

MEASURING SUN-INDUCED CHLOROPHYLL FLUORESCENCE: AN EVALUATION AND SYNTHESIS OF EXISTING FIELD DATA

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ABSTRACT

In this contribution we examined and synthesized a wide dataset of sun-induced canopy level chlorophyll fluorescence measurements collected on the ground in the last 10 years across a range of temporal scales and a range of species and plant functional types. Field measurements have been collected with a state-of-the-art spectrometer setup and standardized methodology, making the comparison of datasets collected at different times and locations easier. Results show that different land cover classes and forest species are characterized by different fluorescence magnitude, with generally the highest fluorescence emissions in crops followed by broadleaf and then needleleaf species.

Successful examples of the use of sun-induced chlorophyll fluorescence for the characterization of the functional status of vegetation are then discussed.

Fluorescence at canopy level was used as an indicator of plant physiological status in the context of control – treatment experiments. Sun-induced fluorescence at the oxygen A-band was proven effective in tracking photosynthesis declines in plants exposed to stress agents (i.e. DCMU and ozone).

Finally, fluorescence was successfully used to improve the estimation of gross primary production using light-use efficiency models in croplands and grasslands.

1. INTRODUCTION

Sunlight absorbed by chlorophyll in plants is mostly used to drive photosynthesis, but energy exceeding the photosynthetic demand can be dissipated as heat or re-radiated at longer wavelengths (660–800 nm). This near-infrared light re-emitted from plants under daylight is termed sun-induced fluorescence. Fluorescence has been found to be strongly correlated to photosynthesis

and for this reason it can be used to monitor the spatial and temporal dynamics of vegetation productivity and vegetation physiological responses during the onset of stress [1-3].

In the last years a wide dataset of sun-induced canopy level chlorophyll fluorescence has been collected through hyperspectral remote sensing observations acquired on the ground across a range of temporal scales and a range of species and plant functional types. Estimating fluorescence from hyperspectral observations requires “ultra-fine” spectral resolution (e.g., 0.1 nm full width at half maximum) in order to characterize very narrow atmospheric absorption features. Such resolution can be achieved by commercial spectrometers at the expenses of the width of the range being investigated [4, 5].

In this contribution we examine and synthesize this dataset of sun-induced canopy level chlorophyll fluorescence collected in the context of different national and international projects, including several ESA funded projects in support to the development of the FLEX mission.

The presented data are of interest for the analysis of vegetation status and functioning as well as the validation of the fluorescence maps derived from the airborne high performance imaging spectrometer HyPlant developed by the Forschungszentrum Jülich and the Finnish company Specim.

2. FLUORESCENCE ESTIMATION

Far-red fluorescence (F_{760}) has been measured with passive spectrometric systems exploiting the atmospheric oxygen absorption band O₂-A positioned at 760.4 nm. Fluorescence has been quantified by measuring the infilling by fluorescence compared to an

unperturbed reference signal using spectral fitting methods. Canopy leaving radiances were measured with two portable HR4000 spectrometers operating in the visible and near-infrared region with different spectral resolution. The first instrument covers the 400-1000 nm spectral range with a full width at half maximum (FWHM) of 1 nm and allows the computation of different vegetation indices and incident Photosynthetic Photon Flux Density (PPFD). The other spectrometer is specifically intended for fluorescence measurements and cover the spectral regions centered on the O₂-A bands with a finer resolution (FWHM = 0.1 nm). The average canopy plane was always observed from nadir with bare fibers (field of view of 25°) at a distance between 100 and 450 cm from the top of the canopy.

Spectral data were acquired with a dedicated software [6]. Spectrometers were spectrally calibrated with a known standard (CAL-2000 mercury argon lamp, OceanOptics, USA) and radiometrically cross-calibrated to a recently calibrated FieldSpec FS FR spectrometer (ASD, USA).

The collected spectral database was processed with an IDL (ITTvis IDL 7.1.1®) application developed for this purpose. The basic processing of raw data includes: correction for CCD detector non linearity; DC subtraction and correction for DC drift using optically black pixels; wavelength calibration and linear resampling; radiance calibration; incident radiance computation by linear interpolation of two “sandwich” measurements.

