Productional Biology: Kinetic Imaging of Plant Chlorophyll Fluorescence

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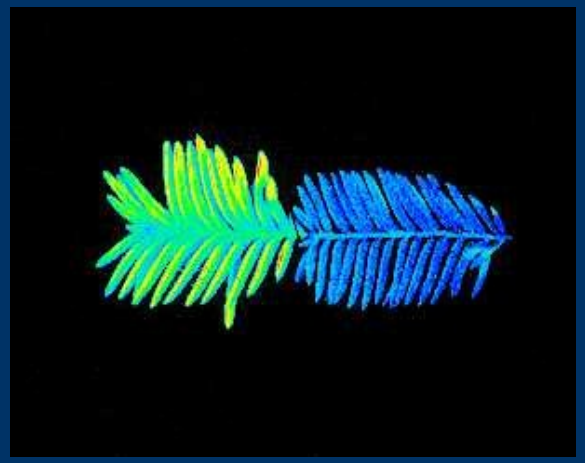
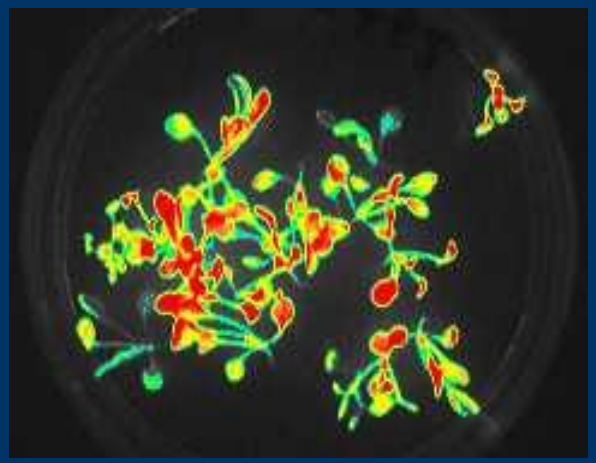
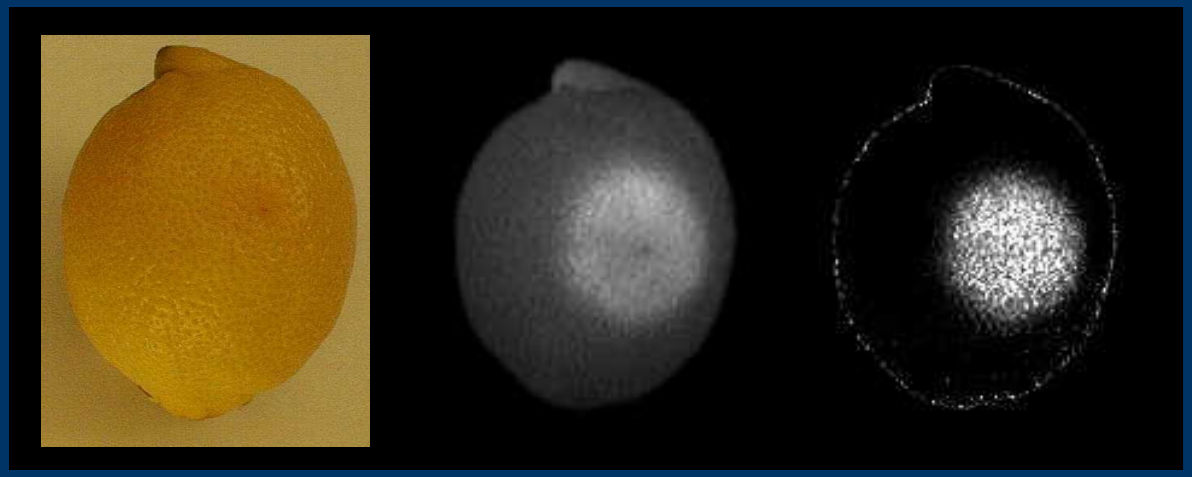
modified by M. Bartak

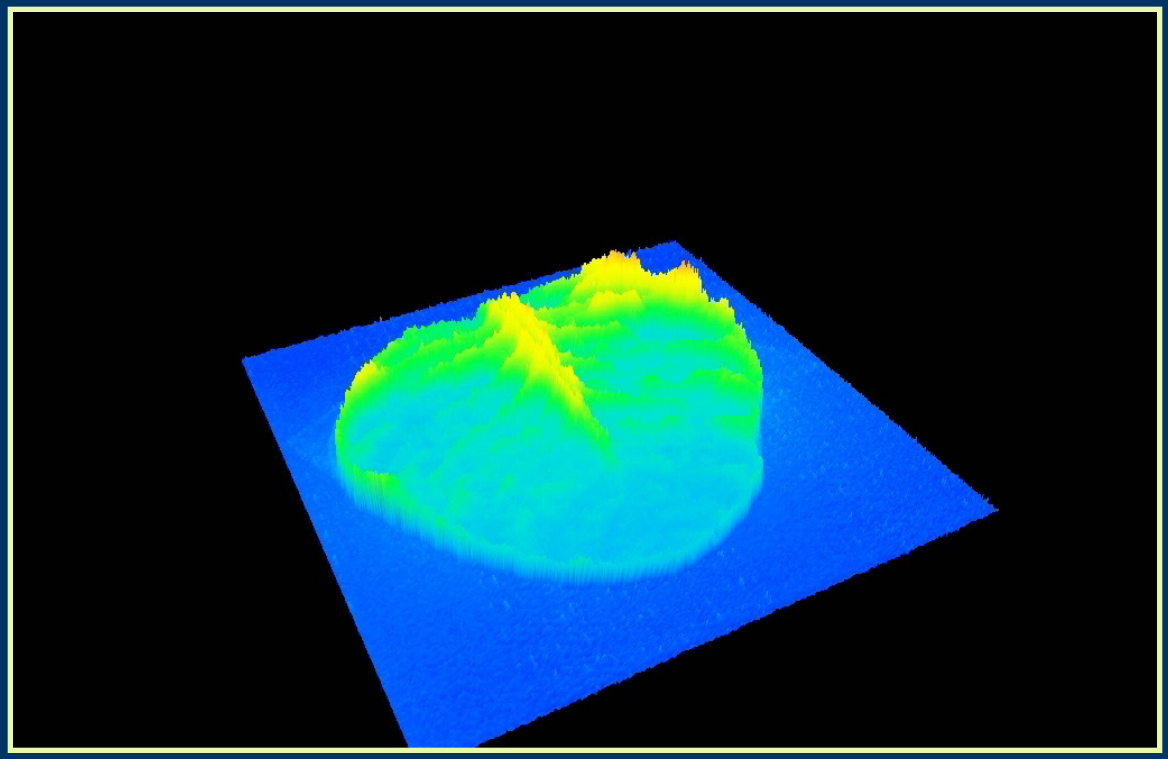




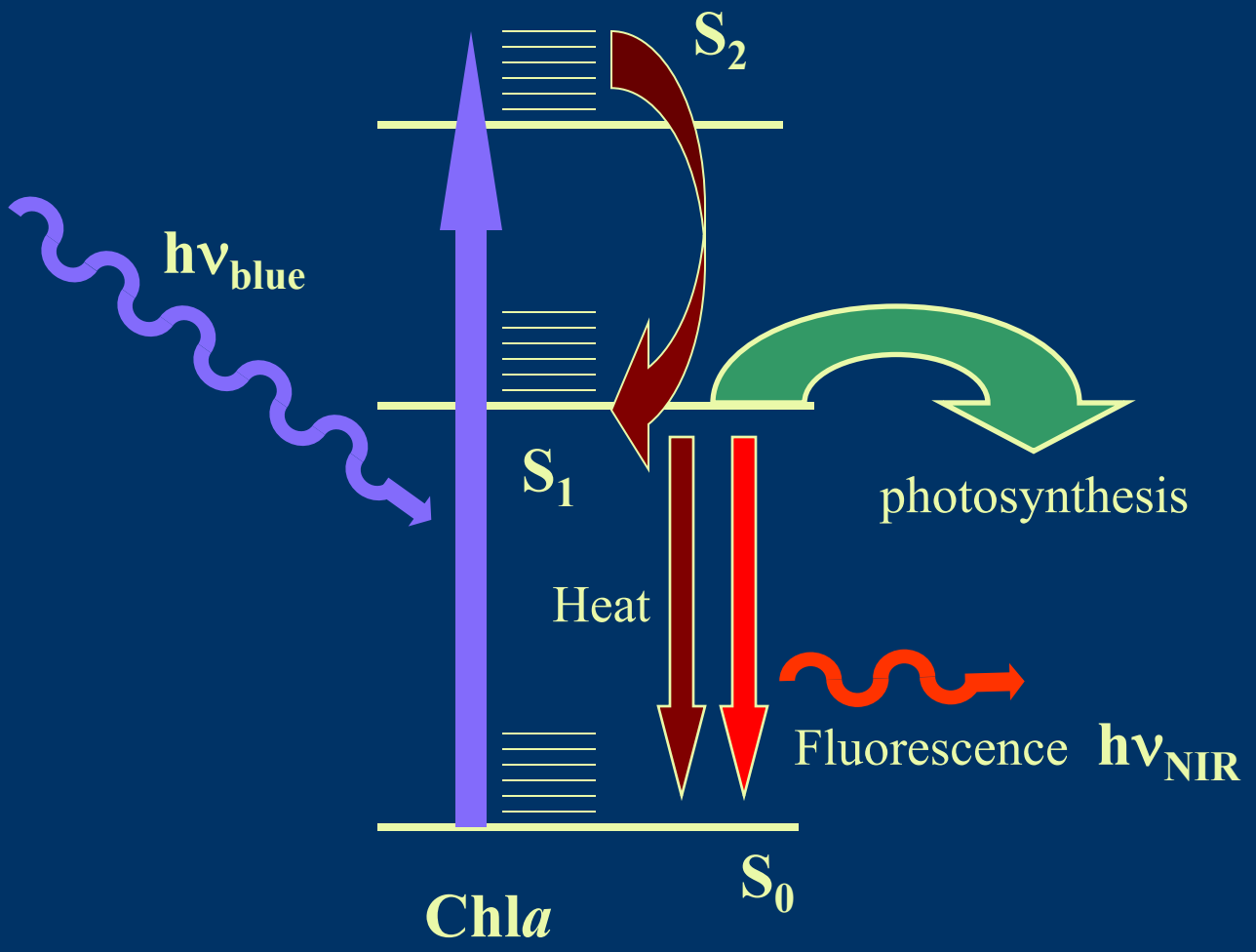


Early Fluorescence Imaging Experiment

Kautsky and Hirsch (1931) irradiated a dark-adapted leaf with a blue light and observed it visually through a dark-red glass. Here is a high-tech presentation of what they saw:



