Sandfish (*Holothuria scabra*) with shrimp (*Penaeus monodon*) co-culture tank trials

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Abstract

Seven tank trials were undertaken involving the co-culture of sandfish and shrimp on sand. Sizes, stocking densities, feeding and other conditions were all varied. Results indicated that powdered sargassum did not support sandfish growth, although it may have prevented starvation, which otherwise killed juveniles within three to six weeks. Sandfish grew well on commercial shrimp starter feed, reaching densities of 300 g m^{-2} or more, but feeding rates much above 1 g total dry weight m^{-2} day^{-1} could lead to anaerobic benthic conditions and sandfish mortalities. Growth was better with higher, rather than lower, water exchange rates. Semi-opaque covers slightly reduced sandfish growth, although appearing to increase survival of small shrimp.

Sandfish were never seen to have a negative impact on shrimp growth or survival. The effect of shrimp on sandfish, however, was more complex. Survival and growth of sandfish were often as good with shrimp as without, and sometimes apparently better. This may be because shrimp improved the benthic conditions by eating excess food, or for other (unknown) reasons. In several trials, however, it seemed that shrimp caused the deaths of sandfish, and in some cases, it was clear that they did so. A range of sizes for which predation did or did not occur is shown.

Introduction

A major objective of the sea cucumber rearing project (2000–2003) of the WorldFish Center (previously ICLARM) and the Vietnam Ministry of Fisheries at RIA3 (Research Institute for Aquaculture no 3), was to look at ways to combine the culture of sandfish (*Holothuria scabra)* with that of shrimp (*Penaeus monodon).* Farming *P. monodon* is an important activity in Khanh Hoa Province, which has over a thousand hatcheries and many areas of sea and brackish-water ponds. As elsewhere in the world, disease problems make shrimp aquaculture a high-risk business in which big profits or big losses can be made. Many ponds stand empty at least part of the time, and farmers look for additional or alternative culture systems and species.

Empty ponds, particularly those in areas of generally high salinity (which many farmers believe are less favourable for shrimp), represent a potential resource that could be used for culturing sandfish (either by itself or in conjunction with shrimp). This approach could lead to either commercial hatchery-based sea cucumber farming or large-scale restocking of overfished sea areas.

Co-culturing is a particularly interesting proposition. By providing an extra crop with few extra costs (apart from the juveniles themselves and in particular using no additional feed), sandfish might make shrimp culture at lower densities more economically attractive. This could help reduce the environmental impact of shrimp farming. Sandfish would live off benthic organic material, and should therefore also clean the pond floors to some extent.

Seven tank trials, examining the co-culture of sandfish and shrimp, are summarised in this report, in chronological order. With the exception of trial C, trials are of the second nursery phase, using sandfish juveniles bred in the project hatchery. These juveniles passed through the first nursery phase in bare outdoor tanks. In these trials they were put on a sand substrate. Sand came initially from a reasonably clean beach source, but was not cleaned during the course of the trials. (Some tank and also pond trials were carried out by RIA3 scientists under a separate DANIDA-funded project, usually with juveniles from the same hatchery and nursery. These are not described here.)

Presentation of the different trials

Trial A: Two stocking densities, with and without shrimp and shrimp feed, and with and without seaweed powder

Eight fibreglass tanks (1.5 m diameter with water depth 65 cm — 1.76 m^2 floor area and 1.15 m^3 volume) were installed outdoors in two adjacent rows (numbered 1, 3, 5, 7 and 2, 4, 6, 8). Tanks were cov-

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ered with a single layer of 60% shade netting. Fine washed beach sand (30 L) was put in each tank, covering the floor to a depth of about 17 mm. Double sand-filtered seawater was supplied via 2 mm nozzles so that flows in all tanks were within 15% of each other even with varying supply pressure. Water generally flowed for 8–12 daylight hours, with a mean exchange rate of about 300–400 L tank^{-1} day^{-1} (i.e. about one third of the tank volumes). Tank water temperature, measured only occasionally, was usually between 29 and 31°C.

On 31 May 2001, the eight tanks were stocked with small sandfish, averaging 0.94 g using matched groups of 36 (low density) or 72 (high density) animals per tank (20 or 40 m^{-2} of tank floor). Four of the tanks were also stocked with 80 postlarval shrimp per tank. Sandfish were weighed after about five minutes of shade drying after 20 and 39 days. Shrimp were sampled after 20 days, and fully harvested and weighed after 39 days.

All four tanks with shrimp were fed with Betagro 501S juvenile shrimp feed, generally given in three equal amounts per day. After an initial overfeeding error in the first two days, feeding rates were cut back and then gradually increased over the course of the experiment as animals grew. Over the first period, feedings averaged 1.5 g tank^{-1} day^{-1} (0.9 g m^{-2}), and over the second period 2.4 g tank^{-1} day^{-1} (1.4 g m^{-2}). Four tanks (two with shrimp and two without) were also fed every second day with a finely ground powder made from dried seaweed, mostly sargassum: those with sandfish stocked at low density received 1.7 g every two days for the first period and 4.6 g every two days for the second, while those with sandfish at high density were fed at twice this rate. Finally, sandfish in two of the tanks with no shrimp received no feed at all.