F₇₆₀ was estimated by exploiting the spectral fitting method (SFM) described in [4, 7], assuming a linear variation of reflectance and fluorescence in the O₂-A absorption band region. The spectral interval used for F₇₆₀ estimation was set to 759.00 - 767.76 nm for a total of 439 spectral channels used.

3. FLUORESCENCE VARIATIONS BETWEEN SPECIES

High resolution top-of-canopy spectral data have been collected from 2005 to present on a number of different crops (corn, rice, alfalfa, sugar beet and sorghum), grasslands, broadleaf (oak, hornbeam, linden and maple) and needleleaf (spruce and pine) forest species. An overview of the field campaigns performed from 2005 on is shown in Figure 1.

Maximum F₇₆₀ values at maximum canopy development ranged from 0.5 mW m⁻² sr⁻¹ nm⁻¹ for spruce forests to 2 mW m⁻² sr⁻¹ nm⁻¹ for broadleaf forests and 2.5-3 mW m⁻² sr⁻¹ nm⁻¹ for crops with alfalfa displaying the highest values among the investigated crops (e.g., sugar beet, maize, sorghum).

F₇₆₀ values higher than 3 mW m⁻² sr⁻¹ nm⁻¹ were measured only in a manipulation experiment consisting in the treatment of a grassland carpet with a solution of DCMU. In this case the maximum F₇₆₀ value recorded was 5 mW m⁻² sr⁻¹ nm⁻¹ for a PPFD of about 1400 μmol m⁻² s⁻¹.

4. FLUORESCENCE FOR EARLY STRESS DETECTION

Different control – treatment experiments were set up to prove the possibility to use F₇₆₀ measured with field spectrometers for early stress detection.

Plants of O₃-sensitive white clover (*Trifolium repens* L. cv. Regal) were exposed to chronic O₃ fumigation (100 ppb O₃, 5 h d⁻¹) for 3 weeks in a controlled environment fumigation facility. Control plants were maintained under the same experimental conditions as O₃-treated plants, but exposed to charcoal-filtered air [8].

Six diurnal cycles of optical properties and leaf physiological measurements were acquired under natural solar illumination. Traditional remote sensing indices (normalized difference vegetation index NDVI, [9]) detected differences between control and treated samples only at the end of the experiment (21 days after

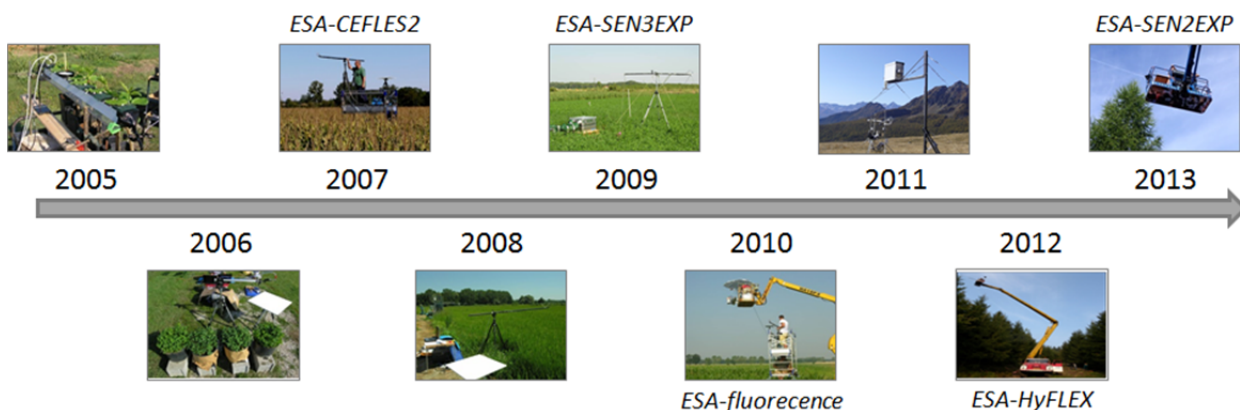


Figure 1. Overview of the field campaigns dedicated to fluorescence estimation performed from 2005 to present. These campaigns have been funded in the context of different national and international projects, including several ESA funded projects.

the beginning of the fumigation treatment) while F_{760} was lower in treated samples from day 1 on. The Photochemical reflectance index (PRI, [10]) tracking the excess energy dissipated as heat was reduced in treated samples (greater de-epoxidation of xanthophyll-cycle pigments) from day 3 on.

