Dynamics of stomatal adaptation in rose leaves exposed to long-term high relative air humidity

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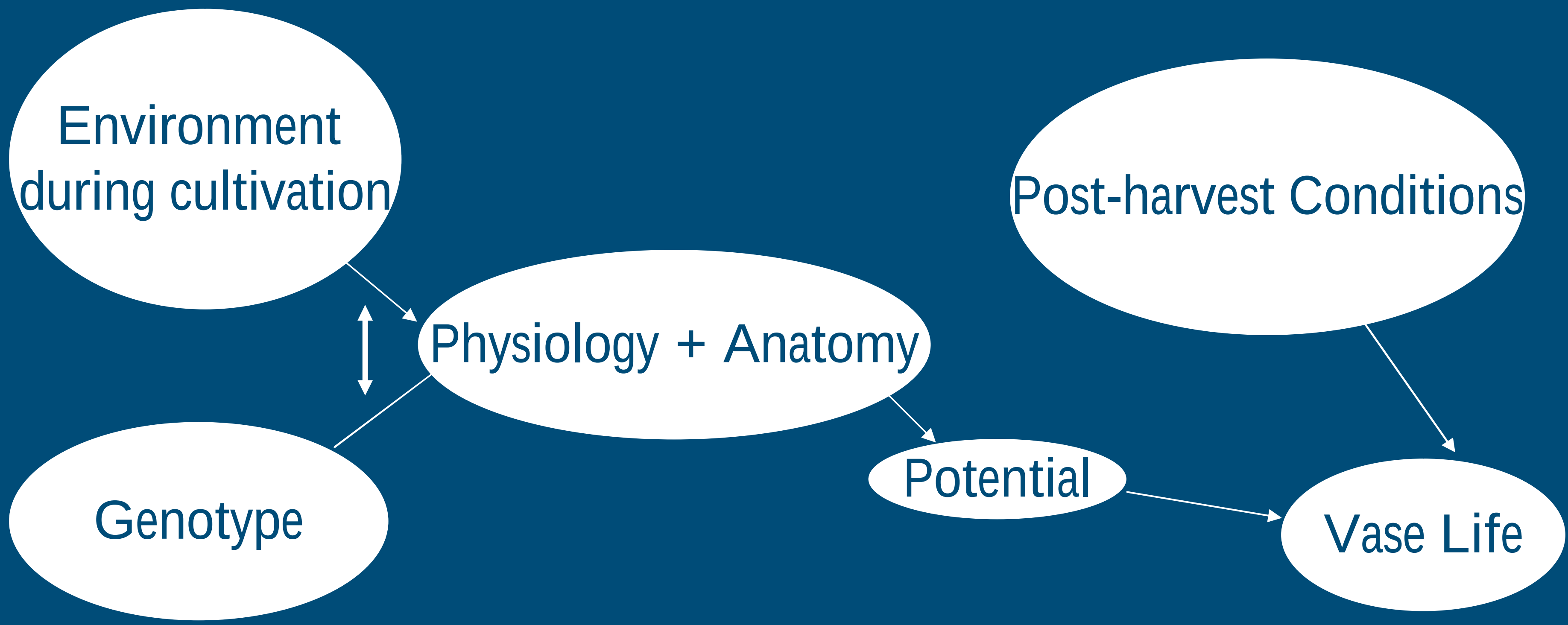
Long-term high relative air humidity (RH>85%) during cultivation downgrades keeping quality in cut flowers



- Light intensity, light quality, CO_2 fertilization, **air humidity**, water stress
- Xylem, stomatal, cuticle properties, hydrosensitivity of Ψ, flower opening

Not only obtain, but sustain quality

High RH is common in winter months in Northern Europe in greenhouse cultivation

Closed greenhouse (closed ventilation windows)
□ high RH levels

High RH during cultivation □ increased water loss during post-harvest (stomatal malfunction)
□ poor keeping quality

Cultivar dependent quality decrease

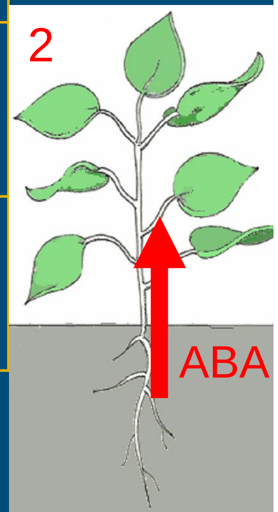
Manipulate humidity
during or *after* leaf development: compensate?

OR

?

