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DETECTION OF BIOLOGICAL AEROSOLS USING INDUCED FLUORESCENCE LIDAR

Seminar

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Abstract

Induced fluorescence light detection and ranging (IF LIDAR) has come of age as a tool for exploring the atmosphere. Transmitting ultraviolet (UV) light and detecting the wavelength-shifted UV fluorescence (UVF) a number of interesting measurements is achieved. Fluorescence emitted by biological material enables distinction between biological and nonbiological aerosol, emission of metal elements makes possible observations of temperature and wind speed in the middle atmosphere. This paper reviews basics of scattering processes, properties of biological material constituents responsible for fluorescence, principles of common LIDAR and form of induced fluorescence LIDAR, examples of its different employability and also theoretical detecting range for real induced fluorescence LIDAR is calculated. Following article was stimulated by activity at Nova Gorica University where induced fluorescence mobile LIDAR was developed.

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Introduction

Over century elapsed since World bear witness to first detection of remote objects by radio-waves. At the outset success was just feasibility of detecting the presence of ship in dense fog without determination of its distance. Since prospective science is instantly perceived and sponsored major development was stimulated by World War II, when technology, already noted as RADAR, was fully exploit as a British defence against German aircraft attacks.[23]

Furthermore, invention of the laser contributed RADAR promotion to LIDAR. As abbreviation LIDAR instead of Radio-wave Detecting And Ranging stands for Light Detecting And Ranging basic RADAR principles of transmitted pulse and range gating its returned signal remained, but difference appeared in emitted wavelength. LIDAR has been from the very begining strongly connected with the progress in optical and electronics technology, consequently modern LIDAR techniques developed the potential to detect parts comparable to light wavelenght at distances of many kilometres. Provision of good spatial, temporal resolution without interference in the processes occuring in air makes LIDAR heighly adeqate for atmospheric research, particularly in meteorology.

Unfortunately war industry development and progressive production of weapons of mass destruction is in proportion to general educational standard, disposal of science and techonology knowledge. Recent ascertainment claims that biological weapons pose a real and potentially immediate modern threat. Several factors contribute to usage of these bioagents. First, the very horror associated with biological weapons serves to make them attractive to modern terrorist groups and has tremendous potential impact as terror. Equipment and knowledge needed to produce these bioagents is relatively to provideand also its manufacture is not expensive. Not the last tempting reason rests in small quantities of bioagents needed to cause huge numbers of casualties.[21]

With regard to upper example of quickened development of RADAR due to war every weapon needs antiweapon or its effective detection. Our new detection challenge represents reduced size of endangering aircrafts to a few microns. Likewise, there is high priority for the military to develop the ability to provide early warning of a remote biological warfare agent attack so that protective measures can be taken before personnel are subjected to infectious or lethal doses of the aerosol cloud. One way to achieve this goal is to use LIDAR sensors that can detect the presence of an aerosol cloud at ranges out to 10 km and discriminate whether the cloud is biological or nonbiological at ranges out to 3 to 4 km.[21] A LIDAR system's discrimination mechanism relies on transmitting ultraviolet (UV) light and detecting the wavelength-shifted UV fluorescence (UVF) that is produced by all biological material. LIDAR systems currently cannot identify a particular biological warfare agent attack within an aerosol, yet they can trigger alarms of possible threat and precaution can be taken as special sensors could be sent to eventually polluted area. Seminar

presents the principles of induced fluorescence LIDAR (IF LIDAR), in addition it reviews theoretical range of IF LIDAR developed at University of Nova Gorica.

Induced fluorescence in biological aerosols

Interest in activity and structure of atmospheres finest particles sooner or later introduces us with scattering processes. At first we should know rough distinction to elastic and inelastic scattering. Rayleigh and Mie scattering processes are elastic processes in which no appreciable energy exchange takes place between the scatterer and the incident photons. Elastically scattered energy has the same wavelength as the incident radiation. It is customary to differentiate between the Rayleigh regime, in which the scatterer is much smaller than the illuminating wavelength, and the Mie regime, in which scatterer is larger or comparable to the wavelength. In the case of inelastic scattering, there is an exchange of energy between the scatterer and the incident photons resulting in a scattered radiation at a wavelength differing from that of the incident radiation. Inelastic scattering includes resonant scattering, ordinary fluorescence, and Raman scattering. Resonant scattering entails absorption at a wavelength corresponding to an electronic transition and re-emission at the same wavelength. Raman scattering results from excitation of either the vibrational-rotational or the pure rotational transitions. Reemission takes place at wavelengths lower and higher than the wavelength of the incident radiation.[30]

1.1 Fluorescence process

As title of current paragraph announces our interest is in fluorescence. Fluorescence usually refers to absorption of radiation at a particular wavelength and re-radiation at one or more longer wavelengths, and can apply to either atoms, molecules or nanostructures.[30] Light is emitted from electronically excited states created by either a physical (for example, absorption of light), mechanical (friction), or chemical mechanism. Generation of luminescence through excitation of a molecule by ultraviolet or visible light photons is a phenomenon termed photoluminescence, which is formally divided into two categories, fluorescence and phosphorescence, depending upon the electronic configuration of the excited state and the emission pathway. Fluorescence is the property of some atoms and molecules to absorb light at a particular wavelength and to subsequently emit light of longer wavelength after a brief interval, termed the fluorescence lifetime. The process of phosphorescence occurs in a manner similar to fluorescence, but with a much longer excited state lifetime.[10] Process of monochromatic light absorption and re-emission at different wavelength illustrates Jablonski diagram, as shown in *Figure 1*. [27]

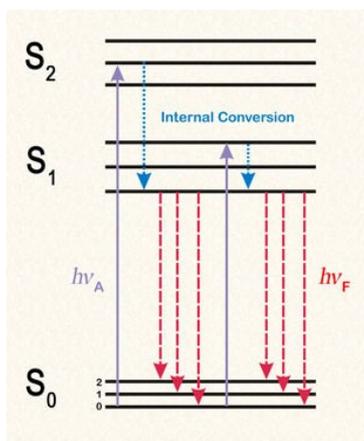
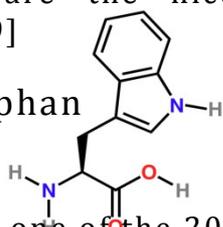


Figure 1: Generalized Jablonski Diagram. Photons are absorbed and re-emitted at different wavelengths [27].

1.2 Compounds responsible for fluorescence

Ultraviolet laser-induced fluorescence is a standard technique used to discriminate between aerosolized particles. Typically, biological materials contain key constituent molecules, which exhibit considerable absorption in UV and characteristic fluorescence emission spectra. Four compounds, enabling discrimination between particles of biological origin and non biological origin are tryptophan, nicotinamide adenine dinucleotide, tyrosine and Phenylalanine. Other sources of fluorescence that absorb at longer wavelengths are the nicotinamide adenine dinucleotide phosphate and riboflavin. [19]

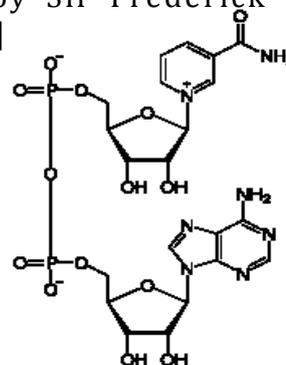
1.2.1 Tryptophan



Tryptophan is one of the 20 standard amino acids found in all forms of life. For many organisms, tryptophan is an essential amino acid, which means that it cannot be synthesized by the organism and therefore must be part of its diet. Humans require about 7 mg of this amino acid per day per kg of body weight. Tryptophan is precursor of neurotransmitter serotonin.[8]

The isolation of tryptophan was first reported by Sir Frederick Hopkins in 1901 through hydrolysis of casein, milk protein.[8]

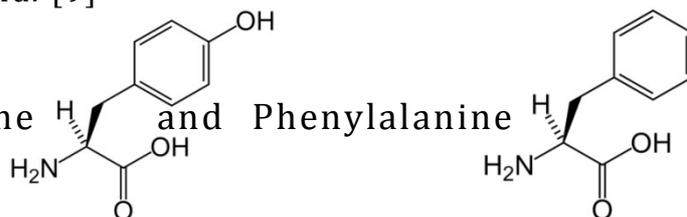
1.2.2 Nicotinamide adenine dinucleotide



Nicotinamide adenine dinucleotide, abbreviated NAD⁺ is a coenzyme found in all living cells. We are interested in reduced form of NAD⁺, which accepts electrons from other molecules and forms NADH. This form can then be used as a reducing agent to donate electrons and represents the main function of

