Gazing at the sea with new X-ray eyes: the starfish as sentinel of climate change

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Pierre Thibault, Università degli Studi di Trieste, Trieste (Italy), originally from Québec (Canada), is an expert in X-ray imaging and tomography. He obtained his PhD at Cornell University and has conducted research in Switzerland, Germany and the United Kingdom. He is now professor at the University of Trieste, where he pursues his EU-funded research project.

On a drizzly morning of February 2020, Ms. Christina Wood, researcher at the National Oceanography Centre of Southampton (United Kingdom) met for the first time Dr. Irene Zanette, a physicist and at that time a Royal Society fellow at the University of Southampton. Ms. Wood was interested in new tools to inspect her precious samples, which she carried in a big jar to show to Dr. Zanette. From this first meeting, a fruitful collaboration started to emerge: the task entailed exactly the type of challenge that Dr. Zanette was ready to take on.

Ms. Wood's samples were small sea stars, *Ctenodiscus crispatus*, commonly called mud stars in English because of the habitat in which they are most commonly found. *C crispatus* is a dominant species across the Norwegian Arctic, comprising more than 3% of the biomass in this region (Wassmann P. et al., 2006) The small sea star - less than 10 cm across - lives in the muddy sea floor along the coast of all the circumboreal regions. It buries itself in sediments and feeds on organic detritus. It is believed to have an important role in "cleaning" the bottom of the sea, through bioturbation, the process of mixing the fine particles that accumulate constantly at the bottom of the sea. The living activities of *C crispatus* - how it feeds, moves and reproduces - contribute to ecological cycles, including carbon capture.

These details were new to Dr. Zanette, an expert in X-ray physics and imaging, as they were to me when I later joined the collaboration. However, we know, as does most of the population by now, that anthropogenic climate change has become an emergency, with observed effects everywhere across the globe. Climate warming and other human-induced environmental changes have complex effects in the balance of all ecosystems. The seas and oceans are not spared. How will *C. crispatus* be affected? How are individuals able to adapt to important changes in their habitat, in particular with regard to their nutrition and reproduction? Can these changes help to monitor and predict the evolution of the ecosystems in these areas? These questions are at the core of Ms. Wood's research project (Solan M., et al., 2020).

Answers can only come from observations and experiments. The stars were obtained by the Norwegian research vessel G.O.Sars, in collaboration with the Institute of Marine Research (IMR) in Norway. During these benthic trawls, the vessel picks up sediments and fauna from the seabed, which are then sieved and preserved for further analysis. Dissection, the standard investigation technique among biologists, is normally used to determine various anatomical details of the collected animal samples. However, irreversibly damaging samples makes it difficult or even impossible to study multiple characteristics on a single individual. Samples are scarce and thus precious, and the expeditions are rare and costly. An alternative to dissection was needed to study the samples without affecting their physical integrity.

Peering into intact, opaque objects has been precisely the defining feature of X-ray imaging since Roentgen's discovery of the up to then unknown *"X radiation",* 125 years ago. Familiar to everyone for medical diagnostics or security screenings, X-ray imaging has become a versatile tool that fulfils a vast range of needs. Recent research in the field focuses on important technical improvements, e.g. for speed, dose reduction, better resolution and better contrast. Fundamental questions are also investigated, in particular the search for imaging procedures that exploit interactions between X-rays and matter that have been ignored so far.

Much of the cutting-edge research is conducted at synchrotron radiation facilities, large particle accelerators designed to generate powerful X-rays that have laser-like characteristics. In 2018, Dr. Zanette received a University Research Fellowship from the Royal Society for a 5-year project. Her goal is to transfer advanced X-ray imaging methods from the synchrotron to conventional X-ray laboratory sources - similar to those found in hospitals or airports - making them broadly available for the scientific community. My work on high-resolution X-ray imaging methods, in particular *ptychography*, is also concerned with synchrotron-based techniques. Thanks to the support from the European Research Council (ERC Starting grant project OptImaX), some of the techniques and analysis software my group has helped develop are now used routinely in synchrotrons throughout the world. With renewed support from the ERC in 2020 (Consolidator grant project S-BaXIT), I moved my laboratory and group to the University of Trieste, where I pursue my research also thanks to the presence of Elettra, Italy's only synchrotron radiation facility.

In December 2020, we had the first opportunity to examine some of Ms. Wood's sea stars at the synchrotron Elettra. This first round of measurements, on juvenile samples between 1 and 2 cm across, took place at the SYRMEP beamline, an instrument dedicated to X-ray imaging and tomography. To study their capacity to adapt, we imaged samples from two different environments: a sheltered fjord, and the open Barents Sea. We assembled an excellent team of young and motivated researchers. Dr. Adriano Contillo, Ms. Sara Savatovic, and Mr. Stevan Vrbaski all contributed to the results that we have obtained up to now. Unfortunately, Ms. Wood could not join because of the COVID-19 pandemic.

A two-dimensional projection of one of the samples is presented in Fig. 1. This type of image provides a phase-contrast enhanced map, where mostly the exoskeleton of the sea star is visible. Preliminary results of the tomographic reconstruction of another specimen are shown in Fig. 2. A slice through the reconstructed volume shows a large amount of sediments inside the animal's stomach, also depicted in blue in the three-dimensional rendering.