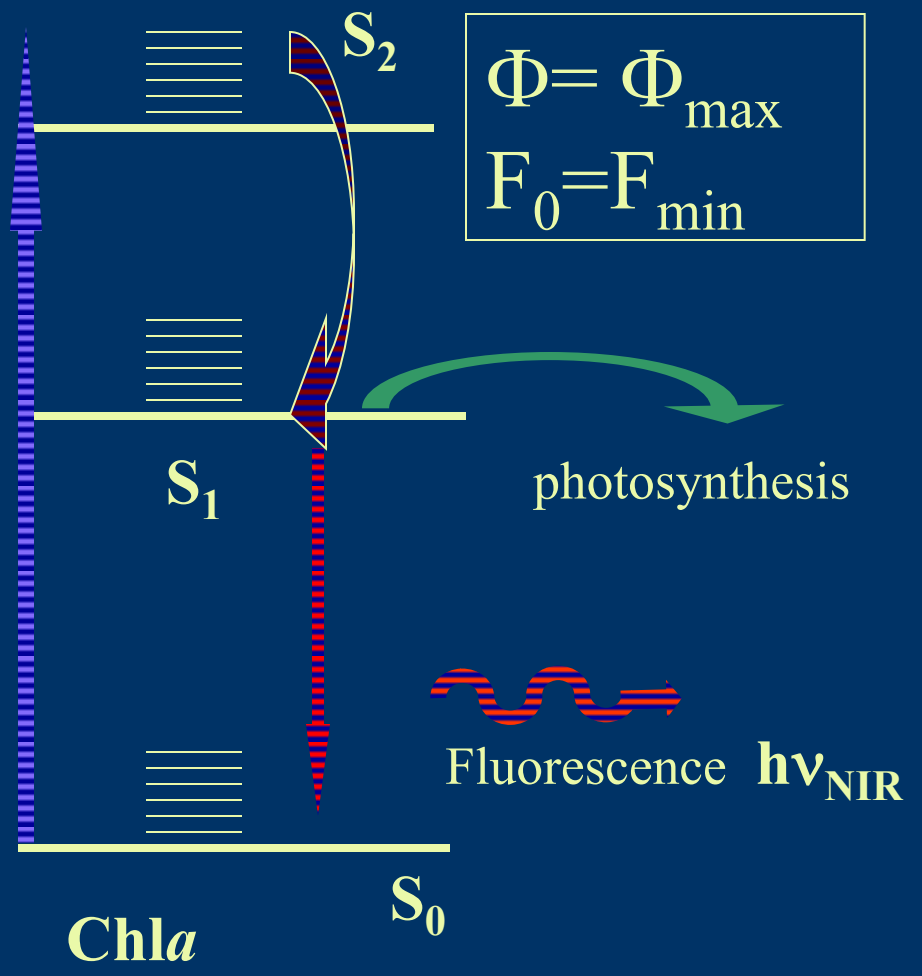
Chlorophyll *a* fluorescence competes with photosynthesis for excitation energy



Role #1 of light in plant fluorescence experiments – measuring light

Aim: Excite the fluorescence-emitting pigment molecules without changing the experimental photo-chemically active object. Fluorescence should be distinguishable from background of the same color.

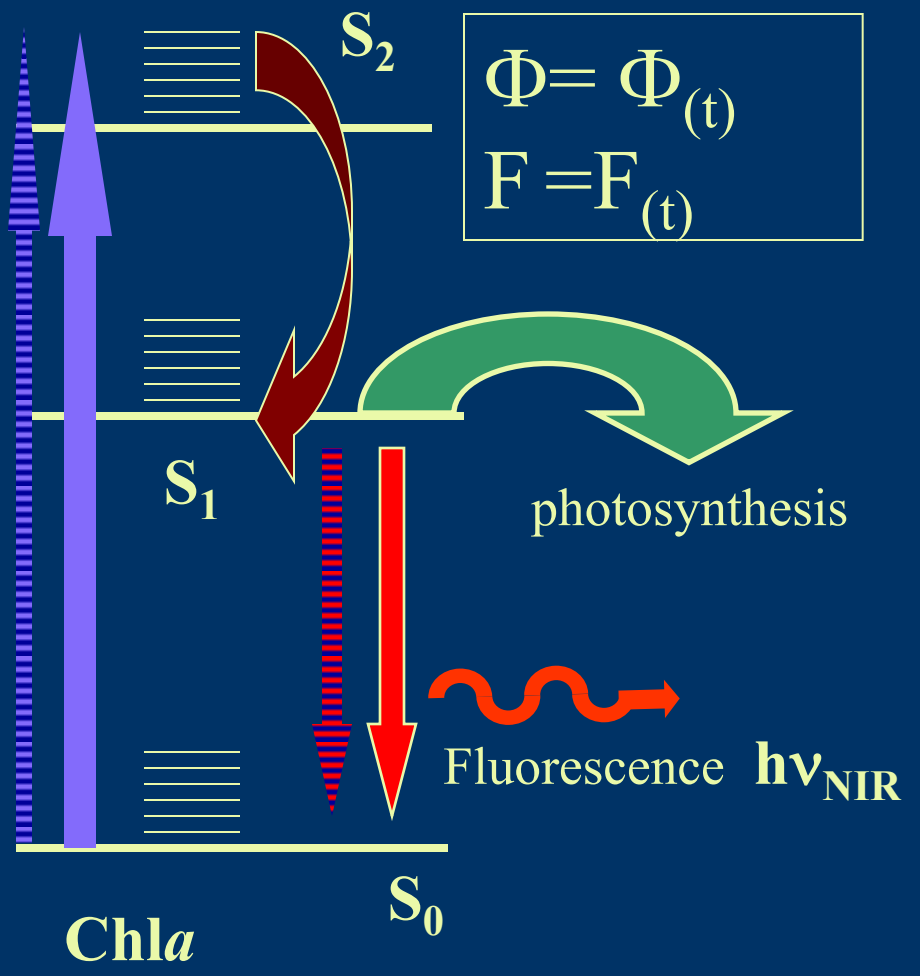
Achieved by MEASURING light: Typically 10-30μs long flashes repeated with a low frequency that



Role #2 of light in plant fluorescence experiments – actinic light

Aim: Excite the fluorescence-emitting pigment molecules without changing the experimental photo-chemically active object. Fluorescence should be distinguishable from background of the same color.

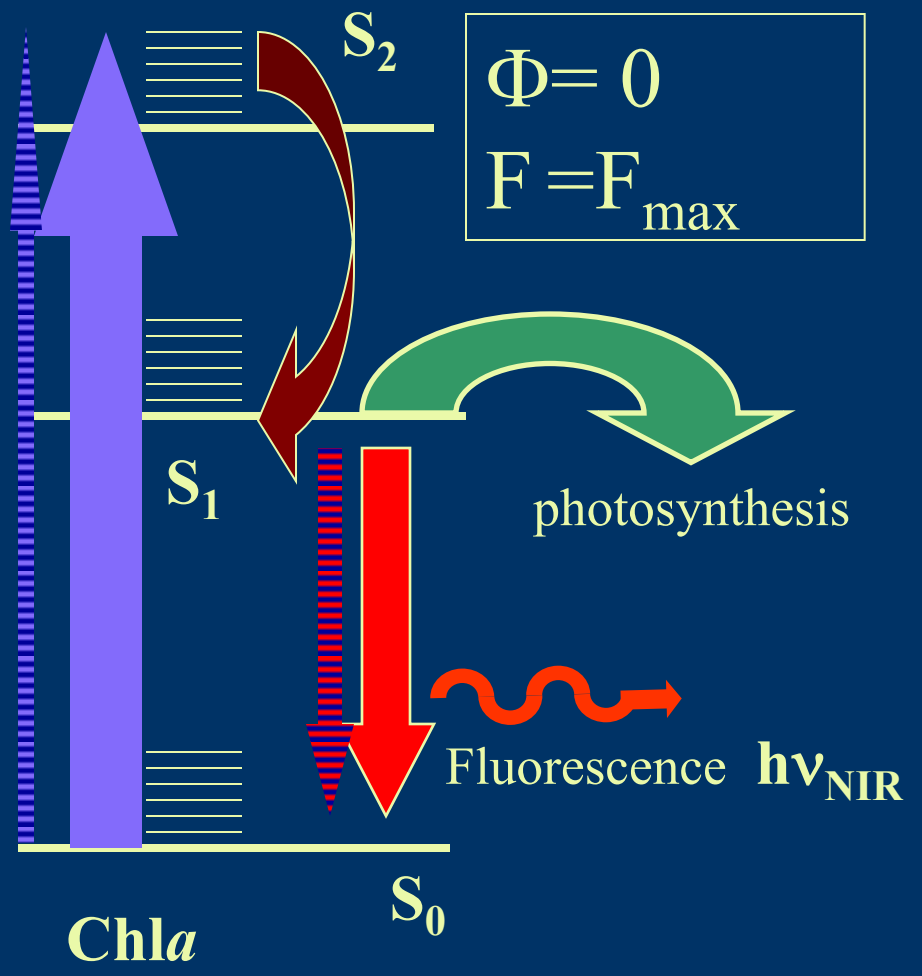
Achieved by MEASURING light: Typically 10-30μs long flashes repeated with a low frequency that



Role #3 of light in plant fluorescence experiments – saturating light

Aim: Excite the fluorescence-emitting pigment molecules without changing the experimental photo-chemically active object. Fluorescence should be distinguishable from background of the same color.

