



## Modeling re-absorption of fluorescence from the leaf to the canopy level

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### ABSTRACT

Chlorophyll fluorescence is widely used as an indicator of photosynthesis and physiological state of plants. Remote acquisition of fluorescence allows the diagnosis of large field extensions, even from satellite measurements. Nevertheless, fluorescence emerging from chloroplasts, the one directly connected to plant physiology, undergoes re-absorption processes both within the leaf and the canopy. Therefore, corrections of the observed canopy fluorescence, taking into account these two re-absorption processes may help to draw accurate inferences about plant health. Here, we show the theoretical development and experimental validation of a model that allows to retrieve the spectral distribution of the leaf fluorescence spectrum from that on top of canopy (TOC) using a correction factor which is a function of both canopy and soil reflectance, and canopy transmittance. Canopy fluorescence spectra corrected by our theoretical approach and normalized shows 95% correlation with the normalized fluorescence spectrum at leaf-level, thus validating the model. Therefore, our results provide a physical explanation and quantification for fluorescence re-absorption within the canopy, a phenomenon which has only been mentioned but never measured up to the date. From a more general perspective, this new analytical tool together with the one previously developed by Ramos and Lagorio (2004) allows to obtain the spectral distribution of chloroplast fluorescence spectrum from that on top of canopy (TOC).

### 1. Introduction

In photosynthetic organisms, chlorophyll-a excited states, produced either by direct absorption or by energy transfer, can decay by three main pathways: electron transfer (which initiates photosynthesis), heat dissipation and light emission as fluorescence. These three processes are competitive with each other and, in consequence, an increase in the efficiency of one of them leads to a decrease in the efficiency of one or both of the other two. This competition is the key for the existing link between photosynthesis and chlorophyll fluorescence (Maxwell and Johnson, 2000).

Thus, analysis of chlorophyll fluorescence from vegetation can give information on photosynthesis and on the physiological state of plants (Maxwell and Johnson, 2000; Moya and Cerovic, 2004; Guanter et al., 2014). This largely contributes to a better understanding of environmental and anthropogenic effects on vegetation physiology and also on carbon fluxes between plants and the atmosphere, thus providing the key for the assessment of greenhouse effect and, ultimately, global warming (Grace et al., 2007). Chlorophyll fluorescence may be studied from different observation scales: ground, airborne and spaceborne. An

excellent review of the methods and equipment used in each case is presented by Meroni et al. (2009).

For the remote sensing of chlorophyll fluorescence from vegetation, passive and active methods have been developed. While passive methods use polychromatic sunlight excitation (Louis et al., 2005; Meroni and Colombo, 2006; Guanter et al., 2007), active procedures use a high energy LASER as irradiation source with LiDAR (Laser imaging detection and ranging system) technology from an airborne (Cerovic et al., 1996; Ounis et al., 2016).

The interaction of light with canopies is a subject of great interest in the remote monitoring of plant health and it was described in literature by means of radiative transfer models (RTM) which are often used to describe the propagation of photons through a medium where they are affected by absorption, emission and scattering processes. Computational methods are required to solve the mathematical equations involved in these models. In particular, for remote sensing of vegetation, several RTM have been developed: PROSPECT simulates reflectance and transmittance of leaves, from 400 nm to 2500 nm (Jacquemoud and Baret, 1990); LIBERTY (*Leaf Incorporating Biochemistry Exhibiting Reflectance and Transmittance Yields*) calculates the

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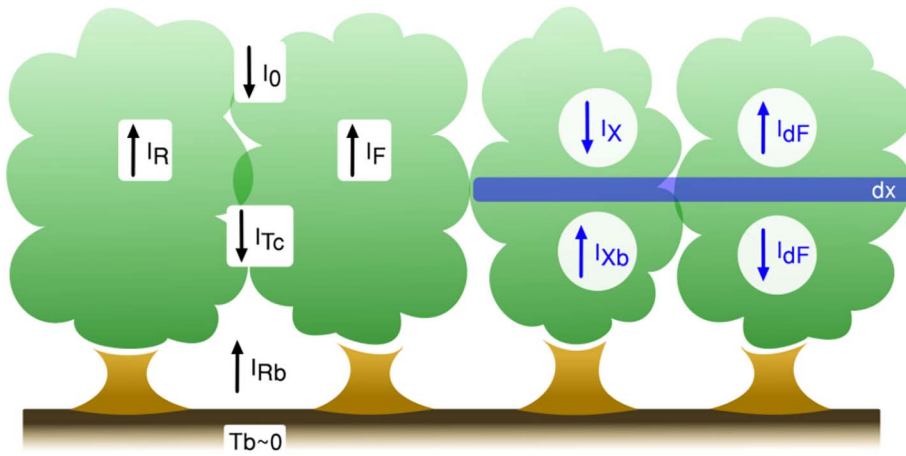


Fig. 1. Canopy model. Photon fluxes involved in the interaction between light and canopy, as assumed by our model. Light attenuation within the canopy was described by a mono-exponential decay in all the cases.

optical properties of conifer needles (Dawson et al., 1998); DLM (dorsiventral leaf radiative transfer model) (Stuckens et al., 2009) is a PROSPECT-like leaf optical model which includes leaves asymmetry; DART (Gastellu-Etchegorry et al., 2012) is a model for remote sensing images and radiative budget and FLIGHT simulates the observed reflectance response of three-dimensional vegetation canopies (North, 1996). Some RTM include fluorescence simulations such as the FluorMODleaf (Pedrós et al., 2010), the Fluspect-B (Vilfan et al., 2016) and FluorWPS (Zhao et al., 2016) models, which predict fluorescence spectra.

