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# Strong links between metal contamination, habitat modification and estuarine larval fish distributions

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# ABSTRACT

Changes to larval fish assemblages may have far reaching ecological impacts. Correlations between habitat modification, contamination and marine larval fish communities have rarely been assessed in situ. We investigated links between the large-scale distribution of stressors and larval fish assemblages in estuarine environments. Larval fish communities were sampled using a benthic sled within the inner and outer zones of three heavily modified and three relatively unmodified estuaries. Larval abundances were significantly greater in modified estuaries, and there were trends towards greater diversity in these systems. Differences in larval community composition were strongly related to sediment metal levels and reduced seagrass cover. The differences observed were driven by two abundant species, *Paedogobius kimurai* and *Ambassis jacksoniensis*, which occurred in large numbers almost exclusively in highly contaminated and pristine locations respectively. These findings suggest that contamination and habitat alteration manifest in substantial differences in the composition of estuarine larval fish assemblages.

# 1. Introduction

A variety of anthropogenic activities contribute to widespread pollution and contamination in the marine environment, which influences the composition and health of ecological communities (Johnston and Roberts, 2009). Estuaries are generally believed to be exposed to the highest levels of contamination of any marine environment due to their proximity to human settlements and their position directly downstream of agricultural and industrial activities (Kennish, 2002; Lotze et al., 2006). Similarly, habitat modification in estuarine systems is widespread, and many estuaries around the world have experienced losses of seagrass, mangrove, saltmarsh and other vegetated habitats (Duke et al., 2007; Lotze et al., 2006; Waycott et al., 2009). Many of these complex estuarine habitats provide a 'nursery' function for ecologically and economically important species of fish (Beck et al., 2001; Boesch and Turner, 1984; Dorenbosch et al., 2004; Robertson and Duke, 1987; Taylor et al., 2005). Thus, it is imperative to understand how the modification of estuaries through contamination and loss of habitat could be impacting the early life stages of estuarine fish. Identifying stressors and monitoring ecological impacts in these communities is critical to managing and conserving native biodiversity in these systems.

It is well documented that toxic contaminants such as metals are found in fish at various stages of their life cycle, often at levels that may potentially reduce growth or survivorship (Alguezar et al., 2006: Guo et al., 2008: Isosaari et al., 2006: Kojadinovic et al., 2007: Miskiewicz and Gibbs, 1994). Evidence also points to the potential adverse effects of toxic substances on reproduction and development of fishes (Arkoosh et al., 1998; Hose et al., 1989; Jones and Reynolds, 1997; Kingsford et al., 1997; Robinet and Feunteun, 2002). The less toxic enriching contaminants (such as nutrients or sewage) may have either a weakly negative or largely positive effect on abundance and diversity of adult fish (McKinley and Johnston, 2010) however, the effects on the egg and larval stages have rarely been studied in situ. There are a small number of quantitative field studies examining these effects at the population and community level (Bervoets et al., 2005). This includes several studies demonstrating changes to the composition and distribution of larval fish communities around sewage plumes (Gray, 1996, 1997; Gray et al., 1992; Kingsford et al., 1997) and one study which found negative effects on embryo and larval development in relation to pulp mill effluent (Karas et al., 1991).

Modification to marine habitats represents another potential stressor of larval fish communities. Habitat degradation is the largest source of ecological modification globally and the greatest threat to biodiversity (Tilman et al., 1994). Australian estuaries have experienced widespread changes to vegetative habitats over the last century,

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with well documented losses of mangrove (Valiela et al., 2001), saltmarsh (Saintilan and Williams, 1999), and seagrass habitats (Walker and McComb, 1992) in south-eastern Australia. The reduced extent of these habitats within heavily modified estuaries arises due to a variety of anthropogenic activities including dredging, increased siltation, nutrient enrichment, contamination, clearing for coastal development, and alterations to natural tidal or fluvial patterns (Saintilan and Williams, 1999; Valiela et al., 2001; Walker and McComb, 1992). Degradation of estuarine macrophytes is likely to lead to changes to larval fish communities, as fishes require estuarine habitats to survive early stages of their life cycle (Beck et al., 2001; Boesch and Turner, 1984; Dorenbosch et al., 2004; Robertson and Duke, 1987).

We explore the impacts of large-scale anthropogenic effects on estuarine larval fish communities across heavily modified and relatively unmodified estuaries in New South Wales, Australia. Specifically, we examine how high levels of modification and contamination in the estuarine environment affect the composition, abundance, and diversity of larvae. In addition, we utilize water quality, sediment metals, and habitat coverage data to estimate the relative importance of these stressors within the broader modification regime.

#### 2. Methods

### 2.1. Study sites and sampling design

Larval fish were sampled in six estuaries along the south coast of New South Wales, Australia. These included three heavily modified estuaries – Port Jackson (33°44.258′S, 151°16.542′E), Botany Bay (33°59.352′S, 151°11.433′E), and Port Kembla (34°28.121′S, 150°54.410′E), as well as three relatively unmodified estuaries – Port Hacking (34°04.680′S, 151°09.311′E), Jervis Bay (35°04.762′S, 150°44.858′E), and the Clyde River (35°44.233′S, 150°14.272′E) (Fig. 1). The three heavily modified estuaries are all highly anthropogenically disturbed environments near large urban and industrial areas and are subject to intense commercial and recreational boating traffic, historic and ongoing contamination, greater recreational fishing activity, and widespread urbanization of their shoreline and catchment (Birch and Taylor, 1999; DPI, 2010; Henry and Lyle, 2003; Scanes, 2010). Compared to the modified estuaries hoating traffic, less urbanization of the coastline and catchment, and virtually no heavy industry (Birch and Taylor, 1999; DPI, 2010; Henry and Lyle, 2003; Scanes, 2010). Both the Clyde River (within Bateman's Bay Marine Park) and Jervis Bay

(Jervis Bay Marine Park) are within marine parks (NSW, 1999). Port Hacking is located between the suburbs of southern Sydney and the forested slopes of Royal National Park, which lines the southern border of the estuary. While not strictly within a marine park, Port Hacking's catchment is largely intact due to its proximity to the Royal National Park and there is no major industrial activity within the estuary (NSWDNR, 2010). Previous monitoring indicates that the heavily modified estuaries are nutrient enriched while nutrient levels in the relatively unmodified estuaries are less elevated (Scanes, 2010).

Each estuary was divided into an inner and outer zone which reflected predicted physio-chemical and contamination gradients. These zones were defined based on their physical and biological characteristics. The inner zone is further up the estuary and represents the lower reaches of the estuarine tributary where brackish waters occur. In this zone turbidity, temperatures, and nutrient levels are higher than in the outer zone (Dafforn et al., in press). The outer zone sites are near the marine entrance to the estuaries where salinity, coastal flushing, wave exposure and oceanic current systems have greater influence. In this zone sediment grain sizes are also larger and there is greater tidal influence (Dafforn et al., in press). Within each estuary six sites with bottom characteristics that allowed uninterrupted trawling were selected, three in each zone. All sampling was replicated over two seasons – the first in the Spring of 2009 and again in the late summer of 2010.

## 2.2. Sampling methods

Larvae were sampled using a benthic sled trawl towed along bare sediment behind a powerboat. Trawls were conducted along relatively flat profiles at a depth of 3–12 m. GPS was used to ensure that all trawls were 250 m in length, towed at a speed of 1.5 knots for approximately 5 min. The trawl was rigged to a four point bridle using an approximate 3:1 warp to depth ratio. The trawl frame consists of a stainless steel sled measuring approximately 1.5 m across and 2 m long. Within the sled frame two plankton nets were mounted in 50 cm diameter stainless steel rings 15 cm off the bottom. Each of these nets consisted of a 50 cm  $\times$  300 cm (long) conical plankton net with 250  $\mu$ m mesh. A 1 L plastic sample jar was affixed to each od end. This yielded two sample jars for each trawl, one of which was processed for data while the other was retained as a backup.

Because it is well known that the vertical and spatial distribution of larvae can be influenced by light conditions and diel period, several precautions were taken to ensure consistency in these variables (Bridger, 1956; Pittman and McAlpine, 2003). All sampling was conducted at night following the incoming tide up the estuary (ie. starting in the outer zone and moving inwards). In order to standardize light conditions sampling was conducted each month within a two week window around the new moon (one week before and one week after the new moon). All samples were immediately preserved in a buffered 5% formalin/seawater mixture for transportation back to the laboratory. Where possible larvae were sorted and identified to species using the current taxonomic standard (Neira et al., 1998). For some taxa larvae were only sorted to genus or family where current taxonomic knowledge is insufficient for species level identification. A variety of *Gobiidae* sp.



