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Editor

Igor Eeckhaut
Marine Biology
6, Av. Champ de Mars
University of Mons-Hainaut
7000 Mons Belgium
Email: Igor.Eeckhaut@umh.ac.be

Production

Information Section,
Marine Resources Division
SPC, BP D5, 98848 Noumea Cedex
New Caledonia.
Fax: +687 263818
Email: cfpinfo@spc.int
www.spc.int/coastfish

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Editorial

New information about sea cucumber biodiversity, ecology and aquaculture is featured in this issue. Reports on research and projects come from various countries, including Fiji, Iran, Oman, Sri Lanka, Madagascar, Philippines and Indonesia. We also present some insights about Mediterranean sea cucumber phylogeny, and a summary of all known saponins.

The first paper is by Cathy Hair et al. (p. 3), who present results from an ACIAR-funded sandfish culture and sea ranching mini-project in Fiji. The main aims of the project were to transfer sandfish hatchery technology to local government and private hatcheries, increase juvenile production, and conduct sea ranching trials with a local coastal community.

Hamilton and Lokani (p. 12) studied sea cucumber populations in the Autonomous Region of Bougainville. They compared results of surveys made in the same areas in 1992 and 2008, which show that the abundance of eight species of sea cucumbers dramatically declined over this 16-year period.

Tsiresy et al. (p. 17) present their results about the growth of *Holothuria scabra* in micro-farms in Madagascar. The authors found that the growth and survival rates are different in three surveyed villages, and discuss the reasons for these differences.

Olavides et al. also (p. 23) worked on *Holothuria scabra* but in Bolinao and Anda in the Philippines. The authors inform us about simultaneous mass spawning in sea ranching sites.

Claereboudt and Al-Rashdi (p. 25) evaluated for the first time, the diversity and abundance of holothuroids along the coast of Oman. One of the most striking results is the large difference in sea cucumber community structure between the Sea of Oman and the Arabian Sea.

Taquet et al. (p. 30) and Dabbagh et al. (p. 35) worked on *Holothuria leucospilota*. Taquet et al. report here about their observations of a large group of *H. leucospilota* juveniles in Manado (north Sulawesi, Indonesia). Dabbagh et al. describe how they successfully induced spawning of *H. leucospilota* individuals in Iran. The authors obtained juveniles after a month of rearing and the survival rate was close to 5%.

The abundance, distribution and some biological aspects of *Holothuria edulis* on the northwest coast of Sri Lanka is the subject of a paper by Dissanayake and Athukorala (p. 39). The findings of this study could be used to develop proper management plans for the sustainable exploitation of this resource in the future.

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| Some insights on the phylogeny of Algerian shallow-water sea cucumber species (Holothuroidea: Aspidochirotida) <i>K. Mezali</i> | p. 45 |
| Review of saponin diversity in sea cucumbers belonging to the family Holothuriidae <i>G. Caulier et al.</i> | p. 48 |
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Mezali (p. 45) gives some insights on the phylogeny of Algerian shallow-water sea cucumber species. The most striking result is certainly the fact that *Holothuria tubulosa*, the most common species and the “best known” species in the Mediterranean Sea, forms a clade with two well-separated populations.

Finally, Caulier et al. (p. 48) reviewed saponin diversity in Holothuriidae. No less than 59 triterpene glycosides are reported in the literature.

We also present at the end of this issue several interesting communications, including a report by Cathy Hair et al. on the use of an algal concentrate as potential substitute for the food usually used for the sandfish culture (p. 60) and a report by Sharon Ng on a recent workshop on the “Feasibility of sea cucumber and abalone farming as an alternative livelihood in Semporna, Sabah”.

As usual, we conclude the issue with some information concerning sea cucumber spawning observations, and a list of abstracts from recent sea cucumber-related publications.

Igor Eeckhaut

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Secretariat of the Pacific Community, Marine Resources Division, Information Section
BP D5, 98848 Noumea Cedex, New Caledonia
Telephone: +687 262000; Fax: +687 263818; cfpinfo@spc.int; <http://www.spc.int/coastfish>

Sandfish culture in Fiji Islands

Cathy Hair,¹ Tim Pickering,² Semisi Meo,³ Tavenisa Vereivalu,⁴ Justin Hunter,⁵ Laisiasa Cavakiqali⁶

Introduction

Reports of decreasing stocks of valuable tropical sea cucumbers worldwide are all too familiar. Throughout the Pacific, as elsewhere, the well-known story describes dwindling numbers, smaller individuals and increasing reliance on exploiting low value species. Management has essentially failed for a host of reasons, with some Pacific Island countries and territories (PICTs) resorting to the extreme measure of fishing moratoria to encourage stock recovery (Kinch et al. 2008; Nash and Ramofafia 2006; Purcell 2010).

Sandfish (*Holothuria scabra*) is a high value tropical sea cucumber species and is regarded as vulnerable to overfishing because of its inshore, mostly shallow, habitat. Hatchery technology for this species has been established in a number of countries in the Asia-Pacific region, including Solomon Islands, New Caledonia, Vietnam and the Philippines, through research supported by the Australian Centre for International Agricultural Research (ACIAR) and the WorldFish Center. The spawning and larval rearing of sandfish up to small juvenile size (>3 g) is now regarded as relatively straightforward. Commercial profits from harvestable adults are, however, far from certain. Hatchery production is a costly intervention. Furthermore, large areas are needed for increasing juvenile production, and grow-out in ponds or the sea is necessary for producing commercial size individuals. ACIAR research on grow-out is also underway. Pond grow-out of juvenile sandfish to market size is being developed in Vietnam (Pitt and Duy 2004; Bell et al. 2007) and sea ranching trials are being conducted in the Philippines. Farther afield, sea-pen farming in Madagascar is showing promise (Eeckhaut et al. 2008; Robinson and Pascal 2009), and a private sector and state government collaboration is planning sea ranching trials in the Northern Territory of Australia.

Recent efforts in the Pacific region involve investigating the potential for sea ranching in Fiji, using locally managed fishing areas or *qoliqoli*. Sandfish are called *dairo* in Fiji and are a traditional food item (Fig. 1). This is in contrast to most PICTs, which export sea cucumbers but do not consume them. The exception is parts of Polynesia where the internal organs may be eaten and the animal left to regenerate (Kinch et al. 2008). Although sandfish is protected by fisheries legislation for domestic consumption in Fiji, there has been widespread export, particularly since the late 1980s. Concerns about decreases in sandfish abundance and size have led to initiatives to promote sea ranching as an income generating resource and also to help rejuvenate wild stocks. We use the term “sea ranching” for a “put, grow and take” operation where Fijian owners can harvest hatchery-produced sandfish grown out in their *qoliqoli* (Bell et al. 2008a). Fortunately, there are some locations where sandfish are still locally abundant, although large mature animals are not easy to find.

This article describes the results of an ACIAR-funded sandfish culture and sea ranching mini-project (see boxed text). The study ran for two years



Figure 1. Sandfish (*dairo*) prepared the Fijian way.

1. James Cook University, Townsville, Australia. Email: cathy.hair@jcu.edu.au
 2. Secretariat of the Pacific Community, Suva, Fiji Islands. Email: TimP@spc.int
 3. Fiji Locally Managed Marine Area Network, Suva, Fiji Islands
 4. Department of Fisheries, Suva, Fiji Islands
 5. J. Hunter Pearls, Savusavu, Fiji Islands
 6. University of the South Pacific, Suva, Fiji Islands

from May 2008 to April 2010 on Fiji's second largest island, Vanua Levu (Pickering and Hair 2008). The main aims of the project were to transfer sandfish hatchery technology to local government and private hatcheries staff, increase juvenile production, and run sea ranching trials with a local coastal community. Country partners included the Fiji Ministry of Primary Industries (Department of Fisheries), J. Hunter Pearls, Fiji Locally Managed Marine Area Network (FLMMA), University of the South Pacific and the Natuvu community of Vanua Levu.

The Australian Centre for International Agricultural Research project "Development of aquaculture-based livelihoods in the Pacific Islands region and tropical Australia" is managed by James Cook University (Townsville, Australia) in partnership with the Secretariat of the Pacific Community, WorldFish Center and University of the South Pacific. This project has initiated a number of aquaculture "mini-projects" throughout the Pacific Islands region dealing with various commodities. Mini-projects are small, targeted research interventions that address bottlenecks to sustainable aquaculture.

Project site

Before the project began, several locations on the south side of Vanua Levu (Caukdrove Province) were inspected for their suitability for sea ranching. Site selection criteria included matching the physical characteristics recommended by Purcell (2004) in addition to human factors such as community enthusiasm, ability to provide security for released sandfish juveniles, and agreement to allow study animals to reach commercial size before harvesting. The site also needed to be located within a few hours travel of the hatchery to ensure minimum transport stress on juveniles prior to release.

The Natuvu community of Wailevu District (population ~250) fulfilled all the requirements (Fig. 2).

Within their *qoliqoli* is an extensive sea grass bed, located immediately in front of their village. This habitat, measuring roughly 750 m long (parallel to shore) and 500 m wide, met the microhabitat release criteria (Purcell and Simutoga 2008; Purcell 2004). The main seagrass species was *Syringodium isoetifolium*, with smaller amounts of *Halodule uninervis* and *Halophila ovalis* closer inshore. The sediment was moderately soft with abundant invertebrate fauna (e.g. other sea cucumber species, sea urchins, sponges) as well as numerous small- to medium-sized sandfish (Fig. 3). No large rivers discharge close to the release site, although older community members recounted stories of freshwater pooling over the seagrass beds, resulting in mass mortality of sandfish during intense storm events (i.e. associated with a cyclone). We considered this potential risk one that would occur at any site and graded the site as "good" to "very good" regardless of flood risk. The Natuvu community's approval was genuine, with everyone showing great interest in the project and voluntarily ceasing harvesting sandfish before the project began in mid-2008. The village is accessible by road or sea from the hatchery.

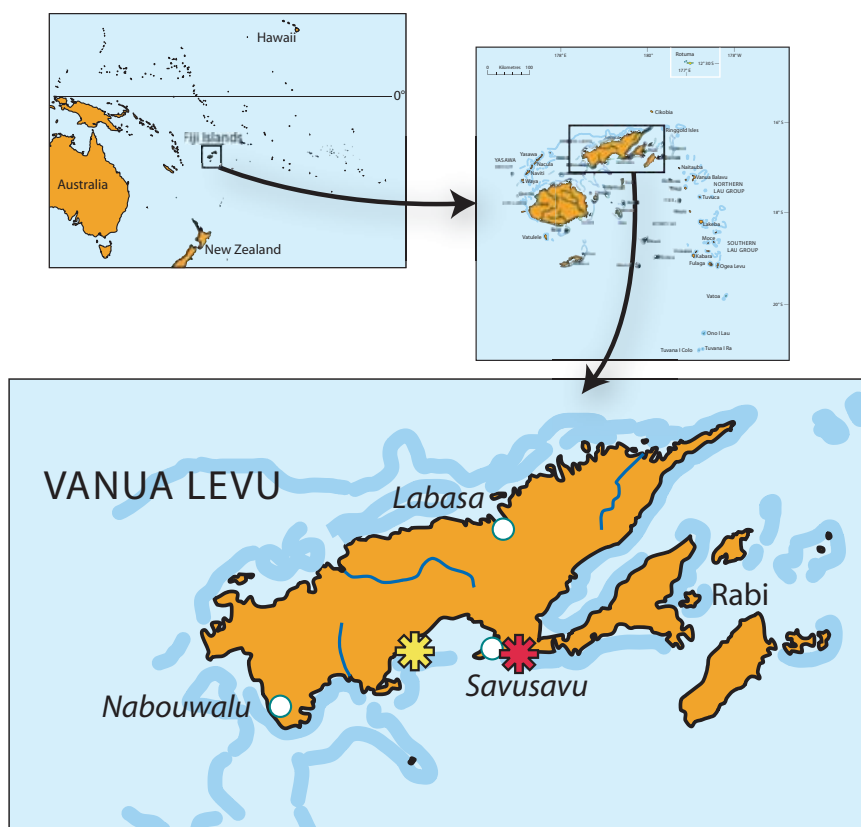


Figure 2. Location of Savusavu township, J. Hunter Pearls hatchery (red star) and Natuvu village (yellow star).



Figure 3. Microhabitat of the release site with small wild sandfish.

Broodstock, hatchery and larval rearing

Mature sandfish broodstock were sourced from within Savusavu Bay to ensure that genetic issues were addressed responsibly (i.e. hatchery juveniles were bred from natal stocks of the release area) (Purcell 2004). Between spawning attempts, a small pond in Savusavu was used to hold broodstock (Fig. 4). Holding groups of adults in earthen saltwater ponds before spawning can result in “conditioning” of sandfish. After conditioning, broodstock become easier to spawn. They may also spawn earlier (and their gonads develop to the same stage of maturity), improving spawning synchrony (Agudo 2006; Duy 2010). This can work well with old shrimp or in fish ponds where the substratum allows feeding and burying of sandfish. However, the 0.2 ha pond was constructed as part of a marina development and was never used for farming. The pond floor was quite rocky and lacked nutrient-rich sediment. Additionally, a broken tidal gate did not control water flow, so water levels rose and fell with the tide. The extensive diurnal tidal flushing did have the positive effect



Figure 4. The pond used for holding sandfish broodstock.

of preventing freshwater stratification during the wet season. Conversely, it also reduced pond productivity by preventing algal blooms. While broodstock survived in the pond, they did not grow and condition as expected. Added to the uncertainty of security, pond use was discontinued after one year.

Larval production was carried out at the J. Hunter Pearls’ blacklip pearl oyster hatchery (Fig. 5), about 15 km east of Savusavu town (see Fig. 2). Spawning induction and larval rearing procedures followed methods developed in New Caledonia by the WorldFish Center (Agudo 2006). Throughout the project, modifications were made to accommodate local conditions and facilities, and to adopt advances in hatchery techniques from Vietnam and the Philippines. The blacklip pearl oyster hatchery met all system requirements for sandfish culture. Microalgae food species were already in production, water quality was excellent and



Figure 5. J. Hunter blacklip pearl oyster hatchery (top). Note the seawater intake pipe near the trees at left of picture and white one tonne *dairo* larval tanks under the white roof. Hatchery manager, Sachin Deo (bottom).

only minor adaptations were needed to proceed with production.

Once the release site was selected and the hatchery prepared, the first training to transfer hatchery technology was held in late 2008. Private and government-sector technicians were trained in all aspects of sandfish culture, including broodstock collecting, spawning, larval rearing and transfer to juvenile grow-out areas (Fig. 6).



Figure 6. A Fiji Department of Fisheries senior aquaculture officer packing broodstock for transport to the hatchery.

In the course of the two-year project (May 2008 to April 2010) there were five attempts to produce sandfish juveniles: two in the summer season of 2008/2009, and three in the summer season of 2009/2010. Each involved multiple spawning inductions, and larvae were produced on each occasion. However, follow-through was variable and larvae were reared to the juvenile stage only from one spawning event in late 2008. On this occasion, about 1,500 small juveniles were transferred into diatom-conditioned raceways, and 500 of these survived to 1–10 g size to be used in sea ranching trials. The failure to produce juveniles in subsequent production runs was due to a combination of human error, unfavourable environmental conditions, and collateral damage caused by two tropical cyclones.

Community sea ranching trial

Because of the small number of available juveniles, the trial was run at an experimental scale in order to gain information to increase the success of

future releases. A University of the South Pacific post-graduate student was responsible for monitoring the trial, which comprised four 100 m² sea pens set up in the Natuvu seagrass meadow. The pens were constructed far enough from shore so that they would not dry out at low tide and close enough to be easily reached by foot. Two pens were each stocked with “large” (3–10 g) and “small” juveniles (between 1 g and 3 g). This design was driven primarily by the low number of available juveniles greater than 3 g — the minimum release size recommended by Purcell and Simutoga (2008) for good post-release survival. In addition, we were releasing juveniles into a very different habitat from other studies, thereby providing an opportunity to test this result under Fijian conditions.

Village involvement was strong, and building and deploying pens was a community affair (Fig. 7). The release of juveniles generated much local interest and was the occasion of a feast attended by many local VIPs (Fig. 8). The release of juveniles into the seagrass bed at Natuvu was carried out according to the methods recommended by the WorldFish Center, based on studies carried out in New Caledonia (Purcell and Eeckhaut 2005; Purcell and Simutoga 2008; Purcell et al. 2006). Juveniles were taken to the site the previous day and left in nets overnight to acclimate to

local conditions and to “de-stress” following transport. In May 2009, 496 juveniles were released: 105 placed into each of the two large size pens (Pens A and C) and 143 into each of the small size pens (Pens B and D). They were removed from the overnight holding nets (Fig. 9), then “planted” in the sediment within the sea pens by digging a small trench with a finger, then placing them gently in it.



Figure 7. Pen construction on the seagrass beds in front of Natuvu Village.



Figure 8. Project staff, community members and VIPs at the occasion of the sandfish release.



Figure 9. Preparing hatchery-reared juveniles for release into the sea at Natuvu in May 2009.

Monitoring began one month after the release in June 2009, and then at approximately one to two-monthly intervals up until the conclusion of the study in April 2010. Project staff were responsible for monitoring, and were assisted by four Natuvuan men who were trained as “*dairo* wardens”. These men helped the scientific staff with monitoring the released sandfish juveniles, and they checked pens regularly (Fig. 10). They were also a valuable source of knowledge regarding local conditions and their *qoliqoli* environment and fauna. During each monitoring session, the number of animals in each pen was counted and their length and width measured. The length and width data were then used to calculate weight using a formula developed by Purcell and Simutoga (2008). On two occasions, weight was checked with an electronic balance that confirmed that the weight calculated from measurements was reasonably accurate.



Figure 10. Fisheries officer collecting *dairo* (top left); Natuvu *dairo* wardens measuring ranched *dairo* 3 months after release (top right); Natuvu *dairo* wardens monitoring ranched *dairo* 11 months after release (bottom left); village chief inspecting *dairo* 11 months after release (bottom right).

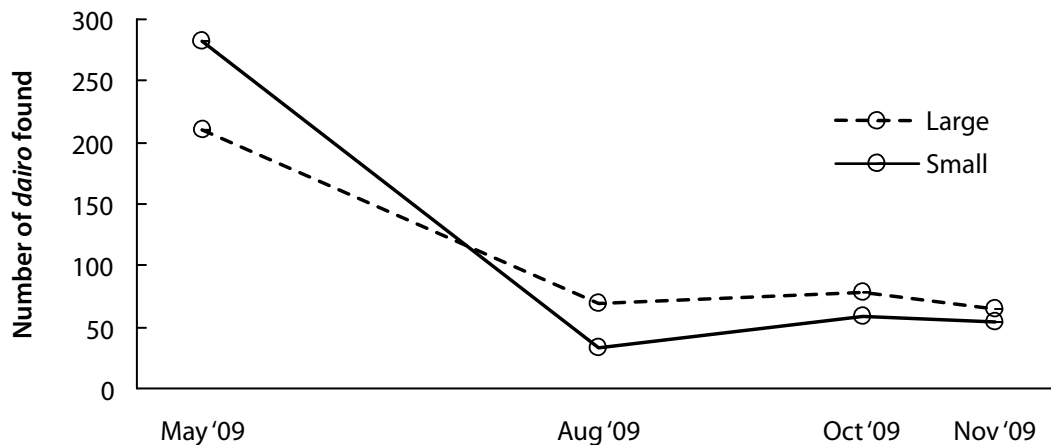


Figure 11. Sandfish survival in the four pens after six months.

The survival rate after six months was around 28% overall (23% for small sandfish and 33% for large sandfish) (Fig. 11). The highest overall survival rate was 41%, recorded from a pen of large sandfish. Mortality (or loss) was greatest in the first three months and then levelled off. Due to pen damage in the latter half of 2009, the six-month average is used as the survival estimate for the trial. Studies by Purcell and Simutoga (2008) also found that most mortality occurs in the first few months after release.

Growth of hatchery-produced sandfish was generally positive throughout the trial, although a period of slow growth occurred between November 2009 and January 2010 (Fig. 12). Measurements are considered most reliable up until nine months after release (February 2010), prior to Cyclone Tomas in March 2010. At this time, average sandfish size was 165 ± 5 g for small sandfish and 167 ± 6 g for large

sandfish. Sandfish were measured after this time (see April 2010 data point in Fig. 12). However, it is possible that some cultured sandfish escaped and wild sandfish entered the damaged pens; therefore, results are unreliable. A data logger placed in the sea-pen area recorded sea temperatures from June 2010 (a month after release) to January 2011.

Potential feasibility for the Pacific

The results to date are promising but due to the small number of animals released, the results may not accurately predict outcomes of a larger-scale release (Purcell and Simutoga 2008; Bell et al. 2008a). The successful collaboration of a private sector business, government fisheries agency, non-governmental organisation (NGO), an educational institution and a community was critical to the outcomes of this project. Experience and lessons learned will be

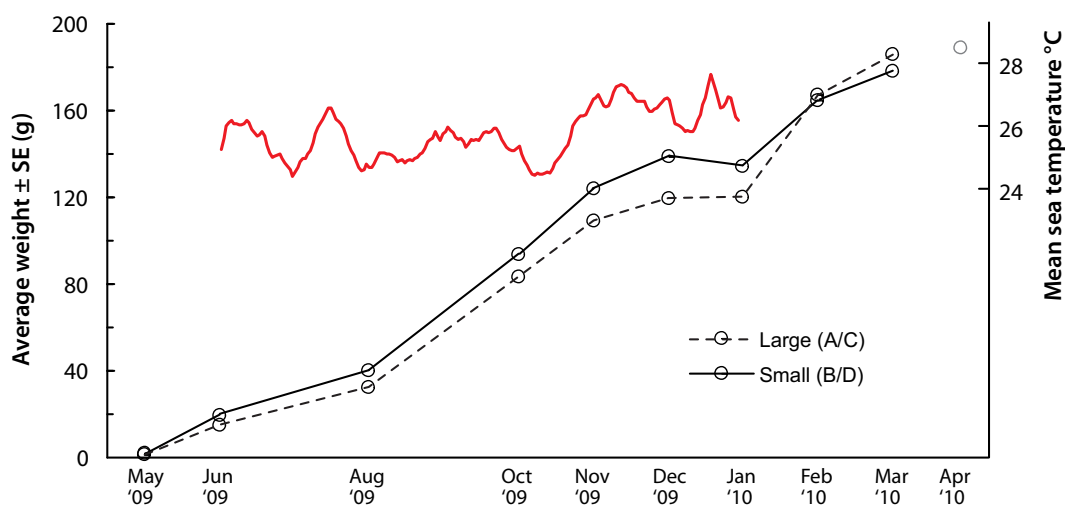


Figure 12. Eleven months of growth data for hatchery-produced sandfish (in the sea) at Natuvu (large sandfish from pens A and C combined, and small sandfish from pens B and D combined). March and April 2010 are sub-samples of the sandfish recorded after Cyclone Tomas and data are to be treated with caution. Red line shows average sea temperature in the sea pens between June 2009 and January 2010.

applied in follow-on work to produce more juveniles and conduct a large-scale release. We hope to be able to expand these results to produce real figures on the feasibility of the activity in Fiji and other PICTs.

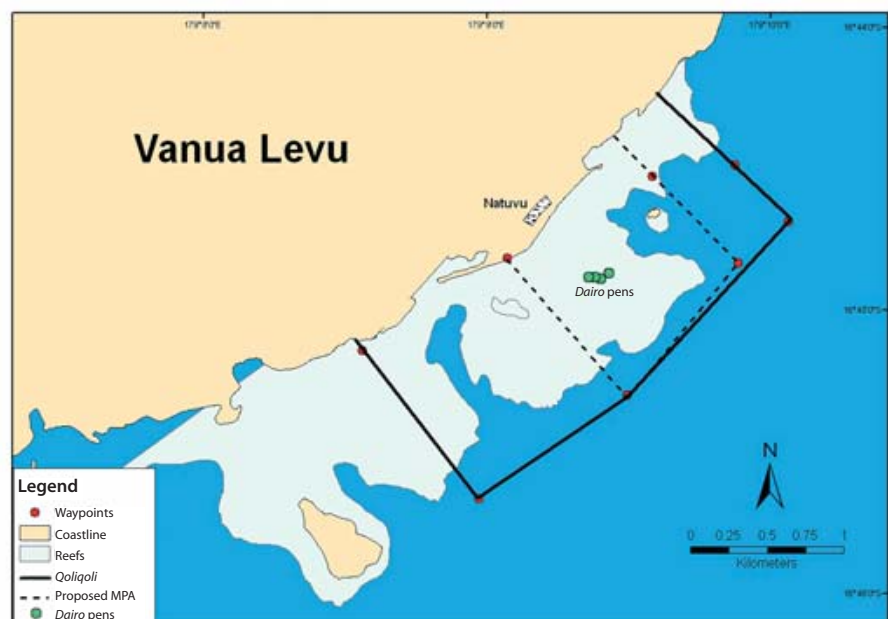
Meanwhile, most PICTs have expressed concern regarding overfishing (Kinch et al. 2008) and hatchery intervention is seen as a way to ameliorate this situation. Bell et al. (2008a) define and discuss the various ways that hatchery-produced sea cucumber juveniles can be used to help restore fisheries: “Restocking” is the release of sandfish into a restricted area and protecting them as a future spawning population; “stock enhancement” is the release of cultured juveniles into wild populations to augment the natural supply of juveniles and boost harvests; “sea ranching” is the release of cultured juveniles into unenclosed marine environments for harvest at a larger size in “put, grow and take” operations (where released animals are not expected to contribute to future spawning biomass). The expected benefits from sea ranching will be compromised if effective resource management is not in place and the economic return to investors (i.e. the community) does not cover production costs. As yet, we do not know the break-even cost for hatchery production of sandfish in PICTs. However, as pointed out by SPC (2009), hatchery intervention is not the only way to restore depleted fisheries or produce more sea cucumbers (Bell et al 2008b; Friedman et al. 2008). There should be a proper investigation into the best and most cost-effective options for each PICT. Ideally, releases of cultured juveniles should add value to other forms of management (Bell et al. 2008a; Purcell 2010).

Nonetheless, restocking and sea ranching are an option for PICTs, and perhaps one of the best ways

to apply the technology is through the release of juveniles into protected areas. There is a history of restocking in some PICTs, where clams have been restocked in marine protected areas or MPAs (these zones may be known by other names, such as special management areas in Tonga, and *tabu* or no-take zones in Fiji). The success of these zones in replenishing overfished populations varies (see references in Tisdell 1992; Gillett 2009). Fisheries agencies and NGOs (e.g. FLMMA) have encouraged communities to set aside no-take fishing areas in order to reduce fishing pressure and conserve marine resources. No-take zones have proven to be an acceptable management measure in the region. Benefits to stakeholders can result from “spillover” of animals from no-take zones into nearby areas that are open to fishing, and increased larval supply from protected broodstock. This second factor is likely to be particularly effective for sandfish by providing a refuge for a largely sedentary animal and promoting maintenance of a large (hence more effective) spawning biomass to increase recruitment outside the MPA. The approach was successful in the Philippines for a sea urchin species (Juinio-Meñez et al. 2008) and a network of MPAs was recommended to enhance larval exchange. Sea ranching, on the other hand, is a popular option due to the immediate need for income-generating activities, particularly in places where stocks have been depleted to a level where a vital livelihood has been lost.

Prior to the start of this study, the Natuvuan chief and community placed a temporary ban on harvesting sandfish throughout their *qoliqoli*. Later, part of the *qoliqoli* was officially declared a no-take zone (Fig. 13), a move supported and ratified by Fiji Department of Fisheries. It is interesting to note

Figure 13. Natuvu village, showing the extent of the *qoliqoli* (solid line) and the MPA within it (broken line). Green dots within the MPA denote sandfish sea pens.



that sandfish broodstock collected from the Natuvu *qoliqoli* showed an increase in average weight during the project — from 320 g in November 2008 to 450 g in March 2010 — possibly due to these fishing restrictions. According to the village chief, there were also noticeably more and larger sandfish and other sea cucumber species in the MPA area (Fig 14). Spawning of hatchery-produced sandfish was observed in November 2009 and March 2010 (Fig. 14), suggesting that ranched sandfish may contribute to future stock biomass, although there is yet no evidence of successful fertilisation and larval dispersal. Locals report that other valuable sea cucumber species have increased in number and size within the MPA (Fig. 14).

There is limited enforcement capacity in many PICTs but customary marine tenure enables communities to exercise some form of control and sustainable management in their areas of jurisdiction. Local ownership can promote good management of the fishery; community surveillance can reduce poaching and overfishing, allow sandfish to reach

A-grade size before harvest and enforce the no-take areas.

Finally, if investment is made in sandfish sea ranching, it is imperative to improve processing and marketing in order to obtain maximum income for the final product, beche-de-mer (Ram et al. 2010; Purcell 2010). This will provide the most benefit at the grassroots level to resource owners and will also offset the costs of juvenile production. The viability of this project rests on rewarding resource owners for the hardships involved in policing their *qoliqoli*, maintaining no-take zones, or delaying harvest until the sandfish reach a large size. If this benefit can be realised, it bodes well for the future of sea cucumber culture in the Pacific.

