LASER AND RADIOACTIVE IRRADIATION AS STRESS SOURCES OF THE COTTON PLANTS AND PHYTOPLANKTON

Dmitry Yu. TSIPENYUK

General Physics Institute of Russian Academy of Sciences, 117942, Vavilova str.38, Moscow, Russia <u>TSIP@KAPELLA.GPI.RU</u>

KEY WORDS: Remote sensing, Real-time, Multy-spectral, Monitoring, Laser/Lidar.

ABSTRACT

The paper describes investigations devoted to study of the possibility of using phytoplancton as a natural sensor of radioactive pollution in the water. It was shown that under influence of radioation in fluorescence spectra of the phytoplancton specific changes occurred. Thus it is possible to use remote sensing methods for express locating areas of radioactive pollutions in the ocean. Also in present paper we investigated possibility to initiate stress in cotton plants by impulse power lasers beam.

1. INTRODUCTION

One of important ecological problems of our world is unfortunately radioactive pollution in the ocean from sources of different types (Wiley 1992). For successful solving of this problem it is important to locate pollution areas precisely using remote sensing methods (Colton 1994). To solve the problem we try to use fluorescence spectra of phytoplankton. It is well known that phytoplankton exists everywhere in the ocean. On the other hand, phytoplankton is very sensitive to temperature, soil concentrations and different chemical pollutions in water. It was shown that the shape of the phytoplankton's fluorescence spectra is very sensitive to the variations of the above mentioned parameters (Bunkin 1984). In other works we can find investigations devoted to monitoring of photosynthetic ozone stress by laser induced fluorescence (Rosema 1992) and laser induced fluorescence in relation to water stress, chlorosis (Chappelle 1984). So we can use phytoplankton as the natural sensor in many cases. It is also well known that under the influence of radioactive radiation different kinds of alive objects appeared qualitatively very similar responses. Under the influence of low dose radiation there were appeared different kinds stimulation effects of radiation (Kuzin 1977) and in the case of high radioactive doses there were stagnation effects (Kudryashov 1982). In the experiments performed we investigated possibility to observe changes in the state of phytoplankton using the changes in the fluorescence spectra of phytoplankton under the action of radiation (mainly high energy gamma-radiation) in a large scale of radiation doses from 15 to 300 000 roentgens. For this purpose we compared fluorescence spectra of samples of phytoplankton that were grown and maintained under the same conditions (lighting, temperature, etc.) but were irradiated in different flux density and the only difference between these samples was different radiation dose obtained.

2. EXPERIMENTAL

The fluorescence spectra of phytoplankton were excited with the nitrogen laser emitting at 337 nm, repetition rate 50-1000 Hz, mean power of one pulse was about 15 kW. pulse duration 6-8 ns. An optical system consisted from two mirrors which directed laser radiation at the temperature stabilised open quartz cup with the sample of phytoplankton. Total flux of photons was about 10 photons/(cm² \cdot s) avoiding damage of phytoplankton and saturation of fluorescence. Fluorescence from the phytoplankton was focused with the help of two quartz lenses onto the entry of the polychromators slit of optical multichannel

spectrum analyser (OMA). A diffraction grating with a relatively weak dispersion (150 grooves/mm) was used in receiver to record simultaneously spectra in a rather wide range of wavelengths (370-720 nm). Spectral region of the spectroanalyzer's sensitivity was from 180 to 950 nm. The sensitivity of the detector was at maximum in the vicinity of 430 nm, where it amounts to 6 photons per count. The samples of phytoplankton for our investigations were grown in the Microbiological Institute of the Russian Academy of Sciences. Phytoplankton samples (in particular Synechococcus cedrorum) were

especially domesticated under appropriate conditions during several days and then were sloped into several small glass cups. As a result we have phytoplankton samples of the same age and of the same stage of a growth. One sample was used as the control one and other samples were irradiated by different flux of radiation. During the experiments special attention was paid to the keeping all the samples of the phytoplankton under the same temperature and lighting conditions. We usually tried to use samples of phytoplankton as fast as possible after obtaining the phytoplankton from the biological laboratory. Usually the timetable of our experiments was the following : 1-3 hours were over after taking phytoplankton from the source before we began to irradiate samples (this usually took about two hours), and one hour later we were already able to measure phytoplankton's fluorescence spectra. All phytoplankton samples were irradiated by laser light during several minutes before the measurements of fluorescence of leafs or phytoplankton, that occurs when a dark adapted leaf is suddenly illuminated) (Rosena 1992).

