STANDOFF DETECTION OF CHEMICAL AND BIOLOGICAL SUBSTANCES USING LASER INDUCED FLUORESCENCE TECHNIQUE

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Abstract

Laser based standoff detection methods are convenient to investigate unknown/hazardous chemical or biological aerosol clouds at a safe distance in order to trigger further counteractions if necessary. Classification and localization of such clouds can be provided by laser induced fluorescence (LIF).

At Lampoldshausen the German Aerospace Center (DLR) operates a 135 m long laser test range, where LIF measurements on aerosols, liquids, and solids can be performed. Fluorescence light of test material, which is illuminated by a laser source with UV wavelengths, is collected by a telescope and analyzed spectrally as well as time resolved by means of a gated spectrograph.

LIF measurements on liquids and aerosols at a distance of 22 m show adequate fluorescence signal strength with a good signal to noise ratio. The data analysis has the ability to distinguish between biological and chemical material with a good prediction confidence level.

Keywords: Aerosol detection; Biological sensing and sensors; Fluorescence, laser-induced; Spectroscopy, ultraviolet; Standoff detection; Hazardous material detection

1 INTRODUCTION

Several attacks in public of varying dimensions, like the disposal of the neurotoxin sarin in a subway in Tokyo 1995 or the transmission of anthrax to american government officials in 2001, show the importance of the ability to early detect chemical, biological, and explosive (CBE) hazardous materials. But also unintentional release of such material, e.g. industrial accidents caused by earthquakes or floods, may occur. The final goal is to identify the substance(s), detect the position of its source, and survey the cloud dimensions and movements at an early stage to trigger the right counteractions and minimize the risk to the public and emergency services. Beside the huge variety of CBE substances, their different physical states and ways of dispersion and contamination further complicate the detection. Biological agents additionally bear the risk of self-replication, which can lead to an exponential growth of the material. This demands high detection sensitivity for low amounts of biological aerosol particles.

A fast and reliable detection of hazardous CBE material on a secure distance to the risk area is provided by laser based standoff detection methods. Laser induced fluorescence (LIF) is one of these methods and has the ability to map and classify aerosol clouds. Even if LIF cannot always be used to identify the material, the information of the location and the propagation of the aerosol cloud can be taken for optimized positioning of point sensors, which may identify the suspected substance.

LIF is a well understood technique. The substance itself is excited by a laser light source and starts to emit radiation with a characteristic spectral and temporal behavior, which allows a classification of the excited molecules. Because typical fluorescence spectra have a broad spectral range, which may lead to ambiguous results, increasing the dimensionality of the measurement data, for instance by using more than one excitation wavelength and/or performing time resolved measurements, is applied ([1], [2], [3]).

In order to apply LIF under real outdoor conditions, several additional effects need to be taken into account. These include e.g. interfering fluorescence spectra from natural surroundings like pollen, dust, and diesel, which have to be considered in the evaluation process. Furthermore, different weather conditions like rain, fog, and background light from the sun also affect the propagation of the laser light. To be flexible in choosing the operation area, the system has to be robust and portable. For operation within public areas, the excitation wavelengths have to be limited to eye-safe ranges, i.e. for fluorescence applications ideally below 400 nm ([4], [5]).

To satisfy these real outdoor conditions, all measurements were performed on a free space optical test range. For a higher dimensionality of the measurement data two different UV excitation wavelengths at 280 nm and 355 nm where used, while time dependent fluorescence spectra of different chemical and biological substances (in fluid and aerosol form) were captured by a gated intensified CCD (iCCD) camera. The time resolved measurements provide a measure for the corresponding fluorescence lifetimes. After background correction, the resulting data is analyzed by pattern recognition software, which leads to a first classification of the various compounds. Following test measurements validate the prior results.

A new feature compared to prior projects is the time decay of the measurements, which leads to an additional dimension in the dataset. Also the combination of the mentioned excitation wavelengths is a unique feature of the system.