Stomatal adaptation (or NOT) to new RH environment in fully developed leaves

Species	Moderate → High RH	High → Moderate RH
Phaseolus vulgaris [1]	√ (7 days)	√ (7 days)
Tradescantia virginiana [2]	√ (4 days)	× (6 days)
Rosa hybrida [3]	× (21 days)	× (21 days)



- Species dependent
- Certain amount of ABA is required **during** and **after** leaf development for functional stomata[2]

[1] Pospisilova (1996); [2] Rezaei Nejad and Van Meeteren (2006); [3] Mortensen and Gislerød (2000)

Hypotheses

- Humidity level
- Root (hormonal & hydraulic) signals

  after leaf development

  do NOT affect stomatal functionality

Materials & Methods

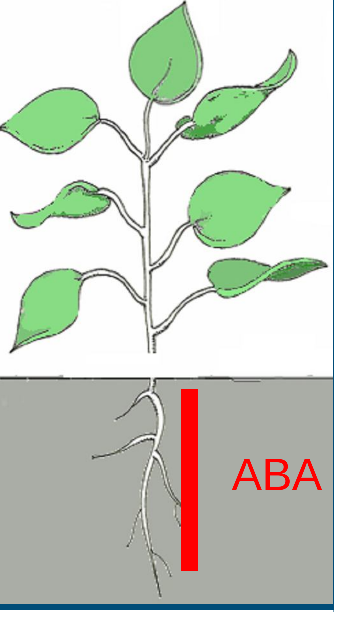
- Three climate chamber experiments ALL WITH:
  - cv: Prophyta (sensitive to high RH)
  - 2 RH: High RH (90%); Moderate RH (60%)
  - Light: 300 ± 20 μmol $m^{-2}s^{-1}$, 18 h/d
  - 19˚C day/night, ambient CO_2

Role of RH and root signals' in fully developed leaves (Exp. 1)

Fully grown plants (flower stage 2):

- transferred to humidity other than growth (60%→90%; 90% →60%)
- cut roses, placed in vases in evaluation room (for 60% RH only) (20˚C, RH: 50%, 10-12 µmol $m^{-2}s^{-1}$, 12 h/d)