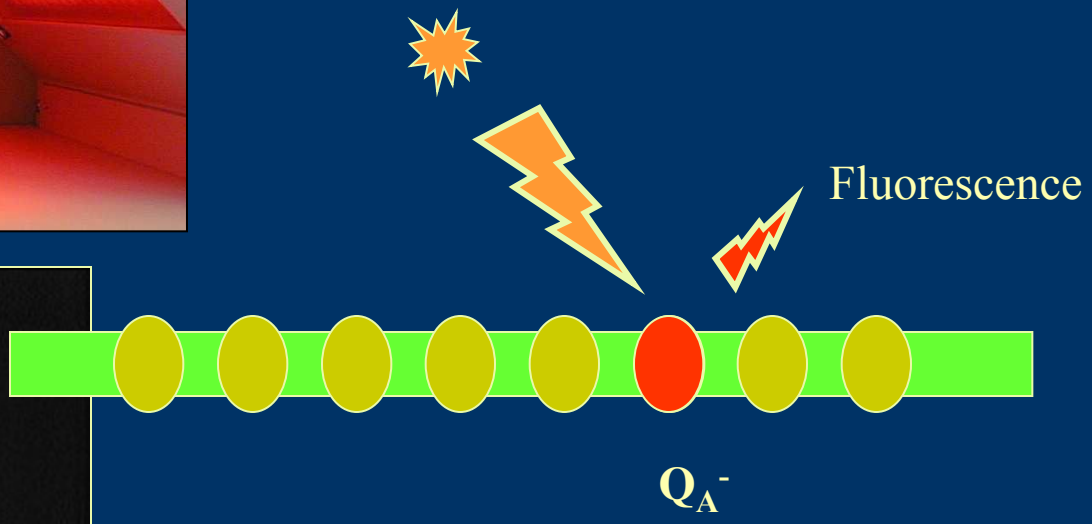
Achieved by MEASURING light: Typically 10-30μs long flashes repeated with a low frequency that

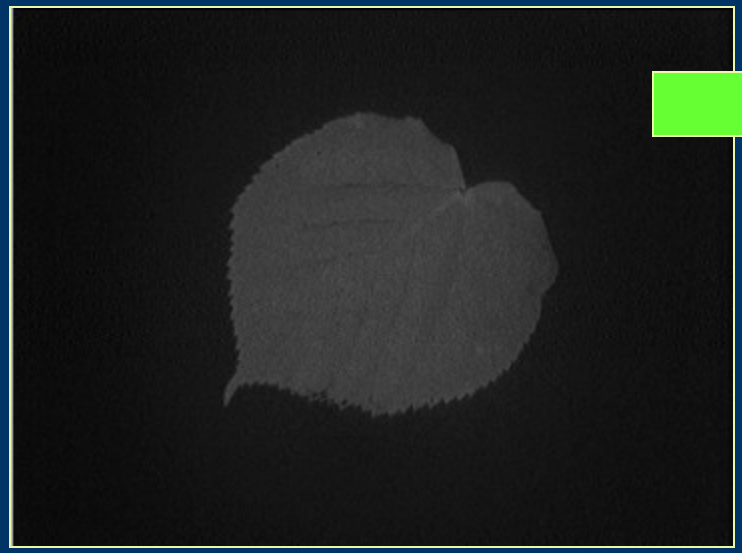


Measuring flashes have little actinic effects

750 LED's are on for 10-200 µs

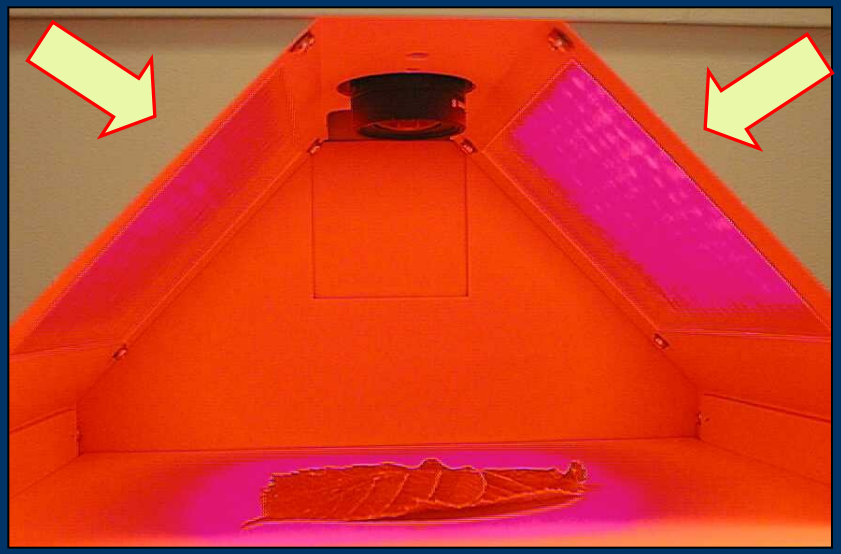
Only few PSII RC's are excited



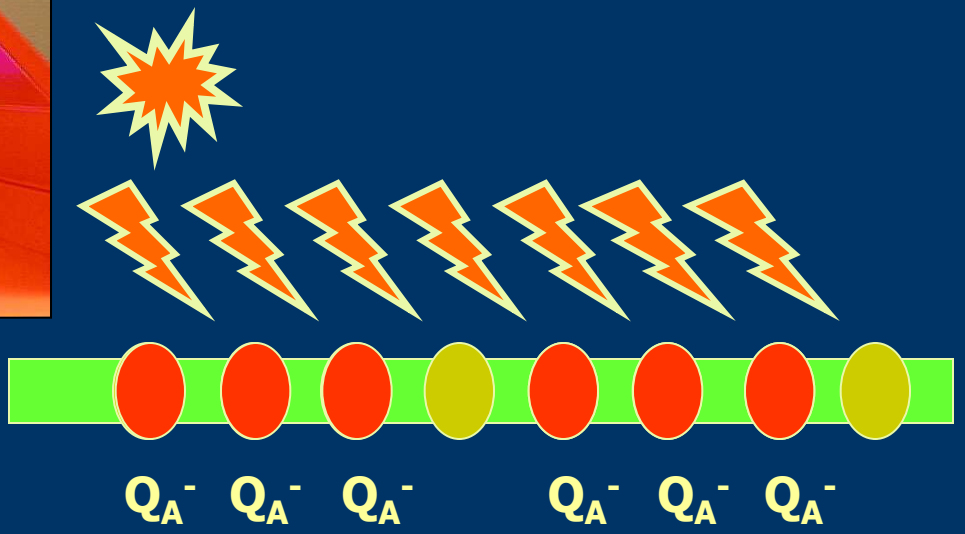


Yet, sufficient fluorescence emission is produced to capture an image

Actinic light is causing fluorescence induction

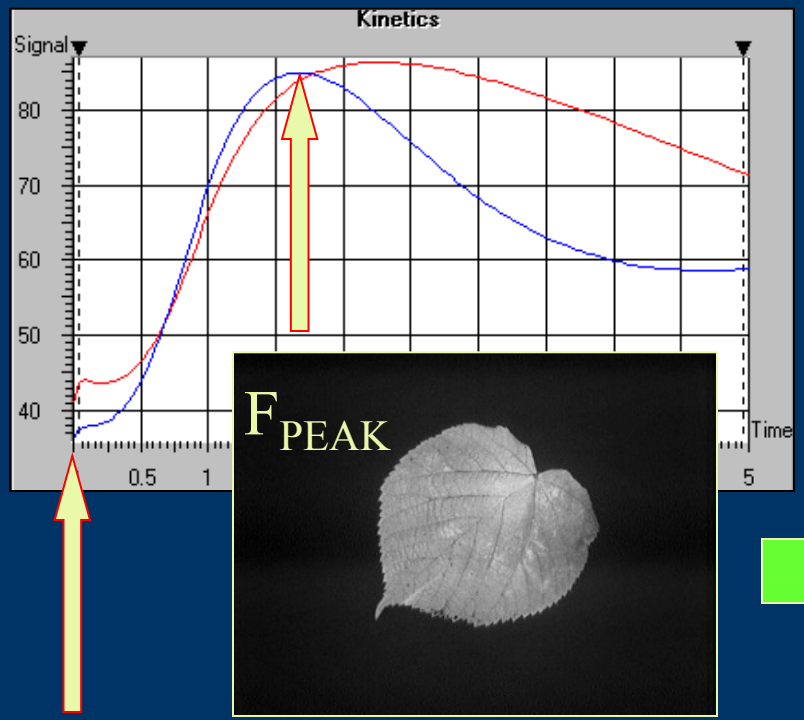


LEDs are on
for seconds to minutes

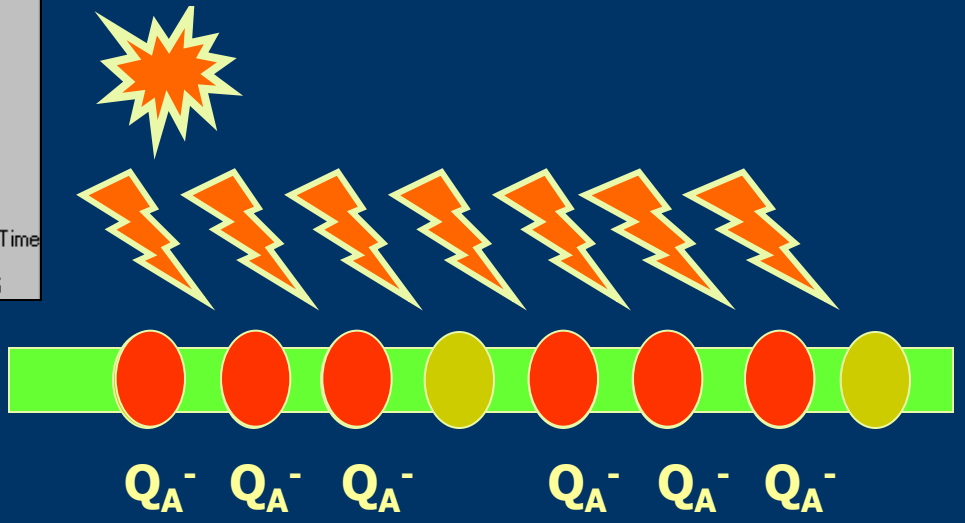


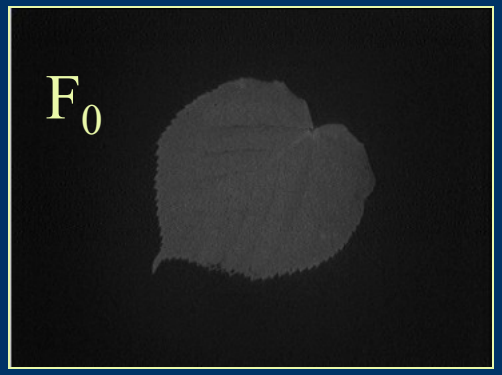
During the actinic light exposure, the continuous excitation keeps some of the PSII RC's closed

Actinic light is causing fluorescence induction



In fluorescence, the actinic light elicits in plants the Kautsky effect of fluorescence induction.