2 EXPERIMENTAL SETUP

2.1 Optical setup

Fig. 1 shows a schematic draft of the optical setup. In this system two alternating excitation wavelengths, 280 nm and 355 nm, with a pulse width of 7 ns and a pulse energy of about 10 mJ, are used. The pulses originate from a neodymium-doped yttrium aluminium garnet (Nd:YAG) laser with several frequency conversion units with a repetition rate of 5 Hz for each wavelength. The fluorescence light is collected by a Newton telescope with an optical diameter of 400 mm. A spectrograph with a grating resolution below 1 nm analyzes the spectra in a wavelength range from 300 nm to 600 nm. Filters in front of the spectrograph block the laser light. Furthermore a gated iCCD camera records time resolved spectra by shifting the gate delay at successive laser pulses. This allows the system to get a measure for fluorescence lifetimes and provides an additional data dimension for the discrimination of the substances.

Realistic outdoor conditions can be tested on the free space transmission test range at a distance between 20 m and 135 m located at the German Aerospace Center (DLR) at Lampoldshausen. For the reported measurements, the distance between the laser/telescope composition and the samples is 22 m. Fig. 2 and Fig. 3 show pictures of the optical setup and the transmission range, respectively.

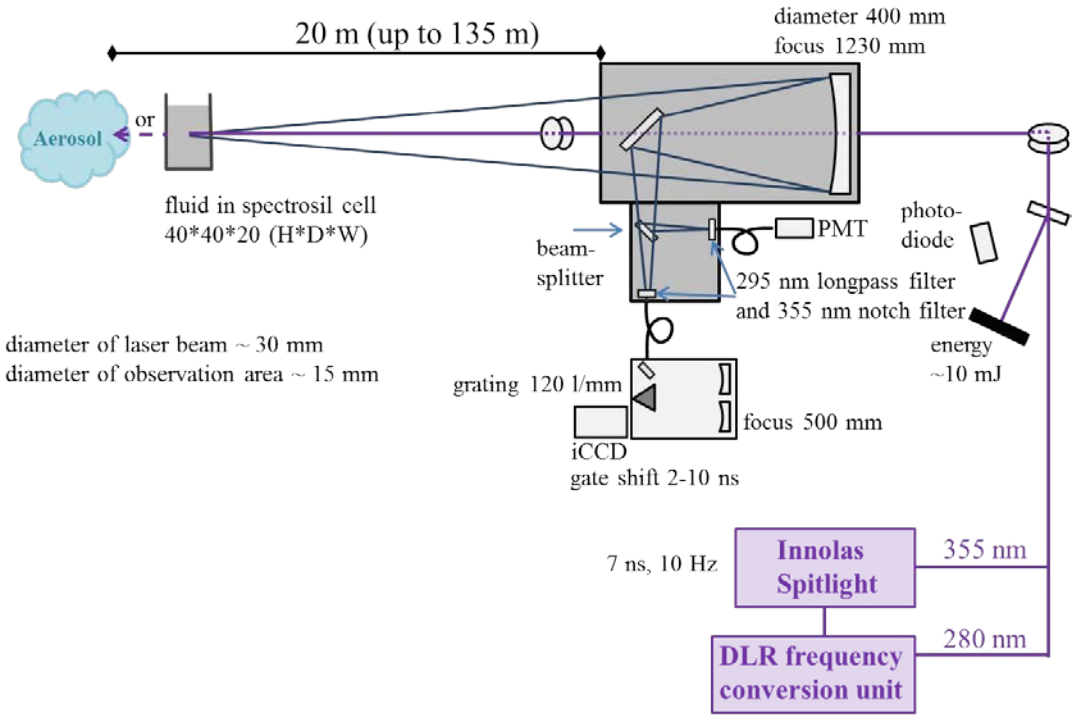

Fig. 1. Schematic draft of the optical setup.

Fig. 2. Picture of the optical setup.

Fig. 3. Picture of the free transmission range at night.

2.2 Data acquisition

The data acquisition system is controlled by a computer program developed using NI LabVIEW® [6], which also manages the data pre-processing. The 10 Hz laser unit runs independently from the data acquisition system and acts as a master trigger. It emits alternatively 280 nm and 355 nm laser pulses. The 200 ns long iCCD camera gate is triggered after every laser pulse with a successive increasing delay, which has an additional offset depending on the sample distance. Furthermore, the camera is triggered between the laser pulses to capture individual background corrections for each spectrum. Fluctuations in the spectra, e.g. caused by laser energy jitter, can be corrected with a wavelength independent fluorescence pulse integration by a photomultiplier tube (PMT). The readout of the integrator is managed by a microcontroller (µC). The microcontroller also gathers the current excitation wavelength status. After data pre-processing, the data is sent to a remote system for analysis and

classification of the measured samples developed by LDI Innovation. Fig. 4 shows a schematic draft of the data acquisition electronics.

