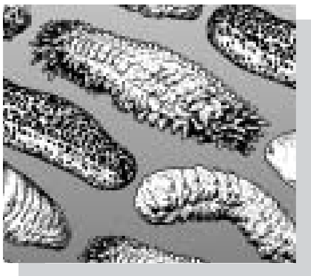

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beche-de-mer

# Larval development and juvenile growth of the Galapagos sea cucumber *Isostichopus fuscus*

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## Abstract

This study presents preliminary results on the early development and growth of the sea cucumber *Isostichopus fuscus* in land-based installations on the coast of Ecuador. This species has been intensively fished along the mainland and around the Galapagos Islands, where efforts at management have always met strong opposition from local communities. Ecuadorian populations of *I. fuscus* have thus been severely depleted over the past decade. The data presented here show that this species can be reared in captivity, thus providing an alternative to fisheries, or a way to maintain sustainable harvests and eventually contribute to the restoration of the natural populations. Data pooled from three trials indicate that juveniles can be grown to a size of ca 3.5 cm in length in less than three months. They are then fit to be transferred to grow-out ponds or to be released in the field.


## Introduction

*Isostichopus fuscus* (Fig. 1) is a deposit-feeding sea cucumber that is mainly found on reefs and sandy bottoms along the western coast of the Americas, from northern Peru to Baja California, Mexico (Castro 1993; Toral 1996; Sonnenholzner 1997; Gutierrez-Garcia 1999). Like many other commercial species, *I. fuscus* has been widely fished over the past decades to meet the growing demand for beche-de-mer on the major Asian markets. As the waters along mainland Ecuador became depleted, the fisheries shifted to the Galapagos Islands, raising international apprehension over the fate of this unique archipelago, which has been recognised as a national park and marine reserve.

**Figure 1.** ***Isostichopus fuscus*** **adults collected along the coast of Ecuador.**

In spite of the worldwide concern, the Galapagos sea cucumber populations became the focus of intensive and poorly managed exploitation in the early 1990s. Since then, governmental attempts at regulating sea cucumber harvests, and banning them in some areas, have met strong opposition from local fishermen in Ecuador. In fact, illegal fish-

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eries have always been a preoccupation and still occur along the mainland, around the Galapagos Islands and elsewhere in the distribution area of *I. fuscus*. Official information on the fisheries and actual total catches are consequently difficult to obtain and remain sparse (Salgado-Castro 1993; Sonnenholzner 1997; Gutierrez-Garcia 1999; Jenkins and Mulliken 1999). Nevertheless, recent data and reports on average capture sizes (Sonnenholzner 1997; Martinez 2001) indicate that *I. fuscus* populations have declined drastically and that natural stocks may irreversibly crash in the near future.

In spite of this alarming situation, a very limited number of studies have been conducted on the reproductive biology, spatial distribution, population structure, growth and survival rate of this species (Herrero-Perezrul 1994; Fajardo-Leon et al. 1995; Toral 1996; Sonnenholzner 1997; Herrero-Perezrul et al. 1999).

Some authors have mentioned that aquaculture and stock enhancement should be investigated as possible solutions to the current *I. fuscus* crisis (Gutierrez-Garcia 1995, 1999; Fajardo-Leon and Velez-Barajas 1996; Jenkins and Mulliken 1999). However, to our knowledge, no results have ever been presented on the captive breeding of the species.

Until recently, aquaculture in Ecuador was largely focused on shrimp. The emergence of white spot disease in 1999–2000 has severely harmed the industry and resulted in the bankruptcy and closing of numerous farms. Consequently, Ecuador now has a lot of shrimp farm infrastructures that could very well be put to use for the development of other species, such as sea cucumbers.

The present paper brings forward preliminary results on the larval development and juvenile growth of *I. fuscus* in land-based nursery systems on the coast of Ecuador. The data show that aquaculture of this species is feasible and that it could potentially be developed as an alternative to fisheries. Then again, it could be used to maintain sustainable harvests and eventually contribute to the restoration of the natural populations. Further research to complement the present work is being conducted on the feeding, growth and reproductive biology of this highly prized sea cucumber, which is a dominant feature of the Ecuadorian marine ecosystem. In time, aquaculture and stock enhancement of *I. fuscus* might provide part of the solution to the Galapagos sea cucumber crisis.