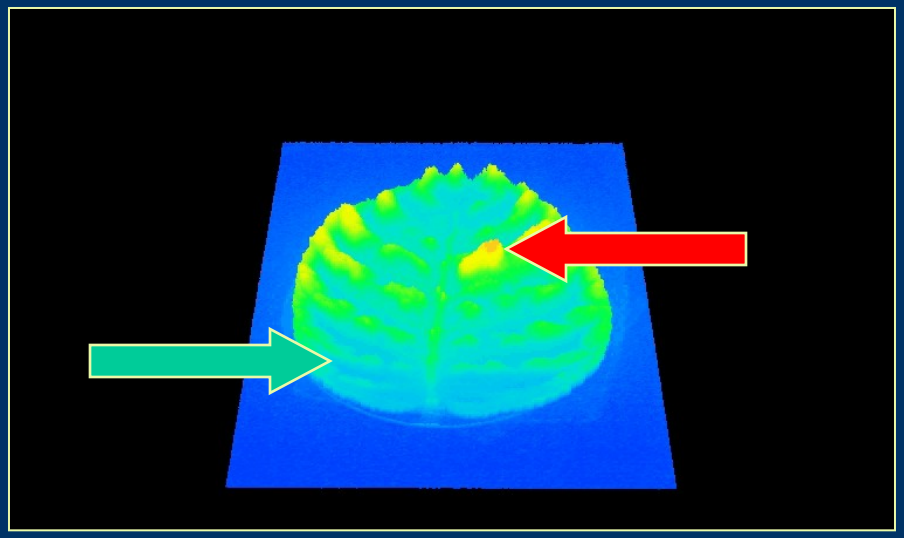




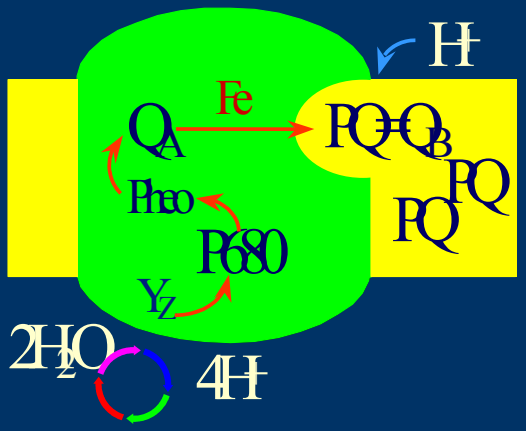
to F_{PEAK} with mostly closed PSII RC's

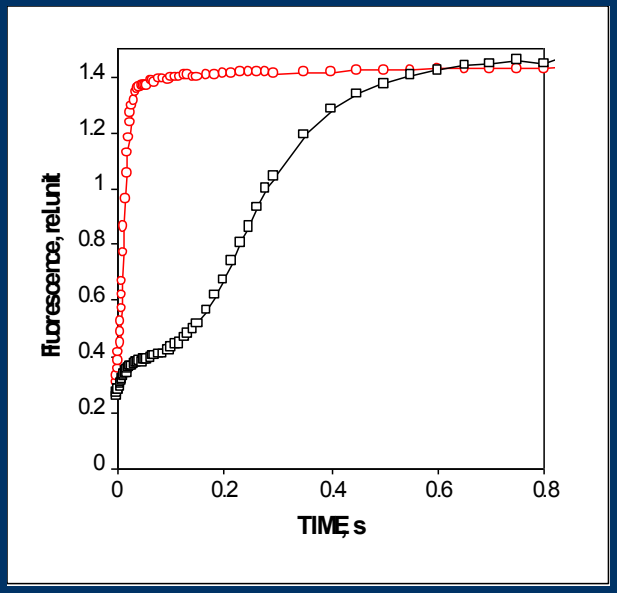
from F_0 with open PSII RC's

Induction in a diuron-inhibited leaf

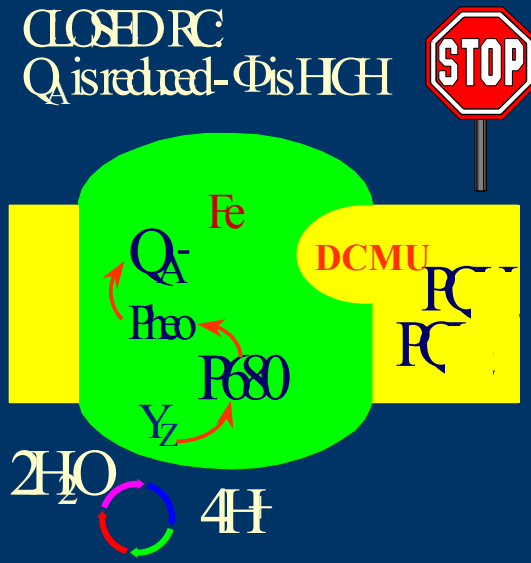


OPEN RC:
Q_A is oxidized - Φ is LOW



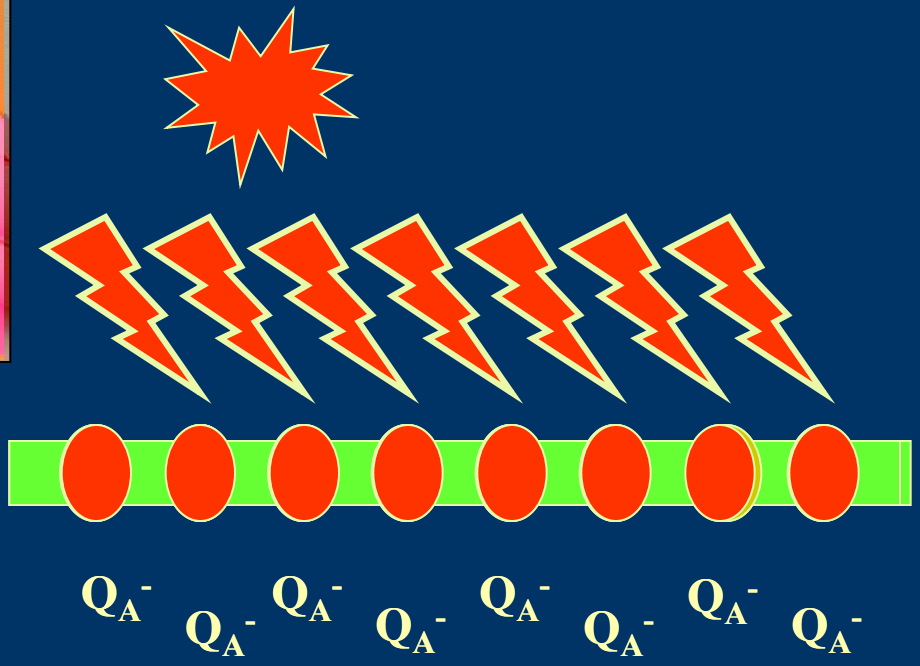


CLOSED RC:
Q_A is reduced - Φ is HIGH



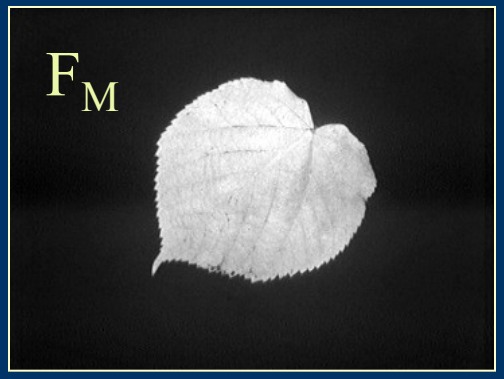
PQ-reducing super pulse

The shutter of the halogen lamp is open typically for 1s



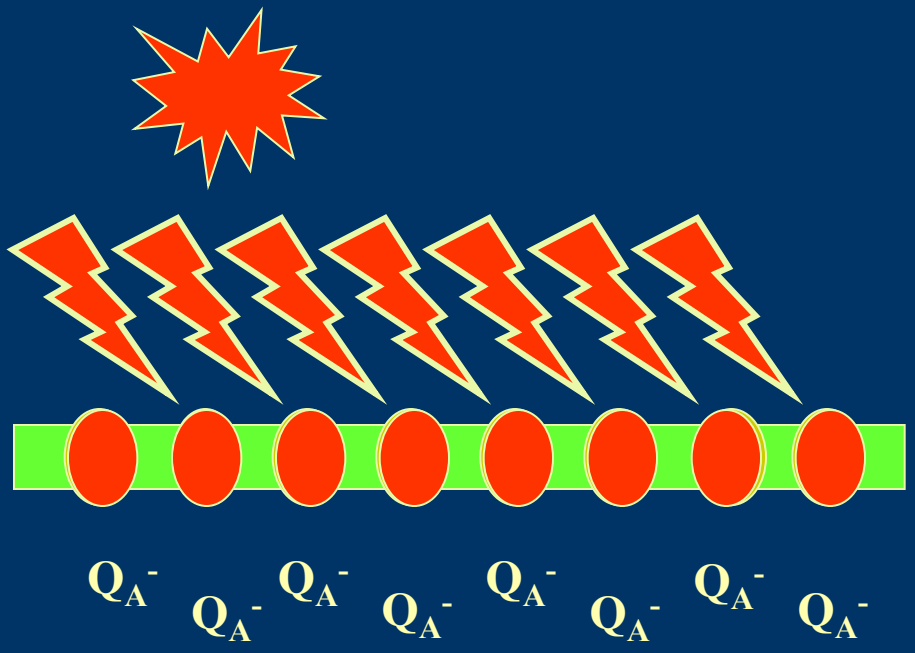
During the pulse, PSII RC's are closed
by a transient reduction of the plastoquinone pool.