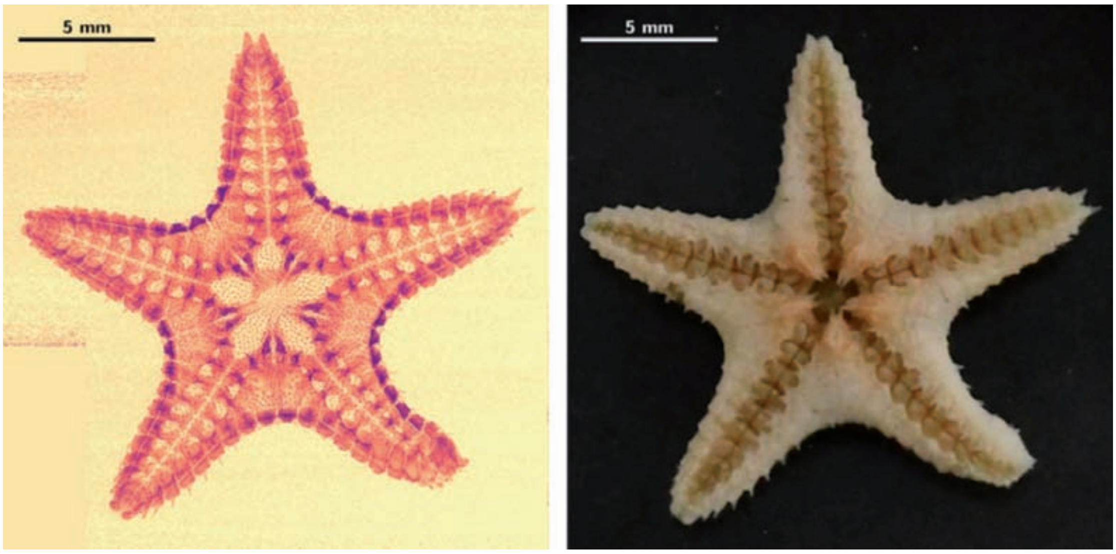

Fig. 1 - Left: radiograph of one of the *C. crispatus* specimens, showing the intricate structure of its exoskeleton. Right: a photograph of the same sample.

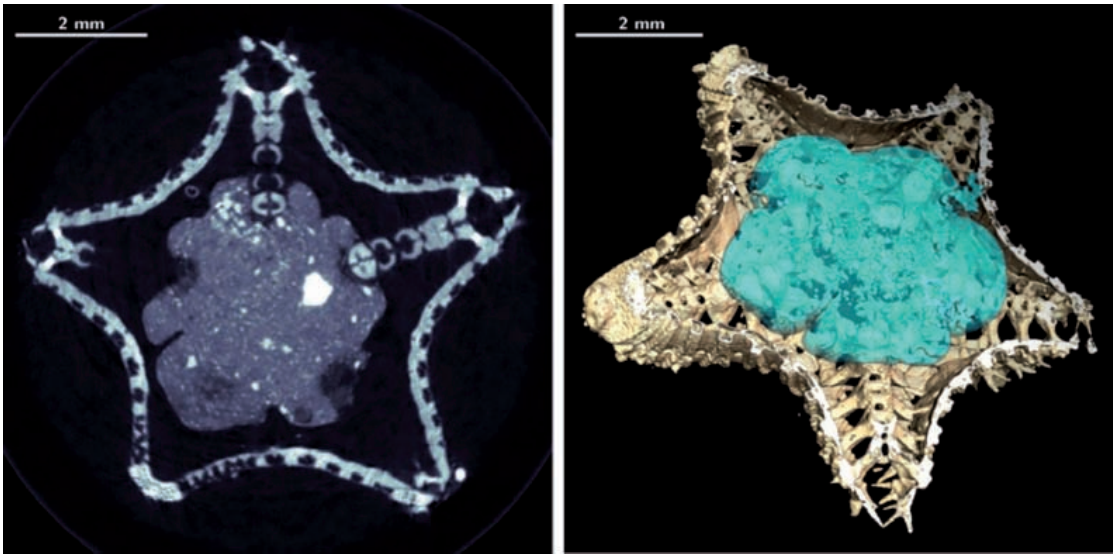

Fig. 2 - Left: tomographic slice of a different *C. crispatus* specimen. The exoskeleton is the brighter part. The grey mass in the centre is sediments and small rocks in the starfish's stomach. Right: three-dimensional rendering of the same dataset.

While the results are already promising, this experiment was the first at SYRMEP for most of the team, with an imaging procedure never tested before on this instrument. During our next measurement campaign, we will apply advanced phase-contrast methods (Zdora M-C. et al., 2020) to push the contrast of soft tissues (e.g. digestive glands and reproductive organs) within the starfish, which are normally much more difficult to distinguish with X-rays.

The collaboration continues. In the near future, more samples from the Norwegian and Barents seas will be scanned. We are also expanding our work to key animals from Adriatic ecosystems, which are rapidly changing.

Within the ERCfunded project S-BaXIT (*Scattering-Based X-ray Imaging and Tomography*), the study of marine samples – among other applications – will be expanded to new and more exotic imaging modalities. Valued at €2.2M, this consolidator grant has as its main objective the development and application of X-ray imaging methods that exploit up-to-now underestimated scattering phenomena that take place in materials. These developments will span the spectrum from theoretical work to simulations, to experimental demonstrations and applications. The synchrotron Elettra, a partner institution in this project, hosts the laboratory for my group, where a dedicated X-ray imaging setup is under development.

C. crispatus, a humble starfish hidden at the bottom of the cold fjords, could hold a crucial role as the sentinel of unfolding climate upheavals. The sea has many stories to tell. It has much to teach us. Trieste, city of science and of the Adriatic, is the ideal location to let X-rays meet the sea.

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