Construction of a Biofluorescence Optical Particle Counter

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1 Introduction

O’Dowd et al. [1] found that submicrometer marine spray aerosol contained a significant fraction of organic matter is associated with the seasonality of plankton biological activity as determined from satellite ocean-colour products. When plankton activity is high, the chemical composition of the aerosol is dominated by organic matter that possesses characteristics similar to organic matter found at the ocean surface. Laser induced fluorescence will probe the organic content of marine aerosols through excitation of fluorescence of chlorophyll-a contained in the phytoplankton. In-vivo chlorophyll-a has a main broad absorbance peak at ~440 nm and two main fluorescence peaks at ~670 nm and ~720 nm. The aerosol is probed with 405 nm 50 mW laser that has a wavelength to allow for maximum absorption and induce a maximum yield of fluorescence. The instrument will stimulate and collect a total fluorescence signal from each probed aerosol particle in order to determine the ratio of elastic scattered light at laser wavelength to fluoresced light. Simultaneously it will allow particle sizing through collection of the elastically scattered laser light, and a flux measurement by counting the particles that pass. The laser choice should, theoretically enable scattering measurements to infer diameters down to 50 nm (but instrumental limitations may prevent this), while still stimulating chlorophyll-a at high efficiency. The energy output of 50 mW is sufficiently high to reliably induce the fluorescence, based on existing experimental data available in the literature, e.g. [2-4]. The fluorescence quantum yield is very low for chlorophyll-a in phytoplankton, ranging from 2% to 7% [3] or 0.6 - 3% [4]. The fluorescence yield depends on, for example, type of phytoplankton, light exposure history, other absorbing pigments in the phytoplankton, competing energy processes, available nutrients and surrounding medium [5, 6]. The fluorescence signature also has a very short lifetime on the order of nanoseconds making the fluorescence output almost simultaneous to the elastic scattered light signal. This necessitates the use of highly sensitive photomultiplier tubes (PMTs) to maximise data signal-to-noise.

Figure 1: A schematic layout of the instrument.

Figure 1 depicts the layout of the laser induced fluorescence optical particle counter (LIF-OPC) instrument. A diaphragm pump draws air through the exhaust tube at a rate of ~3.9 L/min, and supplies the filtered sheath air at ~2.4 L/min. The filtered bleed is independently managed with a valve and its flow rate (~0.8 L/min) is balanced against the sheath and aerosol flow rates to produce a smooth non-turbulent aerosol flow (~0.7 L/min). The filtered bleed flow helps maintain a laminar aerosol flow. The ballast chamber helps smooth the flow produced by the diaphragm pump. The ellipsoid reflector has a protected silver coating to maximize collection of the scattered 405 nm laser light as well as having high collection efficiency for the chlorophyll-a fluorescence. The laser, focused by a cylindrical lens to a sharp vertical line ~0.5 mm by 2 mm is directed by fixed mirrors to the primary focus of the reflector, where it perpendicularly intersects the aerosol flow, and further to a laser dump. High f-
number aspheric lenses are used to collect and collimate the scattered light from the primary and secondary foci, for forward and back scattered measurements respectively, as well as any fluorescence. High efficiency dichroic (long pass) mirrors split the collimated beam at an edge wavelength of 500 nm, passing the scattered light to photodiodes, and the weaker fluorescent emission to highly sensitive Hamamatsu PMTs.

2 Initial Results and Discussion

Calibration and testing of the instrument is currently underway and is producing both sizing and fluorescence measurements. The calibration includes using an aerosol generating system (TSI atomizer and classifier) to supply (ammonium sulphate or sodium chloride) particles of known diameter (from 20 nm to 300 nm) to the LIF-OPC to calibrate the photodiode/PMT scattered light signals. The scattered light from particles increases exponentially with particle size. Because of this the introduction of a logarithmic amplifier has allowed a higher resolution at the submicron region. Microspheres (plain and with fluorescent dye) can also be aerosolized with the same atomizer to calibrate the instrument at sub- and super-micron diameters. The LIF-OPC has sized particles (ammonium sulphate) as low as 167 nm in diameter and also detected fluorescence from (Green 510 PSL spheres from Duke Scientific) particles of 3 µm in diameter.

Chlorophyll-a powder will be used with the aerosol generator to produce chlorophyll particles of known diameter to calibrate the PMT fluorescence signal output. A phytoplankton breeding tank will be created to allow in-house measurements of bubble mediated aerosol from seawater enriched with phytoplankton cultures. After validation the instrument will be brought for flux measurements and chlorophyll detection trials on the Atlantic coast at the Mace Head atmospheric research station.

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