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Laure Carassou, Camille Mellin, Dominique Ponton. Assessing the diversity and abundances of larvae and juveniles of coral reef fish: a synthesis of six sampling techniques. *Biodiversity and Conservation*, 2008, 18 (2), pp.355-371. 10.1007/s10531-008-9492-3 . hal-04410595

**HAL Id: hal-04410595**

**<https://hal.inrae.fr/hal-04410595>**

Submitted on 22 Jan 2024

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# Assessing the diversity and abundances of larvae and juveniles of coral reef fish: a synthesis of six sampling techniques

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Received: 10 July 2007 / Accepted: 18 September 2008 / Published online: 14 October 2008  
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**Abstract** Due to an increasing emphasis for fish population survey and regulation, efficient tools for evaluating the abundance and diversity of fish from various life stages are needed, especially for coral reef species that present a high taxonomic diversity. The characteristics of six different techniques used for sampling pelagic larvae (a plankton-net and two light-traps), newly settled juveniles (one type of artificial reef), and older juveniles (an underwater seine net in seagrass and macroalgal beds, and rotenone poisoning in coral patches) are described in this study. Larvae belonging to 70 families and juveniles belonging to 34 families were collected. An analysis of similarity (ANOSIM) showed that the taxonomic composition of assemblages collected with the plankton-net and the two light-traps were overlapping but clearly different, due to the higher occurrence of Gobiidae in the plankton-net and of Pomacentridae in both light-traps. Larvae being 2–4 mm standard length (SL) dominated in the plankton-net, whereas larvae being 9–11 mm SL dominated in both light-traps. Pomacentridae juveniles were more abundant in rotenone samples, whereas Labridae dominated in the underwater seine. Juvenile fish collected with the artificial reefs, the underwater seine, and rotenone poisoning largely overlapped in size, with mean sizes of 22, 38, and 33 mm SL, respectively. Seven families were caught by the six sampling techniques, but with unequal success. This study provides ecologists and managers with a unique review of six techniques for sampling a wide range of developmental stages of young fish in different habitats of a coral reef lagoon.

**Keywords** Artificial reefs · Coral reef fish · Juveniles · Larvae · Light-traps · Plankton-net · Rotenone poisoning · Sampling · Seine net

## Introduction

The abundance and diversity of adult coral reef fishes mainly depend on the recruitment success (Sale 2002), which is characterized by important seasonal and inter-annual variations

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(Doherty and Williams 1998). Understanding these variations in specific locations is a crucial challenge in reef fish conservation strategies. Recruitment success depends, for a large part, on the number of survivors among pelagic larvae in the water column (Boehlert 1996) and among juveniles in benthic habitats (Doherty et al. 2004). As a consequence, understanding the processes that influence the size and the dynamics of coral reef fish populations requires focusing on the diversity and abundance of their early life stages (Booth and Brosnan 1995). More specifically, studying the processes driving larval and juvenile fish survival under particular environmental conditions may help us to understand the factors influencing the replenishment of adult fish populations in a given location (Doherty 2002; Fuiman and Werner 2002). Such studies are also useful in designing marine protected areas with various levels of connectivity (Cowen et al. 2007) or in evaluating the effect of long-term fishing on a particular species (Hsieh et al. 2006).

Studying the early life stages of fishes is challenging because their size, morphology, and behavior do not only vary greatly between species, but also during ontogeny within species (Moser 1981; Leis 1991). Moreover, the mortality rate also varies during ontogeny and, thus, with size for a given species, which further complexifies the evaluation of young fish abundances over time. Obtaining accurate estimates of the diversity and abundances of fish larvae and juveniles is, thus, problematical (Suthers and Frank 1989), especially in tropical waters where taxonomic diversity is high (Randall 2005). To study larvae and juveniles, different sampling apparatus have to be combined, with each method targeting a specific developmental stage, or size range, to be found in a given habitat (Leis 1991). Small pelagic larvae are usually sampled by plankton-nets (Barkley 1972; Murphy and Clutter 1972), which enable a wide range of taxa in many types of habitats to be caught (Choat et al. 1993). Light-traps are efficient in catching pre-settlement larvae (Doherty 1987), although they collect a narrow range of taxa and can only be used in some habitats (Thorrold 1992). Artificial reefs, moored in different habitats, are useful for sampling larvae at settlement (Leis et al. 2002; Valles et al. 2006). Finally, underwater seine and rotenone poisoning, used in specific habitats, can be used to capture post-settlement juvenile stages (Ackerman and Bellwood 2000; Smith and Sinerchia 2004).

To date, no study has ever synthesized information about the different techniques used to capture the full range of young stages of fishes in coral reef ecosystems. Indeed, most of the published studies generally rely on a single sampling technique, and when comparisons between different techniques have been made, they focus on the apparatus targeting fish larvae only (Choat et al. 1993; Hickford and Schiel 1999; Anderson et al. 2002). This lack of broad comparisons is understandable, as it is difficult to obtain a picture of both larvae and juvenile fish abundances at the same place and time, and to compare techniques used in different habitats and targeting stages that differ in their behavior (Leis et al. 2002). However, a synthesis of the characteristics of a few common sampling techniques is strongly needed. Such a review should help managers and scientists to choose the most suitable technique to catch particular ontogenetic stages or taxa of reef fishes, or young fish that live in particular habitats inside a coral reef lagoon.

The opportunity to provide such a synthesis arose in the context of a research program investigating the relationships between larval and juvenile fish assemblages and the characteristics of their environment in the lagoon of New Caledonia, southwest Pacific. This research program provided data about the taxonomic and size ranges of larval and juvenile fish assemblages caught with six different techniques. These sampling techniques were used in habitats ranging from the water column in coastal embayments, around lagoonal islets and passes, to benthic habitats, such as seagrasses or macroalgal beds and small coral patches. The sampling designs varied according to the specific aims of each study and the

ontogenetic stage under study. In spite of the differences in sampling designs and efforts, the data collected offered a unique opportunity to obtain an overview of the efficiency and limits of each sampling technique.