## Larval development of *Isostichopus fuscus*

*I. fuscus* possess oligotrophic, transparent larvae that follow an indirect development, meaning that the larvae need to feed during their pelagic phase and undergo a series of transformations to reach the juvenile stage (Fig. 2, 3 and Table 1). In most trials, the development, settlement and early growth of the juveniles were somewhat asynchronous, and different stages and sizes could be found simultaneously in the culture. Extreme examples were observed in a few tanks where residual auriculariae neighboured 4-mm long juveniles. Table 1 provides a developmental kinetic that is based on the observation of the bulk of the culture, discarding the asynchronous animals.

Ovulation in *I. fuscus* occurs in the gonadal tubule as the oocytes are released (Fig. 2A). Thus, fully mature oocytes (ca 120 µm in diameter) are expulsed di-

**Table 1. Development of *Isostichopus fuscus*, from fertilisation to 35-mm long juvenile, at a salinity of 34–35, a temperature between 22 and 29°C, a pH of 8.4–8.5 and an oxygen level varying between 5.4 and 6.1 mg $L^{-1}$.**

| Stage | Time |
|---|---|
| Fertilisation | 0 |
| Elevation of the fertilisation envelope | 4 min |
| Expulsion of the first polar body | 7 min |
| Expulsion of the second polar body | 9 min |
| 2-cell | 52 min |
| 4-cell | 70 min |
| 8-cell | 95 min |
| 16-cell | 124 min |
| 32-cell | 140 min |
| Blastula | 3 h |
| Early gastrula | 6 h |
| Hatching | 10 h |
| Late gastrula (elongation) | 14 h |
| Early auricularia | 1–2 d |
| Auricularia | 3–15 d |
| Late auricularia (early metamorphosis) | 16–18 d |
| Doliolaria | 19–24 d |
| Early pentactula | 21–26 d |
| Settlement (metamorphosis completed) | 22–27 d |
| Juvenile, 1 mm | 28 d * |
| Juvenile, 2 mm | 30 d |
| Juvenile, 3 mm | 32 d |
| Juvenile, 4 mm | 38 d |
| Juvenile, 5 mm | 40 d |
| Juvenile, 8 mm | 44 d |
| Juvenile, 10 mm | 47 d |
| Juvenile, 15 mm | 51 d |
| Juvenile, 20 mm | 56 d |
| Juvenile, 25 mm | 63 d |
| Juvenile, 30 mm | 69 d |
| Juvenile, 35 mm | 72 d |

* For the juvenile stages, the time indicated corresponds to the first noteworthy observations of a particular size in the tanks.