Conclusions from trial A

1. Rapid sandfish growth occurred only in tanks with shrimp and shrimp food. Growth continued with sandfish biomass reaching densities of over 300 g m^{-2}. In these tanks powdered seaweed had little effect.
2. In the two tanks with no food, most sandfish died during the second period. In other tanks, sandfish survival rates averaged almost 94%.
3. Seaweed alone may have helped keep sandfish alive, but did not support good growth.
4. Low sandfish stocking densities generally gave better individual growth rates than high stocking densities.
5. Shrimp survival rates averaged better than 60% from postlarvae to above 0.4 g average weight.

Trial B: With shrimp food, with and without shrimp, with and without Gracillaria and chicken manure

Eight 1.5 m diameter tanks were set up as in trial A, although with slightly less sand in each (24 L). On 11 July 2001, tanks were stocked with matched groups of 36 juveniles, averaging 0.83 g per tank (20 m^{-2} of

Table 1. **Treatments and results of trial A**

tank		1	2	3	4	5	6	7	8
treatment	sandfish density	high	low	low	high	high	low	low	high
	shrimp + feed	yes	no	yes	no	no	yes	no	yes
	weed	high	low	no	no	high	low	no	no
20 June									
sandfish	number	71	35	35	64	73	34	36	68
	average wt. (g)	5.6	1.8	7.3	0.7	1.1	5.4	1.2	3.7
	density (g m^{-2})	227	36	144	27	46	103	25	143
	growth rate (g day^{-1})	0.24	0.04	0.32	-0.01	0.01	0.22	0.01	0.14
shrimp	average wt. (g)	0.12		0.09			0.10		0.11
10 July									
sandfish	number	61	36	35	6	70	32	16	66
	average wt. (g)	9.9	4.0	16.0	2.0	1.7	17.2	1.6	10.7
	density (g m^{-2})	342	81	317	7	67	311	14	401
	growth rate (g day^{-1})	0.21	0.11	0.44	0.06	0.03	0.59	0.02	0.35
	survival (%)	86	103	100	9	96	94	44	97
shrimp	number	48		33			58		54
	average wt. (g)	0.41		0.48			0.41		0.43
	survival (%)	60		41			73		68

tank floor). Half the tanks were also stocked with 80 postlarvae shrimp in each (about 45 m^{-2}).

All tanks, including those without shrimp, were fed with Betagro 501S juvenile shrimp feed, generally given in three equal amounts per day. Half the tanks were also fed every second day with fresh chopped and blended *Gracillaria* plus ground and blended (dried) chicken manure. There were, thus, four different feeding treatments, each with two replicates. Feeding rates were gradually increased with time (Table 2). After 22 days all sandfish and samples of shrimp from each tank were weighed. After a further 18 days, the remaining animals were counted and weighed (except, unfortunately, for shrimp from tank 1, which were only sampled).

Shrimp grew well, with survival rates of 59–95% in the three tanks where they were counted. There were heavy sandfish mortalities, however, during the second period in most tanks, with black and smelly sand. Sandfish survival was good only in the two tanks with shrimp and without *Gracillaria* or manure.

The experiment apparently failed in the second period because feeding rates were too high, with all tanks receiving shrimp food at 1.36 g m^{-2} day^{-1} for the last 16 days. This was made worse in tanks that also received large amounts of *Gracillaria* and manure.

Table 2. Feeding rates in trial B

all tanks (g $tank^{-1}$ day^{-1})		tanks 1, 3, 6, 8 (g $tank^{-1}$ 2 $days^{-1}$)		
dates	shrimp food (dry)	dates	*Gracillaria* (fresh)	manure (dry)
12/7	1.50	14/7–24/7	12.50	5.0
13/7–24/7	0.90	26/7–3/8	18.75	7.5
25/7–3/8	1.50	5/8–19/8	30.00	12.0
4/8–19/8	2.40			
average g day^{-1}	**1.64**		**9.75**	**3.9**

Table 3. Treatments and results in trial B

tank		1	2	3	4	5	6	7	8
shrimp		yes	no	no	yes	yes	no	no	yes
Gracillaria + manure		yes	no	yes	no	no	yes	no	yes
02 Aug.									
sandfish	number	34	31	21	34	30	36	35	32
	av. wt. (g)	1.8	2.5	1.7	2.4	2.1	2.5	2.7	3.5
	survival (%)	94	86	58	94	83	100	97	89
shrimp	av. wt. (g)	0.062			0.043	0.046			0.088
20 Aug.									
sandfish	number	0	0	13	32	26	2	0	4
	av. wt. (g)			5.4	10.1	3.2	1.2		5.4
	survival (%)			36	89	72	6		11
shrimp	av. wt. (g)	0.48			0.37	0.26			0.43
	number	?			76	62			47
	survival (%)	?			95	78			59

Conclusions from trial B

1. Feeding rates of shrimp food appear to have been too high (1.36 g m^{-2} day^{-1}) in the second period. The problem was exacerbated in tanks also receiving *Gracillaria* and manure.
2. Sandfish are more vulnerable to foul benthic conditions than are shrimp.
3. The main beneficial effect of shrimp on sandfish was probably just to consume the excess food.

Trial C: Large sandfish with and without shrimp, with shrimp feed, and with seaweed

This experiment was set up to take a preliminary look at interactions between large sandfish (of wild origin) and shrimp, as might occur during growout in ponds.

It was thought that the presence of shrimp might be of direct benefit to sandfish, with, perhaps, shrimp faeces providing a better food source than shrimp feed. On the other hand, large shrimp were seen sitting on large sandfish, and it was feared that the shrimp might damage sandfish skin or prevent wounds from healing.

