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Influence of Stress Conditions on Spectral Signatures in the Visible, Near Infrared, and Thermal Infrared

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Influence of stress conditions on spectral signatures in the visible, near infrared, and thermal infrared

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Introduction

The object of this work is vegetation and its stress. Many reasons lead to vegetation stress: lack of water (water stress), reduction of the number of conducting vessels by disease or insects (biological stress), or high salinity in the soil water (salinity stress). Stress conditions produce changements in living plants, such as variation in water content and in photosynthetic activity, which are monitorable in the vegetation emission spectra.

The main vegetation emissions are of three kinds:

a) black body emission at about 300 °K;

b) elastic light scattering from any natural source illuminating vegetation, like sun;

c) fluorescence emission, induced by broad band excitation, or monochromatic light (laser excitation).

The maximum intensity of a 300 °K blackbody lies in the thermal infrared region, at about 10 μm (Fig. 1), while there is no emission at wavelengths shorter than 3 μm. In the b) case the emission spectral-band lies in the spectral-region of the illuminating source: if solar light is taken into account (Fig. 2), the vegetation spectrum is in the visible, with a peak in the green region (blue light is mostly absorbed by vegetation, and green one reflected), without any contribution in the thermal infrared.

Fluorescence spectra of living plants give quite interesting information on chlorophyll and photosynthetic activity. With a broad band excitation, both elastic scattering and fluorescence emission effects happen, while with a monochromatic excitation the main detectable process is fluorescence.

Moreover, fluorescence signal of plants is quite suitable in a remote sensing application, offering the potential of monitoring the effects of soil and air pollution on the vegetation in a very early, pre-visual stage.

In this paper the main processes involved in the radiation with matter interaction are discussed; then, chlorophyll, as the particular matter with which radiation interacts, is taken into account.

Finally, vegetation emission spectra are presented and some original results discussed.

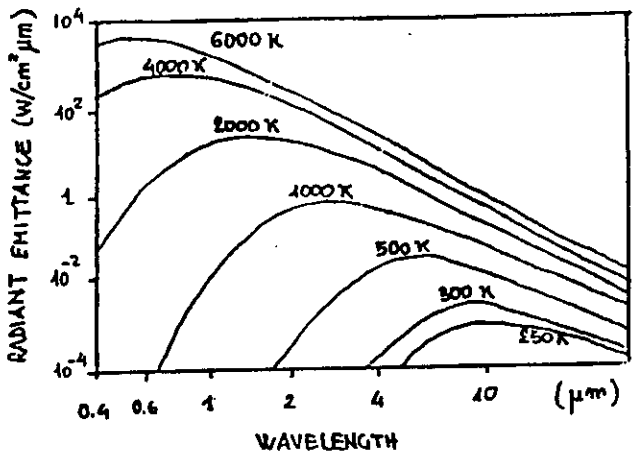

FIG. 1

Black body emission spectra at different temperatures.

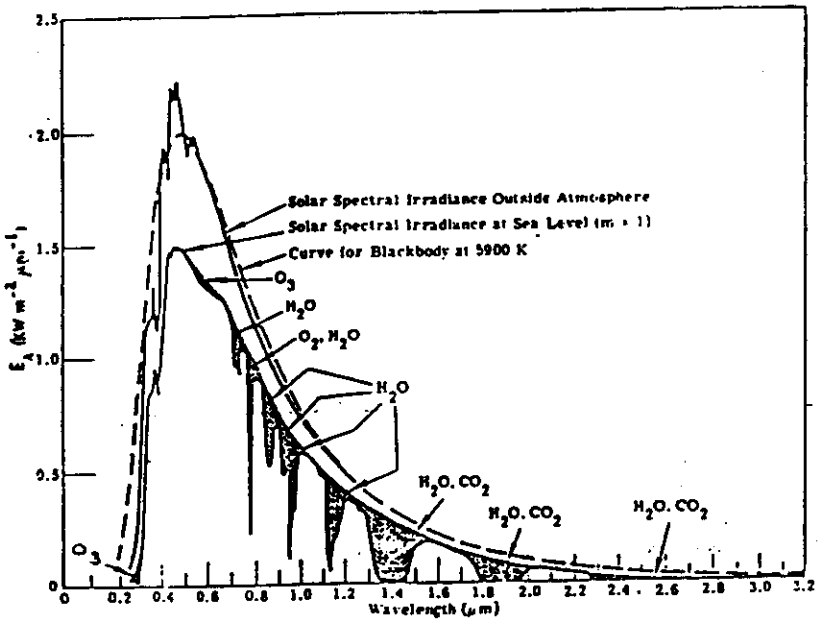

FIG. 2

Spectral distribution curves related to the sun. The shaded areas indicate absorption at sea level due to the atmospheric constituents shown.

