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# Yellow Substance and Chlorophyll Measurements in the Venice Lagoon Using Laser-Induced Fluorescence

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## ABSTRACT

The Venice Lagoon is a particular environment hosting the historical Venice town, the main industries of Marghera, and a large traffic harbour. The water exchange with the open sea is mainly dependent on tidal movements. In this situation a continuous monitoring of the water quality is necessary to avoid, or at least to control, water eutrophication and pollution phenomena, which sometimes reach dramatic levels with sudden algal blooms and anoxia.

Within the framework of the CNR Project "*Salvaguardia del sistema lagunare Veneto*", two experimental campaigns were carried out to point out the usefulness of fluorescence lidar also in this particular marine environment. Both experiments successfully demonstrated the advantages of lidar remote sensing with respect to the traditional sampling and passive sensor techniques in the monitoring of the lagoon water quality. The results are comparable to conventional field measurements with the advantage that in measurements scheduled over an extended time, the need of maintenance is not as costly and tedious as for *in situ* instruments.

## INTRODUCTION

The Venice Lagoon is a vast morphological entity which extends some 50 km in length and 10 km in width, with an

area of about 550 km<sup>2</sup>. Water circulation is mainly controlled by tides through three wide inlets. The widest one, about 1 km wide, is at *Lido* and provides the water exchange in the whole northern lagoon region and part of the central one. This part of the lagoon hosts the town of Venice and faces the industrial area. Furthermore, some water courses flow into the lagoon. The *Dese* river, the most important one, has its outflow in the northern part of the basin, right near the *Burano* and *Torcello* Islands.

Venice Lagoon waters show an average salinity lower than that of the Adriatic Sea with a higher spatial variability depending also on atmospheric conditions. The lagoon waters can be considered as eutrophic and consequently highly productive. This fact brings about difficulties in the use of permanent immersed sensors which are rapidly covered by fouling and require a continuous maintenance. The use of remote sensors can avoid this problem. However, the lagoon is a critical environment for passive optical sensors for both its class 2 waters and shallow waters, which introduce an interference due to the bottom of the lagoon [1]. From passive data observed in shallow waters, or rather from the apparent property of reflectance, it is almost impossible to separate the contribution of the different constituents present in the water and bottoms while active systems on the basis of a selective fluorescent excitation seems probable. In this situation the use of active systems, such as fluorescence lidars, can be recommended.

Within the framework of the Project on the Venice Lagoon System, a research program, called “*Salvaguardia del sistema lagunare Veneto*”, was sponsored by CNR. It includes experiments with remote sensors to define their validity in the detection of those water-quality parameters useful for the study of sea-lagoon processes. Among these experiments, the lidar ones, carried out in the Summer of 1992, were particularly remarkable.

### 1. THE EXPERIMENTS

The locations of the two lidar experiments are shown in Fig. 1, where the location of the first experiment is marked and the ship route of the second experiment is indicated by dashes. The numbers show the sampling stations.

The first experiment was performed on July 13-15, 1992 at *Lio Grando* in *Punta Sabbioni*. This site was appropriately chosen at a point in the channel not too far from the Lido

