Determination of water contents in leaves by a near-infrared multispectral imaging technique

Chieu D. Tran*, Victor I. Grishko

Department of Chemistry, Marquette University, P.O. Box 1881, Milwaukee, WI 53201-1881, USA

Received 19 September 2003; accepted 10 October 2003

Abstract

The kinetics of water desorption from olive leaves was studied using a near-infrared (NIR) multispectral imaging spectrometer. This imaging spectrometer is capable of sensitively and rapidly recording NIR spectral images of leaves because it was constructed with an acousto-optic tunable filter and an InGaAs focal plane array NIR camera. The high sensitivity and fast scanning ability of the imaging spectrometer make it suitable for kinetic determination. The kinetics of water desorption from olive leaves, determined by this multispectral imaging instrument, show that rate of water desorption is strongly dependent on the environment in which the leaves were stored. Water desorbed from leaves faster when leaves were stored under dry conditions. The rate for leaves stored in 0% humidity environment is 1.5× faster than those stored in 50% humidity.

Keywords: Near-infrared; Acousto-optic tunable filter; Remote sensing; Water contents; Olive

1. Introduction

Water content is one of important indicators of the welfare of plants [1–11]. It is therefore, hardly surprising that there is a continuous need to develop sensitive sensors which can remotely and non-intrusively determine water levels in plant leaves [1–11]. The near-infrared technique may be suited for such a sensor as it is known to possess advantages such as wide applicability, non-invasiveness and on-line characteristics. Specifically, the near-infrared region covers the overtone and combination transitions of the C–H, O–H, C=O and N–H groups, and since all organic compounds possess at least one or more of these groups, the technique is applicable to all agricultural and forest products [6,9,12–16]. There is no need for pretreatment of the sample, and since NIR radiation can penetrate a variety of samples, the NIR technique can be used for non-invasive and on-line quality control and assurance as well as for remote sensing. In fact, it was demonstrated recently, that the NIR technique can be effectively used to determine the water contents of leaves [3–5].

*Corresponding author. Tel.: +1-414-288-5428; fax: +1-414-288-7066.
E-mail address: chieu.tran@marquette.edu (C.D. Tran).

and soils [8], to determine the nitrogen concentration in potato tissues [10] and the soluble solids content in processing tomatoes [7], as well as to measure acidity, soluble solids, and firmness of Jonagold apples [11]. However, care must be taken when using the NIR method to analyze agricultural and forest products. This is due to the limitations of the method currently used, namely, NIR measurements are based on point measurements on single fruits or leaves. As such they are complex, time consuming and difficult to integrate. Furthermore, agricultural and forest products such as plant tissue is not spectroscopically homogenous; i.e. the cellular architecture and processes strongly influence light penetration and cause a great deal of light scattering. As a consequence, extrapolation from a single point measurements to whole group of fruits or plant is not only labor intensive but also prone to errors. Thus, NIR chemical characterization of fruits or plant has been limited to relatively small scales. Non destructive NIR method which is capable of rapidly acquiring spectral information on a large spatial scale is therefore desirable. A NIR multispectral imaging spectrometer is needed for such purpose.

Multispectral imaging spectrometer is an instrument that can simultaneously record spectral and spatial infor-
mation of a sample. Chemical information on a large area of a single sample or samples can be elucidated from recorded images. In these instruments, the spatial distribution of the sample is obtained by a camera, and the spectral information is gained by scanning a dispersive element [17–20]. In order to be used for realtime and on-line monitoring, the instrument must be able to sensitively and rapidly record spectral images of a sample. These requirements can be satisfactorily met with the instrument recently developed in our laboratory [21–23]. In this instrument, an acousto-optic tunable filter (AOTF) is used for spectral scanning and an InGaAs camera is for recording NIR images [21–23]. An AOTF is an electronic tunable filter which has rapid scanning ability, high diffraction efficiency, wide spectral tuning range and high resolution [24–27]. Because of these advantages, the multispectral imaging instrument based on an AOTF is compact, has no moving parts and rapid scanning ability. The sensitivity of the instrument is high because of the high diffraction efficiency of the AOTF (i.e. high light throughput) and the high sensitivity of the recently available InGaAs camera (i.e. this InGaAs camera has the highest sensitivity among available NIR cameras such as InSb and HgCdTe in the short NIR region, namely from 1000 to 1700 nm). We were able to use this NIR imaging spectrometer for measurements which to date were not possible. These include the determination of the chemical inhomogeneity of copolymers and the authentication of stock certificates and currency [21–24]. The kinetic and evidence of inhomogeneity of the curing of epoxy by amine and of sol–gel processes can also be measured using this instrument [21–29].

The information presented is indeed provocative and clearly indicates that it is possible to use the AOTF based NIR multispectral imaging instrument for non-destructive and non-invasive determination of water contents of plant leaves. Such consideration prompted us to initiate this study which aims to explore the use of this AOTF based NIR multispectral imaging instrument for the remote, non-intrusive determination of water contents of plant leaves. Preliminary results on the measurements of water contents of leaves of the olive tree, *Olea europaea*, will be reported in this manuscript.