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Figure 14. Signs of a healthy ecosystem within the Natuvu MPA: a hatchery-produced sandfish spawning inside a sea pen in March 2010 (top left), size range of sandfish (bottom left) and commercial-sized curryfish (*Stichopus herrmanni*) (right).

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Severely overfished sea cucumbers in the Autonomous Region of Bougainville

Richard Hamilton¹ and Paul Lokani²

Abstract

In this paper we compare changes in populations of sea cucumbers that were surveyed on reef flats around Buka Island, in the Autonomous Region of Bougainville (ARB) in 1992 and 2008. Over this 16-year period, the abundances of eight species of sea cucumber declined dramatically. By 2008, six of the eight species sighted in 1992 and 2008 — *Actinopyga echinites*, *A. miliaris*, *Bohadschia similis*, *Holothuria atra*, *H. coluber* and *H. scabra* — had declined to between 1% and 5% of their 1992 abundances, providing quantitative evidence that the sea cucumber fishery in the ARB is severely overfished. In 1992, *H. scabra*, *H. atra* and *B. similis* constituted 92% of the sea cucumbers sighted on reef flats. These species were also the three most abundant sea cucumbers in the 2008 survey, comprising 62% of all sea cucumbers seen. Relatively high abundances of *Actinopyga mauritiana* and *Bohadschia vitiensis* were also sighted in 2008, species that were not encountered in the 1992 survey. Our findings provide support for the national moratorium that was placed on the sea cucumber fishery in Papua New Guinea in 2009, and we anticipate that many years of complete closure will be required before stocks in the ARB recover to 1992 levels.

Introduction

Sea cucumbers, or their dried form (beche-de-mer), are either eaten or used for medicinal purposes, and they have been a highly sought-after commodity in Asia for centuries (Toral-Granda et al. 2008). In the Pacific, most commercially valuable sea cucumbers occupy shallow clear seas, making the harvesting of these sessile and often conspicuous animals a relatively simple procedure. The combination of high value plus ease of capture has meant that many sea cucumber stocks around the world are now heavily overfished (Toral-Granda et al. 2008; Friedman et al. 2010). In Melanesia, sea cucumbers provide a very important source of income to rural coastal communities, being one of the few commodities that can be captured, processed locally, and then stored for extended periods of time (Kinch et al. 2008a).

In shallow habitats, sea cucumbers are typically gleaned from reef flats and the reef crest on a low tide; in deeper areas, free divers use masks and fins to pick up sea cucumbers. In water that is too deep to easily access (typically below 20 m), free divers often use “bombs” — a small harpoon or series of straitened hooks inserted into a lead weight — to harpoon sea cucumbers that are beyond their reach. In Melanesia, the sea cucumber fishery is a multi-species fishery. In Papua New Guinea (PNG) for example, at least 26 species of sea cucumbers are

harvested (Kinch et al. 2008b). Like other regions in the western Pacific, many sea cucumber fisheries in Melanesia have exhibited boom and bust cycles. Initially, sea cucumber fisheries target only one or two high value species, but once these stocks are over fished the fishery typically shifts its focus to lower value species (e.g. Lokani 1990; Lokani et al. 1996). Stocks of high value species are often quickly depleted, as evidenced by a case study on the Carteret Islands that are northeast of Bougainville Island. In 1982, a beche-de-mer fishery targeting white and black teatfish (*Holothuria fuscogilva* and *H. whitmaei*) harvested approximately 10 t of sea cucumber from the Carteret Islands, but by 1983, total production had fallen to just over 2,000 kg and large, high grade beche-de-mer was virtually eliminated from the fishery (Dalzell 1990).

Although sea cucumber fisheries typically follow boom and bust cycles, today, their high value, a lack of alternative income opportunities, a wide network of beche-de-mer purchasers, and the ease of processing and storing means that artisanal fisheries can continue to place considerable pressure on these fisheries well after stocks have been overexploited (Friedman et al. 2010). Several recent surveys in Melanesia have shown that sea cucumber stocks are widely overfished (e.g. Friedman et al. 2008; PNG National Fisheries Authority 2007; Ramofafia 2004; Ramohia 2006).

1. The Nature Conservancy, Indo-Pacific Resource Centre, 51 Edmondstone Street, South Brisbane, QLD 4101, Australia.
Email: rhamilton@tnc.org

2. The Nature Conservancy, Papua New Guinea Office, Suite 7, Monian Haus, Nita Street, Tabari Place, Boroko.
Email: plokani@tnc.org

For many locations, however, a lack of historical scientific data (Friedman et al. 2010), coupled with a shifting baseline syndrome — long-term and usually negative changes that are often not immediately or readily apparent to new generations of fishers or scientists working in a data-poor area (Pauly 1995) — make it difficult for many managers to comprehend the scale of changes that have occurred in sea cucumber fisheries in recent decades.

In this paper we present data on the abundances of sea cucumbers on reef flats around Buka Island, ARB in 1992 and 2008. The scale of decline is dramatic, and provides a quantitative example of the magnitude of change that has occurred in this artisanal fishery over a 16-year time frame. It is hoped that this information will provide support for the national moratorium currently placed on all PNG sea cucumber fisheries, as well as providing ARB fisheries managers with targets to aim for (i.e. 1992 levels) when rebuilding their sea cucumber fishery.

Methods

In 1992, one of the authors on this paper (PL) surveyed sea cucumber abundances on three reef flat sites located on outer reefs on the western side of Buka Island (Lokani, unpublished data 1992). In 2008, we resurveyed sea cucumber abundances at nine reef flat sites within the same region (Fig. 1).

These nine sites were resurveyed as part of a larger marine assessment investigating the status of sea cucumbers, reef fishes and coral cover in the northern region of the ARB (Hamilton et al. 2010). In 1992, four 600 m² transects were sampled at each site. In 2008, ten 400 m² transects were sampled at each site. Both surveys were done on foot and transect lengths were measured by laying down a 50-m measuring tape across the reef flat prior to conducting the survey. Transect width was measured by pulling an 8-metre-long rope between two observers, with observers keeping the midpoint of the rope on the 50-m transect line. In each pass of a transect, the number of individual sea cucumber species was recorded onto underwater paper. Sea cucumber abundance estimates per transect were converted to the number of individuals per hectare (ind. ha⁻¹), and the mean abundances by species and year were graphed in SigmaPlot. Mann-Whitney Rank Sum Tests were used to compare differences in the mean abundances of sea cucumbers sighted in 1992 and 2008 because the data failed the assumption of normality.

Results

Species composition

In 1992, 3109 sea cucumbers representing eight commercial species (*Actinopyga echinites*, *A. miliaris*, *Bohadschia argus*, *B. similis*, *Holothuria atra*, *H. coluber*, *H. scabra*, *Stichopus herrmanni*) were counted on 12 transects. In 2008, 740 sea cucumbers representing 11 commercial species (*A. echinites*, *A. mauritiana*, *A. miliaris*, *B. argus*, *B. similis*, *B. vitiensis*, *H. atra*, *H. coluber*, *H. whitmaei*, *H. scabra*, *S. herrmanni*) were counted on 90 transects. The relative abundance of each sea cucumber species recorded on reef flats in 1992 and 2008 is shown in Table 1. In both surveys the three most abundant species were *H. atra*, *H. scabra*, and *B. similis*. In 1992, these three species made up 92% of the sea cucumbers seen, while in 2008, this trio made up 62% of all sea cucumbers seen. In 2008, *A. mauritiana* and *B. vitiensis* comprised 31% of the sea cucumbers sighted; these species were not recorded in the 1992 survey.

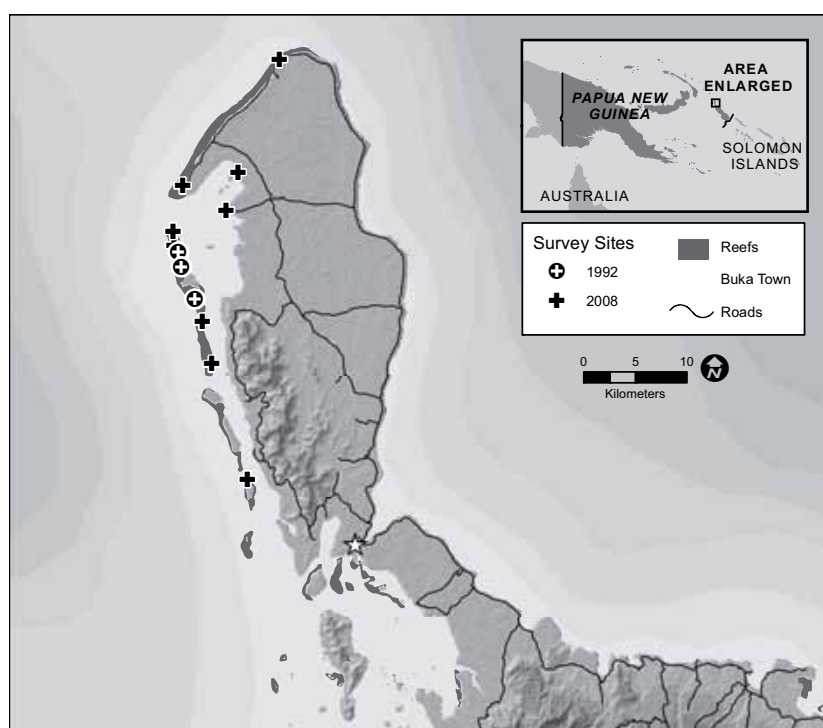


Figure 1. Locations of reef flat sites surveyed in 1992 and 2008 around the western side of Buka Island, Autonomous Region of Bougainville.

Table 1. Relative abundance of each sea cucumber species recorded on reef flats in 1992 and 2008.
Note: Common names follow those used by the Secretariat of the Pacific Community (2003).

| Scientific name | Common name | 1992 relative abundance | 2008 relative abundance |
|------------------------------|--------------------|-------------------------|-------------------------|
| <i>Holothuria scabra</i> | Sandfish | 39.96% | 21.08% |
| <i>Bohadschia similis</i> | Chalkfish | 32.28% | 21.08% |
| <i>Holothuria atra</i> | Lollyfish | 20.35% | 19.46% |
| <i>Actinopyga echinites</i> | Deep-water redfish | 5.24% | 0.95% |
| <i>Actinopyga miliaris</i> | Blackfish | 1.02% | 1.08% |
| <i>Holothuria coluber</i> | Snakefish | 0.96% | 0.54% |
| <i>Stichopus herrmanni</i> | Curryfish | 0.16% | 0.68% |
| <i>Bohadschia argus</i> | Tigerfish | 0.03% | 0.27% |
| <i>Actinopyga mauritiana</i> | Surf redfish | 0 | 18.38% |
| <i>Bohadschia vitiensis</i> | Brown sandfish | 0 | 13.24% |
| <i>Holothuria whitmaei</i> | Black teatfish | 0 | 3.24% |
| Total: | | 100% | 100% |

Table 2. Mean abundance of sea cucumber species on the reef flats in 1992 and 2008. The percentage of each species still remaining on reef flats in 2008 is also shown.

| Species | 1992 (ind. ha ⁻¹) | 2008 (ind. ha ⁻¹) | Percentage remaining since 1992 | Difference significant? |
|-----------------------------|-------------------------------|-------------------------------|---------------------------------|-------------------------|
| <i>Holothuria scabra</i> | 1879.51 | 48.33 | 3% | Yes (P < 0.001) |
| <i>Bohadschia similis</i> | 1392.36 | 43.83 | 3% | Yes (P < 0.001) |
| <i>Holothuria atra</i> | 877.78 | 40.00 | 5% | Yes (P = 0.004) |
| <i>Actinopyga echinites</i> | 226.00 | 1.95 | 1% | Yes (P < 0.001) |
| <i>Actinopyga miliaris</i> | 44.10 | 2.22 | 5% | No (P = 0.069) |
| <i>Holothuria coluber</i> | 41.32 | 1.11 | 3% | Yes (P < 0.001) |
| <i>Stichopus herrmanni</i> | 6.94 | 1.39 | 20% | Yes (P = 0.002) |
| <i>Bohadschia argus</i> | 1.39 | 0.55 | 40% | No (P = 0.220) |

Abundance

By 2008, the densities of the three most commonly sighted sea cucumbers (*H. atra*, *H. scabra*, *B. similis*) were only a fraction of their former 1992 abundances (Fig. 2).

Of the eight species sighted on reef flats in both the 1992 and 2008 surveys, all declined to 1–40% of their former abundance. By 2008, six of the eight species that were sighted in both surveys showed reductions in abundance of 95% to 99% of 1992 levels. For most of these species, these declines were statistically significant (Table 2).

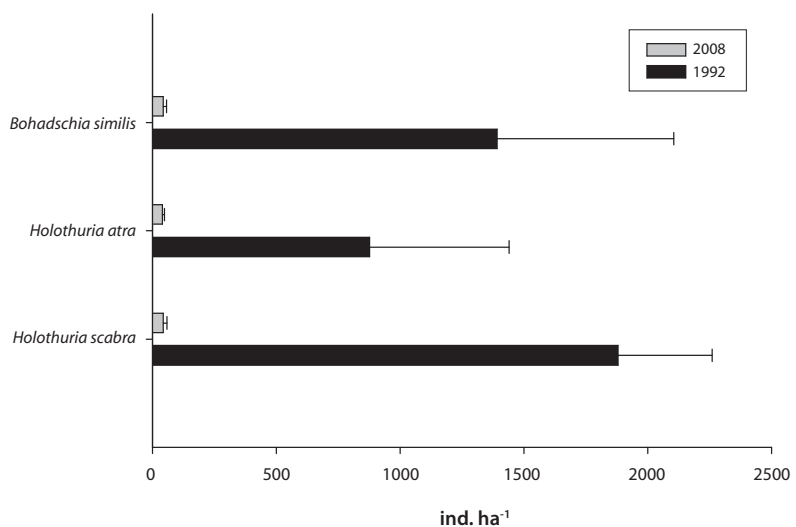


Figure 2. Mean abundance (ind. ha⁻¹) of the three sea cucumber species that were most commonly sighted on reef flats in 1992 (n=12) and 2008 (n=90).

Discussion

A comparison of 1992 and 2008 data provides compelling quantitative evidence that sea cucumber populations around Buka Island have been severely overfished. All species recorded on reef flats in both the 1992 and 2008 surveys showed major reductions in abundance over this 16-year period. These changes were typically extreme, with six of the eight species declining to between 1% and 5% of 1992 levels. While we only have historical data for a small spatial area, it seems highly probable that the declines seen on the reef flats in the Buka region are indicative of the fishery throughout ARB. In the much more intensive 2008 survey, we sampled 80 sites from five habitats (reef flat, reef crest, shallow reef slope, deep reef slope and lagoon) around the northern ARB, and at virtually all sites sea cucumbers were present in low to very low abundances (Hamilton et al. 2010). Several decades previously all of these habitats in the northern ARB had supported large abundances of sea cucumbers (Paul Lokani, pers. observ.). It is possible that declines in other habitats may be even more extreme than what occurred on reef flats. A comparison of sea cucumber densities in 1992 and 2006 in New Ireland Province showed that sea cucumber densities in reef flat environments declined less than in other habitats such as lagoons (PNG National Fisheries Authority 2007).

The only site in the 2008 survey that had a high abundance of sea cucumbers was a single reef crest site in the Tinputs region, which had high densities of *H. atra*. This site had been closed to fishing by the community for several years. The much higher-than-average densities of *H. atra* at this site provide evidence for the positive impact that protection can have for this species (Hamilton et al. 2010). In a recent survey of sea cucumbers in PNG's New Ireland Province, sea cucumber densities in two customary protected "control" sites were compared with densities from 40 other sites around New Ireland that are open to fishing. Similar to this study, the New Ireland survey revealed that at one of the two "control" sites *H. atra* (but not other species) were in far greater abundances on inter tidal habitats than at surrounding sites that were open to fishing (PNG National Fisheries Authority 2007).

Traditionally, many regions in Bougainville placed *tambus* on reefs in order to allow stocks to recover (Lokani 1995). Encouraging the reestablishment of such practices would be one way of managing sea cucumbers at the community level. The current moratorium that was placed on PNG's sea cucumber fishery in 2009 should also allow stocks the chance to recover. However a cautionary note is that even long-term closures have had mixed success at rebuilding overfished sea cucumber fisheries in the western Pacific (Friedman et al. 2010).

Maintaining the current moratorium and conducting follow-up surveys in the future before a decision is made to re-open the sea cucumber fishery in the ARB is clearly desirable.

The 2008 survey recorded three sea cucumber species (*A. mauritiana*, *B. vitiensis* and *H. whitmaei*) that were not recorded in the 1992 survey. Of these three species *A. mauritiana* and *B. vitiensis* were relatively dominant, comprising 31% of all sea cucumbers sighted. Finding higher species diversity on overfished reefs is not what one expects. However, in this instance, it appears to relate in part to patchy distributions of these species and the fact that the 2008 survey covered a larger area than the 1992 survey. A close examination of the 2008 data reveals that nearly all *A. mauritiana* recorded were sighted at two sites just north of the sites surveyed in 1992. These two sites are on a narrow reef flat, and transects placed here intersected the surf zone. This was not the case for the three 1992 sites, which were on wider reef flats. This explains the relative dominance of *A. mauritiana* in the 2008 surveys, because this species is typically found in the surf zone. The higher abundances of *B. vitiensis* are more difficult to explain because this species occupies a similar habitat to *B. similis*, and *B. similis* was dominant in both the 1992 and 2008 surveys. It may be that *B. vitiensis* has reversed the general trend and shown a real increase in abundance over the 16-year period.

Acknowledgements

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An assessment of *Holothuria scabra* growth in marine micro-farms in southwestern Madagascar

Gaëtan Tsiresy,¹ Benjamin Pascal² and Thomas Plotieau³

Abstract

This article presents and compares the results of some initial *Holothuria scabra* rearing cycles in three villages on the southwestern coast of Madagascar. Growth and survival rates differ between these three pilot sites. Survival rates are very low in two of the three villages, although very fast growth is recorded for holothurians at these locations. In the third village, the situation is reversed: the survival rate is higher but first-phase growth is very slow. *In situ* trials and sediment analysis have made it possible to identify a number of parameters that influence production. The main factors are the presence of crabs, which affect juvenile survival, and sediment structure, which has an impact on growth rates. As a result of these observations, some improvements in production techniques were shown to be effective, such as ploughing and crab control systems. The purpose of this article is to review the available knowledge on small-scale farming in the natural environment.

Introduction

In the Toliara region (southwestern Madagascar), sea cucumber harvesting continues to be a widespread traditional activity in coastal communities (Rasolofonirina and Conand 1998). From the 1990s onward, overharvesting has been characterised by declining quality and size in the products sent to market (Conand et al. 1998). In response to environmental and socioeconomic threats arising from the overharvesting of sea cucumbers, hatchery and culture projects are being launched in increasing numbers around the world. Pioneering work by Aqua-Lab on the reproduction and growth of captive *Holothuria scabra* (southwestern Malagasy coast) has opened the way for a second phase applied to rural development: the creation of micro-farms managed by families of traditional fishers. These traditional fishers are currently supported by two non-governmental organisations: Trans-Mad'Développement (TMD) and Blue Ventures (BV) (Robinson and Pascal 2009).

A year after the start of TMD's mariculture project, a technical supervisory team (and the holothurian culture groups it supports in the three coastal villages) provided an initial assessment of the problems they encountered. In particular, the only available information for selecting sites favourable for farming *Holothuria scabra* is incomplete, and relevant identification methods have yet to be defined.

Sites near mangroves, with sandy and/or muddy bottoms partly colonised by algae, are considered to be conducive for culturing sandfish. Past experience, however, indicates that the criteria for identifying favourable sites and our understanding of the related factors require refinement.

Materials and methods

This assessment covers 17 farms — each 900 m² in area, which were supplied with 450 juveniles for the first culture cycle. The farms are located in three coastal villages: five in Sarodrano, seven in Andrevo and five in Fiherenamasay.

The methodology used to assess growth processes involves the practical application of rearing techniques by aquaculturists and project staff, and consideration of the results recorded during production monitoring activities. The study protocol, therefore, results from the various village aquaculture techniques and implementation stages and the monitoring methods used by technicians.

Supply of juveniles

Juveniles were supplied by Madagascar Holothurie Société Anonyme, a company operating in Toliara and Belaza, 25 km south of Toliara (Eeckhaut et al. 2008). *H. scabra* juveniles were transported by sea

1. PhD student at the Institut Halieutique et des Sciences Marines (University of Toliara – Madagascar); responsible for scientific supervision of the sea cucumber culture project (Trans-Mad'Développement). Institut Halieutique et des Sciences Marines (IHSM), University of Tuléar, 601 Tuléar, Madagascar.

2. Doctor of Geography; Coordinator of the aquaculture programme (Trans-Mad'Développement); Associate researcher in UMR208 (IRD/MNHN). Trans'Mad-Development, Besakoa, Antsirasa 601, Toliara, Madagascar. Email: benjmada@yahoo.fr

3. Laboratoire de Biologie marine, University of Mons, 7000 Mons, Belgique.

in plastic crates to Andrevo and Fiherenamasay (a 4–10 hour trip), and by canoes to Sarodrano (a 45-minute trip in a sheltered lagoon).

Because transport time in the north is considerably longer and rougher, transport-related stress is a significant factor for juveniles delivered to Andrevo and Fiherenamasay than for those sent to Sarodrano.

Grain-size analysis and determination of organic matter content

Analyses on grain size and organic matter content were carried out in order to characterise the various sediment structures at the three holothurian farming sites. Sediment samples were collected from each site and dried for 72 hours in an oven at 60°C. A sample of dry sediment was sieved and each fraction collected was weighed so as to establish its percentage of the original sample. To determine the organic matter content, the sediment was charred for four hours at 500°C.

Monitoring farm production

In order to monitor production trends at each site, holothurians were counted at night at low tide and then weighed. Depending on weather conditions, the results of these monitoring operations may have suffered from various kinds of biases. A number of different factors may, in fact, affect holothurian behaviour (e.g. burrowing) and the conditions that influence the detection of animals (e.g. visibility in the water, low tide level, light intensity depending on the moon and available light). In each operation, more than one-quarter of the specimens initially supplied were weighed in order to gauge weight gain (i.e. at least 113 specimens weighed each time out of the 450 initially delivered).

The physio-chemical parameters known to affect sea cucumber growth and survival were assessed monthly during production monitoring work. Seawater temperature and salinity were recorded monthly (alternating with a spring tide), during the day and at night. These two parameters did not significantly vary from one site to another during the initial culture cycle. Their influence was, therefore, not considered when interpreting the results presented in this article. Water salinity remained stable at 35‰ throughout the growth cycle at the three sites. Furthermore, temperature did not differ significantly between the three growth cycles considered in this study.

Targeted trials and identification of improved farming techniques

Controlling predation

We observed a significant decline in holothurian stocks during farming, mainly due to predation by crabs (Fig. 1). To control crab predation, crab traps — made of wire mesh with an entrance hole — and simple traps (plastic buckets with monofilament line attached to the rim, and bait in the middle) were installed. The farmers also regularly organised crab hunts.



Figure 1. A crab attacking a holothurian
(Photo: G. Tsiresy, TMD, 2009)

Because of the crabs' constant presence in the pens, we looked for ways to physically prevent them from approaching the most vulnerable holothurian juveniles. One solution involved setting up 25-m² nurseries (totally enclosed and roofed mini-pens, Fig. 2) in which the newly delivered juveniles were left to grow for the first few weeks before being moved to the larger pen.



Figure 2. Nursery at Belaza, 20 km at south of Toliara, Madagascar
(Photo: B. Pascal, TMD, 2009).

Improving farming techniques

Having observed a very slow growth rate at Fiherenamasay (0.23g day^{-1} of growth), we carried out some experiments to improve holothurian weight gain at this site. Six experimental 4-m^2 sections were constructed within the pens, each containing 50 small holothurians ($< 40\text{ g}$). In two sections, the sediment was ploughed; in another two sections the surface sediment layer was scraped off (5cm); and two sections were left unchanged (i.e. were controls).

After identifying the effects of ploughing on growth, it was decided to extend the ploughing system to cover the entire bottom area of the four pens at Fiherenamasay.

Data analysis

The ANOVA (two-factor) parametric analysis technique was used to compare observed means. The statistical analysis was done using SYSTAT 6.0.1, SPSS INC software.

Results and discussion

Sites offering highly variable aquacultural potential

After five months of growth, major differences could be observed between the three sites, although these differences were obvious from the first month onward, as shown in Figure 3. In terms of survival, differences were significant (Fig. 4). The mortality rate at one site was nearly 40% (Fiherenamasay) while the other sites were over 70% (Sarodrano and Andrevo).

Growth rates varied by a factor of 1 to 8, depending on the site: from 0.23g day^{-1} at Fiherenamasay (where holothurians grew the slowest) to 1.8g day^{-1} at Andrevo (where holothurians grew the most). Five months after seeding, juvenile weights differed significantly, again depending on the site — (2-factor ANOVA; $p = 0.001$) between Sarodrano and Fiherenamasay, and ($p < 0.001$) and between Andrevo and Fierenamasay. In contrast, no significant difference was recorded between Sarodrano and Andrevo ($p = 0.870$).

It should be noted that the curve steepens from about months three and four for the Fiherenamasay site due to a technical measure involving ploughing the site, first to a limited extent by technicians and later to a greater extent by farmers. With these improvements, the growth rate increased from 0.23g day^{-1} to almost 1g day^{-1} . The sediment structure, therefore, seems to have a major effect on growth.

Irrespective of these changes, growth rates were constant at the three sites throughout the farming process as long as the biomass limit was not reached. The differences observed, therefore, relate to growth rates, regardless of the critical biomass specific to each site. Indeed, the curve shows that the environment's maximum capacity is not reached at the three sites (45 g m^{-2} of farmed biomass) due to the low holothurian densities in the pens and their low mass. If the biomass threshold was reached (biomass estimated at 692 g m^{-2} at favourable sites in the region: Lavitra 2008), the curves would look asymptotic, showing a plateau, which would mean that the holothurians would have ceased growing. This is not the case at all and the curves remain linear.

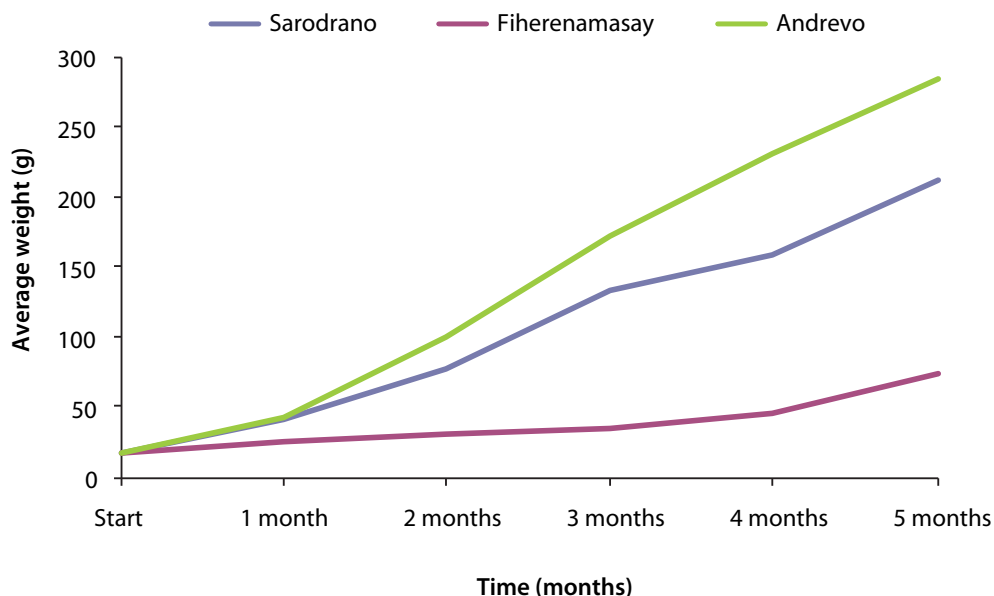


Figure 3. Observed growth rates by site.

Table 1. Sediment characteristics.*

| Particle size (µm) | Sediment fraction | Sarodrano village | | Andrevo village | | Fiherenamasay village | |
|--|-------------------|------------------------|---------|------------------------|---------|------------------------|---------|
| | | Gran T0 | Gran T4 | Gran T0 | Gran T4 | Gran T0 | Gran T4 |
| > 1,000 | Very coarse sand | 13.93% | 12.78% | 17.40% | 14.65% | 24.27% | 32.10% |
| 500 to 1,000 | Coarse sand | 11.92% | 13.83% | 34.55% | 35.82% | 36.39% | 25.15% |
| 250 to 500 | Medium sand | 48.07% | 36.26% | 21.67% | 22.16% | 25.96% | 20.13% |
| < 250 | Fine sand | 26.08% | 37.12% | 26.38% | 27.37% | 13.70% | 22.61% |
| State of the surface sediment layer | | Loose | | Loose | | Compact | |
| Organic matter content T0 | | Min 1.44% Max 1.90% | | Min 1.41% Max 1.80% | | Min 2.74% Max 2.93% | |
| Organic matter content T4 | | Min 2.32% Max 2.68% | | Min 1.83% Max 2.18% | | Min 2.88% Max 3.11% | |
| Growth rate | | 1.3 g d ⁻¹ | | 1.8 g d ⁻¹ | | 0.23 g d ⁻¹ | |

* Grain size profile and sediment organic matter content at the three sites.
Gran = granulometry; T0 = date farming began; T4 = after 4 months of farming.