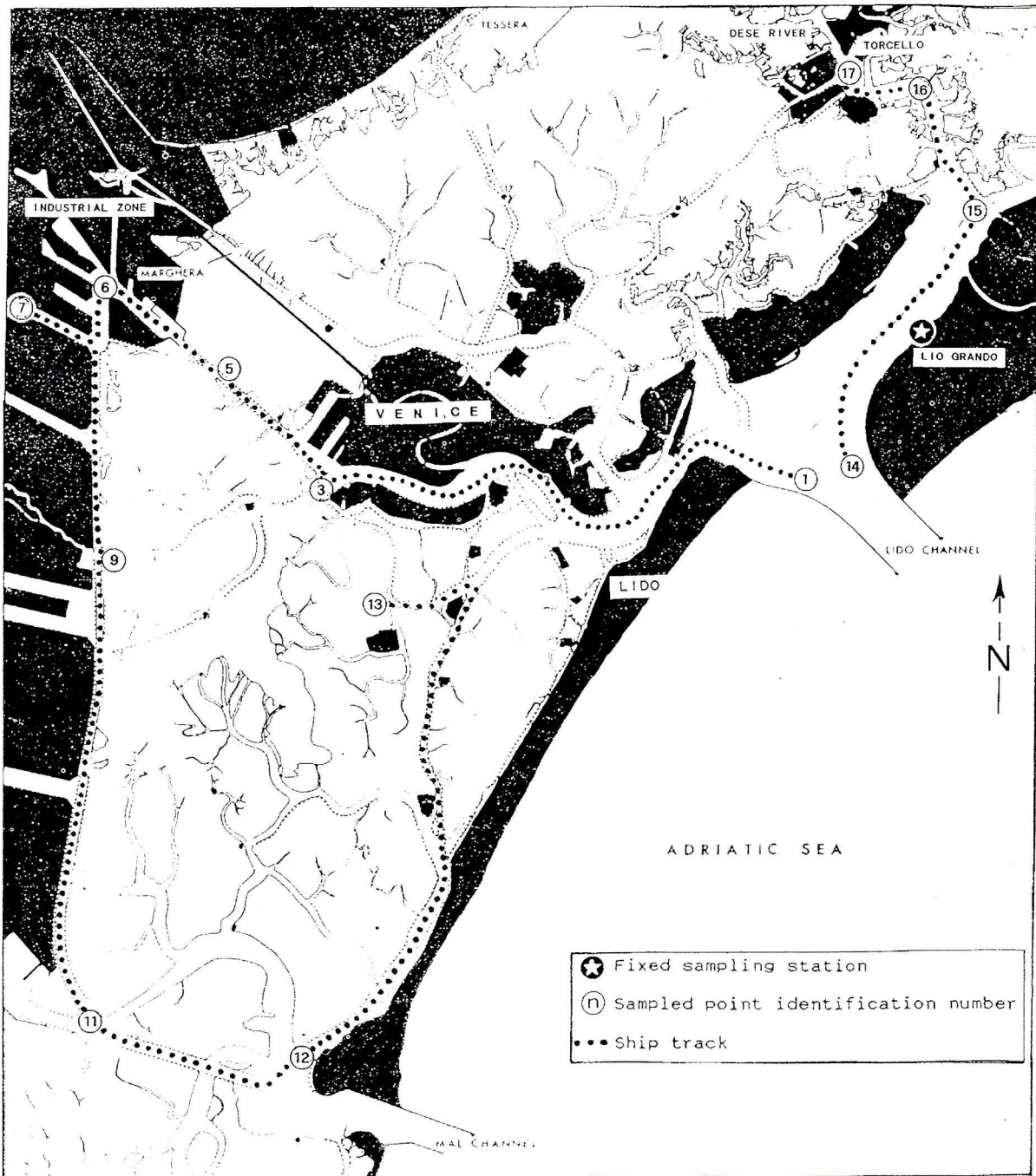


Fig. 1 - The Venice Lagoon with the experimental site of the first lidar experiment and the ship route during the second experiment indicated. Measurement stations along the route are indicated.

inlet. This channel represents an important hydric component controlling all the water exchange in the northern part of the lagoon, including the contribution of the *Dese* River. The purpose of the experiment was to study the water contents as a function of hydrodynamic phase with high spectral resolution fluorescence lidars during two tidal cycles.

Two fluorescence lidars, one [2] from the Lund Institute of Technology (LTH) and the other one [3] from the Istituto di Ricerca sulle Onde Elettromagnetiche (IROE), were operated from a pier extending out toward the centre of the channel (Fig. 2). The lidar measurements were performed during a 24 hour cycle and at the same time *in situ* measurements of water quality parameters were carried out by the research vessel "Litus" of the Istituto per lo Studio della Dinamica delle Grandi Masse (ISDGM). Remote Raman and fluorescence data on turbidity, DOM (Dissolved Organic Matter), and chlorophyll were recorded and compared with the corresponding *in situ* data. Some experiments were also done for the detection of lagoon bottom vegetation.

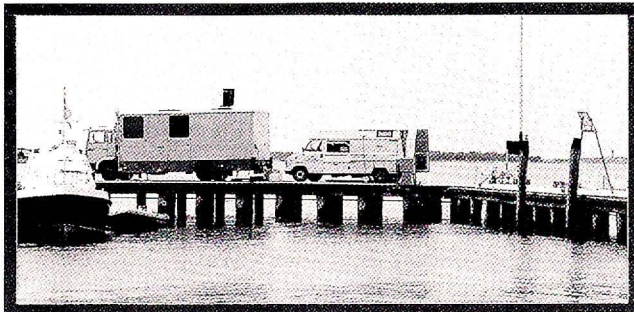


Fig. 2 - A photograph showing the two lidar systems operating from the pier, and the *Litus* moored for data taking.

On August 26-27, 1992 a campaign was carried out using the CNR O/5 (Oceanographic Ship) "D'Ancona" and a lagoon boat. The O/S and the boat travelled together inside the lagoon: the O/S was devoted to *in situ* measurements while the boat carried the IROE lidar-van (Fig. 3). The main objective of this experiment was the test of a high spectral resolution fluorosensor in different lagoon environments, with the additional objective of detecting the spatial variability of water quality parameters in quasi-synoptic conditions. Raman signal, chlorophyll and DOM fluorescence were detected along the path shown in Fig. 1, which included all the most significant parts of the lagoon waters.

## 2. THE MEASUREMENT SYSTEMS

The lidar systems used in the lagoon experiments were described in other papers [2-4]: therefore, only a very short

description of them will be given here, while a more detailed one will be given for the *in situ* measurements.

### 2.1 LIDAR Systems

The IROE FLIDAR-3 (Fig. 4) is a high spectral resolution fluorosensor. For these experiments the lidar was hosted in its dedicated van with motor-generator power supply. The laser beam was directed vertically into the water by means of a folding mirror: in the first experiment the mirror was placed on a tripod on the pier and in the second one on an aluminium frame reaching out in front of the boat.

Fig. 5 shows examples of remote fluorescence spectra of water detected by the FLIDAR-3 with 308 nm (a) and 480 nm (b) excitations.

The fluorescence lidar of the LTH is also hosted in a van and can be used both in a spectrally resolved mode and in a multi-colour imaging mode [4] (Fig. 6). The emitter of the system is a Nd:YAG laser, which is frequency tripled to give 355 nm radiation. The beam is sent out via a retractable dome on the top of the van. The folding mirror in the dome

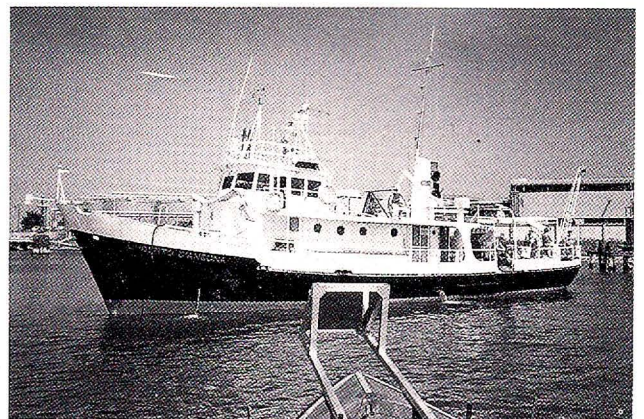
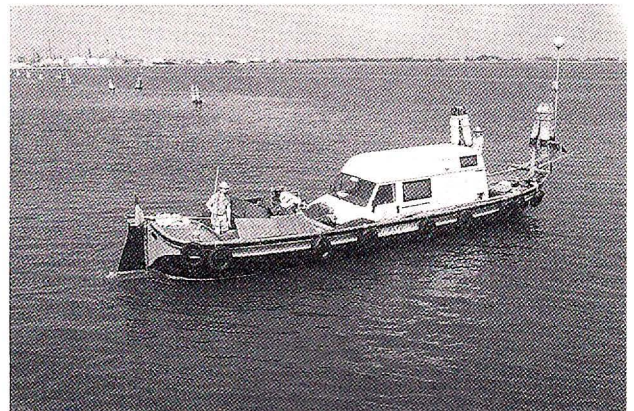


Fig. 3 - a) The lagoon boat hosting the FLIDAR in the Venice lagoon. b) The O/S D'Ancona, taken from the lagoon boat. In the foreground the folding mirror of the FLIDAR, deflecting the laser beam into the water.