2. Experimental

2.1. Samples

Olive leaves were harvested from olive trees (*Olea europaea* L.) in Barcelona, Spain and shipped immediately to our lab by express mail. Upon arrival, they were divided into three groups: group 1 samples were stored at room temperature in a desiccator equipped with silica gel drying agent (dried samples), group 2 samples were stored at room-temperature in a desiccator saturated with water vapor (100% humidity samples) and group 3 samples were stored at room temperature without any desiccator (50% humidity samples).

2.2. Instrument

The AOTF-based NIR multispectral imaging instrument used in this study is similar to the imaging spectrometer used in our previous studies. Spectral images of olive leaves were recorded in the 1000–1700 nm range with a 2 nm interval. NIR spectra at different positions within a leaf were calculated from recorded images using an average of 7×7 square pixels.

3. Results and discussion

It was found that freshly harvested olive leaves have similar absorption spectra in the wavelength range used in this study (1000–1700 nm). Shown in Fig. 1a is the spectrum of an olive leaf. As illustrated, the leaf has an intense and broad absorption band at approximately 1460 nm. Based on results of our previous work and those reported in literature, this band can be assigned to the overtone and combination transitions of O–H and C–H bands. The validity of this assignment can be seen by comparing the leaf spectrum with that of water (Fig. 1b) and of palmitic acid (Fig. 1c) which is known to be one of the main components of olive oil. As illustrated, the absorption bands of the leaf is very similar to the O–H band of water (Fig. 1b) and the C–H band of palmitic acid (Fig. 1c). Based on limited number of
spectra recorded here, it is relatively difficult to resolve the O–H band from that of the C–H band. However, deconvolution is not needed to determine water content of leaves. This is because it is known that the water contents of leaves are known to be strongly dependent on the leaf conditions and readily undergo dehydration (desorption). As a consequence, the rate of water desorption from leaves can be determined by monitoring changes in the NIR absorption spectra of leaves in this region. Shown in Fig. 2 are images of leaves (at 1460 nm), recorded after they were stored under three different conditions (0%, 50% and 100% humidity) for 21 days. It is evident from the images that leaves underwent substantial changes in their chemical compositions when they were stored under different conditions. It is, therefore, expected that the observed changes in the spectra of the leaves are due to the desorption of water from leaves. To determine kinetics of water desorption from leaves, images were recorded for three groups of leaves (0%, 50% and 100% humidity) over a range of wavelength from 1000 to 1700 nm at an interval of 2 nm, and at different times (from t=0 to t=21 days). NIR spectra were then calculated from these recorded images using an average of a square of 7×7 pixels. Shown in Figs. 3–5 are three-dimensional spectra of leaves from three groups (humidity 0%, 50% and 100%) plotted as a function of wavelength and time. As illustrated, for leaves stored under 0% and 50% humidity (Figs. 3 and 4), intensity of the band at 1460 nm decreases substantially as a function of time. For leaves stored under 100% humidity, there does not seem to be any change in the intensity of this band. These results seem to suggest that the decrease in the absorbance of this band is due to changes in the water contents of the leaves, namely when stored under dry and/or 50% humidity, leaves underwent a dehydration effect; water desorbed from the leaves.

Plots of absorbance of the 1460 nm band vs. time are shown in Fig. 6 for leaves stored under 0%, 50% and 100% humidity. As illustrated, for 0% and 50% leaves, water underwent rapid desorption. However, for 100% leaves, there is no change in the absorbance at 1460 as a function of time. Furthermore, changes in the absorbance at 1460 nm for 0% and 50% leaves were found to decay exponentially with time. This is hardly surprising as it is expected that desorption of water from leaves follows first order kinetics (or rather pseudo first order). From the plots, the rate of water desorption was found...
Fig. 6. of changes in absorbance at 1460 nm of leaves stored at 0% (a) and 50% (b) humidity as a function of time.

to be 0.36 day\(^{-1}\) and 0.24 day\(^{-1}\) for 0% and 50% humidity, respectively. It is expected that water desorbed from leaves faster when they were stored under dry conditions (0% humidity) than under normal conditions (50% humidity). However, it is rather unexpected to find out that the difference in rates can be as large as 1.5 times.

Taken together, the results presented clearly demonstrate that the AOTF-based NIR multispectral imaging instrument can be successfully used for the non-invasive measurement not only of the water content of leaves but also of the kinetics of water desorption from leaves. Since AOTF is an all solid state spectral tuning device which has no moving parts, and the InGaAs focal plane arrays NIR camera used in this study is operated at room temperature and can be powered by battery, the entire AOTF-based NIR multispectral imaging instrument including its computer control can be constructed for realtime measurements directly on-field. It can, therefore, be used for on field measurements, in either manned or unmanned states. This is the subject of our intense investigation.

References