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The absorption efficiency of the suspension-feeding sea cucumber, *Cucumaria frondosa*, and its potential as an extractive integrated multi-trophic aquaculture (IMTA) species

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ABSTRACT

Finfish aquaculture commonly releases waste material in the form of excess feed and faeces, which can impact the surrounding environment, often through increased oxygen demand in the benthos as a result of a buildup of organic matter. Integrated multi-trophic aquaculture (IMTA) in the Bay of Fundy co-cultures extractive species such as mussels (Mytilus edulis) and kelps (Saccharina latissima) alongside of the fed finfish to partially mitigate the impacts associated with excess inorganic and organic nutrients. The orange-footed sea cucumber (*Cucumaria frondosa*) is being examined as a potential extractive species to remove additional particulate organic waste in some of the larger particle size categories. Sea cucumbers were exposed to natural (IMTA sites and natural seston) particles and enhanced laboratory diets where the organic content (OC) of the food and faeces were determined to estimate absorption efficiency (AE). AE ranged between 68 and 85% for all the experimental trials but averaged $70 \pm 3\%$ when evaluating their response to only the natural diets. Sea cucumbers were capable of consuming aquaculture waste material when exposed to it in the laboratory and when deployed at an IMTA site, feeding directly upon the particulates released. There was a strong positive relationship ($R^2 = 0.82$) between food and faeces OC, making it possible to predict the faecal OC from the food supply OC. AE was not as readily predictable from the food supply OC although there was a significant positive relationship between food OC and AE. Sea cucumbers are efficient in absorbing organic material $(70 \pm 3\%)$ within the range (>30 and <50% OC) they are typically exposed to in their natural environment. When challenged with particulate material of higher organic content (>60% OC), such as cultured microalgae or salmon food and faeces they exhibit equal or enhanced (>80%) AE's. Our results show that C. frondosa has a great deal of potential to become an effective organic extractive IMTA species and aid in the reduction of organic loading occurring at aquaculture sites.

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1. Introduction

In marine aquaculture, excess food and faeces can lead to organic loading on site beneath the cages (Buschmann et al., 2008; Troell et al., 2003). This can greatly impact the chemical and biological oxygen demand of the substrate leading to a decline of some oxygen-sensitive species and the increase in the abundance of opportunistic species (Buschmann et al., 2008; Edgar et al., 2010; Troell et al., 2003). Integrated multi-trophic aquaculture (IMTA) is one technique that has the potential to help reduce some of the environmental impacts and has been steadily gaining momentum in Canada (Chopin et al., 2001; Ridler et al., 2007) and internationally (e.g. Troell et al., 2003). IMTA involves the culture of traditional finfish (e.g. salmon), but uses the waste products (excess feed and faeces) produced by the finfish as a food source for other commercially viable extractive species (MacDonald et al., 2011; Reid et al., 2010; Troell et al., 2003). Both inorganic extractive species (e.g. seaweeds) and organic extractive species (e.g. mussels) are grown in close proximity to the site to help absorb excess nutrients produced, while creating an additional cash crop for farmers (Chopin et al., 2001; Troell et al., 2003). This multispecies approach appears to be working, in that it has successfully created an additional cash crop, although there still appears to be some controversy on the demonstration of the mitigation of aquaculture wastes (Navarrete-Mier et al., 2010; Reid et al., 2010). These challenges may be due to the fact that some of the feeding niches for the larger organic particles are currently not being filled. As a result, there is a growing interest in increasing the variety of species used for extraction, with the commercially important orange-footed sea cucumber (Cucumaria frondosa) being considered as an additional organic extractive species in open-water IMTA systems.

Some species of sea cucumbers have been observed consuming and reducing aquaculture wastes, such as the California sea cucumber,



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Parastichopus californicus, which has been found to reduce both oyster waste (Paltzat et al., 2008) and salmon pen net fouling (Ahlgren, 1998). Australostichopus (Stichopus) mollis can effectively consume mussel aquaculture waste, leading to an increase in biomass and a faster growth rate for *A. mollis* (Slater and Carton, 2007). However, only deposit-feeding sea cucumbers, which feed by ingesting sediment (Lopez and Levinton, 1987), have been used in aquaculture. Suspension-feeding sea cucumbers, such as *C. frondosa*, which feed on particles suspended within the water column, have not yet been assessed for their potential value in aquaculture systems. *C. frondosa* is already a commercially important species with an established market as a fishery commodity referred to as "beche-de-mer" or "trepang" which is sold to Asia and some European countries (Ke et al., 1987).

C. frondosa is a benthic dendrochirotic echinoderm that primarily inhabits the sub-tidal zone (Singh et al., 1998). It is one of the most abundant and wide spread species of holothurians within the North Atlantic Ocean and the Barents Sea (Russia) (Gudimova et al., 2004). Its geographic range includes the coasts of New England, eastern Canada, southern Iceland, Greenland, northern Europe and Scandinavia to the Faroe Islands (Hyman, 1955; Jordan, 1972; Klugh, 1923; Singh et al., 1998). It is the largest sea cucumber along the eastern coast of North America (Hyman, 1955; Jordan, 1972; Ke et al., 1987) and is known to cover vast areas of the rocky substrate at depths of less than 30 m (Jordan, 1972; Singh et al., 1998), but can be found at depths up to 300 m (Hamel and Mercier, 1999).