Fig. 1. Location of study sites in a) Port Jackson, b) Botany Bay, c) Port Hacking, d) Port Kembla, e) Jervis Bay, and f) Clyde River estuaries. • Indicates outer zone sites • Indicates inner zone sites.

### Table 1

Mean  $\pm$  SE. Water quality and benthic sediment metals values. 'All Metals' represents the normalized total of values for Co, Cr, Cu, Fe, Mn, Ni, Pb and Zn from Dafforn et al. (in press). Individual values are presented for Cu, Ni, and Pb.

|            |              |                | Temp (°C)                        |                                  | Sal ‰                            |                                 |                                 | рН                               |                                 |
|------------|--------------|----------------|----------------------------------|----------------------------------|----------------------------------|---------------------------------|---------------------------------|----------------------------------|---------------------------------|
|            |              |                | Outer                            | Inner                            | Outer                            | Inner                           |                                 | Outer                            | Inner                           |
| Heavily    | Port Jacl    | kson           | $22\pm0.2$                       | $23.5\pm0.6$                     | $36.3\pm0.3$                     | 34.7                            | ± 0.6                           | $8.1\pm0.0$                      | $7.9 \pm 8.5$                   |
| Modified   | Botany l     | Bay            | $\textbf{23.5} \pm \textbf{0.5}$ | $24.8 \pm 0.3$                   | $34.7 \pm 1.2$                   | 29.5                            | ± 2.5                           | $8.2\pm0.0$                      | $\textbf{8.0} \pm \textbf{0.0}$ |
| Estuaries  | Port Ker     | nbla           | $\textbf{22.8} \pm \textbf{0.2}$ | $\textbf{23.3} \pm \textbf{0.2}$ | $\textbf{36.3} \pm \textbf{0.2}$ | 36.1                            | ± 0.2                           | $\textbf{8.2}\pm\textbf{0.0}$    | $\textbf{8.2}\pm\textbf{0.0}$   |
| Relatively | Port Ha      | cking          | $21.8 \pm 0.6$                   | $\textbf{23.6} \pm \textbf{0.5}$ | $\textbf{35.4} \pm \textbf{0.2}$ | 34.9                            | ± 0.7                           | $\textbf{8.1} \pm \textbf{0.0}$  | $\textbf{8.0}\pm\textbf{0.0}$   |
| Unmodified | Jervis Ba    | ay             | $\textbf{22.3} \pm \textbf{0.4}$ | $22.7\pm0.3$                     | $\textbf{35.4} \pm \textbf{0.4}$ | 35.1                            | ± 0.6                           | $8.1\pm0.0$                      | $8.1\pm0.0$                     |
| Estuaries  | Clyde Ri     | iver           | $20.1\pm0.6$                     | $\textbf{23.0} \pm \textbf{0.8}$ | $35.5\pm0.7$                     | 30.8                            | ± 2.5                           | $\textbf{8.1}\pm\textbf{0.0}$    | $\textbf{7.9} \pm \textbf{0.0}$ |
|            |              | All Metals (1  | Normalized Total)                | $Cu (mg kg^{-1})$                |                                  | Ni (mg kg $^{-1}$               | )                               | $Pb (mg kg^{-1})$                |                                 |
|            |              | Outer          | Inner                            | Outer                            | Inner                            | Outer                           | Inner                           | Outer                            | Inner                           |
| Heavily    | Port Jackson | $-3.9\pm0.1$   | 6.3 ± 3.7                        | $9.5\pm3.2$                      | $151.7 \pm 37.7$                 | $1.0\pm0.3$                     | $16.2 \pm 4.1$                  | $11.0\pm0.4$                     | $243.1\pm68.7$                  |
| Modified   | Botany Bay   | $-2.7\pm0.3$   | $5.3\pm1.0$                      | $15.2\pm5.4$                     | $58.6 \pm 1.9$                   | $1.6\pm0.1$                     | $25.0\pm1.6$                    | $13.2\pm4.5$                     | $86.0\pm5.3$                    |
| Estuaries  | Port Kembla  | $15.9 \pm 1.8$ | $\textbf{7.5}\pm\textbf{3.4}$    | $118.1\pm17.4$                   | $163.4\pm64.3$                   | $10.5\pm1.9$                    | $16.0\pm2.0$                    | $\textbf{81.9} \pm \textbf{8.6}$ | $110.1\pm37.4$                  |
| Relatively | Port Hacking | $-1.6\pm0.9$   | $-7.4\pm0.8$                     | $31.6\pm20.0$                    | $\textbf{0.0} \pm \textbf{7.0}$  | $1.8 \pm 1.0$                   | $\textbf{0.9}\pm\textbf{2.2}$   | $17.3\pm10.6$                    | $\textbf{5.2} \pm \textbf{5.4}$ |
| Unmodified | Jervis Bay   | $-5.2\pm0.0$   | $-7.9\pm0.0$                     | $\textbf{7.6} \pm \textbf{2.8}$  | $\textbf{9.8}\pm\textbf{3.2}$    | $\textbf{0.8}\pm\textbf{0.3}$   | $\textbf{0.4} \pm \textbf{0.1}$ | $0.9\pm0.6$                      | $1.3\pm0.6$                     |
| Estuaries  | Clyde River  | $-2.5\pm0.2$   | $-3.7\pm1.3$                     | $1.2\pm0.8$                      | $\textbf{6.9} \pm \textbf{4.4}$  | $\textbf{2.4} \pm \textbf{0.4}$ | $\textbf{9.8}\pm\textbf{3.0}$   | $\textbf{2.2}\pm\textbf{0.3}$    | $10.0\pm3.0$                    |

were distinguished as morphologically distinct varieties and were included as separate 'species' within the analysis. Due to unresolved taxonomic issues, these unidentified goby species are presented as 'unidentified Gobiidae sp.'. See Appendix 1 for detailed species information.

A calibrated flow meter was affixed to the trawl and readings were used to standardize all larval abundance results to 100 m<sup>3</sup> of sea water. At each sampling location four replicate water samples of 1 L each were taken at a depth of 1 m using a water sampling tube. These samples were combined in a plastic container on the vessel and basic water quality data was measured from this water at the time of sampling using a calibrated VSI 6820 V2 sonde. In a parallel study surficial sediments were collected from sites in close proximity to the trawls. Samples were oven-dried before being digested and analyzed with ICP-OES following the methods outlined by Hill et al. (2009). Recoveries were calculated against certified reference materials and all metals used in this study were within accepted recovery limits. Arsenic and mercury recoveries were insufficient for further analysis (Dafforn et al., in press). Vegetative habitat size and cover were calculated using estuarine fact sheet monitoring values (NSWDNR, 2010).

# 2.3. Statistical analysis

All multivariate and univariate analysis was conducted using mixed model PERMANOVA in PRIMER v.6 (Anderson, 2001). Prior to analysis abundance data was log(x + 1) transformed. Bray–Curtis similarity matrices were calculated for multivariate data while Euclidean similarity matrices were used for univariate measures. The PERMANOVA design employed in the course of this analysis consisted of the following factors:

Mo-Modification - Heavily Modified or Relatively Unmodified (2 levels, Fixed).

Ti - Time - November or February (2 levels, Fixed).

Zo - Zone - Inner or Outer (2 levels, Fixed).

Es – Estuary(Modification) – (6 estuaries, Random).

 $Si - Site(Estuary(Modification) \times Zone) - (36 sites, Random).$ 

Monte Carlo *p*-values were used in some places where the number of unique permutations was less than 20. Analysis of water quality, metals, and habitat cover covariates was conducted using the DistLM function of PERMANOVA. This program calculates a distance-based multivariate multiple regression (e.g. dbRDA) for any linear model on the basis of any distance measure, using permutation procedures (McArdle and Anderson, 2001). Covariate factors were analyzed graphically using

Principal Coordinated Ordination (PCO). PCO is a computer program that performs a principal coordinate analysis of any symmetric distance matrix. This analysis is also called metric multi-dimensional scaling (Anderson, 2003).

# 3. Results

# 3.1. Estuary characteristics

Estuaries displayed similar average water quality conditions, though differences were found in most parameters between zones (Table 1). Higher sediment metals values were recorded in the modified estuaries, particularly in the inner zone sites where anthropogenic contamination is greater. See Dafforn et al. (in press) for detailed description and analysis of the sediment metals data. In many of the modified sites sediment metals values were above levels predicted to have biological effects according to water quality guidelines (ANZECC, 2010; Dafforn et al., in press). On average relatively unmodified estuaries had greater coverage of mangroves (22.4%) and seagrass (10.5%) relative to the modified estuaries (1.8%, 2.6% respectively). These vegetated habitats are virtually absent from Port Kembla and Port Jackson (NSWDNR, 2010). The coverage of saltmarsh was similar between relatively unmodified (4%) and modified (4.8%) estuaries though this was due to large saltmarsh patches in Botany Bay (Table 2).

