Fluorescence sensing techniques for vegetation assessment

Lawrence A. Corp, Elizabeth M. Middleton, James E. McMurtrey,
Petya K. Entcheva Campbell, and L. Maryn Butcher

Active fluorescence (F) sensing systems have long been suggested as a means to identify species composition and determine physiological status of plants. Passive F systems for large-scale remote assessment of vegetation will undoubtedly rely on solar-induced F (SIF), and this information could potentially be obtained from the Fraunhofer line depth (FLD) principle. However, understanding the relationships between the information and knowledge gained from active and passive systems remains to be addressed. Here we present an approach in which actively induced F spectral data are used to simulate and project the magnitude of SIF that can be expected from near-ground observations within selected solar Fraunhofer line regions. Comparisons among vegetative species and nitrogen (N) supply treatments were made with three F approaches: the passive FLD principle applied to telluric oxygen (O_2) bands from field-acquired canopy reflectance spectra, simulated SIF from actively induced laboratory emission spectra of leaves at a series of solar Fraunhofer lines ranging from 422 to 758 nm, and examination of two dual-F excitation algorithms developed from laboratory data. From these analyses we infer that SIF from whole-plant canopies can be simulated by use of laboratory data from active systems on individual leaves and that SIF has application for the large-scale assessment of vegetation. © 2006 Optical Society of America

OCIS codes: 120.0280, 120.6200, 120.5700, 170.6280.

1. Introduction

A major goal of the U.S. Carbon Cycle Science program is to monitor carbon dioxide (CO_2) uptake by vegetation. Biological carbon (C) sequestration is driven by nitrogen (N) availability, as N is involved in photochemical processes and is one of the primary resources that regulate plant growth. Large-scale monitoring of these processes is currently possible only with remote-sensing systems that rely heavily on passive reflectance (R) information. Fluorescence (F) emitted from chlorophyll (Chl), or ChlF, is directly related to photochemical reactions and has been extensively used for the elucidation of the photosynthetic pathways. Recent studies have shown that ChlF can be extracted from high-resolution reflectance spectra of vegetation[1–3]; this has been made possible by advances in passive F instrumentation, which facilitate remote acquisition of solar-induced fluorescence (SIF). The goal of this effort is to evaluate the potential of emerging F methodologies for determining vegetation parameters related to photosynthetic function and carbon sequestration dynamics in plants.

Chl is the major plant pigment associated with harvesting solar energy for conversion to photochemical energy and use in CO_2 assimilation for the production of sugars and other organic compounds. Under optimal growth conditions, most light absorbed by plant Chls and carotenes is utilized in photosynthesis; less than 3% is dissipated as heat or as F. The magnitude of F emissions (EMs) varies with exposure of plants to light and other environmental conditions and is governed by Chl concentration. The highest F yield occurs when competing photochemical processes do not drain energy available for photosynthesis and heat dissipation is low. As certain nutrients play primary roles in photosynthesis and

---

L. A. Corp (lcorp@hydrolab.arsusda.gov) and L. M. Butcher are with Science Systems and Applications, Inc., Lanham, Maryland 20706. E. M. Middleton is with the Biospheric Sciences Branch, NASA/Goddard Space Flight Center, Greenbelt, Maryland 20771. J. E. McMurtrey is with the Hydrology & Remote Sensing Laboratory, Agricultural Research Service, U.S. Department of Agriculture, Beltsville, Maryland 20705. P. K. Entcheva Campbell is with the Joint Center for Earth Systems Technology, University of Maryland, Baltimore County, Baltimore, Maryland 21250.

Received 3 May 2005; revised 19 August 2005; accepted 25 August 2005.

0003-6935/06/051023-11$15.00/0

Chl synthesis, their deficiencies can be detected on the basis of changes in F, especially that of N.[4–8]

ChlF is manifested as red fluorescence (RF) and far-red fluorescence (FRF).[9] RF has been attributed primarily to Chls associated with photosystem II, whereas FRF has been attributed to antenna Chls of both photosystems II and I.[10–12] The RF/FRF ratio could relate to changes in the distribution of excitation (EX) energy between the two photosystems and has been extensively studied as an indicator of Chl content and stress condition in plants.[9,13–15] In addition to ChlF, F from vegetation occurs throughout the ultraviolet (UV-A) to visible regions, with EM maxima occurring in the UV-A, blue (BF), and green (GF).[16,17] Several investigations have demonstrated relationships to plant health and growth condition for these F regions and their ratios.[5,16,18–22]