Conclusions are carried out with regard to a potential LIDAR application.

Radiation with matter interaction: an overview

When an electromagnetic field interacts with a material, part of the radiation is absorbed, part transmitted, and a part scattered. The main interest for remote sensing application is for the scattered radiation.

Absorption spectra are characteristic of matter. They occur over a broad portion of the electromagnetic spectrum, from the microwave region where the spectral lines are characteristic of rotational transitions, to the vacuum ultraviolet where they result from outer-shell electronic transitions. The wavelength spread is from 20 cm (or longer) to wavelengths as short as 2.5×10^{-5} cm; and the corresponding energy span is from 6 microeV to 5eV. This wide range is made possible by means of molecular interactions as outlined below.

Far-infrared (25-500 μm) - absorption due primarily to pure rotational transitions and vibration-rotation bands.

Thermal-middle infrared (2.5-25 μm) - absorption due to fundamental (as well as some overtone) and combination vibrational-rotational bands.

Near-infrared (0.7-2.5 μm) vibrational overtone and combination bands. This region contains additional overtones and combinations of the fundamental vibration-rotation bands.

Visible (0.4-0.7 μm) - absorption due to electronic transitions, with vibrational-rotational structure.

Near Ultraviolet (0.25-0.4 μm) - absorption due to electronic transitions, with vibrational-rotational structure. All molecules have electronic absorption bands, but only diatomic molecules and small polyatomic molecules have characteristic resolvable structure in the near ultraviolet.

The spectrum of the scattered radiation contains information about both quantitative and qualitative characteristics of the target. Scattering can be described by means of two main processes, called elastic and inelastic scattering.

The first one (elastic scattering) is the process in which

$$\nu_r = \nu_o \qquad (1)$$

where ν_o indicates the frequency of the impinging electromagnetic field, while ν_r is the frequency of the scattered field.

This kind of interaction occurs without any appreciable energy exchange between incident radiation and internal states of the atoms or molecules constituting the target material. Because of this reason elastic scattering does not lead to a quantitative analysis of the target constituents, but it gives information only about target geometrical characteristics.

Mie and Rayleigh scatterings belong to this kind of interaction: Mie scattering takes place when the dimensions of the particle are close to or

larger than the wavelength of the incident light. The Mie scattered light is concentrated in the forward direction, with a much smaller intensity backward. The cross section of this scattering is customarily very large, so that high sensitivity is achieved for detecting particulated matter such as dust, water droplets, and other aerosols. Rayleigh scattering is known to occur in phase with the incident radiation; the Rayleigh-scattered energy is concentrated along the direction of the incident beam, with equal intensities forward and backward. Since the central wavelength of the Rayleigh-scattered component is the same as that of the Mie-scattered component, the scattered light does not identify the scatterer.

In the latter process (inelastic scattering)

$$\nu_r \neq \nu_o \qquad (2)$$

there is an energy exchange between the two system, from which it is possible to get quantitative information about chemical-physical parameters of the target.

Figure 3 shows these kinds of processes schematized by means of energy level diagrams of atoms or molecules (constituting the target) and the energy of incident radiation.

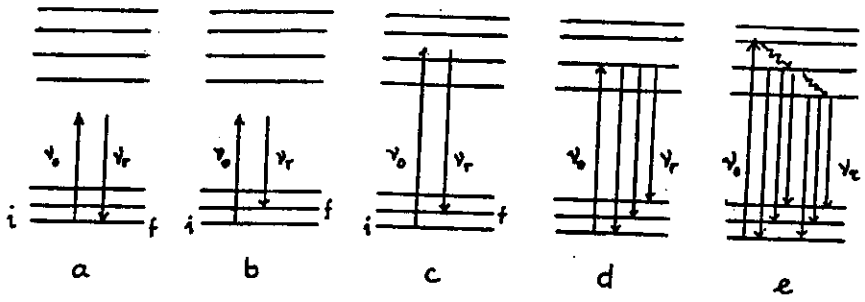

FIG. 3

Radiation with matter interaction: different processes.

Rayleigh scattering is represented in Fig. 3a: ν_o is the incident frequency and ν_r is the scattered one (which in this case are the same). The figure illustrates electronic ground and excited states, with the individual levels designated i for initial, m for intermediate, and f for final. These levels are supposed to be established for atoms by interactions producing fine and hyper-fine structures; and for molecules they correspond to vibrational-rotational levels.

Figure 3 also presents the possible types of transitions involved in the atomic and molecular processes by which a photon is emitted inelastically (inelastic processes).