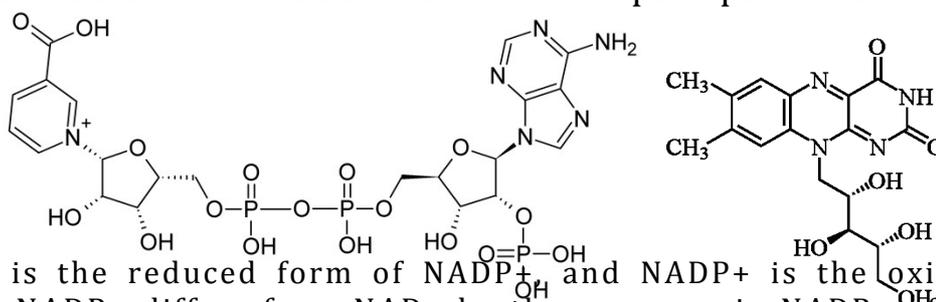
NAD⁺. In organisms, NAD⁺ can be synthesized from the amino acids tryptophan or aspartic acid. [9]

1.2.3 Tyrosine and Phenylalanine



Tyrosine and phenylalanine also belong into group of 20 standard amino acids. Tyrosine is non-essential amino acid and it is found in large quantities in casein. It has a special part by virtue of phenol functionality and plays important role in photosynthesis. Phenylalanine is essential amino acid classified as nonpolar because of hydrophobic nature. Breast milk from mammals is rich in phenylalanine. It is also produced by plants and most microorganisms. L-phenylalanine can also be converted in L-tyrosine.[8]

1.2.4 Nicotinamide adenine dinucleotide phosphate and riboflavin



NADPH is the reduced form of NADP⁺ and NADP⁺ is the oxidized form of NADPH. NADP⁺ differs from NAD⁺ by the presence in NADP⁺ of an additional phosphate group on the 2' position of the ribose ring that carries the adenine moiety.[8]

Nicotinamide adenine dinucleotide phosphate (NADP⁺) is used in anabolic reactions, such as lipid and nucleic acid synthesis, which require NADPH as a reducing agent.[8]

Riboflavin, also known as vitamin B2, is an easily absorbed micronutrient with a key role in maintaining health in humans and animals. As such, vitamin B2 is required for a wide variety of cellular processes. Like the other B vitamins, it plays a key role in energy metabolism, and is required for the metabolism of fats, ketone bodies, carbohydrates, and proteins.[19]

1.3 Contribution of individual residues to biological particle fluorescence

The fluorescence of a folded protein contained or bio-aerosol is a mixture of the fluorescence from individual aromatic residues and coenzyme. Fluorescence is generally excited at 280 nm or at longer wavelengths. Most of the emissions are due to excitation of tryptophan and NADH, with a few emissions due to tyrosine and phenylalanine.[18, 19, 21]

The three residues have distinct absorption and emission wavelengths. They differ greatly in their quantum yields and lifetimes. Due to these differences and to resonance energy transfer from proximal phenylalanine to tyrosine and from tyrosine to tryptophan, the fluorescence spectrum of particle containing the three residues usually resembles that of tryptophan.[18]

Table 1: The table summarizes the fluorescence characteristics of aromatic residues [18, 19].

	Lifetime [ns]	Absorption		Fluorescence	
		Wavelength [nm]	Absorptivity [M-1cm-1]	Wavelength [nm]	Quantum yield
Tryptophan	2.6	280	5,600	348	0.20
NADH	0.4	259/339*	16,900/6,220*	460	/
Tyrosine	3.6	274	1,400	303	0.14
Phenylalanine	6.4	257	200	282	0.04
NAD(H)P	/	355	/	475	/
Riboflavin	/	400	/	525	/

* NADH absorbs also at higher wavelengths

Tryptophan and NADH have much stronger fluorescence and higher quantum yield than the other two aromatic amino acids. The intensity, quantum yield, and wavelength of maximum fluorescence emission of tryptophan is very solvent dependent.[18]

Both NAD⁺ and NADH absorb strongly in the ultraviolet due to the adenine base, where NADH also absorbs at higher wavelengths, with a second peak in UV absorption at 339 nm. NAD⁺ and NADH also differ in their fluorescence, as a matter of fact the oxidized form of the coenzyme NAD⁺ does not fluoresce. [11]

Tyrosine, like tryptophan, has strong absorption bands at 280 nm, and when excited by light at this wavelength, has characteristic emission profile. Tyrosine is a weaker emitter than tryptophan, but it may still contribute significantly to fluorescence because it usually present in larger numbers. The fluorescence from tyrosine can be easily quenched by nearby tryptophan residues because of energy transfer effects. Also, tyrosine can undergo an excited state ionization which may result in the loss of the proton on the aromatic hydroxyl group that leads to quenching of tyrosine fluorescence.[18]

Phenylalanine is weakly fluorescent. Phenylalanine fluorescence is observed only in the absence of tyrosine, tryptophan and NADH. The simple structure of phenylalanine may preeminently demonstrate the effect of structure on fluorescence.[18]

The fact that the reduced form of nicotinamide adenine dinucleotide phosphate, (NADPH), absorbs at a different wavelength from the oxidized form, NADP⁺, provides a potential method for distinguishing between viable and nonviable bacteria. When bacteria die, they convert to the oxidized state. Therefore, the differential between NADPH and NADP⁺ can provide viability information in near real time. Current laboratory efforts are under way to exploit this parameter. If successful, this would significantly enhance the current ability to determine viability at the same time as discriminating biological from nonbiological particles in the environment [19].

Induced fluorescence LIDAR

1.4 Principle of LIDAR Operation

A LIDAR system essentially comprises a transmitter, a receiver, and detection system with controller.

General principle of LIDAR starts with directing laser beam into the atmosphere. Light travelling through the air encounters aerosols of different origin, size and concentration. Regarding wavelength of emitted light aerosol characteristics define the form and intensity of backscattered light. In that sense the origin and size of particles effects the type of scattering process and the wavelength of backscattered light, whereas the concentration of the aerosol effects the intensity of returning light. Telescope orientated in same direction as laser enables us to observe the illuminated region of atmosphere and gathers returned light. Our eye cannot comprehend the information in the backscattered light for which reason light is transformed to electrical signal and analysed by computer.[7]

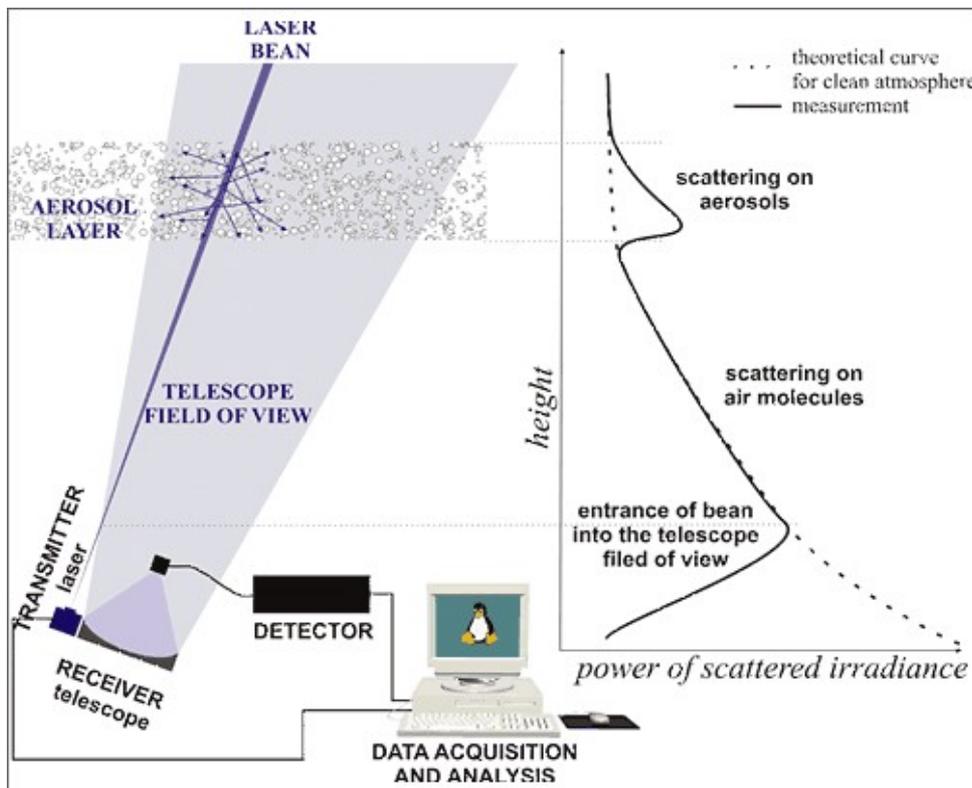


Figure 2: LIDAR operation principle.[27]