# 3.2. Larval fish assemblages

In total more than 10,200 fish larvae were collected and identified during the study. The summarized larval dataset can be found in Appendix 1. The abundance of larval fish was significantly greater in the heavily modified estuaries (p = 0.003). There was a nonsignificant trend towards increased species richness (p = 0.19) and

# Table 2

Size of vegetative habitats and % of estuary with vegetative habitats (NSWDNR, 2010).

|            |              | Estuary Size | Habitat Size | (km <sup>2</sup> ) |           | % Habitat Co | ver      |           |
|------------|--------------|--------------|--------------|--------------------|-----------|--------------|----------|-----------|
|            |              |              | Seagrass     | Mangrove           | Saltmarsh | Seagrass     | Mangrove | Saltmarsh |
| Heavily    | Port Jackson | 49.7         | 0            | 0                  | 0         | 0.00         | 0.00     | 0.00      |
| Modified   | Botany Bay   | 80           | 6.238        | 4.227              | 11.573    | 7.80         | 5.28     | 14.47     |
| Estuaries  | Port Kembla  | 1.6          | 0            | 0                  | 0         | 0.00         | 0.00     | 0.00      |
| Relatively | Port Hacking | 11           | 0.807        | 0.307              | 0.082     | 7.34         | 2.79     | 0.75      |
| Unmodified | Jervis Bay   | 5.3          | 0.972        | 3.314              | 0.521     | 18.34        | 62.53    | 9.83      |
| Estuaries  | Clyde River  | 103.2        | 6.05         | 1.999              | 1.486     | 5.86         | 1.94     | 1.44      |

### Table 3

| Jnivariate analysis of the impacts of modification on a) Larval abundance, b) Species richness, c) Shannon diversity. Factors: Mo = Modification, Zo = Zone (Inner vs. Outer), |
|--|
| fi = Time of Sampling, Es = Estuary, Si = Site. Bold values correspond to plots in Fig. 2. 🗆 Indicates Monte Carlo p value.  |

| Source        | dF | a) Abundanc | e       |         | b) Species Ri | chness  |         | c) Shannon I | Diversity |         |
|---------------|----|-------------|---------|---------|---------------|---------|---------|--------------|-----------|---------|
|               |    | MS          | F       | p-value | MS            | F       | p-value | MS           | F         | p-value |
| Мо            | 1  | 19.36       | 36.598  | 0.003   | 46.722        | 2.273   | □0.19   | 0.36275      | 5.7006    | □0.074  |
| Zo            | 1  | 11.955      | 2.1936  | 0.189   | 4.5           | 0.13776 | 0.767   | 0.28464      | 0.54786   | 0.471   |
| Ti            | 1  | 8.1882      | 1.9609  | 0.254   | 200           | 5.4381  | 0.091   | 1.5799       | 9.8475    | 0.027   |
| Es(Mo)        | 4  | 0.529       | 0.18135 | 0.958   | 20.556        | 0.88729 | 0.473   | 6.36E-02     | 0.18753   | 0.949   |
| MoxZo         | 1  | 10.619      | 1.9486  | 0.243   | 12.5          | 0.38265 | 0.605   | 1.2568       | 2.419     | 0.212   |
| MoxTi         | 1  | 5.6766      | 1.3594  | 0.29    | 80.222        | 2.1813  | 0.204   | 0.58704      | 3.659     | 0.117   |
| ZoxTi         | 1  | 1.238       | 3.0933  | 0.143   | 10.889        | 1.4     | 0.317   | 4.29E-03     | 6.59E-02  | 0.816   |
| Es(Mo)xZo     | 4  | 5.4498      | 1.8683  | 0.131   | 32.667        | 1.4101  | 0.247   | 0.51954      | 1.5311    | 0.218   |
| Es(Mo)xTi     | 4  | 4.1757      | 3.3578  | 0.033   | 36.778        | 5.1719  | 0.007   | 0.16044      | 0.96128   | 0.454   |
| MoxZoxTi      | 1  | 3.7201      | 9.295   | 0.042   | 18            | 2.3143  | 0.18    | 0.27969      | 4.2995    | 0.116   |
| Si(Es(Mo)xZo) | 24 | 2.917       | 2.3457  | 0.024   | 23.167        | 3.2578  | 0.003   | 0.33933      | 2.0331    | 0.044   |
| Es(Mo)xZoxTi  | 4  | 0.40023     | 0.32184 | 0.855   | 7.7778        | 1.0937  | 0.364   | 6.51E-02     | 0.38977   | 0.812   |
| Res           | 24 | 1.2436      |         |         | 7.1111        |         |         | 0.1669       |           |         |

Shannon diversity (0.074) in the heavily modified estuaries (Fig. 2, Table 3).

Multivariate analysis of the community composition found that modified and unmodified estuaries differed significantly (p = 0.028) (Fig. 3). Several of the random interaction terms were also significantly different (e.g. Mo × Zo × Ti). Here and elsewhere, the test of the main effects can still be considered, as the higher level fixed factor effect remains relevant regardless of the outcome of the interaction with a random factor (Quinn and Keough, 2002). Simper analysis revealed that the top six species contributing to this difference were *Paedogobius kimurai* (wide gape paedomorphic goby), *Gobiopterus semivestita* (transparent goby), *Arenigobius* spp. (bridled goby spp.), *Hyperlophus vittatus* (sandy sprat), *Hyperlophus transucidus* (translucent sprat) and *Ambassis jacksoniensis* (Port Jackson glassfish) (Fig. 4, Table 5). Collectively these species accounted for 37% of the difference between the modified and relatively unmodified estuaries.

# 3.3. Covariates analysis – modified vs. relatively unmodified estuaries

Salinity, pH, temperature, all sediment metals, % seagrass, % mangrove, and % saltmarsh cover were all found to have a significant relationship with the larval community composition according to the DistLM analysis (Table 4b). However, DistLM is considered a poor predictor of the relative strength of these effects and so it was used only to identify appropriate covariates to test in the PCO (Anderson, 2003; McArdle and Anderson, 2001). As stated earlier, multivariate analysis of community composition found that modified and relatively unmodified sites differed significantly

(p = 0.028). PCO plots indicate that the major cluster of modified sites correspond strongly to both increased sediment metals levels and decreased coverage of seagrass. Salinity also correlates strongly but did not show a clear trend by modification (Fig. 5a). All sediment metals trended in approximately the same direction and were inversely related to vegetative cover, such that the most contaminated sites occurred primarily in the estuaries with the lowest seagrass cover. This suggests that there is a strong relationship between sediment metals levels, reduced vegetative cover, and community composition in the heavily modified sites showed little relationship to the metals and vegetative cover covariates (Fig. 5a, Table 4).

Fig. 5b plots the species which were highly correlated with the major clusters of modified and relatively unmodified sites (those with a correlation factor >0.6). In order of the power of this relationship, *P. kimurai, G. semivestita, A. bifrenatus*, and *H. transucidus* are more abundant in the sites that are highly metal contaminated and have lower seagrass cover (Fig. 5b). In order of the power of this relationship, *A. jacksoniensis*, and *H. vittatus* are more abundant in the relatively unmodified sties (Fig. 5b). Notably, *P. kimurai* and *A. jacksoniensis* were the 2nd and 3rd most abundant species in this study, and each was encountered almost exclusively in modified/ relatively unmodified sites (respectively).

# 4. Discussion

We documented large differences between larval fish assemblages living in heavily modified and relatively unmodified estuaries. Total abundance of fish larvae was significantly greater in the



Fig. 2. Mean ± SE a) Larval abundance, b) Species richness, and c) Shannon diversity in estuaries of differing levels of modification. Dotted line is average across all samples.



**Fig. 3.** Three dimensional MDS plot of multivariate assemblage composition by modification. Symbols represent centroids of the assemblage composition. Heavily modified includes sites in Port Jackson, Botany Bay, and Port Kembla. Relatively unmodified includes sites in Port Hacking, the Clyde River, and Jervis Bay.

heavily modified estuaries and trends suggest that diversity could also be higher in these modified environments. However, certain species were strongly negatively associated with estuary modification. Differences in community composition were strongly related to both sediment metal levels and reduced seagrass cover in heavily modified estuaries. Contamination levels and seagrass cover are inversely related and experimental work is needed to establish causation and to parse out the relative contribution of each factor to the observed patterns.