Stage 2

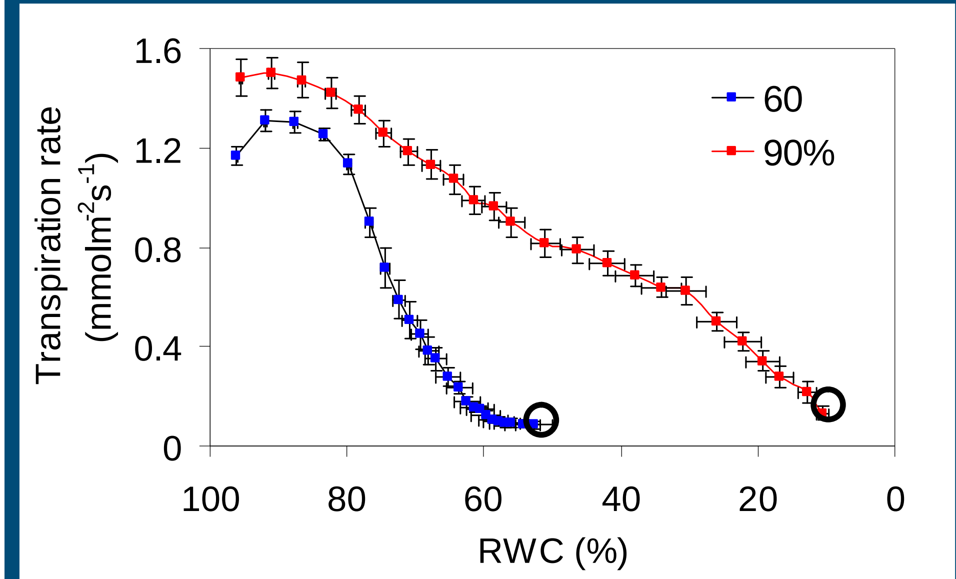


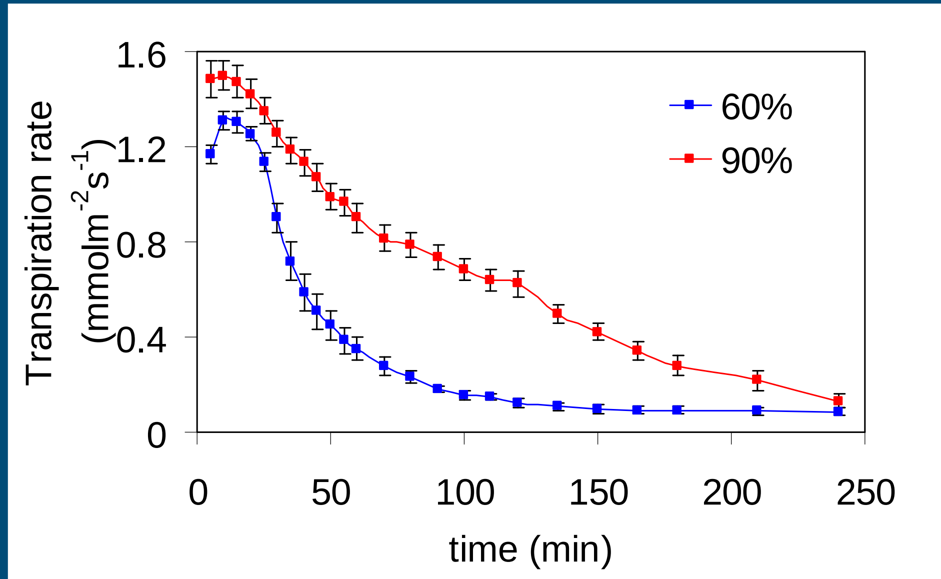
Measurements (Exp. 1)

- Stomatal responses to desiccation were followed every 2d
  - new humidity: 14 replicates, 1 leaf/ plant
  - cut roses: 8 replicates; 1 leaf/ stem

- Rehydrated terminal (detached) leaflets
- T: 21^{o}C, RH: 50±3 % (1.47 kPa VPD),& 50 µmol m^{-2} s^{-1}
- Transpiration rate measured gravimetrically

Stomatal responses to desiccation



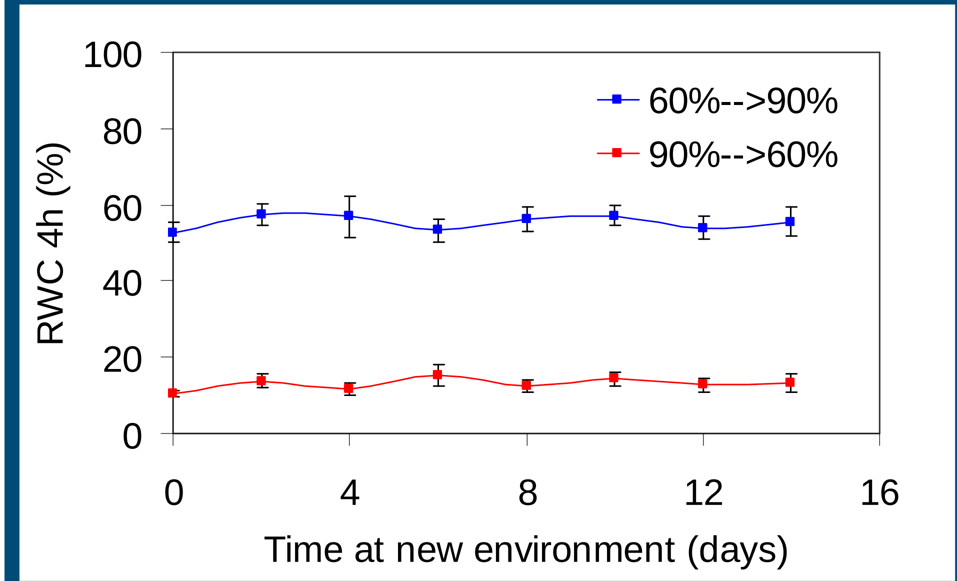


Relative Water Content=[(FW-DW)/SFW-DW)]*100

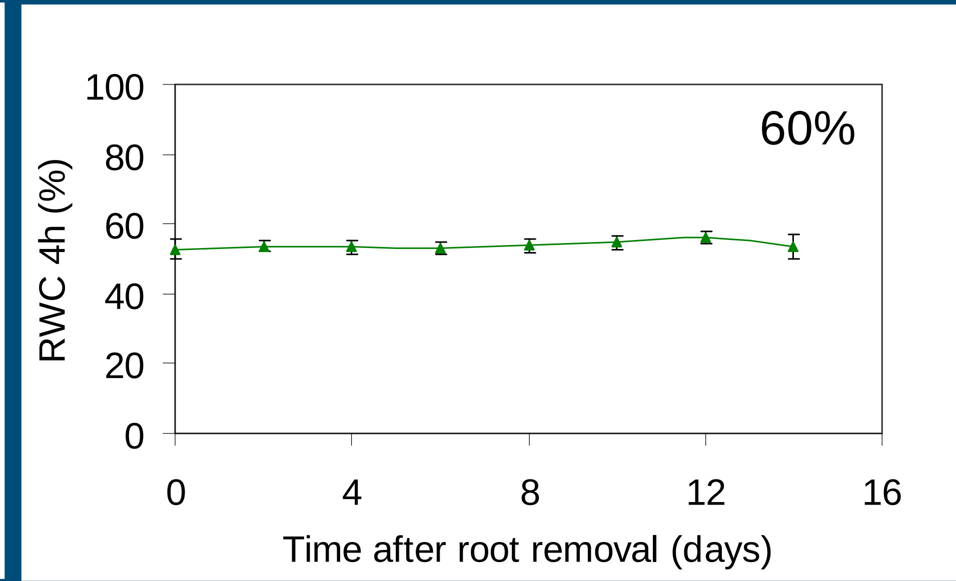
- Iwanoff effect
- High RH affects speed and degree of stomata closure
- End value can be misleading