1.4.1 Transmitter

Transmitter is light source. Pulsed lasers, with their inherently low divergence, narrow spectral width, and short, intense pulses are ideal as the light sources for LIDAR systems and most common transmitter selection for modern LIDARs. The more powerful pulses are, the more distant particles can be observed. However backscattered light magnitude is due to intensive transmission still good enough to be detected in spite of great remotnes. Besides the laser beam backscattered light

the receiver also captures radiation from other sources such as scattered or direct sunlight, starlight, moonlight, airglow, and scattered light of anthropogenic origin. These unfavourable contributions designated as background represent noise to LIDAR observation. Low divergence of the beam enables the detection system to view as small area of the sky as possible in order to keep the background low. Monochromatic light of sufficiently narrow spectral width provided by laser and scattered by a molecular gas or liquid allows the detection optics of a LIDAR better distinction from those of the incident radiation. Receiver spectrally filters collected light, therefore selectively obtains only certain wavelengths in which manner background can be reduced for several orders.[1, 26]

1.4.2 Receiver

Receiver is to collect and process the scattered laser light. Usually optical telescope mirror represents the receiver. After light is gathered to focus point by the primary optic, light is usually optically analyzed in some way before being directed to the detector system. The simplest form of spectral filtering uses a narrowband interference filter that is tuned to specific wavelength. As mentioned in transmitter description this significantly reduces the background, blocks extraneous signals and actually makes signal detectable. A narrowband interference filter that is typically around 1 nm wide provides sufficient rejection of background light for a LIDAR to operate at nighttime. For daytime use, a much narrower filter is usually employed. Follows processing based on wavelength, polarization, and/or range, depending on the purpose for which the LIDAR has been designed. Appropriately transformed light signals are led by optical cable or directly on to the cathode of an optical detector. [26]

1.4.3 Detecting, recording and LIDAR controlling

The detector converts the light from the receiver system into an electrical signal. Photomultiplier tubes (PMTs) are generally used as detectors for incoherent LIDAR systems that use visible and UV light, whereas photodiodes (APDs) are used to presume IR spectral area. The electrical signal from photodetector is amplified and translated to digital format. Computer makes final data acquisition furthermore produces a permanent record of the measured intensity as a function of altitude. Signal intensity corresponds to concentration of the constituent considering backscattered light energy density diminishes by the square of length measured from the source. Precisely recorded time of flight and return executed by LIDAR controller coordinating transmitter and receiver enables us to estimate the distance of scatterer. [26]

1.4.4 Configuration setting

In general, LIDARs are used for the study of aerosols in the atmosphere in either monostatic or bistatic configuration depending upon the application or experimental requirements. In the monostatic configuration, the transmitter and receiver are arranged either co-axially in which the axis of the laser beam is coincident with that of the receiver optics, or in a bi-axial arrangement in

which the laser beam only enters the field of view of the receiver optics beyond some predetermined range. In the case of bistatic configuration, the transmitter and receiver are spatially separated. Although monostatic LIDARs are suitable for the development of mobile systems for vehicles, aircraft and spacecraft installations besides their ground based applications, bistatic LIDARs, on account of their angular scattering measuring capabilities, possess advantages over monostatic LIDARs.[1]

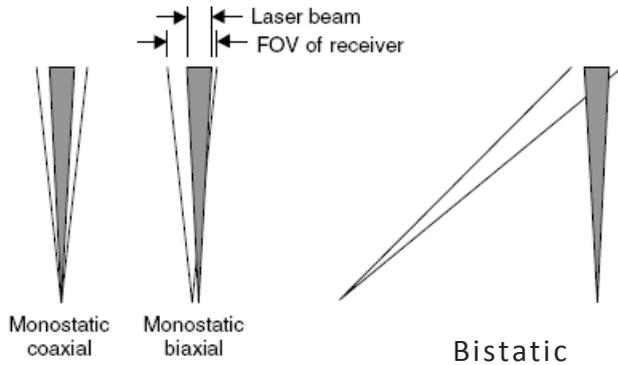


Figure 3: Configuration setting of LIDAR.[1]

1.5 Induced fluorescence LIDAR

Although induced fluorescence LIDAR follows the same general principle of operation as any LIDAR we could say IF LIDAR is completely different device. The subject of observation or better observed process is properly speaking quite different.

Again the operation starts with transmitter sending light pulses into atmosphere. Here also first difference appears. To cause induced fluorescence on target remoted particles laser must provide light in UV spectra between 200 and 400 nm. Light beam travels through atmosphere and comes across aerosols of different origin. When light runs into biological material the most significant process termed fluorescence happens. Light photons cause excitation of molecules contained in bioaerosol, subsequently the result of molecules relaxing from electrically excited state back to its ground state is emission of photons. Emitted light wavelength differs from laser light depending on type of biomaterial.[10] Returning light of consecutive intensities representing concentration of bioaerosol at different layers is captured with telescope and further treated. Expected wavelength of fluorescence is separated from the whole spectrum and transformed in proper way into meaningful information about atmosphere.

It is a fact LIDAR systems can be operated in various ways regarding different technical arrangements. Various LIDAR observations can be perceived exclusively as principle of transmitting and receiving signal ignoring type of physical processes occurring in air. Whether IF LIDAR conquers exceptional position among LIDARs is sense of last statement therefore probably the matter of broadened applicability or just the matter of view. After all not much is required for laser to provide the light in UV specter... still being loyal to physics I continue... yet a lot is acquired when induced fluorescence on the specific targets becomes detectable.

1.6 Usage of induced fluorescence LIDAR

The induced fluorescence signal itself is quite limited for atmospheric research, but combined with other scattering/absorption processes of laser-atmosphere/-hydrosphere/-biosphere interactions is employed successfully for different kind of specific observations.

1.6.1 Remote sensing of organic materials

Investigation on amino acids in the second sections shows the most significant ability of induced fluorescence LIDAR to draw distinction between material of biological origin and nonbiological material. Because of exquisite characteristic the LIDAR has already experienced various applicability. Examples of various scenarios in bioaerosol detecting, pollution tracing, vegetation and marine monitoring as historic monument imaging are presented in this paragraf.

- Detection of biological warfare agents

It is for certain that army does not lead the science, but we should recognize some stimulations. Many a project has been accomplished due to need of army. Military purposes take credit for developing applicability to many scientific ideas and inventions which would otherwise take many years to evolve. Financial encouragement is something the army can provide thus science should make good use of it. And it did. Alternative examples of induced fluorescence LIDAR usage are symbiosis to development in detecting biological warfare agents. Introduction to this article presented the main idea of perceiving biological warfare agents and also explained its restrictions. Making inferences a lot of work awaits to be done in bringing full operating device with bioaerosol type discriminating detector. One possible path has already been traced out by employing induced fluorescence LIDAR as a warning device where additional device is sent in the field to define type of aerosol.[21] However, greater interest preform investigations made on recognising type of bioaerosol by comparing intensity of three induced fluorescence emissions for two different UV exitation wavelengths. The fluorescence signals normalized to the particle elastic scatter intensity for a set of sixteen chambered samples corresponding to different threatening biomaterials were investigated. The summed averaged intensities are shown in *Figure 4* as centroids of ellipses for which the major and minor axes are given by the summed standard deviations for each sample.[26]

The results indicate that bacterial spores, vegetative bacterial cells and proteins can be differentiated based on the two wavelength excitation approach. Anyhow it should be taken into consideration this is an initial study preformed in laboratoric conditions.[26]

