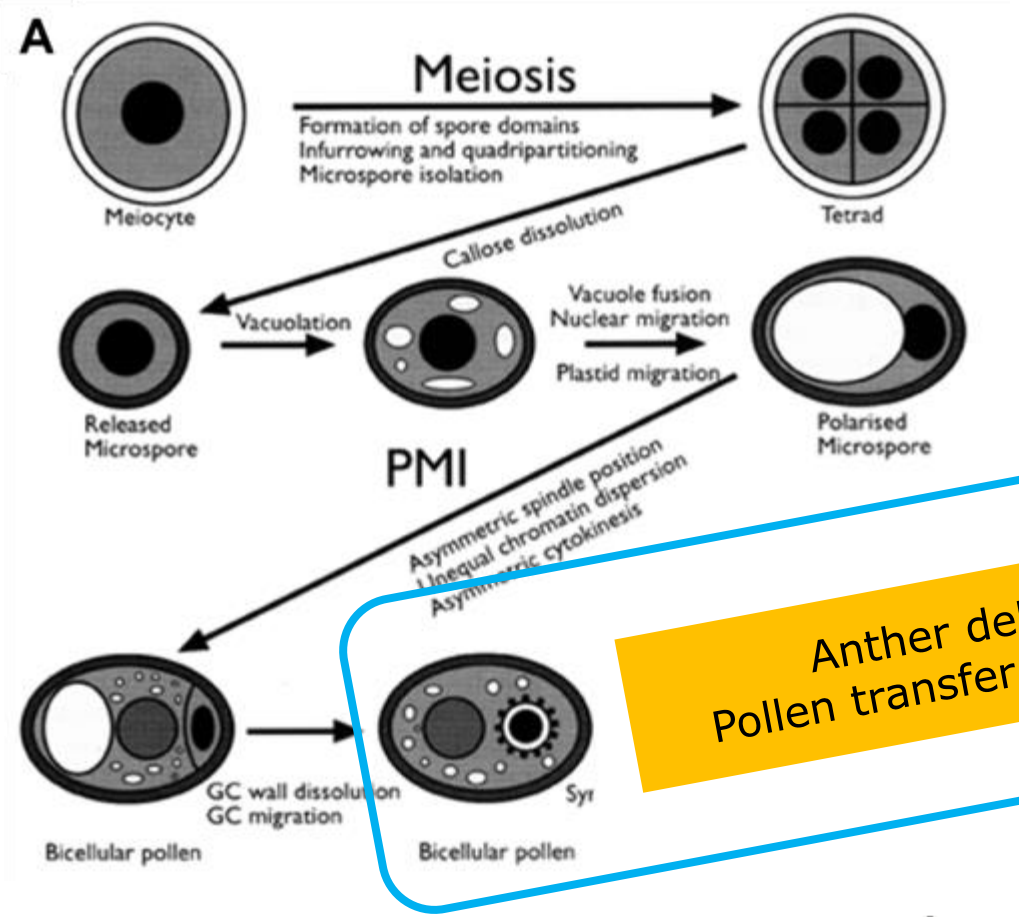




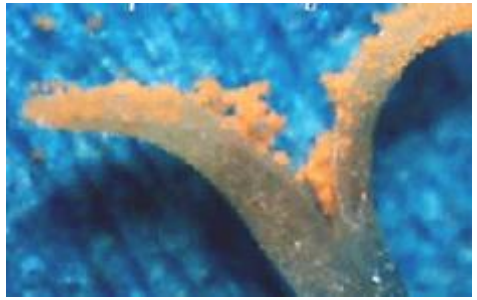
Anther dehiscence and pollen release

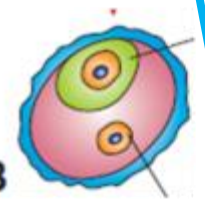


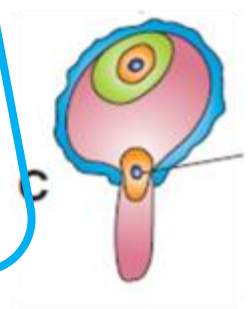
Pollen functionality

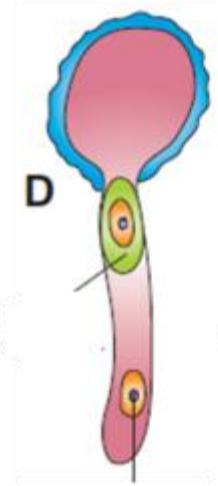


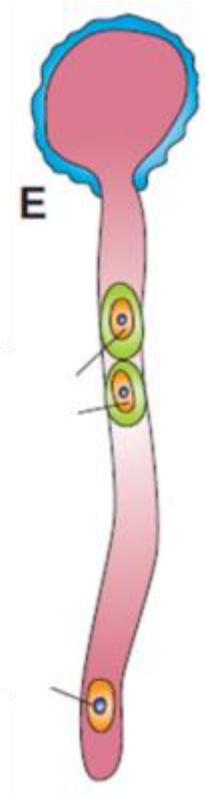
Anther dehiscence
Pollen transfer to the stigma











Time

Temperature

Humidity

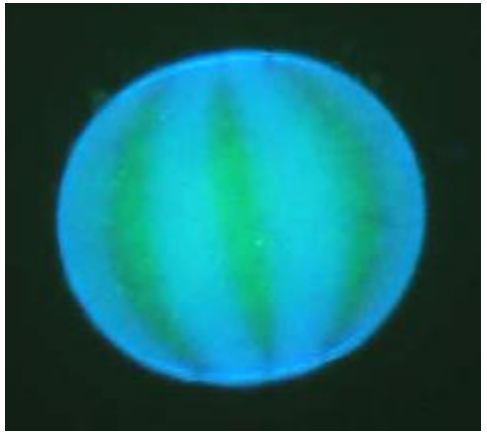
Other environmental factors

Estimation of pollen functionality

Temperature

Humidity

Time



Pollen viability:
Tests to estimate the number of living pollen grains

Pollen germinability:
Tests to estimate the number of pollen grains able to develop the pollen tube

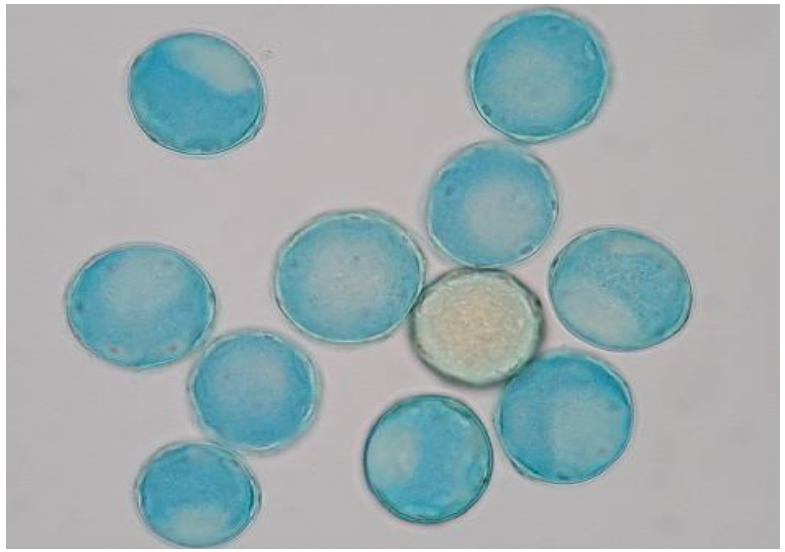
Pollen viability

Viable Pollen

Dead Pollen

Aborted Pollen

Staining tests for pollen cellular content

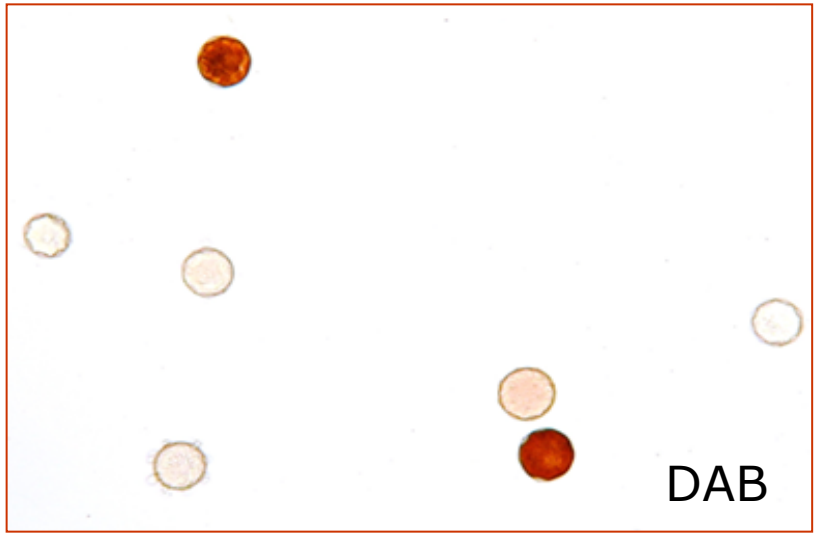


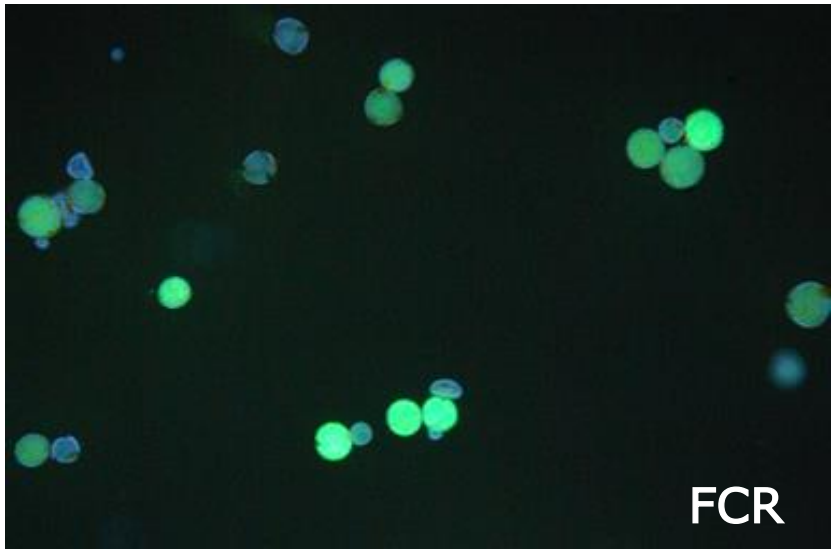
- Lactophenol-cotton blue
- Acetocarmine
- Neutral red
- …
- Alexander’s procedure