The remote sensing of F over land has proved challenging because the relatively weak EMs from vegetation must be differentiated from the more-intense R signals. One solution is to utilize the Fraunhofer line depth (FLD) principle with high spectral resolution R spectra. The FLD principle uses the dark lines in the spectrum of light reaching the Earth's surface reported by Joseph Fraunhofer in 1817 and subsequently identified as either solar or telluric (i.e., atmospheric) in origin. This approach employs the dark regions to potentially differentiate F from the solar irradiance continuum with sufficient resolution for low-Earth-orbit observations by interferometer-type passive satellite systems. This technology has been suggested as a passive method for detecting vegetation stress from orbit.[23,24] In particular, the FLD method has been suggested as a means for extracting ChlF from R spectra in the telluric oxygen (O_2) bands centered at 688 and 760 nm. However, only a few instruments capable of remotely detecting solar-induced ChlF have successfully demonstrated variations in the F signal that can be ascribed to vegetation stress.[25–29]

The focus of this investigation is on the F signal from terrestrial vegetation as viewed from near-ground levels. The current study was designed to (1) develop generalized three-dimensional EX by an EM spectral matrix (EEM) for foliage represented by two herbaceous and three woody plant species from actively induced F studies; (2) present an approach to simulating SIF from the EEM and discrete EX spectral data at the foliar level; (3) determine from field spectra the magnitude of SIF that can be obtained at ground level from terrestrial vegetation assemblages within the telluric O_2 absorption bands; and (4) compare the relative ability of three F approaches to discriminate experimental vegetation treatments by use of the passive FLD applied to the telluric O_2 bands, the simulated SIF at solar Fraunhofer lines, and two active dual-EX algorithms.

5. Conclusions

This research has defined a method for converting monochromatically induced excitation spectra to simulate polychromatic solar-induced fluorescence. Integrated values from actively induced leaf level EX spectra were adjusted, and radiometric estimates of near-ground SIF from vegetation were reported for the primary emission bands. The simulated red SIF intensity at 680 nm ranged from 1 to 5 $\mathrm{mW\ m^{-2}\ nm^{-1}\ sr^{-1}}$, whereas the far red at 740 nm ranged from 4 to 10 $\mathrm{mW\ m^{-2}\ nm^{-1}\ sr^{-1}}$. Of the solar Fraunhofer lines, the H_α line at 656.28 nm and the Fe_I line at 751.1 nm provided the highest signals for near-ground observations of the red fluorescence and far-red fluorescence features. Simulated SIF for the blue–green EM bands were substantially lower for green vegetation but comparable for dead or dormant plant materials, and the H_β line at 486 nm provided the highest blue–green F signal. The simulated SIF data shown here suggest that Fraunhofer line properties have a great effect on the final selection of suitable lines that needs to be balanced with the proximity to the fluorescence EM peaks. Also, simulated SIF from excised plant material showed promise with respect to differentiating plant growth and condition. Increased spatial coverage and repeat temporal measurements should reinforce relationships between solar-induced ChlF and canopy photosynthetic light-use efficiency.

Further sensitivity to plant health parameters can be achieved through active systems and several discrete fluorescence ratios. Maximum EM intensity from blue fluorescence and green fluorescence bands can be achieved from EX at 347 ± 25 and 410 ± 10 nm, respectively, whereas the RF and FRF bands exhibited multiple EX maxima: 430 ± 10, 472 ± 8, and 665 ± 8. These RF and FRF EX maxima were strongly associated with the blue and red absorption of Chl *a* and Chl *b* pigments, which led to the development of spectral F excitation ratios for the *in vivo* determination of Chl *a*:*b* content (Fig. 5). Monitoring the variation in these pigment ratios among species and with stress condition could lead to an improved remote-sensing interpretation of species composition and physiological growth condition.

Based on the results from this study, we recommend that specifications for future active F instrumentation require multiple excitation bands. Further studies are needed to establish a compromise between maximized EM intensity versus sensitivity to plant stresses in additional species and stress conditions. The differences in plant physiology and F response found in corn, soybeans, maple, sweet gum, and poplar used in this experiment are estimated to be in the range of what is indicative of other annual and perennial plant species.