



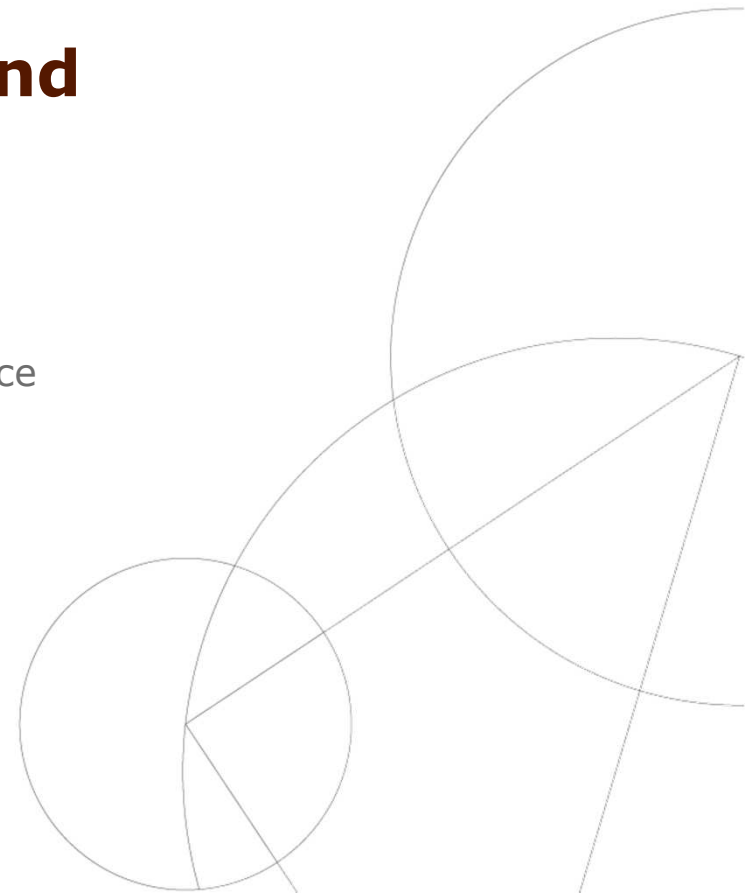
Department of Plant and Environmental Sciences



Wide hybridization of *Kalanchoë* and *Hibiscus* species: pre-fertilization barriers and optimization of pollination

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Kalanchoë and *Hibiscus* plants

Kalanchoë

- family Crassulaceae
- 125 species, mainly succulents
- Africa (Madagascar)
- the most common cultivars → from *K. blossfeldiana*
- most sold potted plant in Denmark
- dwarfism, flower colour and flower morphology

Hibiscus

- family Malvaceae
- ~ 300 species
- small trees, shrubs, herbs
- mainly tropical and subtropical regions
- the most important: *H. rosa-sinensis*
- outdoor and indoor use



Crossbreeding and Wide hybridization

Crossbreeding - breeding method by mean of sexual reproduction, where plants with different characteristics are mating in order to obtain superior progeny

Wide hybridization - breeding method by mean of sexual reproduction, which uses plants belonging to different species

Aim: to merge distant gene pools and increase genetic variability of cultivated plants

