Optical Instrumentation - Transmissometer, OBS, LISST, AC-9, Radiometer, Spectral Radiometer, In-Situ Flow Cytometer

The ocean is moderately transparent to light in the visible portion of the spectrum, more so in the blue than in the red. Pure water and oligotrophic seawater reach an attenuation length of 50 meters in the blue. Further in the ultraviolet, absorption increases as the tail of electronic transitions in the far UV become significant while in the infrared, vibrational and rotational absorption bands increase absorption. Within the transmission window in the visible, spectral absorption by dissolved organic compounds, gelbstoff, may make the water yellow or green, and scattering by particles may decrease transmission roughly proportional to the projected area of the particles. These effects on light transmission can be used to study what is in the water besides inorganic salts.

Scattering of light by small particles is Rayleigh scattering when the particles are smaller than the wavelength of the radiation, geometric scattering when the particles are much larger than the wavelength, and Mie scattering when the size is comparable to the wavelength. The range in sizes in the ocean varies from sub-micron for fine clay and certain plankton and bacteria to centimeter aggregates of marine snow. Mie scattering is relevant to part of this range (red light emitting diode (LED) light is typically 0.6 μ) and in Mie scattering, the optical index of the particle also must be considered. Detailed studies of pure populations of particles might confirm this description of scattering but natural seawater contains particles of many sizes, shapes, and compositions.

Inorganic particles are relatively dense and have a high index of refraction. Organic particles are close to water in density and index of refraction. But aggregates of clay may have an average density much less than the clay of the individual pieces and an index closer to water. So there is a wide range in size, composition, index, and density of particles in the ocean. In fact, it has been observed that the total mass contained in each decade range of size is roughly the same from fine clay to whales. Clearly this can't continue indefinitely and still conserve mass.

Zaneveld produced the first major improvement over the Secchi disk, as far as a widely used instrument, with the optical transmissometer. He observed that forward scattering is sensitive to large particles as well as small particles. A measure of loss in unscattered light is a good measure of total projected area of particles. In the transmissometer, or c-meter, light from a chopped red LED is collimated to 15 mm (to sample a larger volume and to be only 3 milliradians in beam angle). This beam passes through 25 cm of seawater (1 m pathlength and down to 5 cm pathlength instruments have been made) and is refocused on a photodiode. The signal from the photodiode is synchronously detected to reject light not coming from the chopped source. Baffles further reject sunlight. Light scattered more than 18 milliradians is lost. Full-scale output, that possible with pure water and no windows (a theoretical result, not ever realized) would be 5 volts and no transmission is 0 volts. Considering reflective loss at the two glass/water interfaces and 8.7% loss for pure water at 670 nm, 4.565 volts is the highest it can

read. In air, with a higher index mismatch but no attenuation in the fluid, about 4.75 volts is expected and this is the precruise calibration that is monitored unless really clean seawater is available.

The c-meter has a Beer's law dependence on concentration so -log attenuation is proportional to concentration. Transmission is voltage measured/voltage full scale with windows. Attenuation is 1-Transmission. Actual concentration of suspended particles must be deduced from the attenuation signal by calibration against actual samples of population of stuff in suspension, neither aggregated not disaggregated. Such samples are very hard to obtain except if the transmissometer is lowered with bottles on a CTD/Rosette sampler. Then particle sizes can be measured with a Coulter counter, and dry weight of particles can be measured on a filter.

At high concentrations of particles, coastal regions and estuaries, the transmissometer may lose sensitivity at the low end, even a 5-cm transmissometer. Blacker than black. But a backscatter instrument may still have a signal proportional to concentration. The Optical Backscatterance Sensor (OBS) measures light scattered back into a broad cone of backscatter angles. This is insensitive to concentrations of milligrams/liter but still works for gm/l. At extremely high concentrations, 10-100 gm/l, it can work as a transmissometer in a range where signal falls with increasing concentration. It too needs samples of suspended sediment for real calibration of gm/l/volt.

In situ size distributions can be determined with LISST, based upon an inversion of the scattering angle distribution. Since large particles scatter light by small angles and small particles scatter into large angles, a photodiode array can obtain a distribution of scattering angles from a distribution of particles and this angular scattering function can be inverted for size distribution. Unscattered transmission can be measured at the same time for total attenuation.

Spectral properties of the scattering or absorption give information about organic components and living components of seawater. The chlorophyll content can be determined crudely with a fluorometer, in which chlorophyll fluorescence is excited by blue light and green scattering at 90º is sensed. More specifically, the spectral distribution of fluorescence can be determined, as it is in the AC-9 fluorometer, to distinguish living chlorophyll from various degradation products that also fluoresce. An even more sophisticated fluorescence instrument, the Sapphire, excites multifluorescence bands with various excitation wavelengths to obtain a fingerprint of coastal waters capable of identifying river sources by the specific compounds giving specific responses.

