Salting affects the collagen composition of the tropical sea cucumber *Holothuria scabra*

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Abstract

The effect of salting time on the collagen content of the body wall of *Holothuria scabra* (sandfish) is investigated in the present work. Sandfish were harvested from Tavua Bay in Fiji, and processed using an upgraded technique. Sandfish were cooked for the first time followed by immersion in salt and then treated at different salting times of 24 hours (h), 36 h, 48 h and 72 h. The results show that after 72 h, collagen fibers were almost totally destroyed and disappeared from the body wall of sandfish. It is highly recommended that a new and better processing technique be established to prevent post-harvest nutritional losses from beche-de-mer for better value, and to ensure that salting does not exceed 48 h.

Introduction

Sea cucumbers (called beche-de-mer or trepang when dried) are regarded as an authentic cuisine for a number of Asian consumers. It is regarded as a nutritious low-fat food source that is rich in protein and essential lipids. As bottom dweller invertebrates, sea cucumbers tend to bioaccumulate essential nutritional compounds of high value. Sea cucumbers are known to have medicinal benefits such as in the treatment of cancer, arthritis and a number of other medical ailments (Bordbar et al. 2011).

Collagen is a protein that is found in all sea cucumbers, and high-quality is found in tropical sea cucumbers (Dong et al. 2011). Significantly, given the importance of body wall texture in the value of sea cucumbers, collagen has been reported to make up at least 70% of the protein content in sea cucumbers (Saito et al. 2002) and, individually, between 3.4% and 24.3% of beche-de-mer dry weight (Liu et al. 2010; Zhong et al. 2015). Collagen belongs to a family of extracellular matrix proteins that maintain the integrity of various tissues. There are approximately 27 types of collagen found, with 42 distinct types of polypeptide chains and about 20 additional proteins and collagen-like domain, and 20 other collagens modified by enzymes (Kivirikko and Prokop 1995; Myllyharju and Kivirikko 2004). Collagen molecules are composed of three α chains that are mainly stabilised by intra- and inter-chain hydrogen bonds that form the collagen triple helix (Zhang et al. 2013), which contains a repeat of the amino acids glycine, proline and hydroxyproline (Ichikawa et al. 2010). Collagen is used in leather products, biomedical products (e.g. wound dressings, implants and drug carriers), and by the food industry in the production of gelatin (Nam et al. 2008).

There is limited literature on the possible changes in collagen content during the processing of sea cucumbers, and the effects of particular processing steps and conditions (e.g. temperature) on the collagen content of sea cucumber products. Niamnuy et al. (2008) studied changes in the protein composition and the physical effects on shrimp during boiling at different salt solutions. The findings revealed that increased salt concentrations affected the protein quality and texture of the shrimp. When the boiling time and salt concentration both were increased, protein denaturation was more common and the shrimp became harder due to the decrease in myofibrillar, sarcoplasmic and stroma proteins. Niamnuy et al. (2007) reported that boiling time affected shrimp quality through protein denaturation as well as the salt, moisture and protein content of shrimp. The final product's quality can only be determined through organoleptic evaluation, and the extent of quality loss (e.g. appearance, taste, texture and colour due to boiling time and various salt concentrations), affect the overall acceptance of the final product by the customer (Tapaneyasin et al. 2005).

In Fiji, sea cucumber processing is a destructive process, with a series of cooking, salting and drying steps followed by complete dehydration of the sea cucumber for prolonged storage. In Fiji, salting sea cucumbers is done for approximately 48–72 h before further cooking and drying. It is believed that salting leaches away essential proteins and salt-soluble amino acids that make up the proteins.

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The aim of this research was to determine the effect of salting time on the collagen content of the body wall of *Holothuria scabra*.

Methodology

Holothuria scabra were harvested from Tavua Bay in Fiji and were processed using the techniques used by Ram (2017). Briefly, after harvesting, sandfish were laid on a flat surface for approximately 5 minutes (min). Fresh water was heated to a temperature of 80° C and the sandfish were immersed in the water gently and cooked for about 15–20 min, depending on their size. After the first cook, sandfish were immersed in a 3% saline solution for 24–36 h to remove the chalky spicules from the surface. The remaining spicules were gently brushed off to ensurre that the surface was not damaged. The sandfish were processed with different salting treatments of 24 h, 3 h, 48 h and 72 h, using coarse "A" solar dry salt. After the salting treatments, tissue from the sandfish was cut aseptically (1 cm^2) on the ventral side, some 3–4 cm away from the anus, and transferred in a clean vial and stored in 10% formalin at room temperature for initial fixation of the tissues for histology. The samples were then delivered to the Histology Unit at James Cook University in Townsville, Australia, where the collagen composition of the sandfish tissues was analysed using the Picro Gomori staining technique standard university laboratory protocol. After embedding the paraffin, thin sections of the tissue were cut in a microtome and then stained using H&E (Hematoxylin and eosin) stain and orcein combined with either light green or one step Gomori trichrome, and orcein combined with Picro-sirius red for detection of collagen fibers. In the Picro Gomori staining technique, the collagen samples in the tissue were stained green and the tissue as pink.

Results

The results of the four salting treatments (Fig. 1) show that when the sandfish were exposed to 72 h of salting, collagen fibers were significantly lost as compared to 24 h, 36 h and 48 h exposure times.

