# Chlorophyll Fluorescence Detection with a High-Spectral Resolution Spectrometer through *in-filling* of the O<sub>2</sub>-A band as function of Water Stress in Olive Trees

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### Abstract

A high spectral resolution spectrometer of 0.065 nm FWHM in the 680-770 nm range was used for collecting spectral measurements in an orchard of olive trees in Spain under 3 different water stress treatments. The measurements were conducted as part of validation efforts for the FluorMOD project funded by the European Space Agency (ESA) to advance the science of vegetation fluorescence simulation. Diurnal steadystate chlorophyll fluorescence was measured from leaves in the field during summer 2004 using the PAM-2100 fluorometer to study the effects of water stress on chlorophyll fluorescence. Water potential, photosynthesis, and stomatal conductance on trees were also measured in a weekly basis to track the effects of water stress on the tree status and functioning. Infrared Apogee sensors were placed on top of the trees for diurnal thermal data collection, studying the effects of water stress on the tree temperature as an indicator of stress. The Ocean Optics HR-2000 spectrometer was used to measure irradiance and radiance spectra from above tree crowns under different stress conditions. The spectral measurements of irradiance with a cosine corrector and crown radiance with bare fibre were acquired from a pole 7 m in height to collect nadir radiance from the top of tree crowns. Analysis in the red edge covering the 680-770 nm range enabled the study of the chlorophyll fluorescence *in-filling* in the O<sub>2</sub>-A band at 760 nm. Results of the spectral analysis and simulation using the FluorMOD radiative transfer model demonstrate that water stress effects on steady-state fluorescence are detectable at the tree level in the O<sub>2</sub>-A band from reflectance spectra due to the *in-filling* effects.

Keywords: leaf fluorescence; canopy fluorescence; fluorescence in-filling, O<sub>2</sub>-A band

## 1. Introduction

Validation efforts for the FluorMOD project "Development of a Vegetation Fluorescence Canopy Model" launched by the European Space Agency (ESA) require the acquisition of canopy-level spectral radiance and reflectance under different viewing geometries, stress levels, and species. In this study, a part of validation efforts, the main objective was to determine if water stress levels caused by different irrigation treatments in olive tree crops would affect the crown-level natural fluorescence emitted, assessing the potential for its detection through the fluorescence *in-filling* effects in the O<sub>2</sub>-A band at 760 nm. The development of an integrated leaf-canopy model that simulates the effects of solar-induced fluorescence was proposed in FluorMOD as a critical task that would provide a guide to the assessment of fluorescence detection potential from canopy

reflectance. As a result of these efforts, the FluorMOD project (Miller *et al.*, this issue) is making progress on the development of a leaf model that simulates leaf fluorescence, FluorMODleaf (Pedrós *et al.*, this issue), and a canopy level model, FluorSAIL (Verhoef, this issue), which incorporates an excitation-fluorescence matrix computed externally by means of the leaf-level fluorescence model. The FluorMODgui integrated leaf-canopy model (Zarco-Tejada *et al.*, this issue) enables the simulation of diurnal effects under different viewing geometries, atmospheric characteristics and illumination dependency, modelling the effects of natural fluorescence on apparent canopy reflectance. The integrated model provides a theoretical basis for the reflectance measurements obtained from trees under different water stress levels, assessing if the effects observed on the experimental data are reproduced by the physical model as function of the fluorescence contribution.

# 2. Field Data Collection

Reflectance measurements were collected from the top of the olive tree crowns using a 0.065 nm FWHM Ocean Optics HR2000 fibre-optics spectrometer installed on a pole with a head where a cosine (downwelling irradiance) and a bare fibre (upwelling radiance) were attached. The experimental design consisted of a study area of olive trees where 3 irrigation treatments were applied, with 35 m<sup>3</sup> water/week (control treatment R), 3.6 m<sup>3</sup>/week (stress treatment S1), and an intermediate treatment (S2) (Figure 1). Temperature from the crowns was collected continuously using Apogee IRTS-P (Apogee, UT, USA) thermal sensors. Conductance, photosynthesis, water potential, and chlorophyll fluorescence were measured diurnally and weekly over the summer from June to November 2004, monitoring the stress caused by the deficit irrigation scheme. The 0.065 nm FWHM Ocean Optics HR-2000 spectrometer (Ocean Optics, Dunedin, FL, USA) provided spectral measurements in the 680-770 nm range from top of the crowns in the diurnal series (Figure 2). The spectral measurements were made over selected tree crowns under the 3 different water stress treatments, measuring irradiance with a cosine corrector and crown radiance with a 7 m height pole attaching the fibre head at the end to collect nadir radiance.