As an example, the last diurnal cycle measurements of PPF, leaf assimilation measured with CIRAS PP-System and F_{760} are reported (Figure 2). Full and empty symbols refer to control and ozonated canopies, respectively.

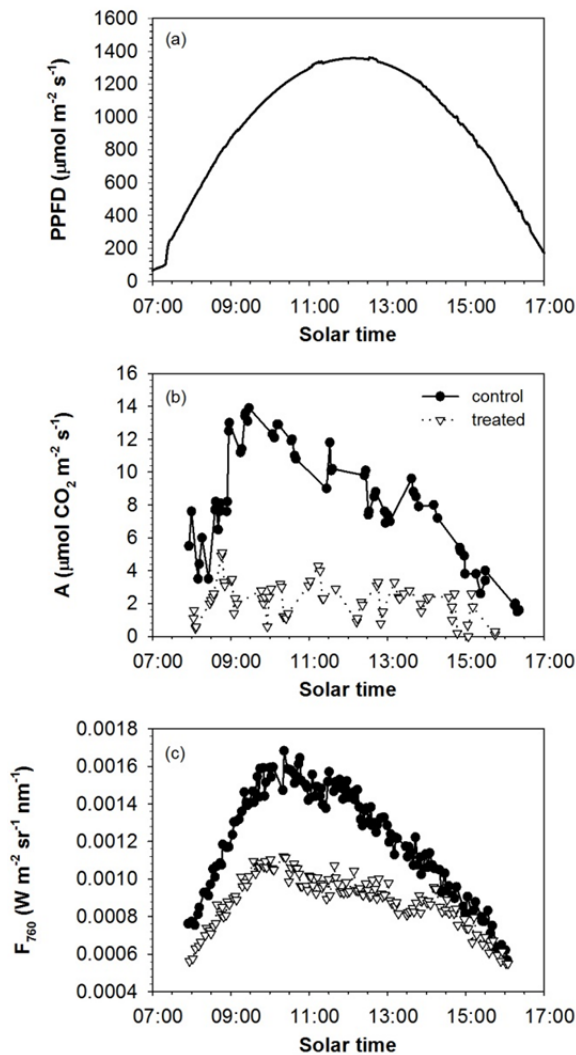


Figure 2. Diurnal evolution of (a) incident Photosynthetic Photon Flux Density (PPFD, $\mu\text{mol m}^{-2} \text{s}^{-1}$), (b) leaf assimilation (A , $\mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$) and (c) sun-induced chlorophyll fluorescence measured at 760 nm (F_{760} , $\text{W m}^{-2} \text{sr}^{-1} \text{nm}^{-1}$). Full and empty symbols refer to control and ozonated samples, respectively.

A second control – treatment experiment was performed in 2012 treating commercial grass with 3-(3,4-dichlorophenyl)-1,1-dimethylurea (DCMU). Two lawn carpets were laid on plastic sheets. One carpet was

treated with DCMU. The control carpet was treated at the same time with ethanol/water without the herbicide. DCMU blocks energy transfer in photosynthesis without changing the pigments and thus is known to increase fluorescence. The application of DCMU had no effect on reflectance and traditional vegetation indices while caused an increase of F_{760} up to $5 \text{ mW m}^{-2} \text{sr}^{-1} \text{nm}^{-1}$, more than the double of the control carpet.

5. FLUORESCENCE FOR THE ESTIMATION OF CROP AND GRASSLAND PRODUCTIVITY

In three terrestrial ecosystems (rice field, alfalfa crop and alpine grassland) spectral observations were collected in parallel with eddy covariance (EC) flux towers measurements providing half-hourly measurements of gross primary production (GPP). The availability of EC measurements allowed to investigate the possibility of monitoring GPP of terrestrial ecosystems from high spectral resolution field spectroscopy measurements based on a Light Use Efficiency (LUE, [11]) model:

$$GEP = \varepsilon \cdot fAPAR \cdot PAR_i = \varepsilon \cdot APAR \quad (1)$$

where the light-use efficiency (ε) represents the conversion efficiency of absorbed energy to fixed carbon, $fAPAR$ is the fraction of photosynthetically active radiation absorbed by vegetation, PAR_i is the incident photosynthetically active radiation and $APAR$ is the incident photosynthetically active radiation absorbed by vegetation. All the parameters in the LUE model can be in principle derived directly from remotely sensed measurements.