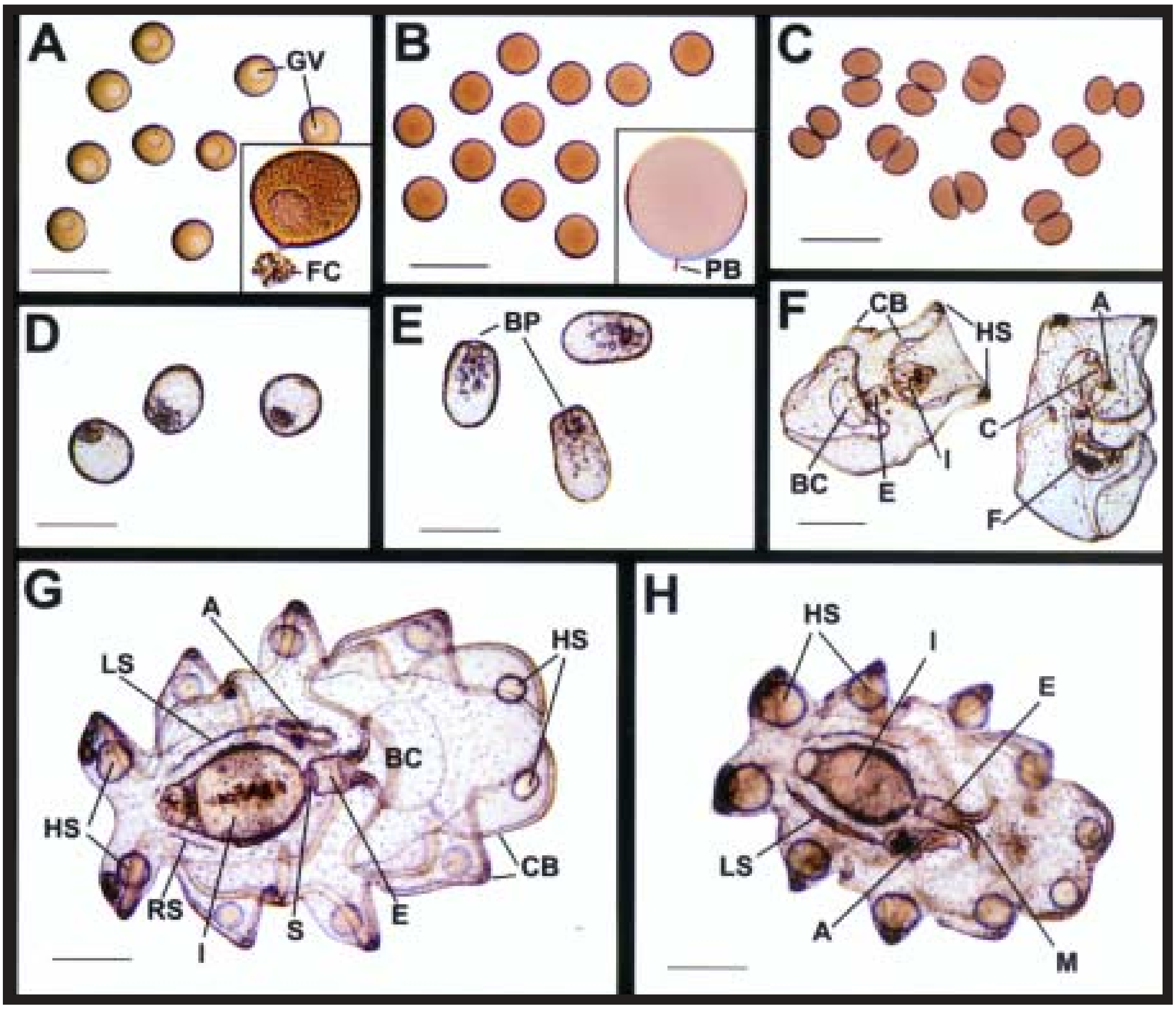


**Figure 2.** **Early development of the sea cucumber *Isostichopus fuscus*.**

The bars represent 200 µm. A. Oocytes collected surgically from a mature gonad. The germinal vesicle (GV) is clearly visible. The insert shows a close-up of an ovulating oocyte with the follicular cells (FC) still attached to it. B. Fully mature, newly fertilised eggs with clear germinal vesicle breakdown. The insert shows the expulsion of the two polar bodies (PB). C. 2-cell stage. D. Newly hatched gastrula. E. Elongated gastrula with visible blastopores (BP). F. Early auricularia on which the ciliary bands (CB), hyaline spheres (HS), buccal cavity (BC), oesophagus (E), intestine (I), cloaca (C) and anus (A) are identifiable. Food items (F) are present in the buccal cavity. G. Ventral view of a fully developed auricularia showing the left somatocoel (LS), axohydrocoel (A), hyaline spheres (HS), ciliary bands (CB), buccal cavity (BC), oesophagus (E), sphincter (S), intestine (I) and the right somatocoel (RS). H. Dorsal view of a metamorphosing auricularia. With a noticeable decrease in size, the buccal cavity disappears and the hyaline spheres (HS) are pulled closer together. The mouth (M), intestine (I), oesophagus (E), left somatocoel (LS) and axohydrocoel (A) are clearly visible.

rectly in the water column at the metaphase-I of meiosis, after the germinal vesicle breakdown.