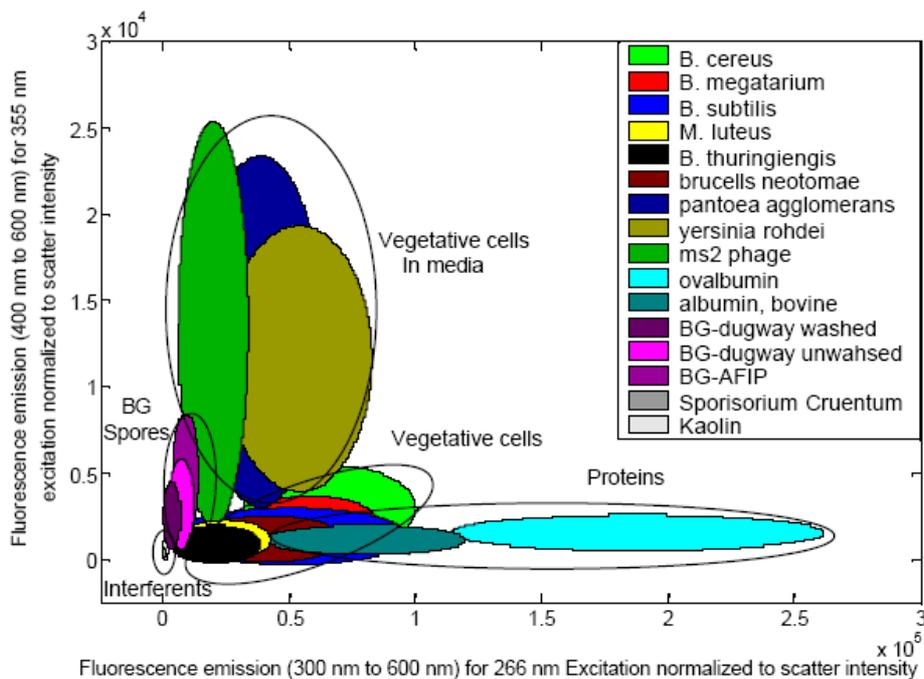


Figure 4: Scatter plot distribution of the 355 nm-excited fluorescence versus the 266 nm-excited fluorescence for the 16 samples. The ovals represent the probability that a particle of the indicated type will fall within that range.[26]

- Biogenic particles in lowermost stratosphere

Bioaerosol is expected to be found in troposphere and we believe there must be infinite palette of various measurements and verifications that can be made. In present case several researchers went further and the lowermost stratospheric aerosol layer have been observed where organic material released by biomass burning from forest fires was detected.[16]

- Remote Sensing of Oil Spills and floating chemicals

Helicopter installation of fluorescence LIDAR was used to monitor water and land areas of oil transportation and storage. The field tests have proved the ability of FLS-A LIDAR to distinguish the fluorescence of minor oil pollution on different spectral backgrounds recorded in airborne measurements. A strong demand can be expected soon for routinely operated, cost-effective remote sensing instrumentation with analytical capabilities that can be placed on small stationary, ground-moving, airborne, aquatic or terrestrial platform [2].

- Fluorescence LIDAR technique for the remote sensing of stone monuments

Fluorescence LIDAR techniques offer considerable potential for remote, non-invasive diagnostics of stone cultural heritage in the outdoor environment. Besides identification of different types of stones and their origin fluorescence hyperspectral imaging has been proven extra useful for the detection of biodeteriogens and their characterisation. The technique can yield data for use in facade status assessment and restoration planning.[20, 22, 21]

- Fluorescent screening of phytoplankton and organic compounds in sea water

With in situ fluorometers, phytoplankton distributions are easily detected measuring the fluorescence emission of chlorophyll. But there is a great variety of physiological processes which influence the chlorophyll fluorescence yield, thus making the quantitative interpretation of these signals very difficult. Various fluorescence methods have been developed to investigate the physiological state of phytoplankton. Findings conclude that cell densities would be much too small for biomass/photosynthetic activity measurements with remote sensing using fluorescence LIDAR. However, predictions are optimistic and these restrictions may be overcome eventually.[26]

- Laser-induced chlorophyll fluorescence spectra, monitoring of vegetation status of plants growing under nickel stress

Laser-induced fluorescence studies of vegetation were used to explore the possibility of using laser as a remote means of measuring vegetation characteristics such as plant vigour, as affected by various stress factors such as drought, natural nutrient deficiency, etc. plant type identification and forest biomass estimation. Certain study demonstrates the use of laser-induced chlorophyll fluorescence spectra in the early detection of heavy metal stress impact on crops, particularly mung. It was found that the fluorescence intensity ratio decreases with increasing chlorophyll content and when the nickel concentration was raised to 1.0 mM, the fluorescence intensity ratio showed increasing trends.[15]

1.6.2 Metal and Nitrogen fluorescence

- Metal fluorescence LIDAR

It has long been a challenge to make in situ measurements of the mesopause region of the Earth's atmosphere (80 to 110 km in altitude), because it is too high for airplanes and balloons, but too low for satellites.

Coincidentally above 80 km the atmosphere is transparent enough for metallic elements to exist in their atomic form. Advantage of the atomic sodium is to fluorescence when illuminated by laser beam. By transmitting laser pulses at another narrow bandwidth temperature and wind can be assessed. These observations are crucial to understanding of solar-terrestrial relations and lay the groundwork for studies of global warming and atmospheric turbulence.[17, 22]

- Auroral ionosphere activity

Fluorescence LIDARs can be used to probe the D region of the ionosphere. Molecular nitrogen fluorescence determine excited or ionized N₂ density in the auroral ionosphere. In addition, the LIDARs provide high spatial resolution diagnostics during ionospheric high power radio frequency heating experiments which are often performed during moderate aurora [13].

IF LIDAR developed at University of Nova Gorica

During three months, from July to September 2007 University of Nova Gorica has been setting up configuration for biaxial, monostatic, mobile induced fluorescence LIDAR. Preparations included several reasoning tasks like calculation about the laser safety and nominal optical hazard zone; the simulation of mobile LIDAR; mechanical ways how to mount the transmitter (laser) and receiver (photometer); optical ideas how to make and assemble the receiver compact and effective; simulation of geometry factor between laser and telescope etc. Simulation of the LIDAR function included calculation of theoretical range.[32]

LIDAR operation principle section intentionally spared the reader from equation representations as some elementary reckoning is made to define detecting range presently. Reach of observation acquisition depends on transmitter efficiency and pulse energy, properties of the atmosphere and its constituents, as optical efficiency of the receiver system.

1.7 Mobile IF LIDAR system description

1.7.1 Transmitter

- Laser

Principal function of transmitter performs pulse laser CFR400 by Quantel Big Sky Laser line of products, which is capable of emitting light of different wavelengths at nominal 1064 nm (IR) as at quadruplicated wavelength 266nm (UV) simultaneously. Ultraviolet light traveling through air diminishes much faster over distance than infrared light therefore ability of laser to emit light also at IR extends the range of LIDAR observation. Reach of detection also depends on pulse intensity. As a matter of fact laser is capable of emitting light also at 532nm, but is not used as it induces some noise on other wavelengths and detection channels. Main laser specifications for both wavelengths are presented in Table 2.[4, 32]

The laser used as the LIDAR transmitter is Class IV laser and belongs to the most dangerous group of lasers. Safety concerns must be taken into account as it may hurt our skin or eyes. Parameters designating laser hazard rate were calculated for laser outgoing wavelengths with different pulse energies, pulse durations, laser beam sizes, divergence angles considering variable eye sensitivity to different light wavelengths. Most important values are nominal ocular hazard distance (NOHD) describing distance at which direct light is still vulnerable to an eye and nominal hazard zone (NHZ) determining radius of threat for our eye to be hurt by diffusive light. Also optical density (OD) for protective eyewear that prevents eye damage by direct laser light was estimated. Beside technical inconvenience safety was also one of the reasons to remove laser light at 532nm since human eye is highly sensitive to it.[4, 32]

Table 2: Laser specifications.[4]

Parameters	Values for UV and IR	
	Wavelength [nm]	226
Pulse energy [mJ]	40	10
Pulse duration [ns]	8	9
Exposure duration [s]	10	10
Pulse repetition frequency [Hz]	10	10
Beam diameter [mm]	7	7
Beam divergence [mrad]	1,2	1,5
MPE[Wcm ⁻²]	3·10 ⁻⁴	2,66·10 ⁻⁵
Optical density of eyewear [OD]	4,443	5,291
Difuse reflection NHZ [m]	0,291	1,55
Intrabeam NOHD [m]	343	1890

- Laser controler

CFR series come equipped with a compact case that includes an air/water cooler and electronics seen in figure 6.

- Additional optics

Transmitter system includes set of optic components that modify emitted laser light in desired manner. Mirrors were assembled to adjust direction of the laser light to receiver field of view in a way that enables fine correction at any time. The major danger at managing with LIDAR represents hazard of laser light entering into eye of eventual passerby. To guarantee appropriate safety exit light energy density was reduced by increasing diameter of the beam leading it through system of lenses. THORLABS company provided mirrors and lenses in transmitter optical system shown in Figure 5.[6]



Figure 5: Pulse laser Quantel CFR400 with additional optic components which are currently dislocated as laser is included in other projects of university.