Raman scattering is a process involving an exchange of a significant amount of energy between the scattered photon and the scattering species, as shown on Fig. 3b, 3c. Thus the Raman scattering component is shifted from the incident beam frequency by an amount corresponding to the internal energy of the species. The cross section for Raman scattering is usually smaller than that for Rayleigh scattering by about three orders of magnitude. The Raman-scattered intensity is proportional to the number of molecules in their initial states producing the spectral band.

Fluorescence is the spontaneous emission of a photon following excitation into an excited state by absorption of incident radiation at a frequency ν_o within a specific absorption line or band of an atomic or molecular species. In Fig. 3d the excited level decays by re-emitting photons via transitions to the original and different lower levels.

These emissions exhibit discrete peaks conventionally called resonance fluorescence. The excited atoms and molecules also suffer collisions which redistribute them into other excited levels through non-radiative transitions, as denoted by the wavy arrows in Fig. 3e. This process usually yields broad fluorescence as a near continuum. The re-emitted radiation is useful in identifying and monitoring the atomic or molecular species responsible for the fluorescence.

Fluorescence is customarily thought as two single-photon processes; that is, a two step interaction consisting of absorption of a single photon with frequency ν_o followed by spontaneous emission of a photon with frequency ν_r.

On the other hand, scattering associated with an individual atom or molecule, like Rayleigh and Raman scattering, is generally considered to be a two-photon process described by a single step interaction yielding effectively the simultaneous destruction of one photon with frequency ν_o and the creation of a different photon with frequency ν_r.

The processes, which are of interest in vegetation stress monitoring, are fluorescence, excited by means of a laser source, and reflectance, which is related to absorption process, excited with a broad band radiation.

Moreover, the black-body emission in thermal infrared, being a measure of vegetation surface temperature, gives information about stress level in plants.

Chlorophyll

Chlorophyll is a complex organic molecule, which is present in all vegetation species. Chlorophyll is also the main responsible of the photosynthetic process in living plants. So, by in vivo chlorophyll detection (that is by means of fluorescence and reflectance spectra), it is reasonable to expect information both on species and vegetation stress.

It is practically impossible to analyze chlorophyll fluorescence in terms of energetic levels of single molecules.

Infact, a complex organic molecule is formed by a high number of atoms; so there are many vibrational levels. Moreover, each vibrational level is splitted in rotational inner-levels, broadened by collisions with neighbouring molecules. It turns out a near-continuum level band, which produces absorption and emission spectra like those in dashed line showed on Fig.4. Because of this reason chlorophyll fluorescence has been studied only empirically.

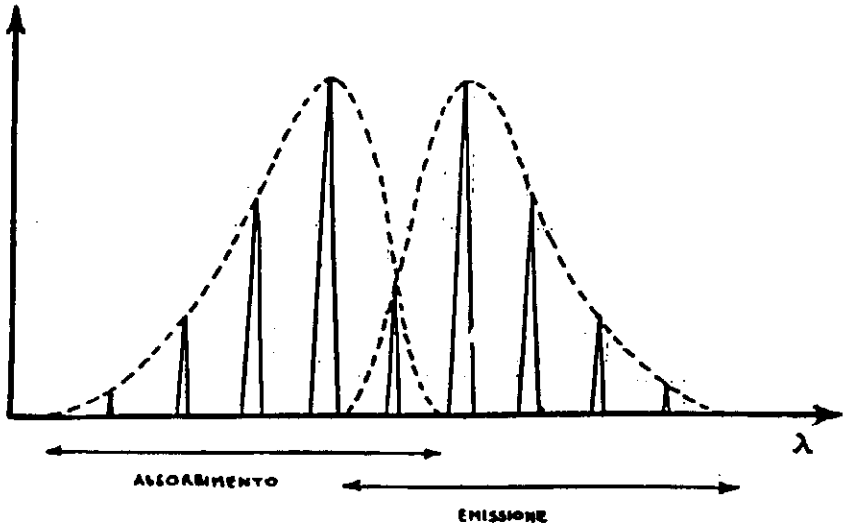

FIG. 4

Typical absorption and emission (fluorescence) spectra of a complex organic molecule.

A complication comes out from the fact that chlorophyll is highly reactive with light and that spectra have to be taken in vivo, where many other parameters are under variation.

Before dealing with spectral signatures of chlorophyll, the role of chlorophyll in photosynthetic mechanism is presented.

In photosynthesis two different phases can be distinguished: one is stimulated by solar light, the other one occurs in the dark.

The first phase involves light-harvesting-system and reaction centers, therefore it involves any kind of processes which are inherent with optical emission.