The development of *I. fuscus* is initiated with the elevation of the fertilisation envelope, roughly 4 minutes after fertilisation. The expulsion of the first polar body occurs ca 3 minutes later (Fig. 2B). The second polar body follows rapidly within ca 2 min. The first cleavage is equal, radial and holoblastic and divides the cell into two equal hemispheric blastomeres (Fig. 2C). The second cleavage again occurs along the animal-vegetal axis, yielding more spherical blastomeres. Embryos hatch from the fertilisation envelope as early gastrulae, ca 10 h after fertilisation (Fig. 2D). These early gastrulae swim with the help of cilia covering their entire surface; they elongate into full-size gastrulae after ca 14 h (Fig. 2E). Auricularia larvae begin to appear ca 24 h after fertilisation; they constitute the first feeding stage. Growing auriculariae can be observed during the next two weeks of culture (Fig. 2F, Table 1). At this stage, they begin to accumulate hyaline spheres. The oesophagus, the sphincter, the intestine, the cloaca as well as the anus are clearly visible. After 16–18 days, the auricularia reaches its maximum size of 1.1–1.3 mm; it has left and right somatocoels, as well as an axohydrocoel (Fig. 2G).

In the following hours, many auriculariae initiate the transformation that will lead to the doliolaria stage (Fig. 2H). In the course of this process, the larvae shrink down to nearly 50 per cent of their initial size, the buccal ciliated cavity disappears and the hyaline spheres are pressed closer together (Fig. 3A). The doliolaria stage is reached ca 19–24 days after fertilisation (Fig. 3B, Table 1) as the larvae stop feeding and the cilia are aligned in five distinct crowns along their cylindrical body. At this time, the movement of the primary tentacles can be observed through the translucent body wall. The somatocoel is also visible. A few days later, the doliolaria transforms into an early pentactula pos-