Useful to estimate pollen abortion

Pollen viability

Enzimatic tests: useful to distinguish viable from dead pollen grains

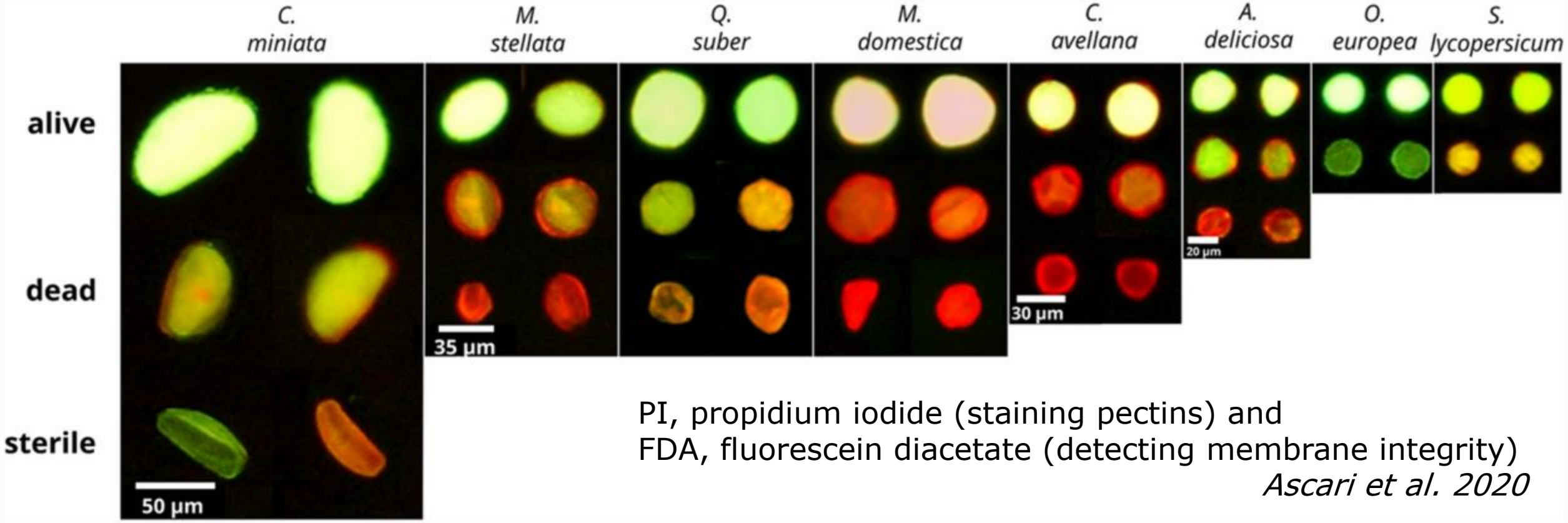




Compound	Metabolic reaction	Living process
Triphenyl Tetrazolium Chloride (TTC)	It is reduced by NADPH to its insoluble purple form	Respiration
Fluorescein Diacetate (FDA)	It is transformed in the polar fluorescent compound that is accumulated in the cell	Membrane Integrity
Diaminobenzidine (DAB)	Become brown after peroxidase reaction	Peroxidase activity

Pollen viability

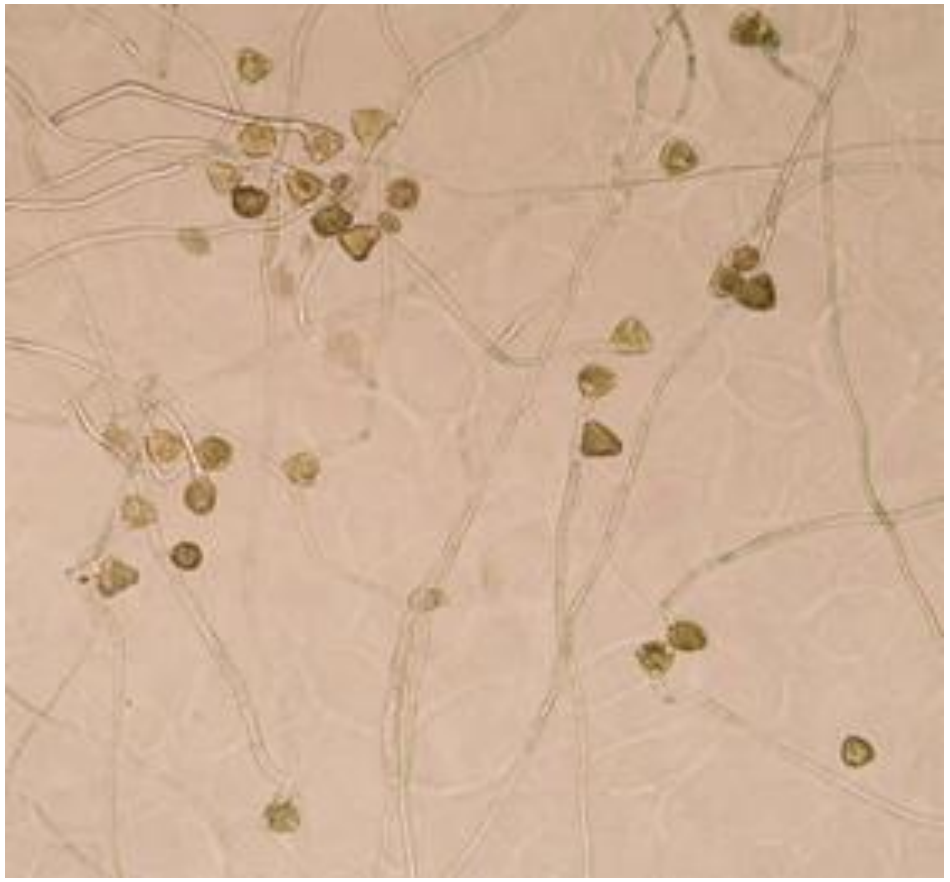
Multiple-reaction tests



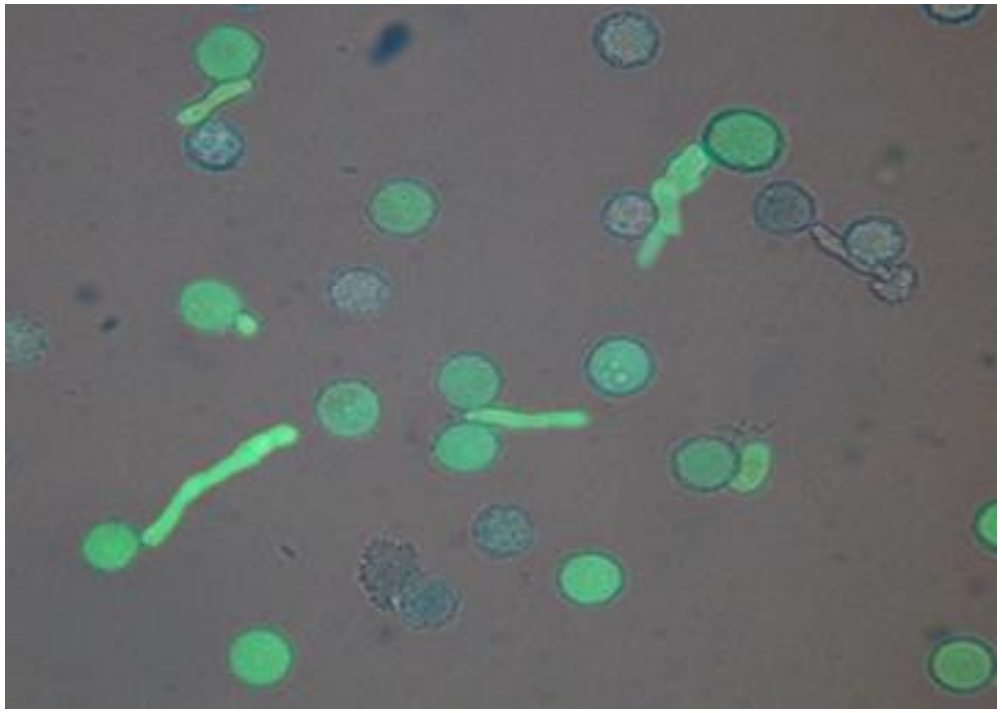
PI, propidium iodide (staining pectins) and
FDA, fluorescein diacetate (detecting membrane integrity)