Discussion

This study investigated the effects of different salt treatments on the body wall of *Holothuria scabra*. The results have shown that extended salting time (72 h) affects the collagen composition in the tissue. Collagen is regarded as an essential part of the protein that forms part of the connective tissue of the human body. Ram et al. (2017) showed the decrease in crude collagen composition from 103 mg/g to 99 mg/g of the dry tissue through the salting treatments at 24 h, 36 h, 48 h and 72 h of *H. scabra* exposure. Their research also showed that as the salting treatment time increased, the crude collagen

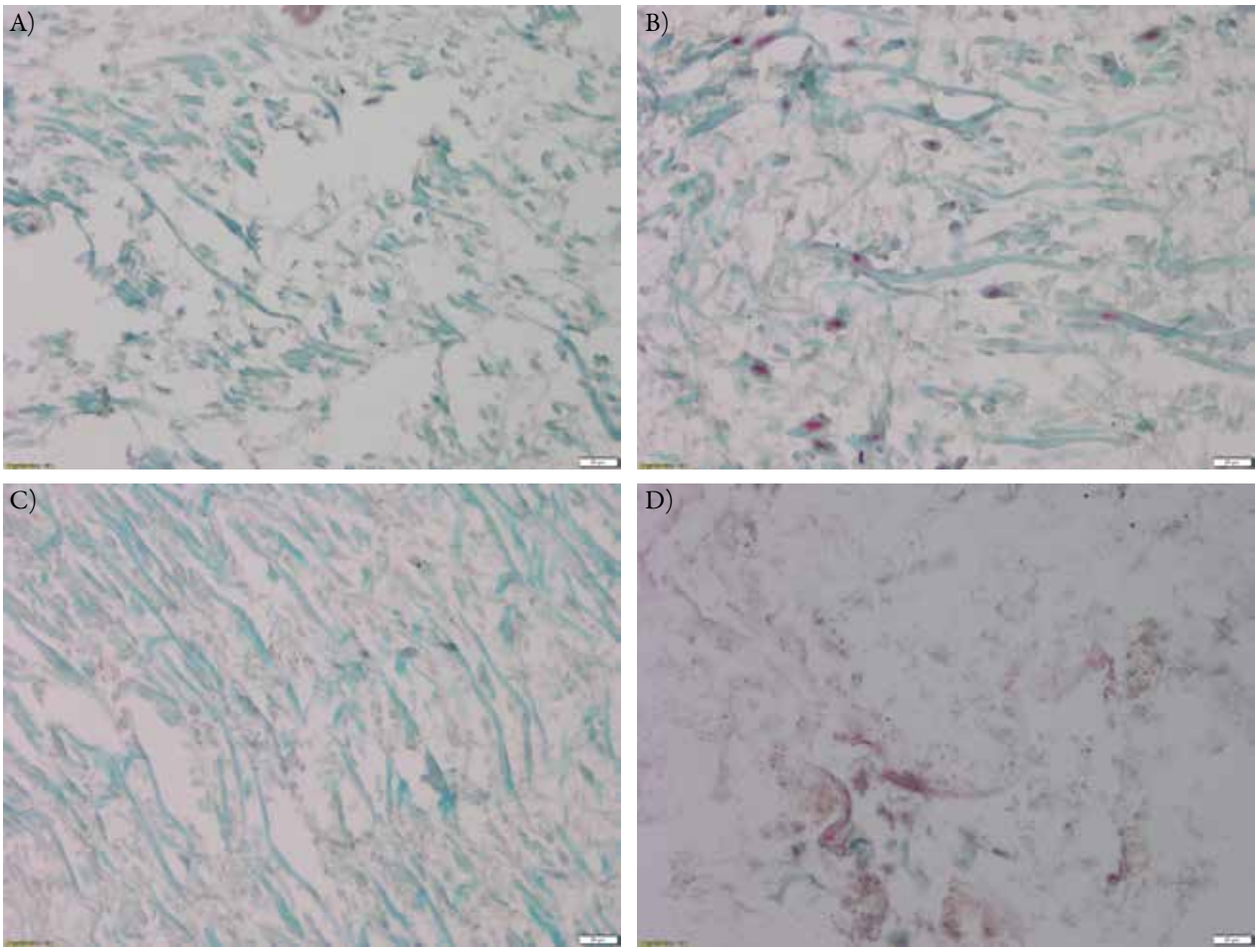

Figure 1. Picro Gomori stain of the body wall of *Holothuria scabra* treated at different salting times using the dry coarse "A" solar salt. A) 24 h; B) 36 h; C) 48 h; and D) 72 h.

concentration decreased in the holothurian body wall, indicating that the salt leached valuable collagen from the body wall of *H. scabra*. Processing sea cucumbers into beche-de-mer requires continuous attention, and the processing time plays a key role in achieving a good-quality product of high value. Exposure at 48 h was observed to be an ideal time for salting sea cucumbers during processing, and it certainly adds weight to the dried product. Twenty-four hours and 36 h of salting exposure are considered to be ideal exposure times because the body wall is still water-logged and the drying process will be problematic because of the higher chances of spoilage as compared to 48 h exposure. Processors in Fiji could have faced this issue during processing and this could have been an ongoing issue with poor-quality and poor income for the communities involved in this trade.

Conclusion

This study focused on assessing the collagen composition of the body wall of *Holothuria scabra*. The study showed that the exposure of sea cucumbers during salting affects the collagen composition and essential nutritional value of the sea cucumbers. The findings revealed that a salting time of 48 h was sufficient during processing. Exceeding 48 h extensively leaches the essential nutrition from the body wall as was reported in the present study. Better processing techniques need to be developed to prevent post-harvest nutritional losses from beche-de-mer.

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