Complexity of Chlorophyll Fluorescence Dynamic Response as an Indicator of Excessive Light Intensity ⋆

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Abstract: The controllability of LED lighting systems for greenhouses and plant factories offers a possibility for light induced diagnose of plant status. Here, a novel method for proximal remote detection of plant light tolerance is investigated. The method is based on an identification of a transfer function model for the measured chlorophyll fluorescence response to a small step variation in blue LED light. It is postulated that the least required model order decreases as the plants become light stressed due to saturation effects at excess light conditions. We apply this method to basil and lettuce plants under different background light intensities, and the results are compared to measured effective quantum yield (y(II)), relative electron transport rate through PSII (ETR(II)) and non-photochemical quenching (NPQ), all reflecting the photosynthetic performance. For both species it is indeed found that the required model order decreases with increasing background light intensity at the same time as the measured reference parameters indicates a decreased photosynthetic efficiency. It is suggested that the light intensity should be such that the chlorophyll fluorescence response requires a model order of 3 or higher to avoid ineffective irradiation of the plants.

Keywords: Photosynthesis, fluorescence, plant sensing, step function responses, dynamic behaviour, system identification, method development, feedback control.

1. INTRODUCTION

With the recent developments in high brightness LED technology, new lamps competing with the traditional high pressure sodium (HPS) lamps are being introduced to the greenhouse market. The high level of controllability of these systems offers a possibility for light induced diagnose of plant status based on chlorophyll fluorescence analysis.

Photosynthesis in plants is driven by light energy absorbed by chlorophyll molecules within the photosynthetic apparatus. The absorbed energy can be (i) used in photochemistry, (ii) dissipated as heat (NPQ), or (iii) be re-emitted as chlorophyll fluorescence (CF). The intricate connections between these three de-excitation pathways cause dynamics in the light-induced chlorophyll fluorescence signal. When a dark-adapted leaf is illuminated with continuous light, the chlorophyll fluorescence shows a transient response with a number of characteristic inflection points. This is called fluorescence induction and the inflection points are labelled OJIPSMT. During the initial fast phase of the induction curve (OJIP) chlorophyll fluorescence rises from the initial low origin level O, via the intermediate inflections J and I, to a peak level P in about 1 s, which is considered to reflect the successive reduction of the electron acceptor pool of PSII. During the following slow phase (PSMT), the fluorescence declines to a terminal steady-state level T in the time-scale of minutes. The decline is often accompanied by a local, semi steady-state, minimum S and a local maximum M, and sometimes more than one pair of S and M inflections (oscillations) can be observed (Walker, 1992; Walker et al., 1983). The slow phase is more difficult to interpret as many different processes linked to e.g. photochemical quenching (qP) and non-photochemical quenching (NPQ) are known to influence the signal. Given the strong connection to photosynthesis and the optical nature of the signal, chlorophyll fluorescence transient measurement is a sensitive and non-invasive tool, widely used for investigating physiological processes of the photosynthetic apparatus in vivo. See reviews by Papageorgiou et al. (2007a), Papageorgiou and Govindjee (2011) and Stirbet et al. (2014) and chapters in Papageorgiou et al. (2007b) for a deeper introduction and more references to the fast and slow chlorophyll fluorescence transients. The slow part of the fluorescence induction, i.e. the PSMT phase, is our main focus here.

The PSMT phase is less known compared to OJIP and challenging to extract reliable information from since the