A subtle combination of factors governing growth mechanisms

What emerges when interpreting the results given in Table 1 is that the amount of organic matter present in the sediment is not an adequate criterion for explaining differing growth rates. While Fiherenamasay shows an organic matter content higher than Sarodrano or Andrevo, growth rates at that site are at least five times slower.

These characteristics point to two types of environment at the three sites. Andrevo and Sarodrano have very loose sediment and high growth rates, while Fiherenamasay has compact sediment (for the surface layer), with a relatively high organic matter content and slow growth.

Research on *H. scabra's* feeding habits has shown that when sediment was ingested, this species was not selective with regard to sediment particles smaller than 2mm (Lavitra 2008). Similarities between the Sarodrano and Andrevo sites in terms of sedimentary structures, however (small proportion of particles larger than 1 mm), and growth rates, and the differences with the Fiherenamasay site, suggest that sediment grain size structure and compactness play a role in growth rates.

Thus, it is legitimate to theorise that, where growth rates (and not critical biomass) are concerned, an environment offering high organic matter content with a grain size structure dominated by coarse particles (even under 2 mm) will be less favourable than a location with a finer particle size but with a low organic matter content. Grain size features could, therefore, be a key factor in understanding growth rates.

In order to consolidate this reasoning, an understanding of organic matter composition and the abundance of its component living micro-organisms would be needed, and this could potentially vary from site to site; the organic matter at Andrevo and Sarodrano may be better suited to the nutritional requirements of *H. scabra*. The experiment carried out at Fiherenamasay supports our theory, however, because ploughing the sediment speeds up the growth rate of cultured sea cucumbers. Modifying the sediment structure has, therefore, produced very similar growth rates as at the two other sites. The sediment ploughing process produces a looser form of sediment that contains a larger proportion of large particles (32.10%) and very small (22.61%) particles, with few medium-sized ones. Skimming off the surface layer reveals a muddier, more powdery sediment. It could be that the truly determining factor is sediment structure (grain size and compactness) because of its indirect action on the associated organic matter.

While the quantity of organic matter present in the sediment is a determining factor in explaining threshold biomass, it appears that sediment structure regulates organic matter content accessibility and, therefore, drives growth rates. The types of organic matter should also be taken into consideration. Growth rate and threshold biomass can be completely independent of each other.

Early mortality

From these initial results, it can be seen that the Andrevo and Sarodrano sites are far more favourable for farming *H. scabra* than Fiherenamasay in terms of growth rates. Farming cycles can be shorter at these two locations. However, from the point of

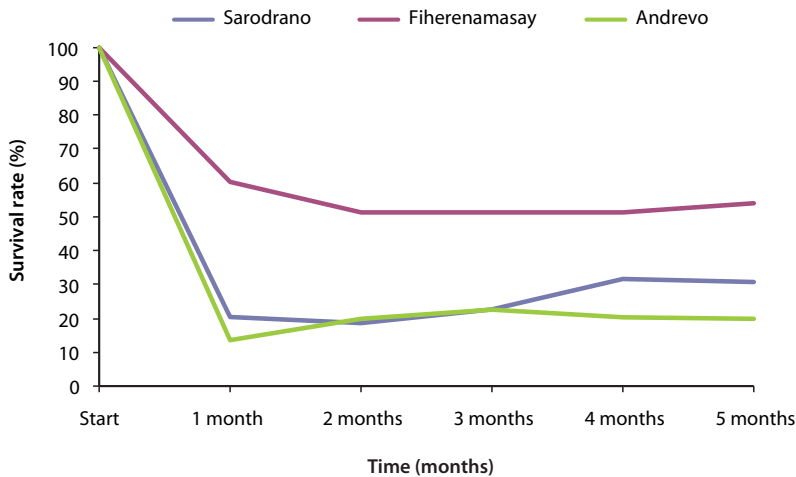


Figure 4. Survival rate by site (estimated from night censuses).

view of aquacultural development, these two sites both have the same drawback: early mortality in farmed holothurians (Fig. 4).

The three curves in Figure 4 show survival rates at the farms, and all indicate that 30 days after seeding, many juveniles die off. The shape of the curves (Fig. 4) shows that mortality occurs during the early weeks of rearing.

It can be noted that survival rates fall rapidly by between 10% and 30% in the villages of Sarodrano and Andrevo compared with 60% for Fiherenamasay. At the fifth month of rearing, the survival rates obtained in the three villages are: 30.71% at Sarodrano, 53.82% at Fiherenamasay and 20% at Andrevo. Stocks do not, therefore, seem to sustain any significant losses after the first month.

Predation: A compelling theory

Three main reasons may explain these poor survival rates: predation, disappearance (due to tidal currents or by juveniles escaping over the top of the fence) or theft.

The theft theory can be discounted immediately. First, cultured holothurians only acquire commercial value above about 100 g in weight, meaning not before the third month of growth. While end-of-cycle losses may be due to theft, first-month losses cannot be ascribed to thieves, who would try and sell the juveniles on the local market. Had these juveniles been stolen by other farmers for further growing, our monitoring work would have revealed a suspicious increase in the pens of thieves, which has not been the case.

Although it offers some probability, the explanation that holothurians disappear from the pens under the effect of marine currents does not (for

now) seem sufficiently convincing. It is true that tidal currents can be quite strong in the rearing zones. Farmers say that they have observed young holothurians floating in and being carried away by currents. The close-interval monitoring we carried out after deliveries, however, has always shown that holothurians do not move very far away from their initial settlement location in the pen. Juvenile scattering toward the fence, due to currents, has never been observed, and juveniles have never been found outside the fence when checks were made.

The disappearance of juveniles because of predation emerges as the most plausible hypothesis. In the natural environment, newly stocked juveniles are frequently attacked and eaten by various fish species (Hamel et al. 2001; Pitt and Duy 2004) and crabs (Pitt and Duy 2004; Lavitra 2008). When the first deliveries were made at night, we observed crabs attacking juveniles in under 10 minutes (Figs. 5a et 5b). Other specimens were sometimes found with tegument wounds, and some remains were found during checks (Fig. 5b). The most surprising discovery was the extraordinarily high level of predation, with the entire stock (450 juveniles) in some pens disappearing in the space of two weeks.



Figure 5. a: Young holothurian after predation; **b:** Holothurian remains found in a pen (Photos: G. Tsiresy, TMD, 2009).

Nursery effect

During the second rearing cycle at Sarodrano and Andrevo — which suffer the greatest losses of juveniles — the introduction of nurseries (smaller pens covered with netting where juveniles were placed) made it possible to significantly restrict losses in the first month. The idea behind these nurseries was to physically prevent crabs from approaching juveniles during the first weeks of growth in the sea.

With this new system, 15 days after stocking, the observed survival rates were 79% for Sarodrano and 70% for Andrevo. Nurseries, therefore, produce better survival rates. Unfortunately, these positive results have led farmers to neglect crab culling, both in nurseries and in the rest of the pens, and losses after the first few days were colossal. Some 35 days after delivery, the survival rate had dropped to 33% at Andrevo, equating to a loss of 37% of stocks in 20 days in comparison with the first count. After 70 days, the survival rate had dropped to 51% at Sarodrano although it was still 79% after 15 days in the sea and 76% after 35 days in the sea.

This new system seems to have no effect on juvenile growth if the stocking density is suitable for the specific biomass of each site.

Conclusion

These initial results clearly reveal the importance of certain factors in production dynamics. Excessive mortality at some sites is a major obstacle to the economic viability of the aquaculture ventures. Unless an effective counter-measure can be found, the sites where crabs occur in abundance will probably not be suitable for profitable farms. Depending on the relevant issues, technical solutions such as nurseries or sediment ploughing can be found and provide worthwhile results but will always be insufficient without close supervision by farmers of their stock.

Our results are best applied to growth mechanisms, and suggest that sediment structure is just as important for growth as high organic matter content. In order to refine the scope of these results, growth tests should be carried out in a range of sediments to validate the effect of grain size features and the role of organic matter in growth mechanisms. The composition of organic matter should also be taken into consideration so as to improve our understanding of growth mechanisms of *H. scabra*. One question remains key to a reliable future for farming: Does the consumption of organic matter over successive rearing cycles reduce the resilience of the environment? If it does, would this mean that the critical biomass threshold shrinks during farming, and that fallow systems would probably have to be brought in to avoid impoverishing the sedimentary bottom soils?

These few results show that site selection and the characterisation of their particularities enable rearing techniques to be adapted and failures to be avoided. A site offering rapid growth will not necessarily have a high critical biomass (stocking capacity) and, conversely, a slow-growth environment may potentially offer a high stocking capacity. This

aspect implies that production models will vary between sites in terms of techniques (e.g. ploughing, nurseries) but also in terms of rearing densities and juvenile delivery frequency. For this purpose, there would be merit in developing a farming site classification guide. A technical protocol would have to be drafted for each type of environment so that optimum husbandry practices can be used. The availability of such a guide would make the work of field support operators much easier and help promote holothurian farming much more successfully.

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Simultaneous mass spawning of *Holothuria scabra* in sea ranching sites in Bolinao and Anda municipalities, Philippines

Ronald Dionnie D. Olavides,¹ Bryan Dave R. Rodriguez, Marie Antonette Juinio-Meñez

The Bolinao-Anda reef system in Pangasinan, north-western Luzon is among the many areas in the Philippines where sea cucumbers and invertebrate fisheries thrived in the 1970s and 1980s. These fisheries, however, have collapsed in the last two decades due to overfishing. With improvements in hatchery technology for sandfish (*Holothuria scabra*), the University of the Philippines Marine Science Institute was able to establish five-hectare sea cucumber ranching sites in the municipalities of Bolinao and Anda (Fig. 1). The first batch of juveniles released in Bolinao and Anda was in December 2007 and December 2008, respectively. We are studying the feasibility of community-supported sea ranching sites to provide supplemental income to our sea ranch operators (composed of marginal fishers), and its potential to help replenish depleted stocks. We have released 16,711 individuals in Bolinao and 20,549 in Anda, to date.

In recent months, we have received anecdotal accounts of solitary sandfish spawners in the managed area from sea ranchers who guard and co-manage the site. For the first time, however, we not only verified spontaneous spawning of sandfish at the sea ranch but also documented a simultaneous mass-spawning event at both the Bolinao and Anda sites on 23 February 2010 from 11:00 to 15:00 (a day after the first quarter moon, and on a rising tide) during our regular site visits. We conducted rapid surveys at both sea ranching sites to estimate the number and density of spawners.

At Anda, we first observed five male spawners at around 11:00. An hour later, a female spawner released gametes in two bursts, followed by four male spawners. In Bolinao, we first observed a spawner at around 12:45 (Fig. 2). Also about an hour later, we found that 15 individuals had

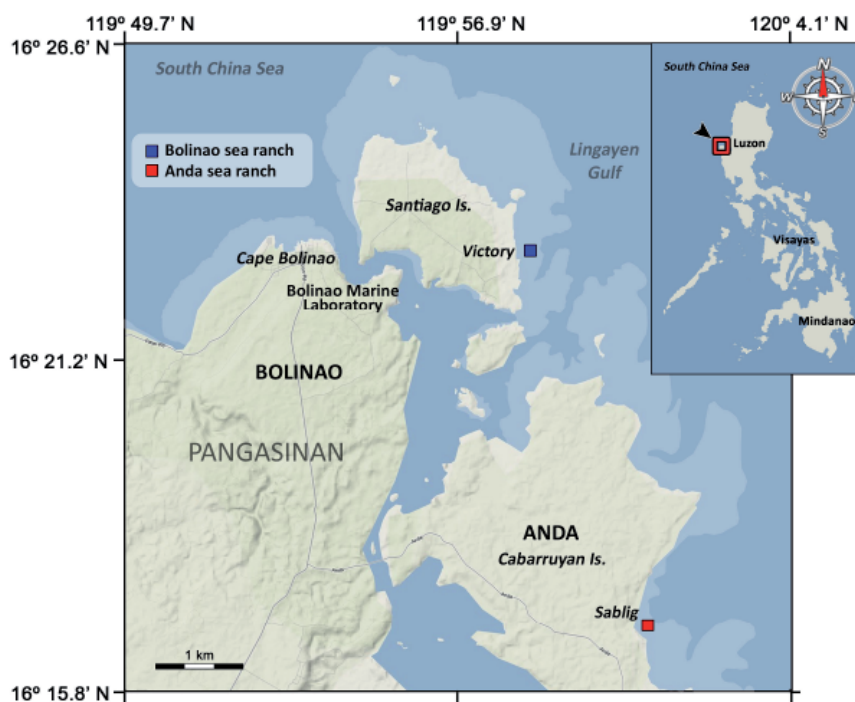


Figure 1. Location of Bolinao and Anda sea ranching sites, off the island of Luzon in the Philippines.

1. The Marine Science Institute, University of the Philippines, Diliman, Quezon City 1101, Philippines. Phone: +63 2 922 3959. Fax: +63 2 924 7678. Email: olavides.ronald@gmail.com



Figure 2. *Holothuria scabra* specimens spawning at the Anda sea ranch site.

Table 1. Summary of observational data on sandfish mass spawning inside sea ranching sites (23 February 2010).

| Site | Search effort | Ratio of spawners | Density of spawners |
|-------------------|------------------------------|--------------------------|---|
| Bolinao sea ranch | ~700 m ² (45 min) | 19.7% (38 out of 181) | 0.0543 ind m ⁻² (estimated 543 ind ha ⁻¹) |
| Anda sea ranch | 1,600 m ² (1 h) | 34% (60 out of 175) | 0.0375 ind m ⁻² (estimated 375 ind ha ⁻¹) |

spawned within 1–5 m of each other inside a 100-m² sea pen, and 23 other spawners in the “nursery” and “buffer” zones of the sea ranch. We observed 60 spawners (weight range = 250–750 g, mean weight = 402 ±76 g) in Anda, and 38 spawners (weight range = 150–400 g) in Bolinao sea ranch. We estimate a ratio of 5 males for every 1 female at both sites.

Synchrony of gamete release, high densities of free-spawners and large population sizes have been reported to increase fertilization success (Levitan and Sewell 1998). While minimum density figures for successful sea cucumber spawning are not yet available, theoretic densities in the range of 10–50 ind ha⁻¹ may be able to avoid depensation for most tropical sea cucumbers, provided that there are groups of more than 10 individuals within 5–10 m of one another (Bell et al. 2008). Spawning densities for both sites are above those suggested by Bell et al. (2008), and the spawning synchrony is, at least, a contributing factor for fertilization success. The natural observation of mass spawning, in addition to the large proportion of adults for Bolinao and Anda sites from empirical data on population parameters (Juinio-Meñez et al. unpublished data), show positive implications on the potential of sea ranching

sites to serve as functional reproductive reserves that could provide sources of larvae and recruits needed for the restoration of stocks.

Acknowledgement

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Shallow-water sea cucumber inventory in the Sultanate of Oman

Michel R. Claereboudt¹ and Khalfan M. Al-Rashdi²

Abstract

The Sultanate of Oman's coastline extends over 3,500 km in three connected bodies of water: the Arabian Gulf (aka Persian Gulf), the Sea of Oman and the Arabian Sea. The aim of this two-year study (from 2007–2009) was to evaluate the diversity and abundance of holothuroids along Oman's coastline. The study was conducted at 11 locations, and in total, 17 species of Aspidochirotida, 2 species of Apoda and 2 species of Dendrochirotida were observed. The most striking result is the large difference in sea cucumber community structure between the Sea of Oman and the Arabian Sea.

Introduction

Although there is a growing number of studies focusing on echinoderms and holothuroids worldwide, the Arabian Sea has not received much attention. There have been a few publications on echinoderms in the region (e.g. Campbell 1988; Price 1982, 1983) but with the exception of recent studies on the fisheries and stock assessment of *Holothuria scabra* (Al-Rashdi et al. 2007a; Al-Rashdi et al. 2007b), there are no (to our knowledge) first hand records of sea cucumbers in the Sultanate of Oman.

The Sultanate of Oman's coastline extends over than 3,500 km in three connected bodies of water: the Arabian Gulf (aka Persian Gulf), the Sea of Oman and the Arabian Sea. The country has always depended on the sea, however human pressure on its natural resources is steadily increasing from fishing, extensive infrastructure development, tourism and industry. Global changes have also taken their toll on marine ecosystems with the passages of the first two recorded tropical cyclones in the Sea of Oman in recent history (Fritz et al. 2010) and an unprecedented and devastating bloom of the toxic dinoflagellate *Cochlodinium polycrikoides* in 2008–2009 (Richlen et al. 2010).

The marine climates in each of the three bodies of water are very distinct. The Arabian Sea coastal ecosystems withstand the full force of a seasonal coastal upwelling during the summer monsoon (June–September) with sea surface temperatures often dropping well below 20°C. The cool upwelled water is accompanied by a steady influx of nutrients that feeds the growth of extensive beds of benthic algae (Barratt 1984) interspersed with rich coral communities. In the Arabian Gulf and the Sea of Oman, sea surface temperatures in the same summer

period often exceed 32°C, whereas in the winter, water temperatures drop to below 22°C. Despite these extremes, coral communities flourish along the hard-substrate shores (Sheppard and Sheppard 1991) and support a rich echinoderm fauna.

There is only one marine protected area (the Daimaniyat Nature Reserve) in the Sea of Oman, and it covers about 20 km² and encompasses a string of nine small islands (Fig. 1). The aim of this study was to evaluate the diversity and abundance of holothuroids in the Sultanate of Oman as a first step toward monitoring and managing this important but fragile resource.

Methods

This study was conducted over two years (from 2007–2009) at 11 locations around the Sultanate of Oman (Fig. 1). At each location, several sites (a minimum of three) were surveyed by scuba from the surface to a maximum depth of 20 m. All holothuroids encountered were identified to the species level on the basis of their gross morphological characteristics, habitat and posture. Species were photographed and some specimens were collected and deposited at Sultan Qaboos University. Species identification followed the description of Clark and Rowe (1971) and more recent reviews of Holothuroidea in the Indian Ocean (Price 1983; Samyn 2003; Samyn et al. 2006).

At each location, at the end of a series of at least three dives in different sites, the abundance of the various species was recorded on a semi-quantitative scale (Braun-Blanquet 1932) from 0 to 5, with 0 = not recorded; 1 = rare (observed once or twice, but not on every dive); 2 = present (observed on nearly every dive at this location); 3 = common (observed several times

1. Sultan Qaboos University. College of Agricultural and Marine Sciences. Box 34, Al-Khod 123, Sultanate of Oman. Email: mclaereboudt@mac.com>

2. Aquaculture Center. Ministry of Fisheries Wealth, PO Box 247, PC 100, Muscat, Sultanate of Oman. Email: omanaba@yahoo.com

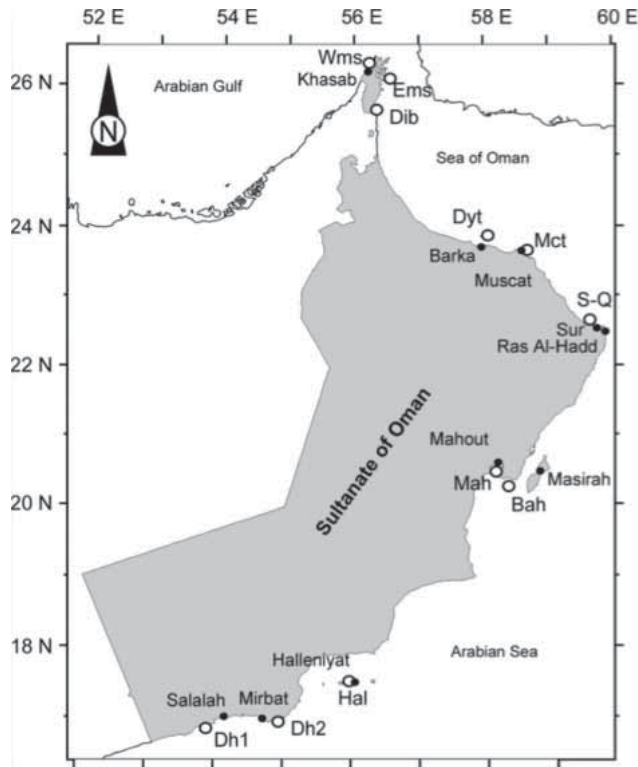


Figure 1. The 11 sea cucumber sampling locations around the Sultanate of Oman. Black, filled circles are cities and towns, and unfilled circles are sampling sites: Dh1 = Dhofar Rakhyut, Dh2 = Dhofar Mirbat, Hal = Halleniyat, Mah = Mahout, Bah = Masirah-Bar Al-Hikman, S-Q = Sur-Qalhat, Mct = Muscat, Dyt = Damanyat, Dib = Dibab, Ems = East Musandam, Wms = West Musandam.

during each dive at that location); 4 = abundant (observed many times during each dive) and 5 = dominant (one of the species encountered in significant numbers during any given dive). Distribution patterns were analyzed by clustering analysis and non-metric multidimensional scaling (MDS) on Bray-Curtis similarity matrix using Primer (Clarke and Gorley 2006).

Results

In total, 17 species of Aspidochirotida, 2 species of Apoda and 2 species of Dendrochirotida were observed during the survey (Table 1). Additional species of Dendrochirotida were observed in the south of Oman but were neither collected nor identified and will require additional fieldwork.

Although eight species were found in all or nearly all locations, many others were only found in four locations or fewer. *Holothuria scabra* (Fig. 2E), for instance, was restricted to two locations on the Arabian Sea near the Island of Masirah, and *Holothuria arenacava* was only found in a few sandy embayments near Muscat (two sites at one location) (Fig. 2A). The overall number of species decreased from the Arabian Sea (19 species) to the Sea of Oman (13 species) to the Arabian Gulf (8 species). Several species were recorded for the first time during the survey: *Holothuria arenacava* (Fig. 2A), *H. nobilis* (Fig. 2C), a common yet unidentified species of *Holothuria* (Fig. 2B), *H. cinerescens*, and a mottled pink *Actinopyga* (Fig. 2D), these last three were from Dhofar (southern Oman). *Holothuria hilla* and *Holothuria impatiens* (Fig. 2F) were also observed.

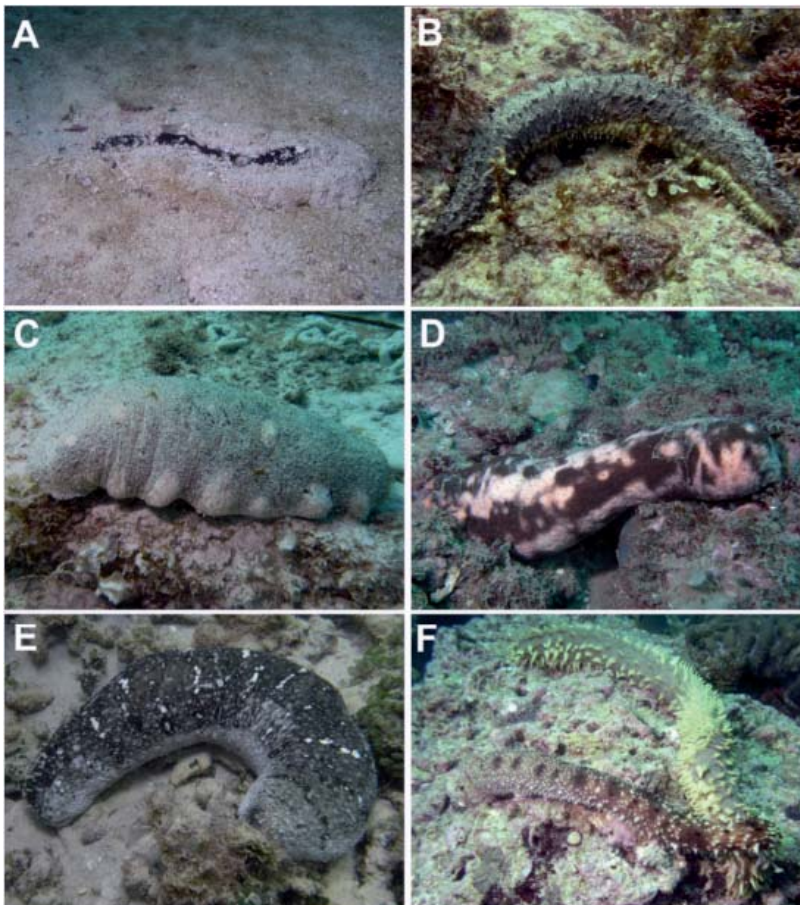


Figure 2.

Some of the holothuroids observed in the shallow waters of Oman.

- A: *Holothuria arenacava*, specimen nearly completely buried;
 B: unidentified species of *Holothuria*;
 C: *Holothuria nobilis*;
 D: unidentified species of *Actinopyga*.
 E: *Holothuria scabra*;
 F: *Holothuria hilla* (larger specimen) and *Holothuria impatiens*.

Table 1. Shallow-water Holothuroidea fauna in the NE Indian Ocean from Price (1982,1983) and relative abundances of species of holothuroids observed during the survey in the shallow waters of the Sultanate of Oman.

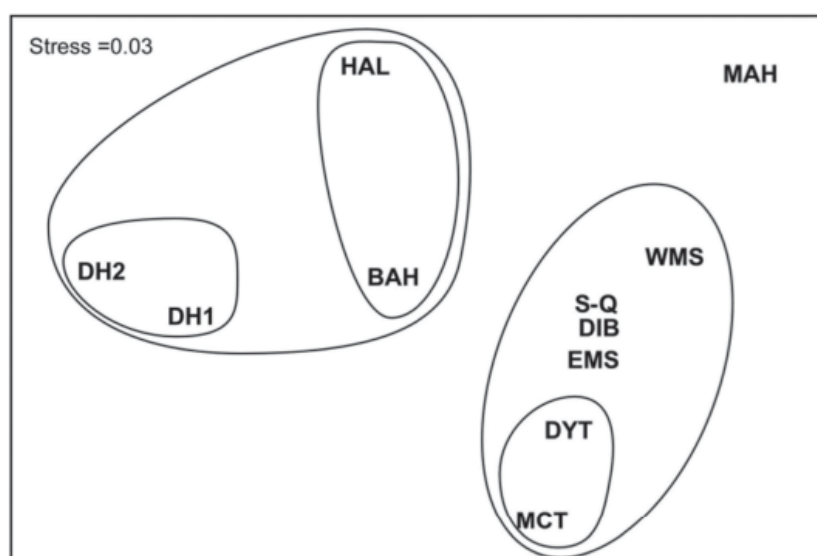
| | Arab. Gulf ^a | Arab. Sea ^a | Arabian Sea ^b | | | | | Sea of Oman ^b | | | | | |
|--------------------------------|-------------------------|------------------------|--------------------------|-----|-----|-----|-----|--------------------------|-----|-----|-----|-----|-----|
| | | | Dh1 | Dh2 | Hal | Mah | Bah | S-Q | Mct | Dyt | Dib | EMs | WMs |
| Holothuroidea | | | | | | | | | | | | | |
| <i>Stichopus variegatus</i> | + | + | 3 | 3 | 3 | 2 | 3 | 3 | 4 | 4 | 3 | 3 | 3 |
| <i>Labidodemas semperianum</i> | + | | | | | | | | | | | | |
| <i>Holothuria atra</i> | + | + | 3 | 3 | 3 | 4 | 3 | 3 | 3 | 3 | 3 | 3 | 3 |
| <i>Holothuria edulis</i> | + | + | 3 | 1 | 2 | 2 | 2 | 3 | 4 | 3 | 3 | 3 | 3 |
| <i>Holothuria rigida</i> | + | | | | | | | | | | | | |
| <i>Holothuria leucospilota</i> | + | + | 3 | 4 | 4 | 3 | 3 | 3 | 4 | 3 | 3 | 3 | 3 |
| <i>Holothuria arenicola</i> | + | + | 0 | 0 | 1 | 1 | 0 | 0 | 0 | 1 | 0 | 0 | 0 |
| <i>Holothuria hilla</i> | + | + | 1 | 1 | 1 | 1 | 1 | 2 | 3 | 2 | 2 | 2 | 2 |
| <i>Holothuria impatiens</i> | + | + | 2 | 2 | 0 | 0 | 1 | 0 | 2 | 1 | 0 | 1 | 1 |
| <i>Holothuria scabra</i> | | + | 0 | 0 | 0 | 3 | 1 | 0 | 0 | 0 | 0 | 0 | 0 |
| <i>Holothuria arenacava</i> | | | 0 | 0 | 0 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 0 |
| <i>Holothuria pardalis</i> | + | | | | | | | | | | | | |
| <i>Holothuria nobilis</i> | | + | 1 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| <i>Holothuria difficilis</i> | | + | 2 | 2 | 0 | 0 | 1 | 1 | 1 | 1 | 1 | 1 | 0 |
| <i>Holothuria cinerescens</i> | | + | 1 | 1 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| <i>Holothuria pervicax</i> | | + | 2 | 1 | 1 | 0 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| <i>Actinopyga milliaris</i> | | + | 2 | 2 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| <i>Actinopyga mauritiana</i> | | + | 2 | 2 | 1 | 2 | 3 | 2 | 3 | 4 | 3 | 3 | 2 |
| <i>Actinopyga sp.</i> | | | 0 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| <i>Holothuria sp.</i> | | | 4 | 4 | 2 | 0 | 2 | 0 | 0 | 0 | 0 | 0 | 0 |
| <i>Thyone dura</i> | + | + | 2 | 3 | 1 | 0 | 2 | 0 | 0 | 0 | 0 | 0 | 0 |
| <i>Oshmimella ehrenbergi</i> | + | + | 1 | 2 | 0 | 0 | 0 | 0 | 1 | 1 | 0 | 0 | 0 |
| <i>Euapta goddefroyi</i> | | + | 0 | 0 | 0 | 1 | 1 | 1 | 2 | 2 | 1 | 1 | 0 |
| <i>Leptosynapta chela</i> | + | | | | | | | | | | | | |

Numbers are semi-quantitative abundances (0–5; Braun-Blanquet 1932). Dh1 = Dhofar Rakhyut, Dh2 = Dhofar Mirbat, Hal = Halleniyat, Mah = Mahout, Bah = Masirah-Bar Al-Hikman; S-Q = Sur-Qalhat; Mct = Muscat; Dyt = Damanyat; Dib = Dibab; Ems = east Musandam; Wms = west Musandam.

