Real-time Monitoring of Pathogens and Nanomarkers in Water using Resonance Laser Spectroscopy Techniques

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INTRODUCTION

Close Corporation SERVET is a multifunctional commercial enterprise, which develops new technologies and produces equipment in the fields of housing and public utilities, environmental protection and health care, nature management and mining operations.

During past ten years our company has been involved in research and development programs in the sphere of medical diagnostics and treatment, manufacturing new drugs and medical equipment, intended for timely detecting infectious diseases and identifying qualitative and quantitative characteristics of infectious agents both in a human organism and in other environments (water, soil, foodstuff, premises, etc).

Having completed a long research work containing different aspects of physics, virology and microbiology, we came to an obvious conclusion, that, the physio-chemical properties (characteristics) of various pathogens (viruses, bacteria etc.) are strictly specific for each stage of their development in an organism or environment

We studied VIS-near IR spectra of a number of pathogens (salmonella, herpes genitalis, hepatitis A and C, grippe viruses A and B) as well as water solutions of nanosilver and superluminescent nanomarkers. The research results enabled us to work out an operating model for a unique medical device for complex noninvasive and aliquot-free express diagnostics (i.e. without blood test and/or laboratory sampling of human tissue) for viruses and infectious disease agents. Currently we are developing equipment for monitoring of drinking water. [1,]

In our contribution, we discuss a new method, allowing real-time monitoring of a number of pathogens in water.

Keywords: SBS, toxicants, laser monitoring, liquid medium.

1 EXPERIMENT

In our experiments, the laser radiation that passed through a quartz cell filled with water solutions of the pathogens, nanosilver or nanomarkers was analyzed by the spectrum analyzer "Agilent", which provides a spectral resolution of 0.5 nm. The solid DPSS lasers (Laser-Export Co. Ltd., Russia) and the semiconducters lasers with the wavelengths of 1017, 810, 670, 532, 480 nm were used as the sources of exciting radiation. The Fig.1. shows the scheme of the experimental stand.

We researched water solution of pathogens (E- coli, strain K12 salmonella, goldish staphylococcus coliphage MS-2 and etc.) with concentrations 10, 10^2, 10^3, 10^4, 10^5, 10^6, 10^7, 10^8, 10^9 m.c/ml (lg tds50). In order to reveal effects, which can impede automatic data-processing, we examined probes of potable water which was given from five different areas of Russia. We analyzed about 700 spectra of the pathogens for exciting wavelength 810 nm, about 100 spectra for wavelengths 670 and 1017 nm.

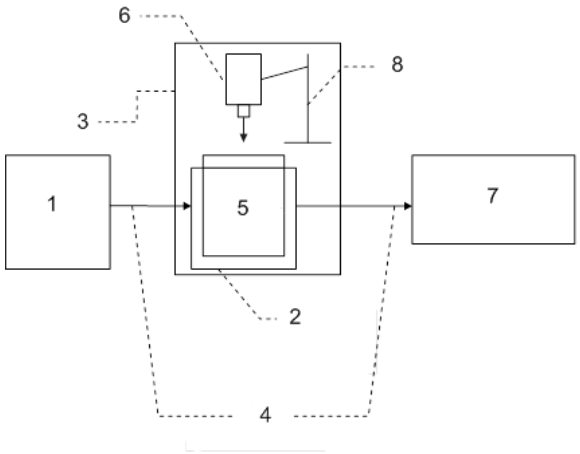


Figure1: The scheme of the experimental stand.

The main setup of this stand is following:

1-the block of the three of semiconductor laser sources with exciting wavelengths λ1 =1017 nm, λ2 = 810 nm, λ3=670 nm; 2-the opaque cuvette holder with windows for input and output of radiation; 3-waveguides for input-output of radiation; 4- the quartz cuvette; 7-spectrum analyzer "Agilent" with the spectral resolution 0,05 nm, equipped with the microcomputer for processing spectra; 8-the additional semiconductor laser source with wavelength λ4 =532 nm; 9- the protective shroud; 10-the holder of the laser source with wavelength λ4.

The input waveguide brings one of theexciting radiations (with wavelength λ1, λ2 or λ3) to the cuvette, containing the sample. If the laser with wavelength λ4 was used as exciting source, then its radiations was propagated through air to cuvette. In this case, laser with wavelength λ4 and cuvette with sample was put in the protective shroud 9. The radiation, which passed through the cuvette and output waveguide, was analyzed by spectrum analyzer "Agilent".

