

# Isolating Nanocrystalline Cellulose from Invasive Island Tunicates

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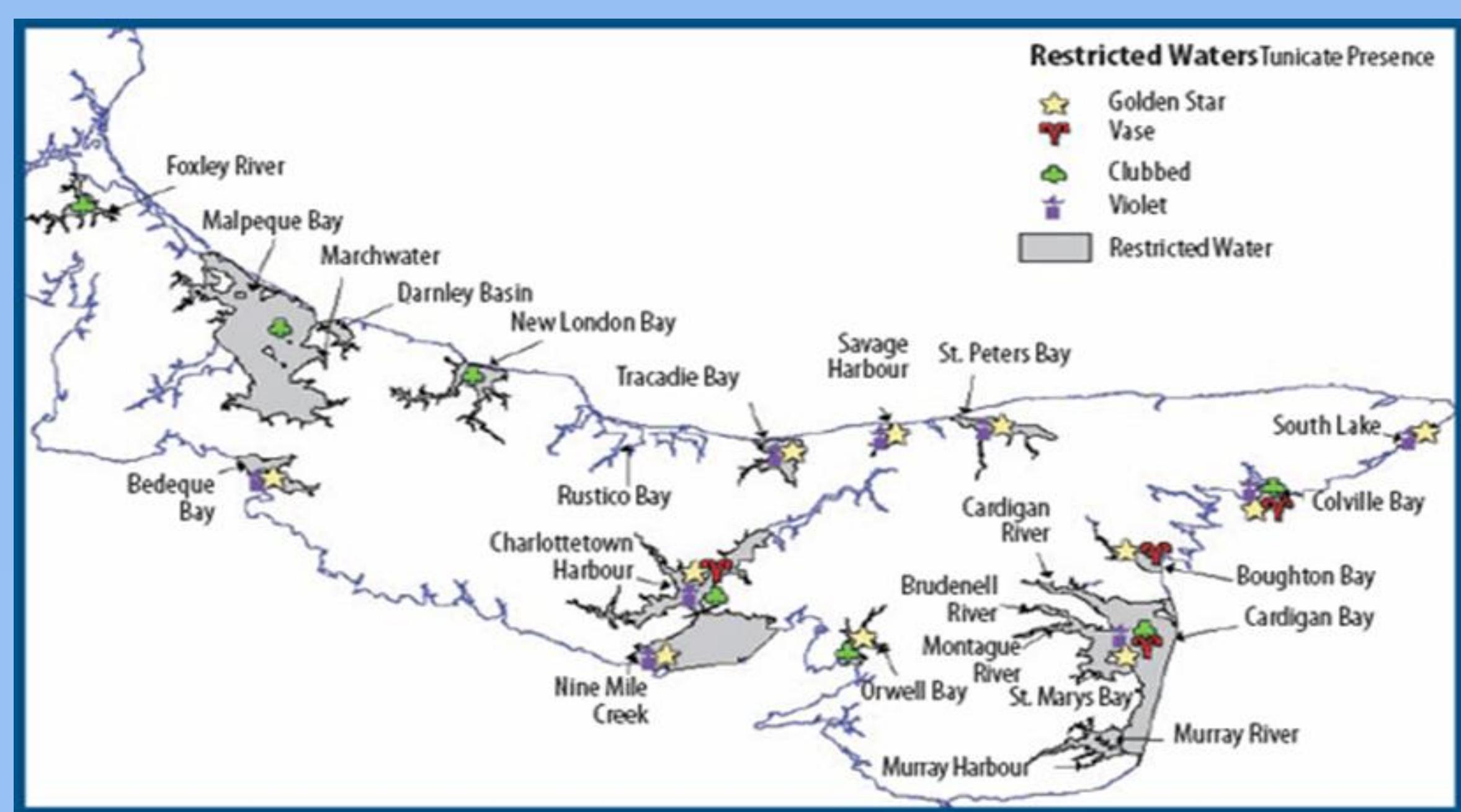
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## Abstract