# 4.1. Positive effects of estuary modification

Increased abundances of fishes in modified environments are unlikely to result directly from either increased levels of metal contamination or reduced cover of seagrass habitat. A far more likely cause of this pattern would be nutrient enrichment. Monitoring indicates that nutrient levels in the three heavily modified estuaries are elevated compared to the relatively unmodified estuaries (Scanes, 2010) although data was not available at sufficient resolution to formally analyze this relationship. Trends in this study suggest that larval fish communities may also be more diverse in the modified estuaries. Several studies have demonstrated that forms of contamination which have an enriching effect (e.g. nutrient run-off, fish farms, sewage, hydrocarbons, etc.) increase both the abundance and diversity of adult fish assemblages (McKinley and Johnston, 2010; McKinley et al., in review). This is the first study to observe positive relationships between anthropogenic modification of estuaries and the abundance of larval fish.

# 4.2. Impacts of metals contamination

A variety of studies indicate that adult fish are fairly resilient in the face of anthropogenic contaminants and adult fish assemblages do not appear to be as sensitive to contaminants as invertebrates or fish larvae (Johnston and Roberts, 2009; McKinley and Johnston, 2010). In most field studies contaminants have been shown to have either weakly negative or a largely positive effect (where enriching contaminants are present) on adult fish abundance and diversity (McKinley and Johnston, 2010). However, most of these studies have focused primarily on adults of large bodied predatory fishes which are highly mobile (McKinley and Johnston, 2010). This contrasts to the larvae examined in this study, which are primarily small bodied species that are comparatively less mobile at maturity (e.g. Gobiidae, Clupeidae and Apogonidae spp.). Whilst large species may accumulate contaminants to a greater degree than larvae or invertebrates due to their high trophic position, they



Fig. 4. Mean ± SE larval abundance by estuary for top six species contributing to differences between heavily modified and relatively unmodified estuaries.

### Table 4

a) Multivariate analysis of the impacts of modification on larval community composition. Factors: Mo = Modification, Zo = Zone (Inner vs. Outer), Ti = Time of Sampling, Es = Estuary, Si = Site. b) Results of DistLM covariate analysis. Bold values correspond to Fig. 3 plot.  $\Box$  Indicates Monte Carlo *p* value.

| Source        | dF   | a) Commu        | nity Composition |                 |
|---------------|------|-----------------|------------------|-----------------|
|               |      | MS              | F                | <i>p</i> -value |
| Мо            | 1    | 18,770          | 2.8208           | 0.028           |
| Zo            | 1    | 7057.6          | 1.6089           | 0.219           |
| Ti            | 1    | 8514.7          | 2.3778           | 0.087           |
| Es(Mo)        | 4    | 6654.1          | 3.0881           | 0.001           |
| MoxZo         | 1    | 5412.3          | 1.2338           | 0.349           |
| MoxTi         | 1    | 4482.7          | 1.2519           | 0.319           |
| ZoxTi         | 1    | 1968.9          | 1.1223           | 0.371           |
| Es(Mo)xZo     | 4    | 4386.7          | 2.0358           | 0.001           |
| Es(Mo)xTi     | 4    | 3580.9          | 2.6272           | 0.001           |
| MoxZoxTi      | 1    | 2231.7          | 1.2721           | 0.316           |
| Si(Es(Mo)xZo) | 24   | 2154.8          | 1.5809           | 0.001           |
| Es(Mo)xZoxTi  | 4    | 1754.3          | 1.2871           | 0.14            |
| Res           | 24   | 1363            |                  |                 |
| Variable      | b) D | istLM Covariate | Results          |                 |
|               | SS   |                 | F                | <i>p</i> -value |
| Temp (°C)     | 8.51 | E+03            | 3.1395           | 0.003           |
| Sal           | 7.00 | )E+03           | 2.5608           | 0.004           |
| pH            | 5.49 | 0E+03           | 1.992            | 0.029           |
| Co 228.616    | 1.82 | 2E+04           | 7.0587           | 0.001           |
| Cr 267.716    | 1.96 | 6E+04           | 7.6637           | 0.001           |
| Cu 327.393    | 1.06 | 6E+04           | 3.9677           | 0.001           |
| Fe 238.204    | 1.68 | 8E+04           | 6.4673           | 0.001           |
| Mn 257.610    | 1.42 | 2E+04           | 5.4071           | 0.001           |
| Ni 231.604    | 1.95 | 6E+04           | 7.6265           | 0.001           |
| Pb 220.353    | 1.36 | 6E+04           | 5.1363           | 0.001           |
| Zn 206.200    | 1.55 | 6E+04           | 5.9537           | 0.001           |
| %Seagrass     | 1.10 | 0E+04           | 4.1003           | 0.001           |
| %Mangrove     | 6.20 | 0E+03           | 2.2589           | 0.011           |
| %Saltmarsh    | 940  | 9.6             | 3.4858           | 0.001           |
| Estuary Area  | 7.35 | 5E+03           | 2.6935           | 0.003           |

can be highly mobile so direct exposure times are not certain, their diets are comparatively diverse, and they have a higher capacity for physiological resistance and tolerance (van der Oost et al., 2003; Wirgin and Waldman, 1998).

In contrast, significant evidence points to developmental and reproductive susceptibility to contaminants in fish populations (Arkoosh et al., 1998; Hose et al., 1989; Jones and Reynolds, 1997; Kingsford et al., 1997; Robinet and Feunteun, 2002). Studies have found reduced egg and larval abundance due to exposure to sewage sludge (Waring et al., 1996), behavioral and metabolic changes with heavy metal exposure (Kienle et al., 2008; Sreedevi et al., 1992), increased incidents of larval deformity due to sewage plumes, chemical effluent and tributylin exposure (Hu et al., 2009; Kingsford et al., 1997; Vetemaa et al., 1997), and reduced fish condition, survivorship and growth with exposure to metals (Bervoets and Blust, 2003; Canli and Atli, 2003; Hutchinson et al., 1994). While few of these effects have been verified in at the community level in wild populations, it is not unreasonable to expect that many of these impacts could be detected in wild larval fish assemblages. As such, it is probable that contamination impacts have directly contributed to differences in the larval fish assemblage between modified and relatively unmodified estuaries.

In this study the abundance of several species were positively correlated with highly contaminated areas, which could imply that these species favor these sites or are comparatively resistant to pollution effects. Notably, *P. kimurai* were very strongly associated with sediment metals levels. This species was extremely abundant in highly contaminated sites; in such areas they accounted for