The second phase consists in the chain of reactions which exploits the assimilation of CO_2 from stomata and of H_2O from soil to synthesize O_2 and nutritive carbohydrate,

$$CO_2 + H_2O \xrightarrow[\text{light}]{} (CH_2O) + O_2 \qquad (3)$$

There are various photosynthetic pigments, whose function is to provide the plants with an efficient system of absorbing light throughout the visible spectrum. This energy is then transferred to the reaction centers, where it is utilized for the photochemical reactions. The bulk of this pigments are called the light harvesting pigments.

There are two kinds of chlorophyll in higher plants: chlorophyll a and chlorophyll b. Chlorophyll a is the major pigment and it is present in all photosynthetic organisms that evolve 02. Several forms of chlorophyll a have been postulated: chlorophyll a 660, 670, 680, 685, 690 and chlorophyll a 700-720.

When a pigment is excited by a quantum of light, there are several ways of deexcitation. Deexcitation occurs by transferring energy to other pigments, by fluorescence emission, by internal conversion — energy loss for heat —, by employing energy in photochemical reaction (redox process sensitized by reaction center).

The energy, by transfer mechanism, reaches the reaction center Ch 1, where it is converted into chemical energy with the production of an oxidizing and a reducing equivalent.

The redox reactions are attributed to two different pigments of chlorophyll a: P700 for system I and P680 for system II.

The fluorescence spectrum of chlorophyll a in vivo has a narrow band at 685 nm and a broad band at 740 nm. This fluorescence is dominant because the other pigments transfer their absorbed energy with a relatively high efficiency.

Now it is clear that there is a deep interconnection between fluorescence and photochemical processes. In fact it is possible to note that characteristic of fluorescence spectra are bounded to the photosynthetic activity, i.e. to the conditions of illumination, of stress, and of the plant aging level.

Fluorescence and reflectance spectra

The experimental setup used for spectra excitation and recording is shown on Fig. 5; it mainly consists of a laser source which illuminates the vegetation sample, a receiving optics which collects the radiation emitted from the target, a 500 mm focal length polychromator with 300 l mm^{-1} ruled grating, and an optical multichannel analyzer, PAR OMA-2, for the real time recording of the spectra.

This experimental system simulates a kind of active teledetection (fluorescence lidar), but it is quite easy to make passive teledetection with the same system by substituting the laser source with a lamp (which is a broadband, incoherent source) and then achieving reflectance spectra.

Two kinds of lasers were used: an He-Ne laser, whose wavelength (632.8 nm) matches reasonably well the absorption band of chlorophyll a, and a N_2 laser, which emits in the near ultraviolet (337.1 nm).

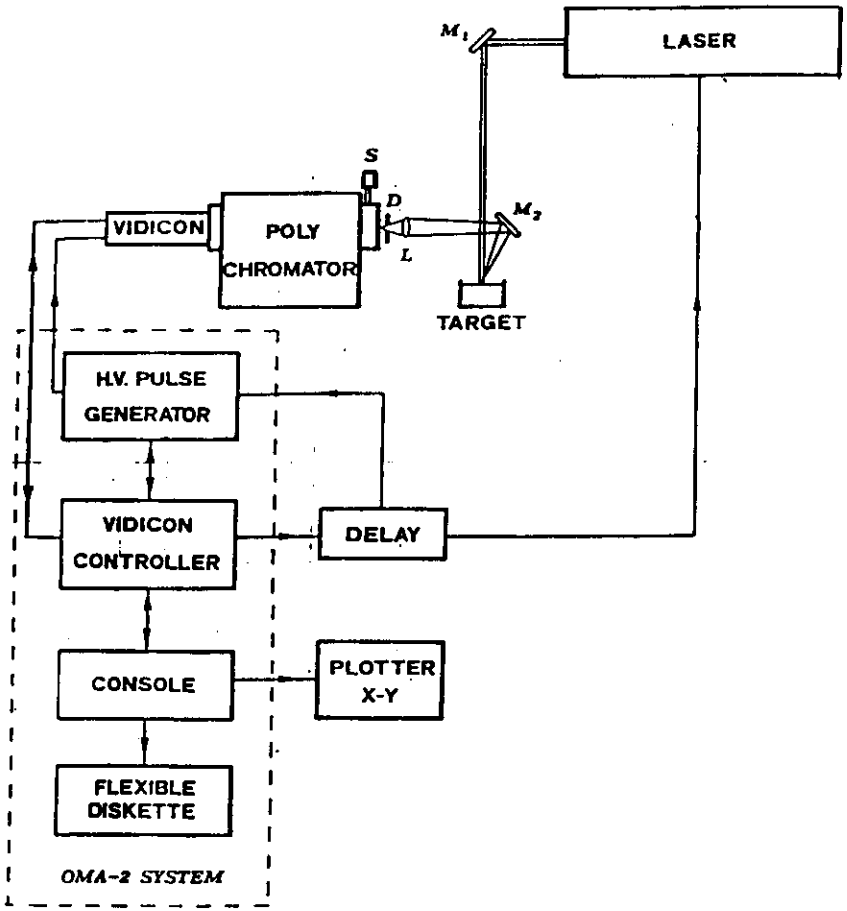

FIG. 5
Experimental set up.