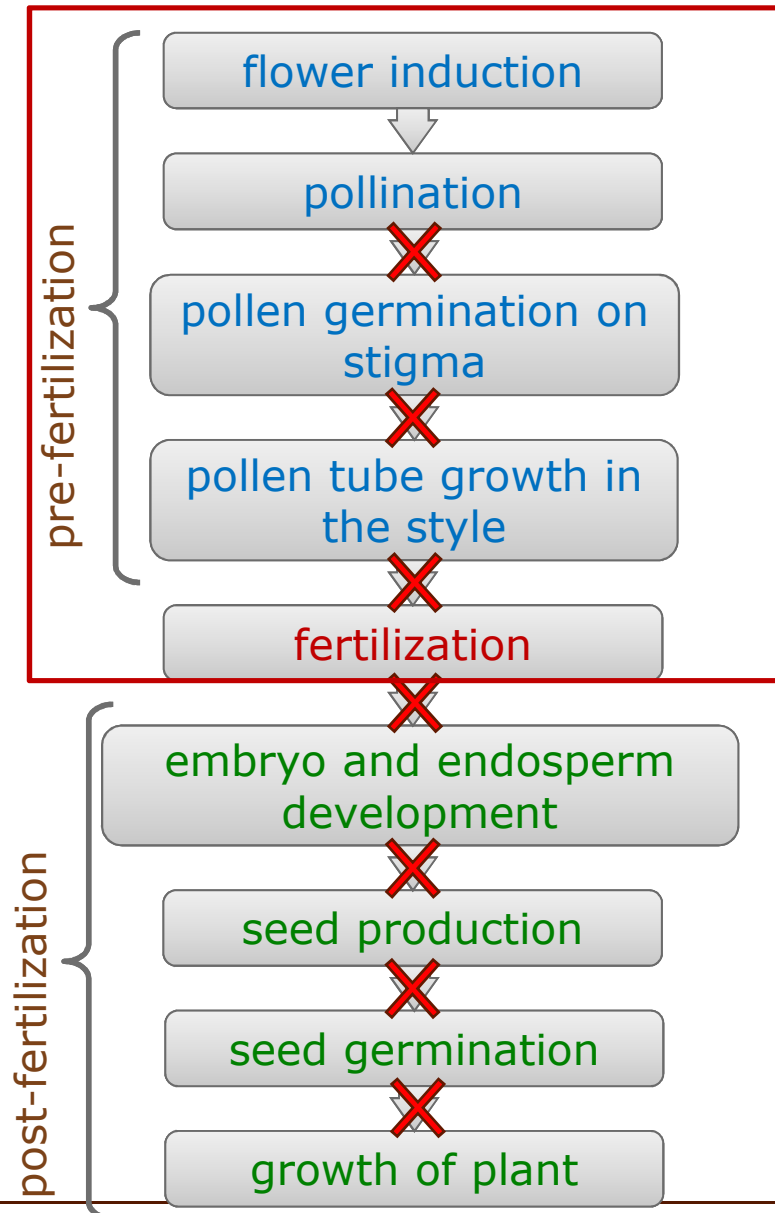
Key parts of the plant in reproductive process:

Pistil: female reproductive part of flower

Pollen grain: structure, which contains male reproductive cells



Reproductive process in wide hybridization



Hybridization barriers:

Pre-fertilization – all mechanisms that prevent fertilization

Post-fertilization – processes reducing hybrid embryo viability

Background of barriers:

active: incompatibility

passive: incongruity

In practice they are difficult to distinguish



Pollen quality

Failure in reproductive process can occur due to low quality or quantity of pollen delivered to stigma

In hybridization programs, pollen quality is routinely determined

Methods to determine pollen quality:

Viability testing

example – staining with 1% acetocarmine (cytoplasmic stain)

Germinability testing

In vitro pollen germination analysis using artificial medium containing basic salts and sucrose



Pollen–pistil interactions

Functions of pistil:

Promotion: it allows capture of pollen and germination of pollen tubes, it facilitates growth of pollen tubes by providing guidance and nutrients

Protection: it represents complex barrier that protects ovules from inappropriate pollen

Fertilization to occur → remarkable cellular coordination



Stigma receptivity

The ability of stigma to capture pollen by adhesion, let it hydrate and germinate pollen tubes

→ state of stigma when successful pollination of compatible plants takes place

Duration from hours to several days

Dry and wet stigmas – different mechanism of pollen adhesion



Examination of stigma receptivity

Morphological key – pistil during maturation undergoes distinct morphological changes;
Developmental stages can be correlated to the pollination success

Expansion of stigma in a receptive stage

Enzymatic key – peroxidase activity

In the receptive stage stigma is characterized by high enzymatic activity; during pistil maturation peroxidase is active in the stigma, and its activity reaches its peak in the receptive stage

Methods to examine presence of peroxidase: application of hydrogen peroxide or colorimetric reaction – peroxtesmo Ko paper test



Pollen tube growth

Possible observed incompatibility reactions in wide crosses:

- inhibition of pollen tube germination on stigma, multiple pollen tubes
- inhibition of pollen tubes in the style
- inhibition and branching of pollen tube in the style/ ovary
- swelling tip of pollen tube
- spiky and thick pollen tubes
- disorientation of pollen tubes in the ovary



Conclusions:

Why do we need knowledge about...?

Pollen quality:

- pollen as an only limiting factor

Stigma receptivity:

- time and duration of stigma receptivity → effective pollination period
- time of action of incompatibility mechanisms

Pollen tube germination and growth:

- occurrence and nature of pre-fertilization barriers
- localization of pollen tube inhibition
- observation of fertilization → post-zygotic barriers