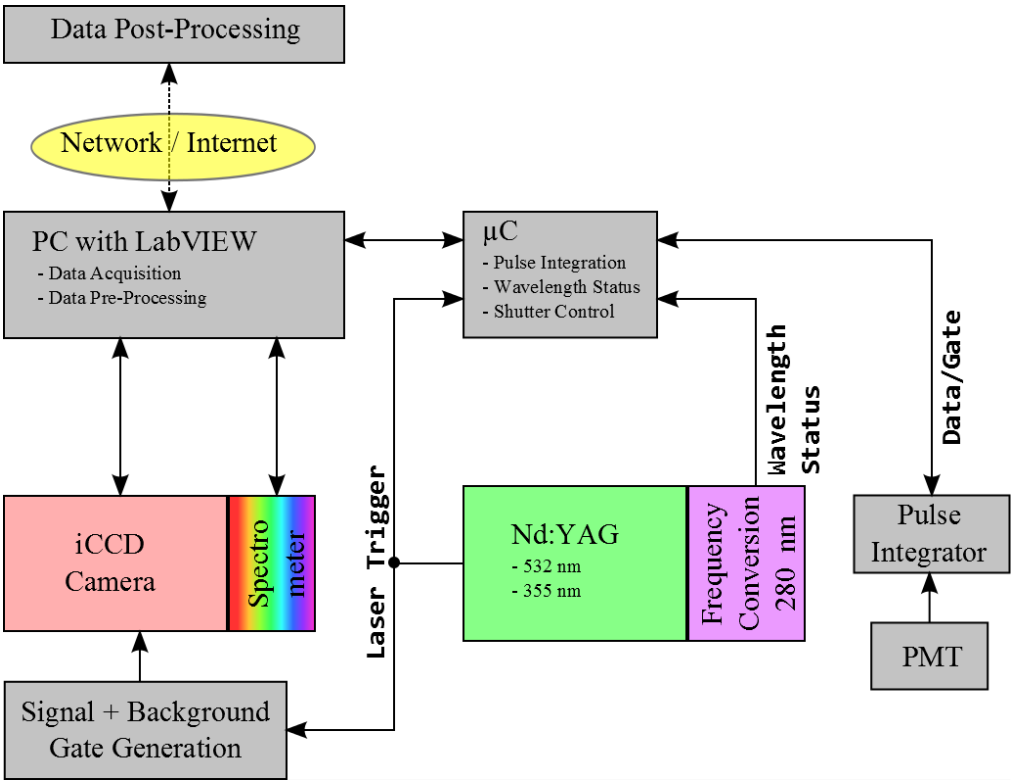

Fig. 4. Schematic draft of the data acquisition electronics.

3 RESULTS

In order to test the capabilities of the LIF system several different biological substances like fungi, bacteria, vitamins, enzymes and aromatic amino acids were measured in liquid and/or aerosolized form. As solvent deionized water was used. To simulate natural background material, hydrocarbons and plants like saffron and dandelion were also measured. Because of the dependence of the substances on the solvent, three different concentrations in the range from 3.75 µg to 3.0 mg per ml were tested. Some substances are not completely soluble in water, which requires to stir them during the measurement to keep the solution in a homogeneous state. To reduce statistical effects, up to 100 single spectra were summed up. To prove the reproducibility and to gather information about a potential alteration of the substance during the illumination, multiple measurements per sample were performed. In addition before and after every measurement absorption spectra of the substances were measured. Each of the following fluorescence spectra is corrected by the spectral profile of all optical parts included in the system. Additionally dark background spectra have been subtracted from every spectrum which provides a zero point correction.