Cellulose is well known as the most abundant organic polymer on Earth. Nanocrystalline cellulose (NCC) is a valuable commodity, which consists of the nanoscale crystalline region of the cellulose polymer. Tunicates are marine invertebrate animals, whose name originates from their unique integumentary tissue known as the tunic. Tunic tissue utilizes cellulose microfibrils which act as a skeletal structure. Tunic tissue constitutes the only known animal source of NCC. This work seeks to isolate NCC from the four species of invasive tunicates on PEI. Laying academic ground work, towards the industrialization of NCC isolation from tunicates on PEI. In an effort to mitigate or eliminate the many problems posed by local invasive tunicates. Particularly to the local aquaculture community, as well as our island economy. Well concurrently opening the door to new value added economic resources, sourced from a harmful invasive species. The resulting NCC is characterized by scanning electron microscopy (SEM), X-ray diffraction (XRD) Energy-dispersive X-ray spectroscopy (EDX) Optical Microscopy and thermal gravimetric analysis (TGA). The percent yield, crystallinity, thermal properties and surface morphology will be discussed. This information will be used to determine which invasive species of tunicate are suitable for industrial scale up.

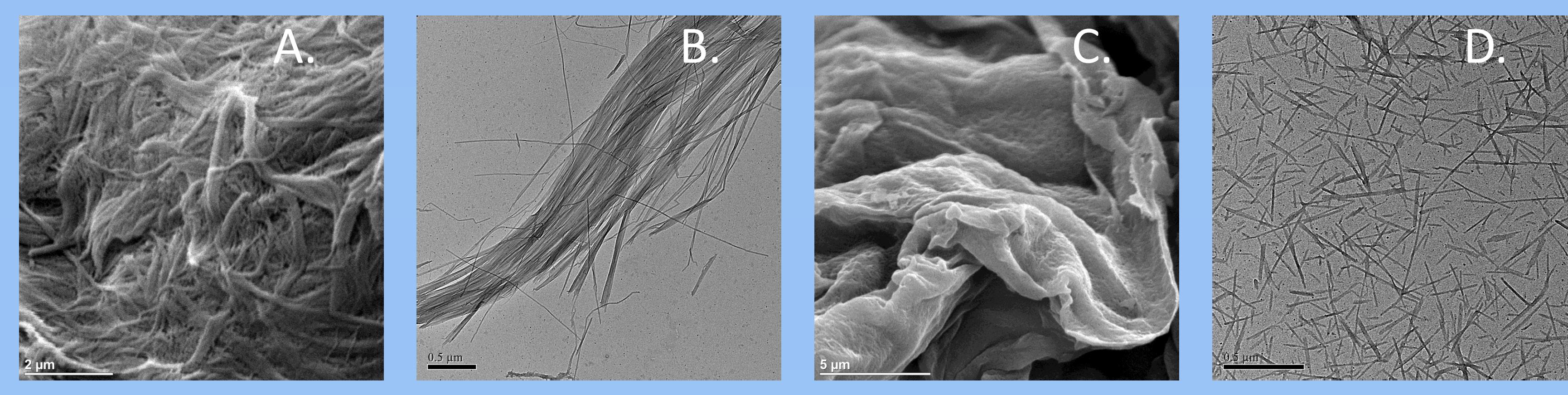
## Introduction

Tunicate sourced NCC is of high commercial value [1]. If researchers can find a large source of tunicates, than the industrial scale isolation of high quality NCC from these tunicates may also be possible. NCC could then be utilized as a value added product, offsetting the cost of implementation. Prince Edward Island possesses a unique set of circumstances which make isolating NCC from tunicates attractive. As the largest mussel producer in Canada, PEI fishermen rely on their ability to pull healthy mussels from our waters [2]. Tunicates have caused a decline in the health of mussels, and in the ease of harvesting them. With four invasive tunicate species and a growing tunicate population, it is imperative to PEI fishermen that a solution be found [3]. Since the mid-1990s, the coastal waters of PEI have become one of the most heavily invaded marine ecosystems in Canada [4].