RWC 4h after desiccation (role of RH and of roots)

Role of RH



Role of roots



- No stomatal adaptation after 14d in new RH environment
- Vase life: stomata stay functional even after 14d without root signals (hormonal & hydraulic)

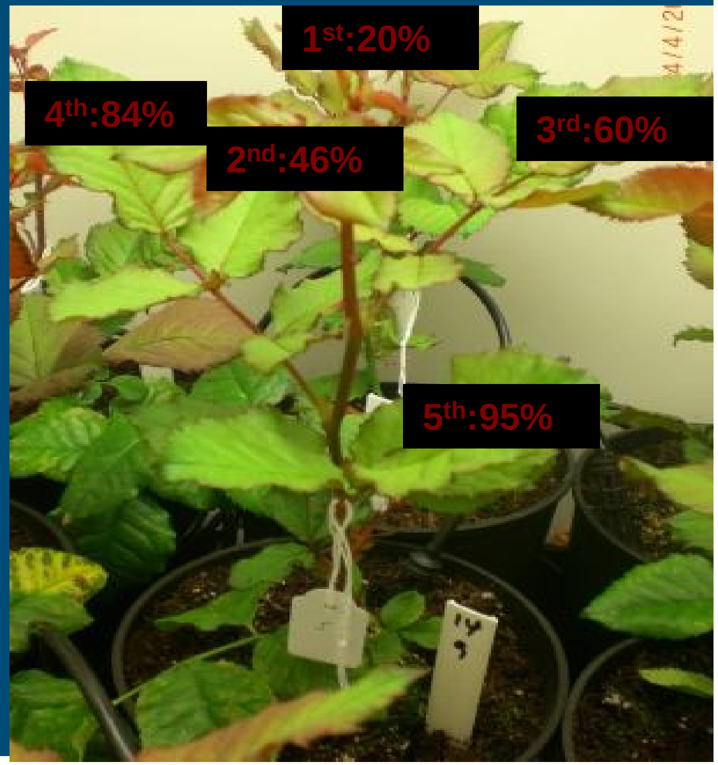
New research questions

- Is the high RH effect on stomatal behaviour gradual during leaf development?

- Are the non fully developed leaves able to adapt to a new RH environment? To which extent?

Role of RH during leaf development (Exp. 2)

Five weeks after bud break (1st leaf≈20% final length), plants transferred to:



- New RH environment (60%→90%; 90%→60%)
- treated with 30µm ABA twice a day (1st leaf only)

Measurements (Exp. 2)

- Leaf expansion was followed daily (measuring leaf length of top 5 leaves)
- Fully developed leaf (100% FLE)→ leaf length is constant for 3 consecutive days