The objectives of the present study are, thus: (1) to describe the six different techniques used for sampling the larvae and juveniles of coral reef fish species in the lagoon of New Caledonia; (2) to statistically compare the taxonomic diversity and the size spectra of the larvae or juveniles caught by each method; (3) to examine the relationship between the diversity of larval or juvenile fish collected and the number of samples performed for each technique; (4) to discuss the efficiency of each technique for capturing particular ontogenetic stages or families of reef fishes, or fish that live in particular habitats.

## Materials and methods

### Definitions

The end of the larval stage of reef fishes is usually defined as the moment when they settle into benthic habitats (Leis and McCormick 2002). In this study, the term “larvae” will refer to all of the young fishes caught in the water column, whereas the term “juvenile” will refer to all of the settled individuals caught on benthic habitats, whatever their actual developmental stage.

### Sampling methods

Fish larvae were sampled in the water column using three different methods, including a plankton-net and two types of light-traps. The plankton-net was 3.60 m long with a 335- $\mu\text{m}$  mesh size and a circular opening of 0.60 m. It was towed horizontally by night  $\sim 0.5$  m below the water surface by a boat that followed a circular trajectory for 2 min at  $\sim 2$  knots. The plankton-net was always towed at the beginning of the night.

The first type of light-trap used in this study was initially developed by the Aquafish Technologies Company, Lattes, France, and will be named “Aquafish LT.” Each Aquafish LT consisted of a water-tight block containing a 12-V battery and a 7-W neon lamp, which was set on top of seven Plexiglas units separated by  $\sim 10$ -mm vertical slits. The base of the light-trap consisted of a removable collector equipped with 2-mm mesh size gauze windows. When the light was on, fish larvae were attracted and entered the trap through the vertical slits; they were then theoretically unable to escape. These light-traps were always set 2.5 m deep. The capacity of the 12-V battery allowed a timer to automatically switch on the lamp 4 h per night from 01:00 to 05:00 am. All Aquafish LT were retrieved early in the morning.

The second type of light-trap was developed by the Ecocean Society, Saint-Clément-de-Rivière, France (INPI deposited patent no. 0208582), and will be named “Ecocean LT.” Each Ecocean LT consisted of a buoyant water-tight block containing a 6-V battery and a 7-W neon lamp, under which a 2-mm mesh size conical net was attached vertically. Theoretically, this design aims at reducing the high abundances of Clupeiform fish, since most engraulids and clupeids tend to remain under the lamp in the open water, whereas coral reef species tend to stay inside the conical net in order to hide<sup>1</sup> (Lecaillon and Lourié 2007).

<sup>1</sup> For details on the procedure, see <http://www.ecocean.fr>.

When the net was pulled out of the water, fishes were retrieved in the removable collector equipped with 330- $\mu\text{m}$  mesh size gauze. The Ecocean LT were set at the surface. The capacity of the 6-V battery allowed a timer to automatically switch on the lamp 7 h per night from 22:00 pm to 05:00 am. All Ecocean LT were retrieved early in the morning.

Juvenile coral reef fishes were sampled using three different techniques: (1) artificial reefs for juveniles newly settled in seagrass or macroalgal beds, or near coral patches; (2) an underwater seine pulled by scuba-divers for older juveniles inhabiting seagrass or macroalgal beds; (3) rotenone micro-poisoning for those inhabiting small coral patches. The artificial reefs consisted of a three-dimensional structure made of a 1-cm mesh size plastic net enclosing a spiral made of a 5-mm mesh size plastic net which enhanced refuge complexity, and three polystyrene buoys which maintained the structure vertically in the water column. Each artificial reef was attached to a 7-kg concrete weight by a 0.8-m long steel cable and moored between 2 and 8 m deep. Juveniles were retrieved 2 days after all reefs had been initially emptied and cleaned. Two scuba-divers wrapped each artificial reef with a 1-mm mesh size bag and then brought it to the surface. Each reef was thoroughly cleaned and all fish were carefully removed from the bag.

The underwater seine was a 10-m long, 1.20-m high, 4-mm mesh size beach seine net that had been equipped with a 50-cm wide, 1-m long, and 2-mm mesh size circular collector. The seine was modified to keep it vertical above the sea bottom by adding small polystyrene buoys onto the headrope and attaching a weighted rope to the footrope. After deployment, the seine was dragged by two scuba-divers over a distance of  $\sim 10$  m. The collector was then tightly closed and brought to the surface, where all the juveniles were carefully removed from the collector.

Rotenone poisoning was performed on small coral colonies  $<1 \text{ m}^3$  in size. The selected coral colony was first entirely covered with a 1-mm mesh size square net, whose perimeter was weighted by a chain to hold it on the substratum and whose apex was held above the colony by small buoys. Then  $\sim 200$  g of rotenone powder, mixed with 1 l of seawater and 20 ml of detergent, was injected into the enclosed area. After  $\sim 10$  min, i.e., the time required for rotenone to asphyxiate the fish, two scuba-divers collected all of the individuals under the net, sometimes using forceps to remove those lodged in coral holes.

Except for the fish larvae caught with the plankton-net and the juveniles sampled with rotenone, all individuals were anaesthetized in a benzocaine solution upon retrieval and immediately preserved in 95% methylated alcohol.