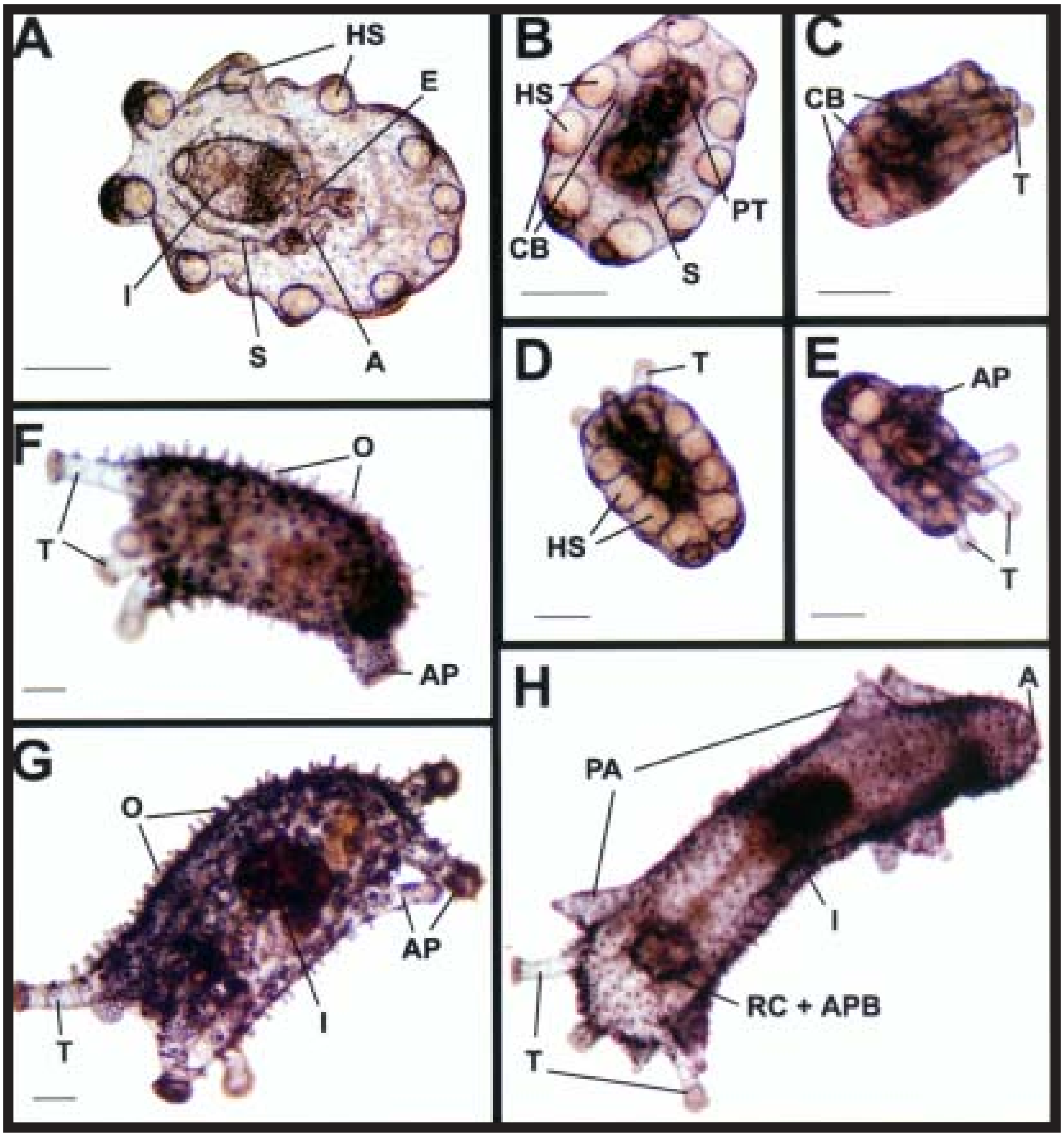


**Figure 3.** **Late development of the sea cucumber *Isostichopus fuscus*.**

The bars represent 200 µm. A. Late metamorphosing auricularia, showing the hyaline spheres (HS), oesophagus (E), intestine (I), somatocoel (S) and axohydrocoel (A). B. Fully developed doliolaria with hyaline spheres (HS), primary tentacles (PT), ciliary bands (CB) and somatocoel (S). C. Early pentactula with 5 tentacles (T) and the still visible ciliary bands (CB). D. Dorsal view of newly settled pentactula with tentacles (T) and hyaline spheres (HS). E. Ventral view of newly settled pentactula showing the first ambulacral podia (AP) and the 5 buccal tentacles (T). F. Early juvenile, measuring 1.5 mm in length, with tentacles (T), ambulacral podia (AP) and ossicles (O). The hyaline spheres have disappeared. G. A 2-mm long juvenile with 5 tentacles (T) and 3 pairs of ambulacral podia (AP). The intestine (I) and ossicles (O) are visible. H. A 3-mm long juvenile showing the tentacles (T), papillae (PA), intestine (I), anus (A) and the ring canal and aquapharyngeal bulb (RC + APB).

sessing five buccal tentacles (Fig. 3C). At this stage, the larvae remain close to the substrate, successively going through swimming and settling phases. Definitive settlement, with the complete loss of cilia, completion of metamorphosis and emergence of the two first ambulacral podia, occurs ca 22–27 days after fertilisation (Fig. 3D, E).

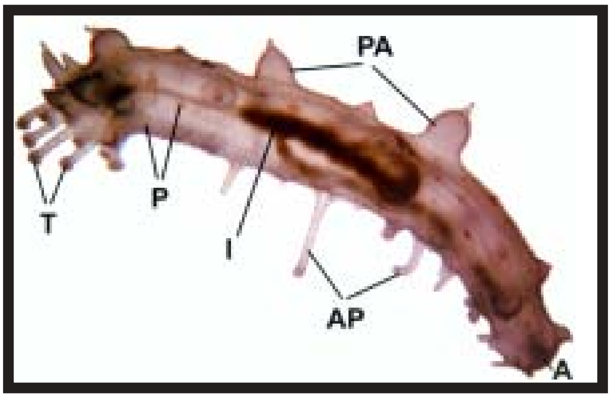


**Figure 4.** **Juvenile sea cucumber *Isostichopus fuscus* measuring 1.5 cm in length and showing the tentacles (T), early body wall pigments (P), intestine (I), ambulacral podia (AP), anus (A) and papillae (PA).**