All the spectra were obtained in vivo by different cultivars, and different water stress levels were investigated.

The fluorescence emission spectrum of chlorophyll in leaves is given with an excitation at 632.8 nm, by the convolution of a peak at 685 nm and a broad band at 720 nm, as shown on Fig. 6; while with an excitation at 337.1 nm the fluorescence spectrum of vegetation shows another intense peak around 460 nm, due to the fluorescence of other pigments, such as NADP (nicotinamide adenine diphosphate) and vitamine K1 (plastoquinone). The fluorescence spectrum with a 337.1 nm excitation is shown on Fig. 7.

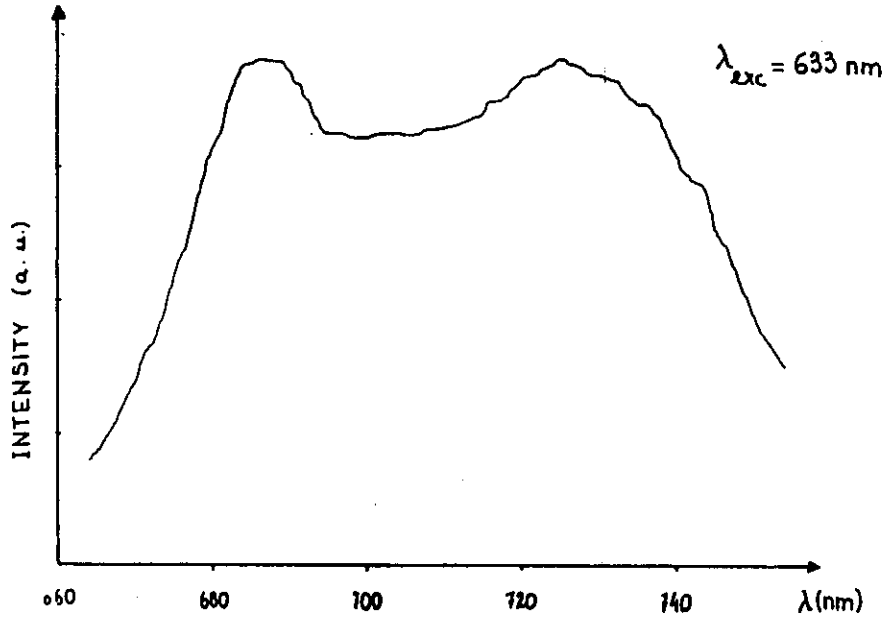

FIG. 6
Chlorophyll fluorescence spectrum in living plants (λ_{exc} = 633 nm).

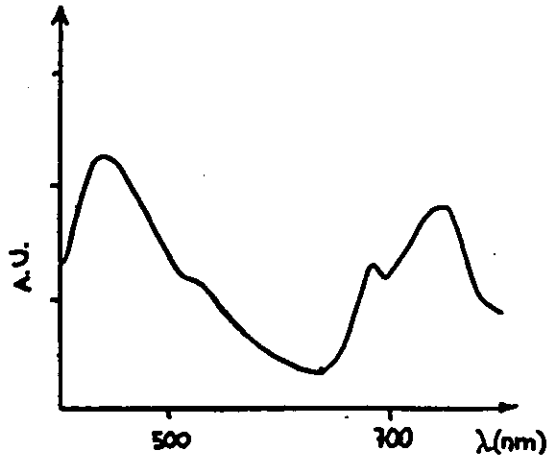

FIG. 7
Chlorophyll fluorescence spectrum with N_2 excitation (λ = 337.1 nm). (From Chapelle, Appl. Opt. 84)

On Figures 8a,b,c it is displayed a sequence of fifteen spectra indipendent of each other; the experiment was carried out in this way: as

the laser source was turned on, the first spectrum was taken and recorded (the OMA system requires 0.7 sec for averaging and recording a set of 10 spectra), then the second spectrum was taken 5 sec later and so on; after the 15th recorded spectrum the laser source was turned off.