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diverse processes involved cause ambiguity. At the same time it contains important information about the status of the plant, which is why many recent studies focus on dissecting the slow fluorescence. The characteristics of the PSMT phase of light induced fluorescence of dark-adapted photosynthetic organisms has shown to reflect in vivo photosynthetic processes. The decay P-to-S has, for example, been attributed to re-oxidation of plastoquinol (Munday and Govindjee, 1969) and induction of the reversible energy-dependent component (qE) of NPQ (Briantais et al., 1979). The succeeding rise S-to-M have been shown to coincide with a rise in O_2 evolution rate (Papageorgiou and Govindjee, 1968b,a), supporting the involvement of photochemical quenching. The S-to-M rise has also been linked to State 2 to State 1 transitions in cyanobacteria and green algae (Kodru et al., 2015). Other processes, such as the Calvin-Benson cycle, are considered to also come into play before reaching the steady-state level (Papageorgiou et al., 2007a). Takayama et al. (2012) studied the PSMT phase as a prospective candidate for plant diagnosis in greenhouses, trying to elucidate the contributions of qP and NPQ in mature tomato leaves. They measured y(II) (effective PSII quantum yield) and NPQ during fluorescence transients induced by weak excitation light (photosynthetic photon flux density (PPFD) less than $100\,\mu\text{mol/m}^2\text{/s}$), and concluded that the shape of the PSM transient is predominantly determined by qP (redox state of PSII electron acceptors), whereas qE strongly influences the shape after the M inflection. Bradbury and Baker (1981, 1984) have previously reported similar results for bean leaves, but also found that at strong excitation light ($1000\,\mu\text{mol/m}^2\text{/s}$) the S-to-M transient is lost and the monotonous quenching from P-to-T is dominated all the way by NPQ processes.

In Carstensen et al. (accepted) a new method for probing the photosynthetic apparatus, based on analysis of the varying dynamic chlorophyll fluorescence response to a relatively small excitation light, is suggested and investigated. In contrary to most commonly used chlorophyll fluorescence analysis techniques the fluorescence signal is detected *remotely* at *canopy level* and *in presence of background lighting*, making the method an interesting option for plant monitoring in greenhouses and plant factories for diagnostics or feedback control of the growth environment. As part of the experimental work rational output error (OE) transfer function models are identified to fit measured PSMT phases, triggered by adding a step excitation light to different background light intensities. For each step a new OE model is identified and the idea is that changes in the plant status will be captured as changes in the identified OE models. It was found that basil plants subjected to higher background light intensities required models of lower order as opposed to plants subjected to lower light intensities. It was also found that basil plants acclimated to different light intensities responded differently, indicating that the characteristics of the PSMT phase reflects the light tolerance of the plants. The theory postulated is that excess light causes saturation effects which effectively imply that states active at low light become obsolete at higher light.

The work presented here further investigates how the model order required to predict dynamic fluorescence responses varies with background light intensity and consequent light stress. The number of background light levels investigated have been increased from three to seven and lettuce is studied in addition to basil. The results are compared to reference measurements of effective quantum yield of photosystem II (y(II)), relative electron transport rate through PSII (ETR(II)) and non-photochemical quenching of fluorescence (NPQ), parameters often used to quantity photosynthetic status. This study is aiming to advance our understanding of plant specificity of fluorescence dynamics and loss thereof, and of the connection to the underlying physiological mechanisms.

As the plant is a highly non-linear and time dependent system, each model identified to a dynamic chlorophyll fluorescence response is to be considered a linear approximation of the system at that time. The method described here tracks the number of poles and zeros of the model required to describe the system at different times, rather than finding one general model to describe the system at all times.

2. MATERIAL AND METHODS

2.1 Plant material and growth conditions

Ocimum Basilicum (basil, Aroma 2) and *Lactuca Scariola* (lettuce, Simpson) were grown in a mixture of 2 parts of potting soil (Proveen Substrate, Mixture Semis Bouturage 2) and 1 part of Vermaculite (Agra). After having germinated under a cold fluorescent light the plants were moved to a light regime with a PPFD of $210\,\mu\text{mol/m}^2\text{/s}$ and an additional $26\,\mu\text{mol/m}^2\text{/s}$ of light within the far red spectral range ($700 - 800$ nm). The red-green-blue (R-G-B) ratio was 69-13-18, where blue is defined as $400 - 500$ nm, green $500 - 600$ nm and red $600 - 700$ nm. The photoperiod was 16 hours and the day/night temperatures were 22/17°C.