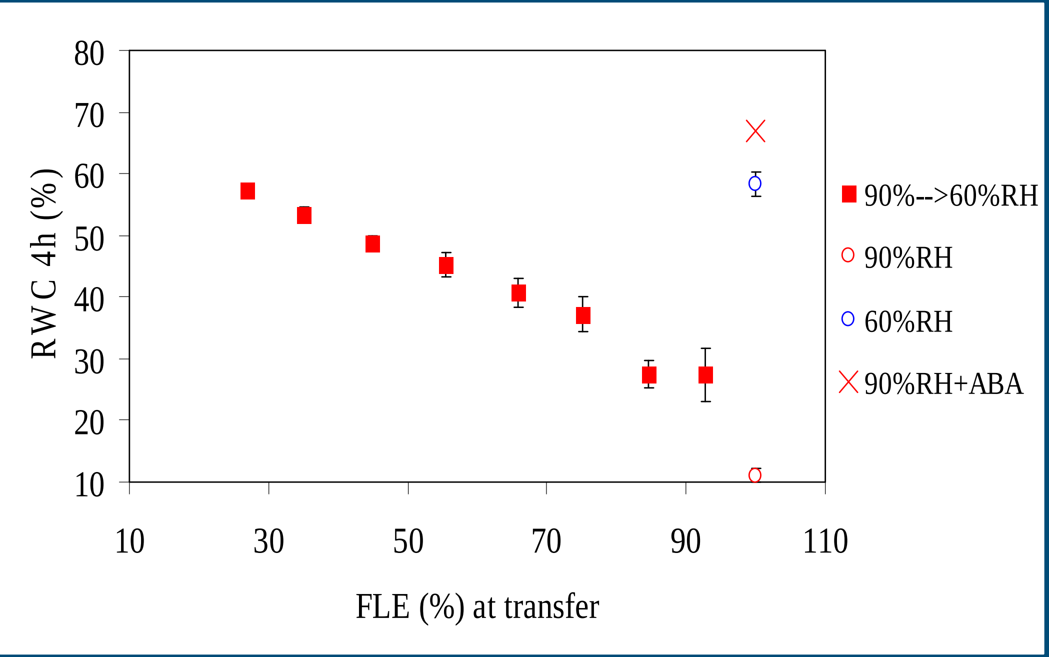
criterion = **100%FLE** and not time in the new humidity (Exp.1)

- Stomatal responses to desiccation in fully expanded leaves
(3 repetitions, n>18)

Similarly to Exp. 1:

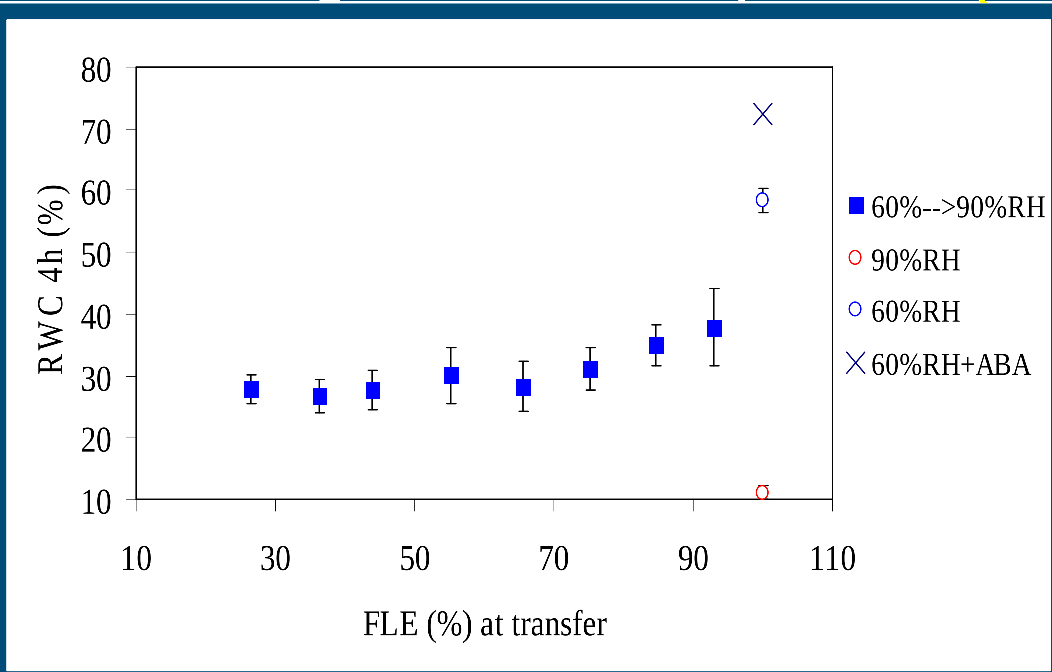
- Rehydrated terminal (detached) leaflets
- RH :50±3 %, T:21^{o}C (1.47 kPa VPD),& 50 μmol m^{-2} s^{-1}
- Transpiration rate measured gravimetrically

Role of RH during leaf development (90%→60%)



- 27% FLE behave as control moderate RH; 10% step in FLE: different behavior (even >90% FLE before transfer is better than high RH control)
- not complete, but partial adaptation
- ABA application induced very active stomatal functioning

Role of RH during leaf development (60%→90%)



- High RH during the last part of leaf expansion is detrimental irrespective the FLE at the time of transfer (FLE<80%)
- No differentiation between different FLE classes
- Not complete, but partial adaptation
- ABA application induced very active stomatal functioning

New research questions

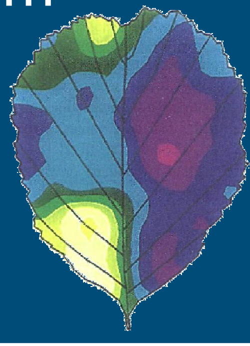
- Is the gradual effect of moderate RH due to gradual stomatal initiation during leaf expansion?
- Does the different humidity effect come from different stomatal initiation/ appearance?