2 THE STUDY OF WATER SOLUTION OF PATOGENS

We examined the different regions and the water showed that the impurities in drinking water, do not lead to the appearance of the Stokes and anti-Stokes components of the spectrum.

We investigated E-coli solutions to receive informative parameters for automatic identification of their existence in water. We found that, if concentration of E-coli is less than 10^3, the peak was Stokes. The peak was anti-Stokes, if concentration of E-coli was from 10^3 to 10^7. The peak was Stokes, if concentration of E-coli exceeded 10^8.. We found out that dependence of peak position from concentration same if solution was stimulated by the laser with wavelength 1017 nm.

We found out, that exciting radiations with wavelengths 1017 and 810 nm induces the stimulated Brillouin scattering in our spectra of the matter, containing pathogen DNAs. Threshold power level for appearance the stimulated Brillouin scattering was achieved by electromagnetic fields from the exciting radiation and the stimulated fluorescence. . We found out that peak positions and widths of are "fingerprints" for pathogens under study, and optical densities of these bands is proportional pathogen content, if this content is less 15%. Thus, Stokes and anti-Stockes bands of the stimulated Brillouin scattering can be used to recognize the pathogens under studies. As example, Fig. 2, 3, 4 show spectra of radiation, passed through the cell with physiological solution) and water solution of mixture of grippes A and B, respectively/ Fig. 4 shows the radiation spectrum goldish staphylococcus and coliphage MS-2.

We researched spectral characteristics of these patogens in water in an amount necessary for the initial identification with 95% probability. Software and device model were built for the optical detection of this object.

For the diagnostics of nanomarkers we suggest to analyze the radiation spectra from matters which are irradiated by two lasers. One of the lasers provides the exciting radiation in near IR region (810 or 1017 nm), the other provides the exciting radiation in visible region (480 or 512 nm). In near IR region, we detected the stimulated fluorescence in the radiation spectra of nanomarkes in water solution being irradiated by these lasers. For example, Fig. 4 shows the radiation spectrum of HEX nanomarkers exposed to the 1017 and 480 nm radiation.

We recorded in the database of the series spectra of salmonella and of salmonella, E-coli, staphylococcus aureus coliphag MS-2 and etc. These studies have allowed us to create a setting automatically detects the presence of pathogenic organisms directly in the water flow.