### Sampling periods

Although each sampling technique was used in the context of different studies, all of the samples were collected during the warm season, from September to February, a period when old fish larvae are abundant in the SW lagoon of New Caledonia (Carassou and Ponton 2007). The plankton-net and the two light-traps were used four nights per month from September 2004 to January 2005, and the two light-traps only were used six nights per month from September 2005 to February 2006. In order to optimize the efficiency of the light-traps, sampling was always performed as close as possible to the new moon period. The artificial reefs were retrieved once per month from September to December 2005. The underwater seine sampling and rotenone poisoning were performed once per month from September 2005 to February 2006. These sampling schemes resulted in a total of 60 samples with the plankton-net, 44 samples with Aquafish LT, and 22 with Ecocean LT in 2004–2005, and 135 samples with Aquafish LT, 136 with Ecocean LT, 192 with artificial reefs, 48 with the underwater seine, and 24 from rotenone poisoning in 2005–2006.

## Fish identification and measurement

Fish larvae were identified to the lowest possible taxonomic level using Leis and Trnski (1989), Leis and Carson-Ewart (2000a), and the Fishpaste electronic database developed by P.J. Doherty (AIMS, Townsville, unpublished database). Juveniles were identified using information found in Randall (2005) for most species, Wilson (1998) for Lethrinids, and Bellwood and Choat (1989) for Scarids. As most fish larvae could not be identified to the species or even the genus level, all fish will be grouped to the family level in the analyses presented here.

All of the individuals were measured (standard length in mm) using an electronic calliper, except for the larvae of Clupeiformes, for which only up to 20 randomly chosen individuals were measured per sample when abundant, and the juveniles of Siganidae and Apogonidae, for which a minimum of 15 randomly chosen individuals were measured per sample when abundant.

## Data analyses

Non-parametric one-way analyses of similarity (ANOSIM) were used to compare the taxonomic diversity and size distribution of the assemblages collected with the three techniques used in the water column on the one hand, and with the three techniques used in benthic habitats on the other hand. In each case, the analyses were based on a Bray–Curtis similarity matrix built on the presence–absence of each family or each size class in the samples. ANOSIM generated a value of  $R$  statistic measuring the degree of difference between groups, i.e., assemblages obtained from different techniques, on a scale of 0 (indistinguishable) to 1 (all similarities within groups are inferior to any similarity between groups). Levels of significance  $P$  of the differences between assemblages were obtained by a permutation procedure on the similarity matrices (Clarke and Gorley 2001). When significant differences were found to occur ( $P < 0.05$ ), analyses of contribution to the dissimilarity (SIMPER) were performed in order to identify the family and/or the size class accounting for the difference (Clarke 1993). The data collected from November 2004 to January 2005 were used to compare the plankton-net and the two light-traps, while the data collected from September to December 2005 were used to compare the artificial reefs, the underwater seine, and the rotenone poisoning technique.

Finally, the total number of different families, which increased as samples were successively plotted, was established every year for every sampling technique. The samples were entered in a random order 999 times. The resulting curves were then averaged to obtain a smooth curve—these calculations being performed with the Primer v5 software (Clarke and Gorley 2001). All of the results were finally synthesized to obtain a qualitative overview of the best sampling techniques for targeting specific ontogenetic stages of different families in specific habitats.

## Results

### Taxonomic composition of catches

Among the 397 samples collected by plankton-net and light-traps and the 264 samples collected with artificial reefs, underwater seine, and rotenone poisoning, a total of 35,072 larvae and 2,370 juveniles belonging to 74 families were collected (Table 1). Among these

**Table 1** Families of fish larvae and juveniles collected with the six sampling techniques

Order and families	Plankton-net	Aquafish light-trap		Ecocean light-trap		Artificial reef	UW seine	Rotenone poisoning
		Y1	Y2	Y1	Y2			
Anguilliformes								
Anguillidae	–	–	–	–	p	–	–	–
Congridae	–	–	p	p	p	–	–	–
Muraenidae <sup>a</sup>	–	–	–	p	p	–	–	p
Nettastomatidae	–	–	–	–	p	–	–	–
Ophichthidae	–	–	–	p	p	–	–	–
Atheriniformes								
Atherinidae	p	p	A	p	p	–	–	–
Notocheiridae	p	–	p	–	p	–	–	–
Aulopiformes								
Synodontidae <sup>a</sup>	–	–	–	–	p	–	p	p
Beloniformes								
Belonidae	p	–	–	–	–	–	–	–
Hemiramphidae	p	–	p	–	p	–	–	–
Beryciformes								
Holocentridae	–	–	–	–	–	–	–	p
Clupeiformes								
Clupeidae	p	A	A	p	A	–	–	–
Engraulididae	D	D	D	A	D	–	–	–
Elopiformes								
Elopidae	–	–	–	–	p	–	–	–
Gadiformes								
Bregmacerotidae	p	–	–	–	p	–	–	–
Gasterosteiformes								
Aulostomidae	–	–	–	–	p	–	–	–
Fistulariidae <sup>a</sup>	–	p	–	p	p	–	p	–
Pegasidae	p	–	–	–	–	–	–	–
Syngnathidae <sup>a</sup>	A	p	p	–	p	–	p	p
Lophiiformes								
Antennariidae <sup>a</sup>	–	–	p	p	p	p	–	p
Mugiliformes								
Mugilidae	p	–	–	–	–	–	–	–
Ophidiiformes								
Bythitidae	p	–	–	–	–	–	–	–
Ophidiidae <sup>a</sup>	–	–	–	–	p	–	–	p
Perciformes								
Acanthuridae <sup>a</sup>	–	–	p	–	p	–	–	p
Ambassidae	p	–	p	–	p	–	–	–
Ammodytidae	p	–	p	–	p	–	–	–
Apogonidae <sup>b</sup>	p	A	A	A	A	D	p	D
Blenniidae <sup>b</sup>	p	p	p	p	p	D	p	p
Caesionidae	–	–	p	–	p	–	–	–
Callionymidae <sup>a</sup>	p	–	–	–	–	–	p	p
Carangidae	p	p	p	p	p	–	–	–
Perciformes								
Chaetodontidae <sup>a</sup>	–	p	p	p	p	p	p	p
Eleotridae <sup>a</sup>	p	–	–	–	–	p	–	–
Gerreidae	p	–	p	–	p	–	–	–
Gobiesocidae <sup>a</sup>	p	–	p	–	p	p	–	p
Gobiidae <sup>b</sup>	D	–	p	p	p	p	A	A
Haemulidae	p	–	p	p	p	–	–	–