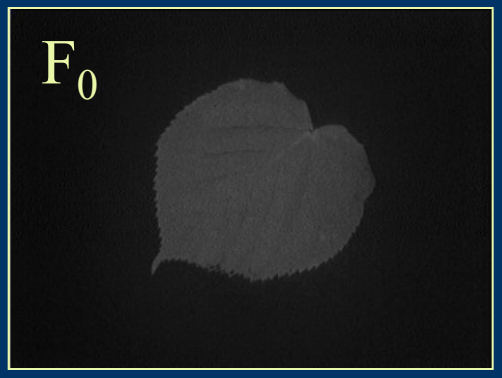
Fluorescence in PQ-reducing saturation pulse.



Fluorescence at the end of the pulse

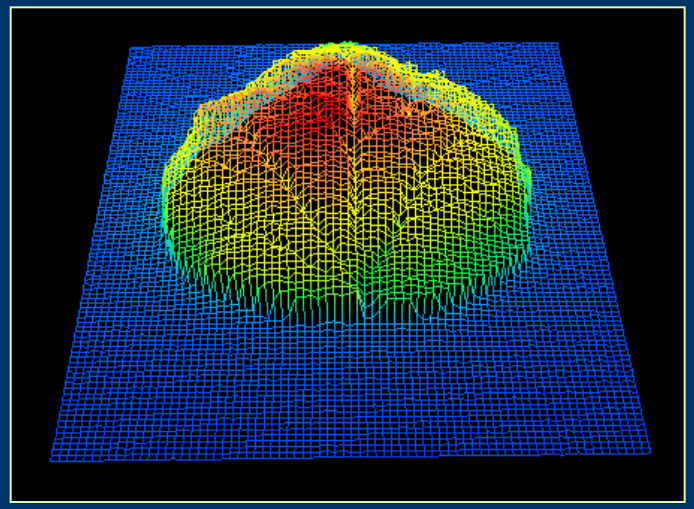
The closure of all PS RC's is reflected by a transient from F_0 to F_M.

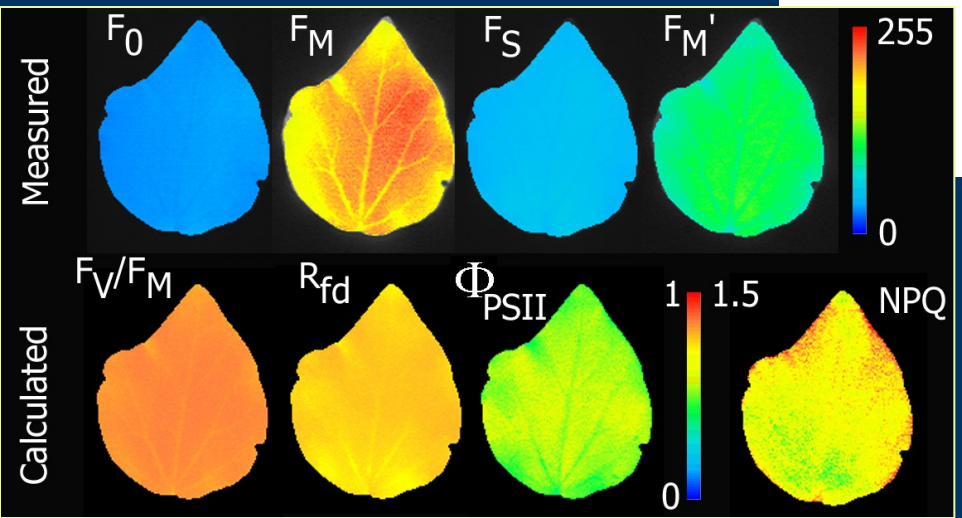


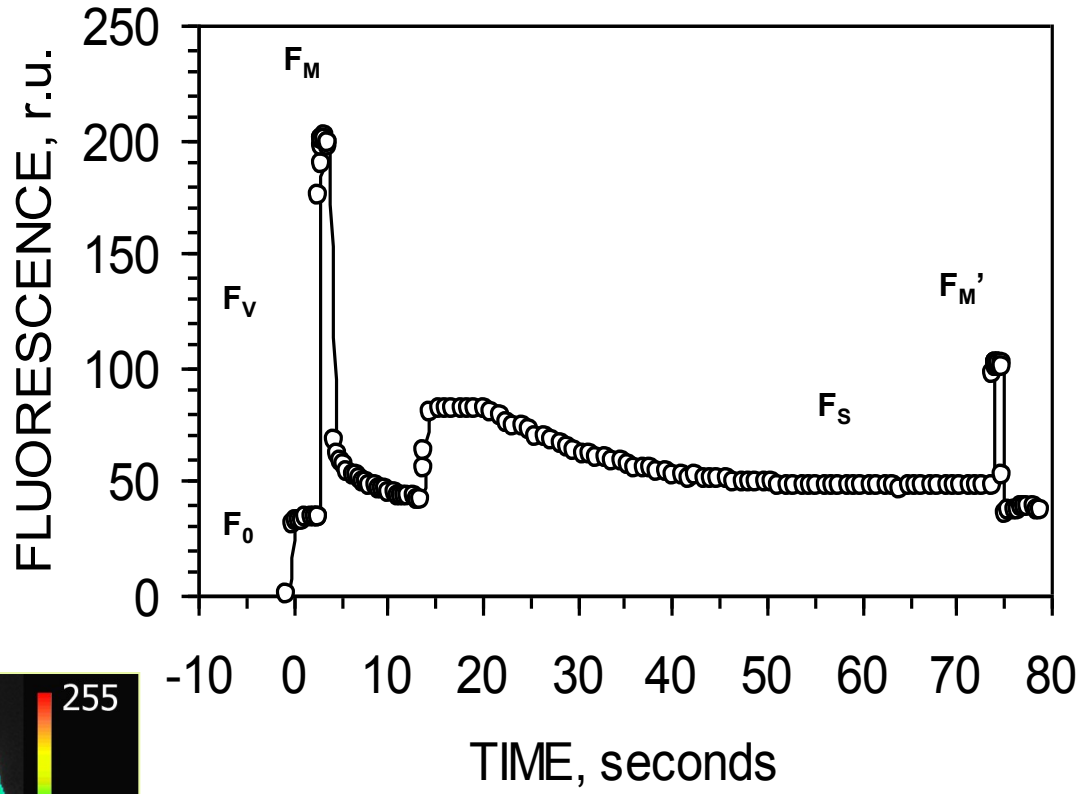


Fluorescence before the pulse

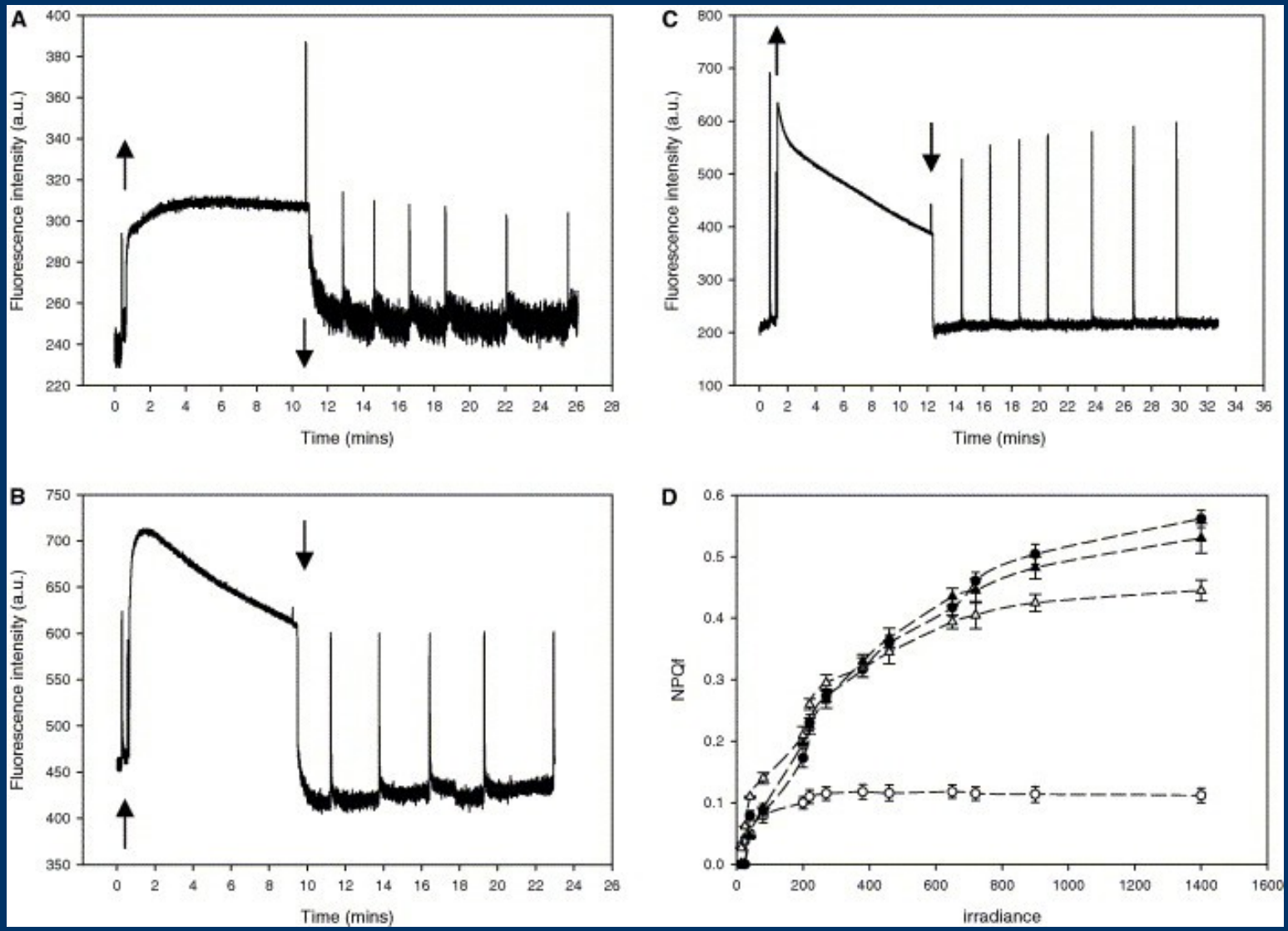
Pixel-to-pixel arithmetic image operations