2.2 Experimental set up

Samples (one pot of basil or lettuce) were placed on a grate approximately 80 cm below two commercially available horticulture LED lamp (LX-series, Heliospectra, Sweden). These lamps have three different individually controllable LED-groups: blue (wavelength peak at 450 nm), red (wavelength peak at 660 nm), and white (colour temperature 5700K), and were used to provide the seven different background light regimes presented in Table 1. Each level was held for 10 minutes, and the levels were separated by a 10 minute dark period. The incoming light was measured at canopy level using a Maya2000 Pro (Ocean Optics, USA) spectrometer equipped via a 50 μm optical fibre, with a cosine corrector giving a 180° field of view, facing upwards towards the lamps.

Five minutes into each level an excitation signal in form of a step increase of $50\,\mu\text{mol/m}^2\text{/s}$ blue light (realised by control of the 450 nm LED-group in the lamps) was applied for a duration of 60 seconds. The excitation signal induced a change in the chlorophyll fluorescence signal. The induced chlorophyll fluorescence and reflected light was measured using another Maya2000 Pro spectrometer, mounted $16 - 18$ cm above the plant canopy, facing

downwards towards the canopy equipped, via a 600 μm optical fibre, with a Gershun tube giving a field of view of 20°. Both spectrometers had a sample rate of 1 Hz. Three repetitions of the experiment were made on different plant samples.

Table 1. Background light regimes

Level	PPFD (μmol/m^2/s)	R-G-B ratio
1	55	22-11-67
2	96	22-10-68
3	168	23-10-67
4	217	23-10-67
5	341	23-10-67
6	533	23-10-67
7	827	22-10-68

2.3 Data treatment

The incoming light spectra was integrated over the $400-700$ nm wavelengths to achieve an input signal $u(t)$ consisting of the incoming PAR as a function of time. The spectra of measured chlorophyll fluorescence and reflected light were integrated over $730-750$ nm, matching the fluorescence peak at 740 nm. This signal is referred to as the output signal $y(t)$ of the system.

To handle transients in the measured output signal, arising as a result of transitions from dark periods to light levels (rather than effects of the excitation signal), the signal was detrended. This was realized by fitting a second order exponential decay to a data set consisting of the 100 samples preceding the application of the excitation signal and the 100 last samples measured at the currently treated light level. The interpolation of the resulting function was then subtracted from the measured chlorophyll fluorescence response. Finally, data sets consisting of 20 samples taken before excitation and 60 samples taken during excitation were selected from the input and output signals on each light intensity level for identification.

2.4 Model fitting

The OE models were identified individually to each excitation step input and output response, using MATLAB's System Identification Toolbox. The models are written

$$y(t) = \frac{B(q^{-1})}{F(q^{-1})}u(t) + e(t), \tag{1}$$

where $y(t)$ is the measured output, $u(t)$ is the measured input and $e(t)$ is a disturbance. B and F are polynomials in the time shift operator q^{-1}. The order of these polynomials determines the number of poles and zeros of the model. In this work only models with equal number of poles and zeros are investigated, and the term *model order* is referring to that number.

The aim of the model fitting was to find the lowest possible model order that could predict the dynamic response of the chlorophyll fluorescence in a *satisfactory way*. This meant that the simulation should (i) have a high fit percentage to measured data and (ii) capture the main characteristics of the dynamic fluorescence response, i.e. the pattern of the PSMT phase of the induced fluorescence curve. Higher model order that results in better fitting percentage as a result of modelling of disturbances were visually identified and discarded. The performance of five model orders were evaluated for each data set, with the first one having 1 pole and 1 zero, the second one having 2 poles and 2 zeros and so forth. The required model order reflects the complexity of the dynamic fluorescence responses at different light intensity levels.