Five, 6 m^3 outdoor concrete tanks were used. Tank floors were covered with washed beach sand to a depth of about 10 mm in all except tank 2, which had sand to about 25 mm depth; the extra sand depth was to allow sandfish to bury themselves in case they needed protection from shrimp. Water flows of about 500–1000 L day^{-1} were provided from a reservoir plus settlement tank via nozzles. Initially water was sand-filtered, although filtration was later omitted.

On 6 September 2001 sandfish were divided into groups of seven animals each. Individual weights ranged from about 40–

440 g, but the groups were "matched" so that the mean of each group was between 160 and 170 g. By using such a wide range of sizes, individuals could be followed through successive weighings. Feeds were either commercial shrimp food (Betagro 503) or seaweed (mainly sargassum), milled dry and then sieved (< 250 μm) to produce a fine powder.

There were five treatments; individually identifiable sandfish provided the only repetitions. Tank 1 contained no shrimp and was given 3 g shrimp food per day. Tanks 2 (extra sand) and 4 were stocked with approximately equal numbers and weights of shrimp (~ 17.4 m^{-2}), averaging about 1.6 g and given 6 g shrimp food per day. Tank 3 contained no shrimp, was given 3 g seaweed powder per day, and tank 5 contained no shrimp and was not given food. Sandfish were collected and weighed on six occasions covering a period of 107 days. Shrimp were counted and weighed only at the beginning and end of the trial.

All sandfish survived and appeared healthy in all tanks with no shrimp. In tank 2, one sandfish eviscerated on final handling (viscera were included in the weighing), perhaps indicating it was weak. In tank 4, on 24 October, one sandfish had skin lesions; on 5 November, there were two sandfish with lesions; and on 22 December, only the three largest sandfish were found.

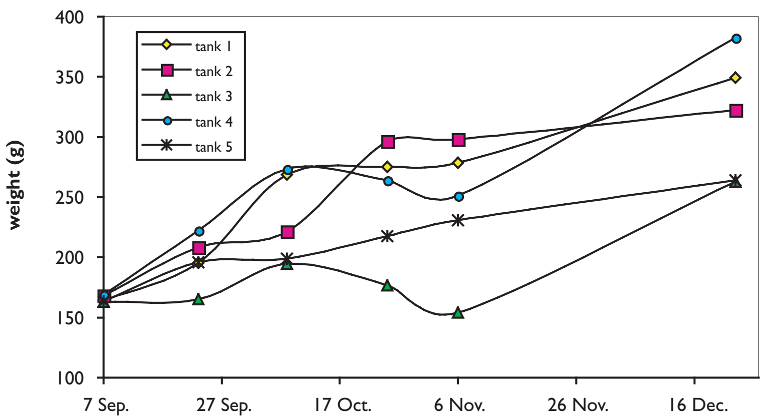

Figure 1. Average weight (g) of sandfish with time in trial C

Conclusions from trial C

1. Reasonable sandfish growth rates were obtained in tanks at densities up to almost 400 g m^{-2}.
2. Shrimp were not needed, shrimp food alone appeared to support this growth.
3. Feeding sandfish with seaweed powder did not appear any more beneficial than not feeding them at all.
4. Shrimp of an average weight less than 6 g may have caused mortalities of 70–190 g sandfish in tank 4.
5. Extra sand or other factors may have protected sandfish in tank 2.
6. It is not clear why shrimp survival was low in tank 2.

Table 4. Tank treatments, sandfish survival, densities and growth rates (ranked) in trial C

tank	sand (mm)	shrimp	feed type	feed (g day^{-1})	survival (%)	initial density (g m^{-2})	final density (g m^{-2})	mean growth (g day^{-1})
1	10	no	shrimp	3	100	187	399	1.700
2	25	yes	shrimp	6	100	192	369	1.410
4	10	yes	shrimp	6	43	193	219	*1.100
5	10	no	none	-	100	187	302	0.920
3	10	no	weed	3	100	186	301	0.916

* for all animals (1.65 for the three largest which survived to end of trial)

Table 5. Shrimp stocking and harvest data in trial C

	stock (6/9/01)		harvest (15/12/01)		
	number	average wt. (g)	number	average wt. (g)	survival (%)
tank 2	107	1.56	21	9.67	19.6
tank 4	102	1.64	67	5.66	65.7

Trial D: Factorial: size, shrimp, tank covers and flow

Sixteen fibreglass tanks (1.5 m diameter, depth to outlet 65 cm) were installed in two adjacent rows outdoors. The site was near a row of coconut trees, but was otherwise unshaded. Beach sand was washed and sieved through 400 x 800 μm mesh. A sample of this sand was sieved while wet, using available screens, and then dried and weighed. Size distribution was > 250 μm: 9.6%, > 140 μm: 78.9%, > 100 μm: 6.3%, and > 50 μm: 5.3%. Eight litres of this sand were put into each tank. The following four factors were tested, in 16 combinations, randomly assigned.

a) Size
Juveniles from bare nursery tanks were sorted into matched groups and then sorted into groups of 24 large or 36 small animals. They were weighed after three to five minutes of drying and after removing remaining excess water with a towel. Total weights of large juvenile groups were within 7% of each other, those of small juveniles were within 20%. Large juveniles averaged 0.53 g, small juveniles 0.12 g.

b) Shrimp
Eighty postlarval *P. monodon*, recently supplied from a hatchery, were stocked in each of eight tanks.

c) Covers
Blue semi-translucent PVC roofing panels (2 m x 80 cm) were put on eight of the tanks, using two slightly overlapping panels per tanks. A timber batten was used to give a slight slope to the panel and a small gap was left at one edge to facilitate feeding and checking. Panels reduced water temperature fluctuations, rain and dirt (mainly coconut tree debris), but they also substantially lowered light levels and, therefore, photosynthesis, which might be important in sandfish feeding.

d) Flow rate
Tanks were supplied with unfiltered seawater via nozzles of two sizes from a header/settlement tank, whose water level varied throughout the day. (Water was pumped from the sea nearly always during daylight hours.) At mid-level, flows were approximately 1800 and 600 ml min^{-1} for the two nozzle sizes, giving retention times of about one and three days.