Ascari et al. 2020

Pollen germinability



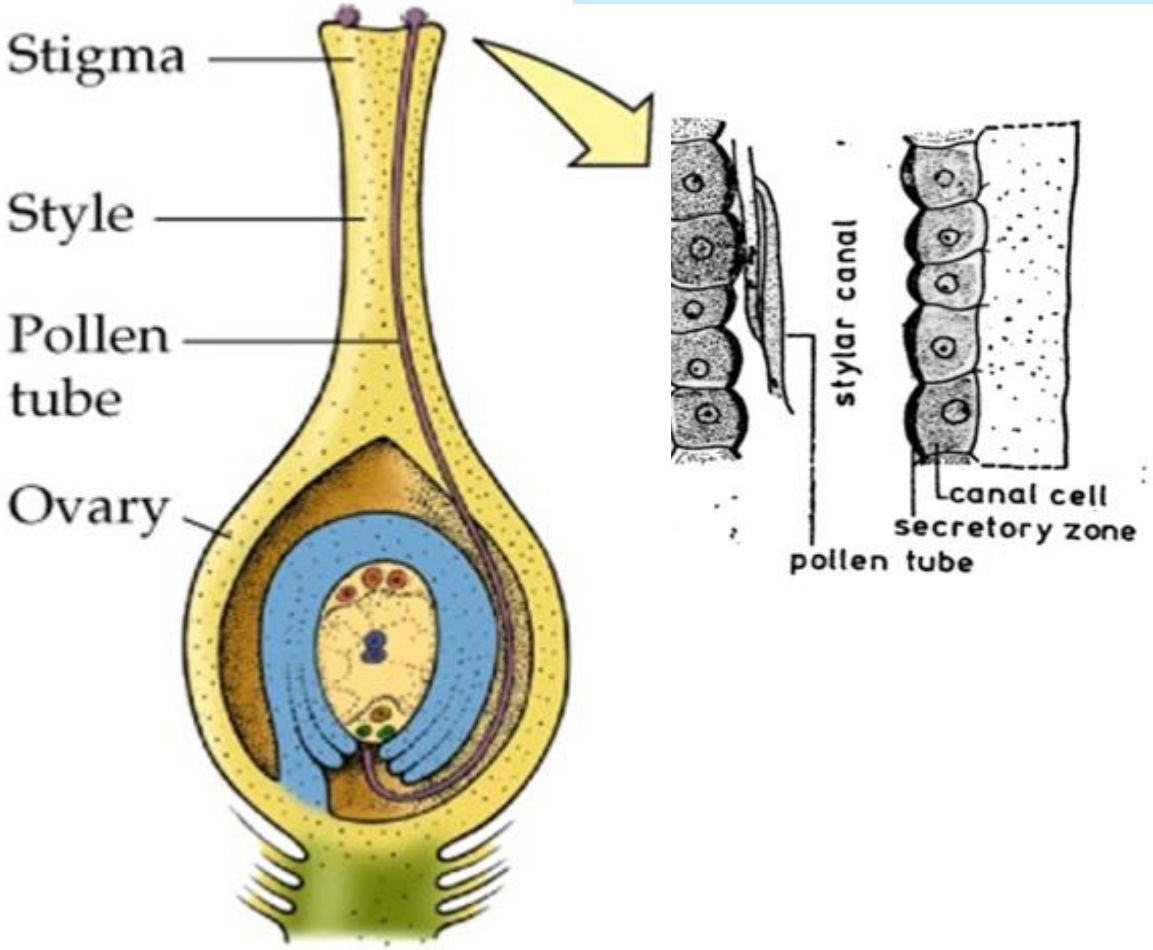
"in vitro" germination tests

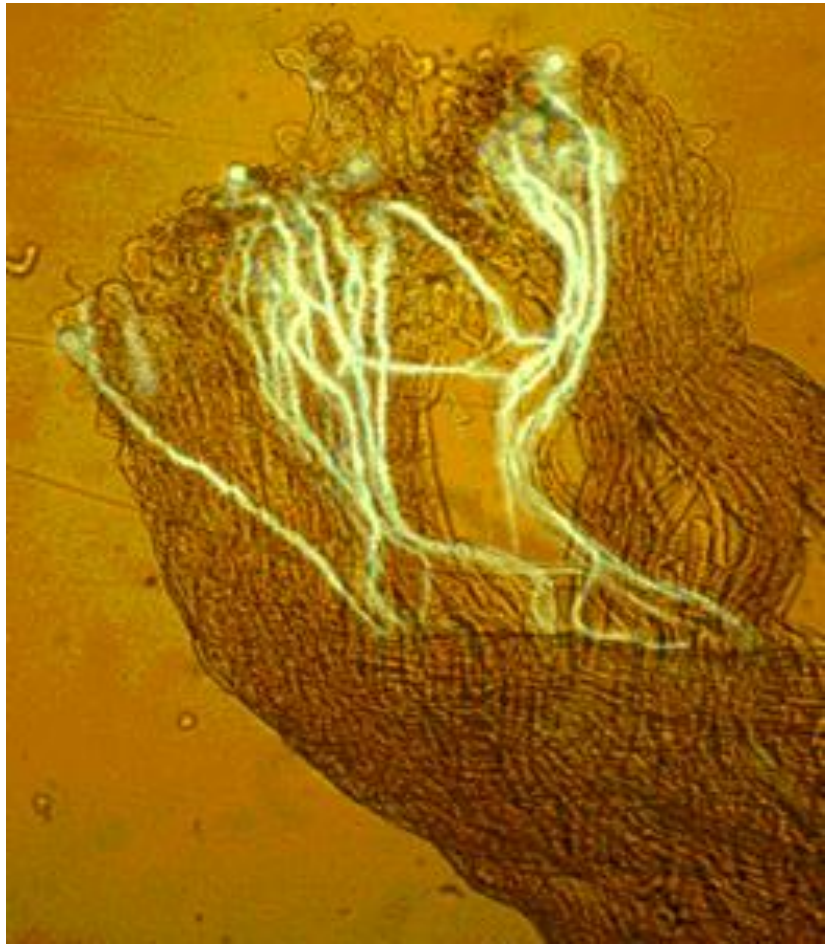


- Identification of the culture medium composition
- Selection of the best method (hanging drop, liquid medium in vials, agar medium in petri dishes …)