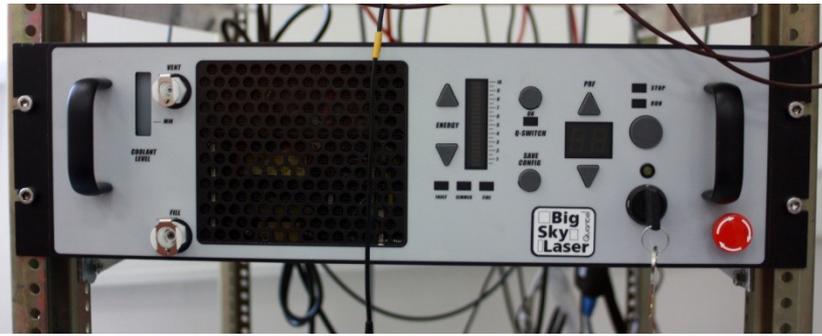


Figure 6: Original laser equipment - case with air/water cooler and electronics.

1.7.2 Receiver

12" Dopsonian telescope by Guan Sheng Optical company serves as a receiver. 302 mm parabolic primary mirror gathers backscattered light and induced fluorescence to its focal length at 1520 mm, where another mirror is placed. Secondary mirror redirects light into detecting system installed outside the telescope. The tube is rolled steel perfect to carry transmitter and detecting system. As telescope was originally produced as astronomic useful finder scope is present and primary mirror is also equipped with fan cooler. Photo exposing interior of telescope, finder scope on upper left and case with detector system above the telescope tube is shown at Figure 7.

1.7.3 Detecting system

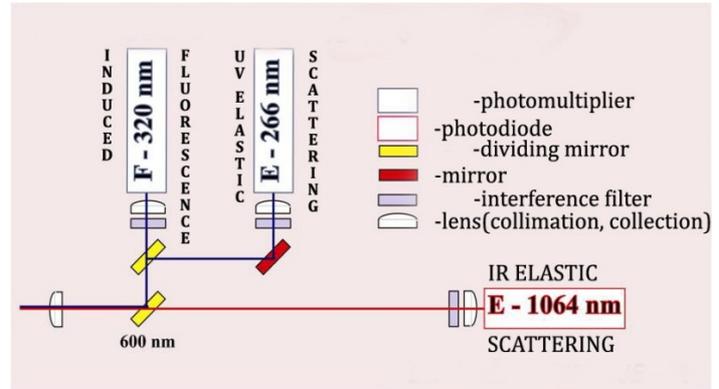
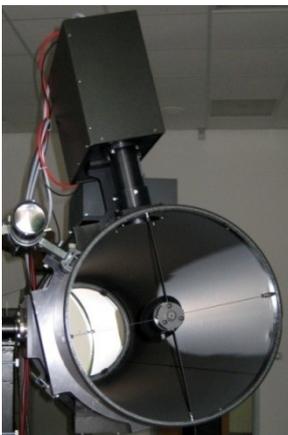


Figure 7: The Guan Sheng Optical 12" Dopsonian telescope with detecting system above. Figure 8: Scheme of detecting system.

- Dichroic mirrors

Dichroic mirrors made by SLS Optics Limited were applied to divide elastic scattering (at 1064nm and 266nm) and induced fluorescence (UV). The first dichroic mirror in the receiver separates UV from IR. Ultraviolet light is divided once more with the second dichroic mirror, where induced fluorescence is separated from elastic scattering. According to theoretical calculation optical properties of dichroic mirrors are presented in figures 9 and 10. At special request mirror characteristics were additionally laboratorically tested where graphs were confirmed.[6]

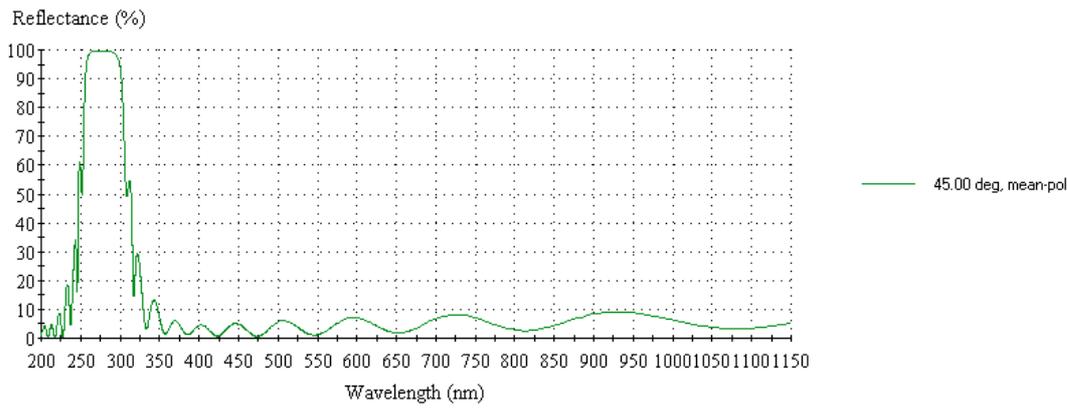


Figure 9: Dichroic mirror reflectance that serves for separation of UV and IR light.[6]

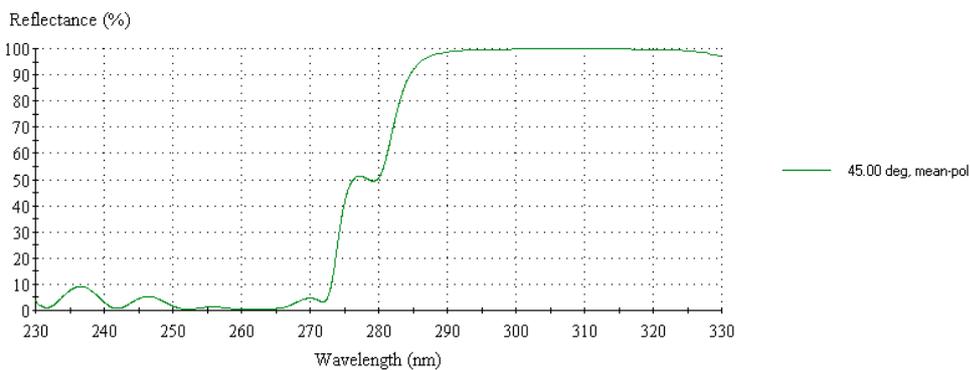


Figure 10: Dichroic mirror reflectance that serves for separation of elastic UV scattering and induced fluorescence. Interference filters.[6]

In order to separate desired signal from background interference filters by BARR Associates were installed. Main properties of interference filters are described by central wavelength (CWL), full width at half maximum (FWHM), maximum permeability, optical density (OD) and size. Values depend on type of scattering we monitor. Table 2 presents characteristics of employed filters for elastic scattering and induced fluorescence.[6]

Table 3: The table summarizes interference filters properties.[6]

Properties	Elastic		Induced Fluorescence	
	IR	UV	UV1	UV2
CWL	1064±0,15 nm	266±0,45 nm	295±0,75 nm	295±1,50 nm
FWHM	1,0±0,20 nm	1,0±0,60 nm	5,0±1,00 nm	10,0±2,00 nm
Maximum permeability	> 70%	> 18%	> 25%	
OD between 200 and 1100 nm	4 (=0.01% light transmission)			
Size	1"			

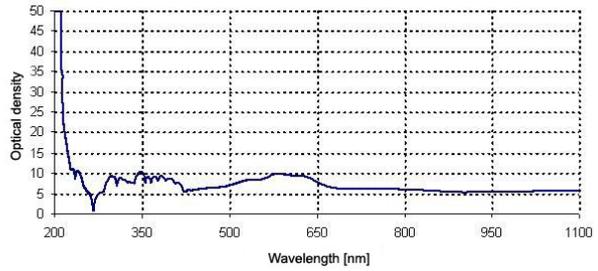
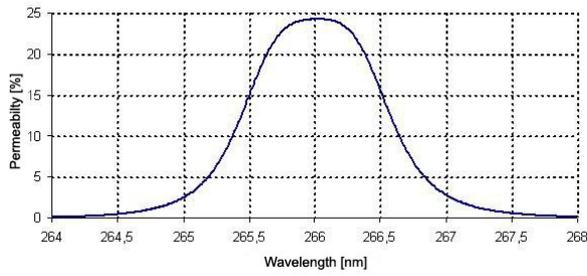


Figure 11: Permeability and optical density dependance on wavelength for interference filter used to select elastic scattered UV light at 266 nm.[6]