**Table 1** continued

Order and families	Plankton-net	Aquafish light-trap		Ecocean light-trap		Artificial reef	UW seine	Rotenone poisoning
		Y1	Y2	Y1	Y2			
Kuhliidae	–	–	–	–	p	–	–	–
Kyphosidae	–	–	–	–	p	–	–	–
Labridae <sup>a</sup>	p	–	–	p	p	A	D	A
Leiognathidae	A	–	p	–	p	–	–	–
Lethrinidae <sup>a</sup>	–	p	p	D	A	p	D	–
Lutjanidae	p	p	p	–	p	–	–	–
Monodactylidae	p	–	–	–	–	–	–	–
Mullidae <sup>a</sup>	p	–	p	–	p	–	p	p
Nemipteridae <sup>a</sup>	p	p	–	p	p	–	–	p
Pinguipedidae	–	–	–	–	–	–	p	p
Plesiopidae <sup>a</sup>	–	p	p	–	p	–	–	p
Pomacanthidae <sup>a</sup>	p	–	–	–	–	–	–	p
Pomacentridae <sup>b</sup>	p	D	D	A	D	p	p	D
Priacanthidae	p	–	–	–	–	–	–	–
Pseudochromidae <sup>a</sup>	–	–	p	p	p	–	–	p
Ptereleotridae	–	–	–	–	–	–	p	–
Scaridae <sup>b</sup>	p	–	p	–	p	p	A	p
Schindleriidae	–	–	p	–	–	–	–	–
Scombridae	p	p	p	–	–	–	–	–
Serranidae <sup>a</sup>	–	p	p	–	p	–	–	p
Siganidae <sup>a</sup>	p	A	p	D	p	p	A	–
Sillaginidae	p	–	–	–	–	–	–	–
Sphyraenidae	p	p	p	–	p	–	–	–
Toxotidae	p	–	–	–	–	–	–	–
Trichiuridae	–	–	p	–	–	–	–	–
Tripterygiidae <sup>b</sup>	p	p	p	p	p	A	p	p
Xenisthmidae	p	–	–	–	–	–	–	–
Pleuronectiformes								
Bothidae	p	–	p	–	p	–	–	–
Cynoglossidae	–	–	–	–	p	–	–	–
Poecilopsettidae	–	–	–	–	p	–	–	–
Soleidae	p	–	–	P	–	–	–	–
Scorpaeniformes								
Platycephalidae <sup>a</sup>	p	–	–	–	–	–	p	–
Scorpaenidae <sup>a</sup>	–	p	p	p	p	–	–	A
Triglidae	p	–	–	–	–	–	–	–
Siluriformes								
Plotosidae	–	–	–	–	–	–	p	–
Tetraodontiformes								
Monacanthidae <sup>a</sup>	p	–	p	p	p	A	p	–
Tetraodontidae <sup>b</sup>	p	p	p	p	p	p	p	p

p = present; A = abundant, i.e., among the five most abundant families collected with the sampling technique; D = dominant, i.e., among the two dominant families collected with the sampling technique

– = absent from the samples collected with the sampling technique

<sup>a</sup> Individuals of this family were caught as larvae and juveniles

<sup>b</sup> Individuals of this family were caught with the six sampling techniques. For the two light-traps, the data are presented for the two sampling years, with Y1 = 2004–2005 and Y2 = 2005–2006

74 families, 30 (40.5%) were caught both as larvae in the water column and as juveniles in benthic habitats; 40 (54.1%) were collected as larvae only and four families (5.4%) as juveniles only (Table 1). Some families were collected with only one sampling technique: ten



with the plankton-net only, two with the Aquafish LT only, nine with the Ecocean LT only, two with the underwater seine, and one by rotenone poisoning only (Table 1). Interestingly, seven families were caught by all six sampling techniques: Apogonidae, Blenniidae, Gobiidae, Pomacentridae, Scaridae, Tripterygiidae, and Tetraodontidae.

The plankton-net allowed the catching of 45 families, the most abundant being the Gobiidae and Engraulidae. The Aquafish LT allowed the catching of 21 families in 2004–2005 and 39 in 2005–2006. Samples from both years were dominated by Engraulidae and Pomacentridae. The Ecocean LT allowed the catching of 25 families in 2004–2005 and 52 in 2005–2006, with Siganidae and Lethrinidae, and Pomacentridae and Engraulidae being the most abundant, respectively (Table 1). The larval assemblages collected with the plankton-net and both light-traps were overlapping but clearly different ( $P = 0.001$ ,  $R = 0.560$  for plankton-net vs. Aquafish LT;  $P = 0.001$ ,  $R = 0.663$  for plankton-net vs. Ecocean LT), with this difference being due to a greater occurrence of Gobiidae, Syngnathidae, and Leiognathidae in the plankton-net samples (Table 2). The composition of assemblages collected with the two light-traps, although largely overlapping, were significantly different ( $P = 0.001$ ,  $R = 0.372$ ; Table 2), with Engraulidae and Clupeidae being more frequently encountered in the Aquafish LT.