| Univariate analys<br>vs. Outer), Ti = Ti | is of ti<br>ime of | he impacts<br>f Sampling       | t of modification of the formula of | ation on the<br>ary, Si = Sit | e abundance<br>te. Bold valu    | t of the top sizes correspon | x species co<br>d to plots i | ontributing 1<br>n Fig. 4. 🗆      | to difference<br>Indicates M | es between<br>lonte Carlo | i heavily m<br>p value.        | odified and           | l relatively    | unmodified                        | l estuaries. l          | Factors: Mo     | = Modifica                        | tion, Zo = Z              | one (Inner      |
|--|--------------------|--------------------------------|---|-------------------------------|---------------------------------|------------------------------|------------------------------|-----------------------------------|------------------------------|---------------------------|--------------------------------|-----------------------|-----------------|-----------------------------------|-------------------------|-----------------|-----------------------------------|---------------------------|-----------------|
| Source                                   | dF                 | a) <i>Ambass</i><br>Port Jacks | is jacksonie<br>on Glassfish  | nsis<br>1                     | b) <i>Paedogo</i><br>Gape Paedo | bius kimurai<br>omorphic Go  | Wide<br>by                   | c) <i>Arenigob</i><br>Bridled Gol | iius spp.<br>by spp.         |                           | d) <i>Gobiopt</i><br>Transpare | erus semiv<br>nt Goby | estita          | e) <i>Hyperlo</i> j<br>Translucen | ohus transuc<br>t Sprat | idus            | f) H <i>yperlop</i><br>Sandy Spra | <i>hus vittatus</i><br>it |                 |
|  |                    | MS                             | Н   | <i>p</i> -value               | MS                              | F                            | <i>p</i> -value              | MS                                | F                            | <i>p</i> -value           | MS                             | F                     | <i>p</i> -value | MS                                | F                       | <i>p</i> -value | MS                                | F                         | <i>p</i> -value |
| Mo                                       | -                  | 27.921                         | 21.09   | 0.005                         | 106.8                           | 5.0101                       | 0.092                        | 45.827                            | 6.2256                       | 0.077                     | 47.307                         | 3.9835                | 0.107           | 29.698                            | 2.9652                  | 0.145           | 0.4408                            | 3.11E-02                  | 0.875           |
| Zo                                       | 1                  | 5.3218                         | 1.0103  | 0.38                          | 11.16                           | 2.5078                       | 0.178                        | 1.1148                            | 0.60947                      | 0.459                     | 41.333                         | 5.1813                | 0.048           | 20.957                            | 3.4281                  | 0.079           | 2.25E-02                          | 7.88E-03                  | 0.941           |
| Τi                                       | 1                  | 4.9414                         | 0.79944   | 0.393                         | 3.3772                          | 1.4373                       | 0.301                        | 11.025                            | 41.378                       | 0.003                     | 0.81232                        | 1.1328                | 0.356           | 1.116                             | 1.6502                  | 0.275           | 2.5402                            | 0.22698                   | 0.689           |
| Es(Mo)                                   | 4                  | 1.3239                         | 0.58894   | 0.657                         | 21.317                          | 17.73                        | 0.001                        | 7.3611                            | 4.3679                       | 0.007                     | 11.876                         | 9.1728                | 0.001           | 10.016                            | 22.901                  | 0.001           | 14.19                             | 7.4158                    | 0.002           |
| MoxZo                                    | 1                  | 2.756                          | 0.5232  | 0.517                         | 12.333                          | 2.7714                       | 0.172                        | 2.257                             | 1.2339                       | 0.298                     | 15.455                         | 1.9374                | 0.314           | 23.701                            | 3.8769                  | 0.048           | 0.21389                           | 7.51E-02                  | 0.817           |
| MoxTi                                    | 1                  | 2.2305                         | 0.36086   | 0.583                         | 9.1559                          | 3.8965                       | 0.132                        | 5.13E-02                          | 0.19262                      | 0.663                     | 0.1117                         | 0.15577               | 0.693           | 0.58669                           | 0.86754                 | 0.362           | 3.7085                            | 0.33138                   | 0.601           |
| ZoxTi                                    | 1                  | 1.4214                         | 10.022  | 0.047                         | 1.3765                          | 0.93777                      | 0.409                        | 0.61466                           | 0.2738                       | 0.634                     | 0.74144                        | 0.8802                | 0.409           | 0.47051                           | 0.63674                 | 0.492           | 2.9029                            | 0.66492                   | 0.457           |
| Es(Mo)xZo                                | 4                  | 5.2677                         | 2.3433  | 0.083                         | 4.4502                          | 3.7012                       | 0.02                         | 1.8291                            | 1.0854                       | 0.365                     | 7.9772                         | 6.1616                | 0.004           | 6.1134                            | 13.978                  | 0.001           | 2.8487                            | 1.4887                    | 0.24            |
| Es(Mo)xTi                                | 4                  | 6.181                          | 3.6434  | 0.02                          | 2.3498                          | 4.6984                       | 0.008                        | 0.26644                           | 0.16116                      | 0.952                     | 0.71707                        | 1.083                 | 0.375           | 0.67627                           | 8.6253                  | 0.001           | 11.191                            | 6.6171                    | 0.002           |
| MoxZoxTi                                 | 1                  | 6.0243                         | 42.478  | 0.003                         | 0.67833                         | 0.46214                      | 0.515                        | 6.9228                            | 3.0837                       | 0.167                     | 8.64E-02                       | 0.1026                | 0.771           | 0.95335                           | 1.2902                  | 3.13E-01        | 9.446                             | 2.1637                    | 0.204           |
| Si(Es(Mo)xZo)                            | 24                 | 2.248                          | 1.3251  | 0.255                         | 1.2024                          | 2.4041                       | 0.021                        | 1.6853                            | 1.0194                       | 0.494                     | 1.2947                         | 1.9554                | 0.052           | 0.43735                           | 5.578                   | 0.002           | 1.9135                            | 1.1314                    | 0.401           |
| Es(Mo)xZoxTi                             | 4                  | 0.14182                        | 8.36E-02  | 0.99                          | 1.4678                          | 2.9349                       | 0.042                        | 2.2449                            | 1.3579                       | 0.29                      | 0.84235                        | 1.2723                | 0.289           | 0.73894                           | 9.4246                  | 0.001           | 4.3658                            | 2.5813                    | 0.064           |
| Res                                      | 24                 | 1.6965                         |   |                               | 0.50012                         |                              |                              | 1.6532                            |                              |                           | 0.66209                        |                       |                 | 7.84E-02                          |                         |                 | 1.6913                            |                           |                 |



**Fig. 5.** Principal Coordinated Ordination (PCO) of correlations between covariate factors and two dimensional plots of community composition by modification. a) Metals contamination, habitat modification, and water quality covariates. (Pearson Correlation > 0.2). b) Plots of top six species contributing to differences between heavily modified and relatively unmodified estuaries (Pearson Correlation > 0.6).

60-95% of the larval assemblage and they were hyper-abundant particularly in the most highly contaminated inner zone sites of Port Jackson and Port Kembla. In contrast, this species was rarely encountered in the relatively unmodified estuaries. P. kimurai occurred in the highly metals contaminated sites both in estuaries where there were virtually no vegetative habitats (Port Jackson and Port Kembla) and directly adjacent to mangrove and seagrass patches (Botany Bay). This implies that this species is particularly successful in highly contaminated sites and may be unusually resistant to contaminants. Contamination resistance in fish has been demonstrated in some cases (Wirgin and Waldman, 2004; Xie and Klerks, 2004). In addition, the distribution of this species is unusually patchy and there is some speculation that it is an invasive originating from South East Asia (eg. Thailand). However, this has not been confirmed (Iwata et al., 2001; Neira et al., 1998). P. kimurai is also unusual among fish as it is sexually mature at a very small size - adults average approximately 1.5 cm in length and females pregnant with eggs are approximately the same size as other gobies' larvae (Iwata et al., 2001). It has been demonstrated that a variety of highly invasive invertebrate and fish species are rapidly maturing and unusually contamination resistant (Alcaraz et al., 2005; Dafforn et al., 2009; Piola and Johnston, 2008). It is possible that this species is also a marine invader displaying these characteristics.

## 4.3. Impacts of vegetative habitat alteration

Correlative studies are limited in their ability to predict the magnitude and relative importance of covarying factors (Shipley, 2002). In this study, sites with high levels of metals in the sediment also tended to have reduced coverage of vegetative habitats; this is likely the case because the core mechanisms of estuary contamination exposure (e.g. run-off, urbanization of shoreline/ catchment, outflows, etc.) also tend to precipitate habitat alteration in estuarine systems (Drinkwater and Frank, 1994; Rogers et al., 2002). Run-off which carries metals and other contaminants is also known to increase turbidity in many cases, hence lowering light levels and impacting plant growth (Longstaff and Dennison, 1999). In many cases increased nutrient levels accompany other forms of contamination (McKinley and Johnston, 2010). Increased

nutrient availability has been shown to increase the relative dominance of epiphytic plants in seagrass beds, often to the detriment of the seagrass community (Harlin and Thorne-Miller, 1981). For these reasons losses of some estuarine vegetative habitats may be strongly correlated with levels of contaminant exposure. Contaminants such as metals are also acutely toxic to some seagrass and other plant species, and trace metal run-off is a well documented cause of seagrass habitat loss (Macinnis-Ng and Ralph, 2002; Prange and Dennison, 2000; Warnau et al., 1995). It is therefore difficult to distinguish the relative effects of contamination vs. habitat loss as these stressors may be intimately related in the estuarine system.

Many of the species which contributed strongly to the trends in this study feed on vegetative matter, lay their eggs on plants, or use estuarine vegetation for shelter during the larval and post settlement stages of their life cycle (Miskiewicz, 1987). It is therefore possible that changes to the larval assemblage have directly resulted from loss of vegetative habitats in the modified estuaries. For example, A. jacksoniensis and F. lentiginosus primarily settle in seagrass beds during their juvenile and adult stages while juvenile and adult Gerres subfasciatus (roach) utilize mangroves, so changes to, or the absence of these habitats could explain decreased abundance of these species in the modified estuaries (Gray et al., 1996; Jelbart et al., 2007; Jenkins et al., 1997; Neira et al., 1998). While most of the larvae sampled in this study were taken over bare sediment and were at the pre-settlement (planktonic) stage of their life cycle, changes to these vegetative habitats could impact these larvae when they reach their juvenile and adult stages. It is therefore possible that losses of vegetative habitats have reduced the available habitat for these species, which would reduce the population size and hence larval abundance in the long term.