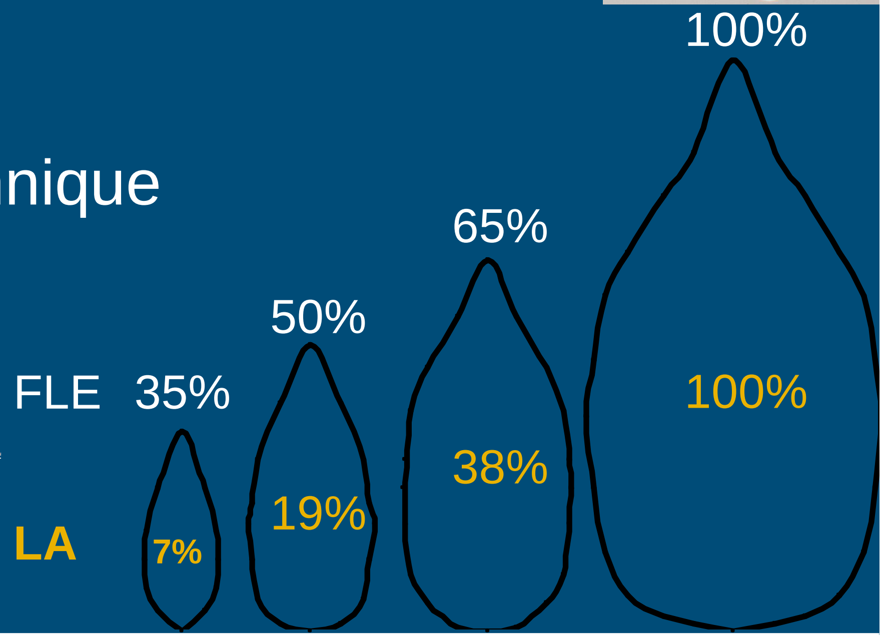
Stomatal initiation and development (Exp. 3)

- 35, 50, 65, 100% FLE (leaf order 1)
  (4 leaflets/ stage and humidity level)
- Paired sampling
  (always control was 100% lateral leaflet)

<u>Measurements:</u>

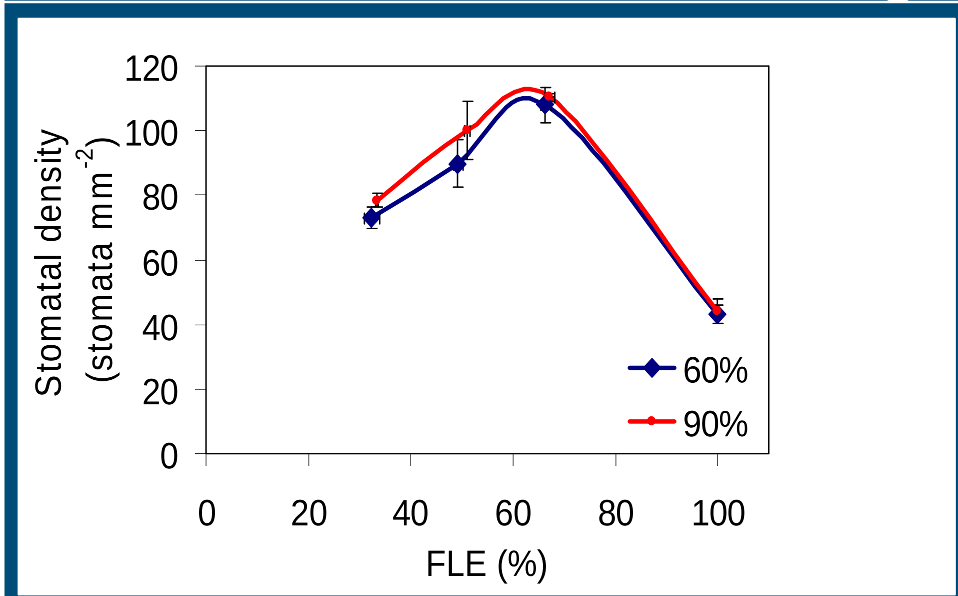
- Silicon rubber impression technique
- [1]Sampling area 1x1 cm
- Stomatal density