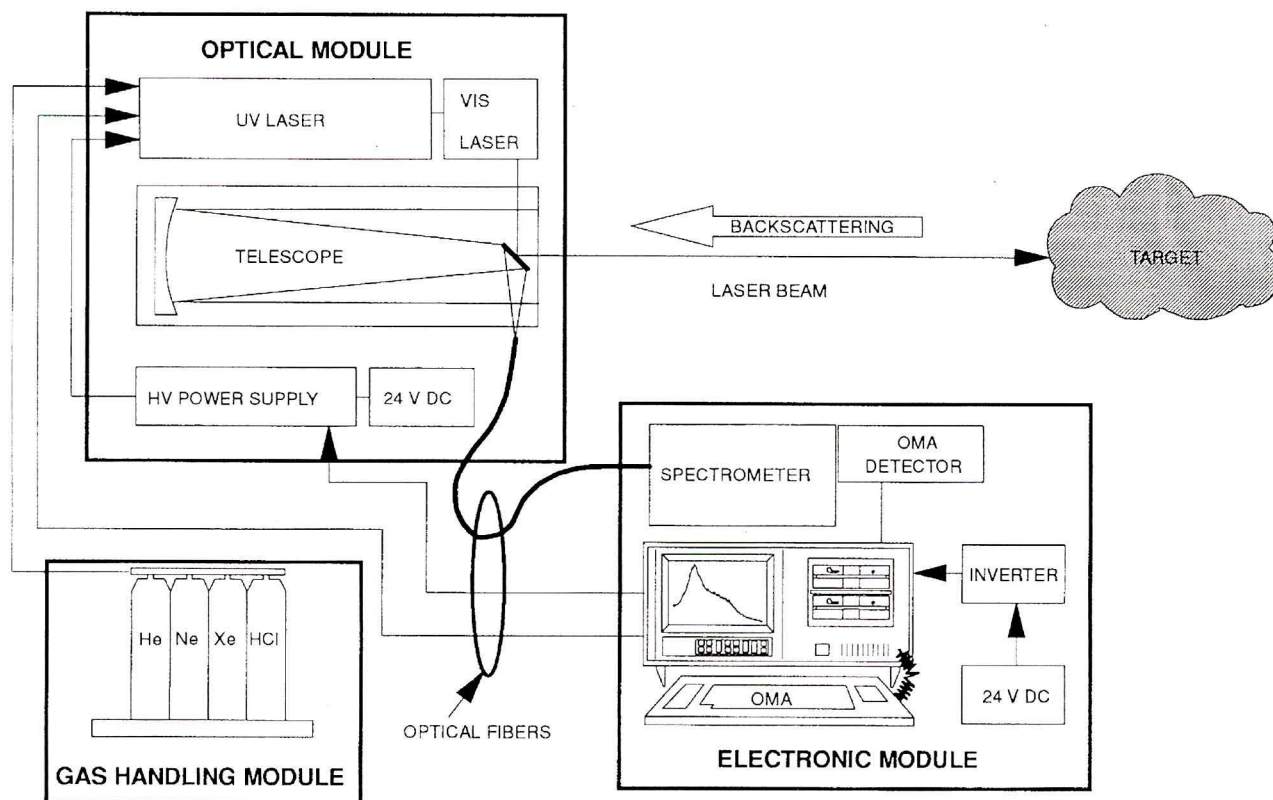


Fig. 4 - Schematic diagram of the IROE FLIDAR-3.

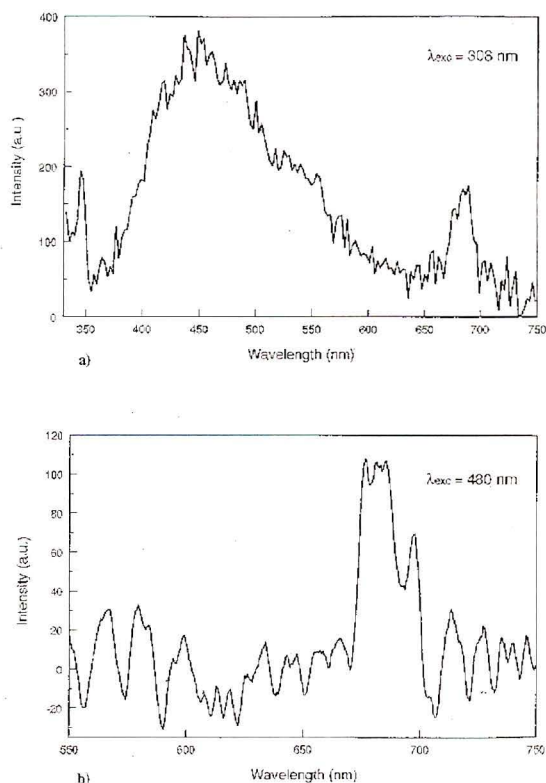


Fig. 5 - Two spectra detected with the IROE system for FLIDAR-3 (a) 308nm and (b) 480nm excitation.

is computer controlled and can direct the beam 360 degrees horizontally and 45 degrees vertically. The backscattered light is received by a vertically mounted Newtonian telescope with a diameter of 40 cm. In the focal plane of the telescope a flip-in mirror is mounted to direct the light into an optical fibre connected to an optical multichannel analyser system.

To measure in the multi-colour imaging mode the mirror is removed and the light is allowed to enter the split-mirror Cassegrainian telescope of the imaging system, where four individually filtered images are formed on a CCD detector, which is preceded by an image intensifier (Fig. 7).

During the *Lio Grando* experiment the laser beam was directed to the water both vertically by means of a folding mirror placed on the pier or directly in a slant direction.