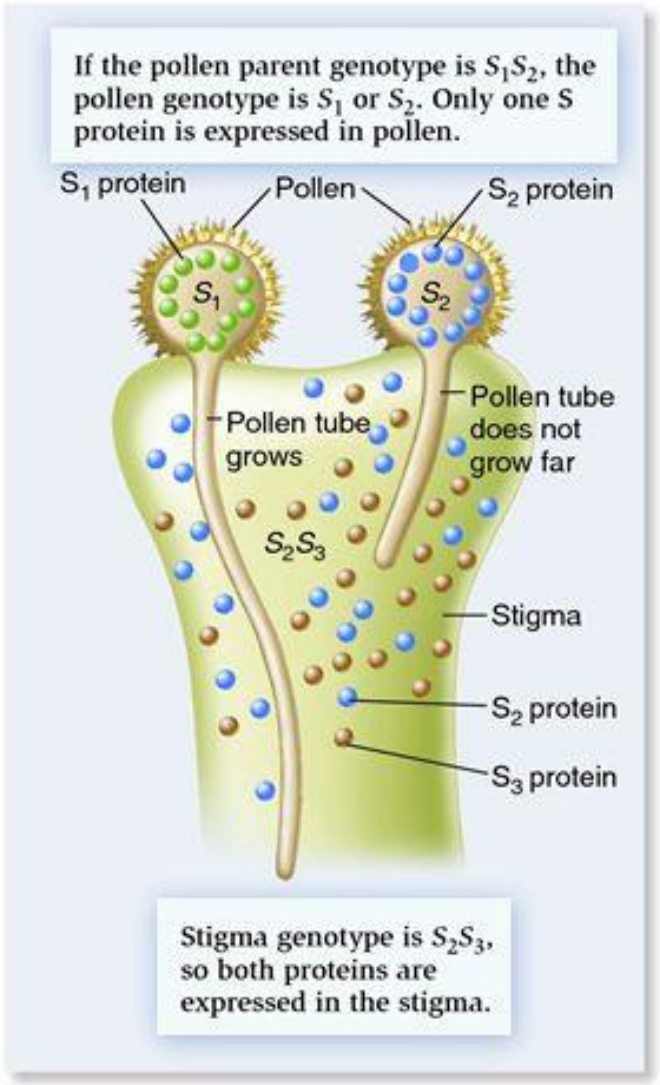
Pollen germinability

"in vivo" germination tests

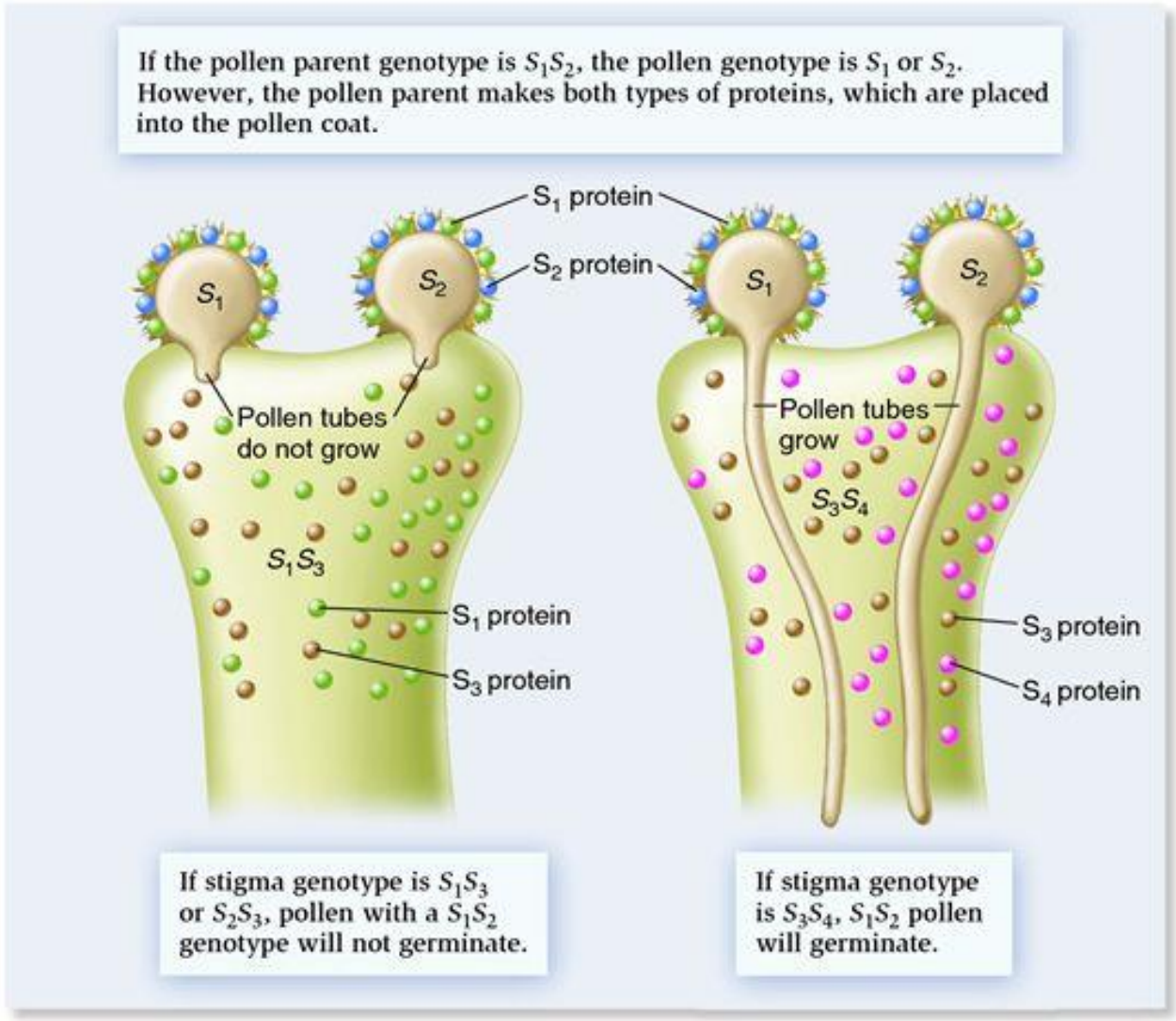




Pollen/Stigma Interaction (Self-Incompatibility SI)



(a) Gametophytic SI: If pollen *S* allele does not match either stigma allele, pollen will germinate.



(b) Sporophytic SI: If pollen coat S proteins do not match either stigma S protein, pollen tubes will grow.

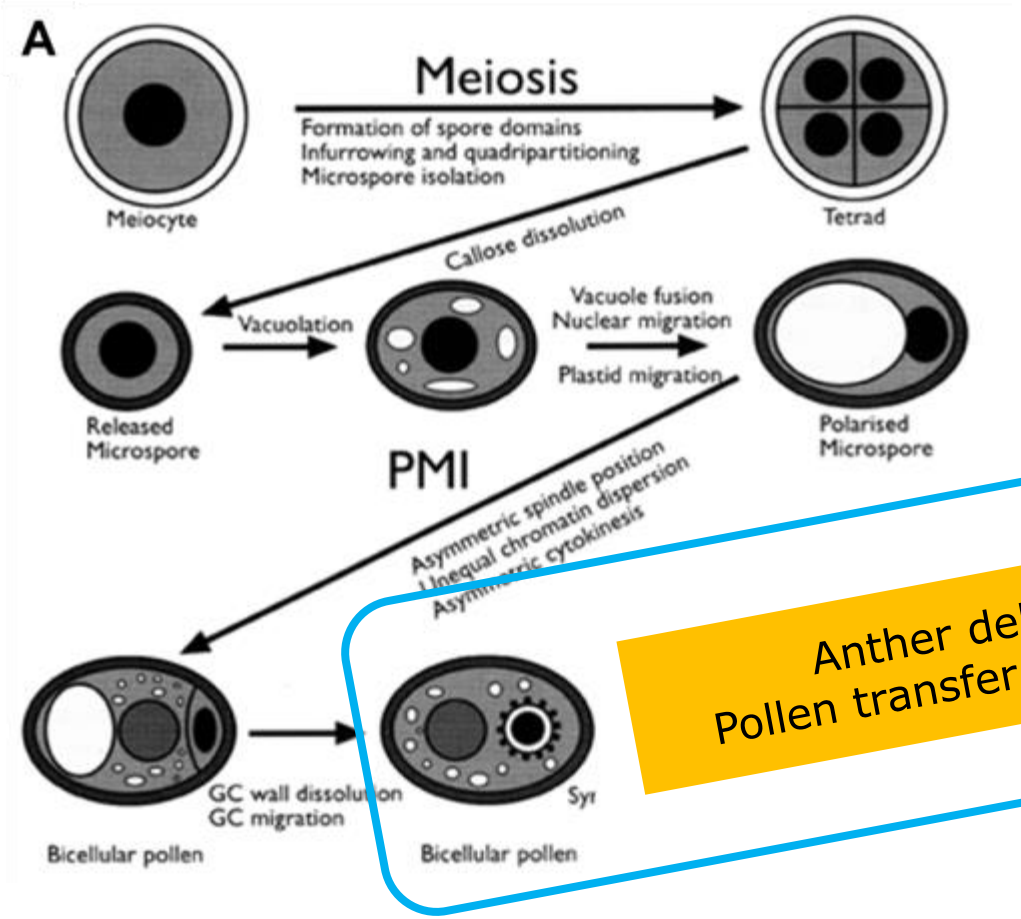
Receptivity of compatible stigmas

The ability of the stigma to support germination of viable, compatible pollen

Receptive stigmas show the activity of several enzymes including esterases, peroxidases, and acid phosphatases (*Dafni, 1992; Dafni and Maués, 1998*).

The presence of these enzymes not necessarily reflect receptivity (*Shivanna and Sastri, 1981*).