2.5 Reference measurements

Reference measurements of photosynthetic parameters were taken on a fully developed leaf from the top layer of the sample canopy using a Dual-PAM-100 Measuring System (Heinz Walz GmbH, Germany). Three repetitions were made on different samples. The instrument was programmed to mimic the light regimes in Table 1 (excluding the addition of the excitation signal) and dark periods used in the previously described experimental setup. Effective quantum yield (y(II)), electron transport rate (ETR(II)) and non-photochemical quenching (NPQ) were measured every 20 seconds for the first three minutes after each light intensity change, and every 60 seconds thereafter.

Electron transport rate light response curves were obtained by plotting the ETR(II) value measured five minutes into each light level versus the PPFD of the incoming light. The potential ETR(II), reflecting maximal photosynthetic quantum yield (Papageorgiou et al., 2007b), was estimated by extrapolation of the slope between the two lowest light intensity levels investigated.

3. RESULTS

3.1 Dynamic fluorescence responses and estimated models

Measured (and detrended) dynamic responses and their corresponding model simulations for three selected light levels of one repetition of basil and one repetition of lettuce are presented in Fig. 1 and 2, respectively. Three types of PSMT patterns were observed under low, mid and high range light intensities, which are exemplified in Fig. 1 and 2 by level 2, level 5 and level 7. The dynamic responses of lettuce and basil were in principle similar, but the change in characteristics of the PSMT pattern occurred at different light intensities for the two different species. Under low range light intensities the P, S, and M parts of the dynamic fluorescence response are distinguishable. This was observed for light intensities up to 217 μmol/m^2/s. For mid range light intensities no S-to-M rise is seen, instead the peak P and the maximum M appears to merge into one "slow" maximum. For lettuce this was first observed at the 341 μmol/m^2/s level, consistent for all three repetitions. For basil it was observed on the 217 μmol/m^2/s level, on the 341 μmol/m^2/s level, and on the 533 μmol/m^2/s level in the three different repetitions. For high range light intensities the peak P is immediately followed by an exponential decay. For lettuce this behaviour was first observed on the 533 μmol/m^2/s level, whilst for basil it was first observed at 827 μmol/m^2/s in two of the repetitions. For the high light intensity range disturbances in the output signal were more prominent. The signal to noise ratio was decreased, which is a result of the fixed excitation amplitude in relation to the increasingly higher background

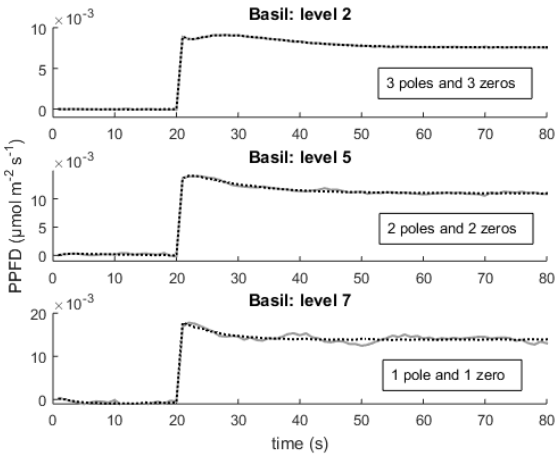

Fig. 1. Measured, detrended fluorescence responses (solid gray) for basil at three selected light levels and the corresponding simulated signals (dashed black).

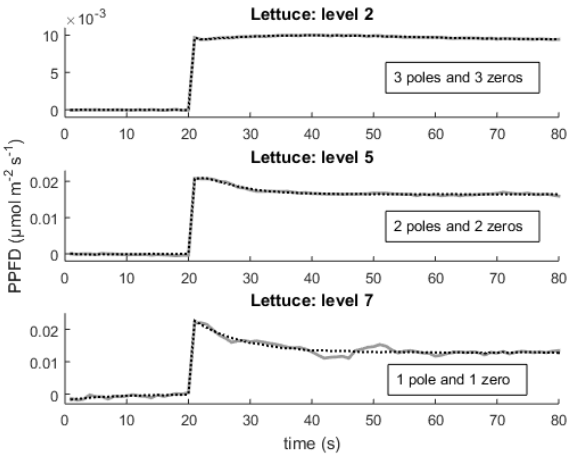

Fig. 2. Measured, detrended fluorescence responses (solid gray) for lettuce at three selected light levels and the corresponding simulated signals (dashed black).

irradiation. Furthermore, oscillation-like behaviour of the fluorescence signal was observed for these light intensities.