^a Arabian Gulf and Arabian Sea from Price (1982,1983).

^b This survey.

Figure 3. Non-metric multidimensional scaling plot (MDS) of the 11 sites with regards to the sea cucumber community structure. The Bray-Curtis similarity index was used on the semi-quantitative abundance data to calculate inter-site similarities. The low stress value (0.03) indicates a good two-dimensional representation of the multidimensional data. See legend of figure one for the names of sampling sites.



From a community standpoint, both the non-metric multidimensional scaling analysis and the cluster analysis, identified a major split in the structure of sea cucumber communities between the Arabian Sea and the Sea of Oman. This first subdivision is mainly due to six species that were restricted to the Arabian Sea coast of the Sultanate: *Holothuria scabra*, *H. nobilis*, *Actinopyga miliaris*, *Actinopyga* sp. (unidentified species), *H. cinerescens* and an unidentified species of *Holothuria* sp. Secondary subdivisions of communities distinguish the northern part of the Arabian Sea coast from its most southern part (Dhofar). In the Sea of Oman, sea cucumber communities from the central part of the Gulf (Muscat) are separated from those in the northern and southern regions (Fig. 3). The holothuroid community found near Mahout Island appeared to be relatively distinct from both that of the Arabian Sea and the Sea of Oman.

Discussion

The holothuroid fauna of Oman is a somewhat impoverished subset of the tropical Indian Ocean fauna (Rowe and Richmond 2004; Samyn 2003). Most genera (and subgenera) of Aspidochirotida are represented with the exception of *Bohadschia* and *Thelenota*. There were also some unusual observations such as “burrowing” *Holothuria arenacava* (Samyn et al. 2001), which was previously only known from coral communities in East Africa. The burrowing habits of this species in sediment, and its superficial resemblance with the abundant *H. leucospilota*, makes it considerably more difficult to observe in the field and probably strongly underestimated in the records elsewhere in the western Indian Ocean. In terms of exploitation, only

H. scabra and, to a lesser extent, *H. atra* (but in the same area and as substitute for *H. scabra*) are reportedly exploited within the country (Al-Rashdi et al. 2007a). *H. scabra* has, unfortunately, nearly disappeared from its very limited range after only a few years of uncontrolled exploitation (Al-Rashdi and Claereboudt 2010), thereby emphasizing the need for stricter management of the sea cucumber fishery.

The most striking result of this survey is the large difference in sea cucumber community structure between the Sea of Oman and the Arabian Sea (Fig. 4). This difference is not surprising given the strong seasonal effect of the monsoon on coastal habitats in the Arabian Sea (Barratt 1984).

Similar observations were made of other groups of marine organisms. Scleractinian corals, for instance, are also impoverished from the Arabian Sea to the Sea of Oman and the Arabian Gulf (Sheppard 1998), and macroalgae have a very similar distribution split across the geographic limits between the Arabian Sea and the Sea of Oman at Ras-Al-Hadd (Schils and Wilson 2006). Two of the sea cucumbers photographed and collected during the survey did not correspond to known shallow-water western Indian Ocean species (Figs 2B and 2D). These may correspond to new species (taxonomic and genetic studies are underway). Although very small in size (only 600 km of coastline), and only superficially surveyed, southern Oman is rich in endemic species such as sea stars (e.g. *Ferdina sadhensis* and *Patriella paradoxa*), the clownfish *Amphiprion omanensis* (Randall 1995), the abalone *Haliotis mariae* (Bosch et al. 1995), the parrotfish *Scarus zhoifar* (Randall 1995) and the coral *Porites decasepta* (Claereboudt 2006).

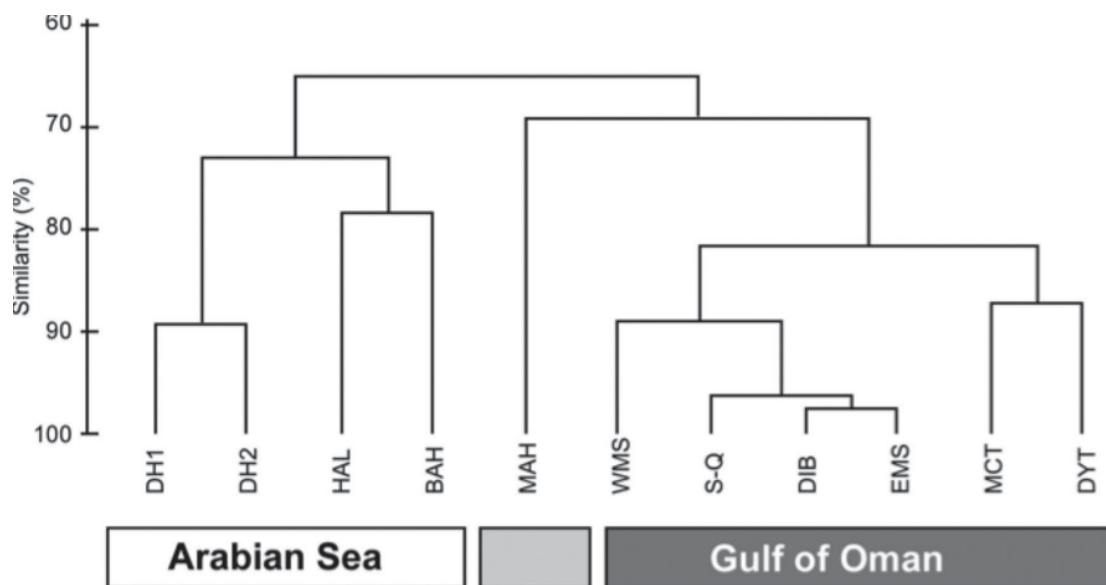


Figure 4. Cluster analysis dendrogram of sea cucumber communities along the coast of Oman. Similarity was measured using the Bray-Curtis index.

While endemic, these species are nearly all relatively common in their limited area of distribution.

The outlier community at Mahout Island (Fig. 1; Mah) is most likely the reflection of a change in fauna due to extensive shallow seagrass beds found near the island and the more limited coral reef development in the area. It is the only place where *Holothuria scabra* has been found in the whole survey and also the only population of sea cucumber in Oman supporting an artisanal fishery. Although, other species of high value (*H. nobilis*) or intermediate commercial value (*Stichopus variegatus*) were recorded, there appeared to be no exploitation of these species. *H. nobilis* is so rare that it could not support any exploitation, while *S. variegatus* on the other hand is relatively common in both the Sea of Oman and the Arabian Sea and could support a fishery but only under a very strict management plan.

Acknowledgements

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First observation of a large group of *Holothuria leucospilota* sea cucumber juveniles at a nursery in Manado (north Sulawesi, Indonesia)

Coralie Taquet,^{1*} Fakhrizal Setiawan,² Nina Yasuda,³ Suharsono⁴ and Kazuo Nadaoka¹

Introduction

The white thread-fish sea cucumber, *Holothuria (Mertensiothuria) leucospilota*, is a tropical holothurian species that is widely distributed in shallow reef areas (e.g. reef flats, shallow costal lagoons, seagrass beds) of the tropical and sub-tropical Indo-Pacific region, including the Red Sea (Conand 1998; Samyn et al. 2006; Conand 2008). Like abalone shells or shark fins, holothurian species constitute both a traditional commodity and a potentially significant commercial resource for coastal populations. In addition to their commercial value as food, holothurian species may also be significant because of their biotechnological properties (Xing and Chia 2000; Tamori et al. 2006; Han et al. 2007; Lawrence et al. 2010).

There is global concern regarding overfishing of *H. leucospilota*. *H. leucospilota* is one of about 60 species of sea cucumber (both temperate and tropical species) that constitute a major part of worldwide holothurian fisheries (Lovatelli et al. 2004; Purcell et al. 2010; Uthicke et al. 2010). In this regard, sea cucumber conservation is a primary concern. As with some other aspidochirots, *H. leucospilota* shows two distinct reproduction modes: 1) sexual reproduction, characterized by dioecism, external fertilization, and a planktonic larval stage (Purwati and Luong-van 2003; Drumm and Loneragan 2005; Gaudron et al. 2008; Kohler et al. 2009); and 2) asexual reproduction by transverse fission (Conand et al. 1997; Purwati 2004; Purwati and Dwiono 2005), which has generated studies on its use in aquaculture and for re-stocking purposes (Purwati and Dwiono 2005).

The diversity of coral reef ecosystems in Southeast Asia and the western Pacific (SEA-WP) presents a strong interest for both global biodiversity conservation and local (human) community development. However, coral reef ecosystems in this region are

deeply threatened due to various factors, including human impacts on the environment and global climate change. Coral reefs of distinct islands are sometimes considered to be independent entities, especially due to the limited movement of adult marine organisms. However, the life history of many marine species, such as holothurians, includes a pelagic larval phase, and so there is an exchange of organisms and species between island ecosystems. Therefore, even within an island-studded region, a global management plan is necessary. Such a global regional management plan within the SEA-WP region, however, requires international coordination.

The SEA-WP Connectivity Project, led by Kazuo Nadaoka (Taquet et al. 2009) aims to provide useful information concerning larval dispersal patterns and reef connectivity, thereby contributing to the identification of important candidate areas for marine protected areas (MPAs). In this study, we used two different approaches: numerical simulation of larval dispersal and a population genetics study. The second approach required extensive tissue sample collection in order to properly infer genetic connectivity in the SEA-WP region. We conducted a sampling campaign in several sites of Indonesia, which allowed us to make other observations. In this paper, we describe a large aggregation of *H. leucospilota* juveniles and some potential implications.

Results and discussion

We explored the general area around Manado (north Sulawesi) in May 2010, and visited Pantai Kalinaun (Fig. 1) on the eastern side of Manado peninsula, north of Bitung. At this site, we observed a large number of very small black sea cucumbers in a seagrass bed (main species *Thalassia* sp.) on a sandy substrate with coral rubble (Fig. 2B and D). Except for their body size, the morphology corresponded

1. Department of Mechanical and Environmental Informatics, Graduate School of Information Science and Engineering, Tokyo Institute of Technology. Email: coralie.taquet@gmail.com.

2. Wildlife Conservation Society (WCS), Indonesia Marine Program, Jl. Toar No. 20 Kec. Wenang, Kel. Bumi Beringin, Kota Manado 95113, Sulawesi Utara, Indonesia.

3. National Research Institute of Fisheries and Environment of Inland Sea, Maruishi 2-17-5, Hatsukaichi, Hiroshima 739-0452, Japan.

4. Research Center for Oceanography, Indonesian Institute of Sciences (LIPI), Jl. Pasir Putih I, Ancol Timur, Jakarta 11048, Indonesia.

* Corresponding author: C. Taquet; Tel: (+81)3-5734-2949 Fax: (+81)3-5734-2650; Email: coralie.taquet@gmail.com



Figure 1. Pantai Kalinaun ($1^{\circ}38'0.04''\text{N}$, $125^{\circ}9'2.62''\text{E}$), Manado area, north Sulawesi, Indonesia (source: ©Google Earth).

exactly with *Holothuria leucospilota* (Conand 1998; Samyn et al. 2006; Conand 2008): long black body, lightly pear shape when contracted, very fine and smooth tegument, Cuvierian tubules. They also presented some characteristics (e.g. reaction and texture to the touch) that are typically found in adult *H. leucospilota* at other sites (Fig. 3). This lead us to consider that those individuals were juvenile *H. leucospilota* (Fig. 2A and C). Indeed, while the mean

body length of adults is about 35 cm (Conand 1998; Kohler et al. 2009), all of the individuals observed in Pantai Kalinaun had a curved body length between 1.5 cm and 3.5 cm, with an average length of 2.37 cm (based on a sample of 35 individuals captured from seagrass bed, Fig. 4). Such length values are consistent with previous observations of *H. leucospilota* juveniles (Shiell 2004). In addition, the length-to-width ratio was similar to that of adults, and the



Figure 2. *Holothuria leucospilota* juveniles observed in Pantai Kalinaun, Manado area, north Sulawesi, Indonesia (Photos: Coralie Taquet).

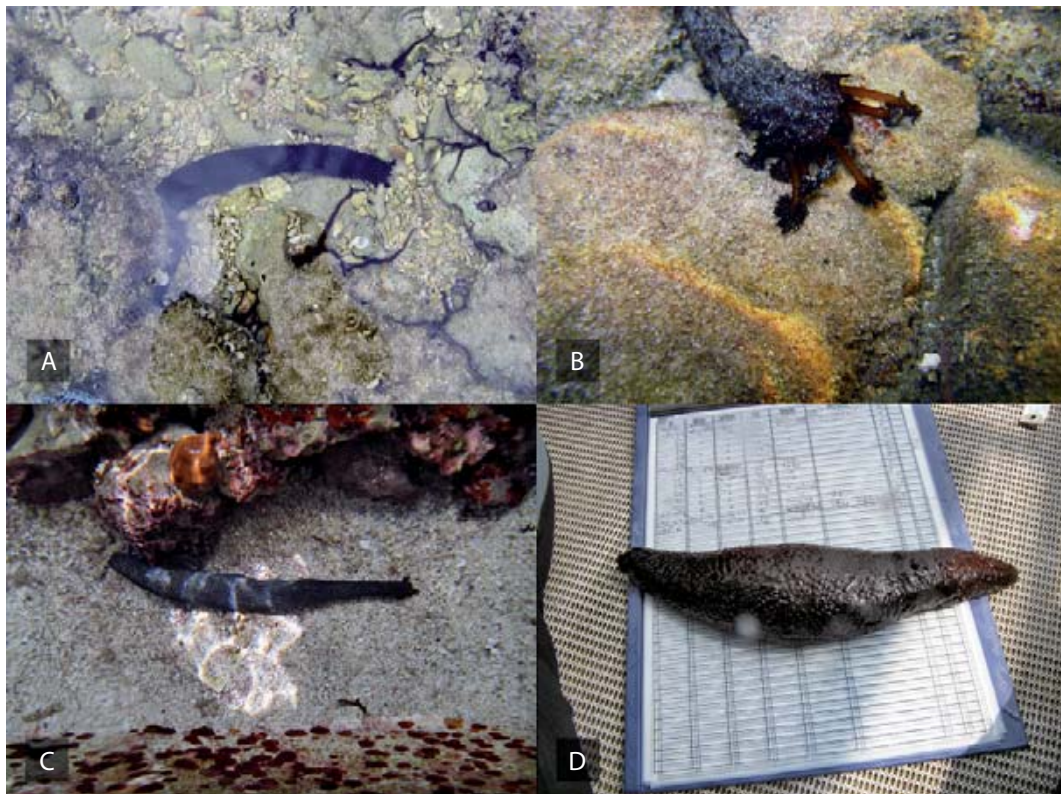


Figure 3. Adult *Holothuria leucospilota* (Photos: Coralie TAQUET): Photos A, B and D are from Kenting Marine Park (Taiwan); and photo B is from Vairao, Tahiti (French Polynesia).

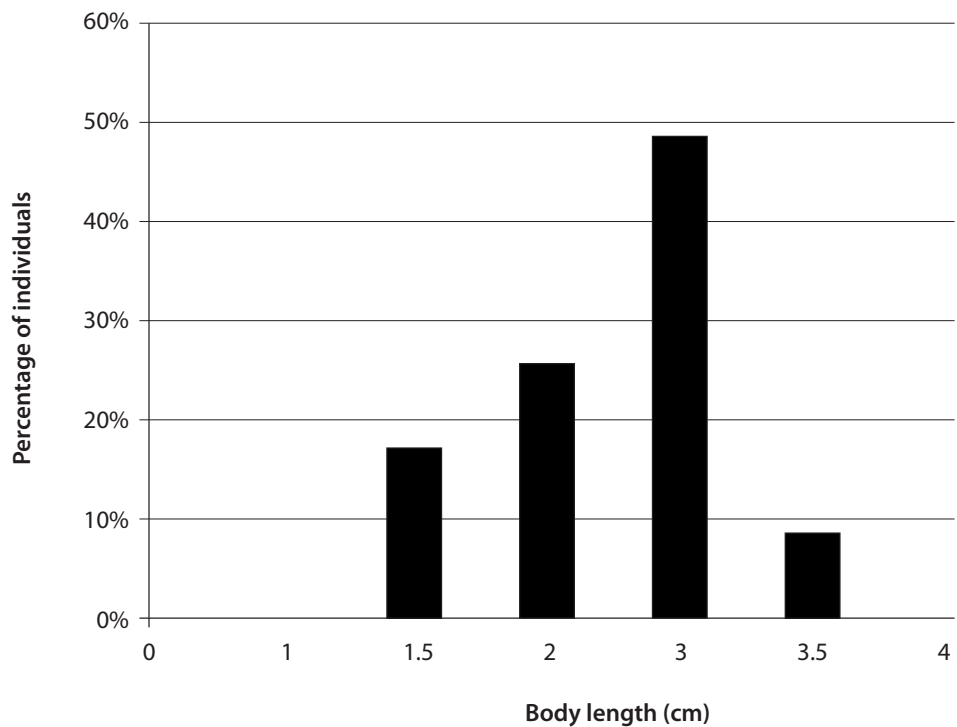


Figure 4. Body length distribution of 35 juvenile *H. leucospilota* in the nursery at Pantai Kalinaun (Manado, north Sulawesi, Indonesia).

absence of a fission “scar” at either end of the body indicates that they were sexually reproduced rather than asexually produced individuals.

Contrary to most of the previous observations of holothurian juveniles (Shiell 2004), we did not observe any adults nearby in Pantai Kalinaun. The holothurian fauna in that small area (approximately 50 m x 40 m) was comprised almost entirely of *H. leucospilota* juveniles. We estimated the mean density of juveniles inside this area to be about 100 ind m⁻². Accordingly, a rough estimate of the total number of juveniles would be 200,000 individuals. This constitutes a huge number given the scarcity of juveniles that have been observed (Purwati and Luong-van 2003) for this species (reputed to reproduce asexually), but shows evidence of spawning in various sites (Purwati and Luong-van 2003; Purwati 2004; Drumm and Loneragan 2005; Gaudron et al. 2008; Kohler et al. 2009). Considering the relatively low number of juveniles observed in some other sites (Shiell 2004) in contrast to the high concentration of juveniles in Pantai Kalinaun, we consider this aggregation to be a real “nursery”.

This observation occurred on 15 May 2010 during a spring flood tide, between 15:00 and 17:00. Depth was about 50 cm. Even with the scant information on larval duration and growth in the wild, we hypothesize that these juveniles could have been produced from the last spawning period in January–April (Purwati and Luong-van 2003; Drumm and Loneragan 2005; Gaudron et al. 2008). Concerning the geographical origin of these new recruits, several hypotheses could be considered due to the complex oceanic currents around Indonesia (Sprintall et al. 2004). On one hand, it is possible that these recruits simply come from nearby sites because we observed adult *H. leucospilota* throughout the Manado area. On the other hand, recruits could come from Mindanao Island (Philippines), north of Kalimantan (Indonesia), or north of Sulawesi (Indonesia) from Indonesian Through Flow and Celebes Sea eddy system (Aditya R. Kartadikaria, PhD student, Tokyo Institute of Technology, pers. comm.). This matter may be addressed by genetic analyses of recruits, and more particularly a comparison between individuals from other spots in the Manado area, but also from elsewhere in Indonesia (e.g. Derawan, Bontang) and the Philippines (Taquet et al. in prep.).

Conclusion

The study of a large nursery of *H. leucospilota* could provide useful information concerning the sexual reproduction of this species, which plays an essential part in maintaining genetic diversity. Such a study could also allow an assessment of factors

that influence settlement and survivorship of holothurian juveniles. This last point is also significant in the context of global holothurian overfishing, because it could facilitate re-stocking through the release of hatchery-produced juveniles in the wild and/or MPA networks.

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Breeding and larval rearing of the sea cucumber *Holothuria leucospilota* Brandt (*Holothuria vegabunda* Selenka) from the northern Persian Gulf, Iran

Abdul-Reza Dabbagh,^{1*} Mohammad Reza Sedaghat,² Hussein Rameshi² and Ehsan Kamrani¹

Abstract

Holothuria leucospilota has been induced to spawn by combining two methods: water pressure and thermal stimulation. Larvae were fed using unicellular algae and *Sargassum* extract. Early juveniles were obtained on day 33. Survival rate was 4.2% to the juvenile stage.

Introduction

In a monograph on echinoderms in the “Fauna of Iran” series, Heding (1940) recorded 17 species of holothurians found in the waters around Iran (Table 1). Among commercial species *H. leucospilota* has a low value (Toral-Granda 2006). This species usually spawns over several months (Jayasree and Bhavana-bayana 1994; Drumm and Loneragan 2005).

In Iran, holothurians are only co-cultured with shrimps, as Amini Rad (2004) has shown a positive synergism between shrimp and sea cucumbers. There have been no studies to date on the sustainable harvesting and propagating of commercial sea cucumbers in Iran; this work attempts to apply methods used elsewhere in the breeding of sandfish (*H. scabra*) to *H. leucospilota*. In this paper we present the first report of successful *H. leucospilota* larval development in Iran.

Material and methods

H. leucospilota for broodstock were collected by snorkeling in depths of 0.5–1.5 m at low tide along the northern Persian Gulf coastal waters of Bandar-e Bostaneh during the summer of 2009. The broodstock was transferred by car to the Persian Gulf Mollusc Research Station at Bandar-e Lengeh, Hormozgan Province, Iran. The work was conducted following the methods outlined in Agudo 2006. Ten sea cucumbers were maintained in a 500-L tank with an 8-cm layer of sand on the floor. The tank was filled with 1-mm filtered and UV-sterilised seawater that was changed daily.

Table 1. Seventeen species of holothurians recorded from the Persian Gulf.

| Species | Commercial value |
|--|------------------|
| 1 <i>Aphelodactyla iranica</i> Heding | |
| 2 <i>Colochirus loppenthini</i> Heding | |
| 3 <i>Halodeima atra</i> Jager | Low |
| 4 <i>H. impatientis</i> Forskal | Low |
| 5 <i>H. monacaria</i> Lesson | |
| 6 <i>H. ocellata</i> Jager | |
| 7 <i>H. paradalis</i> Selenka | Low |
| 8 <i>H. parva</i> Lampert | Low |
| 9 <i>H. spinifera</i> Theel | |
| 10 <i>H. leucospilota</i> Selenka | Low |
| 11 <i>Protankyra magnihamulae</i> Heding | |
| 12 <i>P. pseudo-digitata</i> Semper | |
| 13 <i>Stichopus variegatus</i> Semper | Medium |
| 14 <i>Stolus sacellus</i> Selenka | |
| 15 <i>Thorsonia fusiformis</i> Heding | |
| 16 <i>Thyone festina</i> Koehler and Vaney | |
| 17 <i>T. dura</i> Koehler and Vaney | |

The methods used for inducing spawning in *H. leucospilota* were:

- Heat shock treatment. Water temperature in the spawning tank was raised by 5°C (by adding warmed seawater) and broodstock held under these conditions for 1 hour.
- Combination of water pressure and heat shock treatment. Animals were left in the tank at a

1. Department of Marine and Fisheries Biology, Hormozgan University, PO Box 3995, Bandar Abbas, Iran.

2. Persian Gulf Mollusc Research Station, Bandar-e Lengeh, Iran.

* Corresponding author: Rezadabbagh@yahoo.com

depth of 2 cm for 40 minutes, and then subjected to a powerful jet of seawater for 20 minutes. Subsequently, the water temperature was raised by 5°C for 1 hour. A cover was placed on the broodstock tank and spawning later occurred. After the females spawned, the broodstock tank was left for 1 hour to allow the eggs to be fertilized by sperm from the males.

Spawned eggs were gently siphoned onto an 80-mm sieve into the bucket. The collected eggs were transferred carefully into clean 10-L buckets. To estimate egg density, the water in the buckets was gently stirred to distribute the eggs uniformly. Three 1-mL subsamples were taken from the bucket and eggs were counted under a microscope using a counting cell. Finally, the average density was calculated. The hatching rate was later estimated from the number of early auricularia divided by the number of eggs.

Larvae were stocked in a 300-L tank filled with 1-mm filtered and UV-sterilised seawater (29°C and 40 ppt salinity) at a density of 0.15 larvae mL⁻¹. Feeding was started on day 3 with *Isochrysis* sp. Subsequently, a mixture of algae (*Chaetoceros muelleri*, *C. calcitrans*, *Tetraselmis* sp.) was fed to a density that was gradually increased from 20,000 to 40,000 cells mL⁻¹. The water in the larval tank was changed, performing a complete water change every second day by siphoning the water through a sieve in a bucket (Fig. 1). Growth and survival data were recorded every second day, when siphoning the larvae onto the sieve finished, by taking three 1-mL subsamples.



Figure 1. Sieving the tank water through a filter.

Larvae were reared to the late auricularia stage using unicellular microalgae. When doliolaria were observed, fibreglass settlement plates coated with *Sargassum* extract were prepared, to be used

to induce doliolaria to metamorphose into settled pentactula larvae. *Sargassum* sp. extract was added daily to water in which the plates were immersed (in a separate tank) so that the plates became coated with a fine layer of algal material. These settlement plates were then placed into the larval tank (Fig. 2). After two days, none of the (non-feeding) doliolaria larvae were seen on a sieve during a complete water change. Within a few days, pentactula larvae were formed. At this stage, the larvae were fed daily with *Sargassum* extract.



Figure 2. Settlement plates in the larval tank.

Results

Our specimens were only induced to spawn using the combined treatment of water pressure and heat shock treatment. After 1 hour, the males released their sperm; subsequently, 1 female spawned after 10 minutes, releasing about 45,000 eggs. However, 35,000 early auricularia larvae (Fig. 3) were obtained. The hatching rate was 77.7%. The amount of time for larvae to reach the juvenile stage is shown in Table 2.

Table 2. Development of *Holothuria leucospilota* from fertilization to 1mm juveniles.

| Stage | Time |
|--------------------------------------|---------|
| Fertilisation | 0 |
| Late gastrula | 3d |
| Early auricularia | 4–11 d |
| Mid auricularia | 12–14 d |
| Late auricularia | 17–21 d |
| Doliolaria | 22–27 d |
| Early pentactula | na |
| Settlement (metamorphosis completed) | na |
| Juvenile, 1mm (Fig. 4) | 33 d |

na = data not available



Figure 3. *Holothuria leucospilota* early auricularia larvae.

After 17–21 days of stocking, late auricularia larvae (Fig. 4) were observed. The main features of this stage are that it is transparent, slipper-shaped with ciliated bands, and up to 10 hyaline spheres. Average larval length averaged 1100 μ m and sphere diameter 85 μ m.



Figure 4. *Holothuria leucospilota* late auricularia larvae.

Auricularia larvae metamorphosed to (non-feeding, pelagic) doliolaria on day 22. Doliolaria larvae were dark-brown, barrel-shaped larvae with five ciliated bands around the body. Length and hyaline sphere diameter averaged 590 μ m and 69 μ m, respectively.