## 3. Experimental Results and Model Simulation

Leaf water potential measurements collected from 11 trees over the summer showed large variations in water stress as function of the irrigation treatment (Figure 3). As expected, S1 and S2 trees with lower irrigation doses were more stressed than R trees with nominal irrigation. Consistently, both stomatal conductance and photosynthesis rates were greater for the low-stressed well watered treatment (R). The leaf chlorophyll fluorescence measurements collected from selected trees diurnally and throughout the season showed that steady-state Ft fluorescence values for the low-stressed trees (R) were higher than for the high-stressed treatment (S2, S1) (Figure 4). The relationships between Ft and leaf water potential throughout the season were in good agreement, showing a final recovery across treatments on both Ft and water potential after the first rainfalls.



Figure 1. Airborne hyperspectral CASI image collected over the study area (left), showing the treatment blocks where deficit irrigation experiments were conducted (b).



Figure 2. Schematic view of the methods used for acquiring spectral measurements using the HR2000 high-resolution spectrometer.

Crown temperature measured with Apogee IRTS-P sensors from trees showed up to 4°C differences between stressed and non-stressed trees, showing the larger daily temperature differences between 16.00 and 19.00 GMT. Relationships between temperature and water potential at the tree crown level were obtained throughout the season ( $r^2 \sim 0.7$ ). The diurnal measurements of reflectance conducted with the Ocean Optics HR2000 spectrometer at 0.065 nm FWHM in the 680-770 nm range demonstrated that the fluorescence *in-filling* at the O<sub>2</sub>-A band can be detected on the reflectance signal at the crown level, manifested in a sudden reflectance increment of the reflectance at 760 nm (Figure 5). The amplitude of the 760 nm peak, potentially associated with the emission of natural fluorescence, was compared with steady-state fluorescence measurements collected at the same time from the trees under different stress levels. The agreement obtained in diurnal trials between Ft and the O<sub>2</sub>-A peak amplitude suggests that natural fluorescence can potentially be monitored using reflectance spectra. Nevertheless, the dependency of the emission peak on reflectance BRDF requires critical attention due to the known changes as a function of the viewing geometry and solar angle which accompany diurnal changes. The FluorMODgui radiative transfer model was used to simulate the effects of fluorescence on canopy reflectance, enabling the calculation of the 760 nm peak amplitude with and without fluorescence effects (Figure 5). The simulations show that the 760 nm peak observed on the experimental tree-level canopy reflectance is recreated with the physical model, disappearing when the canopy reflectance is simulated without fluorescence emission.



Figure 3. Measurements of leaf water potential collected from each tree over the summer (left) and averaged for each irrigation treatment (right).

![](_page_4_Figure_0.jpeg)

Figure 4. Steady-state natural leaf fluorescence collected with PAM-2100 from trees under different irrigation levels, showing the variation over the course of the day (left) and over the summer (right).

![](_page_4_Figure_2.jpeg)

Figure 5. Crown reflectance acquired with the HR2000 spectrometer at 0.065 nm FWHM from trees under different stress levels (top), showing the peak at 760 nm due to fluorescence emission. The FluorMODgui model simulation recreated the same effect due to fluorescence emission (bottom).

### 4. Conclusions

Reflectance measurements collected with a high-spectral resolution spectrometer, and simulations conducted with the FluorMODgui model demonstrated that the observed fluorescence *in-filling* at 760 nm, detected as a sudden peak on the reflectance signature, is consistent with the modelled apparent canopy reflectance when fluorescence emission is added. These results obtained at canopy level for water stress monitoring are in agreement with leaf-level studies conducted by Meroni (this issue) and Moya *et al.* (2004), showing that fluorescence emission can be detected at the leaf level and on a corn canopy under diuron herbicide penetration using the  $O_2$ -A band. The model demonstrates the effects of the fluorescence emission and the viewing geometry on the *in-filling* amplitude as simulated by the normal variation in a diurnal setting, showing small effects at the 760 nm band when canopy reflectance is simulated without fluorescence effects. This work suggests the potential application of spectral reflectance for monitoring natural chlorophyll fluorescence emission at the canopy level as an indicator of water stress.

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