The artificial reefs allowed the catching of 15 families only, the most abundant being Apogonidae and Blenniidae. The underwater seine allowed the catching of 21 families of juvenile fish, the most abundant being Labridae and Lethrinidae. Finally, the rotenone poisoning allowed the catching of 26 families, dominated by Pomacentridae and Apogonidae (Table 1). Only did the juvenile assemblages collected with the underwater seine and rotenone poisoning show an overlapping but significantly different composition ( $P = 0.001$ ,  $R = 0.664$ ; Table 2). Pomacentridae and Apogonidae were more frequent in the rotenone samples, whereas Labridae and Lethrinidae were more frequent in the underwater seine samples (Table 2).

### Size composition of catches

The six sampling techniques allowed the sampling of the complementary and successive size spectra of young fish. The size distribution of the larvae collected with the plankton-net was clearly separated from that of the larvae caught by light-traps: small individuals ranging from 2 to 5 mm were significantly more frequent in the plankton-net than in the two light-traps ( $P = 0.001$ ,  $R = 0.788$  for plankton-net vs. Aquafish LT;  $P = 0.001$ ,  $R = 0.809$  for plankton-net vs. Ecocean LT; Table 2). The fish larvae caught with the plankton-net had a mean size of 4.2 mm standard length (SL) and ranged from 0.8 to 64.8 mm SL, with individuals between 2 and 4 mm SL being the most abundant (Fig. 1a, left). The fish larvae caught by the two light-traps presented barely separated size distributions ( $P = 0.002$ ,  $R = 0.157$ ; Table 2): the size range of the larvae caught by the Aquafish LT was 1.3–117.4 mm SL, with a mean size of 17.9 mm SL, and the size range of the larvae caught by Ecocean LT was 1.8–233.2 mm SL, with a mean size of 16.1 mm SL (Fig. 1b, c, left). In both types of light-traps, the most abundant larvae were between 9 and 11 mm SL.

The size distributions of the juvenile fish collected with the three techniques used in benthic habitats overlapped greatly ( $P > 0.05$ ; Table 2). The juvenile fish caught with artificial reefs had a mean size of 22 mm SL and a size range of 8–97 mm SL, with individuals between 10 and 20 mm SL being the most abundant (Fig. 1d, left). The underwater seine and rotenone poisoning allowed the catching of juveniles with a mean size of 38 and 33 mm SL, respectively. The size range of the individuals caught with the underwater seine was slightly narrower, 10–113 mm SL, than that of the individuals caught with rotenone,

**Table 2** Results of the non-parametric analyses of similarities (ANOSIM) testing for significant differences in (a) taxonomic and (b) size composition between assemblages collected with the three techniques used for larvae, and with the three techniques used for juveniles. The results of analyses of contribution to the dissimilarity (SIMPER) are presented when significant differences were found to occur ( $P < 0.05$ ). Only families and size classes which contributed to at least 5% of the dissimilarity were presented

	Global ANOSIM		Pairwise ANOSIM			SIMPER	
	<i>R</i>	<i>P</i>	Pair	<i>R</i>	<i>P</i>	Code	Contrib (%)
<b>(a) Taxonomic composition</b>							
Larvae	0.544	0.001	Aqua × Ecoc	0.372	0.001	Engr	12.2
						Clup	9.2
						Poma	8.6
						Apog	7.3
						Siga	7.0
						Scor	6.1
						Cara	6.0
						Blen	5.1
			Aqua × plkN	0.560	0.001	Gobi	9.5
						Syng	7.8
						Leio	7.2
						Engr	6.3
						Clup	6.1
						Poma	5.8
			Ecoc × plkN	0.663	0.001	Gobi	7.7
						Syng	7.2
						Leio	6.4
						Engr	5.2
						Poma	5.1
Juveniles	0.071	0.020	ArtR × seine	0.053	0.072		
			ArtR × roten	0.051	0.163		
			Seine × roten	0.664	0.001	Poma	15.0
						Labr	11.0
						Leth	8.8
						Apog	7.9
						Scar	7.0
						Gobi	6.8
						Chae	5.9
<b>(b) Size composition</b>							
Larvae	0.699	0.001	Aqua × Ecoc	0.157	0.002	[10–11]	5.0
			Aqua × plkN	0.788	0.001	[2–3]	12.1
						[3–4]	11.0
						[4–5]	6.7
			Ecoc × plkN	0.809	0.001	[2–3]	15.4
						[3–4]	13.7
						[4–5]	8.4
						[5–6]	5.9
						[1–2]	5.5
Juveniles	0.030	0.817	ArtR × seine	0.019	0.307		
			ArtR × roten	0.075	0.910		
			Seine × roten	0.045	0.793		

Aqua = Aquafish light-trap; Ecoc = Ecocean light-trap; plkN = plankton-net; ArtR = artificial reef; seine = underwater seine; roten = rotenone poisoning; contrib = contribution to the dissimilarity

See Table 1 for the family codes. The size classes are given in mm

**Fig. 1a–f** Size distribution (number of individuals per standard length classes, SL in mm) of fish larvae and juveniles caught with the different sampling techniques (*left column*), and size range (SL in mm) and variability of individuals belonging to the seven families commonly collected with the six techniques (*right column*). The upper and lower limits of the boxes on the *right column* correspond to the first and third quartiles, respectively; the *horizontal bars* within the boxes indicate the median, the *errors bars* the 10th and 90th percentile, and the *dots* outside the boxes indicate the values outside the 10–90th percentile range. The *two dotted horizontal lines* on each graph illustrate the upper and lower limits of the most frequent size class represented in the samples from each technique. **a** Plankton-net. **b** Aquafish light-trap (Aquafish LT). **c** Ecocean light-trap (Ecocean LT). **d** Artificial reef. **e** Underwater seine net. **f** Small-scale rotenone poisoning. See Table 1 for the family codes

5–160 mm SL. Similarly, the most abundant individuals were slightly smaller in the underwater seine samples, between 20 and 30 mm SL, than in the rotenone samples, between 35 and 45 mm SL (Fig. 1e, f, left).