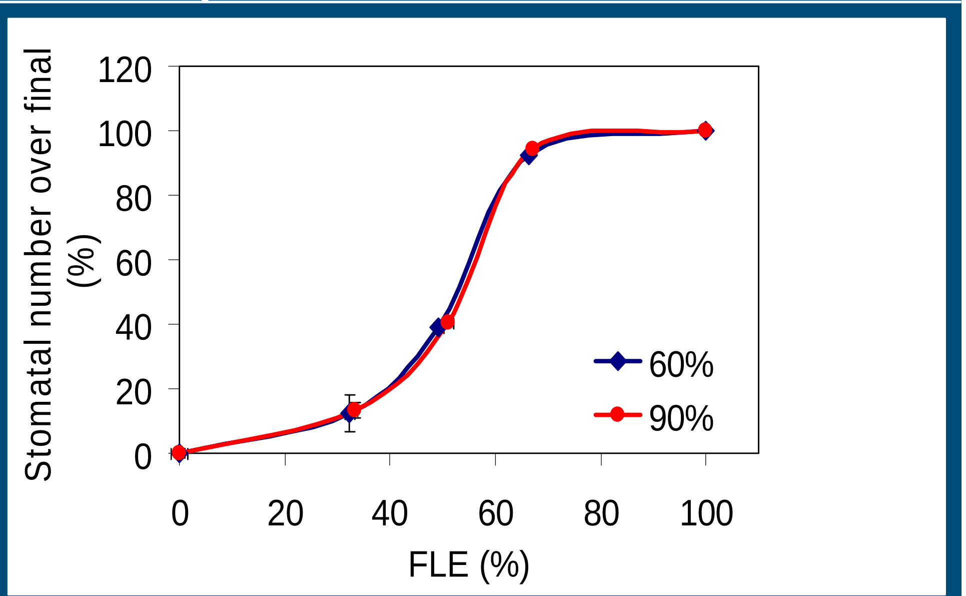




[1]Poole et al (1996)

Stomatal initiation during leaf expansion





absolute stomata number = density × area

- Stomatal initiation pattern was not affected by humidity of growth
- At 65% FLE (or 40%LA) over 95% of stomata are present

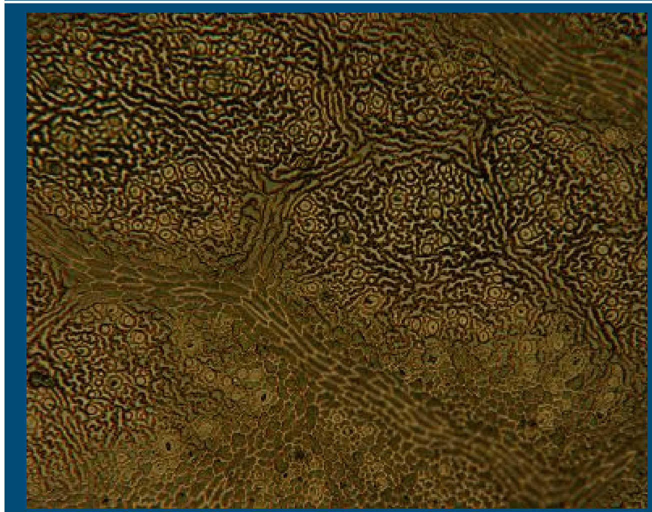
Conclusions

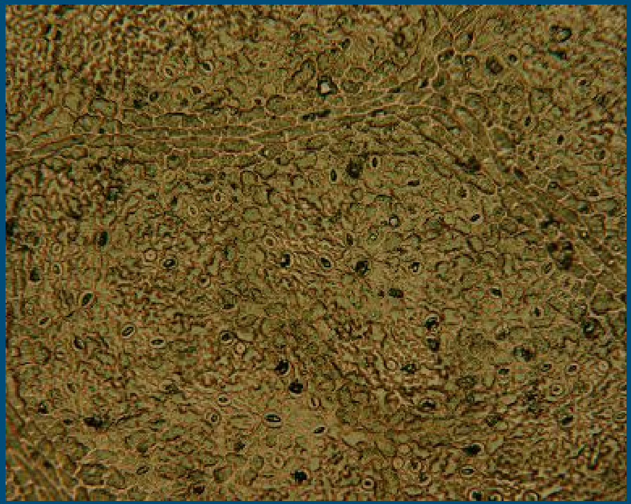
- Stomatal physiology is determined during leaf development
  In fully developed leaves the RH level or presence of roots signals' are NOT important