A high resolution remote spectrum detected with the LTH lidar system in the spectrally resolving mode for 355 nm excitation is shown in Fig. 8. The water OH-stretch Raman signal (denoted by *r*) can be seen superimposed on the broad blue-green fluorescence light distribution due to DOM. The peak intensity of the DOM signal is denoted by *b*. Further, a chlorophyll signal can be observed at 690 nm. Since this signal is frequently weak, it is important to evaluate the background free intensity *c* rather than the total intensity *c'*.

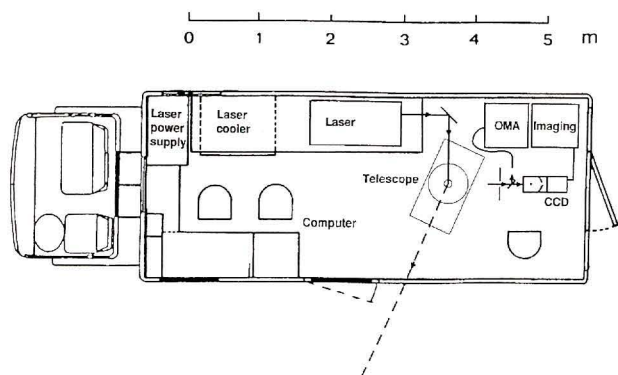


Fig. 6 - Diagram of the LTH lidar van. The laser beam is emitted via the dome on the roof of the van: The fluorescence light collected with the Newtonian telescope is detected by either the OMA (spectrally resolved) system or the imaging system.

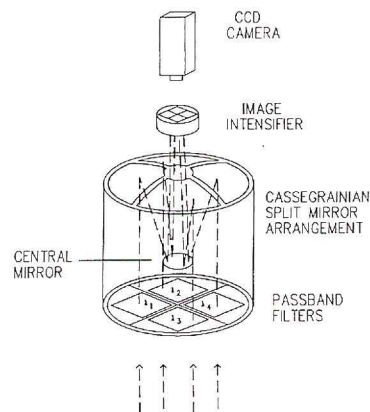


Fig. 7 - Cassegrainian split-mirror telescope mounted on an image-intensified CCD camera. In front of each of the four sections of the first mirror, optical filters are placed to permit simultaneous imaging in four different wavelength bands.

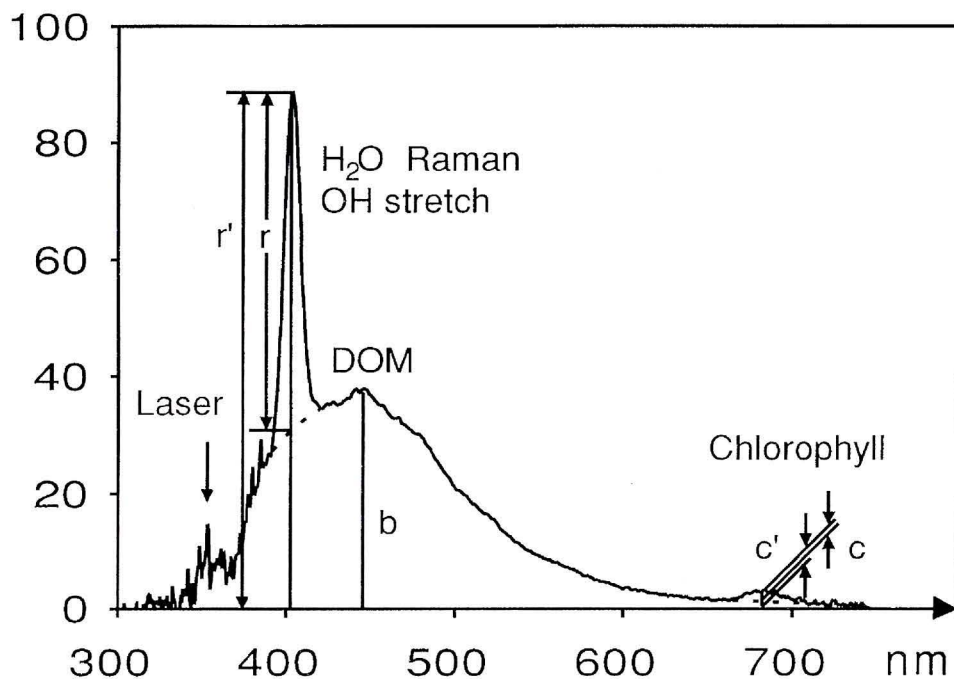


Fig. 8 - An example of a high-resolution spectrum detected with the LTH lidar system. The denotations are explained in the text.

An example of a remote fluorescence recording using the system in the imaging mode is shown in Fig. 9. Since no geometrical image information is expected from the natural water surface, the imaging detector is here used only for intensity recordings. In the figure the round imprint on the water surface of the expanded laser beam is shown at different intensities corresponding to the filter passbands chosen. From recordings of the type shown in Fig. 8 and Fig. 9 for the same water area it was possible, by also taking the experimentally determined filter profiles into account, to evaluate from the imaging detector the quantities  $r$ ,  $b$  and  $c$  with elimination of the background fluorescence signal levels.

### 2.2 In-situ measurements

Direct measurements were carried out on the water simultaneously with the lidar ones. The "Litus" was equipped with a system for the continuous monitoring of water samples taken at about 40 cm below the surface. The parameters, measured every 15 minutes, were: conductivity, salinity, temperature, surface IR temperature, turbidity, fluorescence, chlorophyll, and DOM. The measurement set was completed with the standard meteorological data such as wind speed, humidity, etc.

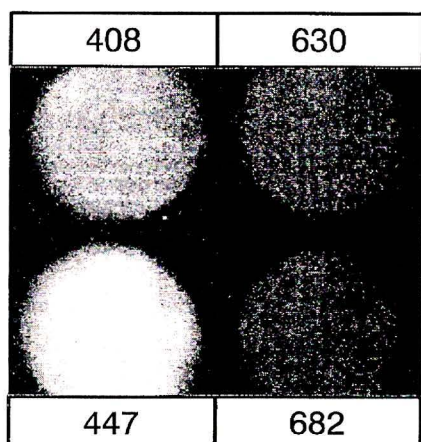


Fig. 9 - An example of a remote fluorescence recording of water in imaging mode. The spatial resolution is not used in this mode, just the intensity.