Pollen functionality



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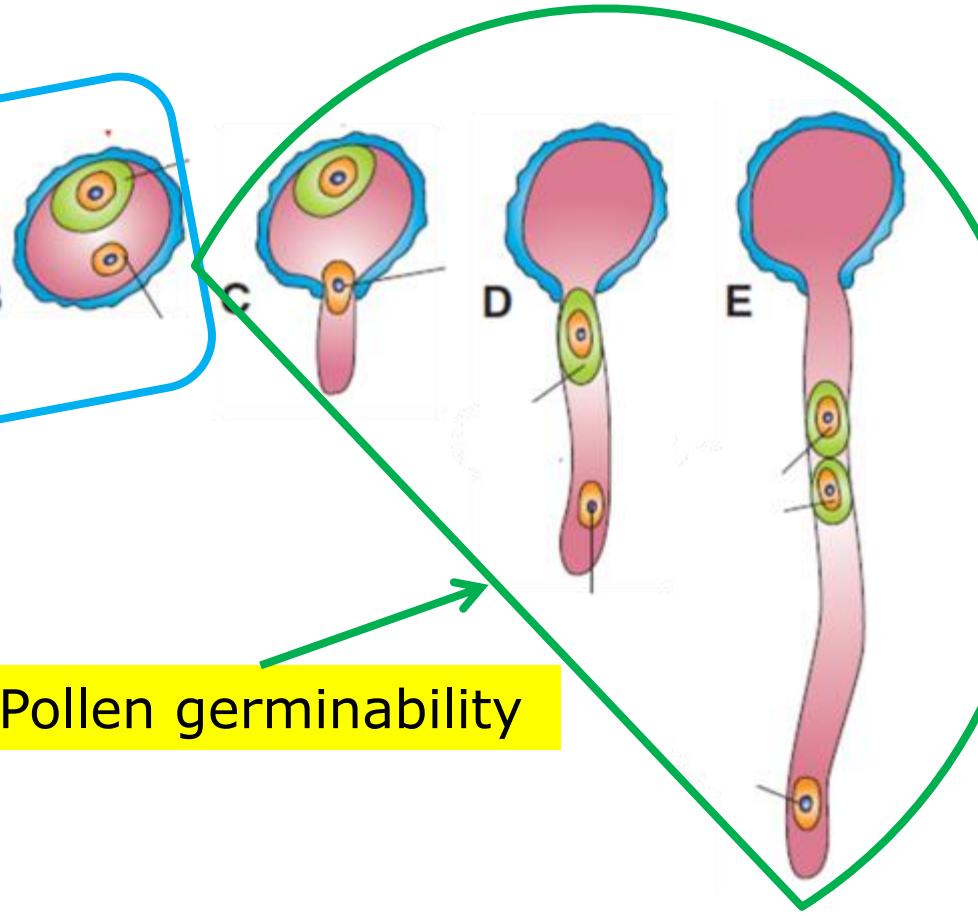
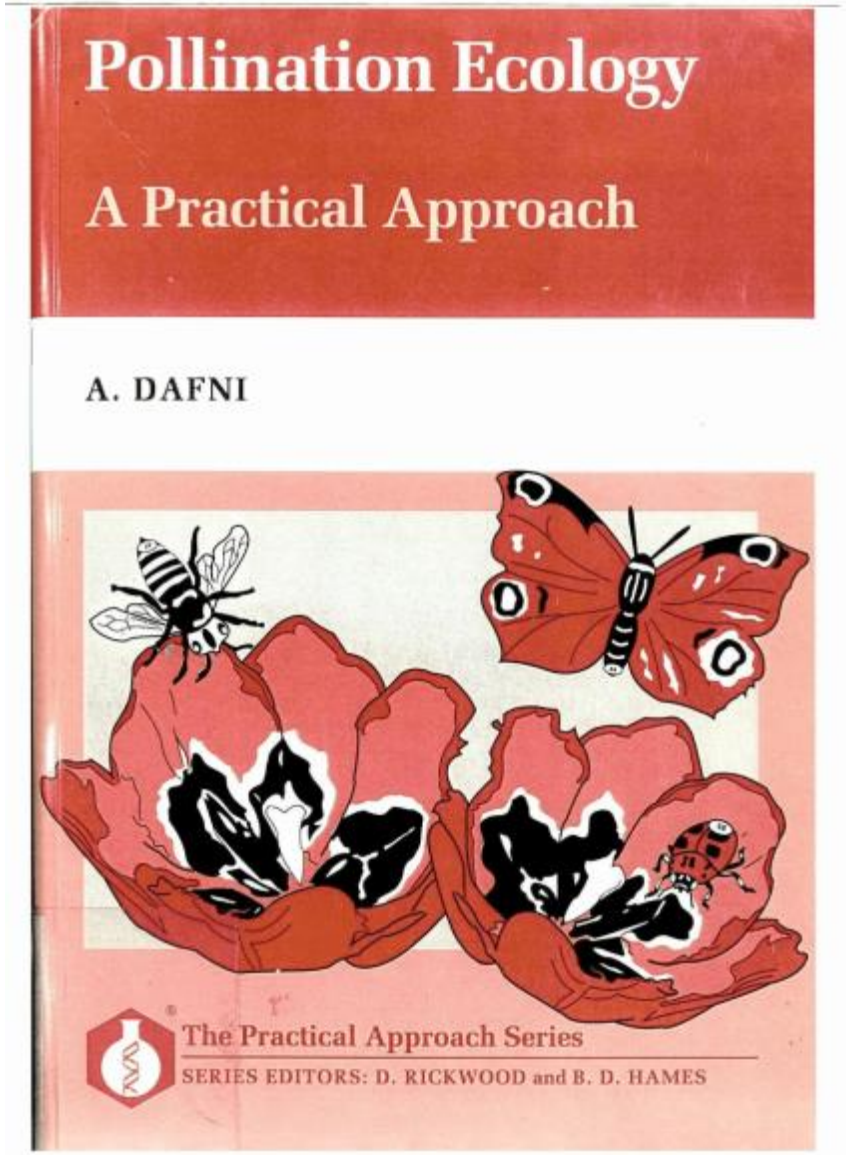
Pollen transfer to the stigma