For gamma-irradiation of the phytoplankton we used bremsstrahlung with endpoint energy 30 MeV or gamma-rays from (Ra-Be)- neutron source. The substance with a phytoplankton was carefully mixed in a flask to ensure its homogeneous distribution over the volume and then was transferred into small glass vessels 25 ml in volume. A few of these vessels were set up at different distances from the bremsstrahlung target (or radioactive (Ra-Be) source). To obtain reliable quantitative results all samples were simultaneously irradiated by different fluxes of gamma-quanta. The control sample was protected by Pb shield from the radiation, but was also situated in the same room. Thus, all samples were all the same time in the same temperature and lighting conditions, but at different radiation fields. In such a way we could avoid possible errors due to different states of phytoplankton and temporal changes of gamma-radiation.

The maximum radiative dose in a vicinity of the bremsstrahlung target was equal approximately to 300 000 roentgens, but the control sample that was out of the radiation field was in the condition of natural background. The phytoplankton was not only exposed by bremsstrahlung when the electron accelerator was in operation, but also by a consequent irradiation from the induced radioactivity in glass and water. The residual radioactivity was due to \Box^+ -decay of oxygen-15 nuclei that were a result of the nuclear reaction ${}^{16}O(\Box,n)$ ${}^{15}O$ and consisted of accompanying 0,511 Mev annihilation quanta. The residual gamma-radioactivity of a glass was resulting from aluminium isotopes formed from a silicon by the reaction ${}^{29(30)}Si(\Box,p)$ ${}^{28(29)}Al$. Total irradiation time of the samples was about 10 min. The level of induced radioactivity was also used for relative comparison of radiative doses received by different samples during the irradiation. After 2-3 hours when the induced radioactivity of the samples were reduced up to the safe level, the samples were transferred to the laboratory for laser induced fluorescence (LIF). Measuremments in the case of irradiation of the samples from (Ra-Be) source, time of irradiation was about 20-23 hours and during 10 hours from that time phytoplankton was additionally eliminated by a light source. The temperature during irradiation was stable (about 20 degr.(C)). Measurements of LIF were made 1-2 hours after the end of irradiation.

3. RESULTS AND DISCUSSION

The LIF of phytoplankton's samples (in particular Synechococcus cedrorum) has several wide maximums between 400 and 750 nm. These maximums are close to 450, 480, 650 and 685 nm (figure 1a).

The maximums at 650 and 685 nm were attributed to the fluorescence of photosystem and some pigments. The two wide maximums in the region 450-480 nm were attributed to some organic substances which were suspended inside and outside (in the surrounding water) the phytoplancton.

It is worth to underline that only the fluorescence maximum near 450 nm was found in the initial solution of several necessary mineral soils solved in a distilled water - in which the phitoplankton was growing up.

When the phytoplankton was seeded in the initial solution and began to grow up, the fluorescence maximum near 480 nm and certainly the maximums at 650 and 685 nm were appeared in addition to the total LIF spectra.

It was observed that all above mentioned fluorescence peaks were very sensitive to the state of the phytoplankton. For example, it was shown that the correlation between temperature of the phytoplankton and LIF took place. Under the best optimal temperature region near 25 degr.(C) (the temperature under which the phytoplankton was grown up) the LIF in regions 450-480, 650 and 685 nm had a minimum in compare to the LIF at nonoptimal temperatures. And in opposite, in the case of temperature changing down from optimum value (to 20 degr.(C)) or continuously up to 45 degr.(C) (stress temperature) the LIF intensity in all regions were continuously and reversibly increased.

These results are in accordance with the principle that under the optimal conditions the LIF of alive photosynthetic organisms has a minimum due to the maximum efficiency of photosynthesis under this conditions (Rosema 1992).

We found that the relative LIF changes in the region 450-480 nm were in several times more than the changes at 650 and 685 nm, although approximately the same qualitative LIF behaviour in all regions was observed.

In the case of phytoplankton irradiation from the radioactive source the stimulation effect was founded under relatively low doses.

We found that there is stimulation effect from (Ra-Be) source which is clearly observed in LIF of the phytoplankton when we compare of the control sample LIF spectra and the spectra of the irradiated sample. The most dramatic changes there are in the region 450-480 nm (diminishing LIF intensity in comparing with control sample about several tens of percents). Much less changes we found in the region 650-685 nm (only several percentages of diminishing intensity of LIF in comparison with the control sample).



Figure 1 - changes in the laser induced fluorescence spectrum of the phytoplankton samples under the action of radiation. Figure 1a - control sample, figure 1b - stimulation effect of low dose (about several tens roentgens), figure 1c - stimulation effect of middle dose (about several hundreds roentgens), figure 1d- stagnation effect of high dose (several hundreds thousands roentgens).