Discussion

Spawning and larval rearing have only been achieved for some species of sea cucumbers (Laxminarayana 2005). Laxminarayana has also reviewed *Apostichopus japonicus*, for which juveniles were produced for over 60 years ago in Japan and China. Nowadays, the focus of studies has been mainly on the breeding and rearing of commercial sea

cucumber species. The development of three commercial sea cucumbers, *Holothuria scabra*, *H. fuscogilva* and *Actinopyga mauritiana* was studied by Ramofafia et al. (2003). Hamel et al. (2003) reported larval development of *Isostichopus fuscus*. James (2004) bred *H. scabra* in India. Spawning and larval rearing of *H. atra* were also reported from Mauritius (Laxminarayana 2005). Thermal stimulation methods were used to obtain fertilized eggs in *H. atra* (Laxminarayana 2005) and in *H. scabra* (James 2004; Ivy and Giraspy 2006).

There have been no documented cases so far on the spawning and larval rearing of the sea cucumber *H. leucospilota*. A diet including *Rhodomonas salina*, *Chaetoceros calcitrans*, *C. mulleri*, *Tetraselmis chui*, *Isochrysis galbana* and *Pavlova lutheri* was used in rearing *H. scabra* in Australia (Ivy and Giraspy 2006). Laxminarayana (2005) fed *H. atra* with unicellular algae (such as *I. galbana* and *C. calcitrans*), algal extract and artificial feeds. Xiyin et al. (2004) used polyethylene film sheets for settling doliolaria larvae of *Apostichopus japonicus*. Rough surface tiles and hard surfaces with available food were used to induce doliolaria larvae to metamorphose into pentactularia (James 2004).

In general, Laxminarayana (2005) reported that *H. atra* larvae that were reared at a salinity of 34–36 ppt reached the pentacula stage by day 20. In the present study, with larvae reared at 40‰, doliolaria larvae were only obtained by day 22. The low growth rate was probably due to high salinity conditions, which prevail in the Persian Gulf. Because no previous rearing work has been carried out on sea cucumbers here, we encountered some problems, such as preparing settlement plates with *Sargassum* extract and determining the density of the *Sargassum* extract. Also in this study, the pentacula larvae were found to be orange. Consequently, it was difficult to distinguish pentacula larvae from juveniles on the plates. In the future, it is hoped to use the experience gained from this study as a stepping stone towards culturing commercial species.

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Abundance, distribution and some biological aspects of *Holothuria edulis* off the northwest coast of Sri Lanka

D.C.T. Dissanayake^{1*} and Sujeewa Athukorala¹

Abstract

The stock status of *Holothuria edulis* off the northwest coast of Sri Lanka was estimated by surveying an area of 1,779 km² (using an underwater visual census technique) in October 2009. Five hundred sampling sites were randomly selected and surveyed. The estimated mean density was 122 ± 365 ind ha⁻¹ while the standing stock biomass was 1,724 t. Uneven distribution of *H. edulis* was observed within survey sites and *H. edulis* was reported in most habitats, including seagrass beds, coral reefs, open sandy bottom areas and macroalgae-rich areas. The population was dominated by a 16.5 cm (mid-length) length class, and asexual reproduction was promising during the survey time. The findings of this study can be used to develop a proper management plan for the sustainable exploitation of this resource in the future.

Introduction

The sea cucumber fishery was introduced to Sri Lanka by the Chinese in late 19th century (Hornell 1917). Even today, sea cucumbers are of interest in coastal, multi-species fisheries throughout the coastal areas of Sri Lanka. Fishing activities, however, are predominant in the northern, eastern and northwestern parts of the island. Fishing is greatly influenced by monsoons; in the east and the north, fishing occurs during the southwest monsoon season (May to September), and in the northwest, fishing occurs during the northeast monsoon season (October to April). Sea cucumbers are mainly harvested by hand either by skin diving or scuba diving with the aid of fiberglass boats powered by 15–25 hp outboard motors (Dissanayake and Wijayarathne 2007).

About 24 sea cucumber species were reported from the coastal waters of Sri Lanka, and 20 species are considered to be commercially important (Dissanayake et al. 2010). *Holothuria edulis* is the most abundant and widely distributed sea cucumber

species around Sri Lanka, and *H. atra* is the second most dominant species (Dissanayake and Athukorala 2009, Dissanayake and Stefansson, 2010). Although *H. edulis* is not frequently exploited on a commercial scale, light exploitation has been carried out from time to time depending on market demand, especially by the aquarium industry (Dissanayake and Athukorala 2009).

According to recent studies, most of Sri Lankan sea cucumbers belonging to the high- and medium-value categories are showing signs of over exploitation, and low-value species are becoming dominant in commercial landings (Dissanayake and Athukorala 2009). As such, there is a potential to exploit *H. edulis* on a large scale from the coastal waters of Sri Lanka as practiced in other Asian countries.

The present study aims to assess the stock status of *H. edulis* off the northwestern coast of Sri Lanka, giving special attention to some biological aspects, including length-weight distribution and reproductive biology.

Materials and method

An underwater visual census was carried out off the northwest coast of Sri Lanka in October 2009.

The study area extended from Mampuri to Vankalai (see Fig. 1). Randomly selected five hundred sampling sites were surveyed within the study area, bounded by Puttlam and Mannar fisheries districts (Fig. 1). The study was confined to water depths of up to 30 m, and the survey area was 1779 km². The survey was carried out at the beginning of fishing season by temporarily closing the commercial fishing activities and the survey period was four weeks. The survey used rapid marine assessment technique that have been employed in sea cucumber surveys in Australia's Torres Strait (Long et al. 1996) and Moreton Bay (Skewes et al. 2002), the Timor MOU Box (Skewes et al. 1999), Papua New Guinea's Milne Bay Province (Skewes et al. 2002) and the Seychelles (Aumeeruddy et al. 2005). Fieldwork was

1. National Aquatic Resources Research and Development Agency (NARA), Sri Lanka.

* Email: chami_dt@yahoo.com

undertaken by a team of divers operating from small boats. Sample sites were located by using a portable global positioning system (GPS). At each site, a diver (or divers) swam along a 100-m transect collecting sea cucumbers and reporting habitat information on a 1-m swath on either side of the transect line. At each site, the substrate was described in terms of the percentage cover of sand, rubble, limestone platform, coral or terrestrial rock and mud. The percentage cover of other conspicuous biota (e.g. seagrass and algae) was also recorded.

H. edulis collected during the survey were brought to the base station where they were individually weighed and measured for total length. Information on maturity stages and reproductive pattern was also collected. Population density, total abundance and standing stock biomass were calculated and mapped.

Results

Abundance and distribution

H. edulis was observed in 23% of sampling sites with an average density (\pm SD) of 122 ± 365 ind ha^{-1} off the northwest coast of Sri Lanka. Total abundance

was 22×10^6 (individuals) while total biomass was 1,724 t.

The distribution of *H. edulis* was highly patchy and the highest densities were recorded close to the lagoon mouth and the upper part of the survey area (Fig. 2). *H. edulis* was found in most habitat types, including seagrass beds (Fig. 3A), coral reefs (Fig. 3B), open sandy bottom areas (Fig. 3C), and macroalgae-rich areas (Fig. 3D). In some areas, *H. edulis* could be found together with *H. atra* (Fig. 3D).

Length-weight frequency distribution

The length and weight frequency distribution of *H. edulis* is summarised in Figure 4. The length of *H. edulis* ranged from 4.5–40.5 cm, and the most frequent length category was 16.5 cm followed by 13.5 cm. The mean length of *H. edulis* was 16.4 ± 4.4 cm and the length frequency distribution was unimodal.

The total weight of *H. edulis* ranged between 10 g and 400 g, and the highest frequency was observed in the 50 g weight category. The mean weight (total) of the northwest population of *H. edulis* was 78.0 ± 15.2 g in 2009.

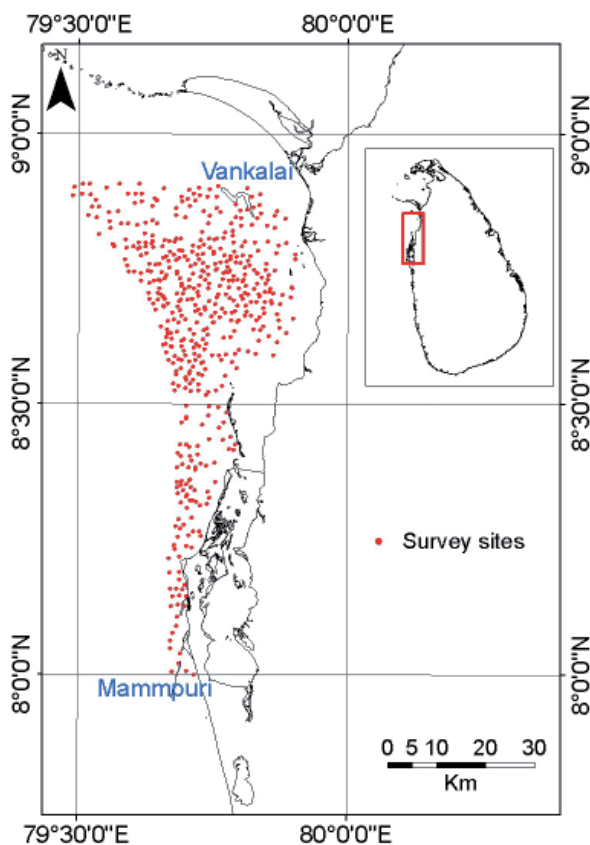


Figure 1. Sea cucumber survey sites off the northwestern coast of Sri Lanka.

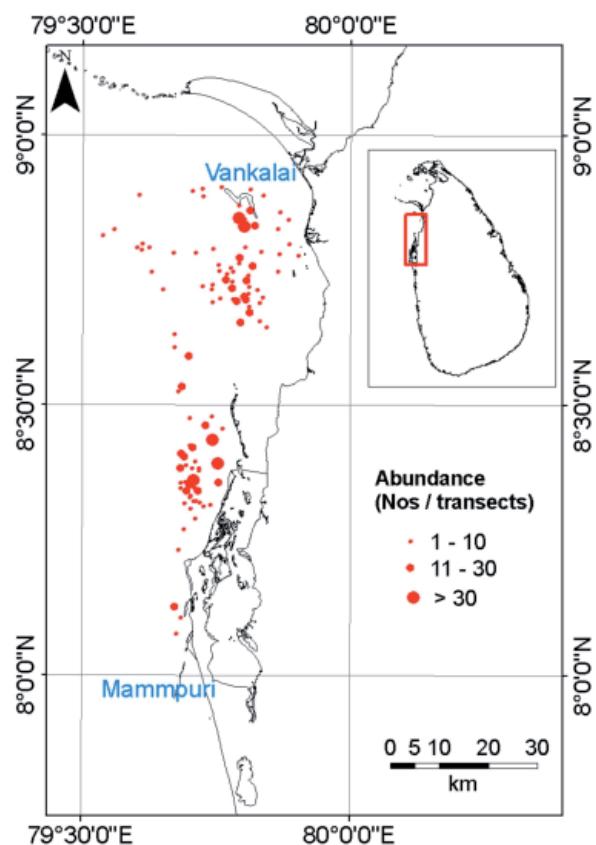


Figure 2. Abundance and distribution of *H. edulis* off the northwestern coast of Sri Lanka in 2009.



Figure 3. Habitat types within survey areas.

- A: *H. edulis* collected from a seagrass bed.
- B: *H. edulis* in a coral associated area.
- C: *H. edulis* with *H. atra* in an open sandy bottom area.
- D: *H. edulis* in macroalgae-rich habitat.

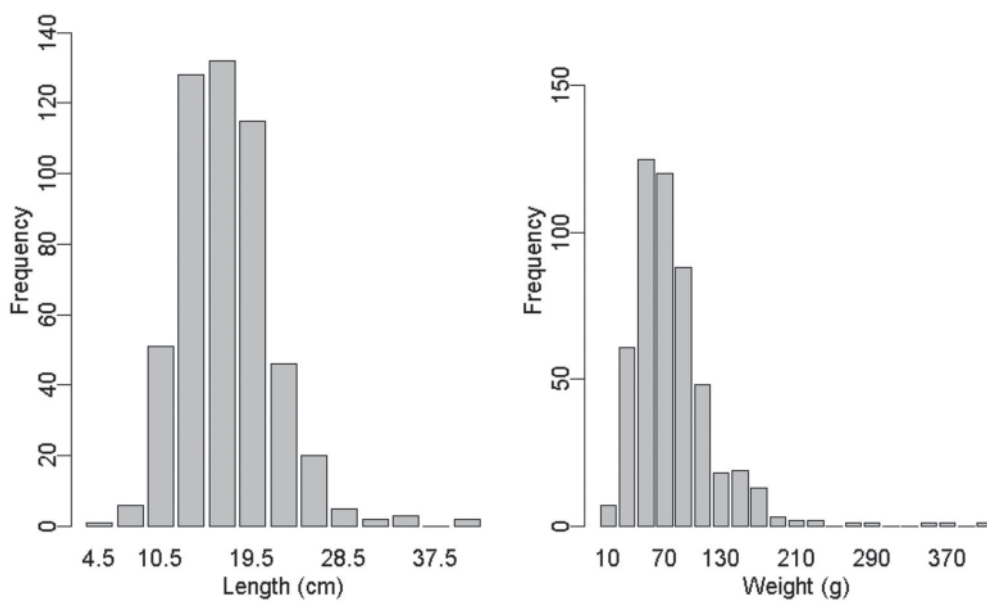


Figure 4. Length and weight (total) frequency distribution of *H. edulis* off the northwestern coast of Sri Lanka.

Reproduction

About 256 individuals of *H. edulis* collected from the survey were dissected in order to study the maturity stages (Fig. 5), although none had mature or developed gonads. However, 23.4% of individuals showed signs of asexual reproduction where the body had divided into two or more parts (Fig. 6).



Figure 5. Dissecting *H. edulis* to study the sexual maturity stages.

Discussion

H. atra is the most common and most abundant sea cucumber species in most parts of the Indian Ocean (Conand and Muthiga 2007), including Mayotte (Pouget 2005) and Reunion Island (Conand and Mangion 2002), and another survey has found that *H. edulis* is the most abundant species in Sri Lanka (Dissanayake and Athukorala 2009). Although *H. edulis* is considered to be a commercial species, it is rarely and seasonally exploited off the coastal waters of Sri Lanka, and this is probably a reason for its higher abundance. The abundance and distribution of *H. edulis* was quite uneven among survey sites, and according to Conand and Muthiga (2007) this is a common phenomenon in other areas of the Indo-Pacific region for all sea cucumber species. The observed discrepancy among survey sites could be linked with the habitat preference of *H. edulis*. In this survey, *H. edulis* was found in different habitat types and was very common in seagrass and coral reef areas. A higher abundance of sea cucumbers in coral reefs and seagrass beds has been discussed by various authors highlighting the importance of these habitats for the protection, sheltering and feeding of sea cucumbers (Sloan and Bodungun 1980; Conand 1990; Conand 2008).

The length distribution pattern of *H. edulis* was unimodal and showed a similar pattern of length-frequency distribution to other holothurian species, including *H. scabra* (Kithakeni and Ndaro



Figure 6. Asexual reproduction (fission) of *H. edulis*.

2002). The results revealed that *H. edulis* can grow up to 41 cm but the population was dominated by individuals that were 16.5 cm (mid-length). These differences in size may be a function of fishing pressure, survey sampling depth, environmental factors and substrate type (Mercier et al. 1999).

Several studies have examined the asexual reproduction of sea cucumbers by fission (Emson and Wilkie 1980; Emson and Maldenov 1987; Boyer et al. 1995; Reichenbach et al. 1996; Uthicke 2001; Conand and Uthicke 2001; Howaida et al. 2004; Conand 1993, 2004; Laxminarayana 2006). Asexual reproduction of sea cucumbers in nature is a very common and seasonal event. According to Uthicke (2001), most holothurian species that reproduce asexually do so by “twisting and stretching” in such a way that the anterior and posterior sections rotate in opposite directions, resulting in a constriction in the holothurian. In the second step, the two halves slowly move in opposite directions until the body wall tears at the constriction point, and the two halves become completely separated. Asexual reproduction of *H. edulis* by transverse fission has been observed by Uthike (1997, 1998, 2001) on the Great Barrier Reef, by Harriott (1980) at Heron Island, and by this study. However, due to a lack of time series data it was not possible to make any conclusion about the sexual reproduction pattern of this species. The absence of mature or developed gonads could be due to the seasonality of sexual reproduction or the species’ exclusively asexual mode of reproduction. However, further studies are needed to confirm these hypotheses.

With the increasing demand for beche-de-mer from the Asian market, there is a possibility of exploiting *H. edulis* on a large scale in the near future. Therefore, the findings of this study can be used to develop a proper plan for sustainably exploiting this resource in the coastal waters of Sri Lanka.

Acknowledgements

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Some insights on the phylogeny of Algerian shallow-water sea cucumber species (Holothuroidea: Aspidochirotida)

Karim Mezali¹

Abstract

Phylogenetic analyses from the fraction of mitochondrial gene (16S mDNA) sequenced for 26 individuals (in general, five species sampled in various shallow-water locations along the Algerian coast) clarified taxonomic uncertainties. This study shows that: 1) the two color morphs of *Holothuria* (*P.*) *sanctori* that have been debated in some of the literature form a well defined clade; 2) *Holothuria* (*H.*) *stellati* whose confusion has always acknowledged, is genetically distinct from the other species; 3) *Holothuria* (*H.*) *tubulosa*, the most common species, and the “best known” species in the Mediterranean Sea, forms a clade with two well separated populations. Few specimens of holothurians analyzed in our collection have given unusual DNA sequences. However, it is clear that one specimen will probably represent either another species previously unknown, or a hybrid between two known species — *H. (R.) polii* and *H. (H.) stellati*.

Introduction

Sea cucumbers (Holothuroidea) in the order Aspidochirotida are a conspicuous and diverse group in marine ecosystems. They inhabit soft sediment and Mediterranean *Posidonia oceanica* meadows (Francour 1990; Coulon and Jangoux 1993; Mezali 2008). They provide important ecosystem services by enhancing nutrient cycling and local productivity in oligotrophic carbonate sediments through their bioturbation and deposit feeding activities (Uthicke 1999). They are also fished for beche-de-mer production (Conand and Byrne 1993; Toral-Granda 2008). Despite being large and often the most dominant mobile invertebrates in Mediterranean shallow-water areas, the taxonomy of many specimens collected from Algerian shallow-water areas is uncertain. This is due to the difficulty in applying traditional taxonomic characters (e.g. body profile, skeleton morphology). We undertook a phylogenetic analysis of sampled holothuroids using sequence data for a mitochondrial gene (16S rDNA). The aims of this study were to provide a systematic revision of the Algerian aspidochirotid holothurians, using molecular phylogenetic systematic methods.

Materials and methods

Sampling

Most holothurians species were collected by scuba diving and skin diving in 2006 across the Bay of Algiers (Tamentefoust) and the Bay of Bou-Ismaïl (Sidi-Fredj) at depths ranging between 1 m and 20 m (Fig. 1). Three additional stations (not shown in Fig. 1) were also explored during 2006 (Stidia and Sidi-Medjdoub in the Bay of Mostaganem and

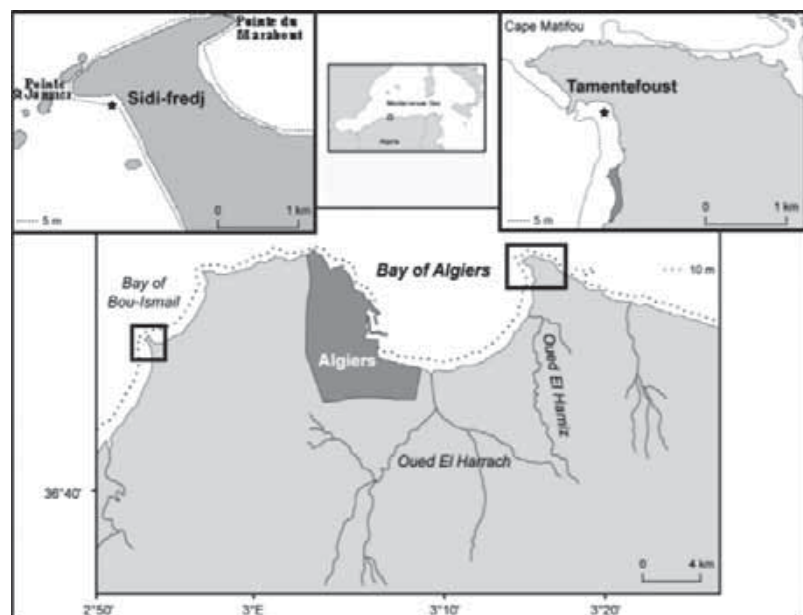


Figure 1. Stations where most of the holothurians were sampled.

1. Département des Ressources Halieutiques, Faculté des Sciences Exactes, de la Nature et de la Vie. Université Abdelhamid Ibn Badis-Mostaganem, BP 300 rue Hocine Hamadou, 27000, Mostaganem, Algérie. Email: mezalikarim@yahoo.fr

Figuiet-plage in the Bay of Zemmouri-Boumerdes). The fresh sampled holothuroids were grouped into 10 morphotypes: 1) The classical *Holothuria (Holothuria) tubulosa A* and 2) *Holothuria (Holothuria) stellati*, characterised by a distinct, round bivium and flat trivium. *Holothuria (H.) stellati* differs from the classical *H. (H.) tubulosa A* by its big protuberances; 3) *Holothuria (Holothuria) tubulosa B* has a flask-shaped integument that is not very thick in a released state; 4) *Holothuria (Holothuria) tubulosa C* has a soft consistency with an arched bivium that has a pointed conical verrucosities; 5) *Holothuria (Holothuria) tubulosa D* has a slightly cylindrical form with a flattened trivium and thick integument. This species presents apparent conical verrucosities laid out in several lines on the bivium; 6) The classical *Holothuria (Roweothuria) poli A* has abundant white pedicels on the trivium; 7) *Holothuria (Roweothuria) poli B* has a similar aspect as *H. (H.) stellati* (large protuberances) and with *H. (R.) poli A* (white pedicels) regularly laid out on the trivium; 8) *Holothuria (Panningothuria) forskali* has a cylindrical soft black body (when it is alive), and a numerous white pedicels on the trivium; 9) *H. (Platyperona) sanctori A* is brown whereas 10) *H. (Platyperona) sanctori B* is easily recognized in water by its white spots. The first seven species described above do not have Cuvierian bodies. Species 8, 9 and 10 have Cuvierian bodies.

DNA extraction, PCR protocols and sequencing

Tissue samples preserved in ethanol (90%) were obtained from the tentacle of each individual. In total, sequence data were generated for 26 individuals (all confused species). Additional sequence data (from the French Mediterranean areas) were obtained from the Florida Museum of Natural History (USA). DNA was extracted from a small piece of macerated tissue (10–20 mg) placed in a 1.5 ml microcentrifuge tube and extracted with 750 μ L of DNAzol and 5 μ L of proteinase K. Sections of the large subunit 16S ribosomal DNA (16S rDNA) genes were amplified using primers 16SA-R (5'-CGCCT-GTTTATCAAAA-CAT-3') and 16SB-R (5'GCCGGTCTGAACT-CAGATCACGT-3') (Palumbi et al. 1991).

PCR amplification was performed in 49 μ L containing: ddH₂O (30.8 μ L); 10X (5 μ L); dNTPs (5 μ L); AR (2 μ L); BR (2 μ L); Taq polymerase (0.2 μ L); MgCl₂ (4 μ L) and 1 μ L template DNA solution. PCR reactions involved denaturation for 60s at 95°C followed by 40 cycles of 30s denaturation at 95°C, 30s annealing at 50°C, and 80s extension at 72°C, and final extension of 10 min. PCR amplicons were purified using PCR DNA Gel Band Purification Kit. All sequencings were done by Interdisciplinary Centre for Biotechnology Research of Florida University (www.biotech.ufl.edu/staff.html). Sequences were aligned using Se-Al 2.011 under default parameters and checked by eye.

Sequence analyses and phylogeny

After trimming some base pairs at the beginning and end of the sequences, sequence size for 16S rDNA was 600 bp. These sequence data used for phylogenetic analyses were treated by Sequencher 4.8 software to make several DNA assemblage sequences relatively short in order to create longer sequences called contigs. Bayesian analyses were performed using MrBayes (v.3.1, Ronquist et al. 2005). Prior to analyses we tested for the most appropriate nucleotide substitution model using Modeltest 3.06 (Posada and Crandall 1998). For the evaluation of the reliability of the reconstituted trees, statistical test was applied using the posterior Bayesian probabilities with GTR (General Time Reversible model). The used out-group sequence was from a species *Cucumaria frondosa*, obtained from Genbank.

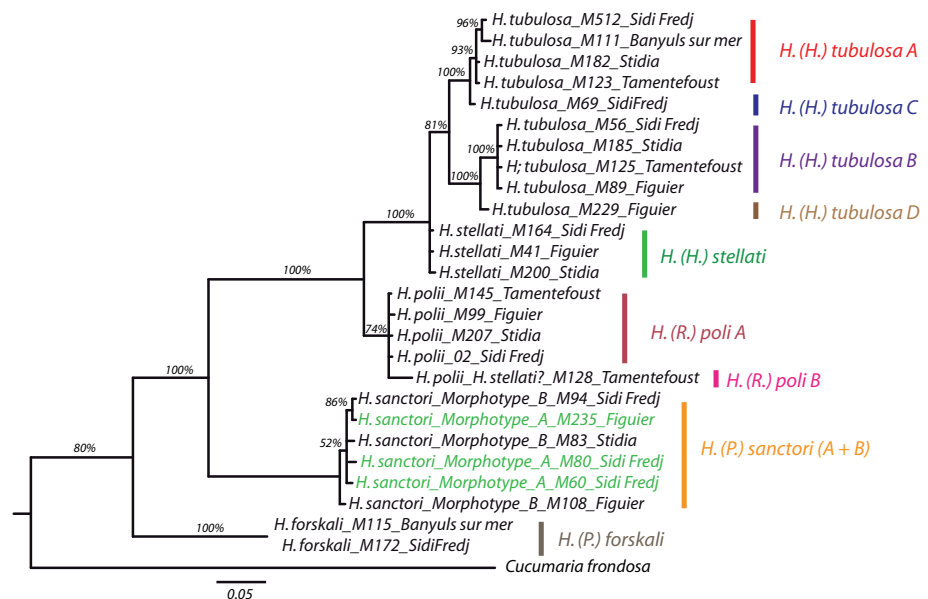


Figure 2. Bayesian consensus tree based on 16S rDNA. The values indicated above the branches represent the posterior probabilities (PP in %).

Results

The Bayesian consensus trees present, in general, five monophyletic clades with high posterior probabilities corresponding to the five examined species (Fig. 2). *Holothuria (H.) tubulosa* is represented by two well supported clades (AC and BD). *Holothuria (H.) stellati* is monophyletic and is well separated from the two groups (A and B) of *H. (H.) tubulosa* (high posterior probabilities 100%). The Bayesian consensus tree shows no differences between the two morphotypes of *Holothuria (P.) sanctori* (A and B). The classical *Holothuria (R.) poli A* is monophyletic and is well separated from the two groups of *H. (H.) tubulosa* (A and B) with high posterior probabilities value (100%). *Holothuria (R.) poli B* included in the clade of the classical *H. (R.) poli A* is slightly different (high posterior probabilities value 74%) and different from the *H. (H.) stellati* (high posterior probabilities value 100%).

Discussion

Phylogenetic analyses using the 16S rDNA provided insights into the relationships within the aspidochirotid holothurians species of Algeria's coastal areas. The most currently recognised species formed separate clades supported by high posterior probabilities values and the species formed clades that agree with taxonomic revisions based on morphology and anatomy (Mezali 2008). The phylogram shows that *H. (H.) tubulosa* have two well separated populations (A and B). *Holothuria (H.) stellati* is well separated from both *H. (H.) tubulosa* (A and B). This molecular results obtained on fresh *H. (H.) stellati* samples contradicts the one obtained by Borrero-Pérez et al. (2009) considering *H. (H.) stellati* to be a junior subjective synonym of *H. tubulosa*. *Holothuria (R.) poli B* occurs in the same clade than the classical *H. (R.) poli A*. *Holothuria (R.) poli B* could be considered as an intermediate form between *H. (H.) stellati* and *H. (R.) poli A* because its morphological characters are common to both species. This led us to suspect that *H. (R.) poli B* is a hybrid species. The taxonomic status of some holothurians specimens analyzed in our collection remains to be confirmed. The two individuals of *H. (H.) tubulosa* (C and D) gave unusual DNA sequences. The use of other molecular markers (i.e. ITS and COI) could be useful to determine in the future the precise taxonomic status of these two specimens.