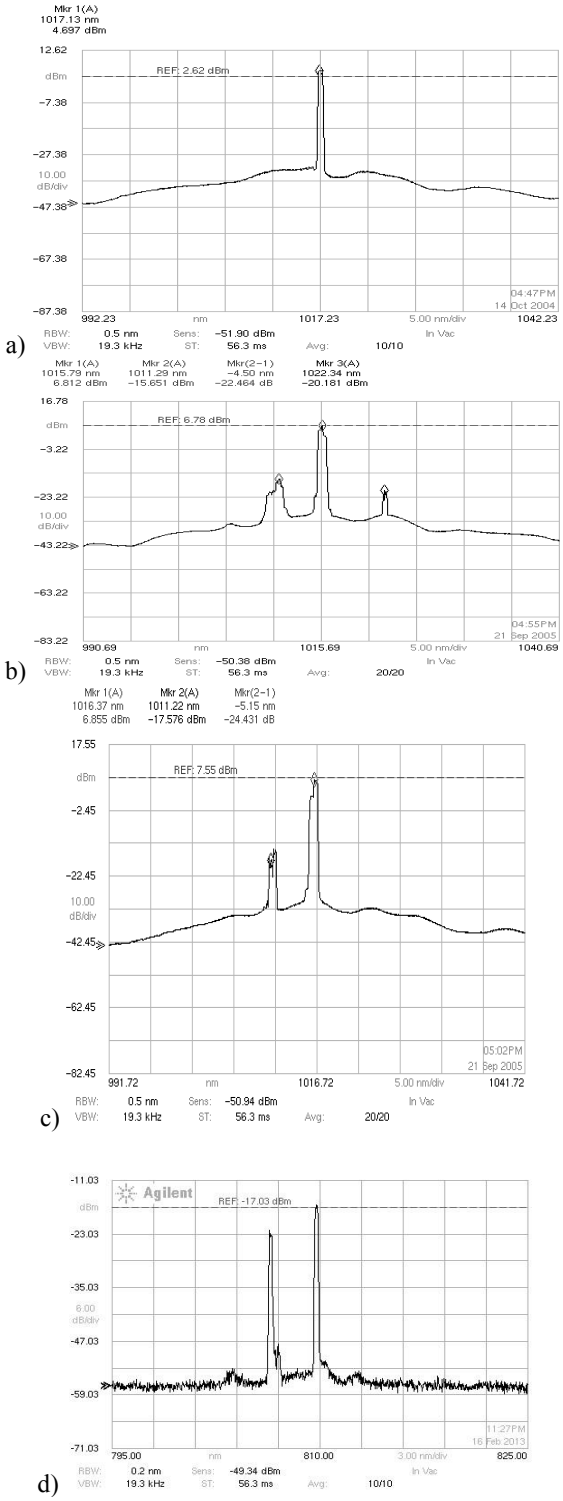
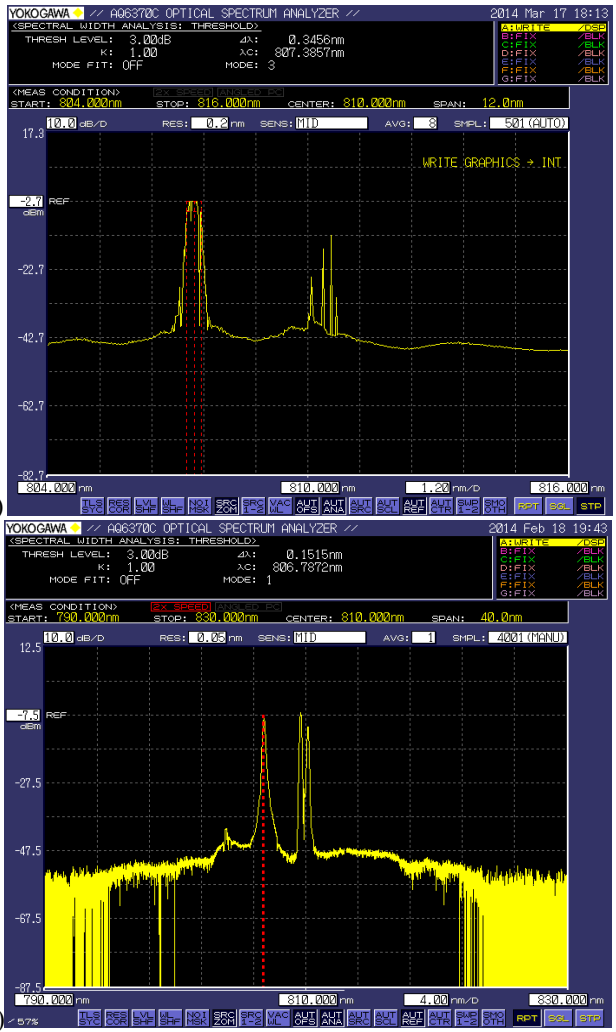


Figure 2: The spectra of radiations, passed through the cell:

with water (a) water solution of mixture

of grippes A and B (b), water solution of herpes genitaler (c).



a)

b)

The spectra of radiations, passed through the cell: with water: a- goldish staphylococcus,

b- coliphage MS-2

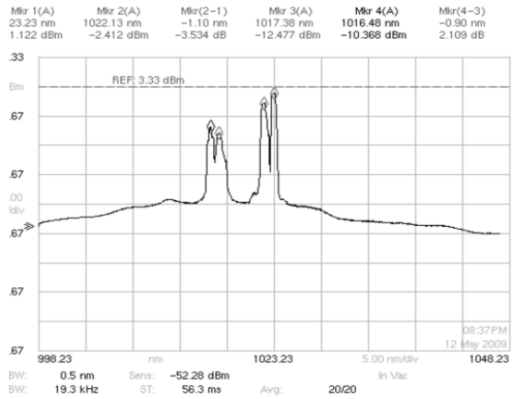


Figure 4: The spectrum of radiation water solution of HEX nanomarkers at excitation by 1017 and 480 nm.

3 CONCLUSIONS

We developed a new method that combines the method of forced luminescence and stimulated Brillouin scattering.

We carried out the statistical spectral analysis of water from different regions in order to determine the statistical errors of the method. The analysis of spectra of water from different regions has shown, that there are not parasitic peaks of a luminescence of water impurity in the chosen range of frequencies (750-1400 nm). The mistake caused by own noise, does not exceed the 0.01%. Therefore the frequency of maximum of peak of intensity can serve as informative parameter at recognition of an image.

We researched spectral characteristics of E coli, salmonella, goldish staphylococcus and coliphage MS-2, in water in an amount necessary for the initial identification with 95% probability.Software and device model were builtfor the optical detection of this object.

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