C. frondosa is a passive suspension feeder that utilises its ten tentacles to capture particles within the water column and is known to feed mainly during the spring and summer when food appears to be more available (Hamel and Mercier, 1998; Singh et al., 1999). C. frondosa is also known to be capable of adjusting its food consumption rate, feeding faster when food concentrations are higher (Singh et al., 1998). Additional studies that investigated seasonal and tidal feeding activity of C. frondosa, confirmed that feeding increased as the quality (chloropigment concentration) of the suspended particulate material increased (Singh et al., 1999). Tentacle insertion rate (TIR) has been determined to be a good indicator of feeding activity and ingestion, and flow rates have been shown to have a significant effect on the feeding activity of C. frondosa within the natural environment (Holtz and MacDonald, 2009; Singh et al., 1999). Beyond this however, very little is known. Absorption efficiency describes the efficiency at which an animal absorbs organic material as it is transferred through the gut (Calow and Fletcher, 1972; Conover, 1966; Penry, 1998; Wang and Fisher, 1999). It was originally termed assimilation efficiency (Conover, 1966) but was later amended to absorption efficiency as it measures the proportion of organic material absorbed from the food within the gut, rather than the incorporation of the material into the animal's tissue (Calow and Fletcher, 1972; Penry, 1998; Wang and Fisher, 1999). While there is a little information on the feeding activities of C. frondosa, there is nothing known about its absorption efficiency and how it is likely to vary under different environmental conditions. Absorption efficiency is an important variable to determine within the IMTA infrastructure in order to quantify the overall reduction in quality (organic content) of the suspended particulate waste material as it is converted and re-deposited by extractive species. With this information it will be possible to estimate the potential for reduction in organic loading by sea cucumbers occurring at IMTA sites compared to that of traditional aquaculture operations.

The purpose of this study is to determine the potential of the sea cucumber, *C. frondosa*, as an effective IMTA organic extractive species. Specifically the objectives are to: (1) quantify the absorption efficiency of *C. frondosa*; (2) verify that *C. frondosa* is capable of consuming aquaculture waste; and (3) to determine if the absorption efficiency of *C. frondosa* can be predicted based on the quality (organic content) of the material available to feed upon.

2. Methods

2.1. Species collection

All sampling and species collection took place within the Bay of Fundy, South-west New Brunswick, Canada from June 16th –July 4th, 2010 and June 16th –September 4th, 2011. Adult sea cucumbers, *C. frondosa*, of a similar size were collected by divers (depth of 12–18 m) at Tongue Shoal (45°03′47″ N, 67°00′47″ W) and held for 1–2 weeks in wire mesh cages suspended off a raft (depth of 8–10 m) at the St. Andrews Biological Station in New Brunswick.

2.2. Laboratory trials

Sea cucumbers were housed at the Huntsman Marine Science Centre (St. Andrews, New Brunswick) in an Aquatic Habitats Benchtop (AHAB) system (Aquatic Habitats[™] Inc., Florida, USA; Model No. B25C-1; Fig. 1). The AHAB system was equipped with ten 10 L tanks (one sea cucumber per tank) with a screen covering each outflow to keep faeces within the individual tank. The AHAB system was run as a recirculation system using ambient seawater (8–11 °C). Flow rates were sufficient to ensure food did not settle out in the experimental chambers, but remained in suspension. Partial water changes were performed following any water sampling activity to replace the volume of water removed from the system. Sea cucumbers (n=10)were transferred to the laboratory and placed in the AHAB system for a period of 24-48 h, prior to experimentation, to acclimate and to void any faeces produced when feeding in the natural environment. Following acclimation, different groups of sea cucumbers (n = 10 per group) were exposed to one of the three diet treatments: (1) a commercial algae diet (Instant Algae®, Reed Mariculture, Campbell, CA, USA) of either T-iso (*Isochrysis galbana*) or Shellfish Diet® which is made up of a mix of 30% Isochrysis, 20% Pavlova, 20% Tetraselmis, and 30% Thalassiosire weissflogii; (2) a modified algae diet consisting of a mixture of diatomaceous earth (Agrogreen® Canada, Ontario) and T-iso used to create a diet of reduced organic content compared to regular algae; or (3) a mixture of particles found at local salmon farms. The mixture of farm particles consisted of ground salmon feed (Skretting® Optiline Microbalance Winter Extruded salmon feed) and dried and ground faeces collected from salmon (Salmo salar) held in a tank and feeding upon this diet. The concentrated mixture of feed and faeces was filtered through a 100 µm sieve and added directly to the AHAB system to distribute food evenly to all individual sea cucumbers. All diet mixtures were added to seawater that was filtered to less than 5 µm, to achieve a final concentration of $20-30 \times 10^3$ cells ml⁻¹. Particle concentrations were verified and monitored throughout the experiments using a Coulter Counter® Multisizer II.

The organic content (OC) of the algal diets (T-iso and Shellfish Diet) was measured by filtering water samples from the AHAB system, however the weight of the algal diets was too low ($<1 \text{ mg L}^{-1}$) at the diet concentrations to reliably estimate food OC (Table 1). The food OC of the algal diets was instead measured by pipetting a small volume (<2 mL) of the concentrated commercial algae solution onto a pre-weighed filter to determine weight after drying and ashing. Water samples (2 L) for the modified algae diet (n=9) and mixture of farm particles (n=5) were taken 20 min after the respective diet mixtures were added to the reservoir of the AHAB system to facilitate uniform distribution of the particles. The amount of diatomaceous earth added to the modified algae diet varied over a 3 day period (June 27-29, 2011) to obtain a range of 40-50% OC, so water samples were taken daily (n=3) and averaged for the exposure period. All diet samples were processed at the St. Andrews Biological Station immediately after collection to determine food OC (Table 1).

Any faeces produced by individuals in the 48–72 h following the addition of either of the 3 diets were collected with a plastic bulb

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