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Review of saponin diversity in sea cucumbers belonging to the family Holothuriidae

Guillaume Caulier,^{1*} Séverine Van Dyck,¹ Pascal Gerbaux,² Igor Eeckhaut¹ and Patrick Flammang¹

Abstract

Saponins are secondary metabolites produced by holothurians. Structurally, they are triterpene glycosides that play an important role in chemical defense and possess a wide spectrum of pharmacological effects. This review highlights the very high diversity of saponins detected in different species of the family Holothuriidae. No less than 59 triterpene glycosides are reported. Several saponins are shared by many species but others are very specific. Overall, most species appear to possess a specific congener mixture. The most evident inter-specific differences that can be highlighted among Holothuriidae are based on the presence or absence of a sulfate group attached to the carbohydrate chain of their saponins. Within a single animal, saponin mixtures also present different concentrations and compositions depending on the organ, with Cuvierian tubules showing the highest saponin concentrations. All of the data combined indicate a complex chemical defence mechanism with different sets of saponins originating from different body compartments and presenting different properties in relation to their ecological role(s).

Introduction

Saponins are an important class of natural products first discovered in higher plants where they are widely spread (Li et al. 2006). In the search for new pharmacologically active substances, saponins have also been isolated from marine organisms such as holothurians (Nigrelli 1952; Yamanouchi 1955), seastars (Mackie and Turner 1970) and sponges (Thompson et al. 1985). Structurally, holothurian saponins are described as triterpene glycosides composed of an oligosaccharide chain and an aglycone based on holostane-3 β -ol (Fig. 1A) (Kornprobst 2005). Saponins of Holothuriidae (Fig. 1B,C) contain a D⁹⁽¹¹⁾ double bond in the aglycone and the carbohydrate chain encloses up to 6 sugars units, including xylose, glucose, 3-O-methylglucose and quinovose, and can be branched only once (Kalinin et al. 2005). Some of these saponins can be sulfated at the level of the sole xylose (Fig. 1C).

Holothurian triterpene glycosides present a high scientific interest in pharmacology and ecology. Indeed, these secondary metabolites have been reported to possess a wide spectrum of pharmacological effects including hemolytic, antitumoral, anti-inflammatory, antifungal, anti-bacterial, antiviral, ichthyotoxic, cytostatic and antineoplastic activities (Kerr and Chen 1995; Kalinin et al. 1996a, 1996b; Prokofieva et al. 2003). Many of these activities are the result of their tensioactive properties. In

ecology, saponins are deleterious for most organisms and probably function as a chemical defense to deter predation (Kalinin et al. 1996a, b; Van Dyck et al. unpubl. obs.).

Saponin diversity in Holothuriidae

This article reviews the diversity of saponins detected in different species of the family Holothuriidae. Table 1 presents all the saponins extracted and characterized from holothurians of the genera *Actinopyga*, *Bohadschia*, *Holothuria* and *Pearsonothuria* during the last 40 years. Saponins detailed in this table have been purified by different methods including liquid-liquid extractions with different solvents, solid phase extraction or chromatography (silica gel or resins), and high performance liquid chromatography. Mass spectrometry-based techniques and nuclear magnetic resonance combined with chemical reactions and chemical evidence were used to highlight the chemical structure of these saponins. Table 1 emphasizes the very high diversity of saponins in holothuriids. Indeed, no less than 59 triterpene glycosides are reported. When these data are gathered from the literature, some nomenclature problems can be identified. It happens that two names have been given independently to the same molecule. For instance, the structure of nobiliside 2a detailed by Wu et al. (2006c) corresponds exactly to desholothurin A described by Rodriguez et al. (1991). Also, authors should homogenize the

1. University of Mons, Marine Biology Laboratory, 7000 Mons, Belgium.

2. University of Mons, Organic Chemistry Laboratory, Interfaculty Center for Mass Spectrometry (CISMa), 7000 Mons, Belgium.

* Corresponding author: guillaume.caulier@umons.ac.be

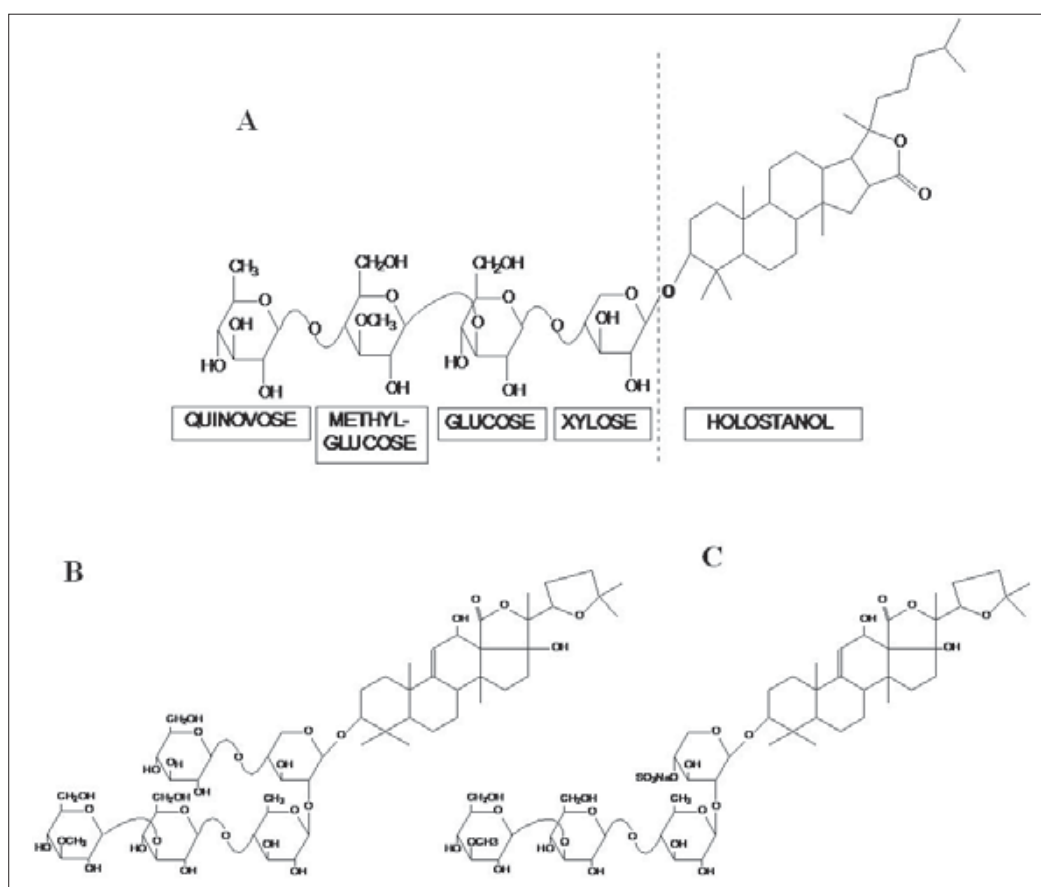


Figure 1. Molecular structure of (A) a hypothetical saponin composed of an aglycone of holostanol (according to the Comparative Toxicogenomic Database) and a linear glycosidic chain constituted by the four most frequent monosaccharides found in holothurian saponins; (B) holothurinoside A, a non-sulfated saponin; and (C) holothurin A, a sulfated saponin.

saponin nomenclature by giving logical names to new molecules, based on the structure of known congeners, rather than names based on the specific origin of the molecules.

The most evident inter-specific differences that can be highlighted among Holothuriidae are based on the presence or absence of sulfate group attached to the carbohydrate chain of their saponins (Kobayashi et al. 1991). The genus *Actinopyga* contains only sulfated saponins (in green in Table 1), the genus *Bohadschia* encloses only non-sulfated ones (in red in Table 1), and the genera *Pearsonothuria* and *Holothuria* present both saponin types. In this last genus, the situation is even more complex with several species containing only sulfated saponins, some others presenting the two types of congeners, and finally one species, *H. forskali*, enclosing exclusively non-sulfated saponin. Several saponins are shared by a lot of species, holothurins A and B for example, but others are very specific like griseaside A or argusides A-E. It must be noted that holothurins A and B were the first to be discovered (Yamanouchi 1955; Kitagawa et al. 1978, 1979) and were subsequently detected in many species of the genus *Holothuria*

(Elyakov et al. 1973, 1975). In the future, one should expect that new studies, using contemporary techniques, will detect additional saponins in these species. Indeed, Table 1 clearly shows that many new congeners have been described only recently. Most species therefore appear to possess a specific congener mixture, a valuable chemotaxonomic character allowing the assignment of a holothuriid species to a specific taxa according to its chemical signature. For example, the taxonomic position of *Bohadschia graeffei* was revised, following the isolation and characterization of its saponins, to a newly established genus *Pearsonothuria graeffei* (Kalinin et al. 2005).

Among the numerous studies dealing with saponins from holothurioids, very few make the distinction between their different body compartments (Table 1) although, within an animal, saponins may present different concentrations and compositions according to the organ considered. Matsuno and Ishida (1969) reported about the distribution of saponins in sea cucumber body compartments. Saponins were found in the digestive organs, the longitudinal retractor muscles, the epidermis, the

Table 1. Saponins of Holothuriidae by species.

| Species | Saponins | BW | CT | References |
|---|--|----|----|-------------------------|
| <i>Actinopyga agassizi</i> | 24-dehydroholothurin A ₃ | x | x | Kitagawa et al. (1982) |
| | Holothurin A | x | x | Kitagawa et al. (1982) |
| | Holothurin A ₃ | | | Kitagawa et al. (1980) |
| | Holothurin B | | | Elyakov et al. (1975) |
| <i>Actinopyga echinites</i> | Holothurin B ₁ | | | Kitagawa et al. (1980) |
| | Fuscocineroside B/C* | x | x | Chapter 2 |
| | Holothurin A | x | x | Elyakov et al. (1973) |
| <i>Actinopyga flammea</i> | Holothurin A ₃ | x | x | Kitagawa et al. (1980) |
| | Holothurin B | | | Elyakov et al. (1973) |
| | Holothurin B ₁ | x | x | Chapter 2 |
| | Holothurin B ₂ | x | x | Chapter 2 |
| | Holothurin B ₃ | x | x | Chapter 2 |
| | Holothurin A | | | Bhatnagar et al. (1985) |
| | Holothurin B | | | Bhatnagar et al. (1985) |
| <i>Actinopyga lecanora</i> | Holothurin A | | | Elyakov et al. (1973) |
| | Holothurin B | | | Elyakov et al. (1973) |
| | Holothurin B ₁ | x | x | Elyakov et al. (1991) |
| <i>Actinopyga mauritana</i> | 24-dehydroholothurin A ₃ | x | x | Kobayashi et al. (1991) |
| | 24-dehydroholothurin B ₁ | x | x | Kobayashi et al. (1991) |
| | Holothurin A | | | Elyakov et al. (1973) |
| | Holothurin A ₃ | x | x | Kobayashi et al. (1991) |
| <i>Actinopyga miliaris</i> | Holothurin B | | | Elyakov et al. (1973) |
| | Holothurin B ₁ | x | x | Kobayashi et al. (1991) |
| | Holothurin A | | | Elyakov et al. (1973) |
| <i>Bohadschia argus</i> | Arguside A | | | Liu et al. (2007) |
| | Arguside B | | | Liu et al. (2008a) |
| | Arguside C | x | x | Liu et al. (2008a) |
| <i>Bohadschia bivittata</i> | Arguside D | x | x | Liu et al. (2008b) |
| | Arguside E (Desholothurin A ₁)** | | | Liu et al. (2008b) |
| | Bivittoside A | x | x | Ohta and Hikino (1981) |
| | Bivittoside B | x | x | Ohta and Hikino (1981) |
| <i>Bohadschia marmorata</i> | Bivittoside C | x | x | Ohta and Hikino (1981) |
| | Bivittoside D | x | x | Ohta and Hikino (1981) |
| | 17-hydroxy impatienside A | | | Yuan et al. (2009) |
| | 25-acetoxy bivittoside D | | | Yuan et al. (2009) |
| <i>Bohadschia subrubra</i> | Bivittoside C | | | Yuan et al. (2009) |
| | Bivittoside D | | | Yuan et al. (2009) |
| | Marmoratoside A (Impatienside A)** | | | Yuan et al. (2009) |
| | Marmoratoside B/Holothurin B | | | Yuan et al. (2009) |
| <i>Bohadschia tenuissima</i> | Arguside C | x | x | Chapter 2 |
| | Bivittoside B | x | x | Chapter 2 |
| | Bivittoside C | x | x | Chapter 2 |
| | Bivittoside D | x | x | Chapter 2 |
| <i>Bohadschia vitiensis</i> | Bivittoside A | | | Radhika et al. (2002) |
| | Bivittoside B | | | Radhika et al. (2002) |
| | Bivittoside C | | | Radhika et al. (2002) |
| | Bivittoside D | | | Radhika et al. (2002) |
| <i>Holothuria arenicola</i> | Holothurin A | | | Elyakov et al. (1973) |
| | Holothurin B | | | Elyakov et al. (1975) |
| | Holothurin A | | | Kobayashi et al. (1991) |
| | Holothurin B | | | Kobayashi et al. (1991) |
| <i>Holothuria atra</i> | Holothurin A | | | Kobayashi et al. (1991) |
| | Holothurin B | | | Chapter 2 |
| | Holothurin B ₁ | x | x | Kobayashi et al. (1991) |
| | Holothurin B ₂ | x | x | Chapter 2 |
| <i>Holothuria axilloga</i> (Microthele) | Axillogoside A | | | Yuan et al. (2008) |
| | Holothurin A | | | Kobayashi et al. (1991) |
| | Holothurin A ₃ | | | Kobayashi et al. (1991) |
| | Holothurin B | | | Yuan et al. (2008) |
| <i>Holothuria cinerascens</i> | Holothurin A | | | Elyakov et al. (1973) |
| | Holothurin A | | | Elyakov et al. (1973) |
| | Holothurin B | | | Elyakov et al. (1973) |
| | Holothurin A | | | Elyakov et al. (1975) |
| <i>Holothuria cubana</i> | Holothurin A | | | Elyakov et al. (1975) |
| | Holothurin B | | | Elyakov et al. (1975) |
| | Holothurin A | | | Elyakov et al. (1973) |
| | Holothurin B | | | Elyakov et al. (1973) |
| <i>Holothuria difficilis</i> | Holothurin A | | | Elyakov et al. (1991) |
| | Holothurin B | | | Stonik (1986) |
| | Holothurin A | | | Stonik (1986) |
| | Holothurin B | | | Stonik (1986) |
| <i>Holothuria edulis</i> | Holothurin A ₁ | | | Stonik (1986) |
| | Holothurin A ₂ | | | Stonik (1986) |
| | Holothurin A ₃ | | | Stonik (1986) |
| | Holothurin B | | | Stonik (1986) |
| <i>Holothuria floridana</i> | Holothurin A ₁ | | | Stonik (1986) |
| | Holothurin A ₂ | | | Stonik (1986) |
| | Holothurin A ₃ | | | Stonik (1986) |
| | Holothurin B ₁ | | | Elyakov et al. (1982) |
| <i>Holothuria forskali</i> | Des-holothurin A (Nobiliside 2a)** | x | x | Rodriguez et al. (1991) |
| | Des-holothurin A ₁ (Arguside E)** | | | Chapter 1 |
| | Holothurin A | | | Rodriguez et al. (1991) |
| | Holothurin B | | | Rodriguez et al. (1991) |
| <i>Holothuria impii</i> | Holothurin A | | | Rodriguez et al. (1991) |
| | Holothurin B | | | Rodriguez et al. (1991) |
| | Holothurin C | | | Rodriguez et al. (1991) |
| | Holothurin D | | | Chapter 1 |
| <i>Holothuria marmorata</i> | Holothurin A | | | Rodriguez et al. (1991) |
| | Holothurin B | | | Rodriguez et al. (1991) |
| | Holothurin C | | | Rodriguez et al. (1991) |
| | Holothurin D | | | Chapter 1 |

Table 1 (continued)

| Species | Saponins | BW | CT | References |
|--------------------------------|------------------------------------|----|----|------------------------|
| <i>Holothuria forskali</i> | Holothurinoside E | x | x | Chapter 1 |
| | Holothurinoside E ₁ | x | x | Chapter 1 |
| | Holothurinoside F | x | x | Chapter 1 |
| | Holothurinoside F ₁ | x | x | Chapter 1 |
| | Holothurinoside G | x | x | Chapter 1 |
| | Holothurinoside G ₁ | x | x | Chapter 1 |
| | Holothurinoside H/Marmoratoside B* | x | x | Chapter 1 |
| | Holothurinoside H ₁ | x | x | Chapter 1 |
| | Holothurinoside I | x | x | Chapter 1 |
| | Holothurinoside I ₁ | x | x | Chapter 1 |
| | Holothurinoside L*** | | | Chapter 4 |
| | Holothurinoside M*** | | | Chapter 4 |
| <i>Holothuria fuscocinerea</i> | Holothurin A | | | Zhang et al. (2006) |
| | Holothurin B | | | Elyakov et al. (1973) |
| | Fuscocineroside A | | | Zhang et al. (2006) |
| | Fuscocineroside B | | | Zhang et al. (2006) |
| | Fuscocineroside C | | | Zhang et al. (2006) |
| | Pervicoside C | | | Zhang et al. (2006) |
| <i>Holothuria gracilis</i> | Holothurin A | | | Elyakov et al. (1973) |
| | Holothurin B | | | Elyakov et al. (1973) |
| <i>Holothuria grisea</i> | 17-dehydroxyholothurinoside A | | | Sun et al. (2008) |
| | Griseaside A | | | Sun et al. (2008) |
| | Holothurin A | | | Elyakov et al. (1975) |
| | Holothurin A ₁ | | | Elyakov et al. (1975) |
| | Holothurin B | | | Elyakov et al. (1975) |
| <i>Holothuria hilla</i> | Hillaside C | | | Wu et al. (2006c) |
| | Holothurin A | | | Elyakov et al. (1973) |
| | Holothurin B | | | Elyakov et al. (1973) |
| <i>Holothuria impatiens</i> | Bivittoside D | | | Sun et al. (2007) |
| | Holothurin A | | | Elyakov et al. (1973) |
| <i>Holothuria leucospilota</i> | Impatienside A (Marmoratoside A)** | | | Sun et al. (2007) |
| | Bivittoside C | x | x | Chapter 2 |
| | Bivittoside D | x | x | Chapter 2 |
| | Desholothurin A (Nobiliside 2a)** | x | x | Chapter 2 |
| | Fuscocineroside B/C* | x | x | Chapter 2 |
| | Holothurin A | x | x | Kitagawa et al. (1979) |
| | Holothurin B | x | x | Kitagawa et al. (1978) |
| | Holothurin B/B4* | | | Chapter 2 |
| | Holothurin B ₁ | x | x | Chapter 2 |
| | Holothurin B ₂ | x | x | Chapter 2 |
| | Holothurin B ₃ | x | x | Chapter 2 |
| | Holothurinoside E ₁ | | | Chapter 2 |
| | Leucospilotoside A | | | Han et al. (2007) |
| | Leucospilotoside C | | | Han et al. (2008) |

| Species | Saponins | BW | CT | References |
|--------------------------------|-------------------------------------|----|----|-------------------------|
| <i>Holothuria lubrica</i> | Holothurin A | | | Yasumoto et al. (1967) |
| | Holothurin B | | | Yasumoto et al. (1967) |
| <i>Holothuria mexicana</i> | Holothurin A | | | Elyakov et al. (1975) |
| | Holothurin B | | | Elyakov et al. (1975) |
| <i>Holothuria moebi</i> | Holothurin A | | | Matsuno and Iba (1966) |
| <i>Holothuria monacaria</i> | Holothurin A | | | Matsuno and Iba (1966) |
| <i>Holothuria nobilis</i> | Holothurin A | | | Elyakov et al. (1973) |
| | Holothurin B | x | | Radhika et al. (2002) |
| | Nobiliside 1a | | | Wu et al. (2006a) |
| | Nobiliside 2a (Desholothurin A)** | | | Wu et al. (2006a) |
| | Nobiliside A | | | Wu et al. (2006b) |
| | Nobiliside B | | | Wu et al. (2006b) |
| | Nobiliside C | | | Wu et al. (2006b) |
| <i>Holothuria pervicax</i> | Holothurin A | | | Elyakov et al. (1973) |
| | Holothurin B | | | Elyakov et al. (1973) |
| | Pervicoside A | x | x | Kitagawa et al. (1989) |
| | Pervicoside B | x | | Kitagawa et al. (1989) |
| | Pervicoside C | x | | Kitagawa et al. (1989) |
| <i>Holothuria polii</i> | Holothurin A | x | | Silchenko et al. (2005) |
| | Holothurin B | x | | Silchenko et al. (2005) |
| | Holothurin B ₂ | x | | Silchenko et al. (2005) |
| | Holothurin B ₃ | x | | Silchenko et al. (2005) |
| <i>Holothuria pulla</i> | Holothurin A | | | Elyakov et al. (1973) |
| | Holothurin B | | | Elyakov et al. (1973) |
| <i>Holothuria scabra</i> | 24-dehydroholothurin A ₂ | x | | Kobayashi et al. (1991) |
| | Holothurin A | x | | Dang et al. (2007) |
| | Holothurin A ₂ | x | | Dang et al. (2007) |
| | Holothurin A ₃ | x | | Dang et al. (2007) |
| | Holothurin A ₄ | | | Dang et al. (2007) |
| | Holothurin B | | | Elyakov et al. (1973) |
| <i>Holothuria squamifera</i> | Holothurin A | | | Stonik (1986) |
| <i>Holothuria surinimensis</i> | Holothurin A | | | Elyakov et al. (1975) |
| | Holothurin B | | | Elyakov et al. (1975) |
| <i>Holothuria tubulosa</i> | Holothurin A | | | Silchenko et al. (2005) |
| | Holothurin B | | | Silchenko et al. (2005) |
| <i>Pearsonothuria graeffei</i> | Bivittoside D | | | Chapter 2 |
| | Desholothurin A (Nobiliside 2a)** | x | x | Chapter 2 |
| | Fuscocineroside B/C* | x | x | Chapter 2 |
| | Holothurin A | x | x | Elyakov et al. (1973) |
| | Holothurin A ₂ | x | x | Stonik (1986) |
| | Holothurin B | x | x | Elyakov et al. (1973) |
| | Holothurin B/B ₄ * | | | Chapter 2 |
| | Holothurinoside C | x | | Chapter 2 |

BW = body wall; CT = Cuvierian tubules; sulfated saponins in green; non-sulfated saponins in red.

* Isomeric saponins.

** Different names for the same structure.

*** Extracted from seawater in which the animal stayed.

intestinal hemal vessels, the ovaries, the testes and the Cuvierian tubules. Amounts of saponins (expressed by the hemolytic index) were different between body compartments and the body wall and Cuvierian tubules showed the highest values (ovaries also presented high saponin concentrations but they varied with the reproductive cycle of the animal; Matsuno and Ishida 1969). Van Dyck et al. (2010) also highlighted a variation of saponin quantities between the Cuvierian tubules and the body wall in several species of Holothuriidae. Triterpene glycosides appear to be particularly concentrated in the Cuvierian tubules, a specialized defense system developed by some species within the family Holothuriidae (Matsuno and Ishida 1969; Elyakov et al. 1973; Kobayashi et al. 1991). This organ located in the posterior part of the animal consists of multiple tubules that can, in some species, be expelled by the individual after stimulation (Bingham and Braithwaite 1986; Hamel and Mercier 2000; Becker and Flammang in press).

In terms of composition of the congener mixture, although for a same species many saponins are common to both body wall and Cuvierian tubules, some congeners seem to be organ-specific (Table 1). Some species possess more saponin congeners in the Cuvierian tubules than in the body wall (e.g. *H. leucospilota*), some less (e.g. *A. echinites*), and some have roughly the same number of saponins in the two organs (e.g. *B. subrubra* and *P. graeffei*) (Kobayashi et al. 1991; Van Dyck et al. 2009, 2010).

The large number of different saponins within a species as well as the intra-individual variations in saponin mixtures raises the question of the specific functions of these molecules. One holothuriid species can indeed contain numerous different saponins (more than 20 in *H. forskali*), the different congeners varying in the number, position and nature of the monosaccharide units and also in the number and position of double bonds, hydroxyl, acetate, sulfate and other functional groups on the aglycone and the carbohydrate chain (Kornprobst 2005; Kalinin et al. 2005). Possessing such a molecular diversity should be a selective advantage for the animal, different molecular structures seemingly conferring different properties to the saponins. According to Kalinin (2000), the presence of a sulfate group enhances the hydrophilicity of the saponin while the length and composition of the carbohydrate chain is important for its membranolytic action. This could explain at least partly the variation of saponin composition between the body wall and the Cuvierian tubules in a single species. To make the picture even more complex, it has been shown recently that, in the Cuvierian tubules of *H. forskali*, a prolonged stress induces the modification of some congeners into others by addition of a disaccharide (Van Dyck et al. in press). All the data taken together therefore

indicate a complex chemical defence mechanism with, for a single species, different sets of saponins originating from different body compartments and presenting different responses to stress. This presumably finely tunes the saponin properties according to their ecological role(s).

Acknowledgements

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Juveniles and natural spawning observations

Observations of fission in La Reunion, Indian Ocean

From: Philippe Bourjon (seizhavel@wanadoo.fr)

On La Reunion Island, fission of *Holothuria atra* and *Stichopus chloronotus* has been studied and was first observed for *H. leucospilota* (Conand et al. 1997; Conand 2004; Kohler et al. 2009). The present observations confirm those of previous studies.

The interest in the inner reef flat of Planch'Alizés on Reunion Island lies in the high density of *H. atra* and *H. leucospilota* populations. This site has an average depth of less than 1 m, consists of a sandy substrate with some coral rubble, and is considered to be damaged. Eutrophication increases the concentration of organic matter, especially within the substrate. Eutrophication and constant pounding by "swimmers" walking in this popular shallow area may account for some of the fission inductors.

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Figure 1. Regenerating *Holothuria atra* (6.5 cm specimen) observed on the site of Planch'Alizés. Numerous juveniles and some individuals in the regeneration process were mixed with many *H. leucospilota* and *H. atra* adults.



Figure 2. *Holothuria atra* during the fission process. This specimen is on a sandy bottom substrate with coral rubble and living corals. Fission takes place at roughly 30% of the anterior portion of the body.

Observation of a juvenile of the commercial species *Thelenota ananas*, in La Reunion, Indian Ocean

From: Florence Trentin, *Vie Océane* (florence.trentin@orange.fr)

Tropical sea cucumber juveniles are rarely observed *in situ*. However, it is vital to have information on their ecology so as to understand the phenomenon of recruitment, which is still very poorly known in this group of invertebrates.

Thelenota ananas is a fairly uncommon species on Reunion Island reefs. Isolated adult specimens have been seen occasionally but never in dense populations.

They are generally found on sandy bottoms at depths of more than 25 m (one specimen observed on 12/10/2008 on volcanic sand bottom off the coast of St Leu at a depth of 45 m in the area known as “Arche de Noé” – minimum length : 70 cm; another observed on 18/12/2008 on a volcanic slab at “La pointe au sel”, at a depth of 27 m). These different observations lead us to think that adults do not live in coral zones.

The photos of juveniles (Figs 2A and B) were taken at 3:30 p.m. on the 15th of January 2009 at a depth of 15 m in a relatively healthy coral zone near a sandy bottom, south of Pointe au sel (Saint Leu reef), in an area known as “Sec jaune”. The specimen measured about 14 cm in length. A photo of a juvenile of this species taken on a reef in New Caledonia was published by Conand (1981). This raises the question that while this species does not seem to be directly found on coral reefs in Reunion Island, it may need reefs for the development of its juveniles. It would be worthwhile, then, to confirm that adult specimens are really never found in coral zones and that juveniles can be found on coral reefs during the warm season.



Figure 1. Adult *Thelenota ananas* – depth: 45 m.

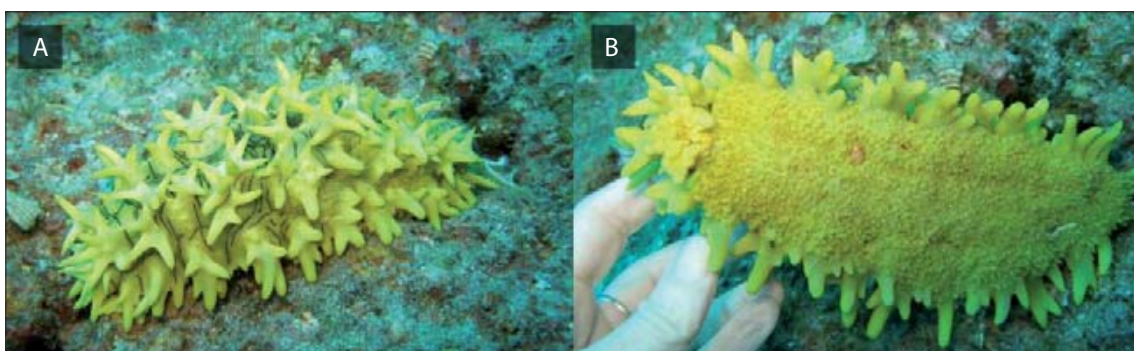


Figure 2. Dorsal (A) and ventral (B) views of a juvenile *Thelenota ananas*.

