



# Spectrofluorescence and reflectance properties of Arctic benthic algae as future LiDAR targets

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## Context

Due to overall ice coverage decrease caused by warmer air and ocean temperatures, a regime shift (Fig 1a) in Arctic macroalgal communities has since been observed, where brown and red macroalgae (Fig 1c) are seen covering or replacing encrusting algae and sessile organisms (Fig 1b) (Svalbard)<sup>1</sup>. Increased light availability to the benthic environment is also believed to contribute to this change.

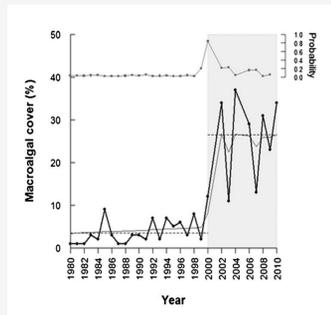


Fig 1a



Fig 1b



Fig 1c

Newly recruited macroalgae (circled)

<sup>1</sup> Kortsch, S et al. 2012. "Climate-Driven Regime Shifts in Arctic Marine Benthos." *PNAS* 109, no. 35 (August 28, 2012): 14052-57.

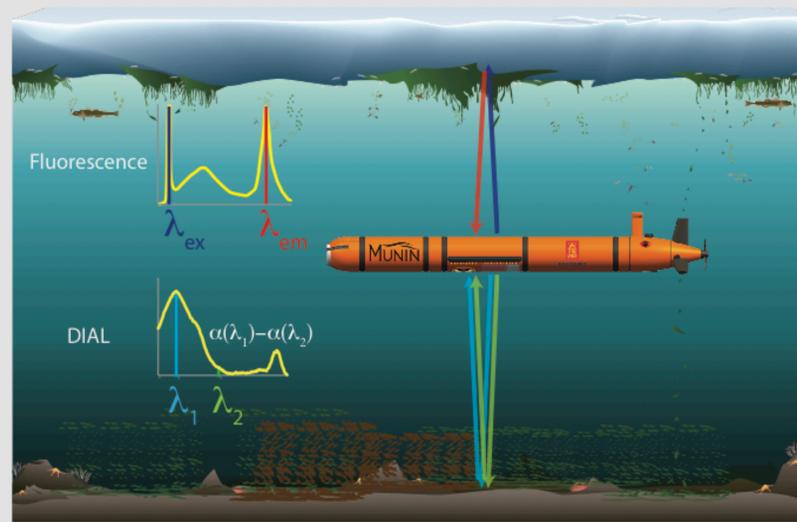


Fig 4. Sentinel North subproject 2.7: Development of an underwater scanning LiDAR built for integration onto an Autonomous Underwater Vehicle (AUV) for the study of benthic and under-ice substrates by their fluorescence and differential absorption (DIAL) properties

## Objectives

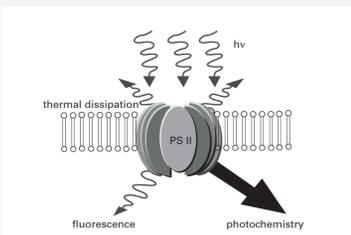
Remote detection of photosynthetic organisms (i.e. phytoplankton) via light stimulated fluorescence is well understood, where fluorescence signal intensity can be linked to in water chlorophyll a (chl-a) concentration. Macroalgae, however are different in their 3-D morphology (e.g. blade, filament), surface characteristics (e.g. flat, rough) and packaging of chl-a, factors possibly affecting fluorescence and absorption return signals when observed by remote methods.

Following the AUV LiDAR approach (Fig 4), macroalgae must be studied whole. Hence, we initially attempt to characterize their fluorescence properties as encountered by our LiDAR. Our initial approach is to investigate fluorescence response via spectrofluorometry analyses on macroalgae samples.

We describe the response to multiple wavelengths of light to guide LiDAR component selection (e.g. laser source  $\lambda = 473\text{nm}$ ,  $\lambda=532\text{nm}$ ), as well as better understand how target structure may influence fluorescence detection.

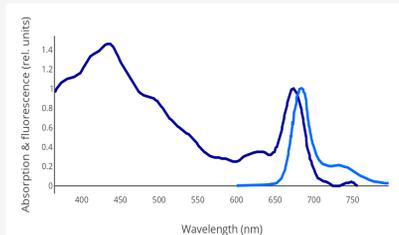
## Theory

### I. Fluorescence



Chl-a use of light (hv) for photosynthesis (photochemistry) and byproducts (thermal dissipation + fluorescence)

Source (adapted): Huot, Y. and Babin, M., 2010. In *Chlorophyll a fluorescence in aquatic sciences: Methods and applications* (pp. 31-74).



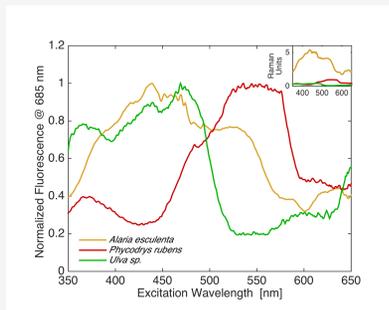
Chl-a light absorption (dark blue) and fluorescence emission (light blue) spectrum showing partial overlap

### II. Nature - pigment assemblage

Fluorescent response at typical chl-a peak emission wavelength (i.e. 685 nm) to excitation wavelength (350 - 650 nm) in brown, red and green macroalgal groups.