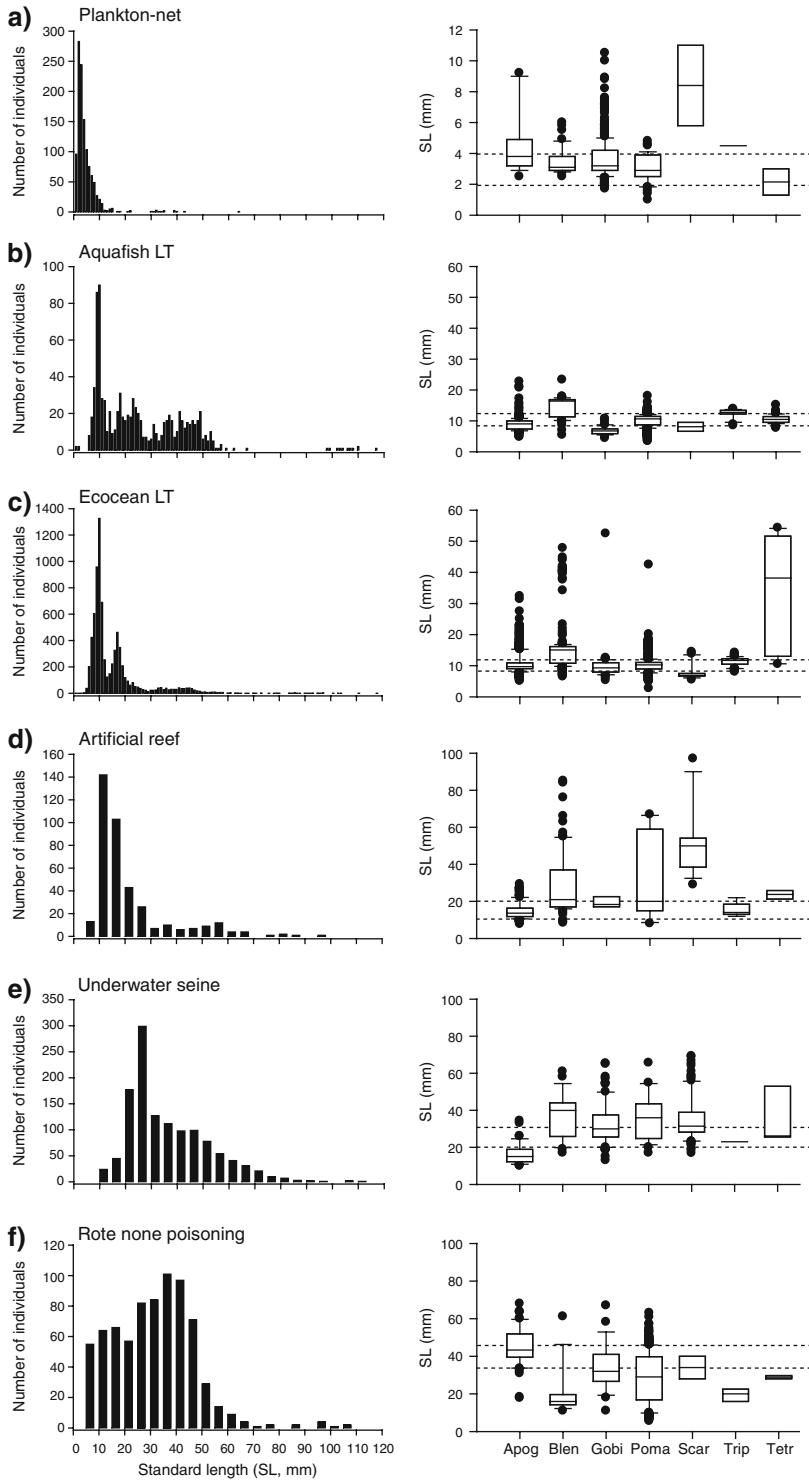
The sizes of the larvae and juveniles of the seven families collected by all six sampling techniques often ranged within the most abundant size class for each technique (Fig. 1, right). However, the Scaridae and Tripterygiidae collected with the plankton-net, the Tetraodontidae collected with the Ecocean LT, and the Scaridae collected with the artificial reefs were larger (Fig. 1a–e, right), and the Blenniidae, Tripterygiidae, and Tetraodontidae juveniles collected by rotenone poisoning were smaller (Fig. 1f, right).

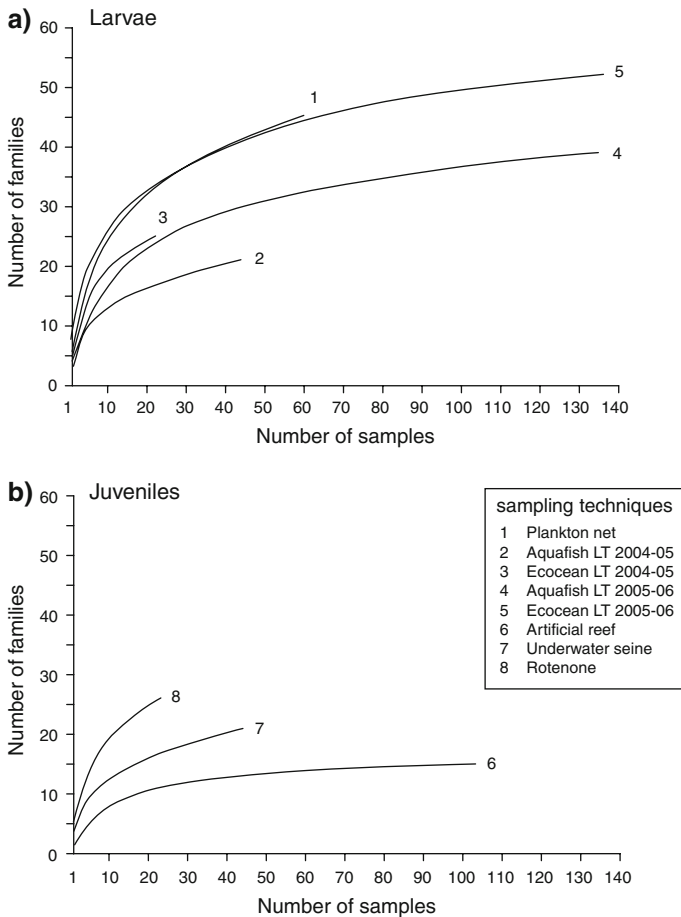
#### Relationship between the number of families caught and the number of samples