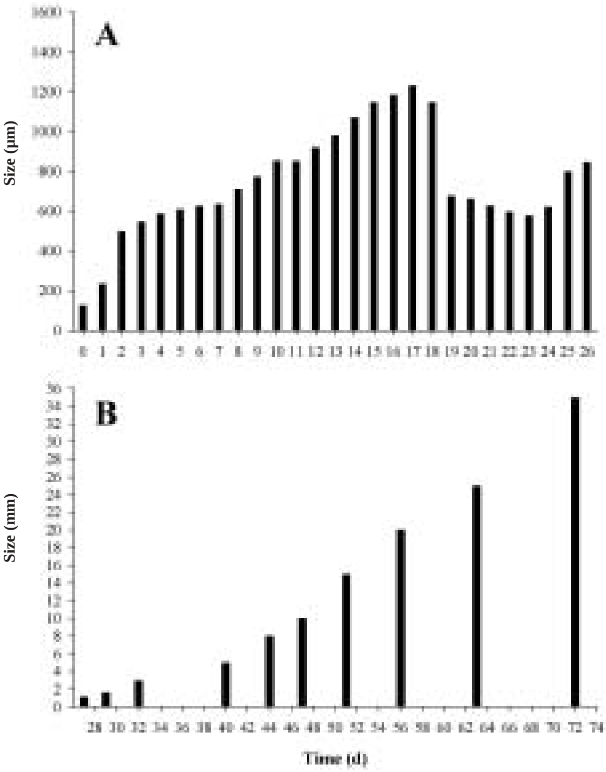


**Figure 5.** **Average growth of the larvae (A) and juveniles (B) of the sea cucumber *Isostichopus fuscus*. Note that the x axis in B is a prolongation of the one in A, with a slightly different scale, and that size is expressed in µm in A and in mm in B.**

## Juvenile growth of *Isostichopus fuscus*

Although the first settled juveniles can be observed as early as day 22, a majority of juveniles measuring 1–1.5 mm in length are generally recorded in the tanks after 28 days of culture (Fig. 3F, Table 1). They reach ca 2–3 mm only a few days later (Fig. 3G, H), and 5 mm after ca 40 days. The juveniles continue to grow at a rate of ca 0.5–1.0 mm per day for the next 3–4 weeks. When they are ca 5 mm in length, the juveniles start to accumulate reddish-brown pigments. In 8-mm long juveniles, the tip of the tentacles becomes ramified. After 52 days of culture, the juveniles are 1.5–1.8 cm long and 4 mm wide (Fig. 4). They possess several papillae and an elongated intestine that already exhibits strong peristaltic movements. The body wall becomes more opaque as the ossicle density and the tegument thickness increase. When the juveniles reach ca 2 cm in length, the whitish coloration that characterises the early stages of life is gradually replaced by a brownish tinge similar to the one observed in adults. After approximately 72 days of culture, the juveniles are ca 3.5 cm long and 1 cm wide and are ready to be released in outdoor ponds, or in the field, to complete their growth.

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# Papua New Guinea sea cucumber and beche-de-mer identification cards

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## Introduction

Since September 2001, the Papua New Guinea beche-de-mer fishery has been governed by the National Beche-de-mer Fishery Management Plan. Management measures include regulations on access, size and catch limits, storage and export. To facilitate the monitoring of this fishery and enforcement of the regulations, the National Fisheries Authority (NFA) decided to produce a booklet showing the different sea cucumber species traded in the region. They approached the Secretariat of the Pacific Community (SPC) Marine Resources Division Information Section to seek their assistance for this project.

During the last 19 years, SPC has published several versions of a sea cucumber identification guide: *Bêche-de-mer of the South Pacific Islands* (1975); *Bêche-de-mer of the tropical Pacific* (1979); and *Sea cucumbers and beche-de-mer of the tropical Pacific – A handbook for fishers* (1984). This last edition presents pictures, in full colour, of 15 holothurian species and their corresponding beche-de-mer (dried) product. Thousands of copies of have been distributed through the Pacific, and copies of the last edition are still regularly sent upon request. Unfortunately, it is now almost out of print and some of the information it contains is out of date: several species that were not listed as of commercial value in 1994 are now traded and some of the species that were categorised as low value in the handbook have since "climbed the ladder". The handbook was not fully adapted to NFA needs, so it was decided to produce a new sea cucumber identification guide, specially designed for PNG.

An identification guide is mostly used in the field, so it has to be easy to carry around and waterproof. Therefore, instead of a handbook, we decided to make a set of pocket-size (95 x 135 mm) cards printed on plastic (as in credit cards) and bound together by a plastic pin. Each card presents one species of sea cucumber, with a full-colour underwater picture of the live animal on one side and two pictures (ventral and dorsal views) of the corresponding dried product (beche-de-mer) on the other side (Fig. 1). It also contains some basic information on the species (preferred habitat and depth, average sizes) and a short description of the dried product. The complete set contains 24 cards, presenting 20 different species, giving some basic information on beche-de-mer processing and listing the main regulations pertaining to the beche-

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