Observations of reflectance spectra of Antarctic krill (*Euphausia superba*) from different environments in the Scotia Sea, Antarctica: *Final report to Antarctic Science Ltd*

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Project Overview
Estimates of krill biomass in the Southern Ocean are highly uncertain due to their wide areal distribution (~19 million km^2) and the very patchy and overdispersed distribution of krill swarms. Traditional krill survey methods utilise nets which suffer problems of net avoidance. Although newer acoustic methods can provide more synoptic scale measurements, converting acoustic returns to krill biomass is not trivial and requires knowledge of a number of parameters which are difficult to measure *in situ*. Additionally, as transducers are often hull-mounted, most acoustic estimates miss the krill in the surface layer (upper 16m). Remote detection via platforms such as, satellites and UAVs (unmanned aerial vehicles), offer a way of collecting synoptic data over large spatial scales and, with continued improvements in spatial, temporal and spectral resolution and could provide information on these poorly sampled surface krill populations. As yet however, the reflectance spectra of live krill collected *in situ* have not been measured, which is the necessary first step for remote detection of krill swarms. The aim of the study was make key initial reflectance measurements of krill, to determine if krill have a characteristic reflectance signature that could be utilised for remote detection.

Field Campaign
Krill reflectance experiments were carried out during research cruise JR19001 aboard the *RRS James Clark Ross* in November/December 2019. Krill swarms were located using a SIMRAD multifrequency echo-sounder (Figure 1A) and sampled using a Rectangular Midwater Trawl (RMT8). Three main regions were sampled, the South Orkney Islands, the South Georgia shelf, and northwest South Georgia. This provided krill from a variety of feeding environments.

From each net haul sampled, a subsample of krill were taken for pigment analysis via extraction in acetone (Figure 1B). Krill were homogenised, and left in the dark at -20 °C for 24 hours, before measuring the absorbance of each sample on a Cary60 UV-Vis spectrophotometer. These measurements were conducted to assess for the presence of the pigment astaxanthin, which is known to be a dominant pigment in krill.

Additionally, lab experiments were carried out on board to measure the reflectance spectra of live krill. These measurements were carried out in a bucket of underway seawater using an ASD FieldSpec Pro Spectroradiometer (Figure 1C). Krill were added sequentially, with reference readings taken at the start and end of each experiment using a 20% grey spectralon panel. Direct measurements were also attempted using a contact probe connected to the ASD FieldSpec Pro. 3-5 krill were placed on a black tray and the reflectance spectra measured by holding the probe against the krill.