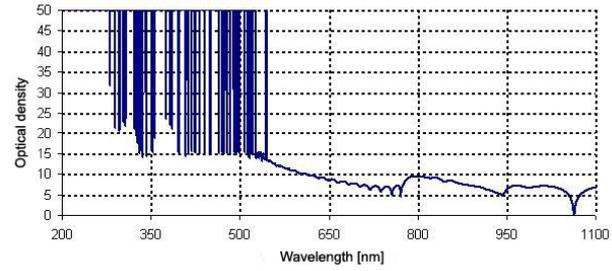
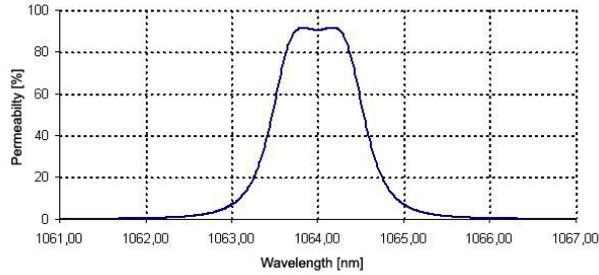


Figure 12: Permeability and optical density dependance on wavelength for interference filter used to select elastic scattered IR light at 1064 nm.[6]

- Photomultipliers

Percipitation of elastic scattering in UV spectrum and fluorescence induced on aerosols of organic origin is held by two photomultipliers Hamamatsu R7400-06. Apperance of the device is shown in Figure 13 and their characteristics in Figure 14.

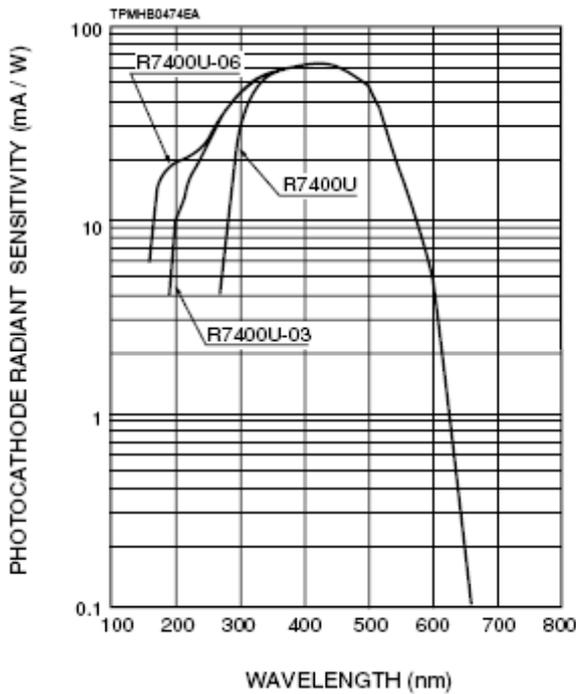


Figure : Characteristic chart of spectral response for photomultiplier Hamamatsu R7400-06.[6]

Figure : Photo of photomultiplier Hamamatsu R7400-06.[6]

Photomultipliers that now represent part of detecting system of induced fluorescence mobile LIDAR were previously proved on existent LIDAR detecting system of Otlica LIDAR observatory, which has been also established as one of projects directed by University of nova Gorica. LIDARs uses lasers of the same producer where one at Otlica uses much powerful type of laser that sends 5ns pulses of 120 mJ with frequency of 20Hz and transmits light of wavelenght at 355 nm. Despite the difference between transmitters the results of experimental measurements taken with new detectors installed on Otlica LIDAR were sufficient to make a good comparison to measurements with standard equipment of Otlica LIDAR. As there had not been noted significant differences in measurements due to usage of different photomultipliers, the selection was confermed. All the measures were taken during clear day and example of experimental measurement with one of multipliers is presented in Figure 15 as S-function.[6]

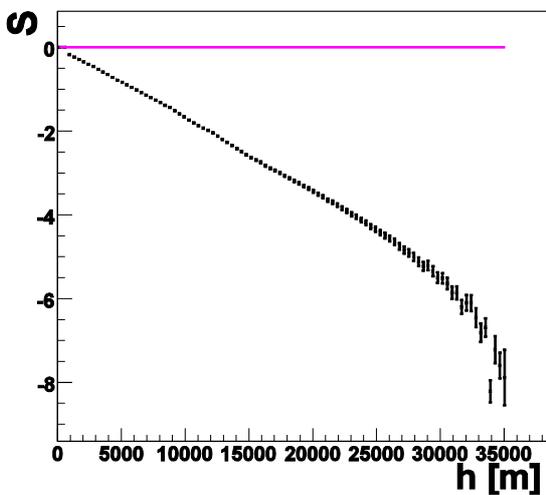


Figure 15: Example of experimental measurement with photomultiplier Hamamatsu R7400-06 perfomed by LIDAR at Otlica observatoy with S-function.[6]

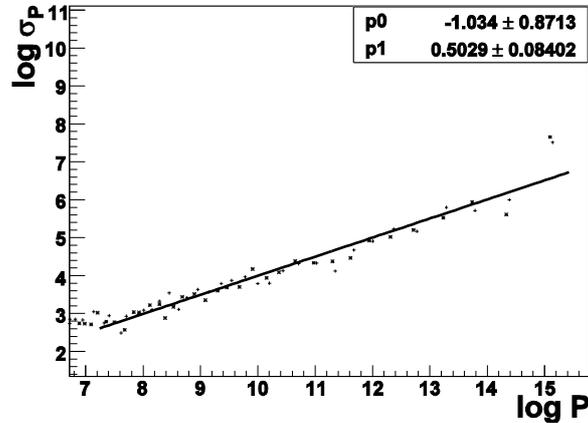


Figure 16: Relation between tested photomultiplier signal error $\sigma_P(r)$ and signal intensity $P(r)$. [6]

S-function takes into consideration that light intensity diminishes by the square of travelled distance. LIDAR equation for elastic scattering is generally written

$$P(r) = P_0 k \frac{c \tau_0}{2} \beta(r) \frac{A}{r^2} T^2, \quad [1]$$

where $P(r)$ represents signal intensity of backscattered light from distance of r at time t , P_0 is emitted light intensity at time t_0 , k designates system efficiency, c light velocity, τ_0 pulse duration, A effective surface of the receiver, r distance between LIDAR and scatterer, $\beta(r)$ backscattering coefficient and $T(r)$ atmosphere permeability for laser light between receiver and scaterer. Permeability depends on diminishing coefficient $\alpha(r)$ found in equation describing optical density $\tau(r)$.

$$T(r) = e^{-\tau(r)} = e^{-\int \alpha(r) dr} \quad [2]$$

Neglecting observations taken below certain distance h , where laser does not enter into receiver field of view yet, S-function can be simplified to dependence on nothing but backscattering coefficient and optical density.

$$S = \ln\left(\frac{P(r)r^2}{P(s)s^2}\right) = \ln\left(\frac{\beta(r)T^2(r)}{\beta(s)T^2(s)}\right) = \ln\left(\frac{\beta(r)}{\beta(s)}\right) - 2(\tau(r) - \tau(s)) \quad [3]$$

Persuming homogeneous atmosphere as regards scattering properties

$$\frac{dS}{dr} = \text{const.} = \frac{d\beta(r)}{\beta(r)} - 2\alpha(r) \quad [4]$$

we get linear form of S-function to distance of 30 km as shown at Figure. Measurement was taken in horizontal direction during clear day. Linear S-function dependance confirms that scattering and absorption atmosphere properties were constant at the time of experimental measurement. Conditions were therefore optimal for detector testings.[6]

As any measurement also photomultiplier measurement has certain error, which has been statistically estimated. Figure 16 confirms photon perception is statistical process corresponding to Poisson distribution. According to average over 20 measurement statistical estimation of error is shown at Figure as vertical line.[6]

- Photodiode

General description of LIDAR in upper section quotes photomultipliers serves as detector of ultraviolet and visible light while infrared light is detected by photodiodes. Si APD S8890-30 by Hamamatsu is our selected photodiode. Device belongs to silicone avalanche photodiode series and is optimized for infrared 1,06 μm YAG detection. Figures 17 and 18 shows photo of photodiode and its characteristics.[6]