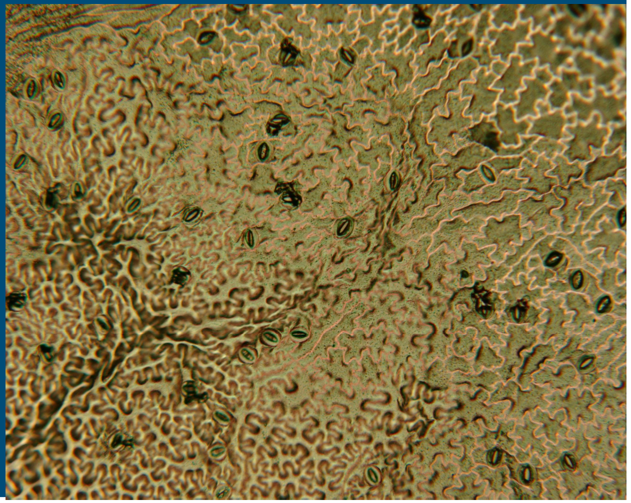
- When last part of leaf development = moderate RH:
  the earlier the switch (i.e. lower FLE) the better
  (*partial adaptation;* **different potential**)
- When last part of leaf development = high RH
  the time of the switch (FLE<80%): minor effect
  (*partial adaptation;* vulnerable till 95%FLE)

- ABA application during leaf development: induces very active stomatal control
- Adaptation dynamics: stomatal development (maturation)
  NOT stomatal appearance (initiation)

THANK YOU FOR ATTENTION!







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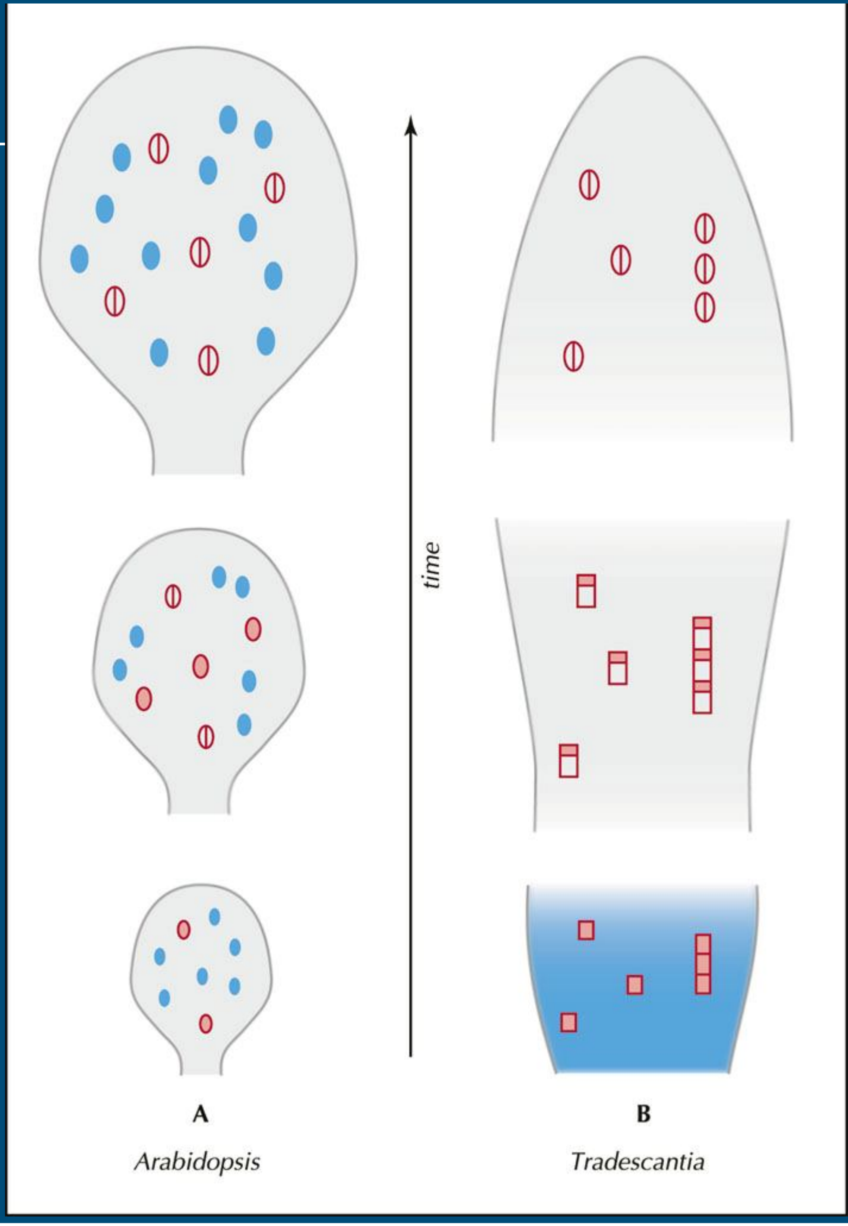

Coxdale (2000)