Among the recent arrivals are four non-indigenous tunicate species:

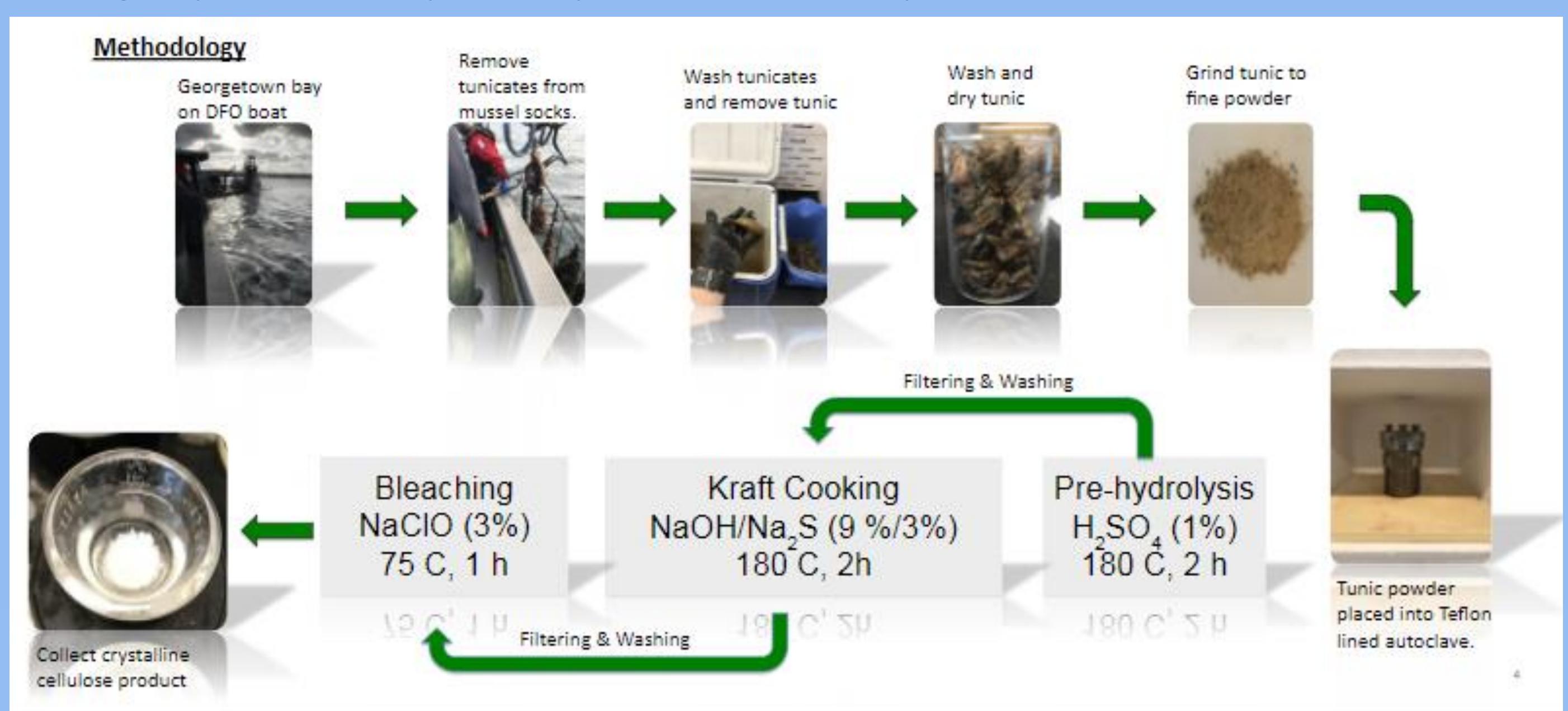
- ❖ Club tunicate (*Styela clava*, Herdman, 1881)
- ❖ Gold Star tunicate (*Botryllus schlosseri*, Pallas, 1766)
- ❖ Violate tunicate (*Botrylloides violaceus*, Oka, 1927)
- ❖ Vase tunicate (*Ciona intestinalis*, Linnaeus, 1767)



SEM (a) and TEM (b) of Club product, SEM (c) and TEM (d) of Vase product.