Fig. 5 and Fig. 6 show examples of measurements in two different conditions (liquid and aerosolized). Fig. 5a and Fig. 5b show spectra of dissolved yeast in deionized water with a concentration of 1 mg/ml, illuminated by excitation wavelengths of 280 nm and 355 nm, respectively. The different colors represent spectra with different iCCD camera gate delays. A comparison of both plots indicates the difference between the emitted spectra with different excitation laser wavelengths (280 nm and 355 nm). Fig. 6a and Fig. 6c show spectra of the amino acid tryptophan, suspended in deionized water. Its spectra and fluorescence lifetimes differ from the features of yeast, which demonstrates the possibility of discrimination. As an example for another way of substance output Fig. 6b and Fig. 6d show spectra of aerosolized tryptophan. The

aerosol spectra were measured with constant camera gate and show accordance with the spectra of the suspension.

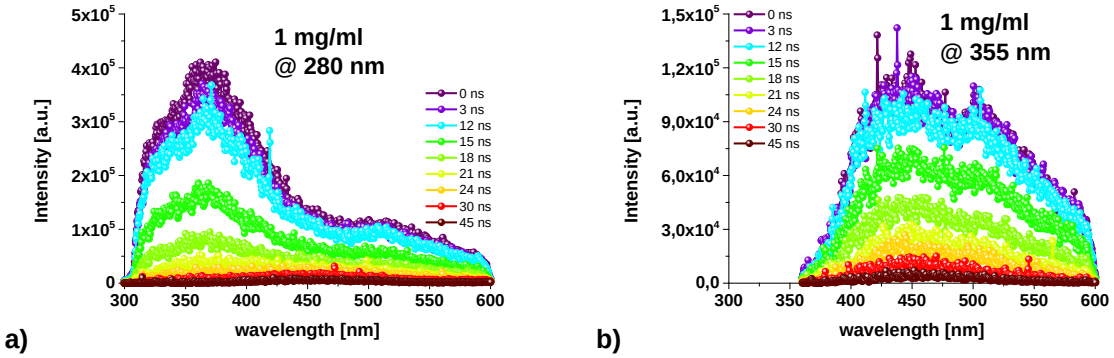

Fig. 5. LIF spectra of yeast, dissolved in deionized water with a concentration of 1 mg/ml. a) Excitation wavelength @ 280 nm, sum of 100 single spectra. b) Excitation wavelength @ 355 nm, sum of 10 single spectra. In both cases camera gate delays were shifted from 0 to 45 ns with 3 ns steps (without offset).