The number of samples required to obtain a similar number of fish families greatly differed between techniques (Fig. 2). For instance, the number of samples required to obtain 20 different families of larvae varied from six with the plankton-net to 37 with the Aquafish LT in 2004–2005 (Fig. 2a). For juveniles, the number of samples required to obtain ten different families varied from three with rotenone poisoning to five with the underwater seine and 16 with artificial reefs (Fig. 2b). Interestingly, the artificial reefs were the only technique for which the number of families caught rapidly reached a plateau when the sampling effort increased. Conversely, rotenone poisoning did not only collect a higher number of fish families when similar numbers of samples were performed, but the curve was still far from reaching a plateau after 30 samples (Fig. 2b).

#### Efficiency, selectivity, and habitat requirements of each technique

All of the samples collected with the plankton-net contained at least one larva (no empty sample). Only 7.6% of the samples performed with the Ecocean LT contained no fish (12 empty samples), compared with 15% for the Aquafish LT samples (27 empty samples). Conversely, over 46% of the samples collected with the artificial reefs contained no fish (89 empty samples), whereas less than 1% of the underwater seine samples and only 4.2% of the rotenone poisoning samples contained no fish (four and one empty samples, respectively; Table 3). The underwater seine and rotenone poisoning are the only techniques that cannot be used on all of the habitats of the lagoon (Table 3). Among the seven families that were caught with the six sampling techniques, some were abundantly caught with a specific technique, whereas others were abundantly collected regardless of which technique was used. For example, Tetraodontidae were abundant as pre-settlement larvae in Aquafish LT, whereas they were rarely caught as juveniles. Conversely, Pomacentridae and Apogonidae were easy to collect in abundance at all ontogenetic stages with all six techniques, except for the young larvae of Apogonidae, which were harder to collect with the plankton-net (Table 3).





**Fig. 2a, b** Increasing total number of different families caught by each technique and year as the sampling effort increases. The samples were entered in a random order 999 times. The resulting cumulative curves were then averaged to obtain a smooth curve for (a) larvae and for (b) juveniles

## Discussion

The selection of a technique for sampling young fishes is problematic, since it must deal with many correlated parameters, such as the targeted taxa, characterized by specific behaviors and associated catchability, the latter varying according to the ontogenetic stage and, thus, on the size (Murphy and Willis 1996). Moreover, since mortality rates also vary according to the ontogenetic stages (Leis 1991), the evaluation of young fish abundances in a specific location requires the association of several techniques targeting successive ontogenetic stages (Murphy and Clutter 1972). The first consideration to address will, thus, be the taxa, and size range, that are targeted (Murphy and Willis 1996). If this taxonomic range and/or size range are large, several techniques have to be associated. For example, our results indicate that 30 out of the 70 families collected at the larval stage were caught by the plankton-net when they were young and by light-traps when they were older. This result, which is consistent with a previous study on the Great Barrier Reef (Choat et al. 1993), is based on the fact that

**Table 3** General overview of the sampling techniques used for targeting different ontogenetic stages of young fish in different habitats

Developmental stage	Pre-settlement		Settlement		Post-settlement	
	Young larvae	Older larvae	Young juveniles	Older juveniles	Underwater seine	Rotenone poisoning
<b>Habitats</b>	Plankton-net	Aquafish light-trap	Artificial reef			
Coral reefs	No <sup>a</sup>	Yes <sup>b</sup>	Yes <sup>d</sup>	No	Yes <sup>e</sup>	
Seagrasses	Yes	Yes <sup>b</sup>	Yes <sup>d</sup>	Yes	No	
Seaweed beds	Yes	Yes <sup>b</sup>	Yes <sup>d</sup>	Yes	No	
<b>Families</b>						
Apogonidae	Present	Very abundant	Very abundant	Abundant	Very abundant	
Blenniidae	Abundant	Abundant	Very abundant	Abundant	Abundant	
Gobiidae	Very abundant	Present	Rare	Very abundant	Very abundant	
Pomacentridae	Abundant	Very abundant	Abundant	Abundant	Very abundant	
Scaridae	Rare	Rare	Abundant	Very abundant	Rare	
Tripterygiidae	Rare	Present	Very abundant	Rare	Present	
Tetraodontidae	Rare	Abundant	Rare	Rare	Rare	
Relative occurrence of empty samples (%)	0	15	~50	<1	<5	

<sup>a</sup> “Yes” indicates that the technique is well suited to the type of habitat; “No” indicates that other methods should be favored. A qualitative index of abundance from “rare” to “very abundant” is provided for the seven families which were caught with the six sampling techniques. Fishes of a given family were considered as very abundant if the family was among the five most abundant for a sampling technique, abundant if the family was among the ten most abundant for a sampling technique, or present otherwise. The family was considered as rare if less than five individuals were caught. The relative occurrence of empty samples is given as a mean to estimate the efficiency of each sampling technique