Different precision levels (from 1 to 3 dimensions) of canopy description may be found in the RTM. 1D models are the simplest approaches assuming a horizontal layer of vegetation and they are appropriate for homogeneous canopies. 3D models include canopy heterogeneity and they are more realistic (North, 1996; Gastellu-Etchegorry et al., 2012). However, to take into account canopy heterogeneity, 3D models require larger amounts of field data compared to 1D models (Ligot et al., 2014).

Chlorophyll fluorescence from plants is characterized by two peaks at about 685 and 735 nm (Mazzinghi et al., 1994). The red emission peak is due to the radiative deactivation of excited photosystem II (PSII) while the far-red emission one has contributions of both photosystems (PSI and PSII) (Agati, 1998; Pfündel, 1998; Franck et al., 2002; Iriel et al., 2014). The quotient between the red and far-red maxima fluorescence is usually referred to as fluorescence ratio and largely used as an indicator of the plant physiological state (Wittenberghe et al., 2014; Rossini et al., 2016). This ratio was connected with the underlying mechanism of photosynthesis and was reported as an instant monitor of CO<sub>2</sub> uptake by plants (Freedman et al., 2002; Rascher et al., 2009; Damm et al., 2010). Moreover, fluorescence emerging from chloroplasts suffers re-absorption processes in the leaf (Agati et al., 1993; Gitelson et al., 1998; Ramos and Lagorio, 2004; Cordon and Lagorio, 2006). In turn, fluorescence photons emerging from the leaves undergo additional re-absorption in the canopy (Porcar-Castell et al., 2014). As a result, the spectral distribution of fluorescence observed by a remote sensor is completely distorted by this phenomenon. An important point in the analysis of chlorophyll fluorescence is that it is actually the fluorescence of the chloroplasts (and not the “observed” fluorescence from a canopy) which is directly connected to the physiological state of the plant. Several groups have previously developed and applied models to perform corrections for light re-absorption processes in leaves (Agati et al., 1993; Ramos and Lagorio, 2004; Cordon and Lagorio, 2006) but, until now, there are no available models that shed light on these complex processes occurring in canopies.

Rossini et al., 2016 already modeled the fluorescence ratio for different canopies. These authors observed that the ratio between red and far-red fluorescence peaks was considerably lower at canopy level than the one measured on single leaves, attributing this fact to the re-

absorption of the red component within the canopy layers. Julitta et al. (2016) recently published a detailed analysis of the red/far-red fluorescence ratio remotely measured by different spectroradiometers. They compared each result with the leaf level value to detect which spectrometer provided the most accurate quantity. They made it clear, however, that the observed fluorescence ratio for the canopy should be lower than the observed fluorescence ratio at leaf level, as the red emission was more affected by re-absorption in the canopy than the far-red band. Fournier et al. (2012) compared the fluorescence emission of a natural grass canopy with the leaf level fluorescence spectrum and they found that the red-to-far-red fluorescence ratio decreased from the leaf to the canopy level. They attributed this effect to a preferential re-absorption of the red fluorescence band but no quantitative estimation of this effect was performed. Another recent work reporting fluorescence re-absorption in a canopy was published by Daumard et al. (2012) who performed passive fluorescence measurements at 687 and 760 nm on sorghum. They effectively observed a decrease in the fluorescence ratio red/far-red for the canopy compared to the leaf-level. Again, preferential light re-absorption in the red was correctly argued but a quantitative support was lacking.

Therefore, although the re-absorption of light within a canopy was profusely cited or qualitatively described in many works, it was never quantitatively corrected. The goal of this work was to present a method to correct active fluorescence measurements by the processes of light absorption that take place in the canopy, filling an important vacancy in the literature. More precisely, the scope of the present study was the development and validation of a model that corrected fluorescence re-absorption within a canopy and that allowed to retrieve the fluorescence spectral distribution at leaf level. Moreover, in combination with other pre-existing approaches, it allowed to recover the fluorescence spectral distribution of the chloroplast from that of the plant cover.

## 2. Model description and deduction

A pictorial description of the canopy physical model is presented in Fig.1. The physical approach developed in this work considered the following assumptions:

i) the system is composed by the canopy and the soil, ii) no light is transmitted through the whole system (canopy + soil), iii) the light suffers mono-exponential attenuation within the canopy, iv) the plant canopy is composed by fluorescent units (leaves), v) the fluorescence emitted by leaves is reabsorbed in the canopy before leaving it, vi) the re-absorption process also produces a mono-exponential attenuation of fluorescence within the canopy, vii) fluorescence is emitted isotropically within the canopy, viii) lateral light losses are neglected, ix) light scattering is taken into account by considering an effective path length.

The model described here is based on mathematical treatments

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