Table 6. **Treatments and results of trial D**

tank		treatment			15 Dec.		14 Jan.				7 Feb.				28 Feb.					
					sandfish		sandfish			shr.	sandfish			shr.	sandfish			shrimp		
	size	shr.	cover	flow	no.	av. wt. (g)	no.	av. wt. (g)	grow. (g day^{-1})	av. wt. (g)	no.	av. wt. (g)	grow. (g day^{-1})	av. wt. (g)	no.	av. wt. (g)	grow. (g day^{-1})	no.	av. wt. (g)	surv. (%)
1	small	-	cover	low	36	0.12	32	2.73	0.09		37	10.1	0.31		37	14.1	0.19			
2	large	-	open	high	24	0.52	23	5.76	0.17		24	17.6	0.49		24	19.8	0.10			
3	small	-	open	high	36	0.11	35	3.03	0.10		36	12.6	0.40		36	20.2	0.36			
4	small	+	cover	low	36	0.12	23	7.52	0.25	0.07	23	33.7	1.09	0.27	23	36.7	0.14	60	0.68	75
5	large	+	open	high	24	0.55	24	12.67	0.40	0.13	24	41.8	1.21	0.22	24	50.6	0.42	45	0.69	56
6	large	+	open	low	24	0.55	24	12.04	0.38	0.13	24	20.8	0.37	0.32	22	32.0	0.53	38	0.82	48
7	large	-	open	low	24	0.54	23	6.83	0.21		24	15.4	0.36		23	12.7	-0.13			
8	large	+	cover	high	24	0.55	22	14.68	0.47	0.13	22	41.4	1.11	0.29	20	40.5	-0.04	75	0.66	94
9	small	-	open	low	36	0.12	34	2.88	0.09		36	10.8	0.33		36	10.9	0.00			
10	large	+	cover	low	24	0.54	23	14.26	0.46	0.10	23	36.3	0.92	0.39	20	30.0	-0.30	60	0.65	75
11	large	-	cover	high	24	0.54	21	10.14	0.32		24	18.7	0.36		24	28.0	0.44			
12	small	+	cover	high	36	0.12	36	7.31	0.24	0.15	36	24.3	0.71	0.37	31	23.7	-0.03	71	0.61	89
13	small	+	open	low	36	0.11	33	5.42	0.18	0.10	36	16.3	0.45	0.27	30	12.0	-0.20	52	0.71	65
14	small	-	cover	high	36	0.11	28	4.46	0.15		35	12.1	0.32		35	8.3	-0.18			
15	large	-	cover	low	24	0.54	24	8.83	0.28		24	17.2	0.35		24	19.4	0.11			
16	small	+	open	high	36	0.13	30	8.00	0.26	0.14	31	29.6	0.90	0.41	30	27.9	-0.08	44	0.79	55

Tanks were stocked on 15 December 2001. Sandfish tanks were given dry seaweed powder once a day. Initially, tanks with large juveniles received 1.5 g day^{-1}, and tanks with small juveniles received 0.75 g day^{-1}. From 1 January 2002, this was increased to 2 g day^{-1} for all tanks. Tanks with shrimp were also fed commercial feed (Betagro 501S or Betagro 501) at 1.5 g day^{-1}. For about two weeks, this was divided into three feeds per day, and then reduced to two feeds per day. The rate was not increased throughout the trial to avoid overfeeding. Animals were monitored for three growth periods: they were weighed on 14 January, 7 February and 28 February.

For each factor, there were eight pairs of tanks that differed only with respect to that factor, and which received similar treatments with respect to the other three factors. While the absence of replication prevented statistical analysis (according to the best available advice), growth rates of sandfish in these pairs of tanks could be ranked for each growth period, and for the overall experiment. The early mortality in tank 4 led to later higher-than-expected growth among the remaining sandfish, so for the later and overall periods, this tank pair was left out of these comparisons. The fraction of tank pairs for which the growth rate advantage was in the direction indicated is shown in Table 7.

Conclusions from trial D

1. Some tanks with shrimp reached surprisingly high sandfish densities, up to 690 g m^{-2}.
2. The heavier juveniles at the beginning of the experiment generally remained heavier, in part because fewer were stocked (a trivial finding).
3. The apparent growth gain due to the presence of shrimp may well have been due (mainly or entirely) to the better feeding in these tanks, which received shrimp food as well as seaweed powder.
4. The four tanks stocked with small sandfish and shrimp had slightly lower sandfish survival rates. Mortality first occurred when sandfish averaged 0.12–0.26 g and shrimp 0.07–0.15 g. It is not clear whether predation by shrimp was to blame.
5. Sandfish growth was slightly better without covers than with covers.
6. High water exchange was beneficial to sandfish growth.
7. Mean shrimp survival was higher in covered tanks (83.1%) than open tanks (44.8%), and growth slightly lower.