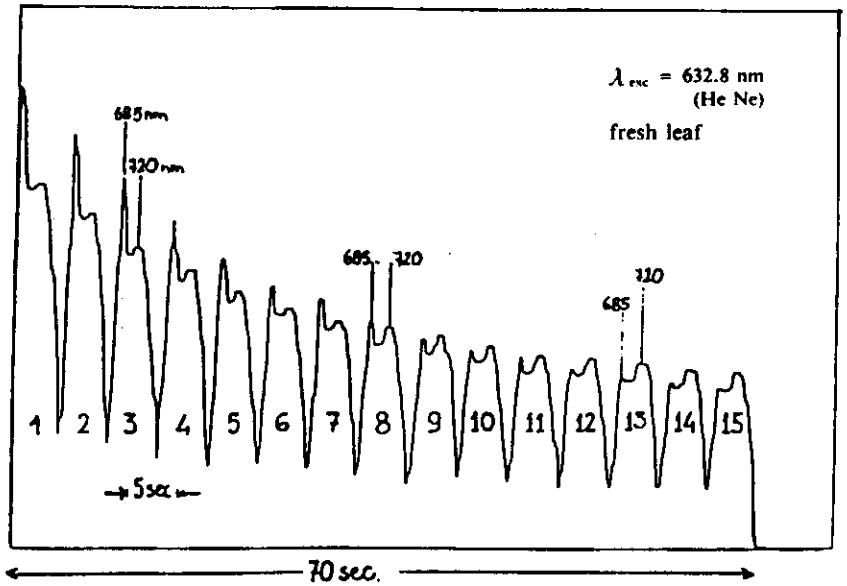

FIG. 8a

The spectra displayed in Fig. 8a refer to a fresh leaf, kept in dark for about 12 hours. The sequence of these fluorescence spectra shows an intensity quenching; moreover there is a progressive intensity reversal between the 685 and 720 nm bands as the time increases. The time decay is not well described by a single exponential; for example, the time behaviour of the 685 nm peak in the reported spectra is described by at least 2 exponential, with time constant of about 130 and 200 sec for the first and second part respectively.

Figure 8b is the analogous of Fig. 8a, but it is obtained from a fresh leaf exposed to solar radiation for 10 min. Solar radiation was simulated in laboratory by means of a solar-emission lamp with a power density of about 20 mW cm^{-2}.

The fluorescence intensity is almost time constant and the behaviour of each single spectrum corresponds to that of the last spectrum of Fig. 8a with 720 nm band higher than the 685 nm one.