## Experimental

NCC was isolated from two invasive tunicate species via a modified prehydrolysis-kraft cooking-bleaching sequence first reported by Koo et al [5]. This process is summarized below.

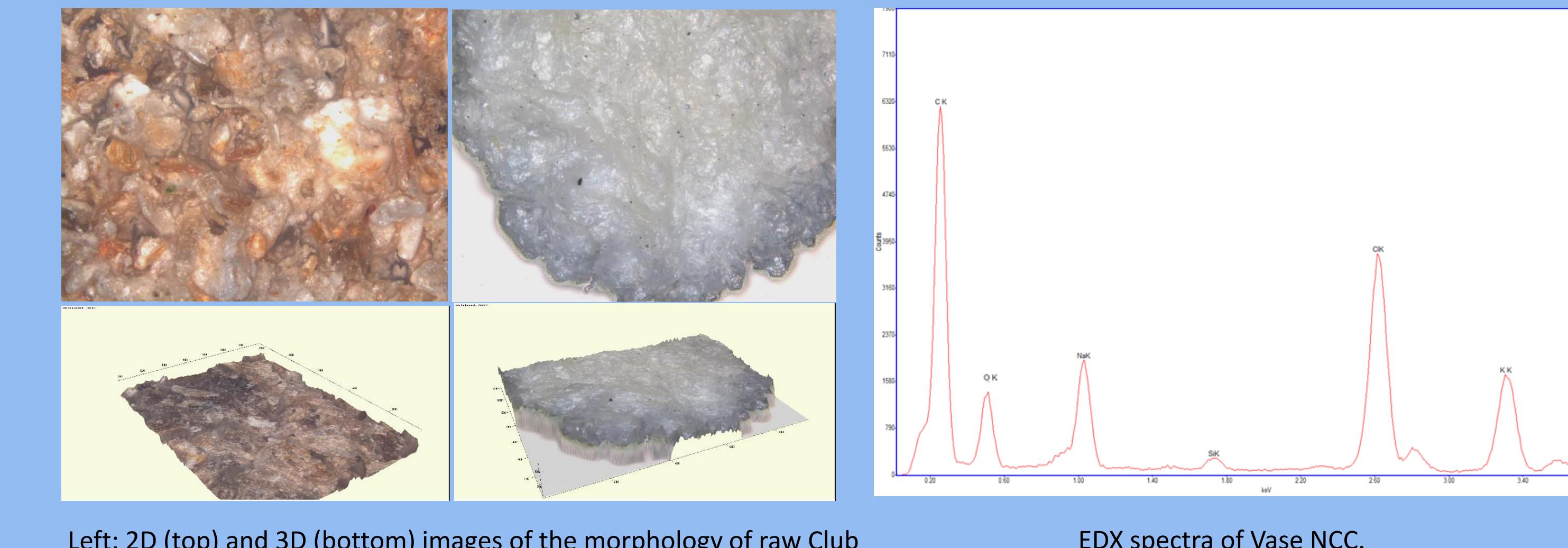
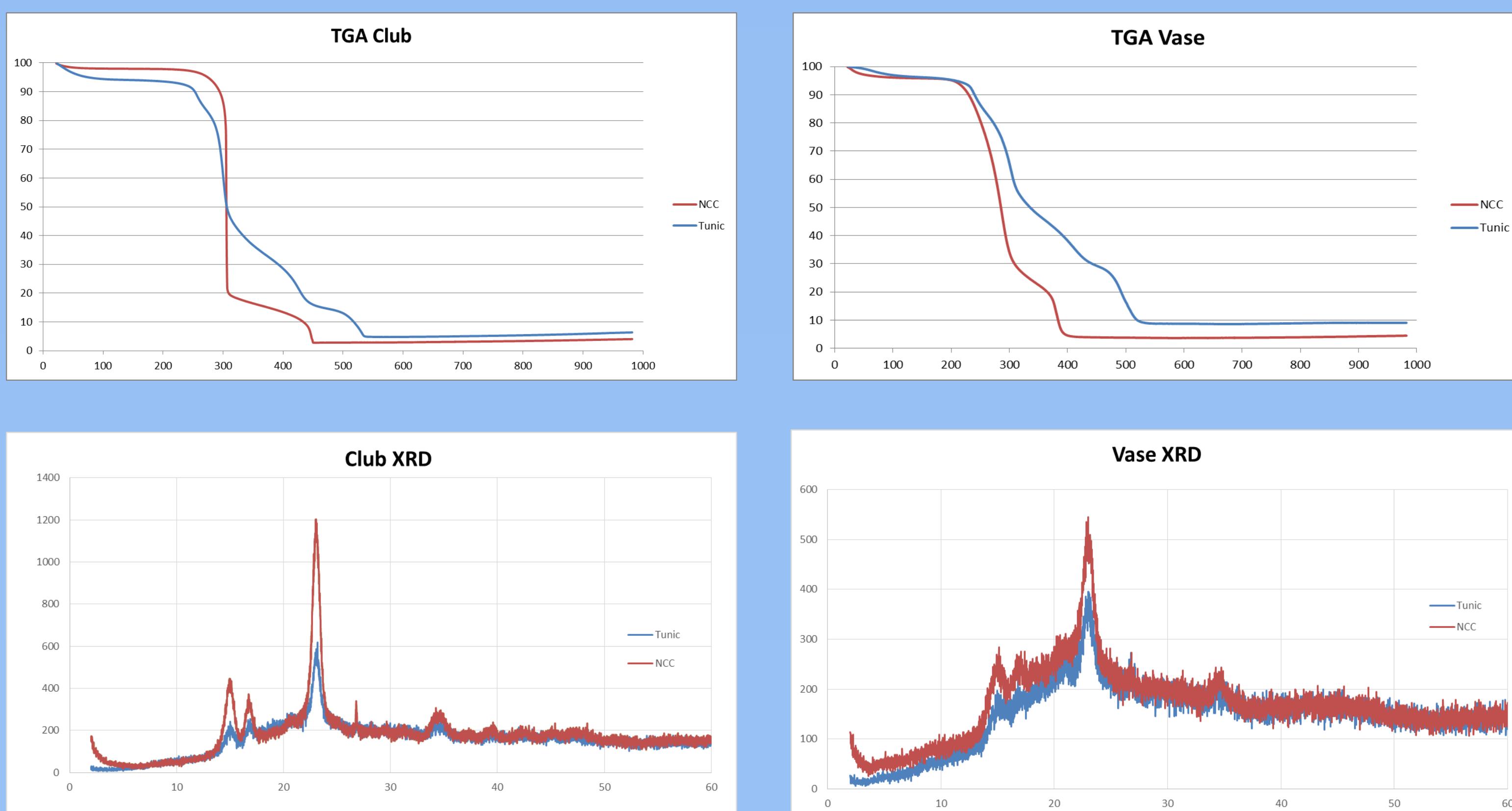