### 4.4. Estuarine opportunists vs. truly estuarine species

It is well known that a variety of fish species utilize estuaries during their life cycle. Many 'estuarine opportunist' species spawn at sea and find their way into the estuarine environment using oceanic currents for transport, entering the estuary at the preflexion and postflexion stages (after hatching) based on a variety of environmental cues (Neira and Potter, 1992; Norcross and Shaw,

1984; Potter et al., 1988). In contrast, truly estuarine species live within the estuary for their entire life cycle, including when spawning. Of the species which contributed most to the differences between heavily modified and relatively unmodified estuaries four are truly estuarine species (P. kimurai, G. semivestita, Arenigobius *spp.*, and *H. transucidus*) while the other two (*A. jacksoniensis* and *H. vittatus*) live primarily within the estuary but spawn at sea (Miskiewicz, 1987: Neira et al., 1998). All of these species spend the majority of their life cycle in the estuarine environment and so changes to that environment are likely to have an impact, regardless of whether or not they spawn at sea or within the estuary. However, it is possible that high levels of contamination and habitat modification disproportionately impact species which spawn in the estuary environment itself. Estuarine opportunist species which spawn at sea and enter estuaries later in their life cycle may be less affected by these stressors as they may be less exposed to contaminants during their early (pre-hatching) growth stages. It is therefore possible that impacts experienced during larval stages of estuarine fishes' could reduce their relative competitiveness and hence increase the dominance of taxa which spawn at sea. It is also possible that some of the estuarine opportunist species are descended from parents who lived in other estuaries and may not have been exposed to contamination levels reflective of the conditions of the estuary in which they were found during the larval phase of their life cycle.

It should be noted that the temporal and spatial variability of larval fishes has been found to be a significant issue in previous studies (Gray, 1996, 1997; Gray and Miskiewicz, 2000). In this study we sampled over a large spatial scale with a relatively high level of spatial replication both within and between estuaries. Despite significant differences between the random factor of sites for many analyses, our sampling design and level of replication was sufficiently robust to show clear differences by estuary and modification. We have also sampled across two seasons, during which a large proportion of estuarine species would have been breeding (Neira et al., 1998). Despite a high degree of temporal variability between these rounds of sampling, time was not sufficiently strong to produce a significant result in most of our analyses. For these reasons we do not believe our results to be strongly spatially or temporally restricted.

# 5. Conclusion

It is clear that there are large-scale differences between the larval fish assemblages living in heavily modified and relatively unmodified estuaries. Differences in larval fish community composition were strongly related to both sediment metal levels and reduced seagrass cover in heavily modified sites. We believe that it is likely that habitat alteration and estuarine sediment contamination are interrelated stressors which have contributed to the observed differences between modified and relatively unmodified estuaries. Ultimately changes to larval fish assemblages may have far reaching ecological impacts both for the adult fish community and other organisms. Notably, the impacts of stressors at the larval stage of economically and culturally important fish species are poorly studied and little understood. The absence of studies examining anthropogenic impacts on estuarine fish larvae represents a major gap in the environmental impact literature and further investigation and monitoring is warranted.

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# Appendix

### Appendix 1

Average abundance data identified to lowest taxonomic level by zone and estuary. Gobiidae sp. represent the total of all observed morphologically distinct taxa which could not be identified to species.

| Family                | Taxon                  | Botany | Bay   | Port Jac | kson  | Port Ke | mbla  | Clyde R | iver  | Jervis B | ay    | Port Ha | cking |
|-----------------------|------------------------|--------|-------|----------|-------|---------|-------|---------|-------|----------|-------|---------|-------|
|                       |                        | Outer  | Inner | Outer    | Inner | Outer   | Inner | Outer   | Inner | Outer    | Inner | Outer   | Inner |
| Ambassidae            | Ambassis jacksoniensis | 26.32  | 1.12  | 5.02     | 0.33  | 6.62    | 3.35  | 20.78   | 10.86 | 570.10   | 33.34 | 46.69   | 6.66  |
| Glassfish             | Port Jackson Glassfish |        |       |          |       |         |       |         |       |          |       |         |       |
|                       | Ambassis marianus      | 0.00   | 0.00  | 0.34     | 0.00  | 0.00    | 0.00  | 0.00    | 0.00  | 0.00     | 0.00  | 0.00    | 0.00  |
|                       | Estuary Glassfish      |        |       |          |       |         |       |         |       |          |       |         |       |
| Apogonidae            | Foa sp.                | 0.00   | 0.00  | 0.00     | 3.08  | 0.00    | 0.00  | 0.00    | 0.00  | 0.00     | 0.00  | 0.00    | 0.00  |
| Cardinalfish          | Cardinalfish sp.       |        |       |          |       |         |       |         |       |          |       |         |       |
|                       | Siphamia cephalotes    | 0.36   | 0.00  | 3.47     | 0.00  | 0.00    | 0.00  | 0.00    | 0.00  | 18.96    | 2.68  | 0.00    | 0.00  |
|                       | Wood's Siphonfish      |        |       |          |       |         |       |         |       |          |       |         |       |
|                       | Apogonidae sp. A       | 2.51   | 0.00  | 1.98     | 0.00  | 0.00    | 0.00  | 0.00    | 0.00  | 0.37     | 0.00  | 0.00    | 0.64  |
|                       | Cardinalfish sp. A     |        |       |          |       |         |       |         |       |          |       |         |       |
|                       | Apogonidae sp. B       | 0.00   | 0.00  | 2.36     | 0.33  | 0.00    | 0.00  | 0.00    | 0.00  | 0.67     | 0.27  | 0.36    | 0.00  |
|                       | Cardinalfish sp. B     |        |       |          |       |         |       |         |       |          |       |         |       |
|                       | Apogonidae sp. C       | 0.00   | 0.00  | 0.99     | 0.99  | 0.00    | 0.42  | 0.00    | 0.00  | 0.00     | 0.00  | 0.00    | 0.00  |
|                       | Cardinalfish sp. C     |        |       |          |       |         |       |         |       |          |       |         |       |
| Atherinidae sp.       | Atherinidae sp.        | 0.73   | 1.25  | 0.00     | 0.00  | 0.00    | 0.00  | 0.00    | 0.00  | 0.00     | 2.15  | 0.39    | 0.00  |
| Old World Silversides | Hardyhead sp.          |        |       |          |       |         |       |         |       |          |       |         |       |
| Belonidae             | Hempheridae sp.        | 0.73   | 1.25  | 0.00     | 0.00  | 0.00    | 0.00  | 0.76    | 0.49  | 0.00     | 0.00  | 0.39    | 0.00  |
| Needlefish            | Garfish sp.            |        |       |          |       |         |       |         |       |          |       |         |       |