When two different samples were simultaneously irradiated by different low level doses (about 15 and 50 roentgens) we found that the stimulation effect of the irradiation was qualitatively proportional to the dose received. The most intensive was the LIF of the control sample, thr intermediate intensity has LIF of the sample which received about 15 roentgens and the lowest intensity belongs to the LIF of the sample that absorbed about 50 roentgens.

Thus we can see that the effect of stimulation is qualitatively proportional to the radiative dose received by the phytoplankton.

During the experiments with bremsstrahlung at the microtron the same kind of stimulation effects was also found at relatively low doses (up to several hundreds of roentgens) figures 1 b,c. In the case of high dose (figure 1d, several tens or hundreds thousands absorbed roentgens) stagnation of phytoplankton, decay of organic matter and appropriate decreasing of the LIF in the region 450-480 nm (decay under direct influence of radiation) and increasing of LIF in region the 650-685 nm (stress conditions) were observed.

We also investigated possibility to initiate a stress in cotton plants by impulse power laser beam. Three experimental and one control groups of four-month-old cotton plants were irradiated by light of the second harmonica of YAG:Nd laser (wavelength 532 nm, laser pulse duration 10 ns, pulses frequency 1.5 Hz). First and second groups were irradiated during 2 and 5 minutes with impulse power 100 kW and third group was irradiated during 15 minutes by 1 MW pulses. The treatment of plants causes the acceleration of leaf fall, which depends on the total irradiation intensity. The high level of correlation (more than 99%) in leaf apparatus state and treated plants with high dose of irradiation has been shown (Voronkov 1988).

4. CONCLUSIONS

We have found in our experiments that very specific changes were relevant in the fluorescence spectra of phytoplankton under influence of radiation in registered range of wavelengths 400-750 nm.

It has been shown that LIF of phytoplankton under influence of radiation appear the same qualitative behaviour as under other types of stress that affect on the phytoplankton state.

The lowest investigated dose was about 15 roentgens but we need to take into account that we measured LIF not more than 24 hours after radiation influence. Probably in the case of continuos growing up of phytoplankton in the vicinity of less intensive radiative source during a long time some well observed changes in a phytoplankton state and hence in LIF will occur.

Thus the laboratory experiments show the possibility to use active and passive remote sensing methods of registration of phytoplankton's fluorescence for express remote location areas of radioactive pollutions in the ocean from satellites or aircrafts.

ACKNOWLEGEMENTS

This work was impossible without great assistance and collaboration from a lot of peoples from the Microbiological Institute and the Institute for Physical Problems, Russian Academy of Sciences.

REFERENCES

Bunkin, F.V., Vlasov, D.V., Gerasimenko, L.M., Slobodyanin, V.P., 1984, Use of Phytoplancton Fluorescence Spectra as a Sensor of the Nonfluorescencing Substances. Sov.J. Kvantovaya Electronica, II, No.6, pp. 1253-1256, (in Russian).

Chapelle, E.W., Wood, F.M., McMurtrey, J.E., and Newcomb, W.W., 1984, Laser Induced Fluorescence of Green Plants.1: A Technique for Remote Detection of Plant Stress and Spies Differention. Applied Optics, 23,134-138.

Colton, D.P., and Louft, H.L., 1994, Location and Identification of Radioactive Waste in Massachusetts Bay. Proceedings of the Second Thematic Conference Remote Sensing for Marine and Coastal Environments held in New Orleans, USA, on 31 January- 2 February 1994, Vol.I, pp.70-84.

Kudryashov, Yu.B., Berenfeld, B.S., 1982, Principals of the Radiation Biophysics. (Moscow University Press) (in Russian).

Kuzin, A.M., 1977, Stimulation Influence of the Ionisation Radiation at Biological Objects. (Moscow: Atomizdat) (in Russian).

Rosema, A., Cecchi, G., Pantani, L., et.al., 1992, Monitoring Photosynthetic Activity and Ozone Stress by Laser Induced Fluorescence in Trees. Int.J.Remote Sensing, Vol.13, No.4, 737-751.

Tsipenyuk, D.Yu., 1992, Development of Laser Remote Sensing Methods Based on Fluorescence and Emission Spectra. Ph.D. Thesis General Physics Institute, Moscow, pp.142.

Voronkov L.A., Tsipenyuk D.Yu., 1988, Influence of laser irradiation on leaf apparatus of cotton plants. Biological scienses (in Russian), N 11, pp.24-27.

Wiley, D.N., et al., 1992, Location Survey and Condition Inspection of Waste Containers at the Massachusetts Bay Industrial Waste Site and Surrounding Areas.U.S. EPA/Region I: Grant No.X001549-01.0. North Falmouth, Massachuset: International Wildlife Coalition.