The model order required (Table 2 and 3) differed between the three light intensity ranges. The low range light levels (coloured light gray) required a model of order 3 or more. For midrange light levels (darker gray) the model order could be decreased to 2. For the high light intensity range (darkest gray) the exponential decay of the responses could be simulated with a model of order 1. Higher model orders resulted in undesired modelling of the disturbances. In two of the lettuce repetitions the disturbances in the output signal at the highest light level were too prominent, and no suitable model could be found. This is indicated with an "X" in Table 3.

Table 2. Model order required for basil

Level	PPFD (μmol/m^2/s)	Rep. 1	Rep. 2	Rep. 3
1	55	4	3	3
2	96	3	3	3
3	168	4	3	3
4	217	3	2	3
5	341	3	2	2
6	533	2	1	2
7	827	1	2	1

Table 3. Model order required for lettuce

Level	PPFD (μmol/m^2/s)	Rep. 1	Rep. 2	Rep. 3
1	55	5	4	3
2	96	3	3	3
3	168	3	3	3
4	217	3	3	3
5	341	2	2	2
6	533	1	1	1
7	827	X	1	X

3.2 Reference measurements

The measured reference parameters for basil and lettuce are presented in Fig. 3 and 4. The effective quantum yield of photosystem II (y(II)) decreased with increasing light intensity level. The relative electron transport rate through PSII (ETR(II)) on the other hand increased with

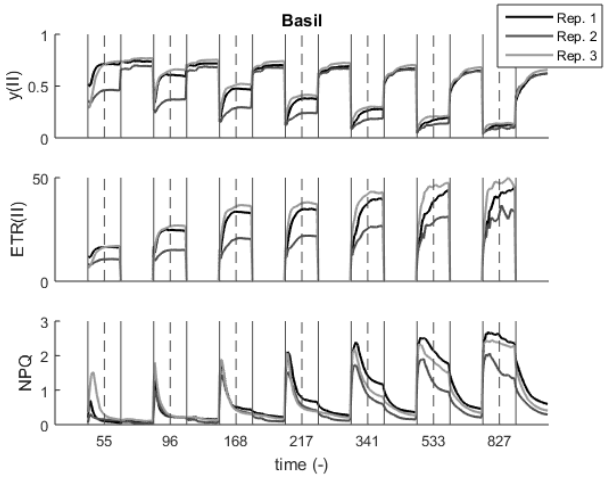

Fig. 3. Reference parameters for three repetitions of basil. The x-axis shows unitless time but is labelled with the different light intensities used. The vertical, solid, gray lines indicates light switches occurring 10 minutes apart.

increasing light level, but saturated to some extent for the mid and high range light intensities. This saturation is clearly visible in Fig. 5, where the ETR(II) level five minutes into each light level is plotted versus light intensity. This figure also shows the potential ETR(II). As the ETR(II) saturates, its deviation from the potential ETR(II) increases drastically. The NPQ (Fig. 3 and 4) was instantly increased upon each change in light level followed by a relaxation. This relaxation was fast at the low range light intensities, where it was almost completely relaxed after five minutes, and slow to nonexistent at mid and high range light intensities.