Measurements of turbidity were carried out by a Hach/Ratio 2000 turbidimeter with a circulating cell intercalibrated with standard solutions of *formazin*. Chlorophyll *in vivo* and DOM were measured on two Turner 111 fluorimeters with flow cells. The excitation wavelengths were 450 nm and 340 nm, respectively, with corresponding fluorescence detection bands at 670 nm and 450 nm. The data are in arbitrary units.

Every hour samples were taken for the laboratory determination of chlorophyll, TSM (Total Suspended Matter), and DOM. The chlorophyll content was computed according to the Strickland and Parson method, the TSM by weight, and DOM by absorption between 320 nm and 440 nm.

The same type of equipment was used on board the O/S "D'Ancona" during the second experiment. The samples for laboratory investigations were taken in correspondence of the stations indicated in the map of Fig. 1.

### 3. EXPERIMENTAL RESULTS

From a general point of view the comparison between the lidar measurements and the *in situ* data shows good agreement. Some discrepancies may be attributed to the distance between the point in which the *in situ* measurements were carried out and the location where the laser beam entered the water. Even if this distance was taken as small as possible, it may play an important part, especially in the measurements done at the *Lido* inlet. Actually, the variation of the parameters is mainly determined by the tidal level and topographical position of the sampling point. These aspects determined a shift between the trend of the measured tide level and the trend of the parameters. Due to the oscillating movement of the sea water in the lagoon, the same water is

measured twice when the tide changes direction. Actually, the last outgoing water from the lagoon (during low tide) comes back at the beginning of the incoming tide.

#### 3.1 Lio Grando experiment

Fig. 10 shows the change of the chlorophyll and DOM signals with the lidar beam orientation with respect to the pier, as measured with the LTH system.

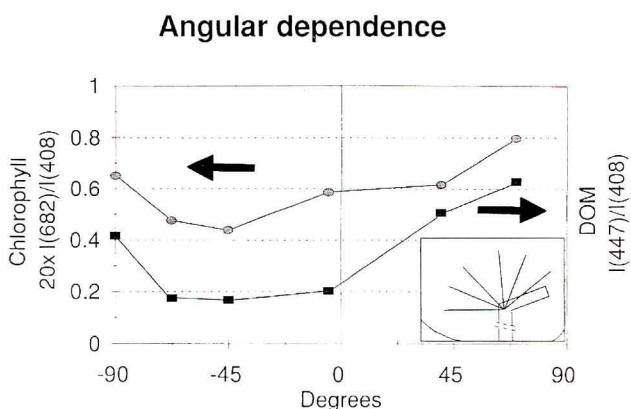


Fig. 10 - The normalized chlorophyll and DOM fluorescence measured as a function of angle with the LTH lidar system. The lidar system was parked at the outer end of the pier, as shown in the insert.

Fig. 11 shows the *in situ* turbidity and the inverse of the amplitude of the Raman peak as a function of time, as measured with the IROE system. The behaviour of the two curves is similar. The differences may be attributed to some algal patches which were floating on the water surface, particularly at the time of the maximum tidal flow.

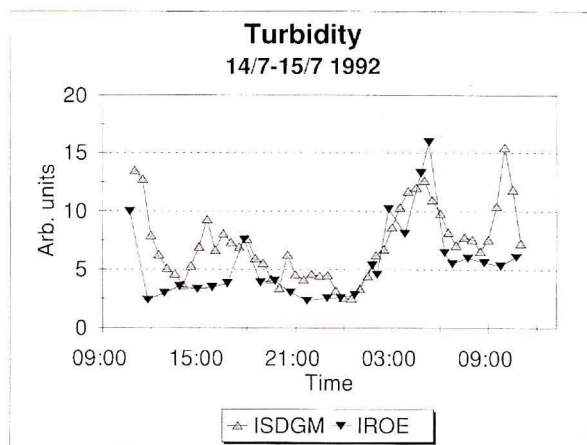


Fig. 11 - The turbidity measured as a function of time with the IROE lidar system and the ISDGM point monitoring system.

Figs. 12 and 13 show data for DOM and chlorophyll measured with the IROE and the LTH lidar system, respectively. Data on the tide status are also included.

Fig. 12 shows data taken with the IROE system. The data are evaluated as the peak fluorescence intensity for DOM at about 450 nm, divided by the intensity at the Raman peak. For the chlorophyll data, the background free intensity (see Fig. 8) at 685 nm is divided by the Raman peak intensity. This procedure eliminates the influence of the effective sampling volume, determined by, e.g. turbidity and slant angle. The peak that can be seen in the chlorophyll panel at three o'clock in the morning of the 15th is certainly due to floating benthic algae.

In Fig. 13 the data of the LTH system are shown. In the two upper panels of the figure the black squares denote measurements made with the spectrally resolved system, and the

grey ovals denote measurements made with the imaging system. The data of the spectrally resolved system are evaluated in the same way as that for the IROE system. The data from the imaging system are evaluated in a similar way, but in order to do a correct background subtraction, the data were fitted together with the data from the spectrally resolved system in the points where they coincided in time. In the evening of July 14 1992 the spectrally resolved system failed, but the measurements could continue with the imaging system. It was thus important to be able to fit the data of both systems together in order to complete the daily cycle measurements. The LTH data cover a longer time span than the IROE data. As can be seen, the DOM and chlorophyll data are quite similar, but they are slightly out of phase with the tidal cycle.