Table 7. Summary of effects of factors on sandfish growth in trial D

factor	period 1	period 2	period 3	overall	average of ratios of mean final weights
large>small	8/8	6/7	3/7	6/7	1.53
shrimp>none	8/8	7/7	3/7	7/7	1.91
open>covered	6/8	6/7	3/7	4/7	1.15
high flow>low	6/8	7/7	5/7	6/7	1.87

Trial E: Two sizes (and none) of sandfish juveniles, shrimp

Sixteen 1.5-m diameter tanks were set up without covers. Mean water exchange was about 300–400 L tank^{-1} day^{-1}, one third of the tank volume. Tanks were stocked on 15 March 2002 using eight regimes (with two replicates): large, small or no sandfish juveniles with ongrown (large), postlarval (small) or no shrimp (omitting the treatment with neither sandfish, nor shrimp).

Sandfish were stocked to equal biomass/tank. Thus, 12 large, or about 168 small juveniles, were placed in groups of approximately 170 g total weight/group; care was taken that the weight range of individuals was also similar in different groups. Shrimp were stocked by number, with groups of 54 postlarvae (30 m^{-2}) or 27 ongrown animals (15 m^{-2}).

Tanks with shrimp were fed starter food (Betagro 501S). Other tanks received a mixture that was being used as the standard feed for sandfish during second nursery phase, consisting of two parts dry spirulina powder, one part Betagro 501S and one part dried ground seaweed. The seaweed was mainly sargassum — locally collected, sun dried, ground and seived through a 250-μm screen. In either case, two daily feeds of 1 g tank^{-1} were given.

Results of trial E

On 25 March, many dead or sick/dying sandfish were seen in tanks with large shrimp and small sandfish. They were left in the tanks and feeding continued. On 10 April, all tanks were emptied. Sandfish from the "large" group were weighed individually, shrimp and sandfish from the "small" group were counted, and the total weight measured.

Sandfish losses were negligible in most tanks and growth was good, averaging over 1 g day^{-1} for large juveniles and

0.1 g day^{-1} for small juveniles. Sandfish biomass densities reached around 300 g m^{-2}. Shrimp survival was also good, 70–80% for both ongrown animals and postlarvae. Shrimp growth was about 0.03 g day^{-1} for ongrown animals and 0.007 g day^{-1} for postlarvae.

Sandfish mortalities, however, were noticed from the tenth day in the two tanks stocked with small sandfish and large shrimp. Final sandfish survival in these two tanks was only 9%, and there was an overall slight drop in average weight. If shrimp growth was roughly linear, at the time these mortalities started, the average weight of shrimp in the two tanks would have been about 0.95 g, similar to the starting weight of the sandfish.

Conclusions from trial E

1. Shrimp of about 1 g caused the death of sandfish of a similar size.
2. Shrimp feed could support good sandfish growth in the absence of shrimp.
3. The survival of small shrimp was highest with large sandfish, otherwise the presence or absence of sandfish had little effect on shrimp growth and survival.

Trial F: Four sizes of sandfish, three sizes of shrimp and no shrimp

Sixteen 1.5 m diameter tanks were set up (unshaded) as before, but with only six litres of sand

Table 8. **Results grouped by treatment in trial E**

treatment			15 March				10 April						
tank	sandfish	shrimp	sandfish		shrimp		sandfish				shrimp		
			av. wt. (g)	no.	av. wt. (g)	no.	av. wt. (g)	no.	growth (g day^{-1})	survival (%)	av. wt. (g)	no.	survival (%)
12	large	large	14.40	12	0.580	27	44.5	12	1.16	100.0	1.25	20	74.1
13	large	large	14.30	12	0.620	27	37.2	12	0.88	100.0	1.44	18	66.7
2	large	small	14.30	12	0.008	54	42.0	12	1.06	100.0	0.23	47	87.0
14	large	small	14.50	12	0.008	54	37.7	12	0.89	100.0	0.18	52	96.3
3	large	none	14.20	12			47.6	12	1.29	100.0			
16	large	none	14.30	12			43.2	12	1.11	100.0			
7	small	large	0.98	168	0.560	27	0.24	13	-0.03	7.7	1.44	21	77.8
15	small	large	0.98	167	0.600	27	0.33	17	-0.02	10.2	1.66	16	59.3
4	small	small	0.99	168	0.008	54	3.36	169	0.09	100.6	0.20	45	83.3
10	small	small	0.99	163	0.008	54	3.40	163	0.09	100.0	0.19	47	87.0
6	small	none	0.98	166			4.69	166	0.14	100.0			
9	small	none	0.96	170			3.12	169	0.08	99.4			
5	none	large			0.610	27					1.31	16	59.3
11	none	large			0.610	27					1.23	26	96.3
1	none	small			0.008	54					0.25	34	63.0
8	none	small			0.008	54					0.19	42	77.8

Table 9. **Averaged results of treatment pairs in trial E**

tanks	treatment		sandfish			shrimp		
	sandfish	shrimp	survival (%)	final density (g m^{-2})	growth rate (g day^{-1})	survival (%)	final density (g m^{-2})	growth rate (g day^{-1})
12, 13	large	large	100.0	277.3	1.02	70.4	14.4	0.028
2, 14	large	small	100.0	270.7	0.98	91.7	5.7	0.007
3, 16	large	none	100.0	308.4	1.20			
7, 15	small	large	9.0	2.5	-0.03	68.5	16.0	0.037
4, 10	small	small	100.0	317.5	0.09	85.2	5.1	0.007
6, 9	small	none	99.7	369.6	0.11			
5, 11	none	large				77.8	15.0	0.025
1, 8	none	small				70.4	4.7	0.008

(3.5 mm depth). Filling started on 26 July 2002, and tanks were stocked on 29 July with four size groups of sandfish bred at RIA3. Some were nursed for two months in seabed babylon cages, with and without snails, while the smaller sizes came directly from the RIA3 onshore nursery tanks.