Observation of wild *Stichopus chloronotus* juveniles

From: Nina Yasuda,¹ Coralie Taquet,² and Katuki Oki³

¹ JSPS Post-doctoral fellow, National Research Institute of Fisheries and Environment of Inland Sea, Hiroshima, Japan.

² Post-doctoral fellow, Department of Mechanical and Environmental Informatics, Graduate School of Information Science and Engineering, Tokyo Institute of Technology, Tokyo, Japan. Email: coralie.taquet@gmail.com.

³ Tida Design Limited Company, Amami Island, Japan.

Species: *Stichopus chloronotus*, Brandt, 1835.

Location: Ohama (28°24'07.77"N, 129°27'15.20"E), Naze-koyado, Amami-city, in Amami Island (Kagoshima-Prefecture), located between the four main islands of Japan and Okinawa Archipelago (Japan). The climate is subtropical.

Date and time: 10 July 2010, day time (11:00–13:00, local time).

Moon phase: New moon

Observer/Photographer: Nina Yasuda (Fig. 1).

Depth and tide: ~ 1 m, high tide (spring tide).

Bottom: Sandy bottom with coral rubble.

Note: Morphology very close to that of an adult (Conand 1998). Curved body length: 5–10 cm. With length/width ratio similar to that of an adult; they are distinguished from one another by thick, short individuals produced from asexual reproduction. There were many juvenile *Stichopus chloronotus* together with adults on the Ohama coast (total density of *Stichopus chloronotus* was about 0.05 ind m⁻², and almost 37 out of 90 individuals were juveniles with less than 10 cm of curved body length). Corals at Ohama were abundant before the coral bleaching event in 1998. *Stichopus chloronotus* is mostly found on dead coral rubble. We found only 1 juvenile out of 86 individuals in the nearby *Stichopus chloronotus* population at Cape Ayamaru (28°24'28.88"N, 129°43'00.74"E) during the same survey (11 July 2010).

Reference: Conand C. 1998. Holothurians. p. 1157–1190. In: Carpenter K.E. and Niem V.H. (eds). The living marine resources of the western Central Pacific. Volume 2. Cephalopods, crustaceans, holothurians and sharks. FAO, Rome. 4218 p.

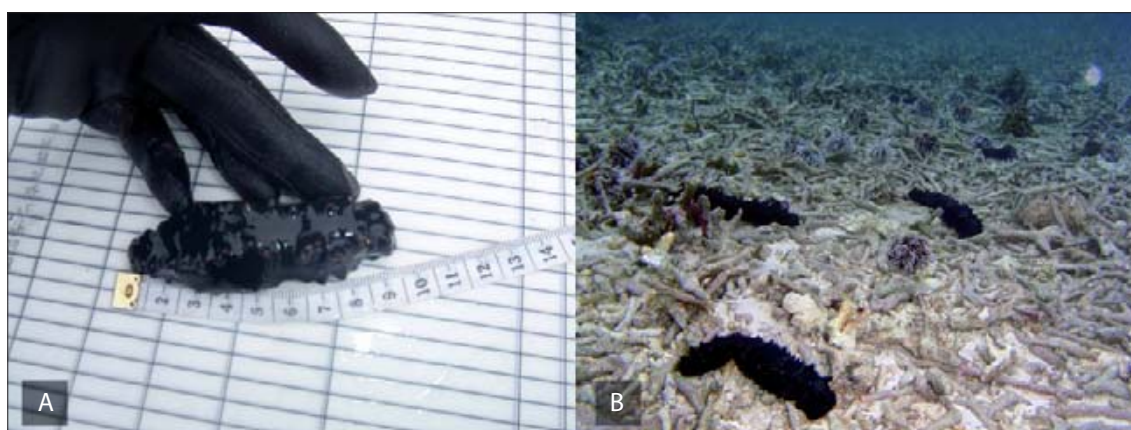


Figure 1. A: Juvenile *Stichopus chloronotus*; B: *Stichopus chloronotus* adults on dead coral rubble at Ohama, Amami Ohshima, Japan.

Observation of a wild *Holothuria scabra* juvenile

From: Coralie Taquet,¹ Fakhrizal Setiawan,² Nina Yasuda,³ Suharsono,⁴ and Kazuo Nadaoka¹

¹ Post-doctoral fellow, Department of Mechanical and Environmental Informatics, Graduate School of Information Science and Engineering, Tokyo Institute of Technology, Tokyo, Japan. Email: coralie.taquet@gmail.com.

² Marine conservation officer, Wildlife Conservation Society (WCS), Indonesia Marine Program, Kota Manado, Sulawesi Utara, Indonesia

³ JSPS Post-doctoral fellow, National Research Institute of Fisheries and Environment of Inland Sea, Hiroshima, Japan

⁴ Director of the Research Center for Oceanography, Indonesian Institute of Sciences (LIPI), Jakarta, Indonesia

Species: *Holothuria* (*Metriatyla*) *scabra*, Jaeger, 1833.

Location: Pantai Kalinaun (1°38'0.04"N, 125° 9'2.62"E), east side of Manado peninsula (north of Bitung, terrestrial protected area), Manado, North Sulawesi, Indonesia. The climate is tropical.

Date and time: 15 May 2010, day time (15:00–17:00, local time).

Moon phase: New moon on 14 May 2010.

Observer/Photographer: Coralie Taquet (Fig. 1).

Depth and tide: ~ 10 cm, low tide (spring tide).

Bottom: Seagrass bed area (main species *Thalassia* sp.) on a sandy bottom with coral rubble.

Note: Morphology very close to that of an adult (Conand 1998): oval body (trivium slightly flattened), brass color with black stripes for the bivium, and whitish trivium. However, the wrinkles on the bivium were not yet noticeable. Curved body length: 10 cm.

Reference: Conand C. 1998. Holothurians. p. 1157–1190. In: Carpenter K.E. and Niem V.H. (eds). The living marine resources of the western Central Pacific. Volume 2. Cephalopods, crustaceans, holothurians and sharks. FAO, Rome. 4218 p.

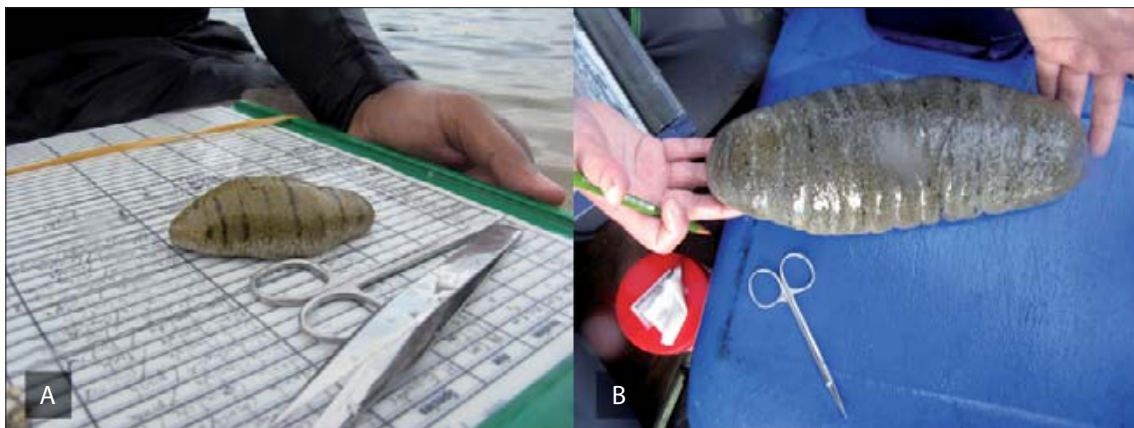


Figure 1. A: Juvenile *Holothuria scabra* in Pantai Kalinaun (Manado, north Sulawesi, Indonesia); B: Adult *H. scabra* in Pulau Pari (Kepulauan Seribu Archipelago, north Jakarta, Indonesia).

Natural spawning observation of *Actinopyga mauritiana*

From: Katsuki Oki,¹ Coralie Taquet,² and Nina Yasuda³

¹ Tida Design Limited Company, Amami Island, Japan.

² JSPS Post-doctoral fellow, National Research Institute of Fisheries and Environment of Inland Sea, Hiroshima, Japan.

³ Post-doctoral fellow, Department of Mechanical and Environmental Informatics, Graduate School of Information Science and Engineering, Tokyo Institute of Technology, Tokyo, Japan. Email: coralie.taquet@gmail.com.

Species: *Actinopyga mauritiana* (Kuriironamako), Quoy and Gaimard 1833.

Location: Kise (28°25'41.60"N, 129°39'21.65"E), Kasari-town, Amami-city, in Amami Island (Kagoshima-Prefecture), located between the four main islands of Japan and Okinawa Archipelago (Japan). The climate is tropical.

Date and time: 11 July 2010, day time (around noon, local time).

Moon phase: New moon.

Observer: Katsuki Oki (unfortunately, it was not possible to take a picture of spawning individuals).

Depth: ~ 1 m; the area is never exposed, even during a spring tide.-

Bottom: Shallow, sandy (coral rubble and sand) bottom with some sea algae.

Tide: High tide (spring tide), almost no tidal current at that time.

Note: Fine day with almost no cloud cover; seawater was also clear and bright. There were four individuals (possibly all males) spawning at the same time in this area. They curved their bodies, and spawned (i.e. emitted a white string that gradually muddied the water) from the tip of the vertical part of their bodies, which were slowly rocking.

PIMRIS is a joint project of five international organisations concerned with fisheries and marine resource development in the Pacific Islands region. The project is executed by the Secretariat of the Pacific Community (SPC), the South Pacific Forum Fisheries Agency (FFA), the University of the South Pacific (USP), the Pacific Islands Applied Geoscience Commission (SOPAC), and the Pacific Regional Environment Programme (SPREP). This bulletin is produced by SPC as part of its commitment to PIMRIS. The aim of PIMRIS is to improve the



availability of information on marine resources to users in the region, so as to support their rational development and management. PIMRIS activities include: the active collection, cataloguing and archiving of technical documents, especially ephemera ('grey literature'); evaluation, repackaging and dissemination of information; provision of literature searches, question-and-answer services and bibliographic support; and assistance with the development of in-country reference collections and databases on marine resources.

Communications

From: Cathy Hair,¹ Teari Kaure,² Paul Southgate,¹ Timothy Pickering³

¹ James Cook University, Townsville, Australia. Email: cathy.hair@jcu.edu.au

² Department of Fisheries, Galoa, Fiji Islands

³ Secretariat of the Pacific Community, Suva, Fiji Islands

Potential breakthrough in hatchery culture of sandfish *Holothuria scabra* by using algal concentrate as food

This note reports on a potentially important advance in larval feeding strategies of the commercially valuable sea cucumber *Holothuria scabra* (sandfish). Reliable and routine methods for sandfish hatchery production have been developed (James et al. 1994; Agudo 2006; Duy 2010) and are based on the provision of live microalgae as the larval food source. Although a mixed-species diet composed of two to three species is generally considered preferable (Agudo 2006; Duy 2010), *Chaetoceros muelleri* can be effectively used alone throughout the larval culture period (Duy 2010). Duy (2010) also reported that Tahitian *Isochrysis* (*T. iso*) is a suitable diet for the first few days of larval rearing, but this species is inadequate as a complete diet (Knauer in prep.). Following settlement, sandfish pentactula and early juveniles are fed live uni-celled algae or diatoms (e.g. *Nitzschia*, *Chaetoceros*, *Skeletonema* species), as well as commercially available dried algae such as “Algamac 2000” and *Spirulina* (Agudo 2006; Duy 2010).

Culturing live microalgae can be unreliable and is expensive and technically demanding. Lack of appropriate resources and technical skills is a major constraint to hatchery-based mariculture development in many tropical countries. This is particularly the case in Pacific Island countries. While a variety of materials have been investigated as potential replacements for live microalgae as a food source for invertebrate larvae (Knauer and Southgate 1999), dried microalgae and concentrated microalgae in paste form have shown particularly promising results (Southgate 2003). Commercially available microalgae concentrates manufactured in the USA (Instant Algae®, Reed Mariculture) have been used as a sole food source for successful hatchery production of the winged pearl oyster in Tonga for the past three years as part of an Australian Centre for International Agricultural Research (ACIAR)-funded project (Teitelbaum and Fale 2008). Clearly, these products may have potential as a food source for the larvae of other marine invertebrates, including sea cucumbers.

A recent technology transfer project in Fiji (funded by ACIAR), attempted hatchery production of sandfish at the Department of Fisheries marine hatchery at Galoa, Viti Levu. Attempts to rear larvae were threatened by a low volume of available live microalgae and its relatively poor quality. But this provided a serendipitous opportunity to assess the nutritional value of the Instant Algae® product called Shellfish Diet 1800® for sandfish larvae. Shellfish Diet® is composed of a mixture of *Isochrysis* (30%), *Tetraselmis* (20%), *Pavlova* (20%) and *Thalassiosira weissflogii* (30%) (see manufacturer’s information: <http://www.reed-mariculture.com>). It is available in a one-quart bottle and has a cell concentration of around 2 billion cells mL⁻¹. It does not contain preservatives.

This trial was unplanned and feeding began in the normal way with the assumption that sufficient live algae would be available. From two 1,000 L larval culture tanks, tank 1 was provided with live algae (20 K cells mL⁻¹ of *T. iso*) on day 2 (first day of feeding) and a small amount on day 5 (10 K cells mL⁻¹ of *T. iso*). The bulk of the diet for the other 10 days of feeding (approximately 90% of that received by the larvae) was Shellfish Diet® (up to 45 K cells mL⁻¹). In tank 2, approximately 60% of the feed was provided by Shellfish Diet® over the same period. Larvae in two 500 L tanks (tanks 3 and 4) were fed predominantly live microalgae (*T. iso* and *Chaetoceros*) (approximately 60% over the culture period) supplemented with Shellfish Diet® as necessary. The larvae reared mainly on Shellfish Diet® (tanks 1 and 2) were present in reasonable densities at late auricularia stage (approximately 30% survival at day 9). They progressed to doliolaria stage (day 11) and settled as pentactula by day 15. The late auricularia and doliolaria had well-developed hyaline spheres, reached maximum sizes (as indicated in the literature), and appeared normal.

This is the first report of successful hatchery culturing of sandfish on a diet composed not exclusively of live microalgae. Indeed, our results, although preliminary, indicate that the majority of a live microalgae diet can be replaced by Shellfish Diet® without compromising growth and survival of sandfish larvae. This finding has major implications for further developments in hatchery culture technology for sandfish and tropical sea cucumber generally. Broad use of Instant Algae® as a food for sea cucumber larvae would be a cheaper, easier

and more efficient option for hatchery culture in Pacific Island countries and would allow diversification of low technology hatchery facilities within the region; a development that would be unfeasible if reliant on live microalgae production. Instant Algae® products have a shelf life of several months when refrigerated, and last up to two years frozen for selected species. Although relatively expensive to purchase per container, when compared to the running cost of a microalgae culture unit, there are significant cost benefits.

Much more research is required before such development could be considered, however. For example, there are questions about the physical behaviour of Instant Algae® in the water column (i.e. cells do not swim and are negatively buoyant) and whether the feeding regimes and culture systems (e.g. aeration) developed for use with live microalgae need to be modified to account for this. Purposely designed nutritional studies should be undertaken to determine if Instant Algae® provides the required nutrients for all development stages of *H. scabra* larvae (i.e. live microalgae may be required at some critical developmental stages), and to assess the relative nutritional value of the five products within the Instant Algae® range for sandfish larvae. These and other aspects relating to the use of these products with sandfish larvae will be addressed in ongoing research at James Cook University.

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Caballero-Ochoa and Laguarda-Figueras: Zoogeography of holothurians (Echinodermata: Holothuroidea) of the Mexican Pacific Ocean [oral presentation].

Caulier, Flammang, Gerbaux, Rakotoarisoa and Eeckhaut: When a repellent becomes an attractant: Harmful saponins are kairomones that maintain the symbiosis between the Arlequin crab and their sea cucumber hosts [oral presentation].

Hennebert and Flammang: Echinoderms don't suck: Evidence against the involvement of suction in tube foot attachment [oral presentation].

Mezali and Paulay: About the taxonomic status of aspidochirotid holothurians inhabiting the *Posidonia oceanica* meadow in the Algerian area [oral presentation].

Navarro, Tuya and González: Population density and size distribution of *Holothuria sanctori*, *Holothuria dakarensis* and *Holothuria arguinensis* (Holothuroidea: Aspidochirotida) on Gran Canaria, Canary Islands (Central–Eastern Atlantic) [oral presentation].

Palazón-Fernández: Casual hermaphroditism in a population of the gonochoric sea cucumber, *Isostichopus bacionotus* Selenka (Echinodermata: Holothuroidea) of north eastern Venezuela [poster presentation].

Reich and O'Loughlin: Modern holothurian calcareous ring anatomy and stereom structure — the need for more detailed studies and research [poster presentation].

Reich and Stöhr: Hard-part morphology of the sea cucumber *Eupyrgus scaber* (Holothuroidea: Molpadiida: Eupyrgidae) [poster presentation].

Rogacheva, Gebruk and Alt: Swimming in deep-sea holothurians: An adaptation to the mid-ocean ridge environment? [oral presentation].

Thandar and Arumugam: A new family within the holothuroid order Dactylochirotida [oral presentation].

From: Sharon Ng, Irwin Wong and Poh Sze Choo

WWF – Malaysia, 49, Jalan SS23/15, Taman SEA, 47400 Petaling Jaya, Selangor, Malaysia

Report on WWF – Malaysia Regional Workshop on “Feasibility of sea cucumber and abalone farming as an alternative livelihood in Semporna, Sabah”

A regional workshop that was organized and sponsored by WWF – Malaysia on the “Feasibility of sea cucumber and abalone farming as an alternative livelihood in Semporna, Sabah” was held from 14–16 April 2010 in Semporna. The workshop was attended by about 40 participants from Malaysia, Singapore and the Philippines, who included scientists, government officials, fishers, traders and representatives from non-governmental organisations (NGOs) and local coastal communities. This report focuses on the agenda and recommendations made specifically for sea cucumbers by workshop participants.

The objectives of the workshop were to:

- discuss the feasibility of sea cucumber and abalone farming as a supplementary livelihood in Semporna;
- review lessons learned and best practices in sea cucumber and abalone farming locally and overseas;
- identify the potential for a pilot project and partners for the project.

Papers presented at the workshop that are relevant to sea cucumber farming included:

- Introduction to Sulu-Sulawesi Marine Ecoregion (SSME), Semporna and objectives of the workshop: Kenneth Kassem;
- A review of sea cucumber and abalone fisheries in Malaysia: Choo Poh Sze;
- Sea cucumber ranching and lessons learnt in the Philippines: Ruth Gamboa; and
- Commercial sea cucumber farming proposal: Teng Seng Keh.

Main points of workshop discussion and recommendations

- It was felt that pen culture of sea cucumber is more suitable in Semporna because of soil conditions. The bedrock there is very near to the surface and building ponds in these places is a problem.
- A sea cucumber hatchery is necessary and there are two options to acquire broodstock: 1) buy from fishers or others in Sabah, or 2) import from neighbouring countries such as Indonesia and the Philippines.
- The Sabah Parks hatchery in Bohey Dulang is considered to be a suitable site to locate a sea cucumber hatchery.
- The state government should take the lead in setting up the sea cucumber hatchery.
- The state government should designate an area for sea cucumber farming. Locating farms in one particular area may also help aggregate broodstock and facilitate natural sea cucumber recruitment.
- It is felt that financing — in the form of micro-credit or interest-free loans for farmers — is necessary and should be provided by the government or NGOs. Rearing individuals to the minimum marketable size of 250 g may take up to 1½ years of culturing, and farmers may need financial support during the grow-out period.
- Government aid through the Projek Rakyat Termiskin project, which is directed at the very poor) and Projek Luar Bandar (Rural Development Project) should be explored for financing farmers.
- Integrated farming such as sea cucumbers with fish, or sea cucumbers with seaweed could be considered, especially in existing fish or seaweed farms.
- Field trips conducted on 16 and 17 April identified Nusa Tengah and Omadal as suitable sites for sea cucumber farming.



Participants of the regional workshop on “Feasibility of sea cucumber and abalone farming as an alternative livelihood in Semporna, Sabah” held in Semporna from 14–16 April 2010.



Participants were taken to several locations to observe the sea cucumber fishery in Semporna.

From: Choo Poh Sze

Sulu-Sulawesi Marine Ecoregion (SSME) consultancy for “A study on the feasibility of sea cucumber and abalone farming as a supplementary livelihood in Semporna, Sabah”: Project-5000-MY025611 and 5140-MY025641-INTL).

As the title suggests, this report explores the feasibility of farming abalone and sea cucumber in Semporna, Sabah. The report begins with a short review on the declining status of the abalone and sea cucumber fishery in Sabah, and suggests why farming these two species is important.

A literature review of sea cucumber and abalone fisheries in Sabah shows that natural populations of both resources are declining. Average annual sea cucumber landings from 2000–2005 was 137 tonnes while that for abalone from 2003–2005 was 39 tonnes. The high-value sandfish and teatfish fisheries appear to be very threatened and the sea cucumber fishery in Sabah comprises mainly lower-value species such as tigerfish, blackfish, curryfish, elephant trunkfish and lollyfish. The main abalone species exploited commercially is the donkey-ear abalone.

Presently, there are no fishing regulations specific to sea cucumber and abalone fisheries. As a result, overfishing of both abalone and sea cucumber has occurred. In the present study, 74% of the fishers surveyed indicated that abalone and sea cucumber resources have declined, and suggested that the fisheries should be regulated by imposing a minimum size at harvest.

There is an urgent need to stop the decline of both fisheries through aquaculture and sea ranching efforts. Fishing for sea cucumber and abalone provides livelihood options for many who are poor and landless; sustaining the fisheries will ensure that the poor are not deprived of a source of much needed income. Only 10% of respondents interviewed in this study reported monthly earnings of above RM1000 (RMD 1.00 = USD 0.30) — an amount exceeding the monthly poverty line household income of RM960 for Sabah.

Although culturing sea cucumbers has been conducted since the early 2000s, no sustained large-scale seed and grow-out production has been realized. Trials on grow-out in sea pens are restricted by an insufficient wild seed supply, and hatchery-produced seed are not available locally. Abalone farming has attracted participation from the private sector where species from temperate waters (*Haliotis iris* and *H. discus*) have been farmed successfully on a small scale. Farming of tropical and subtropical species (*H. asinina* and *H. diversicolor supertexta*) has also been successfully carried out on a commercial scale by a company in Labuan in East Malaysia. About 90% of fishers interviewed indicated their interest in farming abalone and sea cucumber.

A draft pilot project proposal to initiate the farming of abalone and sea cucumber (involving various government agencies and NGOs in Semporna) is also included in this report.

From: Martin Brogger

Dear friends,

The first Latin American Echinoderm Congress will be held from 13–18 November 2011 at Centro Nacional Patagónico, Puerto Madryn, Argentina. The second circular is available on the webpage of the meeting (in Spanish) at: www.cle2011.com.ar

The planned schedule includes: Sunday, 13 November, arrival and evening reception; Monday, Tuesday, Thursday and Friday half day, oral and poster sessions; Wednesday, field trips and evening banquet. Presentations in Spanish, Portuguese and English are welcomed.

Please do not hesitate to contact us if you want more information about the congress.

We hope see you in Puerto Madryn.

From: Cathy Hair

The “Asia-Pacific tropical sea cucumber aquaculture symposium” will be held from 15–17 February 2011 at the Secretariat of the Pacific Community (SPC) headquarters in Noumea, New Caledonia. Organising agencies for the symposium are the Australian Centre for International Agricultural Research (ACIAR) and SPC.

The symposium is an ACIAR initiative that will focus on the current status of research on tropical sea cucumber aquaculture, covering a range of topics such as recent advances in hatchery production technology; release strategies and farming techniques; management practices; post-harvest technologies for value adding; and supply chains and marketing.

The symposium will identify gaps in current knowledge and generate recommendations for future research to support development of sustainable tropical sea cucumber aquaculture. Symposium proceedings will be published following the meeting. This symposium provides a valuable opportunity to bring together key participants who are actively engaged in progressing tropical sea cucumber culture technology and developing its potential to deliver benefits to coastal communities. Requests for further information about the symposium program should be emailed to: Geneviève Mirc (GenevieveM@spc.int) or Cathy Hair (Cathy.Hair@jcu.edu.au).

From: Christine Mae A. Edullantes and Marie Antonette Juinio-Meñez

Department of Science and Technology through the Philippine Council for Aquatic and Marine Research Development for funding support.

The sea cucumber fishery has been a source of livelihood to many coastal villagers in the Philippines. However, due to its high demand on the international market, the landed catch and production of sea cucumber products have declined. *Stichopus* spp. locally known as *hanginan* is among the many sea cucumber species in the country that has a high economic value. The animal is usually collected in the intertidal zone, and is processed, dried and sold as trepang.

Part of the ongoing efforts of the Sea Cucumber Research Program at the University of the Philippines' Marine Science Institute is to develop management and culture technologies to rebuild depleted populations of commercially important species aside from *Holothuria scabra*. Culture trials for *Stichopus* spp. started in March 2009 in the outdoor hatchery of the Bolinao Marine Laboratory. Three batches of larvae have been produced from a spontaneous spawning in the hatchery, and a few thousand juveniles were successfully reared to >5 cm from the last batch of larvae. Early juveniles (~1 cm) were reared in submerged hapa cages at ~2 m depth. Growth was quite variable, some juveniles grew up to about 7 cm after three months of rearing in hapa cages while some juveniles were clearly smaller (Fig. 1). Further spawning trials and grow-out studies will be conducted to improve our understanding of the biology and ecology of the animal.

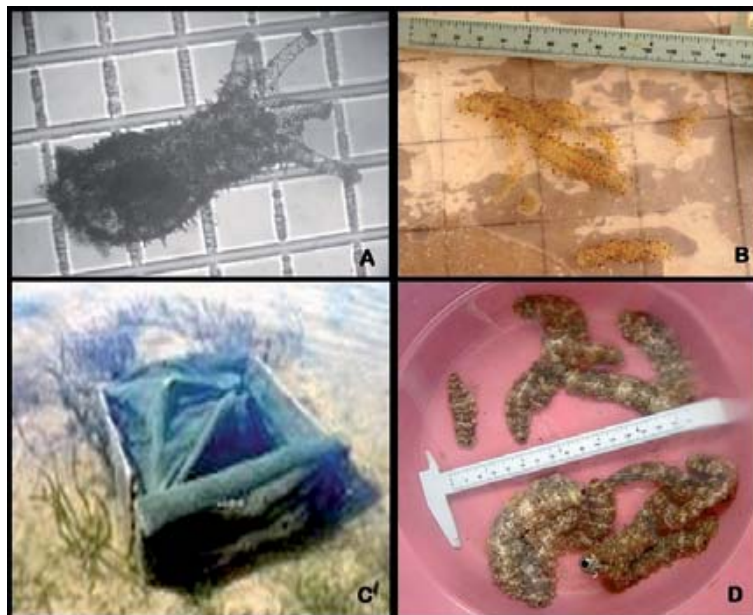


Figure 1. *Stichopus* spp. raised at the Bolinao Marine Laboratory.

From: Beth Polidoro, Marcelo Tognelli, Heather Harwell, Cristiane Elfes (IUCN Facilitators), Amancay Cepeda, José F. González-Maya, Diego A. Zárrate-Charry (ProCAT Hosts) and Juan José Alvarado, Milena Benavides, Chantal Conand, Erika Paola Ortiz, Ruth Gamboa, Jean-Francois Hamel, Annie Mercier, Steve Purcell, Veronica Toral-Granda (scientists)

IUCN Red List workshop for sea cucumbers

The International Union for Conservation of Nature (IUCN) Sea Cucumber Red List Workshop was held in Cartagena, Colombia from 17–21 May 2010. The workshop was convened by the Global Marine Species Assessment, an initiative of IUCN and Conservation International, and hosted by local NGO ProCAT Colombia. The primary goal of this five-day workshop was to bring together regional and international scientific experts to assess the conservation status and probability of extinction for approximately 300 species of sea cucumbers (Echinodermata: Holothuroidea: Aspidochirotida) for the first time, by applying the IUCN Red List Criteria. Results, in the form of Red List Assessments from this important workshop, will provide much needed information for marine conservation efforts for these species around the world, and will be made publicly available on the IUCN Red List of Threatened Species (<http://www.iucnredlist.org>).

The IUCN Red List assessment approach is based on assessing complete clades of species, in order to better understand the status of complete taxonomic groups and to provide baseline data for all species within a group. The assessment process relies heavily on the expertise and participation of hundreds of scientists and institutions. Sea cucumber workshop participants met at the Hotel Bahia in Cartagena, Colombia for five days to create draft Red List assessments for over 300 species of sea cucumbers. The goals of the workshop were to assess the 66 UN Food and Agriculture Organization recognized commercial species, as well as the remaining 250 species within the order Aspidochirotida. Based on a literature review and expert knowledge, participants provided information on each species' taxonomy, distribution, population status, habitat and ecology, major threats and conservation measures in place or recommended. This information was then used to collectively apply the IUCN Red List Categories and Criteria (IUCN 2001), which uses quantitative thresholds to determine each species' probability of extinction expressed as a Red List Category.