Figure 8c shows the dry leaf case. The intensity level is greater than

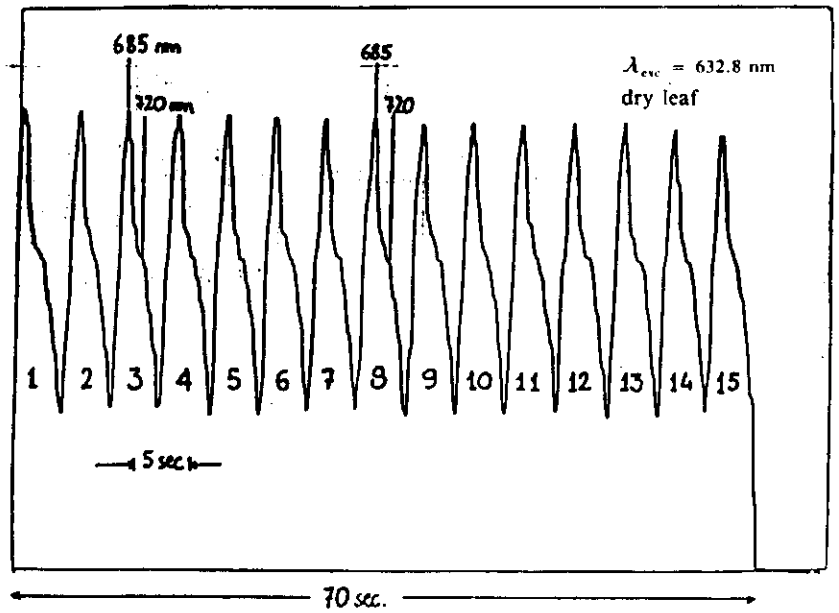

FIG. 8b

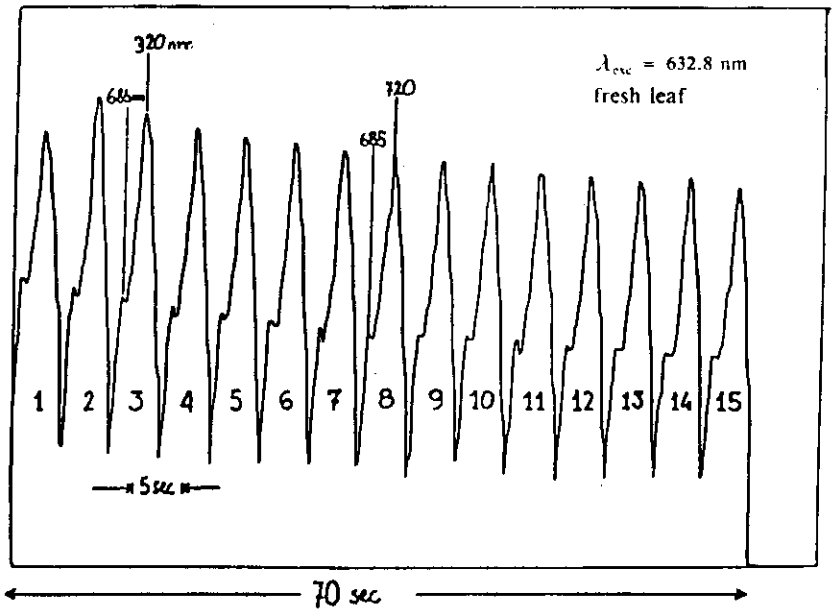

FIG. 8c

those of Fig 8a,b, moreover the fluorescence intensity is almost time constant, as in Fig. 8b, but here the 720 nm hand is lower than the 685 nm one, that is the opposite of what happens for a fresh leaf when fluorescence reaches its steady state. The spectra of a dry leaf result quite similar for exposition or not to solar radiation: the leaf does not react anymore to light, and photosynthetic process is inhibited.

On the basis of these preliminary results, other experiments were carried out. A fresh leaf was cut from the plant; then spectra were taken until dryness came out.

The results are shown on Fig. 9a: this figure represents the behaviour of the two peaks ratio (685 nm intensity/720 nm one) as the time increases.

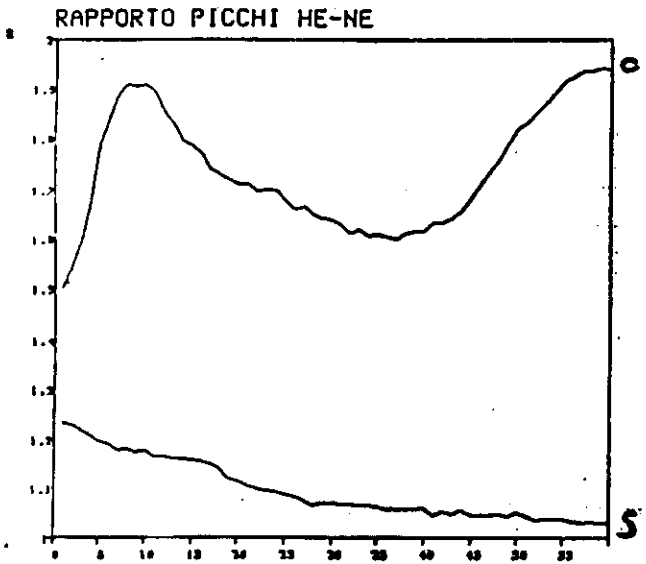

FIG. 9a

Two cases are reported:

1) the leaf was continuously exposed to solar radiation (c - curve);
2) the leaf was solar irradiated only during laser excitation (s - curve).

The fluorescence integrals of each spectrum taken in this kind of experiment are shown on Fig. 9b.

Then reflectance spectra were taken in the same way, but only with a solar lamp as exciting source (without laser exposure). The results are shown on Fig. 10; here the same ratio as in Fig. 9 is displayed versus time increasing. The denomination «c» and «s» refers to a continuous exposure to solar lamp all the experiment long and to a discrete exposure to solar