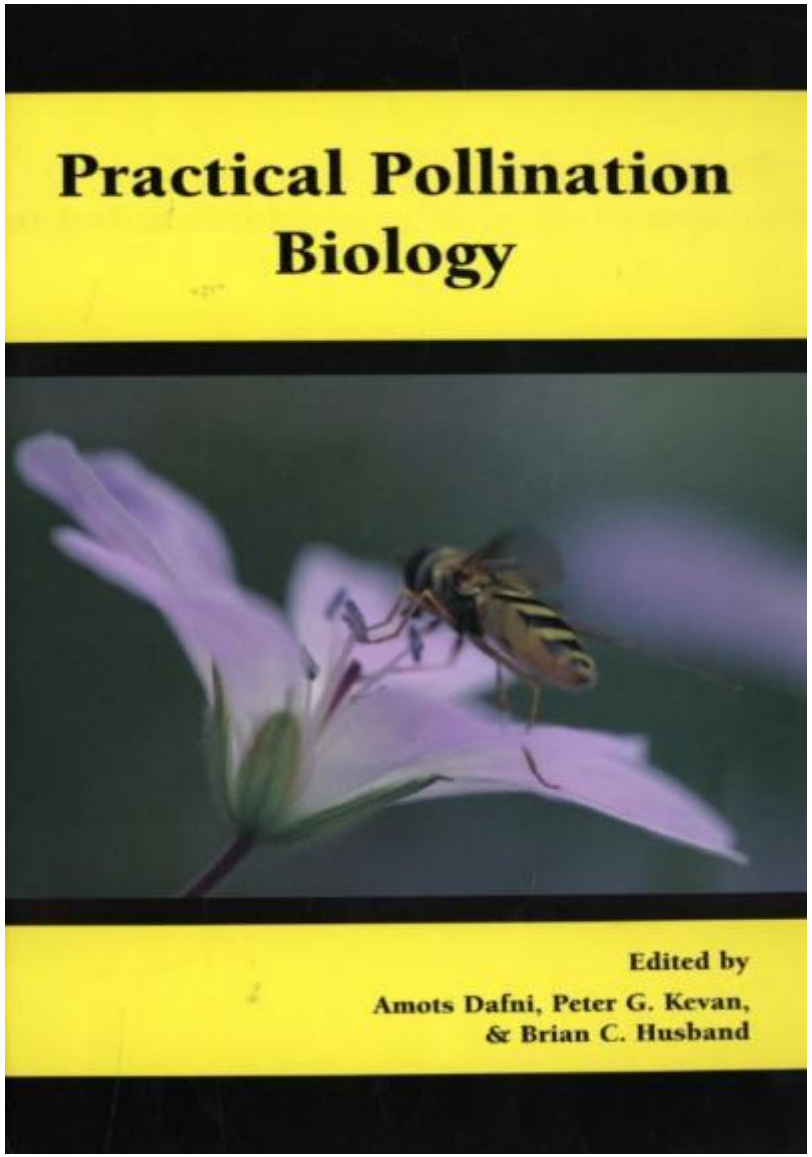


Pollen viability

Pollen germinability

Handbooks for pollen biology/ecology research





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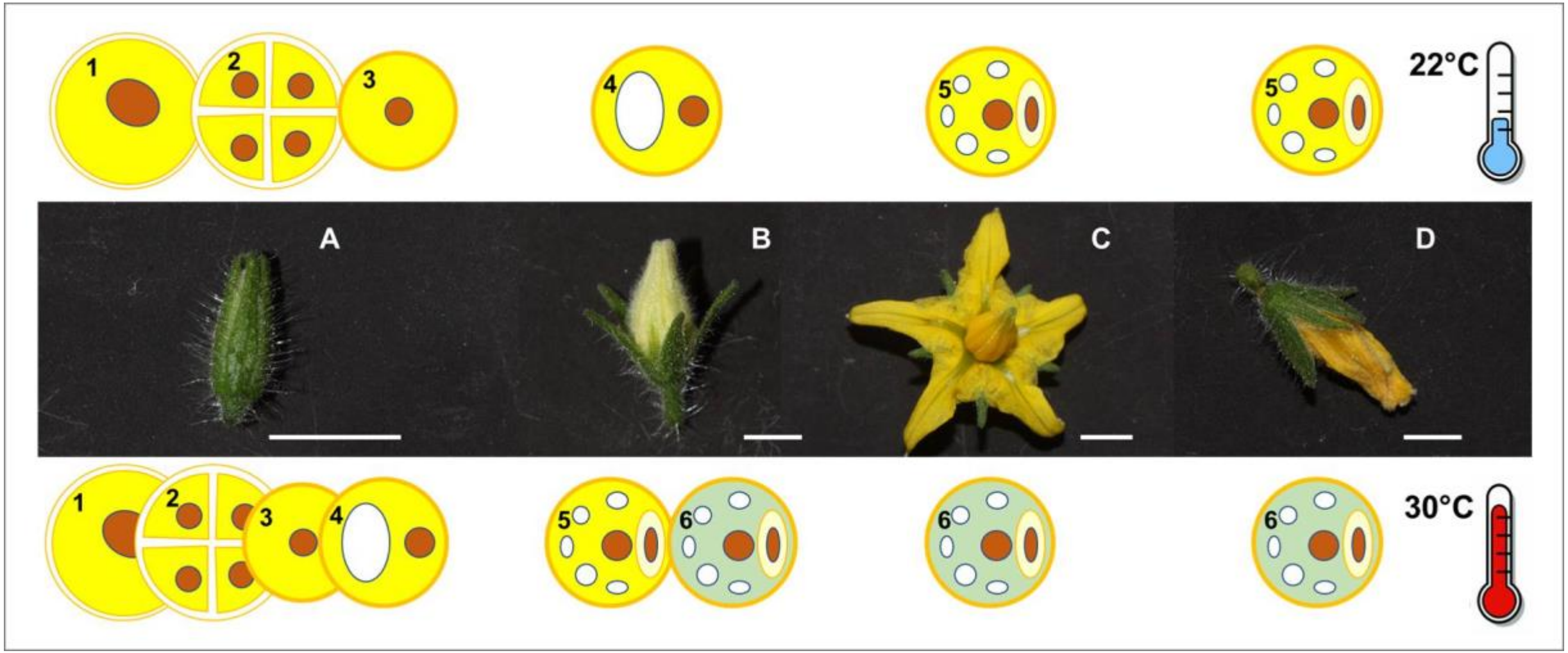
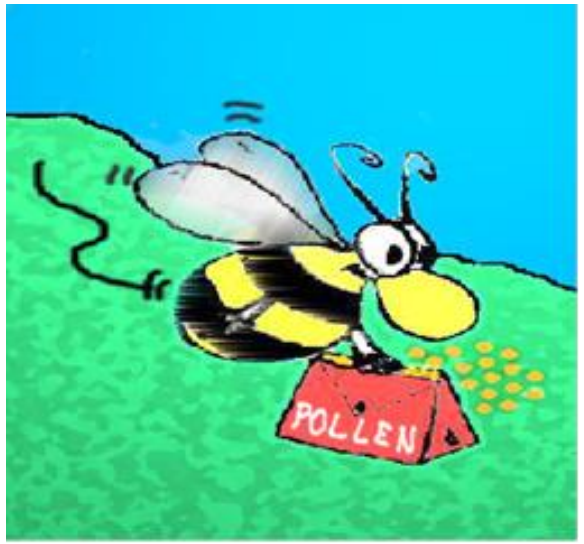
The study case of *Solanum lycopersicum*