For the two larger sizes of sandfish, tanks were stocked with 12 juveniles each; for the two smaller sizes there were 18 sandfish per tank. Treatments with shrimp (hatchery-bred and pond-grown) had 18 sandfish per tank, about 10 m^{-2}. Care was taken to equalise both the average weights and size distribution of the sandfish groups, and to minimise the spread. With the shrimp, average weights were matched. The 16 treatments were assigned randomly (by drawing paper slips from a beaker). All tanks were fed 1 g shrimp starter food (Betagro 501S) twice daily. In the first couple of days the large shrimp that died were replaced (generally by smaller animals, since they were all that were available).

On the second weighing (12 August) all sandfish were collected but only total weights were measured. Survival then was 100%, but two days later, some sandfish in tank 6 were sick. Those that died were not removed. Ten shrimp from each tank were also caught and weighed. On the third weighing (26 August) all surviving animals were collected. Sandfish were weighed individually, shrimp counted and weighed as groups.

In tanks with small, medium or no shrimp, sandfish survival averaged 99%, but with large shrimp only 69%. (It is unfortunate that the experiment was not continued for one more period to see if these losses continued.) Tanks with small shrimp had the best sandfish growth. Survival rates of shrimp were reasonable, at 79.6%. Small shrimp almost doubled in size over the course of the experiment, while large shrimp hardly grew at all. (Perhaps they were underfed, or unable to use the small-size feed efficiently.)

Conclusions from trial F

1. The size of sandfish had no apparent effect on growth or survival of the shrimp.
2. Large shrimp (about 5 g average weight) may have reduced the survival of sandfish in the second period in the small, medium and large sandfish groups (7.6–28.1 g average weights).
3. The presence of small shrimp (up to about 1.5 g average) may have had a slight positive effect on the growth of sandfish.

Table 10. **Results arranged by treatments in trial F**

treatment			29 July			12 August				26 August							
tank	sandfish	shrimp	sandfish		shr.	sandfish			shr.	sandfish					shrimp		
			no.	av. wt (g)	av. wt (g)	no.	av. wt (g)	period one, growth (g day⁻¹)	av. wt (g)	no.	av. wt (g)	period two, growth (g day⁻¹)	overall growth (g day⁻¹)	surv. (%)	no.	av. wt (g)	surv. (%)
12	XL	none	12	30.8		12	46.3	1.11		12	41.3	-0.36	0.38	100			
3	XL	small	12	30.9	0.8	12	42.3	0.82	1.1	12	47.9	0.40	0.61	100	15	1.5	83
13	XL	med.	12	30.9	2.1	12	37.2	0.45	3	12	38.4	0.09	0.27	100	14	2.7	78
9	XL	large	12	29.8	5.0	12	36.7	0.49	5.7	11	42.5	0.41	0.45	92	12	5.1	67
6	large	none	12	10.3		12	23.8	0.97		11	24.4	0.04	0.50	92			
10	large	small	12	10.2	0.9	12	25.4	1.09	1.1	12	34.3	0.63	0.86	100	14	1.8	78
5	large	med.	12	9.4	2.1	12	24.3	1.06	2.6	12	27.7	0.24	0.65	100	14	3.1	78
4	large	large	12	9.5	4.5	12	23.4	1.00	4.6	8	28.1	0.34	0.67	67	16	4.8	89
16	med.	none	20	3.2		20	15.0	0.84		20	20.4	0.38	0.61	100			
7	med.	small	20	3.6	0.9	20	12.1	0.61	1.1	20	19.0	0.49	0.55	100	13	1.5	72
14	med.	med.	20	3.3	2.3	20	9.5	0.44	2.3	20	13.3	0.28	0.36	100	14	2.6	78
1	med.	large	20	3.1	4.7	20	13.4	0.73	5.4	15	17.1	0.27	0.50	75	16	5.8	89
2	small	none	20	1.1		20	7.9	0.49		20	7.3	-0.04	0.22	100			
8	small	small	20	1.0	0.9	20	7.6	0.47	1.1	20	18.1	0.75	0.61	100	13	1.5	72
11	small	med.	20	1.1	2.2	20	8.5	0.53	2.8	19	15.4	0.49	0.51	95	14	3.0	78
15	small	large	20	1.0	4.9	20	7.6	0.47	4.6	10	9.2	0.12	0.29	50	17	4.8	94

Trial G: Two sizes of sandfish and two sizes of shrimp in tanks and aquaria

Tanks

Fifteen small fibreglass tanks (85 cm internal floor diameter (0.57 m^2) and 46 cm water depth (280 L)) were arranged outdoors in three rows with partial shading (60%). Unfiltered water was supplied from a settlement tank (of varying head and with varying demand on the line) using uniform nozzles, for about 10 hours per day. This gave a mean water retention time of about 24 hours. Tank floors were covered with a thin layer of sieved beach sand. All tanks were covered with a further lightweight net (about 4.5 mm mesh) to prevent shrimp from jumping out (and tanks with shrimp had outlet screens). Tanks received a 0.5 g ration of fine shrimp starter (CP 9000) once a day, starting on the first day. Feedings were kept quite low and infrequent to test the behaviour of shrimp towards sandfish when hungry.