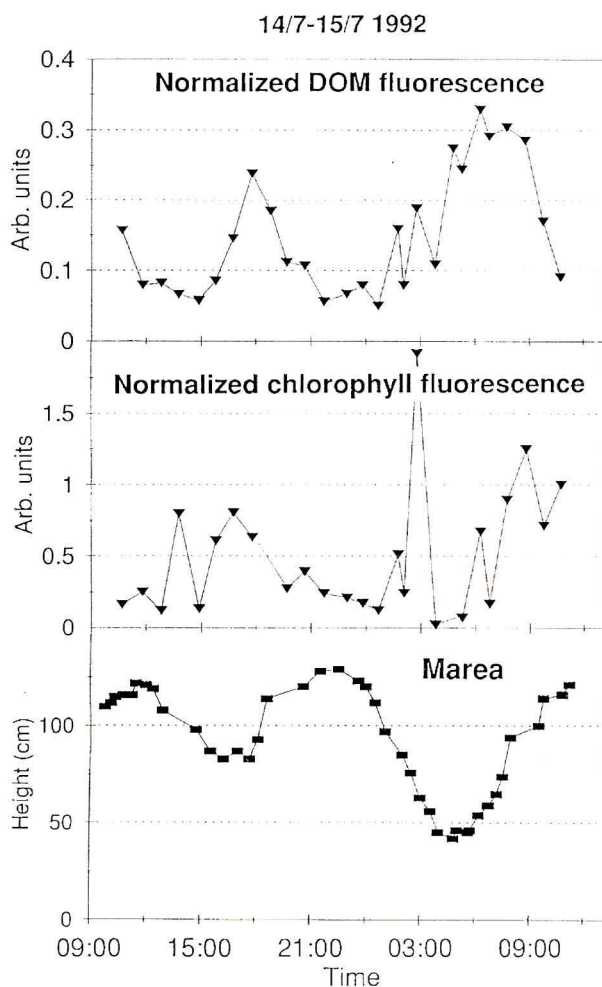


Fig. 12 - The normalized chlorophyll and DOM fluorescence measured during one day and night with IROE lidar system. The bottom panel shows the tide.

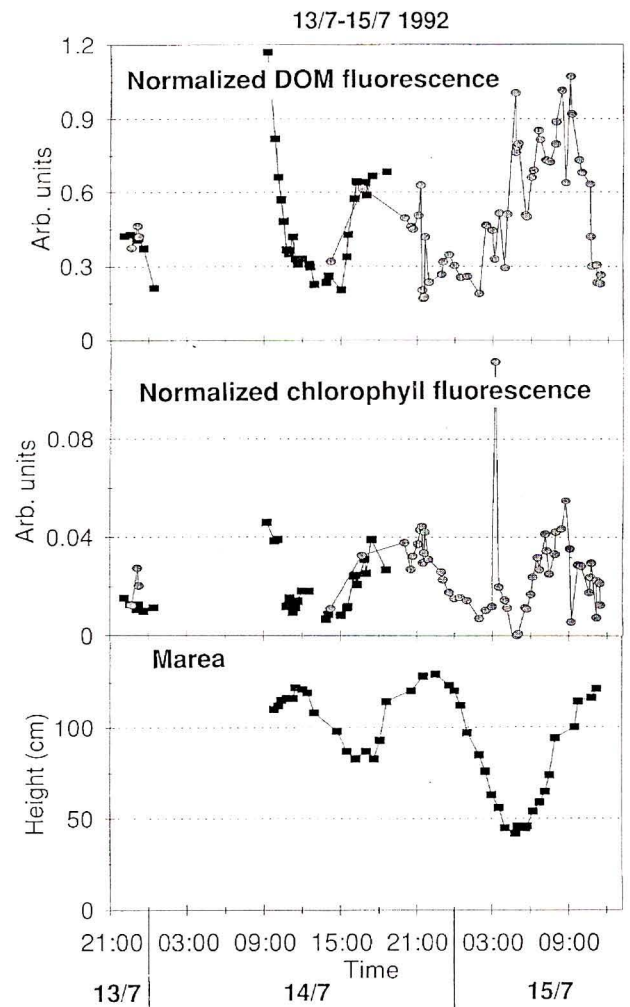


Fig. 13 - The normalized chlorophyll and DOM fluorescence measured versus time with the LTH lidar system. The black squares denote measurements performed with the spectrally resolved system, and the grey ovals denote measurements performed with the imaging system.



Normalized DOM data from both lidar systems and from the *in situ* sampling are compared in Fig. 14. Since the *in situ* data had an unknown offset value the shape of that curve has been fitted to the lidar data. As can be seen, the agreement is very good.

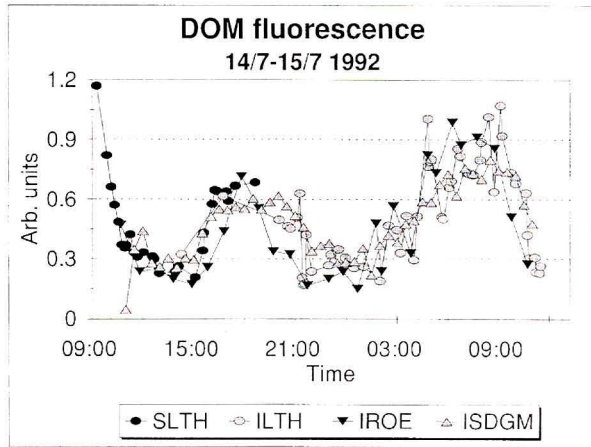


Fig. 14 - The DOM fluorescence measured as a function of time with all systems during 24 hours.

Fig. 15 shows the fluorescence spectra of two benthic algae samples. These samples were floating in front of the pier. They were fished and placed, without drying them, on the pier for the measurement. The observed structure of the spectra is due to the pigments' content, which is different for the two macroalgal samples. Fig. 15 (a) refers to a green alga (*Zoostera*) which contains only chlorophylls. The spectrum shows the typical fluorescence peaks, due to chlorophylls, ranging from 685 nm to 750 nm. Fig. 15 (b) shows the spectrum of a red alga (*Gracilaria*), which contains also phycoerythrin and phycocyanin. The presence of these pigments is in fact responsible for the two fluorescence bands at 580 nm and 660 nm, respectively.