Scheme of the tunicate cellulose preparation. [1]

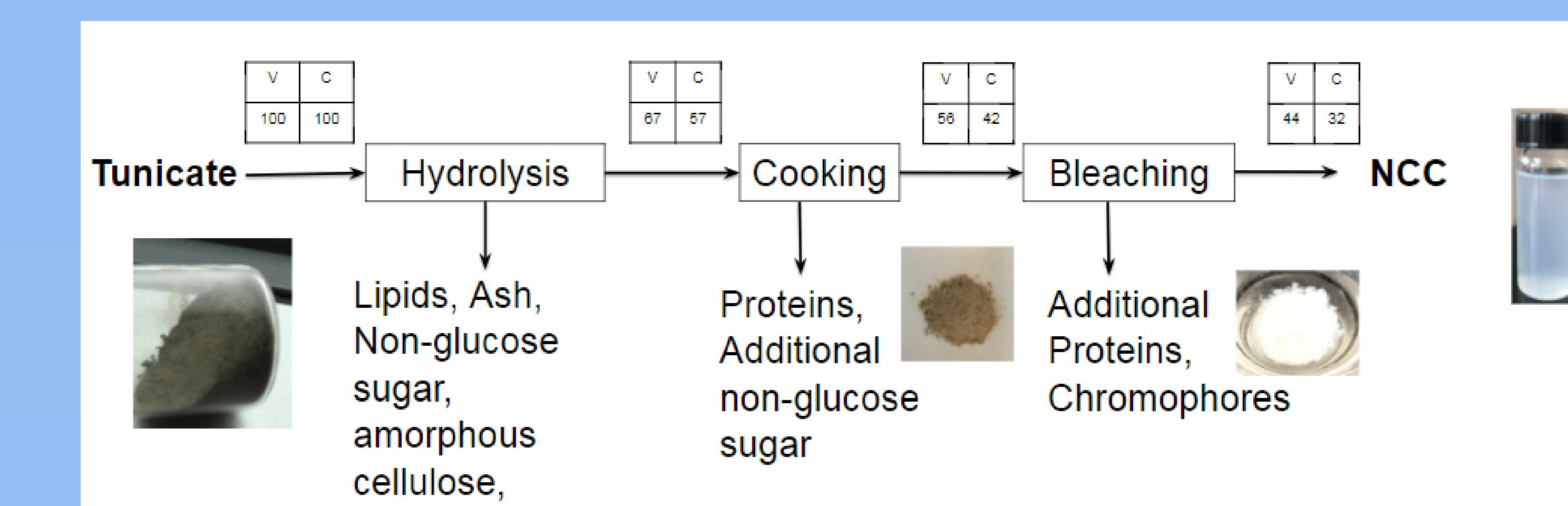
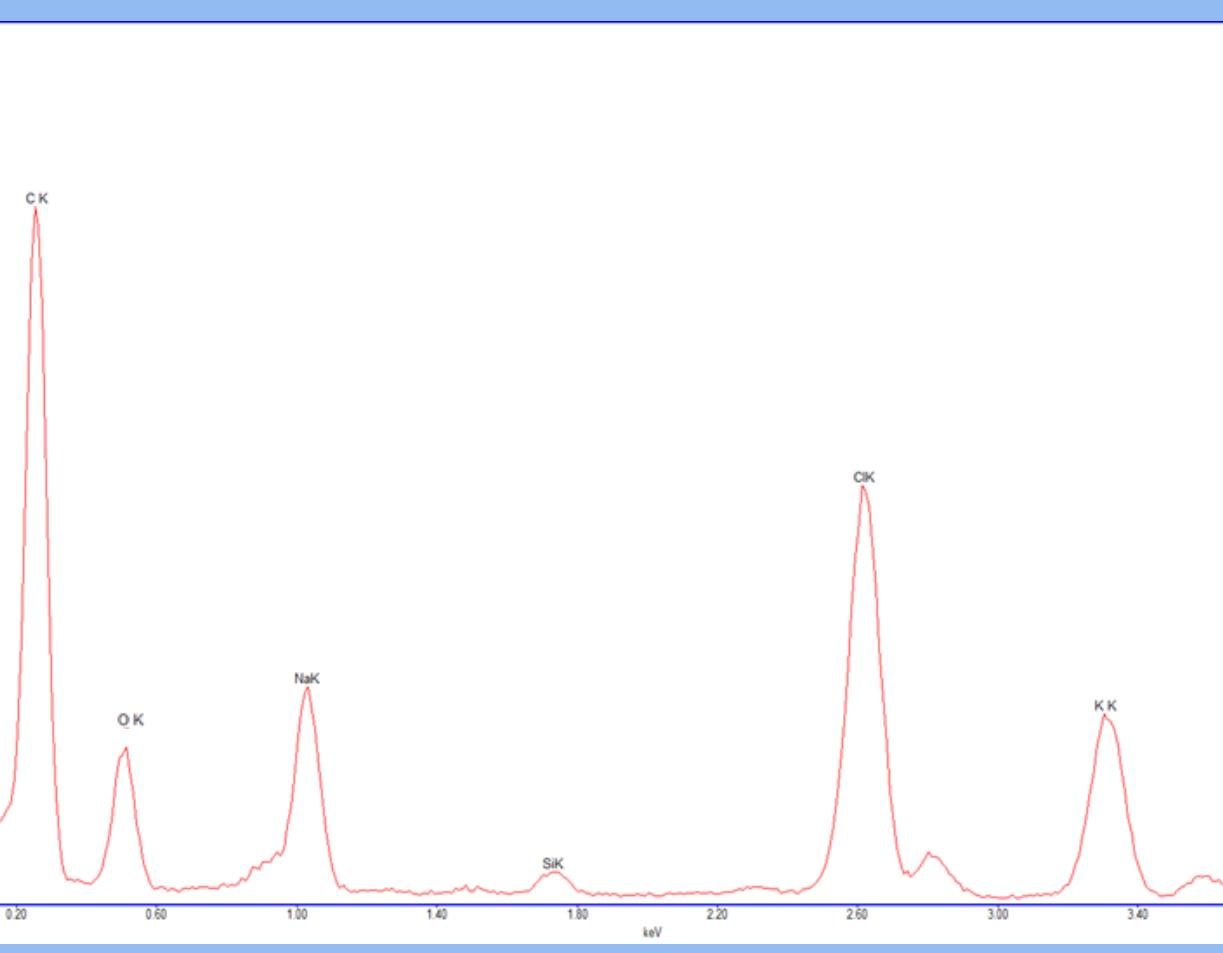
The crystalline cellulose obtained from the prehydrolysis-kraft cooking-bleaching method was washed by centrifugation. Dialysis is then utilized to purify the crystalline cellulose. Finally ultrasonication of the colloidal suspension yields NCC.