3.3 Discussion

The development of the measured fluorescence responses with respect to increased light intensities was similar for basil and lettuce. The modelling of the dynamic responses required different model orders at different background light intensities. Based on the required model orders a

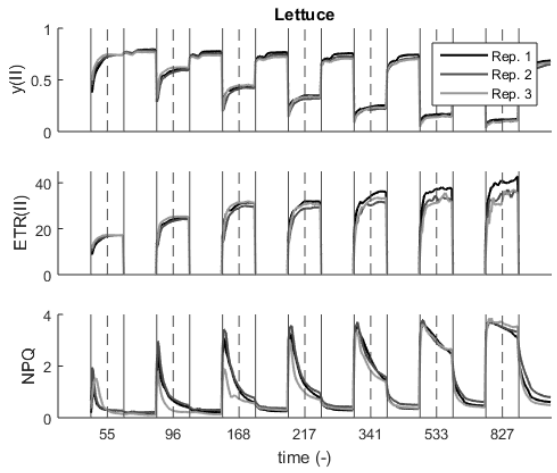

Fig. 4. Reference parameters for three repetitions of lettuce. The x-axis shows unitless time but is labelled with the different light intensities used. The vertical, solid, gray lines indicates light switches occurring 10 minutes apart.

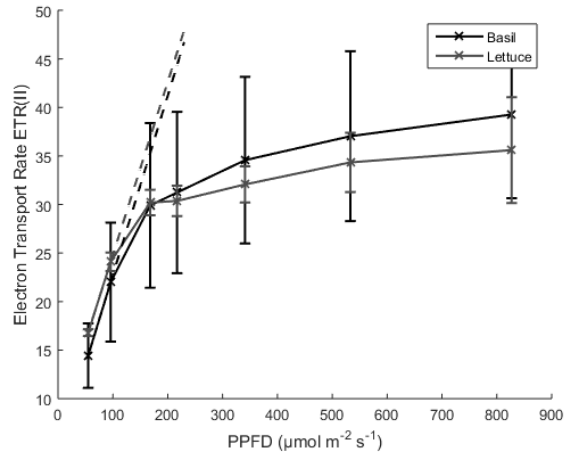

Fig. 5. Electron transport rate light response curve, showing measured (solid) and potential (dashed) ETR(II) for basil and lettuce for different light intensities.

classification of dynamic responses into low, mid and high light intensity ranges was motivated, referring to the background light intensities at which these transients were collected. This classification has interesting connections to the plant physiology at the different background light intensities as seen through the reference measurements.

Within the low light intensity range, covering the light intensities up to the growth light intensity, the y(II) and ETR(II) depends almost linearly on the background light intensity. Within this range the ETR(II) lies fairly close to its potential, which can be considered an indication of low excessive light energy (Papageorgiou et al., 2007b). Furthermore, within this range the NPQ relaxes quickly and to a lower value compared to at light intensities above this range. In the low light intensity range the relaxation of NPQ is completed within the first five minutes after a change in light level, indicating that there is a large potential for the NPQ to change in response to the excitation signal applied (five minutes into each light intensity level) in the remote sensing experiments. This potential of fast response in the NPQ is possibly one explanation of the high complexity of the fluorescence transient response seen in the low light intensity range. Hence, the model order (3 or more poles and zeros) needed at these light intensities, is probably connected to the plants ability to cope with the light intensities within this range.

Within the mid light intensity range both the y(II) and ETR(II) are saturating and the ETR(II) is deviating from its potential. The deviation from the ETR(II) potential indicates that the light energy exceeds the plants' capacity of light utilization (Papageorgiou et al., 2007b). This is consistent with the increase in NPQ observed within this range, indicating that the plants are dissipating more energy as heat as a protective mechanism. Furthermore, within this range the relaxation of the NPQ is slower, indicating a limited potential of fast NPQ response that could be seen as a saturation. Saturations within the system are possibly part of the explanation of the less complex dynamics in the PSMT transient. Hence, the decrease in model order to 2 poles and zeros for this light level range is related to the plants decreased capacity of utilizing the light. Also notable is that Bradbury and Baker (1981) refers to the S-to-M rise for dark-adapted plants as an indication of non-saturating intensities being observed. Assuming that it plays a similar role for the light-adapted plants investigated here, the absence of S-to-M rise for the mid range light intensities might indicate saturating intensities.