- <sup>a</sup> Cannot be used in shallow waters if pulled behind a boat, but see Strydom (2007) for other pulling methods
- <sup>b</sup> Cannot be used when water depth <4 m, but see Fisher and Bellwood (2002b) for a more compact design; efficiency may be limited by turbidity and current speed (Anderson et al. 2002; Lindquist and Shaw 2005)
- <sup>c</sup> Sampling is limited to the upper water layer
- <sup>d</sup> Cannot be used when water depth <2 m, but different types of benthic standard unit exist (see Valles et al. 2006)
- <sup>e</sup> Potentially destructive method and should be limited to collections on small coral colonies. Can be replaced by clove oil (but see Ackerman and Bellwood 2002)

plankton-nets and light-traps operate on different principles. A plankton-net actively catches larvae (Brogan 1994) whose taxonomic composition and size structure depend on the net's mouth diameter, towing speed, and mesh size (Barkley 1972; Morse 1989). Conversely, the efficiency of light-traps mainly depends on the ability of larvae to see a light, be attracted by this light, and swim towards it (Milicich et al. 1992). Small larvae that are poorly developed (Leis and Carson-Ewart 2000a) are more easily caught by plankton-nets than older larvae, which are very active swimmers and present highly developed sensory abilities (Leis and Carson-Ewart 2000b; Fisher and Bellwood 2002a). As a consequence, the adequacy of each technique depends more on the size of the larvae than on the family to which they belong (Choat et al. 1993). This confirms Brogan's conclusions (1994) that larval size is the most important consideration when choosing between techniques for sampling larvae in the water column. However, the taxa themselves must also be considered, since light-traps, for example, only target the species which are actively attracted by the light at the larval stages, so if the taxa targeted are not, light-traps will be inefficient (Doherty 1987). In the latter case, less selective passive techniques, such as crest-nets, may be used (McIlwain 2003). However, crest-nets are designed for sampling specifically at the barrier reef, whereas light-traps may be used in all lagoonal habitats. Depending on the objective of the sampling, light-traps or crest-nets may provide alternative methods for sampling various taxa of settlement-stage larvae in coral reefs (Hair et al. 2000).

A large overlap in the taxonomic diversity and size of the juveniles caught by the different sampling techniques was observed in the present study. Converse to what has been observed for larval fish, these sampling techniques, thus, seem to select juveniles mainly from their behavior. Positive thigmotaxis, i.e., an affinity of juveniles for physical structures that provide a mechanical stimulus (Chapman and Clynick 2006), as well as their swimming capabilities, or even more simply, their presence in a given habitat, may all play a role. Among the different techniques used for the sampling of reef fish juveniles, artificial reefs appear to be less efficient in terms of successful samples and the number of families caught. Indeed, about half of the samples contained no fish after 48 h and only 15 families were collected, a result identical to what has been observed on the Great Barrier Reef of Australia (Leis et al. 2002). However, the number of juveniles caught per artificial reef, and the number of taxonomic groups to which they belong, strongly increase with mooring time (Mellin 2007). Yet, when the mooring time is increased, the problem becomes to disentangle the part of the assemblage resulting from settlement processes and the part resulting from post-settlement interactions between individuals (Mellin 2007). Nevertheless, one important advantage of artificial reefs is that, like light-traps for fish larvae (Doherty 1987), they can be used to sample juvenile fish synoptically in different locations. Light-traps and artificial reefs are, thus, particularly interesting for investigating the spatial distributions of old larvae and young juveniles over large areas. The composition of the assemblages of juveniles obtained with the underwater seine also appears to be influenced by fish behavior, especially the position of the species in the water column (Murphy and Willis 1996) and their swimming capabilities (Allen et al. 1992). In seagrasses or macroalgal beds, the underwater seine tends to be more efficient for capturing juveniles swimming in the first meter above bottom and not trying to escape under or around the seine (Mellin, personal observation). As a consequence, Lethrinidae and Siganidae, although abundant in the samples, were caught with less efficiency in comparison to Labridae or Scaridae, for which nearly all of the observed individuals were successfully caught (Mellin, personal observation).

The present work provides a synthesis about various ways and means to capture the full range of young coral reef fishes in different habitats and gives an overview of their efficiency and limits. Larval and juvenile fish surveys are important, as they allow the

evaluation of recruitment success and, thus, the inter-annual variations in the replenishment of fish populations in specific locations (Fuiman and Werner 2002). Such surveys represent a crucial contribution to conservation and management plans of reef fish populations which are increasingly harvested throughout the world (Hughes et al. 2003). Conversely to previous studies, the present work provides a quantitative and qualitative evaluation of the efficiency of six techniques, targeting a broad range of ontogenetic stages of young reef fish. For reef fish larvae, our results, which are consistent with several studies, suggest that the principal factor to be determined before selecting a sampling technique is the size range of the targeted individuals (Barkley 1972; Thorrold 1992; Choat et al. 1993). For juveniles, our review suggests that the behavior of the individuals, as well as habitat characteristics, are the main factors that should determine the choice of a sampling technique. Further information about the effect of the timing of sampling, i.e., time of night or day, on the taxonomic and size composition of catches may be useful in order to fully evaluate the taxonomic and size selectivity of the sampling apparatus examined. To conclude, we hope that similar reviews will be compiled in the future for other sampling techniques in order to provide a broader overview of the most suitable techniques for sampling larvae and juveniles of different taxa and sizes in different coral reef habitats.

**Acknowledgments** We are most grateful to J. Baly and G. Mou-Tham for modifying the Aquafish light-traps, and the field and laboratory work. We also thank the different captains of the NO Coris and Aldric: M. Clarke, N. Colombani, and S. Tereua. A.M. Lassalette (University of the French West Indies, Martinique) helped in the editing of the English text. Thanks go to the two anonymous reviewers for their comments and detailed suggestions.

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