On 6 June 2003, tanks were stocked using five stocking combinations. Small shrimp had been grown from postlarvae in a tank, large shrimp had been grown in a farmer's pond. There were three replicates of each tank treatment (randomly assigned within rows so that each row had one of each treatment). The treatments were:

1. 12 medium sandfish (av. wt. 3.6 g, density 21 m^{-2}) with 18 medium shrimp (av. wt. 1.4 g, density 32 m^{-2}).
2. 12 medium sandfish (av. wt. 3.6 g, density 21 m^{-2}) with no shrimp.
3. 18 small sandfish (av. wt 0.5 g, density 32 m^{-2}) with 18 medium shrimp (av. wt. 1.4 g, density 32 m^{-2}).
4. 18 small sandfish (av. wt 0.5 g, density 32 m^{-2}) with 24 small shrimp (0.02 g average weight, density 42 m^{-2}).
5. 18 small sandfish (av. wt. 0.5 g, density 32 m^{-2}) with no shrimp.

Aquaria

Six small glass aquaria (30 cm x 20 cm x 30 cm deep) were set up using similar sizes of sandfish and shrimp. Two treatments without shrimp were omitted. The floor area of an aquarium was about one tenth that of a fibreglass tank, and the stocking density per unit area about 3.3 times higher — volume stocking densities in aquaria were about 4.4 times more than in tanks. The aquaria were given an approximate daily ration of 20 mg each of fine shrimp starter, beginning on the second day. Water changes were carried out only occasionally.

Results of trial G

A couple of days after stocking, a small sandfish was found bitten in half in aquarium 6 (which contained medium shrimp). On 14 June, more small sandfish appeared to have been attacked by medium shrimp: some had eviscerated, and some were dead (and partly eaten).

Table 11. **Tank results grouped by treatments in trial G**

			06 June				16 June						
tank	combination		sandfish		shrimp		sandfish				shrimp		
	sandfish	shrimp	no.	av. wt (g)	no.	av. wt (g)	no.	av. wt (g)	growth (g/day)	survival (%)	no.	av. wt (g)	growth (g/day)
2	medium	medium	12	3.7	18	1.40	3	5.7	0.20	25	18	1.390	-0.010
8	medium	medium	12	3.6	18	1.40	0			0	17	1.820	0.040
14	medium	medium	12	3.5	18	1.40	1	3.0	-0.05	8	18	1.560	0.010
3	medium	none	12	3.5			12	7.9	0.44	100			
6	medium	none	12	3.6			12	8.6	0.50	100			
12	medium	none	12	3.7			12	9.6	0.59	100			
4	small	medium	18	0.5	18	1.40	0			0	18	1.610	0.020
7	small	medium	18	0.5	18	1.40	0			0	18	1.500	0.010
11	small	medium	18	0.5	18	1.40	0			0	18	1.560	0.010
5	small	small	18	0.5	24	0.02	18	2.7	0.21	100		0.064	0.004
10	smalll	small	18	0.5	24	0.02	17	1.6	0.11	94			
13	smalll	small	18	0.5	24	0.02	18	2.5	0.20	100			
1	small	none	18	0.5			18	2.8	0.23	100			
9	small	none	18	0.5			18	2.3	0.18	100			
15	small	none	18	0.5			18	2.6	0.21	100			

On 16 June, sandfish of both sizes, as well as medium shrimp, were collected from tanks, counted and weighed. In tanks with medium shrimp most sandfish were either completely flat and flaccid, dead, or had disappeared. Some remains of viscera were found. In the six tanks without shrimp (or with small shrimp), nearly all sandfish had survived and grown. Sandfish growth was low in tank 10 (small sandfish, small shrimp), but otherwise sandfish appeared to be unaffected by the presence of small shrimp. Unfortunately, the small shrimp were not counted, and only a sample from tank 5 were weighed, so their survival and most growth data was lost. Nearly all medium shrimp in tanks survived, though growth was poor.

All sandfish with medium shrimp died except for those in aquarium 4, which lost weight. Sandfish with small shrimp all survived. Six shrimp also died under these rather severe conditions of high density and temperatures, with little water exchange.

Conclusions from trial G

1. Shrimp of about 1.5 g average weight caused the deaths of juvenile sandfish, of average weight 2.8–3.7 g.
2. Shrimp of 0.02 g average weight did not harm sandfish of 0.5 g.
3. Small shrimp did not appear to affect sandfish growth.

Discussion

Taken together, the results of these seven trials indicate that co-culturing *Holothuria scabra* and *Penaeus monodon* should be possible in many situations, and that any adverse interactions are likely to be at the expense of sandfish, not shrimp. In addition, scientists at RIA3 have reared sandfish and shrimp together (to market size) in ponds on at least two occasions (Thu 2003). In these larger-scale trials, there was reasonable survival to harvest size of both shrimp and sandfish in two out of four ponds using wild-collected sandfish, and in one of two ponds with our hatchery-produced juveniles.