Within the high light intensity range the relaxation of the NPQ is almost non-existent. Hence, the plants potential of fast NPQ response to the excitation light is expected to be very low at these light intensities. This coincides with the decreased complexity of the PSMT transient implying that the decreased model order needed at high light intensities is related to saturated fast NPQ and excessive light. The disturbance in the chlorophyll fluorescence signal at these light levels may be partly attributed to SM oscillations, as mentioned in the introduction. Such oscillations can be induced by sudden external perturbations (Stirbet et al., 2014), in this case abrupt high intensity re-illumination. The underlying mechanisms are not well understood but processes that regulate the Calvin-Benson cycle might be involved (Stirbet et al., 2014), which makes the disturbance an interesting target for further research.

Recently, PSMT patterns of dark-adapted bean leaves subjected to different levels of heat stress were reported (Levykina and Karavaev, 2016) with results similar to ours. For example, the time until the second maxima (M) occurred decreased gradually with leaf temperature and the curve acquired a unimodal character at high temperatures. The relative fluorescence quenching $((F_P - F_T)/F_T)$ was identified as a suitable slow fluorescence induction parameter as it was responsive to the treatments and has been reported in the literature to be linked to photochemical activity and CO_2 assimilation (Tuba et al., 1994). In the case of light-adapted plants the rise to P, which is attributed to reduction of primary electron acceptors of PSII (Krause and Weis, 1991), is compromised as the reaction centres of the PSII are not fully open prior to the excitation. This makes the relative

fluorescence quenching an unsuitable parameter for light-adapted plants. Instead of using the relationship between two points of the induction curve, the method deployed in this work take the entire response into account, which is arguably more robust and potentially more informative.

4. CONCLUSION

In this study it was explored how the complexity of light induced chlorophyll fluorescence transients relates to the light intensity plants are exposed to and how it reflects the plants photosynthetic status. The study was performed on basil and lettuce under seven different background light intensities. The complexity was studied by identifying transfer function models of different orders estimating the relation between remotely measured fluorescence transients and a small step variation in blue incoming LED light. It was found that for both basil and lettuce the model order required in to capture the dynamics of the fluorescence transient decreased with increasing background light intensity. This loss of complexity coincided with a saturation in the capacity of photosynthesis, seen as a saturation of ETR(II), as well as with increased photoprotection, seen as an increase in NPQ.

Based on these results it is suggested that the light intensity should be such that the chlorophyll fluorescence response requires a model order of 3 to avoid ineffective irradiation of the plants. The method investigated here shows potential of tracking photosynthetic performance in basil and lettuce remotely and could possibly be deployed in a feedback control system regulating the growth environment in greenhouses and plant factories.

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REFERENCES

Bradbury, M. and Baker, N.R. (1981). Analysis of the slow phases of the in vivo chlorophyll fluorescence induction curve. Changes in the redox state of Photosystem II electron acceptors and fluorescence emission from Photosystems I and II. *BBA - Bioenergetics*, 635(3), 542–551. doi:10.1016/0005-2728(81)90113-4.

Bradbury, M. and Baker, N.R. (1984). A quantitative determination of photochemical and non-photochemical quenching during the slow phase of the chlorophyll fluorescence induction curve of bean leaves. *BBA - Bioenergetics*, 765(3), 275–281. doi: 10.1134/S0006297914040014.

Briantais, J.M., Vernotte, C., Picaud, M., and Krause, G.H. (1979). A quantitative study of the slow decline of chlorophyll *a* fluorescence in isolated chloroplasts. *Biochimica et biophysica acta*, 548(1), 128–138. doi: 10.1016/0005-2728(79)90193-2.

Carstensen, A.M., Pocock, T., Bånkestad, D., and Wik, T. (accepted). Remote detection of light tolerance in basil through frequency and transient analysis of light induced fluorescence. *Computers and Electronics in Agriculture*.