## Results and Data

The results presented here support the successful isolation of NCC from two local invasive tunicate species. Both the Club and Vase tunicate have been utilized to obtain NCC yields ranging between 30-45%. Some preliminary data is displayed below.



Left: 2D (top) and 3D (bottom) images of the morphology of raw Club tunic powder. Right: 2D (top) and 3D (bottom) images of the morphology of Club tunic sourced crystalline cellulose.



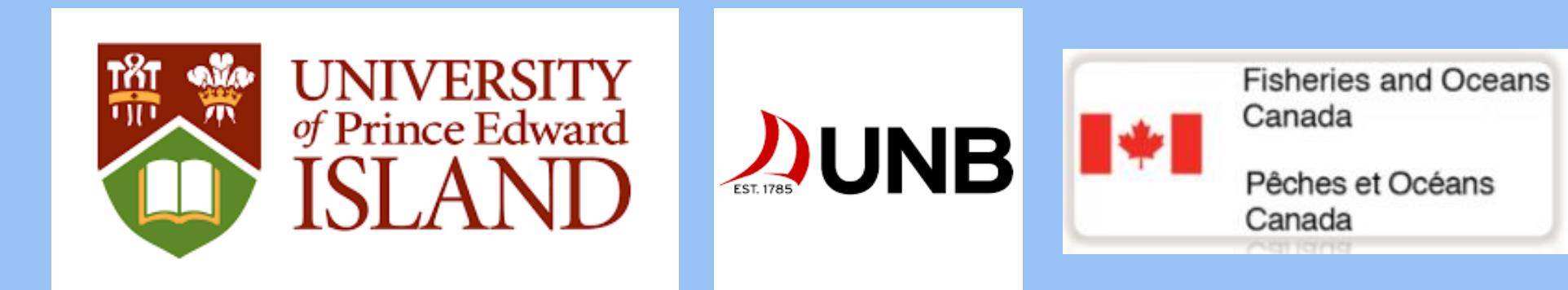
Flowchart of Results Obtained.

## Conclusions

NCC was obtained from two species of local invasive tunicates. The yield for this process was found to be between 30 and 45%. The products have been well characterized, and the resulting data confirms the isolation of NCC was successful. This work suggests that the Club tunicate is a better source for NCC than Vase tunicate. Both in terms of yield and quality of produced NCC. This provides valuable insight towards the future industrialization of this process. Current work focuses on scale up and modification of this process. The potential applications of tunicate sourced NCC in sustainable nanocomposites, superabsorbent materials and 3D-bioprinting are also being investigated.

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