# Appendix 1 (continued)

| Family                             | Taxon                                       | Botany | Bay    | Port Jac | kson   | Port Kei | nbla   | Clyde R | iver  | Jervis Ba | ay    | Port Ha | cking |
|------------------------------------|---|--------|--------|----------|--------|----------|--------|---------|-------|-----------|-------|---------|-------|
|                                    |   | Outer  | Inner  | Outer    | Inner  | Outer    | Inner  | Outer   | Inner | Outer     | Inner | Outer   | Inner |
| Blenniidae                         | Omobranchus anolius                         | 5.03   | 3.73   | 0.50     | 0.00   | 0.35     | 0.00   | 0.00    | 0.60  | 0.00      | 0.00  | 0.00    | 0.00  |
| Blennies                           | Oyster Blenny<br>Omobranchus rotundicens    | 1 09   | 0.00   | 0.00     | 5 30   | 0.00     | 0.00   | 0 38    | 0.77  | 0.00      | 0.00  | 0.00    | 0.00  |
|                                    | Combtooth Blenny                            | 1.05   | 0.00   | 0.00     | 5.50   | 0.00     | 0.00   | 0.50    | 0.77  | 0.00      | 0.00  | 0.00    | 0.00  |
|                                    | Petroscirtes lupus                          | 0.00   | 0.00   | 0.00     | 0.34   | 0.00     | 0.00   | 0.00    | 0.00  | 0.00      | 0.00  | 0.00    | 0.00  |
| Callionymidae                      | Callionymidae sp.                           | 0.00   | 0.00   | 0.34     | 0.00   | 0.34     | 0.00   | 0.38    | 0.24  | 0.00      | 0.00  | 0.00    | 0.00  |
| Dragonets                          | Dragonet sp.                                |        |        |          |        |          |        |         |       |           |       |         |       |
| Carangidae<br>Iacks/Iack Mackerels | Trachurus novaezelandiae<br>Yellowtail Scad | 0.00   | 0.00   | 0.00     | 0.00   | 0.00     | 0.00   | 0.00    | 0.00  | 0.00      | 0.00  | 0.40    | 0.00  |
| Clinidae                           | Cristiceps sp.                              | 0.00   | 0.00   | 0.00     | 0.00   | 0.00     | 0.00   | 0.00    | 0.00  | 0.00      | 0.00  | 0.79    | 0.00  |
| Clinids                            | Weedfish sp.                                | 0.00   | 0.00   | 0.00     | 0.00   | 0.71     | 1 7 4  | 0.00    | 0.00  | 0.00      | 0.00  | 0.00    | 0.00  |
| Herrings/Sprats                    | Maray                                       | 0.00   | 0.00   | 0.00     | 0.00   | 0.71     | 1.24   | 0.00    | 0.00  | 0.00      | 0.00  | 0.00    | 0.00  |
|                                    | Herklotsichthys castelnaui                  | 0.00   | 0.00   | 0.00     | 4.88   | 0.00     | 0.00   | 0.00    | 0.00  | 0.00      | 0.00  | 0.00    | 0.00  |
|                                    | Southern Herring<br>Hyperlophus transucidus | 1.82   | 109.62 | 0.00     | 34.29  | 0.00     | 0.00   | 0.00    | 0.00  | 0.00      | 0.00  | 0.40    | 0.00  |
|                                    | Translucent Sprat                           |        |        |          |        |          |        |         |       |           |       |         |       |
|                                    | Hyperlophus vittatus<br>Sandy Sprat         | 8.33   | 5.26   | 7.06     | 0.34   | 16.15    | 42.96  | 67.92   | 58.51 | 116.68    | 40.50 | 7.34    | 0.79  |
|                                    | Sardinops sagax                             | 0.00   | 0.00   | 0.00     | 0.00   | 5.89     | 0.00   | 0.00    | 0.00  | 0.00      | 0.00  | 0.00    | 0.00  |
|                                    | Pilchard                                    | 0.00   | 0.00   | 0.00     | 0.00   | 0.00     | 2.24   | 0.00    | 0.00  | 0.00      | 0.00  | 1 5 7   | 0.00  |
|                                    | Blue Sprat                                  | 0.00   | 0.00   | 0.00     | 0.00   | 0.00     | 5.54   | 0.00    | 0.00  | 0.00      | 0.00  | 1.57    | 0.00  |
| Creediidae                         | Creedia haswelli                            | 0.00   | 0.00   | 0.00     | 0.00   | 0.00     | 0.00   | 0.00    | 0.00  | 0.37      | 0.00  | 0.00    | 0.00  |
| Sandburrower<br>Engraulidae        | Slender Sandburrower<br>Engraulis australis | 0.00   | 0.50   | 0.00     | 2.35   | 3.20     | 7.66   | 0.00    | 0.00  | 0.00      | 0.70  | 0.00    | 0.00  |
| Anchovies                          | Australian Anchovy                          |        |        |          |        |          |        |         |       |           |       |         |       |
| Gerreidae<br>Silver Biddies        | Gerres subfasciatus<br>Roach                | 30.54  | 0.00   | 6.74     | 7.02   | 12.78    | 0.82   | 27.76   | 10.77 | 8.33      | 5.77  | 12.54   | 1.52  |
| Gobiesocidae                       | Gobiesocidae sp.                            | 0.00   | 0.00   | 0.00     | 0.00   | 0.00     | 0.00   | 0.00    | 0.00  | 0.34      | 0.00  | 1.59    | 0.00  |
| Clingfish<br>Cobiidae              | Clingfishes sp.<br>Afurcagobius tamaronsis  | 0.00   | 0.00   | 0.00     | 0.00   | 0.00     | 0.00   | 0.20    | 0.26  | 0.00      | 0.00  | 0.00    | 0.00  |
| Gobies                             | Tamar Goby                                  | 0.00   | 0.00   | 0.00     | 0.00   | 0.00     | 0.00   | 0.59    | 0.50  | 0.00      | 0.00  | 0.00    | 0.00  |
|                                    | Arenigobius spp.                            | 9.67   | 48.63  | 10.20    | 18.65  | 43.89    | 74.49  | 3.06    | 8.25  | 16.50     | 1.12  | 8.09    | 2.50  |
|                                    | Half Bridled and<br>Bridled Goby            |        |        |          |        |          |        |         |       |           |       |         |       |
|                                    | Favonigobius lentiginosus                   | 2.96   | 0.00   | 4.84     | 0.33   | 1.13     | 0.41   | 0.39    | 5.06  | 3.95      | 7.19  | 36.41   | 0.96  |
|                                    | Long Finned Goby<br>Cobiidae sp             | 13.62  | 13.62  | 13 62    | 13.62  | 13.62    | 13.62  | 13.62   | 13.62 | 13.62     | 13.62 | 13.62   | 13.62 |
|                                    | Unidentified Goby sp.                       | 15.02  | 15.62  | 15.02    | 15.02  | 15.02    | 15.02  | 15.02   | 15.02 | 15.02     | 15.02 | 15.02   | 15.02 |
|                                    | Gobiopterus semivestita                     | 5.29   | 214.77 | 0.68     | 244.13 | 1.39     | 1.10   | 0.00    | 1.50  | 0.00      | 7.39  | 0.00    | 0.32  |
|                                    | Paedogobius kimurai                         | 0.78   | 1.55   | 26.29    | 477.99 | 37.67    | 164.78 | 0.35    | 0.00  | 0.34      | 0.00  | 0.79    | 4.18  |
| California                         | Wide Gape Paedomorphic Goby                 | 0.00   | 1.25   | 10.70    | 1.10   | 0.00     | 2.00   | 2.22    | 1 20  | 0.24      | 0.00  | 0.00    | 0.00  |
| Gobies                             | Eastern Bluespot Goby                       | 0.00   | 1.25   | 12.73    | 1.16   | 0.00     | 2.06   | 2.23    | 1.30  | 0.34      | 0.00  | 0.00    | 0.00  |
|                                    | Redigobius macrostoma                       | 3.60   | 4.31   | 0.00     | 1.74   | 1.06     | 4.56   | 5.42    | 9.45  | 13.18     | 22.01 | 0.00    | 0.53  |
|                                    | Large Mouth Goby<br>Schindleriidae sp.      | 2.51   | 0.00   | 0.00     | 0.00   | 0.00     | 0.00   | 0.00    | 0.00  | 0.00      | 0.00  | 0.00    | 0.00  |
|                                    | Schindler's Goby                            |        |        |          |        |          |        |         |       |           |       |         |       |
|                                    | Tridentiger trigonocephalus<br>Trident Coby | 0.00   | 0.00   | 0.00     | 0.00   | 1.68     | 3.24   | 0.00    | 0.00  | 0.00      | 0.00  | 0.00    | 0.00  |
| Gonostomatidae                     | Gonostomatidae sp.                          | 0.00   | 0.00   | 0.00     | 0.00   | 0.00     | 0.00   | 0.38    | 0.00  | 0.00      | 0.00  | 0.00    | 0.00  |
| Bristlemouths<br>Kyphosidae        | Bristlemouth sp.<br>Cirella tricuspidata    | 0.00   | 0.00   | 0.00     | 0.00   | 0.00     | 0.00   | 0.00    | 0.00  | 0.00      | 0.63  | 0.00    | 0.00  |
| Sea Chubs                          | Luderick                                    | 0.00   | 0.00   | 0.00     | 0.00   | 0.00     | 0.00   | 0.00    | 0.00  | 0.00      | 0.05  | 0.00    | 0.00  |
| Leptoscopidae                      | Lesueurina platycephala                     | 0.00   | 0.00   | 0.34     | 0.00   | 0.34     | 0.00   | 0.00    | 0.00  | 0.00      | 0.00  | 5.09    | 0.00  |
| Lutjanidae                         | Lutjanidae sp.                              | 0.00   | 0.00   | 0.34     | 0.00   | 0.00     | 0.00   | 0.00    | 0.00  | 0.00      | 0.00  | 0.00    | 0.00  |
| Snappers                           | Snapper sp.                                 | 0.00   | 1.00   | 0.00     | 1.00   | 0.00     | 0.00   | 0.00    | 0.00  | 0.00      | 0.00  | 0.00    | 0.00  |
| Leatherjackets                     | Fan Belly Leatherjacket                     | 0.00   | 1.68   | 0.00     | 1.09   | 0.00     | 0.00   | 0.00    | 0.00  | 0.00      | 0.00  | 0.00    | 0.00  |
| -                                  | Monacanthidae sp.                           | 0.00   | 0.00   | 0.34     | 0.00   | 0.00     | 0.00   | 0.38    | 0.00  | 0.00      | 0.00  | 0.40    | 0.00  |
| Monodactvlidae                     | Leatherjacket sp.<br>Monodactvlus argenteus | 0.00   | 0.00   | 0.00     | 0.00   | 0.00     | 0.00   | 0.00    | 0.36  | 0.00      | 0.27  | 1.98    | 0.00  |
| Moonfish                           | Diamondfish                                 | 0.00   | 5.00   | 5.00     | 5.00   | 5.00     | 5.00   | 5.00    | 5.50  | 5.00      | 5.27  | 1.50    | 0.00  |
|                                    | Schuettea scalaripinnis<br>Fastern Pomfret  | 0.00   | 0.00   | 0.00     | 0.00   | 0.00     | 0.00   | 0.76    | 0.00  | 0.00      | 0.00  | 0.00    | 0.00  |
| Mugilidae                          | Liza argentea                               | 0.00   | 0.00   | 0.00     | 0.00   | 0.35     | 0.42   | 0.00    | 0.00  | 0.00      | 0.00  | 0.00    | 0.00  |
| Mullets                            | Flat Tail Mullet                            | 0.00   | 0.00   | 0.00     | 0.00   | 0.00     | 0.00   | 0.00    | 0.00  | 1 20      | 0.00  | 0.00    | 0.00  |
| Weed Whitings/Cales                | Weed Whiting sp.                            | 0.00   | 0.00   | 0.00     | 0.00   | 0.00     | 0.00   | 0.00    | 0.00  | 1.38      | 0.00  | 0.00    | 0.00  |