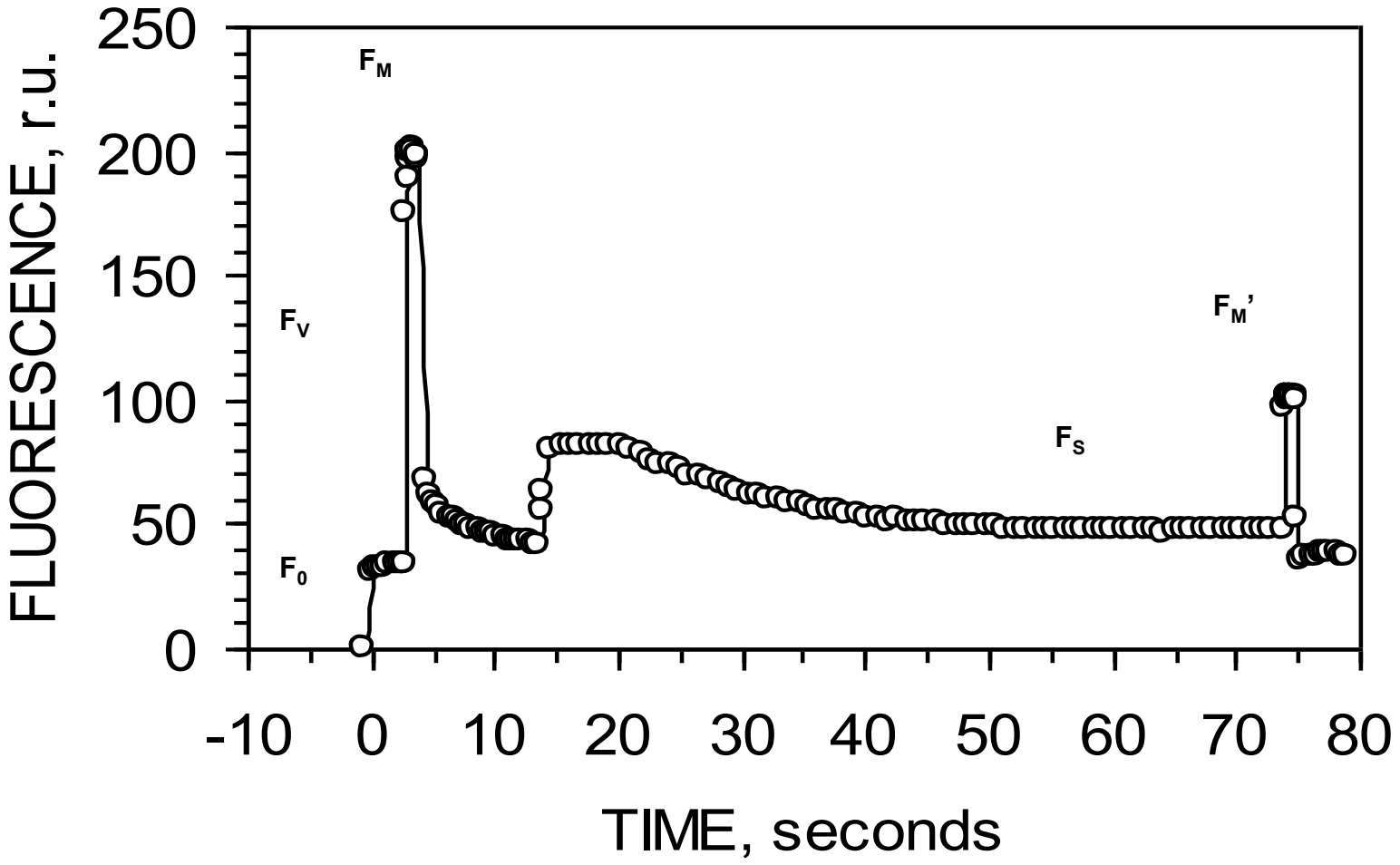




„Cyanobacterial“ Chl fluorescence kinetics



- Source: http://www.sciencedirect.com/science/article/pii/S0014579304014991



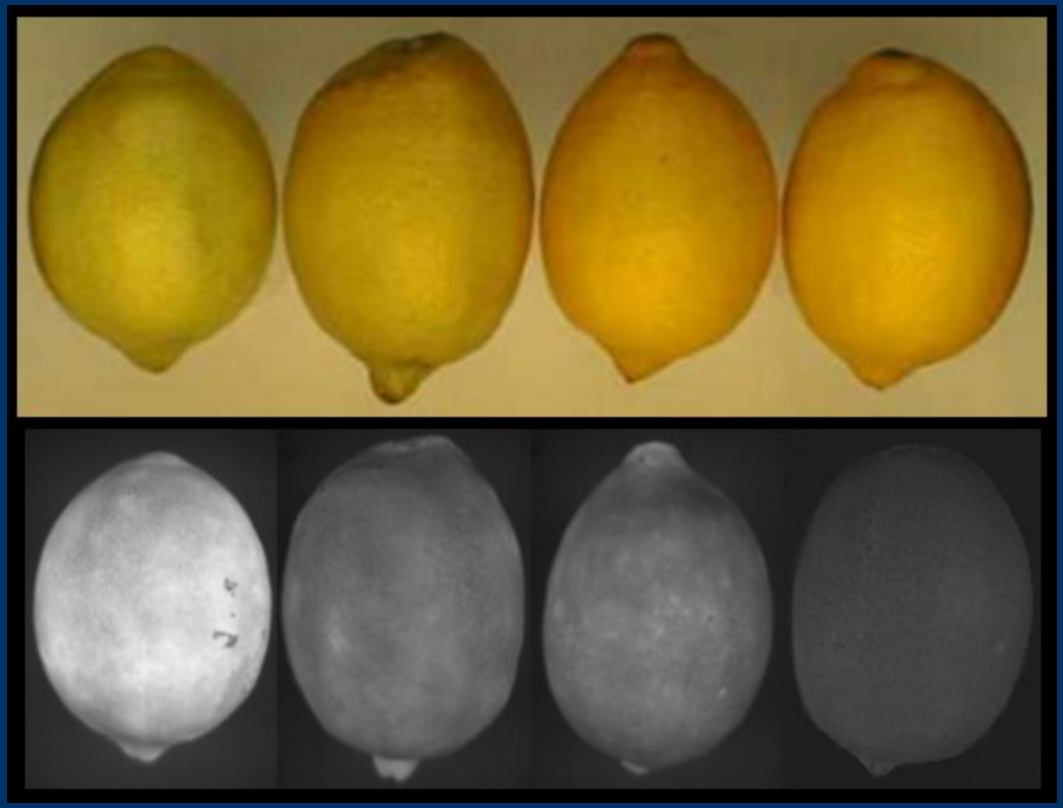
Microbiology and Molecular Biology Reviews