Despite the generally promising results, predation of sandfish by *P. monodon* was a real and rapid phenomenon under certain conditions. Thu and coworkers (pers. comm.) have run tank trials that appear to indicate that shrimp may attack sandfish when stocked at high density, but not at low density. Predation of sandfish juveniles by young swimming crabs and by rabbitfish has also been observed (Pitt and Duy 2004).

It may be that sandfish are not a natural or preferred food source for shrimp, but rather a feed they can learn to use when hungry. Shrimp may be able to induce evisceration of sandfish, and thus feed on the viscera. This is supported by our observations of flat, empty-looking sandfish juveniles with unbroken skin in some co-culture tanks.

A summary of cases where predation of sandfish during co-culture did or did not occur is shown in Figure 2. It is clear that mortality occurred in a minority of cases and that it was not simply restricted to combinations of relatively small sandfish and relatively small shrimp. Rather, it appears that certain rearing conditions may promote aggression of shrimp towards sandfish.

We caution that the results presented here need to be interpreted with care. While the large number of trials provides a measure of confidence that the results represent the range of interactions that can be expected between *H. scabra* and *P. monodon*, most of the experiments had little or no replication. Thus, there is no certainty that the results of individual experiments were not due to chance alone. We strongly encourage further, more rigorous, research on this subject using sufficient replicates to investigate the effects of different sizes of sandfish

Table 12. **Aquarium results grouped by treatments in trial G**

			06 June				16 June						
aquarium	combination		sandfish		shrimp		sandfish				shrimp		
	sandfish	shrimp	no.	av. wt (g)	no.	av. wt (g)	no.	av. wt (g)	growth (g/day)	survival	no.	av. wt (g)	growth (g/day)
1	medium	medium	4	3.0	6	1.40	0			0	5	1.6	0.016
6	medium	medium	4	2.8	6	1.40	0			0	3	2.0	0.056
4	small	medium	6	0.5	6	1.40	6	0.3	-0.017	100	5	2.6	0.116
5	small	medium	6	0.5	6	1.40	0			0	5	1.8	0.036
2	small	small	6	0.5	8	0.02	6	1.2	0.067	100	8	0.063	0.004
3	small	small	6	0.5	8	0.02	6	0.8	0.033	100	8	0.063	0.004

and shrimp at different densities, under different feeding regimes, etc. The results of such experiments may then form the basis of a guide to growers regarding safe size combinations for co-culturing the two species.

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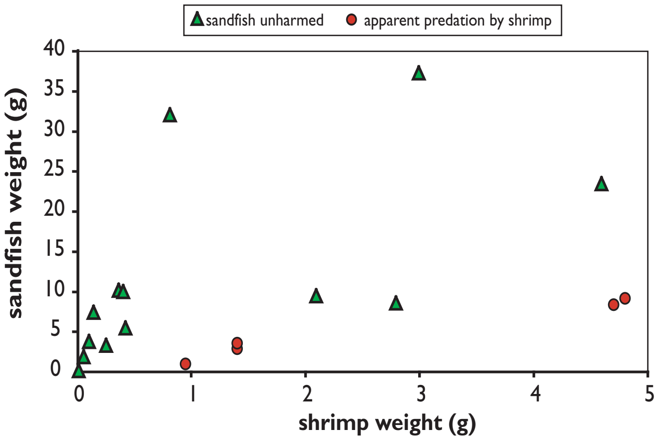

Figure 2. **Mean sizes in tanks where sandfish were unharmed, and where they appear to have been killed by shrimp.**

Monitoring a fissiparous population of *Holothuria atra* on a fringing reef on Reunion Island (Indian Ocean)

Chantal Conand[1]

Introduction

Holothuria atra is the most common and abundant sea cucumber species on the fringing reefs of Reunion Island, which is generally the case with other Indo-Pacific reefs (Conand 1996; Conand and Mangion 2002; Jaquemet et al. 1999; Uthicke 2001). In Reunion, its density varies depending on the site and reef zone studied and its populations show a variety of structures (Conand 1996). In addition, this species' role in the ecology of soft bottoms of Reunion Island reefs is now understood. It plays an important role in remodelling sediments as the studied population ingests some 78 kg m^{-2} annually (Mangion et al. in press).

This is one of the sea cucumbers species that can reproduce asexually by fission, with a range of modes depending on the study site (Chao et al. 1994; Conand 1996; Jaquemet et al. 1999; Uthicke 2001). The scope of asexual reproduction is a key to understanding population genetics. In fact, genetic studies on this species have shown that in spite of the significance of asexual reproduction, sexual reproduction is vital for large-scale dispersion of larvae (Uthicke et al. 2001). However, asexual reproduction is a very widespread mechanism in this species and its influence on population abundance and specimen size has been studied at several sites in the Great Barrier Reef in Australia (Uthicke 1997, 2001), Taiwan (Chao et al. 1994), New Caledonia (Conand 1989) and Reunion (Conand 1996; Jaquemet et al. 1999).

The results presented here deal with the continuation in 1998, 1999 and 2000 of the sampling conducted between November 1993 and November 1997; sampling that had demonstrated a relative stability in densities and mean specimen weights (Jaquemet et al. 1999). This study involved monitoring the influence that asexual reproduction by fission has on population dynamics, in particular on density and mean specimen sizes (weights).

Materials and methods

Sampling was carried out at the back-reef station at Planch'Alizés (Saline Reef), once a year, during the hot season. The methods were the same as those used previously (Conand 1996; Jaquemet et al.

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