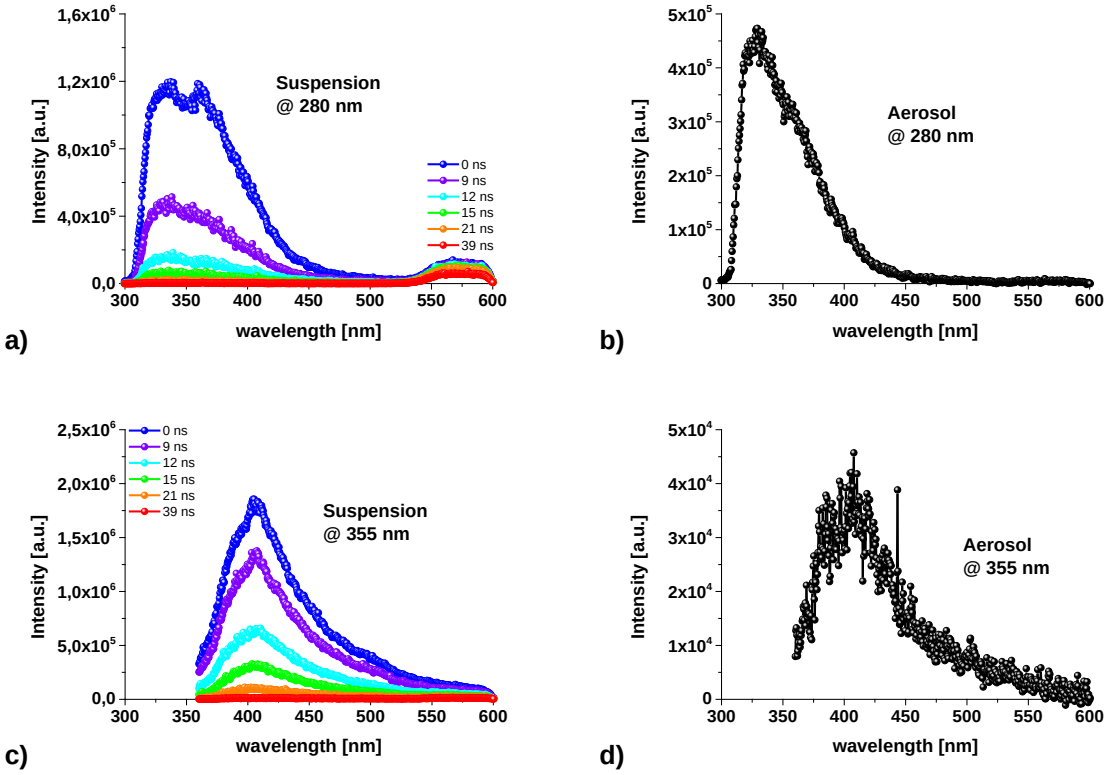

Fig. 6. LIF spectra of the amino acid tryptophan, upper part @ 280 nm and lower part @ 355 nm excitation wavelength. a, c) Suspension in deionized water with camera gate delays from 0 to 39 ns and 3 ns steps (without offset), sum of 10 single spectra. b, d) Aerosol, sum of 100 single spectra.

The additional measurement of fluorescence lifetimes can improve the discriminability of the substances. But this method is limited by the laser pulse width (7 ns), which should be smaller than the fluorescence lifetime. In Fig. 7 the fluorescence lifetimes of two different substances (diesel and dandelion) are shown. The significant difference

between the fluorescence lifetimes of both substances increases their discriminability additionally.

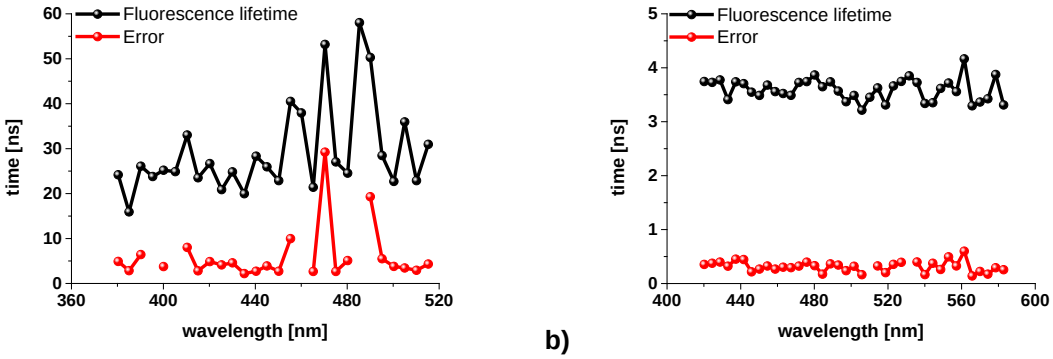

Fig. 7. Fluorescence lifetimes (black) with the corresponding errors (red) of a) diesel and b) dandelion. Both measurements were performed with an excitation wavelength of 355 nm.

4 SPECTRAL CLASSIFICATION

The main objective of the spectral analysis is to develop an algorithmic system, which has the ability to classify measured substances into disjoint classes. In this case four different classes were chosen: plant material, poly-aromatic hydrocarbons (PAHs), live bacteria, and chemical aerosols. The first two classes represent natural background agents, whereas the last two classes include the potential hazardous material. There are different methods of classification, e.g. the Principal Component Analysis (PCA) which is implemented for instance by the Swedish Defence Research Agency (FOI) for a project, described in [1]. In the current work the spectral data is analyzed using a structural extraction combined with statistical classification.

In the first step several parasitic features like artificial spikes and Raman scattering lines are filtered out of all spectra. Because of the high dimensionality (high amount of spectral data points or features, respectively), which is not reasonable for machine learning techniques, it is needed to break down the spectral data to a few significant features. Statistical and structural methods, which are explained in [7], [8], and [9], reduce the number of features by a factor of approximately 60 without losing information about the spectra. The remaining features can be seen as data points of the spectra, which represent specific spectral regions and can be used to reconstruct the original spectra with good accordance. An excellent performance of such a hyperspectral feature extraction was already shown in [10].