Kodru, S., Malavath, T., Devadasu, E., Nellaepalli, S., Stirbet, A., Subramanyam, R., and Govindjee (2015). The slow S to M rise of chlorophyll *a* fluorescence reflects transition from state 2 to state 1 in the green alga *Chlamydomonas reinhardtii*. *Photosynthesis Research*, 125(1-2), 219–231. doi:10.1007/s11120-015-0084-2.

Krause, G.H. and Weis, E. (1991). Chlorophyll fluorescence and photosynthesis: The basics. *Annual Review of Plant Physiology and Plant Molecular Biology*, 42(1), 313–349. doi:10.1146/annurev.pp.42.060191.001525.

Levykina, I.P. and Karavaev, V.A. (2016). Fluorescence induction changes in bean leaves after heat treatment. *Moscow University Physics Bulletin*, 71(1), 112–118. doi:10.3103/S0027134916010124.

Munday, J.C. and Govindjee (1969). Light-induced changes in the fluorescence yield of chlorophyll *a* in vivo. III. the dip and the peak in the fluorescence transient of *Chlorella pyrenoidosa*. *Biophysical Journal*, 9(1), 1–21.

Papageorgiou, G.C., Tsimilli-Michael, M., and Stamatakis, K. (2007a). The fast and slow kinetics of chlorophyll *a* fluorescence induction in plants, algae and cyanobacteria: a viewpoint. *Photosynthesis Research*, 94(2-3), 275–290. doi:10.1007/s11120-007-9193-x.

Papageorgiou, G. and Govindjee (2011). Photosystem II fluorescence: slow changes – scaling from the past. *Journal of Photochemistry and Photobiology B: Biology*, 104(1-2), 258–270. doi:10.1016/j.jphotobiol.2011.03.008.

Papageorgiou, G. and Govindjee (1968a). Light-induced changes in the fluorescence yield of chlorophyll *a* in vivo. I. *Anacystis nidulans*. *Biophysical Journal*, 8(11), 1299–1315. doi:10.1016/S0006-3495(68)86557-9.

Papageorgiou, G. and Govindjee (1968b). Light-induced changes in the fluorescence yield of chlorophyll *a* in vivo. II. *Chlorella pyrenoidosa*. *Biophysical Journal*, 8(11), 1316–1328. doi:10.1016/S0006-3495(68)86558-0.

Papageorgiou, G.C. et al. (2007b). *Chlorophyll a fluorescence: a signature of photosynthesis*, volume 19. Springer Science & Business Media.

Stirbet, A., Riznichenko, G.Y., Rubin, A.B., and Govindjee (2014). Modeling chlorophyll *a* fluorescence transient: relation to photosynthesis. *Biochemistry (Moscow)*, 79(4), 291–323. doi: 10.1134/S0006297914040014.

Takayama, K., Miguchi, Y., Manabe, Y., Takahashi, N., and Nishina, H. (2012). Analysis of Ψ_{PSII} and NPQ during the slow phase of the chlorophyll fluorescence induction phenomenon in tomato leaves. *Environmental Control in Biology*, 50(2), 181–187. doi: 10.2525/ecb.50.181.

Tuba, Z., Lichtenthaler, H.K., Csintalan, Z., Nagy, Z., and Szente, K. (1994). Reconstitution of chlorophylls and photosynthetic CO2 assimilation upon rehydration of the desiccated poikilochlorophyllous plant xerophyta scabrida (Pax) Th. Dur. et Schinz. *Planta*, 192(3), 414–420. doi:10.1007/BF00198578.

Walker, D.A. (1992). Concerning oscillations. *Photosynthesis Research*, 34(3), 387–395. doi: 10.1007/BF00029813.

Walker, D.A., Sivak, M.N., Prinsley, R.T., and Cheesbrough, J.K. (1983). Simultaneous measurement of oscillations in oxygen evolution and chlorophyll *a* fluorescence in leaf pieces. *Plant Physiology*, 73(3), 542–549.