Campbell D et al. Microbiol. Mol. Biol. Rev. 1998;62:667-683

Chlorophyll fluorescence from ripe lemon fruits

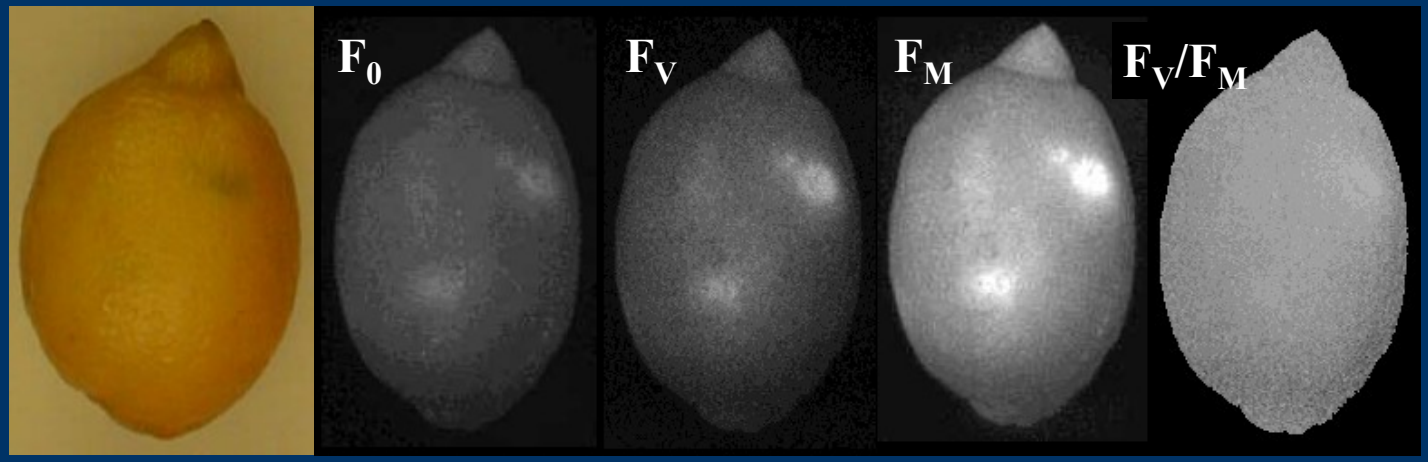
Color photograph

Fluorescence F_M image



Heterogeneous lemon pigmentation

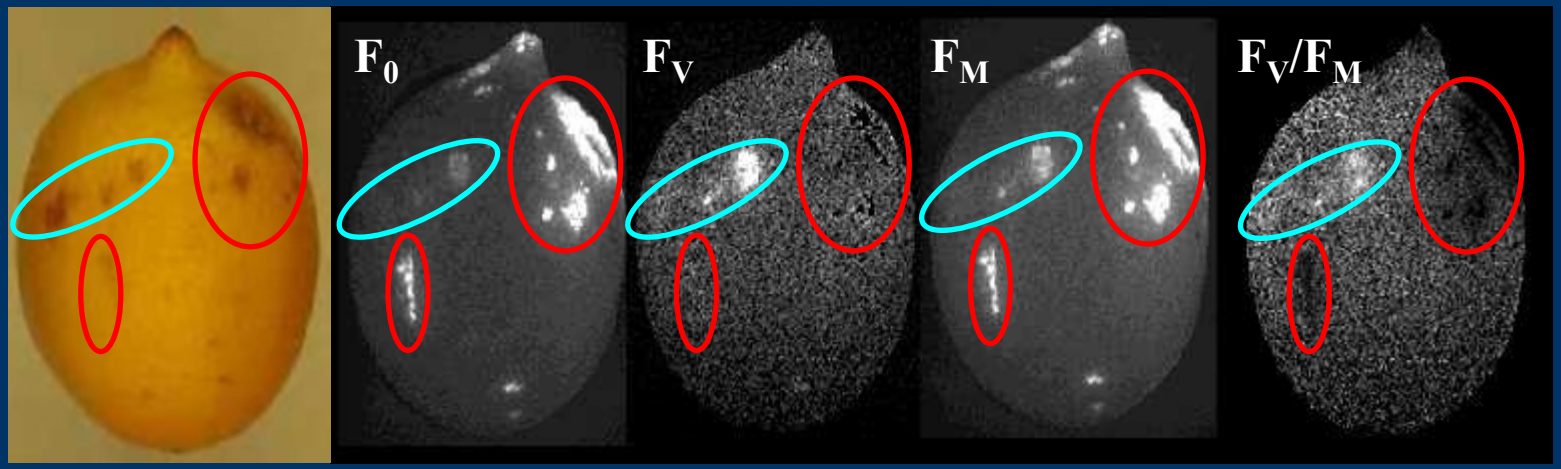
Color photograph



Fluorescence images

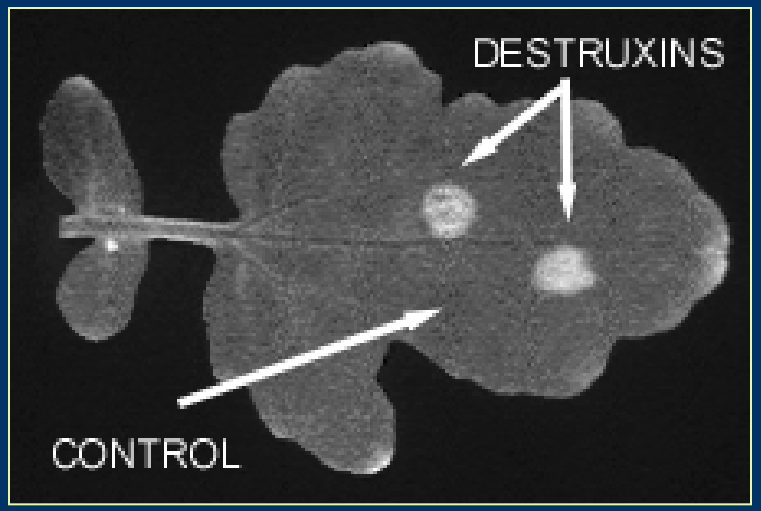
Post-harvest lemon damage

Color photograph

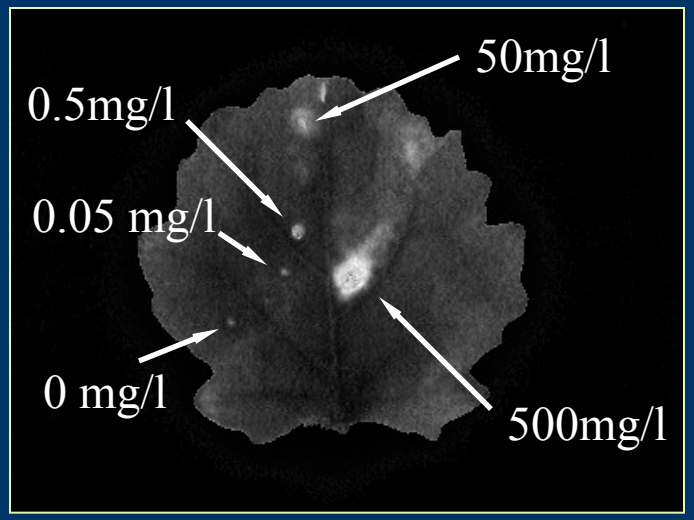


Fluorescence images