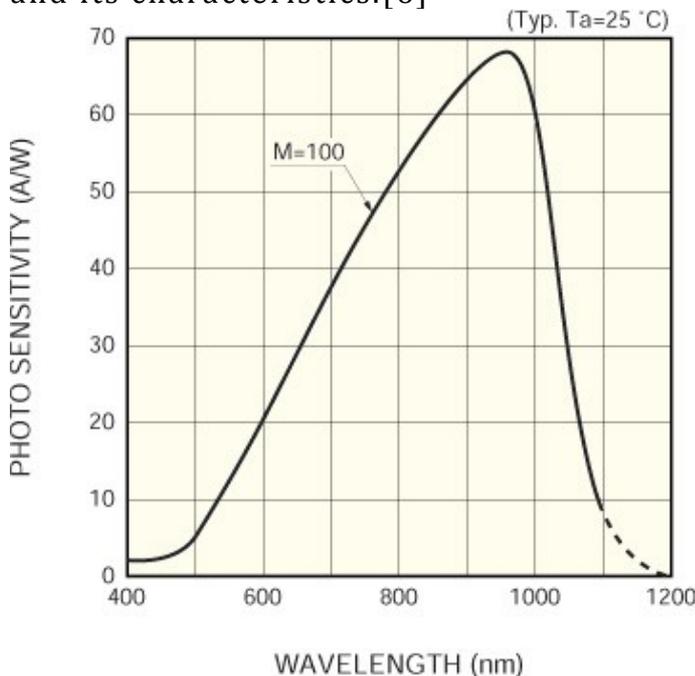


Figure 17: Characteristic chart of spectral response for photodiode Hamamatsu Si APD S8890-30.[6]

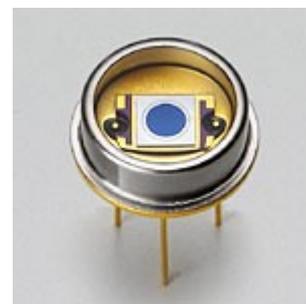


Figure 18: Photo of photodiode Hamamatsu Si APD S8890-30.[6]

- Analog-to-digital converter

Received scattered light and received induced fluorescence travels through described optics to detectors, abovementioned photomultipliers, which are further connected to analog-to-digital converter. In our case device LICEL GmbH TR-40-160 is used, which operates at 12 bits with frequency of 40MHz. LICEL as analog-to-digital converter transforms electrical signal received from photomultiplier into digital information. Selected converter allows for both analog detection, as well as detection by photon counting which makes device particularly suitable for observations of large dynamic range, observations typical for LIDAR. Similar digitalisators are employed in Otlica LIDAR observatory for which reason contemporary version of device used in mobile LIDAR was also previously installed at stationary LIDAR and tested. To perform a trial of new equipment modification of certain programs for data capture had to be done. Full operating LIDAR is sensing three signals therefore needs three digitalisators. As shown in Figure 19 two analog-to-digital converters are combined and installed into one housing becoming two-channell device, while third LICEL is separated in another rack above the first.[6]

- Computer software

Purchase of Licel 40 MHz A/D included computer software package. LabView procedure (Acquis, LiveDisplay, Track) provides primal capture of LIDAR data. Additional procedures at LabView were developed for data analysis and visualisation. Analysis includes basic dependence on signal deminishing by the square of distance. Comparance of signals from different distances enables determination of relative aerosol concentration. Analysis and visualisation is performed individually for every channel and can be shown as a composition of all three sources(UV elastic scattering, IR elastic scattering and UV induced fluorescence). Software is still being developed. Main computer program interface shows Figure 20.[6]

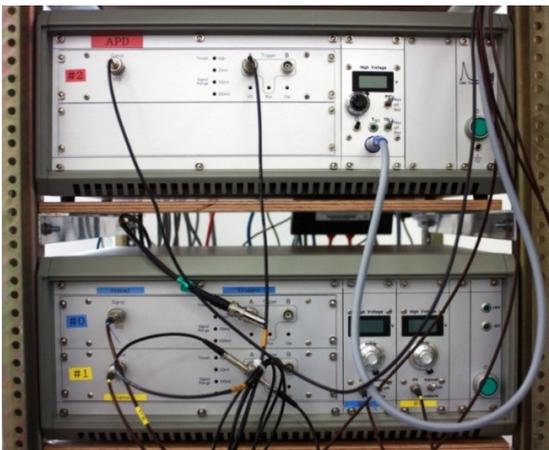


Figure 19: Three LICEL GmbH analog-to-digital converters.

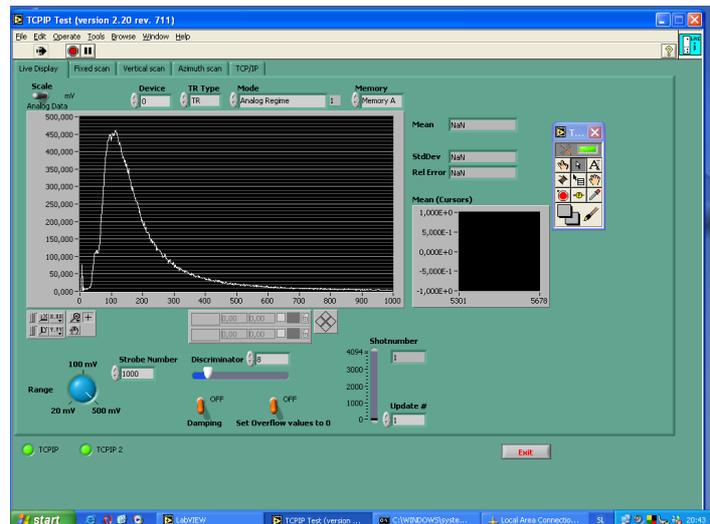


Figure 20: Visualisation of main signal with software tools developed for capturing data, analysis and visualisation of measurement taken by mobile IF LIDAR.

1.7.4 Mechanical parts

Developing sensitive device as LIDAR is important task was to put all the components together in reasonable and accurate way. Appropriate framework supporting telescope was made, adjusting all optics and detector system several holders and housing were manufactured, laser was fixed on the telescope tube and more. Robust, yet still mobile framework was equipped with servomotors which enable managing LIDAR orientation by computer. Successive observations taken during systematical motion of LIDAR can produce three dimensional scanning of atmosphere. Appearance of LIDAR is shown in Figure 21.[6]

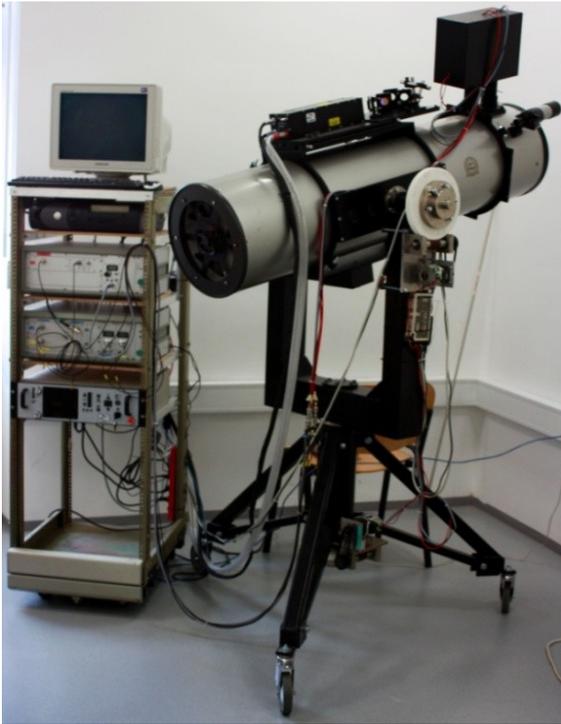


Figure 21: Mobile IF LIDAR developed at University of Nova Gorica.

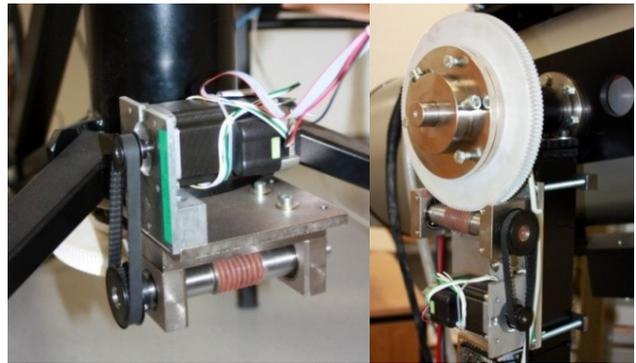


Figure 22: Servomotors serve to orientate LIDAR.