Here the normalized spectra of three typical species are shown for better visualization, while fluorescence is shown corrected to Raman units (true intensity) in figure inset.

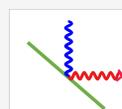
Color groups are characterized by their differences in photopigment assemblages and absorption properties.



### III. Structure - complexity

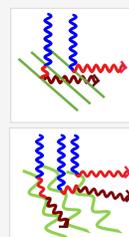
How target substrate structure may affect light interaction with matter (i.e. algae)

Source light (photons in blue sinusoidal line - i.e. 473 nm) reaching algal surface (green) can mostly be emitted (red) at usual 685 nm peak and minimally reabsorbed in simple structures (left figure).



Increasingly complex algal structures (top right - simple layers; bottom right - irregular complex layers) can allow emitted light (red) to be in part reabsorbed and reemitted at longer wavelengths (purple) by the algae.

Absorption and scattering within the structure and medium (water) can also affect observed fluorescence (light reabsorbed and energy reduced).

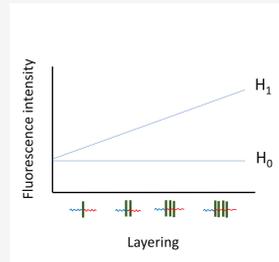


## Hypotheses

### Manipulative experiments

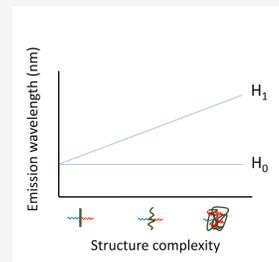
#### I. Layer / thickness effect

Increasing macroalgae layering in optical path will increase detected fluorescence signal intensity (H1):



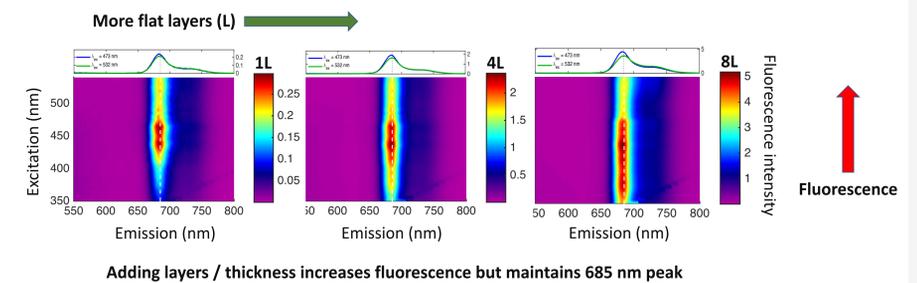
#### II. Structure complexity effect

Increasing macroalgae structure complexity in optical path will affect detected fluorescence emission peak (H1):



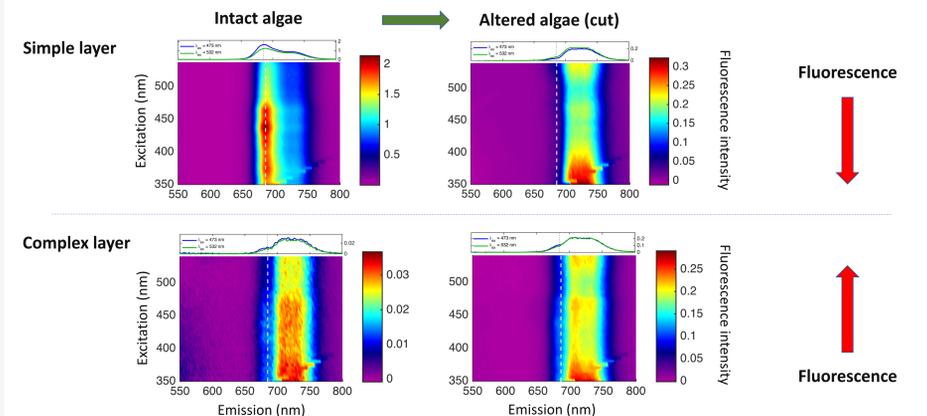
## Results

### I. Layer effect



Adding layers / thickness increases fluorescence but maintains 685 nm peak

### II. Structure complexity effect



Increasing complexity changes fluorescence intensity and peak wavelength

**Conclusions:** In brown kelp *Alaria esculenta*, observed fluorescence increases with the addition of flat layers, while emission peak remains at 685 nm regardless of changes in thickness. In more complex surfaced *Agarum cribosum* (and altered *A. esculenta*), effects of water reabsorption near 685 nm (possibly related to an increased optical path in water surrounding the sample), and possible reabsorption by chl-a at 675 nm (see chl-a - Theory) of initially emitted fluorescence + reemission at 705-730 nm are thought to be observed. This latter phenomenon appears to result in a visible shift + widening of the fluorescence emission band, and reduces total fluorescence energy.

### IV. Experimental setup effects

Fluorescence signal detection and intensity may be affected by experimental conditions. For example, the path length of light through water in and surrounding a sample: a long path length equals more absorption and less signal for the same initial light output!

## Acknowledgments

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## Upcoming

Laser benchtop experiments on live samples!

- Fluorescence
- Reflectance - BRDF
- Polarization - retroreflection

### Polarized reflectance / retroreflection ideas