In Fig. 16 an imaging recording of three algal samples lying on the pier is shown: the same algal samples are reproduced in the photograph in the lower right part of the picture. The laser beam was made divergent to cover a circle with a diameter of about 60 cm. The samples are *Ulva Rigida*, *Zoostera* and *Gracilaria*. In the two left parts of the multi-colour fluorescence image, corresponding to 408 nm and 447 nm, the water signal can be seen as a semicircle. The spectrum for water is shown in the lower left part of the picture. The reason for the signal of 447 nm being stronger than the one of 408 nm is that the sensitivity of the detector

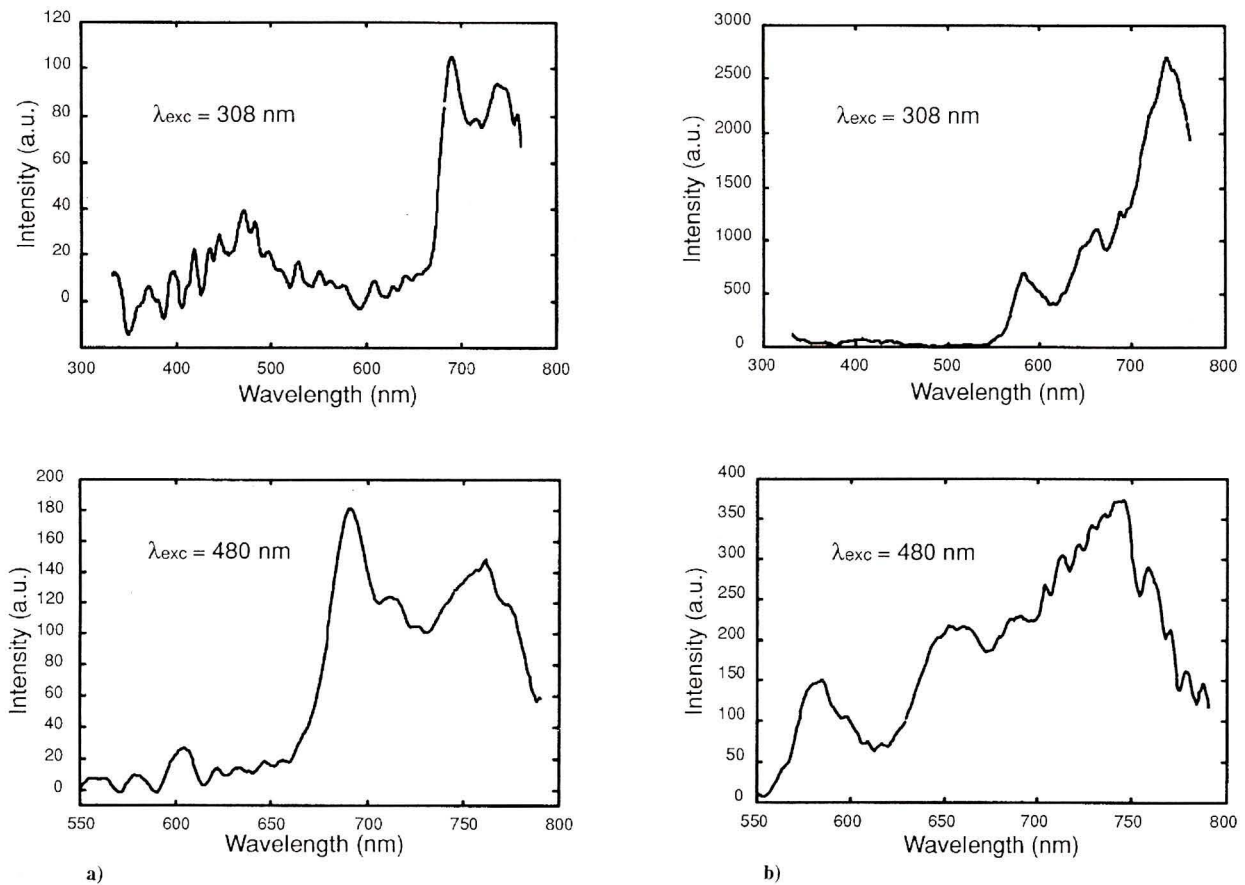


Fig. 15 - Fluorescence spectra of two types of benthic algae, both with 308 and 480 nm excitation. a) *Zoostera*, b) *Gracilaria*.

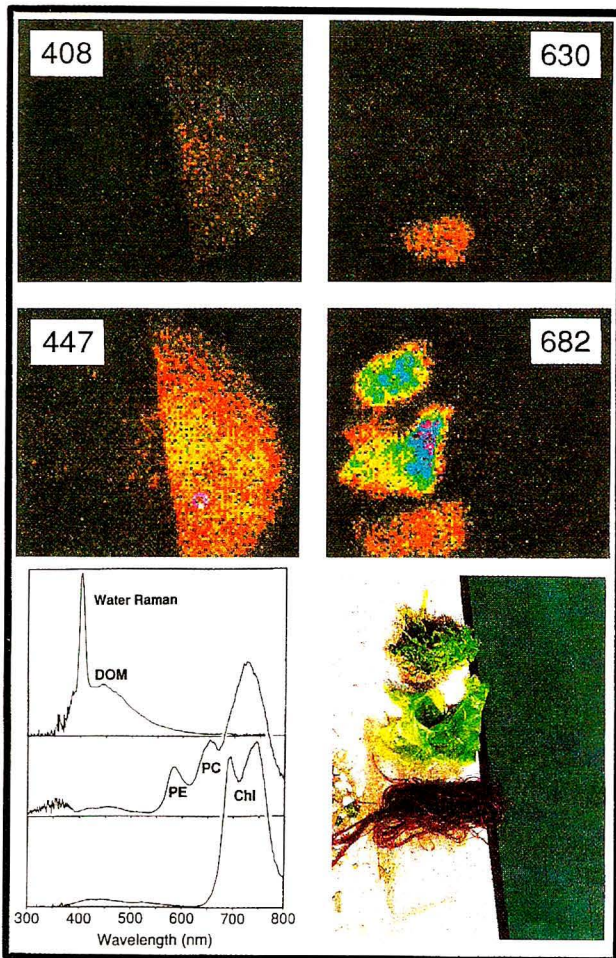


Fig. 16 - An image recording of three algal samples lying on the pier. The spectra are fluorescence recordings of water, the red alga and one of the green algae. The chlorophyll fluorescence can be seen for both algae, but for the red alga also peaks corresponding to phycoerythrin and phycocyanin are seen. A colour photograph of the scene is also shown. The measuring distance was about 20 m.

is less at 408 nm than at 447 nm. In the lower right quadrant of the multi-colour fluorescence image, corresponding to 682 nm, all three algae can be seen. The filter is chosen so as to match the chlorophyll peak. Remote fluorescence spectral recordings of green and red algae are given in the lower left part of the figure. In the upper right quadrant only one alga can be seen, the red one. Red algae contain phycoerythrin as well as phycocyanin, that overlap with the selected filter, centered at 630 nm.