We used the well-known normalized difference vegetation index (NDVI, [9]) as a proxy of the $fAPAR$ component that is effectively used for photosynthesis (photosynthetic $fAPAR$). As a proxy of photosynthetic canopy $APAR$ we used F_{760} as suggested in recent studies [12]. To estimate ε we investigated the use of two optical signals: the Photochemical Reflectance Index (PRI, [10]) and the apparent fluorescence yield at 760 nm (Fy_{760}), computed as the ratio between F_{760} and PAR_i . Spectral indexes were computed from the canopy reflectance spectra measured between 11.00 a.m. and 1.00 p.m. solar time and successively averaged for each half-hourly period, in order to obtain data comparable to EC and meteorological data.

Results showed that the inclusion of a variable ε derived from remotely sensed data improves GPP estimation suggesting that ε is a dynamic variable that should be taken into account to estimate GPP. Both PRI and Fy_{760} can be used for this purpose, demonstrating that besides PRI also Fy_{760} is a promising remotely sensed parameter that can be used to directly quantify light-use efficiency. The best performances for the three sites have been obtained estimating $APAR$ as a function of F_{760} and ε as

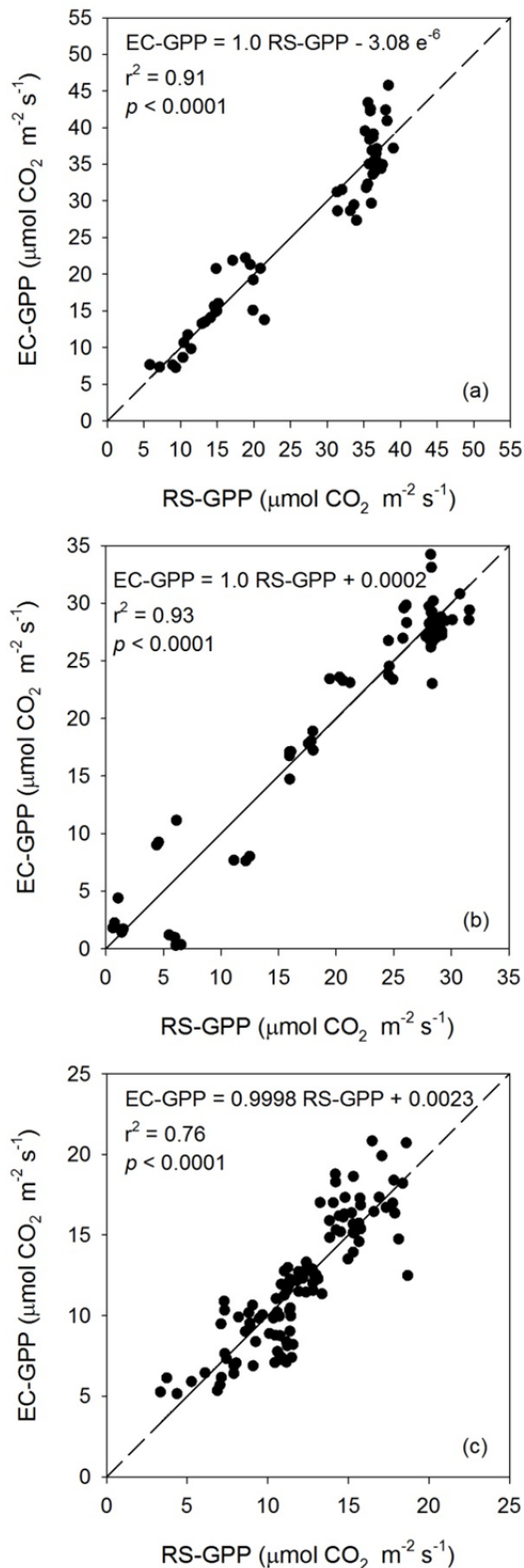


Figure 3. Relationship between GPP values estimated from EC measurements (EC-GPP) and modeled (RS-GPP) with the best-performing model in rice (a), alfalfa (b) and grassland (c). Linear regression equations, determination coefficients and p values are reported.