Effect of Temperature on pollen viability



The study case of *Fragaria vesca*

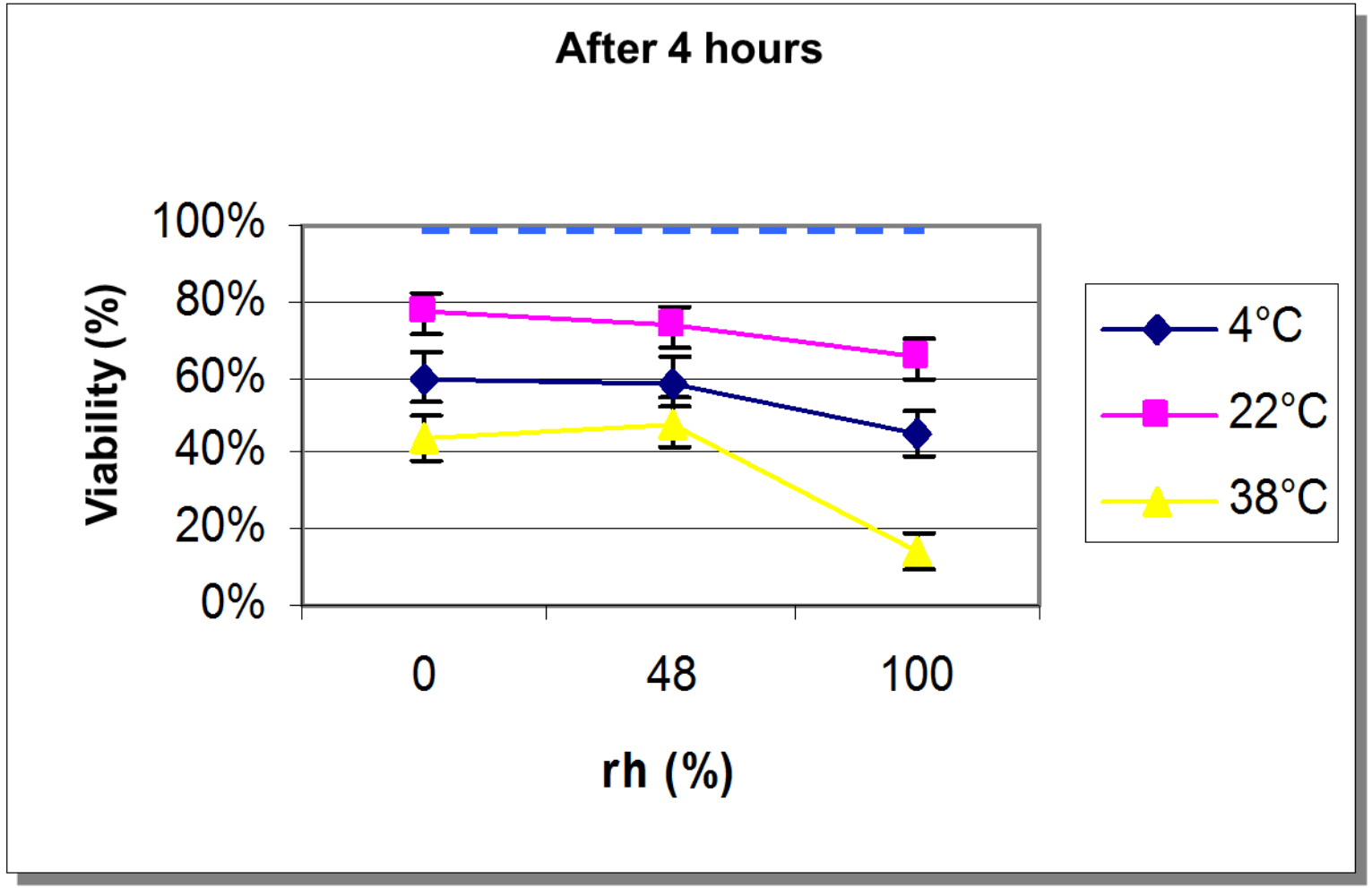


Efficient cross-pollination is not enough for a successful fruit production

Pollen transferred on to the stigma must be viable

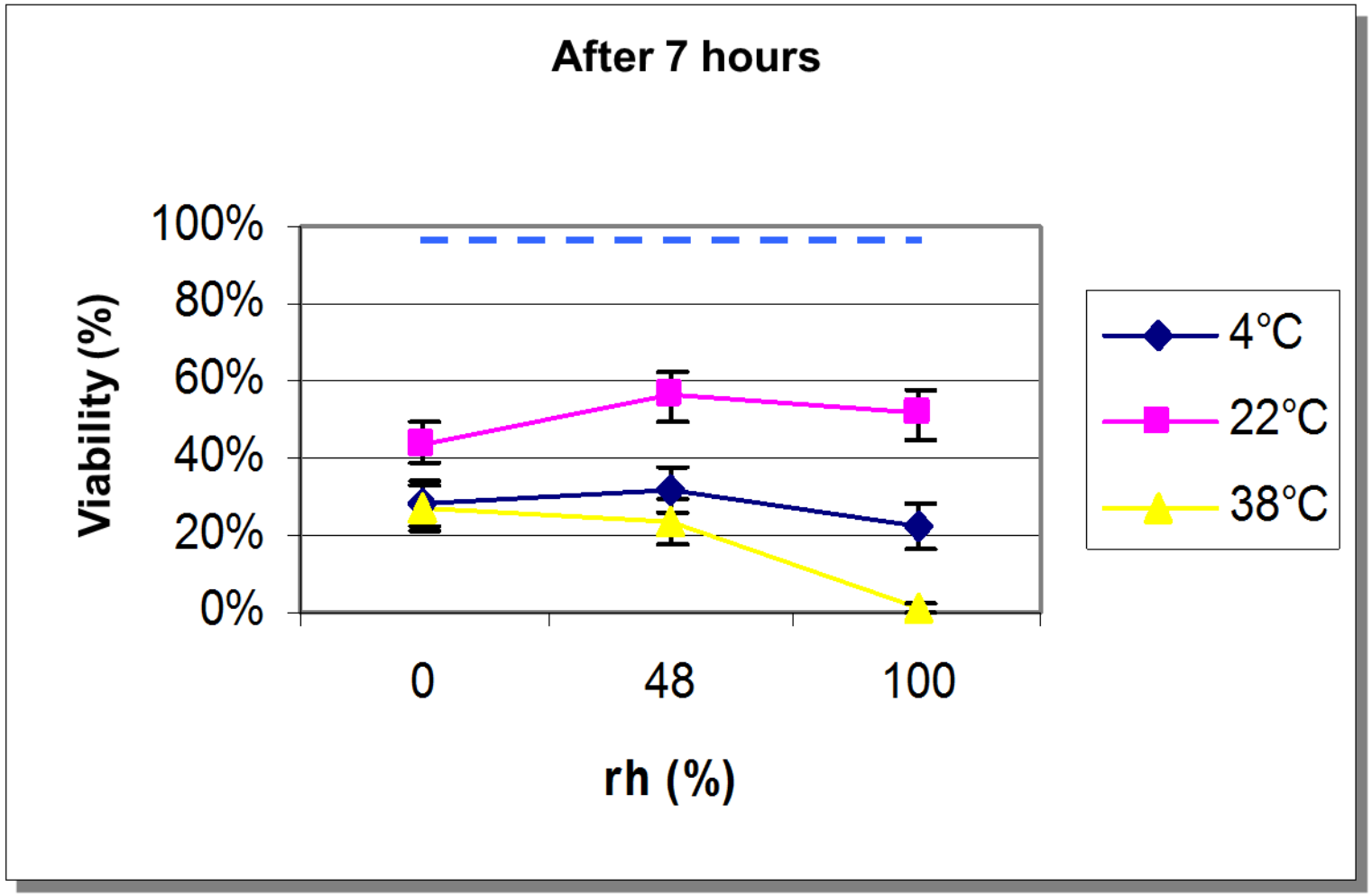
The study case of *Fragaria vesca*

Effect of Temperature and Humidity on pollen viability



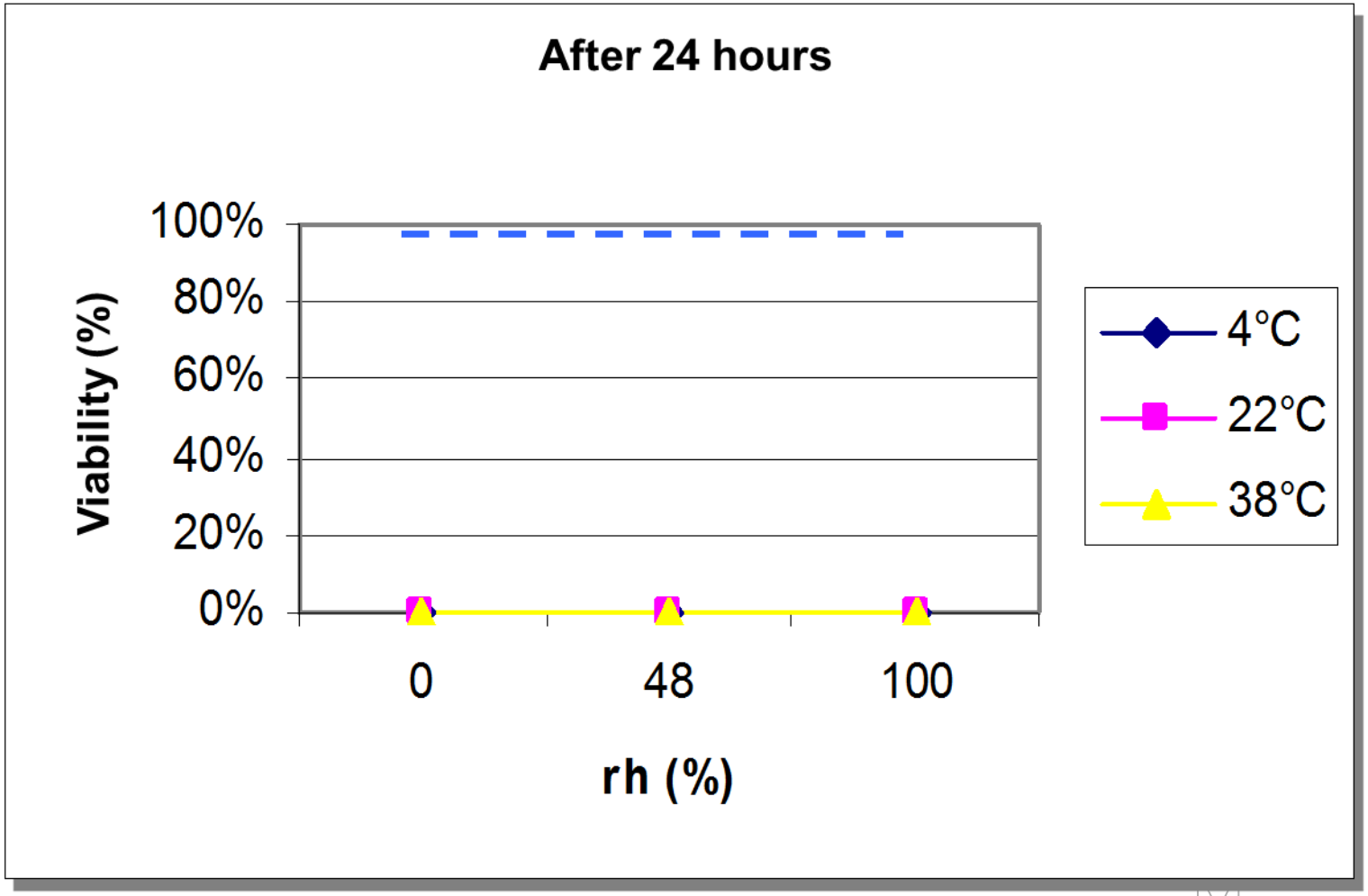
The study case of *Fragaria vesca*

Effect of Temperature and Humidity on pollen viability



The study case of *Fragaria vesca*

Effect of Temperature and Humidity on pollen viability



The study case of *Primula palinuri*

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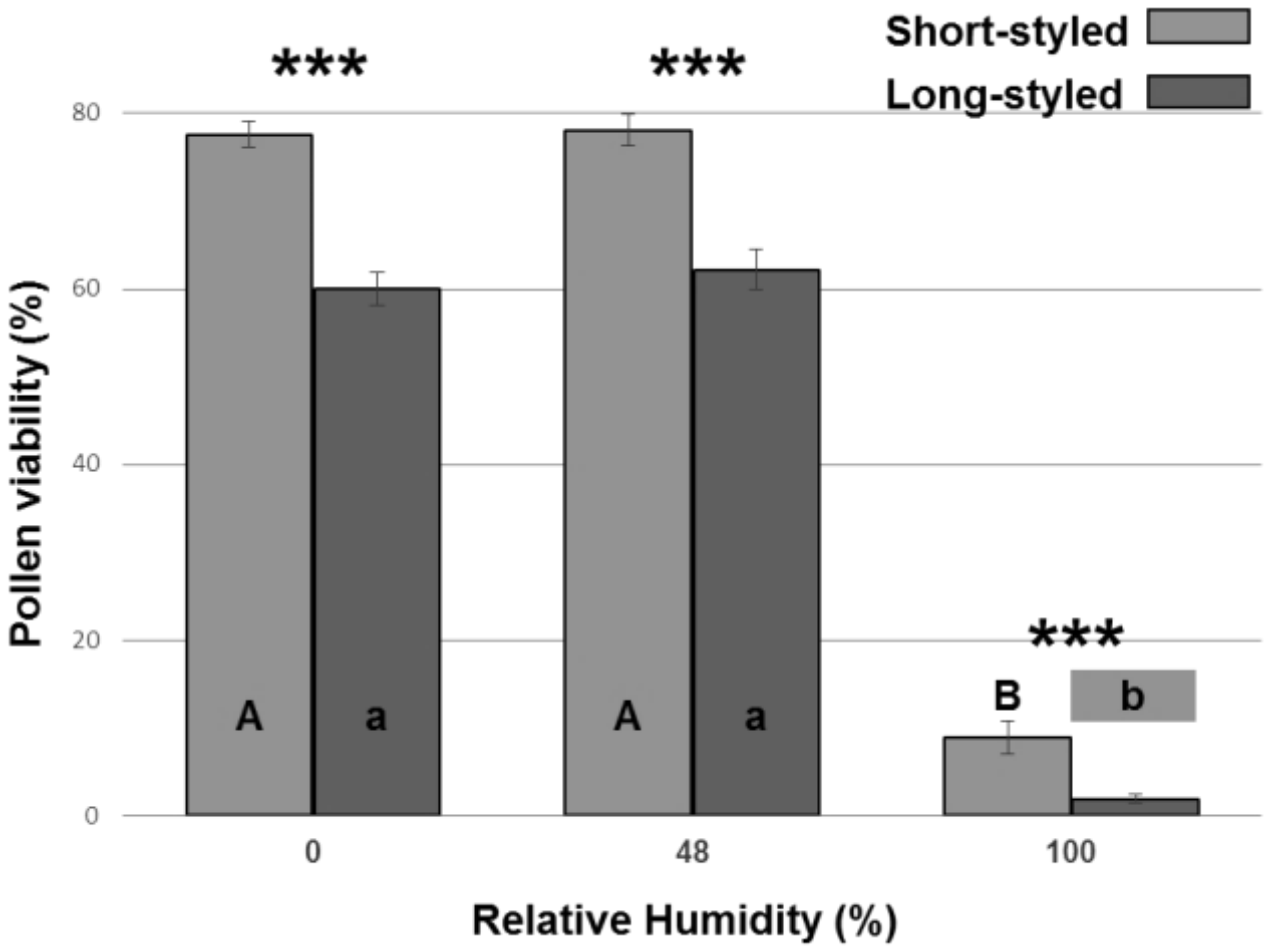
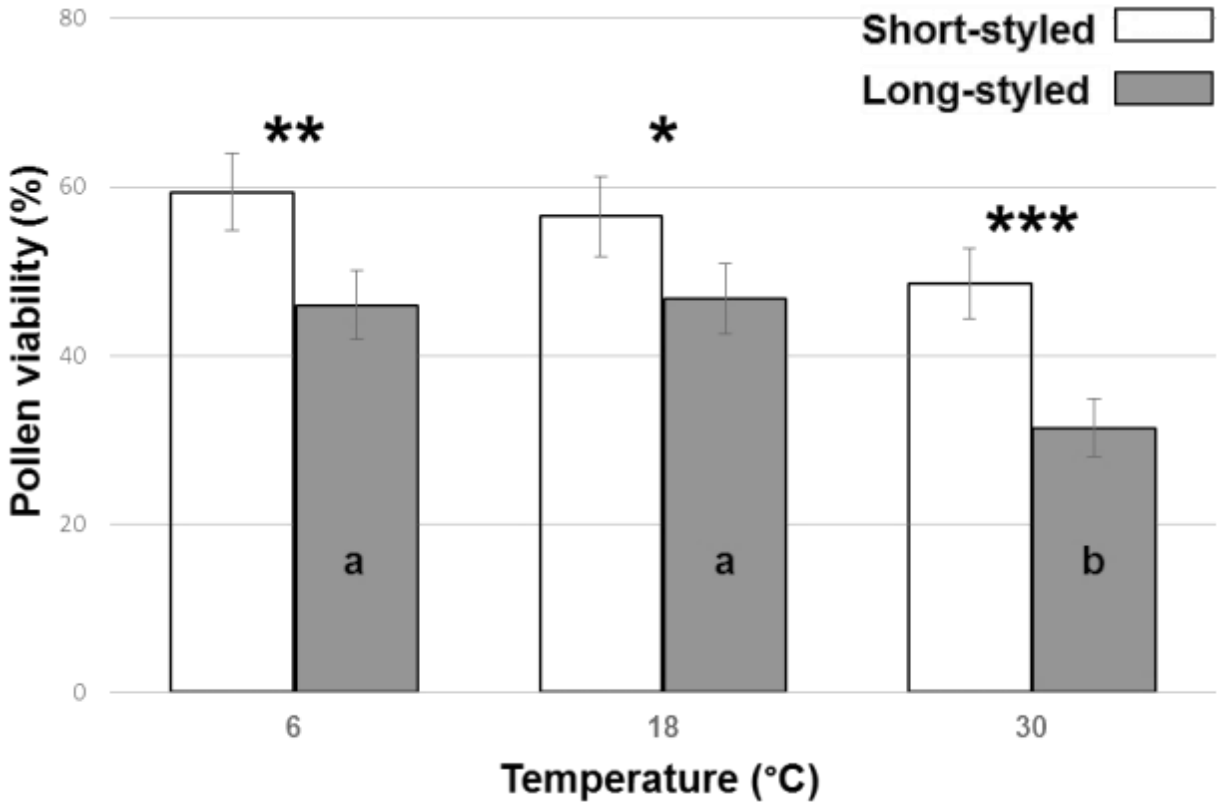