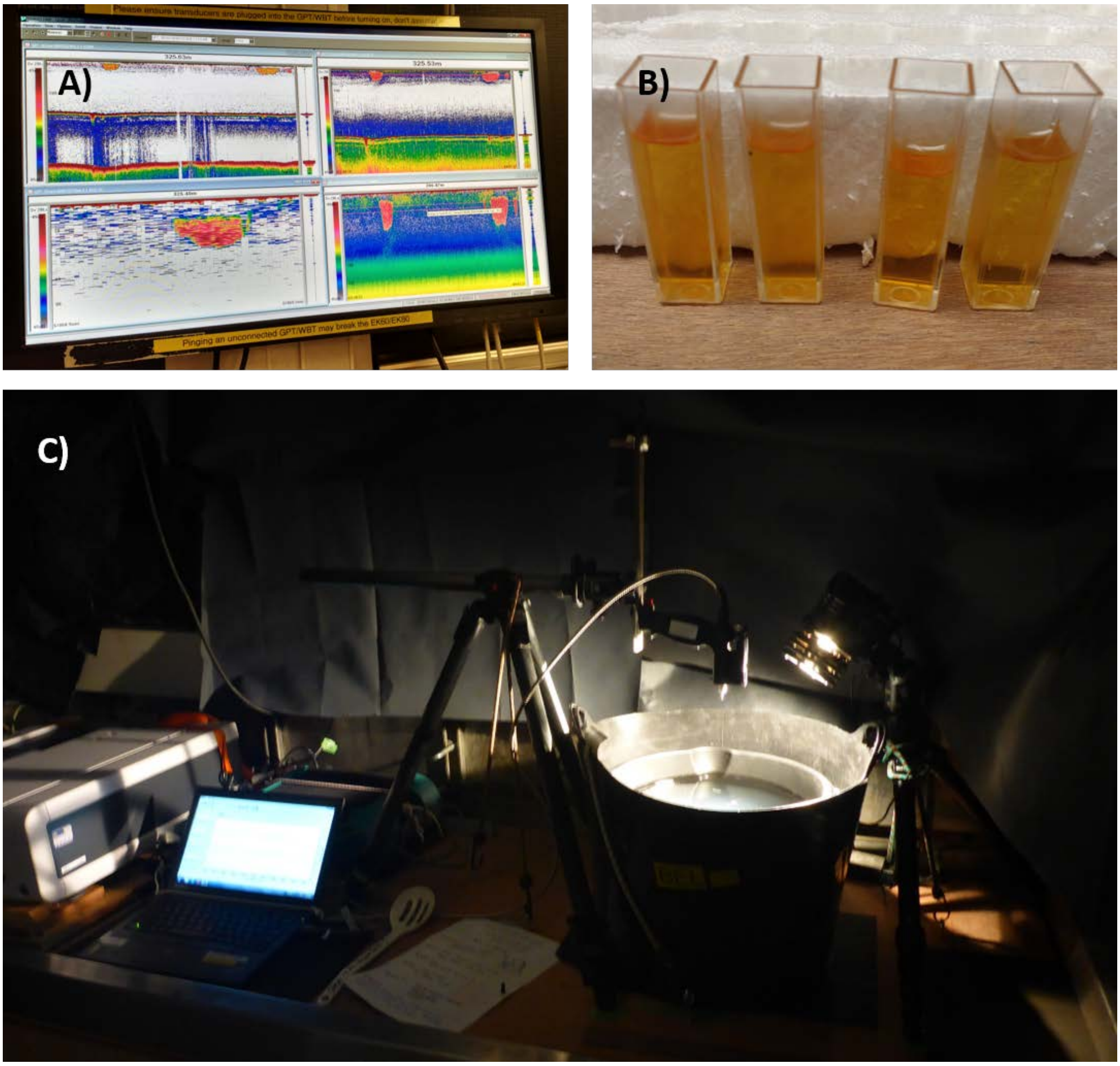

Figure 1: A) Searching for krill swarms using the echo-sounder, B) Krill pigment extraction, C) The lab set up to measure krill reflectance- nicknamed 'Dexter's Lab'

Initial results

Sampling was successful during JR19001, with reflectance experiments conducted at 10 stations (Table 1). Pigment extractions in acetone revealed a defined peak in absorbance at 480 nm which corresponds well to the pigment astaxanthin. Reflectance spectra measurements from lab experiments showed clear differences between the spectra of underway seawater only, and that of underway seawater with krill. A strong reduction in reflectance was measured between ~450-600 nm, agreeing well with pigment extractions and the absorption peak of the pigment astaxanthin. Contact probe measurements were inhibited by the fogging up of the sensor due to the heat of the bulb and water content of the krill, and so data are not reliable from this method.

Table 1: Details of stations sampled for reflectance measurements during research cruise JR19001

Time (GMT)	Latitude (°N)	Longitude (°E)	Event number	Krill sampled
13/12/2019 03:02	-53.5901	-40.4784	75	Juvenille
12/12/2019 20:47	-52.83	-40.1391	74	Juvenille
11/12/2019 04:40	-53.7699	-38.4103	66	Juvenille, adult
11/12/2019 01:35	-53.7393	-38.1479	65	Female, male
08/12/2019 05:23	-53.72	-38.4086	48	Juvenille, adult
08/12/2019 00:39	-53.7446	-37.9203	46	Juvenille, adult
05/12/2019 04:47	-53.7631	-38.8312	30	Juvenille, adult
03/12/2019 02:48	-53.7987	-38.3458	19	Adults
02/12/2019 20:00	-53.7674	-38.0254	15	Juvenille, male, female
24/11/2019 18:30	-60.3936	-46.5036	7	Juvenille

Next steps

Analysis of the field data has advanced and a manuscript is currently in draft for submission to Antarctic Science. The results represent the first step towards being able to detect krill remotely, and also highlight some of the hurdles that remain to be tackled. Obtaining clear satellite imagery in the Southern Ocean is a challenge due to cloud cover, particularly to match field sampling. We suggest the need for further studies, combining targeted surface sampling for krill with UAV surveys, in the hope to allow collection of true field reflectance (i.e. without possible interference from the lab environment) and to ground truth these data.

Figure 2: Team science on research cruise JR19001: Spot those doing their best impression of krill