Over the next year, all species accounts and results from the workshop will undergo a thorough post-workshop review and consistency check before being finalized. During the review, additional experts will be consulted to fill in missing information gaps where possible, and all species account information and Red List Category classification will be checked for technical accuracy. In addition, a geo-referenced digital distribution map will be produced for each species to allow for further spatial analyses of results. Final products, including digital distribution maps, comprehensive species information, and final Red List Category for each species, are scheduled to be published on the 2011 IUCN Red List of Threatened Species.

The IUCN Red List Categories and Criteria are the most widely accepted system for classifying extinction risk at the species level. Comprehensive results from this first-ever Red List assessment of commercial and non-commercial sea cucumber species in the order Aspidochirotida will transform conservation priorities for these species across the globe, and will provide the most up-to-date review of each species' current state of knowledge and global population status. Once published, IUCN Red List assessments for a species can be regularly updated, depending on the availability of better or new data, and any subsequent changes in a species' Red List Category will serve as an important indicator of the success or failure of conservation actions.

Reference

IUCN. 2001. IUCN Red List categories and criteria version 3.1. Available: http://www.redlist.org/info/categories_criteria2001.html.



Participants of the IUCN workshop.

From: Chantal Conand

Dear friends, I would like to point you to DORIS, a new website run by the French Federation of Underwater Studies and Sports. Even if this website is in French, I'm sure our English-speaking colleagues will find some useful resources. The following presentation has been translated from their homepage.

DORIS: Underwater biodiversity as seen by divers...

DORIS (*Données d'Observations pour la Reconnaissance et l'Identification de la faune et de la flore Subaquatiques* – Observation data for recognising and identifying underwater flora and fauna) is a participatory site (<http://doris.ffessm.fr>) run by the French Federation of Underwater Studies and Sports (FFESSM). Designed by divers for divers but open to all, the DORIS site uses illustrated factsheets to make it possible to identify and inventory animal and plants species spotted during dives or at the edge of the water in French departments and territories. These species are described in a way that is both precise and comprehensible for the general public. For each one, the species' living conditions and locations are explained along with its most important biological characteristics and distinguishing features. Any need to protect it and any dangers it might present are explained and identified by logos. Each species factsheet is illustrated by a large number of original photos of the plant or animal on site. Here is an example of part of the factsheet on the sea cucumber *Thelenota ananas*.

The DORIS forum has become a meeting place for everyone who is interested in aquatic biology and its primary goal is to share knowledge and make it accessible to the public. The editorial team takes great care in verifying the information published, thanks to the support of informed amateurs or scientists, who review all the sheets. The same care is taken with the factsheets' illustrations through deliberately unusual and varied images.

With the support of some 700 volunteers, DORIS offers a French-language data and image base on aquatic life; a base that is unique in its kind and is constantly expanding: more than 1400 sheets on-line, 12,000 original illustrations, 690 terms explained in the glossary!

Having gained the recognition of the scientific community, DORIS has become a partner site for the French Inventory of Natural Heritage at the French Museum of Natural History in Paris. DORIS received the educational website award at the World Festival of Underwater Pictures in Antibes in 2007.

The DORIS team is open to all offers of assistance, which are, in fact, a basis for our success. We are always looking for people to draft factsheets, specialists to verify them and photos to illustrate them. If you would like to join us, even for a limited time, please do not hesitate to contact us!

The screenshot shows the DORIS website interface. At the top, there is a search bar and navigation links. The main content area displays a factsheet for *Thelenota ananas* (Jaeger 1833), also known as the spiny sea cucumber. The factsheet includes a large photograph of the species, a detailed description of its morphology and habitat, and smaller images showing close-ups of its papillae and internal structure. The text is in French and provides scientific details about the species.

Figure 1. Part of the DORIS factsheet on *Thelenota ananas*.

Abstracts and new publications

First report of the development of microsatellite markers for a tropical sea cucumber (*Stichopus chloronotus*)

C. Taquet, S. Nagai, N. Yasuda and K. Nadaoka

Source: Accepted as technical note in Conservation Genetics Resources (2010).

We isolated new 10 polymorphic microsatellites from the tropical sea cucumber *Stichopus chloronotus*. These loci provide one class of variable genetic marker as the number of alleles ranged from 2 to 6 and the observed and expected heterozygosity ranged from 0.083 to 1.0 and from 0.081 to 0.724, respectively. We consider that these loci are potentially useful for revealing clones (resulting from asexual repro) and then participate in detailing the genetic structure and gene flow among *S. chloronotus* populations.

Managing sea cucumber fisheries with an ecosystem approach.

S.W. Purcell

Source: Edited/compiled by Lovatelli A., Vasconcellos M. and Yimin Y. FAO Fisheries and Aquaculture Technical Paper. No. 520. Rome, FAO. 2010. 157 p. <http://www.fao.org/docrep/012/i1384e/i1384e00.htm>

Sea cucumbers are important resources for coastal livelihoods and ecosystems. At least 60 species are fished from more than 40 countries and most of the harvests are processed then exported to Asian markets. Sea cucumbers generally appear to have slow rates of population turnover and are easily harvested in shallow waters in the tropics. With retail prices of up to USD 300–500 per kg (dried), exploitation has often been indiscriminate and excessive. Overfishing in recent years has led to local extinction of high-value species in some localities and prompted closures of many national fisheries to allow stocks to recover and to allow more sustainable management plans to be established. Apart from a few developed countries, only a small number of sea cucumber fisheries are currently being managed sustainably. Sea cucumber fisheries differ greatly in the scale of the fishing activities, status of stocks and the capacity of the management agency. Consequently, some management measures will be appropriate in some fishery scenarios but not others. This document presents a logical framework to assist fishery managers in choosing an appropriate suite of regulatory measures and management actions and elaborates on the uses, limitations and ways to implement them. This document contains five main sections. The first provides an overview of the biology and ecology of sea cucumbers, the international market for beche de mer market, types of sea cucumber fisheries and their global status (i.e. population abundance). The second section summarizes fisheries management principles and approaches, with an emphasis on the ecosystem approach to fisheries (EAF). The third section provides the “roadmap”, by way of instructions, flow diagrams and tables, to lead fishery managers along the path of choosing management measures appropriate to their fishery. The fourth and fifth sections discuss the application of each regulatory measure and management action – with Examples and lessons learned boxes to illustrate management problems and potential solutions from various fisheries. Improved management of sea cucumber fisheries is an imperative. It will be best achieved by applying an EAF, in which multiple regulatory measures and management actions are applied in full consideration of the sea cucumber stocks, the ecosystems in which they live and the socioeconomic systems that drive exploitation. The commitment of governments, fishery managers and scientists to develop, apply and strictly enforce EAF will be crucial to sustaining sea cucumber populations for current and future generations.

Sea cucumber aquaculture — promising opportunity for sustainable sea cucumber fishery in South-East Asia

D.A.B. Giraspy and G.W. Ivy

Source: Asian-Pacific Aquaculture 2009, Kuala Lumpur, Malaysia

Overexploitation is affecting sea cucumber population worldwide, and aquaculture and restocking programs are needed to bring back the depleted fisheries to sustainable level. The demand for beche de mer is significantly increasing in Asia, making the sea cucumbers more vulnerable for exploitation. The sea cucumbers sandfish (*Holothuria scabra*) and the golden sandfish (*H. lessoni*) are the most valuable tropical species with highest prices on the international market and these species are available in Malaysian waters. Also there are traditionally valuable species such as gamat species (*Stichopus horrens*) that has more value on the Chinese culture. There has been plenty of interest in recent days for the aquaculture of economically valuable tropical sea cucumbers such as *H. scabra* and *H. lessoni* are distributed in throughout Southeast Asia.

Relatively warm water temperature, wide availability of these species, high water quality, suitable coastal environments and potential sea-based sites for grow out in these regions and the availability of commercial technology makes sea cucumbers in aquaculture look very promising. The typical commercial sea cucumber aquaculture operation includes; a. Broodstock collection and management, b. Spawning stimulation and Fertilisation, c. Larval rearing and Feeding, d. Larval development and settlement, e. Nursery rearing of juvenile, f. juvenile pre grow-out and g. Sea ranching or pond grow-out. The Asian demand for human food and medicinal products from sea cucumbers remains strong and on the increasing trend, promising a ready market for output from commercial sea cucumber culture operations. With long standing experience in sea cucumber aquaculture development, Sea cucumber Consultancy has developed the hatchery technology for mass production of two commercially most important tropical sea cucumbers. Using the state of the art technology, millions of *H. scabra* (sandfish) and *H. lessoni* (golden sandfish) juveniles can be produced routinely in the specialized hatchery. These juveniles can be used for restocking depleted wild fishery or grow them in ponds or lagoons to meet the growing market demands. The hatchery production of indigenous gamat species is also possible with the available technology and that would support local gamat industry. Sea cucumber culture could contribute to the restoration of depleted wild populations and allow sustainable fishery.

Diel burying by the tropical sea cucumber *Holothuria scabra*

S.W. Purcell

Source: Marine Biology 157:663–671 (2010)

Understanding concealment behaviour of marine animals is vital for population surveys and captive-release programmes. The commercially valuable sea cucumber *Holothuria scabra* Jaeger 1833 (Holothuroidea) can display a diel burying cycle, but is it widely predictable. Circadian burying of captive *H. scabra* juveniles, and both juveniles and adults in the wild, was examined in New Caledonia. Groups of ten cultured juveniles in mesh chambers in a tank were monitored for 24 h. Small juveniles (1–5 g) displayed an expected diel cycle of epibenthic foraging in the afternoon and night then burial in sediments in the morning. Burial was related significantly to both light and temperature in combination. Similar groups of juveniles were handled once or three times a day for one week then frequency of emergence during another week was compared to unhandled controls. Handling stress, whether occasional or frequent, significantly suppressed the frequency of their afternoon emergence from sediments for 4 days. In a coastal seagrass bed, burial and emergence of *H. scabra* were monitored during days of opposing tidal cycles in three seasons. Adults seldom buried during the day except in the cool season. At that site, most small hatchery-produced *H. scabra* juveniles were buried during most of the day, while larger juveniles showed little diurnal burying. This study underscores that the circadian behaviours of marine animals can exhibit substantial spatial variation, may be absent at certain sites or seasons, and can be mediated by a complexity of factors that vary over short timescales.

Genetic barcoding of commercial bêche-de-mer species (Echinodermata: Holothuroidea)

S. Uthicke, M. Byrne and C. Conand

Source: Molecular Ecology Resources 10:634–646 (2009)

There are more than 47 species of holothurians used for bêche-de-mer production, many of which are locally overfished. With three exceptions, all bêche-de-mer species are Aspidochirotrida and species identification of many of these is difficult. We analysed available genetic information and newly generated sequences to determine if genetic barcoding with the mitochondrial COI gene can be used to identify bêche-de-mer species. Although genetic data were available for $\pm 50\%$ of bêche-de-mer species, sufficient information and within-species replication were only available for six species. We generated 96 new COI sequences extending the existing database to cover most common species. COI unambiguously identified most bêche-de-mer species providing a genetic barcode for the identification of known species. In addition, conspecific (1.3%) variation and congeneric (16.9%) divergence were well separated ('barcoding-gap') albeit with a small overlap, which may lead to some error if genetic sampling alone was applied for species discovery. In addition to identification of adults, COI sequences were useful to identify juveniles that are often morphologically different. Sequence data showed that large (deep) and small (shallow) morphotypes of *Holothuria atra* are the same species, but suggested potential cryptic species within this taxon. For bêche-de-mer, the COI barcode proved useful in species clarification and discovery, but further genetic and taxonomic work is essential for several species. Some bêche-de-mer clades were problematic with morphologically disparate specimens sharing the same barcode. Our study indicated the presence of undescribed species (*Bohadschia* sp.) and species that constitute separate species in the Indian and Pacific Ocean (e.g. *Holothuria fuscogilva*).

Royal cucumber (*Stichopus regalis*) in the northwestern Mediterranean: Distribution pattern and fishery

R.M. Ramón, J. Leonart and E. Massutí

Source: Fisheries Research Fisheries Research 105:21–27 (2010)

Stichopus regalis is a common sea cucumber that is found in a wide depth range in the Mediterranean beyond 50 m depth. Its five longitudinal muscular bands are a culinary delicacy. It is the most expensive seafood product on the Catalan market, and it can reach up to 130 EUR/kg. Despite its ecological and economical importance, information related to this species is very scarce. The population inhabiting the Mallorca and Menorca continental shelf and slope was studied using data from several different surveys conducted from 2001 to 2009. The spatial distribution of the royal cucumber is strongly aggregated. The population showed a multimodal length–frequency distribution, with individuals ranging from 65 to 295 mm in length. Abundance was highest between 100 and 299 m depth and sizes were largest between 50 and 299 m depth. The length–weight relationship indicates a negative allometry, explained by the fact that the thickness of some parts of the body wall appears to be independent of the size of the individual. Although *S. regalis* has a high price, due to its relatively low abundance it is not a particular target species of the multispecies trawl fisheries, and is captured as a by-catch, with a mean catch per unit effort of 1.78 kg per boat and day. The edible part corresponds to 9.81% of the drained weight of the whole animal.

AChE and EROD activities in two echinoderms, *Holothuria leucospilota* and *Holothuria atra* (Holothuroidea), in a coral reef (Reunion Island, South-western Indian Ocean)

J. Kolasinski, D. Taddei, P. Cuet and P. Frouin

Source: Journal of Environmental Science and Health Part A 45:699–708 (2010)

AChE and EROD activities were investigated in two holothurian species, *Holothuria leucospilota* and *Holothuria atra*, from a tropical coral reef. These organisms were collected from 3 back-reef stations, where temperature and salinity were homogeneous. The activity levels of both AChE and EROD varied significantly between the two species, but were in the range of values determined in other echinoderm species. AChE activity levels were higher in the longitudinal muscle than in the tentacle tegument. Among the several tissues tested, the digestive tract wall exhibited higher EROD activity levels. Sex did not influence AChE and EROD activity levels in both species. Animal biomass and EROD activity levels were only correlated in the tegument tissue of *H. atra*, and we hypothesize a possible influence of age. EROD activity did not show intraspecific variability. A significant relationship was found between AChE activity and Cuvierian tubules time of expulsion in *Holothuria leucospilota*. Individuals collected at the southern site presented both lower AChE activity levels and Cuvierian tubules time of expulsion, indicating possible neural disturbance. More information on holothurians biology and physiology is needed to further assess biomarkers in these key species. This study is the first of its kind performed in the coastal waters of Reunion Island and data obtained represent reference values.

Phylogeny of sea cucumber (Echinodermata: Holothuroidea) as inferred from 16S mitochondrial rRNA gene sequences

K. Kamarul Rahim, H. Ridzwan and U. Gires

Source: Sains Malaysiana 39(2):209–218 (2010)

This study aimed to determine phylogenetic relationship between and among selected species of sea cucumbers (Echinodermata: Holothuroidea) using 16S mitochondrial ribosomal RNA (rRNA) gene. Phylogenetic analyses of 37 partial sequences of 16S mitochondrial rRNA gene using three main methods namely neighbour joining (NJ), maximum parsimony (MP) and maximum likelihood (ML) showed the presence of five main genera of sea cucumbers: *Molpadia* from order Molpadiida and four genera of order Aspidochirotida namely *Holothuria*, *Stichopus*, *Bohadschia* and *Actinopyga*. All of the 17 species obtained from Malaysia distributed among the main genera except within *Actinopyga*. Interestingly, *Holothuria excellens* was out of *Holothuria* group causing *Holothuria* to be paraphyletic. High bootstrap value and consistent clustering made *Molpadia*, *Stichopus*, *Bohadschia* and *Actinopyga* monophyletic. The relationship of *Actinopyga* with the other genera was un-clarified and *Stichopus* was sister to *Molpadia*. The latter finding caused the resolution at order level unclear. The pairwise genetic distance calculated using Kimura 2-parameter model further supported and verified findings from the phylogenetic trees. Further studies with more samples and different mitochondrial DNA genes need to be done to get a better view and verification on the molecular phylogeny of sea cucumbers.

Qualitative and quantitative saponin contents in five sea cucumbers from the Indian Ocean

S. Van Dyck, P. Gerbaux and P. Flammang

Source: Marine Drugs 8:173–189 (2010)

To avoid predation, holothuroids produce feeding-deterrent molecules in their body wall and viscera, the so-called saponins. Five tropical sea cucumber species of the family Holothuriidae were investigated in order to study their saponin content in two different organs, the body wall and the Cuvierian tubules. Mass spectrometry techniques (MALDI- and ESI-MS) were used to detect and analyze saponins. The smallest number of saponins was observed in *Holothuria atra*, which contained a total of four congeners, followed by *Holothuria leucospilota*, *Pearsonothuria graeffei* and *Actinopyga echinites* with six, eight and ten congeners, respectively. *Bohadschia subrubra* revealed the highest saponin diversity (19 congeners). Saponin mixtures also varied between the two body compartments within a given animal. A semi-quantitative approach completed these results and showed that a high diversity of saponins is not particularly correlated to a high saponin concentration. Although the complexity of the saponin mixtures described makes the elucidation of their respective biological roles difficult, the comparisons between species and between body compartments give some clues about how these molecules may act as predator repellents.

The ecological role of *Holothuria scabra* (Echinodermata: Holothuroidea) within subtropical seagrass beds

S.-M. Wolkenhauer, S. Uthicke, C. Burrige, T. Skewes and R. Pitcher

Source: Journal of the Marine Biological Association of the United Kingdom, Cambridge University Press 90:215–223 (2010)

Some sea cucumbers species are heavily exploited as bêche-de-mer for the Asian food industry and the global decline of certain highly sought after species has generated an interest in determining the ecological function of those animals within their ecosystem. This study investigated the ecological role of *Holothuria scabra*, a commercially valuable tropical species closely associated with seagrass beds. Seagrass productivity, seagrass and benthic microalgae (BMA) biomass and organic matter (OM) were measured during two exclusion experiments conducted using *in situ* cages deployed for two months both in 2003 and 2004. Density of *H. scabra* was manipulated in caged exclusions (near-zero density, 'EX'), caged controls (natural densities, 'CC') and uncaged controls (natural density, 'NC'). Seagrass growth was lower when holothurians were excluded (5% in 2003, 12% in 2004). Seagrass biomass decreased in all treatments, but reduction was greater in EX than in controls (18% in 2003, 21% in 2004). Both BMA biomass and OM increased in EX compared to NC/CC (in 2004). From a multivariate perspective, a principal component biplot separated EX from both types of controls in 2004, and multivariate tests based on four attributes supported this separation. These results indicate that seagrass systems may suffer in the absence of holothurians; however, the effect size varied between the two experiments, possibly because experiments were conducted at different times of the year. Nevertheless, our results suggest that holothurian over-fishing could have a negative impact on the productivity of seagrass systems.

Management of sea cucumber stocks: patterns of vulnerability and recovery of sea cucumber stocks impacted by fishing

K. Friedman, H. Eriksson, E. Tardy and K. Pakoa

Source: Fish and Fisheries. Article first published online: 10 Sep. 2010. DOI: 10.1111/j.1467-2979.2010.00384.x

Identifying rates of change in the abundance of sea cucumbers under differing management regimes is fundamental to estimating commercial yields, identifying ecological interactions and facilitating management. Here, we review the status of sea cucumber stocks from a range of Pacific Island countries (Samoa, Tonga, Palau, Fiji and Papua New Guinea), some of which have had a moratorium on exports for up to a decade. We use a time-series approach to look at variation in sea cucumber presence, coverage and density from survey and re-survey data. Results give an appreciation of variation between 'high' status (less impacted) and depleted stocks. Survey data show marked declines in coverage and abundance as a result of artisanal fishing activity, and although species groups were not lost at a country level, local extirpation and range restriction was noted. Resilience and 'recovery' following cessation of fishing varied greatly, both among locations and among the species targeted. Worryingly, even after extended periods of moratorium, the density of some species was markedly low. In many cases, the densities were too low for commercial fishing, and may be at a level where the effective population size is constrained due to 'Allee' effects. From these results, we suggest that management regimes presently employed are generally not well aligned with the level of response to fishing mortality that can be expected from sea cucumber stocks. New adaptive, precautionary approaches to management are suggested, which would allow more timely interventions to be made, while refined information on stock dynamics is sought.

Torres Strait sea cucumber survey, 2009.

T. Skewes, N. Murphy, I. McLeod, E. Dovers, C. BurrIDGE and W. Rochester

Source: CSIRO Wealth from Oceans Flagship Final Report. Report available at: <http://www.pzja.gov.au/resources/publications/scientific.htm>

The Torres Strait sea cucumber fishery has been characterised by boom and bust cycles as the result of resource depletion or price fluctuations. Previous research has resulted in the closure of three highly targeted species (*Holothuria scabra* (sandfish), *H. whitmaei* (black teatfish) and *Actinopyga mauritiana* (surf redfish)), and catch limits for two other high value species (*H. fuscogilva* (white teatfish) and *Thelenota ananas* (prickly redfish)). However, the populations had not been assessed since 2005, and given the likely low fishing activity since then; there was a possibility of a recovery for depleted species.

The sea cucumber populations were surveyed at 113 sites in 5 out of 6 zones in east Torres Strait during a 10 day survey in March 2009. The aim of the survey was to assess the current size and status of sea cucumber stocks, especially focussed on the recovery of closed species, *H. whitmaei* and *A. mauritiana*, and to determine the species split for the Actinopyga clade, with particular emphasis on the *A. echinites* – *A. mauritiana* split.

Sixteen commercial species were observed during the survey. The overall average density of commercial holothurians on the reefs was 329.1 per Ha (± 144.9 , 90 % CI), which equated to a total live wet weight of 18,828 t ($\pm 9,014$ t, 90% CI). The overall species composition in 2009 was similar to previous surveys. The most abundant commercial species in the study area was the low value *H. atra* (lollyfish), followed closely by *Stichopus chloronotus* (greenfish) — together these two species make up 79.1% by number and 50.7% by weight of the commercial sea cucumbers in the study area.

The survey found that the density of *H. whitmaei* had increased significantly since 2005, and was the greatest since surveys began in 1995. Their average size was also the largest of any survey carried in Torres Strait. Comparisons with regional density data indicate that the *H. whitmaei* populations in Torres Strait may have recovered to near natural (unfished) densities, and corroborate with islanders reports of a widespread recovery for this species since it was closed in 2003. This is an important example of the recovery of a depleted sea cucumber population, a recovery period of 7 years, and one of the few thus far documented.

Other high value species, *H. fuscogilva* and *Thelenota ananas*, and an important medium value species, *A. echinites*, were either at stable or higher densities than in previous surveys, therefore this represents a healthy fishery with the potential to provide moderate long term income to local Islander communities, provided it is managed carefully. *A. mauritiana* were still uncommon, however, it is now unlikely that this species was ever a large component of the catch. It is more likely that the surf redfish reported in previous catches was made up of *A. echinites* and *A. miliaris*. These two later species were observed at moderate but highly variable densities.

We consider that most other species are still at virgin or near virgin biomass levels, however, some may have a relatively low fishery stock biomass, and the status of *Bohadschia vitiensis* (brown sandfish) is very uncertain due to the burrowing of this species during the day.

We used the density trend and fishery stock estimate data to recommended conservative Torres Strait wide TACs that could be used in conjunction with developing co-management harvest strategies. Re-opening Black teatfish will likely see renewed interest in the fishery. However, the open ended nature of fishing effort (any Torres Strait Islander can theoretically fish the fishery), and the possibility of large pulses in fishing effort due to community interest and momentum partially spurred on by buyer interest, could see at least localised overexploitation of sea cucumber populations.

The introduction of co-management harvest strategies that limit effort pulses, mitigate localised depletion and collect fishery and fishery-independent data should be part of an ongoing harvest strategy. Such strategies could provide the necessary protection to sea cucumber populations.

Additions to the sea cucumber fauna of Namibia and Angola, with descriptions of new taxa (Echinodermata: Holothuroidea)

A.S. Thandar, M.L. Zettler and P. Arumugam

Source: Zootaxa 2655:1–24 (2010)

This paper records several species of holothuroid echinoderms dredged from shallow waters of the Angolan-Namibian-coastline by the Leibniz Institute for Baltic Sea Research Warnemünde in Germany. Altogether seven species are dealt-with, including five new species, of which one is referred to a new genus *Lanceophora*. The new species include two in-the cucumariid subfamily Colochirinae (*Ocnus placominutus* and *O. paracorbulus*), two in the cucumariid subfamily Cucumariinae (*Panningia pseudocurvata* and *Lanceophora lanceolata*) and one in the family Rhopalodinidae- (*Rhopalodinaria bocherti*). Other species included are

Pseudocnus thandari Natasen Moodley, 2008 and an indeterminate species of *Cladodactyla* in the subfamily Cucumariinae and *Trachythyone fallax* Cherbonnier, 1958a in the subfamily Colochirinae.

Aggregations and temporal changes in the activity and bioturbation contribution of the sea cucumber *Holothuria whitmaei* (Echinodermata: Holothuroidea)

Shiell G.R. and Knott B.

Source: Marine Ecology Progress Series 415:127–139 (2010)

The population density, activity and bioturbation contribution of the sea cucumber *Holothuria whitmaei* was investigated on Ningaloo Reef, Western Australia. Two methods, stratified manta tows and blanket manta tows, recorded population densities between 11.4 and >100 ind. ha⁻¹. Further analysis revealed a heterogeneous pattern of distribution, with individuals tending to aggregate (Moran's I; 0.039; $p < 0.05$) on the outer reef lagoon and particularly in areas of high flow. Densities within aggregations were up to 7.2 times greater than those obtained after blanket manta tows (17.1 ind. ha⁻¹), and 4.5 to 6.3 times greater than those obtained after traditional stratified manta tows (19.3 to 27.1 ind. ha⁻¹). Behavioural studies, including investigations of activity and bioturbation, were conducted over a 2-yr time frame (2002–2003) incorporating 3 monitoring periods within each year: January, April and August. Rates of activity varied diurnally, increasing between morning and afternoon, and seasonally, increasing in April, relative to January and August. Temporal patterns of feeding were more difficult to characterise; although rates of sediment egestion were in many cases higher in the morning, no conclusive diurnal or seasonal patterns could be established. Subsequent regression analysis, however, did find a significant positive correlation between the distance travelled and the volume of sediment egested. The volume of sediment bioturbated by *H. whitmaei* at a population level was found to represent only a small fraction of the sediments available (ca. 2 to 14% per annum), even though the contribution per individual was greater than that of smaller sea cucumber species. However, at maximum densities and typical rates of activity, *H. whitmaei* makes physical contact with approximately 2 times the available coral reef sediments per annum per hectare, simply by crawling. This may represent an important ecological contribution, particularly in light of previously documented links between sea cucumber activity, nutrient recycling and the enhancement of benthic microalgal communities.

The oldest synallactid sea cucumber (Echinodermata: Holothuroidea: Aspidochirotida)

M. Reich

Source: Palaeontol Z. DOI 10.1007/s12542-010-0067-8

Aspidochirote holothurian ossicles were discovered in Upper Ordovician-aged Öjlemyr cherts from Gotland, Sweden. The well-preserved material allows definitive assignment to the family Synallactidae, a deep-sea sea cucumber group that is distributed worldwide today. The new taxon *Tribrachiodemas ordovicicus* gen. et sp. nov. is described, representing the oldest member of the Aspidochirotida. The further fossil record of Synallactidae and evolutionary implications are also discussed.

Diversity of the holothuroid fauna (Echinodermata) in La Réunion (Western Indian Ocean)

Conand C., Michonneau F., Paulay G. and Bruggemann H.

Source: Western Indian Ocean Journal of Marine Science (2010)

Echinoderms are conspicuous components of the tropical fauna and play important roles in the functioning of coral reefs. However their diversity is not as well documented as that of other conspicuous reef organisms, like corals or fishes. We review current knowledge of the diversity of the class Holothuroidea on La Réunion. Several recent initiatives, including the Masma (Conand and Muthiga 2007) and BIOTAS projects, have considerably augmented the number of species known from the island. As a result of these surveys, the recorded holothuroid fauna was doubled. Thirty-six species are now recognized, 17 of which are new records for the island. The order Aspidochirotida, which includes the largest and most conspicuous holothuroids, is the most diverse, with 28 species. Six species of Apodida and two species of Dendrochirotida round out the fauna. These latter groups, especially, may prove more diverse with further investigation. The island's holothuroid fauna is compared with data available from other areas of the Western Indian Ocean and the Indo-west Pacific to evaluate its biogeographic relationships.

The sea cucumber resources and fisheries management in the Western Indian Ocean: Current status and preliminary results from a WIOMSA regional research project.

Conand C. and Muthiga N.

Source: Echinoderms: Durham. Harris et al. (eds) Taylor and Francis, London. 575–581. (2010)