Aronne et al 2021 Annali di Botanica DOI: https://doi.org/10.13133/2239-3129/17157

The study case of *Primula palinuri*



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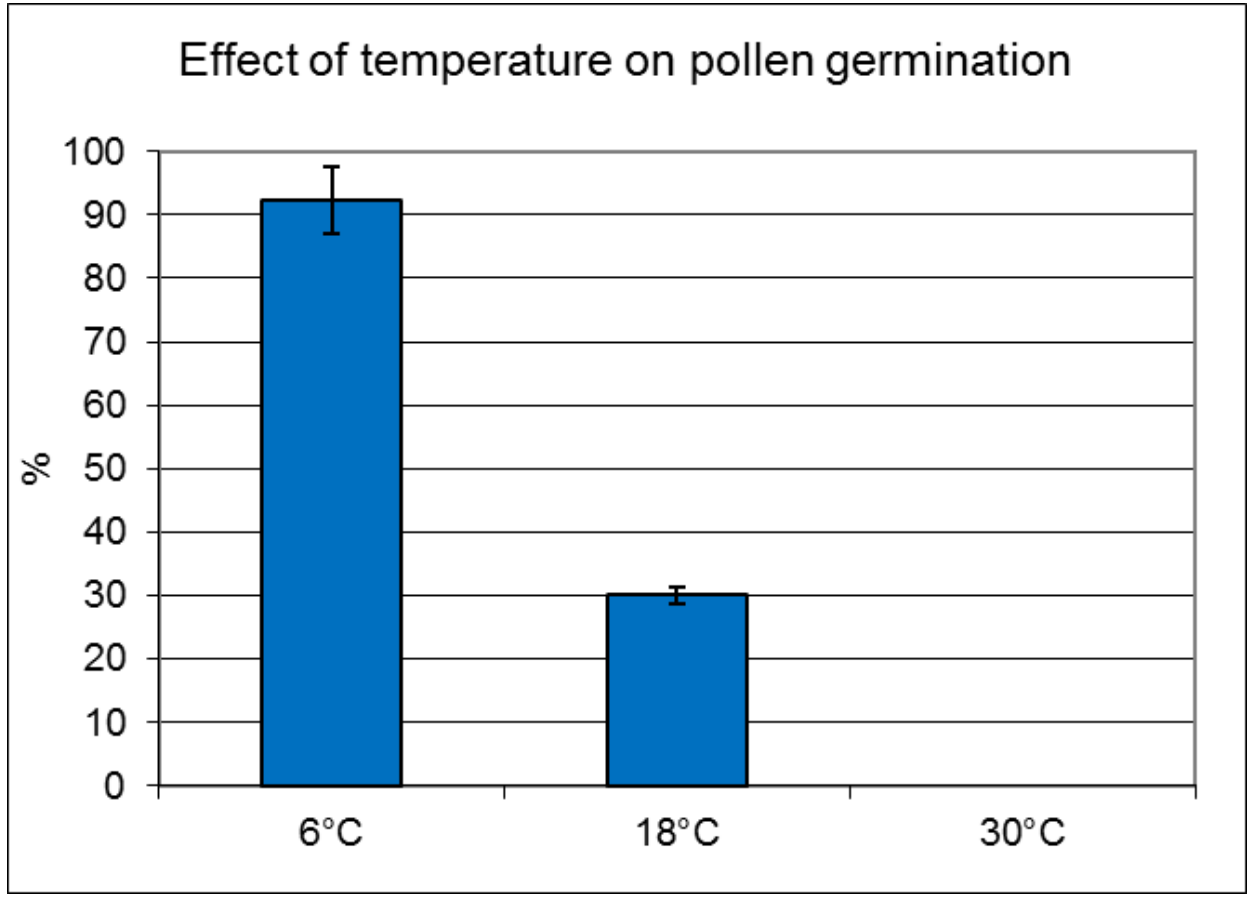

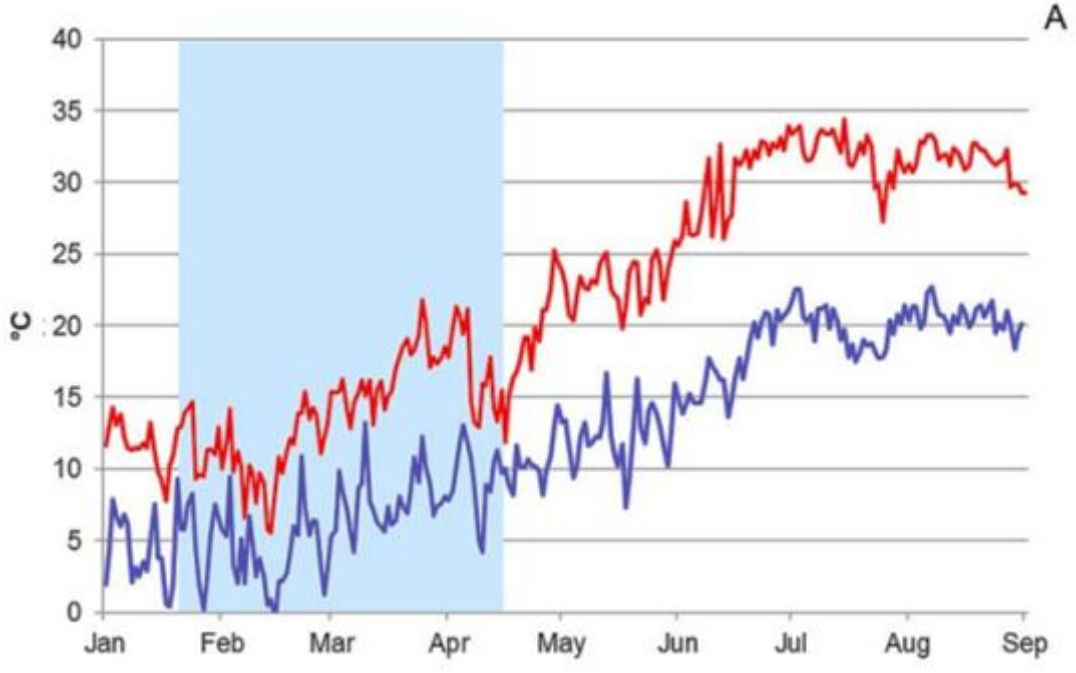

Aronne et al 2014 Plant Biology 17(2), 535-544

Plant reproductive success and species conservation