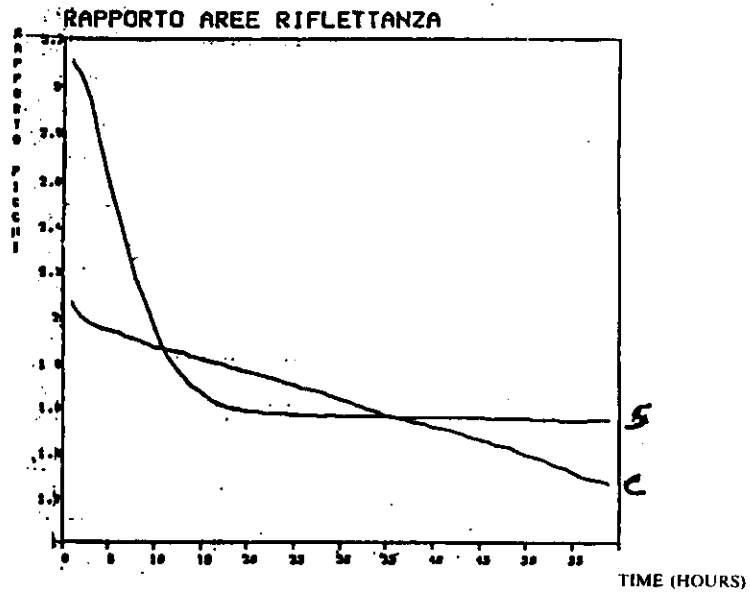

FIG. 9b

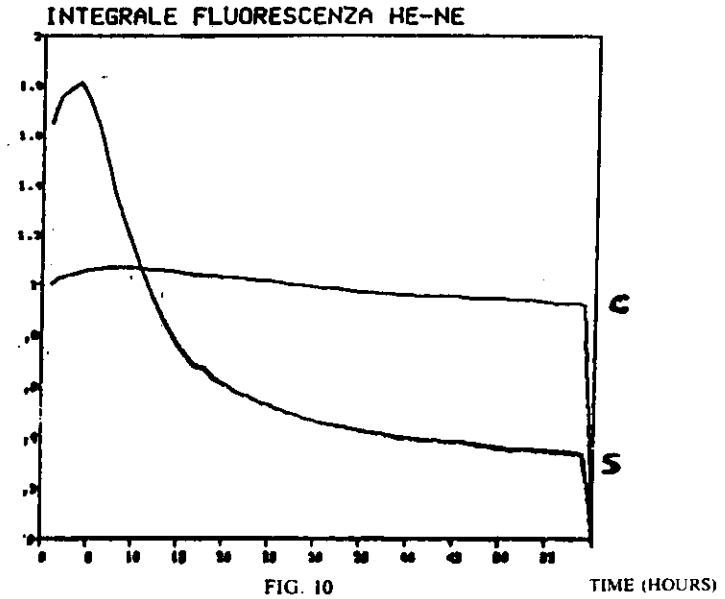

FIG. 10

lamp during only the time of recording a spectra.

From a comparison of the Figures 9b and 10 it is possible to see a concordant behaviour: the measure of the ratio of reflectance bands and the fluorescence area seems to give the amount of chlorophyll acting in the leaf.

As a conclusion it is possible to affirm that since the plant is a living structure, it reacts to the light excitation and fluorescence (like reflectance) results quite modified by the health state of foliage.

In fact for a fresh leaf, part of the impinging light energy is used in the photosynthetic process, which produces a decrement of fluorescence quantum efficiency; while for a stressed leaf a higher quantum efficiency is reached because of the photosynthetic process inhibition. This situation is showed in the fluorescence spectra of Fig. 11. taken at different stress leaf level.

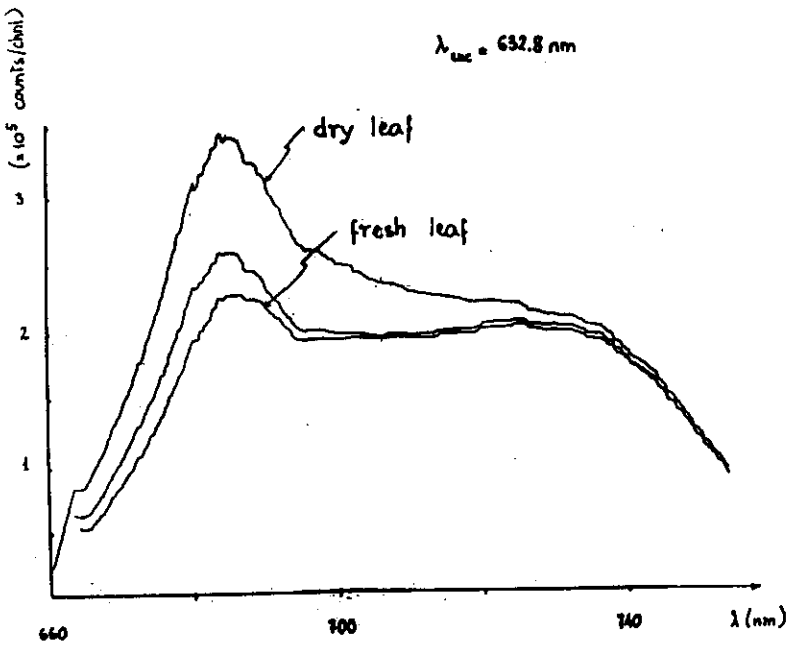

FIG. 11

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