a function of PRI. The comparison between GPP values estimated from EC measurements (EC-GPP) and those modeled with the best-performing RS model (RS-GPP) for each of the three ecosystems is reported in Figure 3. A significant linear relationship with coefficient of determination ranging from 0.76 to 0.93 ($p < 0.0001$) and a slope close to 1.0 was found for all sites.

6. CONCLUSIONS

This manuscript summarises recent results obtained within the context of the activities related to the estimation of sun-induced chlorophyll fluorescence on the ground using high resolution commercial spectrometers.

Fluorescence emission levels of different land cover classes have been characterized. The highest fluorescence emissions were measured in crops followed by broadleaf and then needleleaf species. The detection of an ongoing stress was proven feasible in different control – treatment experiments (DCMU administration and ozone fumigation in our experiments): fluorescence was able to track photosynthesis decline before any visible symptom occurs. Chlorophyll fluorescence data was then used to provide more reliable estimations of GPP in crops and grasslands compared to those obtained with traditional vegetation indices.

The possibility of exploiting the remotely sensed fluorescence to infer the actual photosynthetic rate is crucial for the success of future space missions dedicated to fluorescence measurement (e.g., the FLuorescence EXplorer mission by the European Space Agency).

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REFERENCES

- [1] Damm A., Elbers J., Erler A., et al., 2010, "Remote sensing of sun induced fluorescence to improve modelling of diurnal courses of Gross Primary Production (GPP)," *Global Change Biology* 16, 171-186.
- [2] Guanter L., Zhang Y., Jung M., et al., 2014, "Global and time-resolved monitoring of crop photosynthesis with chlorophyll fluorescence," *Proceedings of the National Academy of Sciences*, March 25.
- [3] Rascher U., Agati G., Alonso L., et al., 2009, "CEFLES2: the remote sensing component to

quantify photosynthetic efficiency from the leaf to the region by measuring sun-induced fluorescence in the oxygen absorption bands," *Biogeosciences* 6, 1181-1198.

- [4] Meroni M., Colombo R., 2006, "Leaf level detection of solar induced chlorophyll fluorescence by means of a subnanometer resolution spectroradiometer," *Remote Sensing of Environment* 103, 438-448.
- [5] Malenovský Z., Mishra K.B., Zemek F., et al., 2009, "Scientific and technical challenges in remote sensing of plant canopy reflectance and fluorescence," *Journal of Experimental Botany* 60, 2987-3004.
- [6] Meroni M., Colombo R., 2009, "3S: A novel program for field spectroscopy," *Computers & Geosciences* 35, 1491-1496.
- [7] Meroni M., Busetto L., Colombo R., et al., 2010, "Performance of Spectral Fitting Methods for vegetation fluorescence quantification," *Remote Sensing of Environment* 114, 363-374.
- [8] Meroni M., Rossini M., Picchi V., et al., 2008, "Assessing steady-state fluorescence and PRI from hyperspectral proximal sensing as early indicators of plant stress: The case of ozone exposure," *Sensors* 8, 1740-1754.
- [9] Rouse J.W., Haas R.H., Schell J.A., et al., 1974, "Monitoring the vernal advancements and retrogradation of natural vegetation," Greenbelt, MD.
- [10] Gamon J.A., Peñuelas J., Field C.B, et al., 1992, "A narrow-waveband spectral index that tracks diurnal changes in photosynthetic efficiency," *Remote Sensing of Environment* 41, 35-44.
- [11] Monteith J.L., 1972, "Solar radiation and productivity in tropical ecosystems," *Journal of Applied Ecology* 9, 747-766.
- [12] Rossini M., Meroni M., Migliavacca M., et al., 2010, "High resolution field spectroscopy measurements for estimating gross ecosystem production in a rice field," *Agricultural and Forest Meteorology* 150, 1283-1296.