As a tracer of water mass and as a probe of water mass transformation, multispectral techniques are valuable, but as a simple detector of organic vs. inorganic sediment, fluorometers suffice and can be used in conjunction with scattering or transmission instruments to estimate organic transport.

The downwelling radiation from daylight is important for growth of phytoplankton in the photic zone. Measurements of light at the surface and at depth can be made with a radiometer. Directional information is useful to model the radiative transfer through an absorbing and scattering medium but the phytoplankton are mostly influenced by total illumination. Nonetheless, downwelling and upwelling radiation is measured with lowered radiometers. From surface light levels to bioluminescence at depth, the range is about 10 orders of magnitude, a real problem for a sensor. Shutters with pinholes are sometimes useful for such a large range. Log response with photomultipliers can be achieved by adjusting the voltage and thus the electron multiplier gain for constant output and measuring the voltage. Alternatively, the gain can be switched to one of several ranges and the range and output recorded. But in the upper ocean, the range of light levels is within the range of photosensors without shutters. Log amplifiers can be used to obtain the dynamic range required without range switching. These use the exponential relation between current and voltage in a forward biased diode to compress the range much as the constant current output of a photomultiplier can give a voltage proportional to the log of illumination.

The spectral radiation upwelling from the sea is useful for remote sensing by satellite. of phytoplankton abundance. Absorption of light in the blue by chlorophyll relative to the green and red can be detected remotely by color. Spectral upwelling and downwelling radiometers are used to support satellite programs. The light available after dispersion by a prism or grating is much less than the total downwelling radiation and high gain is needed in the detector. Of greater importance, since we live in a signal to noise limited world, is the scattered light. Inside a spectrometer, all the light not being detected at the receiver must be extinguished and not allowed to scatter by multiple bounces into the detector. One solution is to build a double monochromator. The first monochromator passes the wavelength of light to which it has been tuned with little attenuation but the wrong wavelength light that has found its way out the exit aperture is attenuated by a factor of 100 or 1000. The second monochromator passes the tuned wavelength with little attenuation as well but the wrong wavelength light that enters is again attenuated by 100 or 1000 which is sufficient to put it below the level of concern. If a single monochromator is used, great care must be taken with baffles to absorb the unwanted light. A reflection of the incident beam before dispersion can swamp the spectral signal. But even the dispersed light can be a problem if it is scattered to the detector.

The spectral radiometer developed by the NIO group in Goa uses a thermoelectrically chilled CCD array as a photodetector. The lowered temperature reduces the dark current, allowing a longer integration time to increase sensitivity without blooming from dark current. But the value of receiving all the wavelengths simultaneously, as in a film spectrometer, permits reasonable integration times, as 1 second or less. Second order dispersed light in the blue part of the spectrum can fall on the red end of the CCD array. This is a problem that can be ameliorated with a blocking filter at the red end of the CCD. The entire spectrometer can be made compact which keeps the optical speed high while keeping the weight down.

The Goa spectral radiometer did not have a perfect cosine-integrating window. The purpose of the window is to weight the downwelling radiation appropriately. That light which falls on a horizontal surface is the downwelling radiation. Frosted hemispheres and frosted plane windows are used to obtain the desired directional sensitivity. Each seems to decrease the available light an unacceptable amount but the absence of some diffuser gives irregular directional sensitivity. Upwelling radiation tends to be dim and blue and the sensitivity has to be quite high to see a signal above dark current from more than a score of meters deep.

Optical fluorescence is a valuable tool in the lab for phytoplankton identification. The flow cytometer measures the optical properties of single cells as they pass through a cuvette. In the lab, the stream of cells is concentrated into a fine column by flow focussing. In this technique, the cell containing flow is surrounded with a cell-free sheath of filtered water. Then both are accelerated by a constriction in the channel diameter and the cells are constrained to a column only about 10 μ in diameter. This region is illuminated by a laser (formerly another bright light) and the scattering at 90º is measured, the fluorescence at several wavelengths measured, and the responses categorized to define a specific phytoplankton. Big green fluorescing cells may be one type while small red fluorescing cells are another. This tool has opened up the study of nanoplankton and picoplankton at sea and now flow cytometers are taken to sea as well as used ashore to process samples taken at sea. A development is underway to put the flow cytometer in the sea for tows and profiles.

The biggest problem foreseen for the in situ flow cytometer has been the fluid focussing requiring filtered water and precise pumping. For the in situ instrument, a coincidence requirement has been implemented that selects only cells on the flow axis and in the center for measurement. There is no sheath flow. Rather, two infrared beams focussed in the center of the flow, one slightly above the other, are used to select particles that by chance are in the center of the channel. Then the scattered light from a frequency doubled laser (blue/green) is measured and the fluorescence at two wavelengths. These are recorded and the other signals are ignored.