With these remaining features a robust classifier can be created by applying the method called “bootstrap aggregation” (bagging, [11]), which delivers additional features for the classifier training set, when the experimental dataset is limited in size. The goal is to generate a robust classifier out of many “weak”/”okay” classifiers. With only one initial dataset (extracted features of e.g. three measurements) bagging allows to reproduce many replicas of this original dataset. Each replica is created randomly by selecting N samples with replacement from the original data, where N is the data size. This method produces new datasets which represent additional imaginary measurements and can be used as training datasets for the weak classifiers. The method grows a decision/classification tree model on each dataset, which slightly differs from the others. The decisions (nodes of the tree) are based on the features of a dataset and have binary answers. Each leaf of the decision tree represents a class label. After the classifier training, the answer for this ensemble of datasets is created by voting among all decision trees. Bagging improves the classification by combining the

answers of weak classifiers ([12]). For the implementation of the bagging algorithm, the "Framework for Ensemble Learning" from the "Matlab Statistics Toolbox" ([13]) is used.

The plot in Fig. 8 represents the clusters of the data in scaled coordinates, which are calculated using the Matlab method "Proximities Matrix Multidimensional Scaling" ([13]) from the decision tree ensemble. The plot shows the high discrimination capabilities of the algorithm. The prediction confidence level lies between 92 % and 98 %.

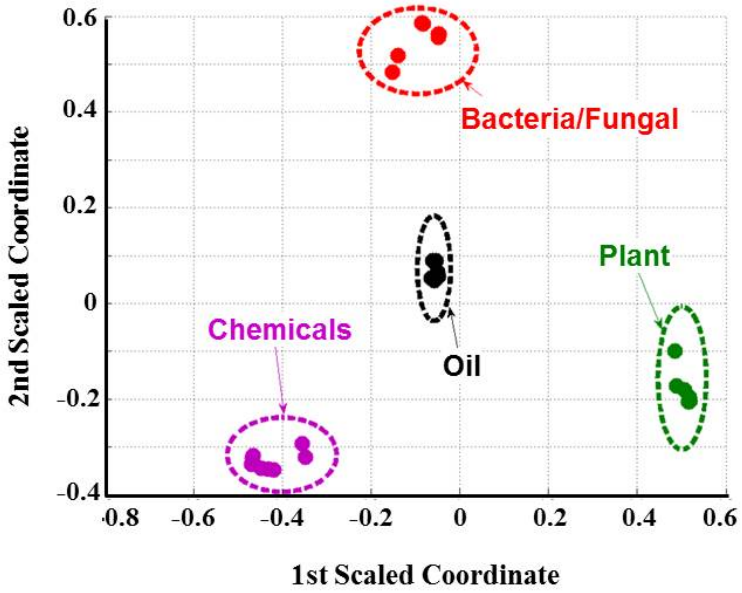

Fig. 8. Plot of the measured samples. The axes represent two eigenvectors of the proximity matrix (scaled coordinates). The clusters are divided into four classes: chemical, biological, oil, and plant material.

5 CONCLUSION

To investigate the time dependent fluorescence properties of a wide variety of chemical and biological material by using the LIF technique under realistic outdoor conditions, the substances were measured on a free space transmission test range in liquid and aerosolized form. Time resolved measurements increase the data dimensionality, compared to prior projects. For the excitation process two different laser wavelengths at 280 nm and 355 nm were used, which is a unique feature of the system. Afterwards, the spectral data is pre-processed and analyzed by pattern recognition software. The data analysis is based on a feature extraction from the spectra, followed by a bootstrap aggregation method to add new imaginary measurement data to the experimental data for better statistics. With these dataset replicas weak classifiers are trained and combined to a robust classifier, which shows a good prediction confidence level. Thus, LIF is a promising standoff measurement method for first classification of hazardous substances and provides information for subsequent identification techniques and counteractions.

As an outlook we plan to enlarge the list of substances to consolidate the prediction power of the pattern recognition system. More substances also bring the advantage to expand the number of classes and define sub-classes, in order to differentiate between various living organisms.

6 ACKNOWLEDGEMENTS

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