(continued on next page)

### Appendix 1 (continued)

| Family                | Taxon                               | Botany | Bay    | Port Jac | kson   | Port Ke | mbla   | Clyde R | iver   | Jervis Ba | ау     | Port Ha | cking |
|-----------------------|-------------------------------------|--------|--------|----------|--------|---------|--------|---------|--------|-----------|--------|---------|-------|
|                       |                                     | Outer  | Inner  | Outer    | Inner  | Outer   | Inner  | Outer   | Inner  | Outer     | Inner  | Outer   | Inner |
| Paralichthyidae       | Pseudorhombus jenynsii              | 0.00   | 0.00   | 0.00     | 0.00   | 0.00    | 0.00   | 1.83    | 0.94   | 0.00      | 0.27   | 0.00    | 0.47  |
| Large Tooth Flounders | Small Tooth Flounder                |        |        |          |        |         |        |         |        |           |        |         |       |
| Pempheridae           | Pempheridae sp.                     | 0.42   | 0.00   | 0.00     | 0.00   | 0.00    | 0.00   | 0.00    | 0.00   | 0.37      | 0.00   | 0.00    | 0.00  |
| Sweepers/Bullseyes    | Bullseye sp.                        |        |        |          |        |         |        |         |        |           |        |         |       |
| Platycephalidae       | Platycephalus fuscus                | 0.00   | 0.00   | 0.00     | 0.00   | 0.00    | 0.35   | 1.14    | 2.20   | 1.60      | 0.00   | 2.29    | 0.00  |
| Flatheads             | Dusky Flathead                      |        |        |          |        |         |        |         |        |           |        |         |       |
|                       | Platycephalus sp.                   | 0.00   | 0.00   | 0.87     | 0.00   | 0.00    | 0.00   | 0.00    | 0.00   | 0.00      | 0.00   | 0.00    | 0.00  |
|                       | Flathead sp.                        |        |        |          |        |         |        |         |        |           |        |         |       |
| Sciaenidae            | Argyrosomus japonicus               | 0.00   | 0.00   | 0.00     | 0.00   | 0.35    | 0.00   | 0.76    | 0.00   | 0.00      | 0.00   | 0.00    | 0.00  |
| Drums and Croakers    | Mulloway                            |        |        |          |        |         |        |         |        |           |        |         |       |
|                       | Atractoscion aequidens              | 0.00   | 0.00   | 0.00     | 0.00   | 0.35    | 0.00   | 0.00    | 0.00   | 0.00      | 0.00   | 0.00    | 0.00  |
|                       | Teraglin                            |        |        |          |        |         |        |         |        |           |        |         |       |
| Silliginidae          | Sillago ciliata                     | 5.45   | 13.44  | 1.01     | 0.43   | 2.22    | 0.00   | 4.93    | 0.47   | 35.50     | 10.80  | 3.61    | 1.46  |
| Whitings              | Sand Whiting                        |        |        |          |        |         |        |         |        |           |        |         |       |
|                       | Sillago flindersai                  | 17.99  | 1.00   | 2.82     | 0.00   | 0.00    | 1.20   | 1.14    | 0.00   | 2.27      | 0.00   | 3.90    | 0.00  |
|                       | Eastern School Whiting              | 1.00   | 0445   | 0.00     | 0.40   |         | 0.44   | 4.40    | 0.04   | 1.00      | 10.01  | 4 5 5   | 0.00  |
|                       | Sillago maculata                    | 1.09   | 34.15  | 0.00     | 8.46   | 1.41    | 0.41   | 4.12    | 0.24   | 1.68      | 12.61  | 1.57    | 0.32  |
| 6-1-11-               | Trumpeter Whiting                   | 0.00   | 0.00   | 0.00     | 0.00   | 0.00    | 0.00   | 0.00    | 0.20   | 0.27      | 0.00   | 1.00    | 0.50  |
|                       | Soleidae sp.                        | 0.00   | 0.00   | 0.00     | 0.00   | 0.00    | 0.00   | 0.00    | 0.36   | 0.37      | 0.00   | 1.98    | 0.53  |
| Sparidao              | Sole sp.<br>Acanthonagrus australis | 0.00   | 0.00   | 0.00     | 0.00   | 0.00    | 0.00   | 0.20    | 0.00   | 2 20      | 4 5 0  | 0.00    | 0.00  |
| Soa Proams            | Vallowfin Program                   | 0.00   | 0.00   | 0.00     | 0.00   | 0.00    | 0.00   | 0.59    | 0.00   | 5.20      | 4.55   | 0.00    | 0.00  |
| Sphyraenidae          | Sphyraena sp                        | 0.00   | 0.00   | 0.00     | 0.00   | 0.00    | 0.00   | 034     | 0.00   | 0.00      | 0.00   | 0.00    | 0.00  |
| Barracudas            | Barracuda sn                        | 0.00   | 0.00   | 0.00     | 0.00   | 0.00    | 0.00   | 0.54    | 0.00   | 0.00      | 0.00   | 0.00    | 0.00  |
| Syngnathidae          | Stigmatonora nigra                  | 0.00   | 0.00   | 0.00     | 0.00   | 0.00    | 0.51   | 0.00    | 0.83   | 0.40      | 0.27   | 3 49    | 0.00  |
| Pinefish/Seahorses    | Wide Rodied Pinefish                | 0.00   | 0.00   | 0.00     | 0.00   | 0.00    | 0.51   | 0.00    | 0.05   | 0.40      | 0.27   | 5.45    | 0.00  |
| Tipejishjseunorses    | Urocampus carinirostris             | 0.79   | 0.00   | 0.00     | 0.00   | 0.00    | 0.00   | 0.38    | 0.47   | 0.00      | 0.96   | 1.96    | 0.00  |
|                       | Hairv Pipefish                      |        |        |          |        |         |        |         |        |           |        |         |       |
|                       | Vanacampus margaritifer             | 0.00   | 0.00   | 0.00     | 0.00   | 0.00    | 0.00   | 0.00    | 0.00   | 0.00      | 0.00   | 0.40    | 0.00  |
|                       | Mother of Pearl Pipefish            |        |        |          |        |         |        |         |        |           |        |         |       |
| Synodontidae          | Synodontidae sp.                    | 0.00   | 0.00   | 0.00     | 0.00   | 0.00    | 0.00   | 0.00    | 0.45   | 0.00      | 0.00   | 0.00    | 0.00  |
| Lizardfishes          | Lizardfish sp.                      |        |        |          |        |         |        |         |        |           |        |         |       |
| Terapontidae          | Pelates sexlineatus                 | 0.00   | 0.00   | 0.00     | 0.00   | 0.00    | 0.00   | 0.00    | 0.00   | 0.00      | 0.36   | 1.57    | 0.47  |
| Grunter Perch         | Six Lined Trumpeter                 |        |        |          |        |         |        |         |        |           |        |         |       |
| Tetraodontidae        | Tetraodontidae sp.                  | 0.00   | 0.00   | 0.00     | 0.00   | 0.00    | 0.00   | 0.00    | 0.00   | 1.60      | 0.00   | 0.00    | 0.00  |
| Pufferfish            | Toadfish sp.                        |        |        |          |        |         |        |         |        |           |        |         |       |
| Tetrarogidae          | Centropogon australis               | 0.00   | 3.18   | 1.01     | 0.00   | 0.34    | 0.00   | 0.39    | 0.45   | 1.60      | 1.26   | 1.59    | 0.00  |
| Waspfish              | Fortesque                           |        |        |          |        |         |        |         |        |           |        |         |       |
| Tripterygiidae        | Tripterygiidae sp.                  | 28.05  | 0.00   | 1.04     | 4.58   | 1.33    | 0.00   | 0.38    | 0.00   | 2.97      | 1.84   | 1.93    | 0.00  |
| Triplefin Blennies    | Triplefin Blenny sp.                |        |        |          |        |         |        |         |        |           |        |         |       |
| Total Abundance       |                                     | 169.70 | 460.31 | 104.91   | 831.40 | 153.17  | 326.95 | 160.77  | 128.57 | 816.66    | 170.60 | 163.16  | 34.96 |

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