1.8 Detector system optical efficiency

We need to consider backscattered light travels through numerous optical components before attains the detector. In the last section simulation of mobile IF LIDAR is done. To estimate detecting range estimation system optical efficiency must be defined. According to *Figure 23* optical efficiency for different wavelengths are calculated.[4, 7, 32]

IR light at 1064 nm: $PMR \times SMR \times PCX1 \times BS1 \times IF1 \times DCX = APD$

$$0.80 \times 0.80 \times 0.92 \times 0.90 \times 0.70 \times 0.94 = \underline{34.9\%}$$

UV light at 266 nm: $PMR \times SMR \times PCX1 \times BS1 \times BS2 \times IF3 \times PCX3 = PMT1$

$$0.50 \times 0.50 \times 0.92 \times 0.98 \times 0.98 \times 0.18 \times 0.98 = \underline{3.9\%}$$

IF light at 295 nm: $PMR \times SMR \times PCX1 \times BS1 \times BS2 \times IF2 \times PCX2 = PMT2$

$$0.50 \times 0.50 \times 0.92 \times 0.98 \times 0.98 \times 0.25 \times 0.98 = \underline{5.4\%}$$

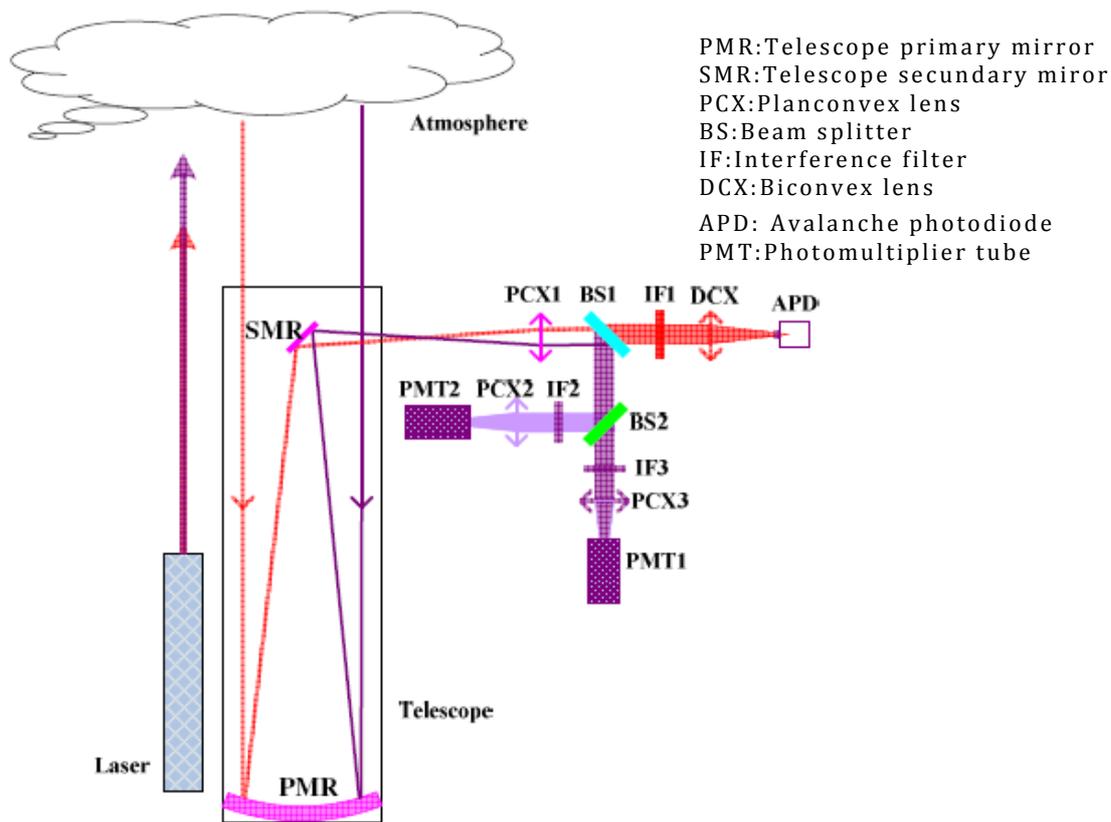


Figure 23: Illustration of optical components that contribute to optical efficiency of the system.[7]

Clearly, signal entering detector is significantly weakened compared to backscattered light signal captured by primary mirror. Imposing role plays also wavelength of light treated by lens system as signal reduction of IR light is minor than UV light. Low optical efficiency was expected as early as project started therefore calculations were made previously in order to select proper components.

1.9 Detecting range estimation for clear atmosphere

Besides transmitter intensity and receiver optical efficiency atmosphere permeability for transmitted, backscattered or fluoresced light and background contribution effects LIDAR detecting range as well. All influential factors are associated in general LIDAR equation:

$$P_s(\lambda, R) \cdot R^2 = P_L(\lambda_L) \cdot [\beta(\lambda, \lambda_L, \theta, R) \Delta R] \cdot A \cdot [T(\lambda_L, R)T(\lambda, R)][\eta(\lambda, \lambda_L)G(R)] + P_B \cdot R^2 \quad [5]$$

P_s - Received signal

P_L - Laser intensity

β - Volume backscatter coefficient

ΔR - Pulse length

A - Area of receiver

T - Atmosphere permeability

η - System optical efficiency

G - Geometrical factor

P_B - Background contribution

Ensuring backscattered signal surpasses detector minimum intensity threshold successful detection is achieved when received signal $P_s(\lambda,R)$ exceeds the background P_b and signal to noise ratio(SNR) is greater than 1 respectively. All equation factors taken into account SNR for individual wavelengths in clear atmosphere is estimated and presented in *Figure 24*. [4]

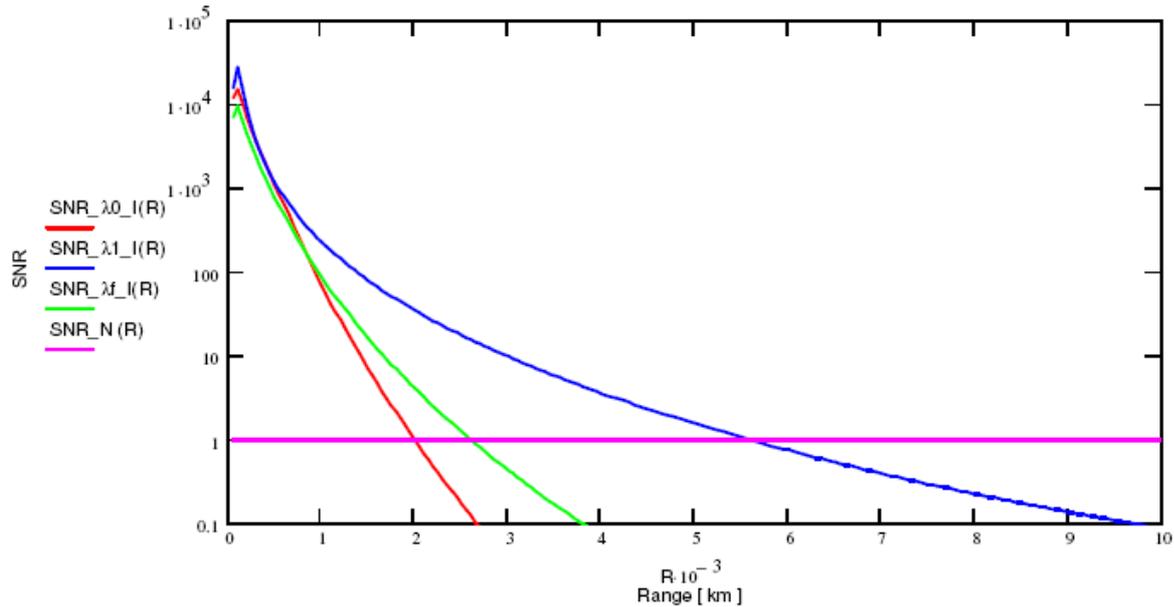


Figure 24: SNR for IR elastic scattered light ($\lambda_1=1064\text{nm}$), UV elastic scattered light ($\lambda_0=266\text{nm}$), fluoresced light ($\lambda_f=290\text{-}300\text{nm}$) for clear, aerosol free atmosphere [4].

LIDAR expected detecting range for UV components designed to distinguish biological and nonbiological aerosols is 2 to 3 km accordingly. Estimated range for IR LIDAR components appointed to observe dynamics of aerosol spreading in atmosphere is 5 to 6 km. Real measurements with aerosol presented scattering is more intensive thus detecting could reach a bit further. [4]

4 Conclusions

Theoretical point of view of operating induced fluorescence LIDAR was presented in seminar. Overview its practical usage and real project example has been made as well. Hopefully gathered information about comprehensive applicability of IF LIDAR inspires anyone to perform further investigations into development of existing projects or to contrive new ways of its employability.

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