Phytotoxin response visualized by fluorescence

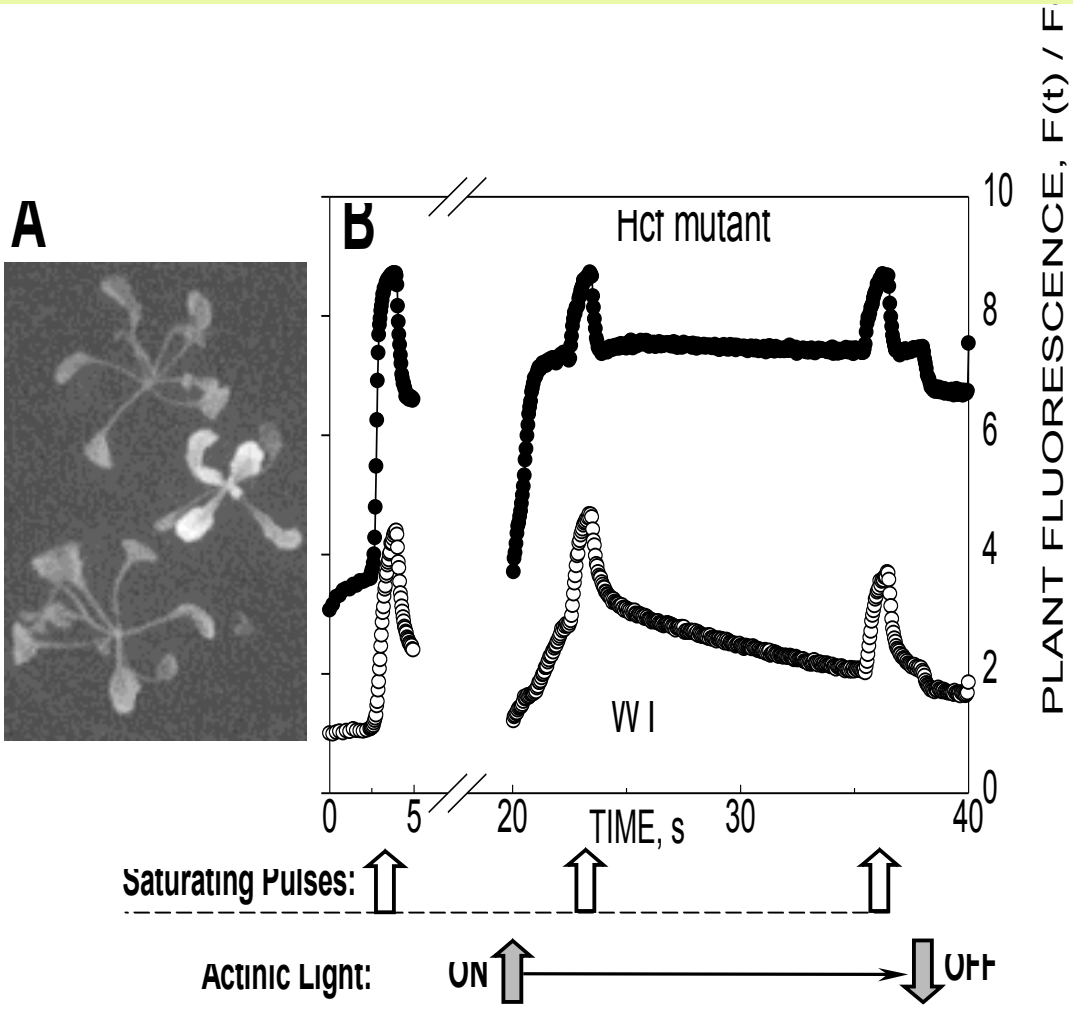


Sinapis alba
60 h, 2000 mg/l destruxin

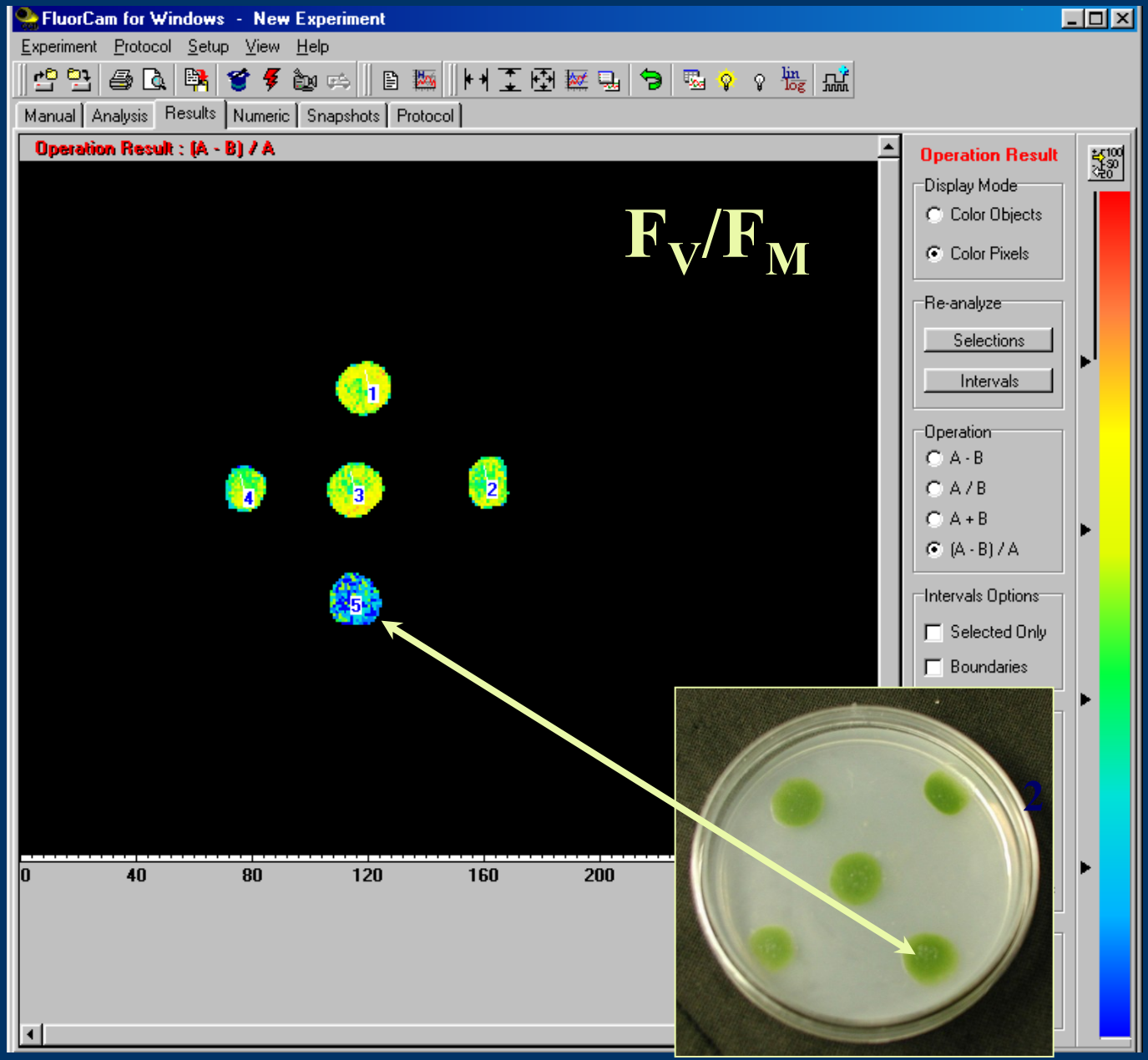


Brassica oleracea
60 h, 0-500 mg/l destruxin

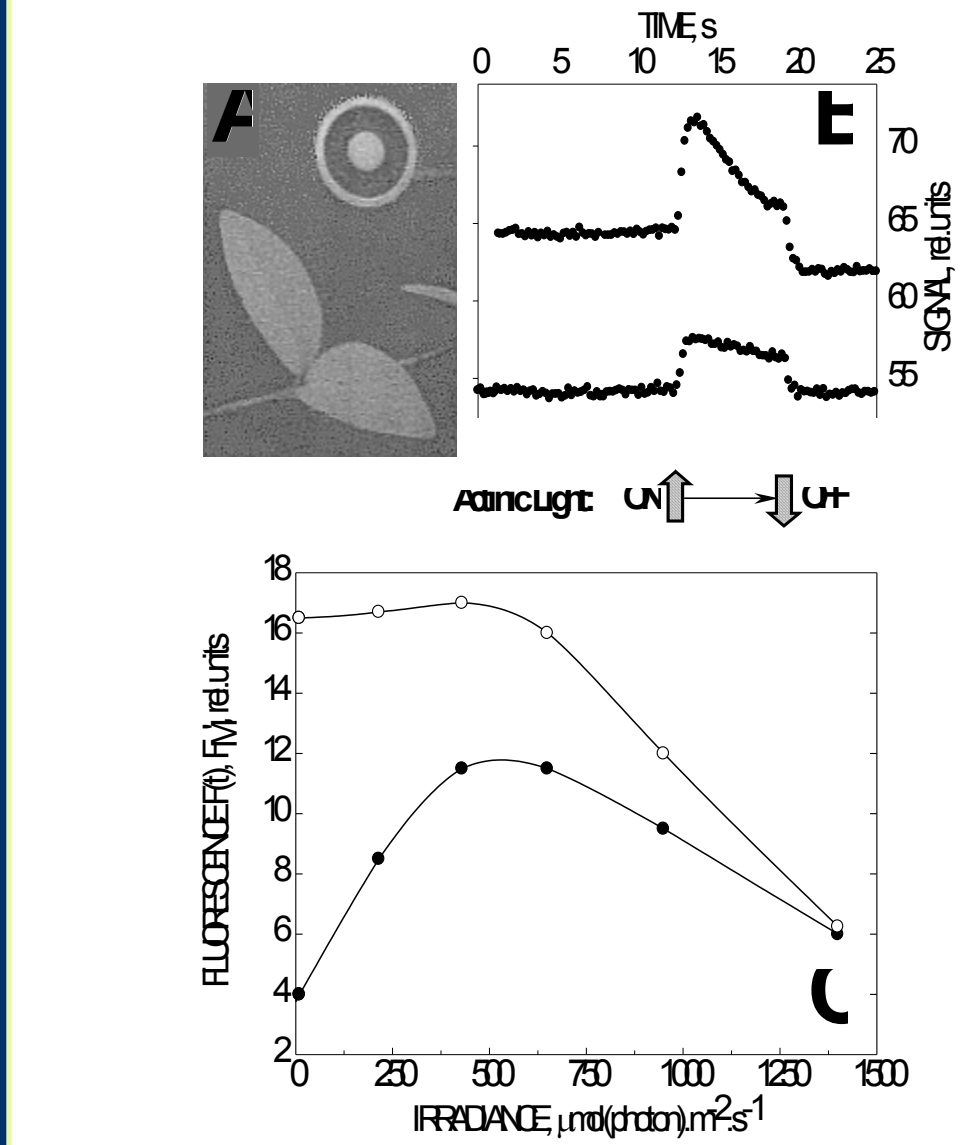
Mutant selection



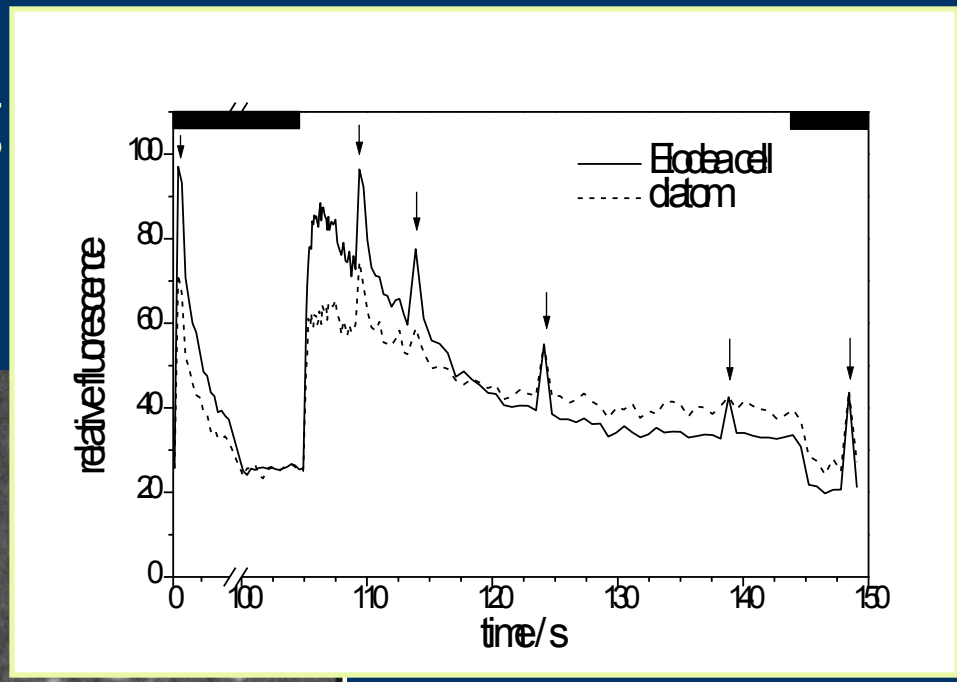
High-light stress sensitivity

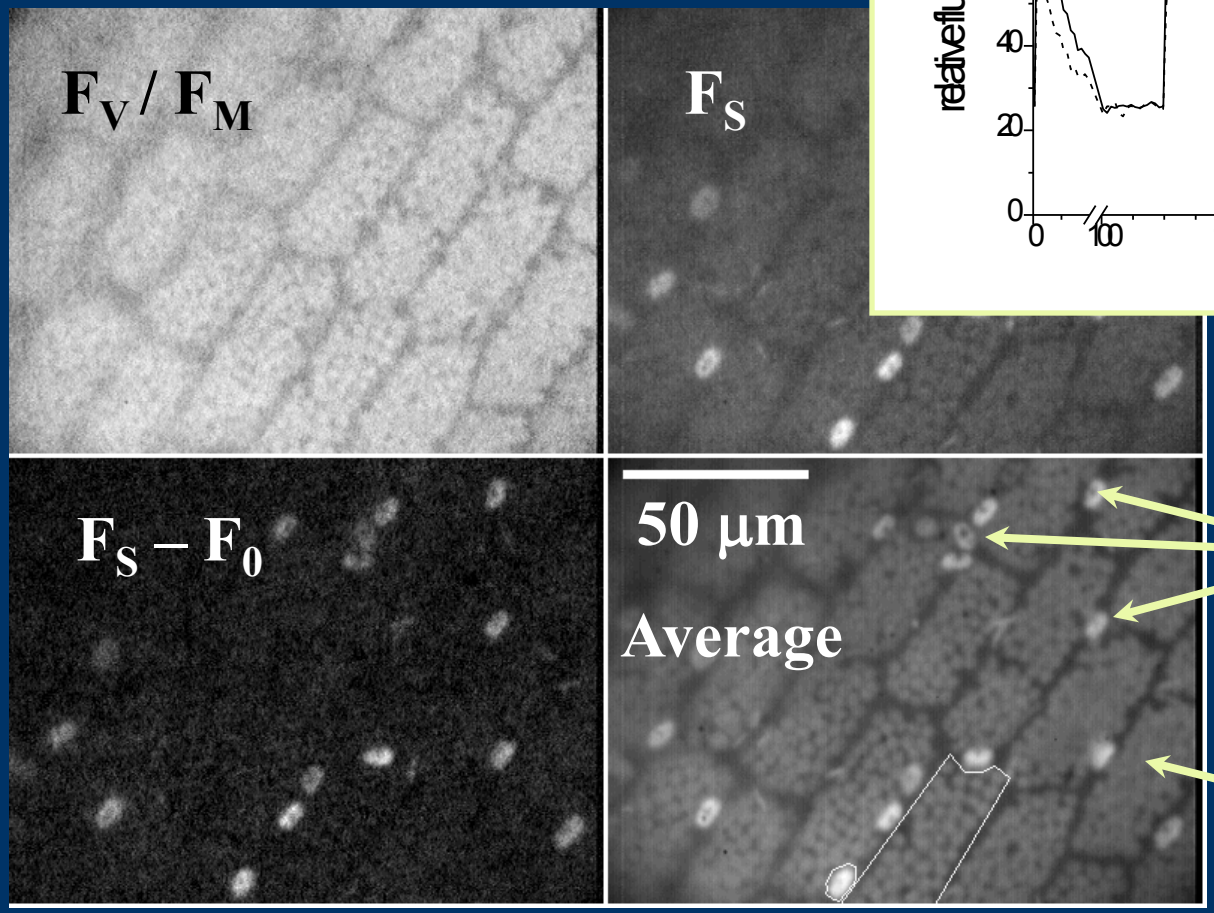


Field operation



Microscopic kinetic fluorescence imaging





diatoms

Elodea chloroplasts