### 3.2 Lagoon experiment

In the lagoon cruise experiment a good agreement was generally found between the lidar and *in situ* data. Some of the discrepancies, as mentioned before, can be attributed to

the distance between the “D’Ancona” and the lidar boat. This fact could be remarkable especially in some cases with crossing ships, which cause a sudden turbidity of the water.

Fig. 17a shows the lidar data on chlorophyll, DOM, phycoerythrin, and phycocyanin for the five stations indicated in the map of Fig. 1. These stations are particularly representative of the lagoon environment since the first one is near the center of the town, the second one is near the *Lido* inlet, and the last three ones are in the northern part of the lagoon. These last three stations correspond to three different areas: two are quite closed while the third one is near the *Dese* estuary where the main part of the fresh water enters the lagoon.

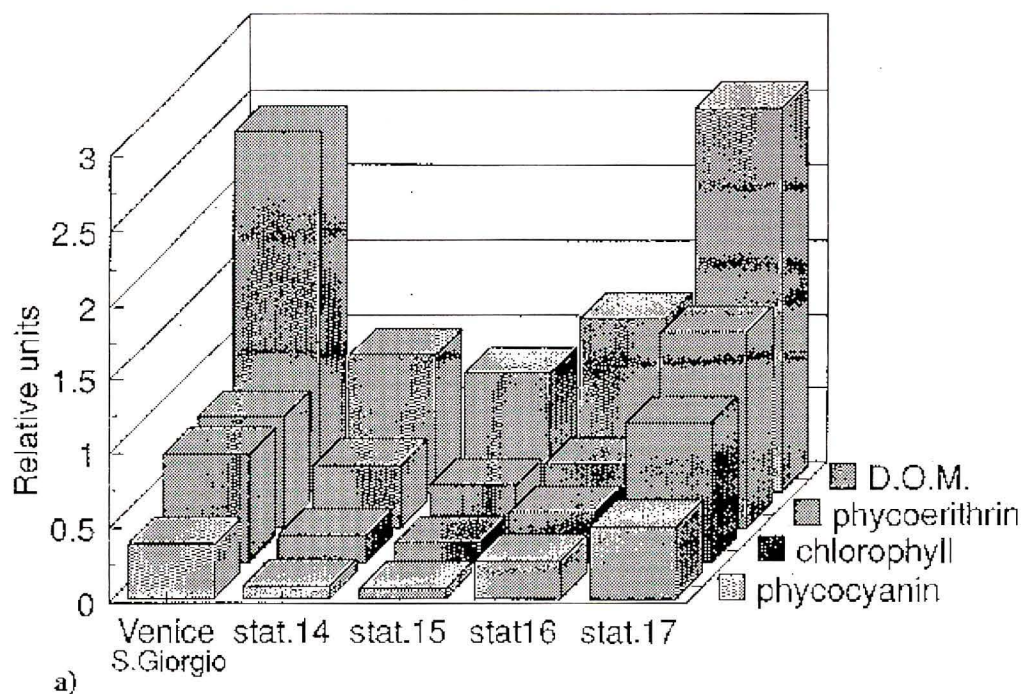
Fig. 17b shows examples of continuous “sea-truth” measurements performed from the “*Litus*” ship. The left panel shows chlorophyll fluorescence and the right panel shows DOM fluorescence. The sampling points along the FLIDAR track shown in Fig. 1 are marked by small numbered circles.

## 4. CONCLUSIONS

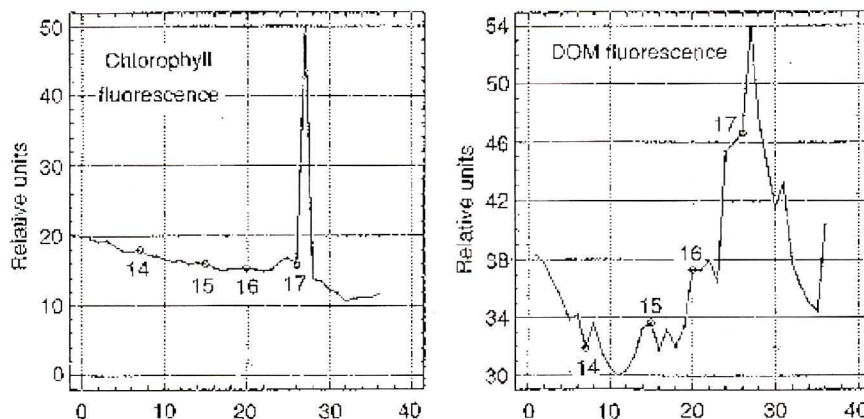
Even if a deeper analysis of the data is necessary and further experiments are needed, the results of these two experiments were:

- A fair agreement between the *in situ* data and the lidar measurements.
- The identification of some spectral signatures for the detection of benthic algae by high spectral resolution lidars and by multicolor imaging lidar.
- The monitoring of the variation of the water quality parameters during a tidal flux.

These results demonstrated that the fluorescence lidar with high spectral resolution is a suitable sensor for the remote control of the lagoon water quality, both from dedicated boats and coastal platforms. That was also confirmed and extended by the flying tests carried out in 1992-93 with the FLIDAR-3 system [5,6]. The use of a lidar monitoring eliminates the problems connected with immersed sensors and the ambiguities which are present in passive colour images in this particular environment.



a)



b)

Fig. 17 - Data at five sampling stations in the Venice lagoon.

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