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Abundance and distribution of the non-indigenous *Acentrogobius pflaumii* and native gobiids in a temperate Australian estuary

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Abstract

Non-indigenous species are recognised as a significant threat to estuaries as they can negatively impact native fauna and ecosystem functioning. This study compared the abundance and distribution of the introduced gobiid, *Acentrogobius pflaumii*, with native gobiids in the shallow, nearshore and deeper, offshore waters of the Swan-Canning Estuary over 31 consecutive months and determined the influence of salinity and water temperature on the gobiid assemblage. Eight gobiid species were recorded (six native and two non-indigenous). Each species was present in offshore waters, with five also occurring in nearshore waters. The contribution of *A. pflaumii* to the gobiid fauna varied markedly between depths; not being recorded in the 1,240 samples from nearshore waters, but representing almost half of all gobiids caught in offshore waters. The percentage contribution of this species to the total gobiid abundance declined with distance from the estuary mouth, from 96% in the most downstream region to only 0.4% in the uppermost. The overwhelming contribution of *A. pflaumii* in the lower estuary, which historically contained few gobiids, indicate that it has successfully exploited an unoccupied niche. Gobiid composition, in both depths, differed mainly among regions, with species partitioning themselves along the longitudinal axis of the estuary. In offshore waters, *Favonigobius lateralis* and *A. pflaumii* dominated the lower regions and *Arenigobius bifrenatus*, *Favonigobius punctatus* and *Pseudogobius olorum* the regions further upstream. The distributions of these species are influenced by salinity and, to a lesser extent, temperature. While there is spatial overlap between *A. pflaumii* and native gobiids species, its preference for deeper, more saline waters may limit the impact of this non-indigenous species.

Introduction

Non-indigenous, or introduced, species are those that have been translocated to environments outside of their natural distribution, typically due to anthropogenic activities (Williams and Grosholz 2008; Ojaveer et al. 2014). Such species are recognised as a pervasive and significant threat to aquatic habitats globally (Carlton 1999; Thresher 1999; Bax et al. 2003; Molnar et al. 2008). Estuaries and coastal waters can harbour large numbers of non-indigenous species, given their subjection to human colonisation and the resulting anthropogenic modifications, such as the construction of ports/canals and their associated shipping activities and aquaculture. For example, Port Phillip Bay, Australia contains 99 introduced and 61 cryptogenic species, which together represent 13% of all species in that system, and in San Francisco Bay, USA, these numbers are double those in Port Phillip Bay, i.e. 234 introduced and 125 cryptogenic species (Cohen and Carlton 1998; Hewitt et al. 2004). On a larger spatial scale, nearly 1,000 non-indigenous species have been recorded in the Mediterranean Sea (Katsanevakis et al. 2014a). Moreover, rates of new colonisations of such species have accelerated in recent decades due to rapid globalization and increasing volumes of trade, travel and transport (Hulme 2009; Katsanevakis et al. 2016).

Once established, non-indigenous species can cause a number of deleterious impacts, ranging from single species interactions and reduction in individual fitness of a population of a co-occurring native species to population declines, local extinctions and changes in community composition. These species can also have effects on entire ecosystem processes and wider ecosystem function, e.g. fishery productivity, the extent and quality of nursery habitats and declines in the provision of ecosystem services, e.g. filtering and detoxification (Worm et al. 2006; Blackburn et al. 2014; Katsanevakis et al. 2014b). Although temperate estuaries are already recognised as the most degraded of all aquatic habitats (Jackson et al. 2001; Warwick et al. 2018), non-indigenous species can further reduce the ability of management agencies to effectively maintain the health and sustainability of these important ecosystems (Simberloff 2003; Crowder and Norse 2008).

Globally, members of the Gobiidae have successfully colonised non-natal systems more frequently than any other fish family; due to their broad physiological tolerances,

generalised feeding habits and flexible reproductive strategies(Wonham et al. 2000). Examples of invasions by non-indigenous gobiids include *Acanthogobius flavimanus* in New South Wales, Australia and California, USA (Middleton 1982; Workman and Merz 2007), *Arenigobius bifrenatus* in New Zealand (Willis et al. 1999) and *Proterorhinus marmoratus* and *Neogobius melanostomus* in the North American Great Lakes, as well as numerous large rivers in Europe, e.g. the Scheldt, Danube and Rhine systems by *N. melanostomus* (Jude et al. 1992).

One such example of a non-indigenous gobiid is the Striped Sandgoby *Acentrogobius pflaumii*, which can grow to 109 mm total length, live for ~4 years and attains sexual maturity within the first year of life (Baeck et al. 2004; Hogan-West et al. submitted; Wang et al. 2015). This gobiid inhabits soft substrates, sand and seagrass beds in estuaries and coastal embayments in China, Korea, Japan, Taiwan and the Philippines (Masuda et al. 1984; Shao et al. 1993; Kanou et al. 2004; Nan et al. 2009; Horinouchi and Sano 2000; Horinouchi 2008). It has also been recorded in Australia in Port Phillip Bay (Melbourne), Botany Bay (Sydney) and Morton Bay (Brisbane) and in New Zealand in the Waitemata and Whangapoua Harbours (Auckland), with ballast water discharge the suspected vector in all invasions (2001, 1999; Francis et al. 2003; Gomon et al. 2008). *Acanthogobius pflaumii* was first recorded in Port Phillip Bay in 1996, but only a year later was one of the most abundant species and the most widely distributed of all introduced fishes in that bay (Lockett and Gomon 2001). It was first recorded in Western Australia in 2004, in Cockburn Sound, a marine embayment near Perth (Mead-Hunter 2005) and found in the Swan-Canning Estuary (~20 km to the north) later that same year (Maddern and Morrison 2009). A number of concerns have been raised around *A. pflaumii* including: competition for habitats and dietary resources and hybridisation with native gobiids (Kanou et al. 2004; Maddern and Morrison 2009; Rowe et al. 2008). In a review of the impacts of *A. pflaumii* and another non-indigenous gobiid (*Acanthogobius flavimanus*) in Australia, Rowe et al. (2008) stated that studies to identify their potential impacts are urgently required and that these studies should initially compare the distribution and abundance of these non-native species to other small benthic fishes.

Together with atherinids, gobiids dominate the shallow, nearshore waters of the microtidal estuaries of south-western Australia, such as the Swan-Canning Estuary (Valesini

et al. 2014; Potter et al. 2016; Veale et al. 2014). Typically, these estuaries are separated into a number of distinct regions on the basis of morphology, water quality and sediment conditions (Gill and Potter 1993; Potter and Hyndes 1999; Loneragan et al. 1989). Differences in environmental conditions are mirrored by corresponding changes in ichthyofaunal composition, particularly of atherinids and gobiids (Prince et al. 1982; Gill and Potter 1993; Potter et al. 2015b). The spatial segregation of members of these two families reflects their life-cycle charcteristics, with marine-spawning species (e.g. marine stragglers) occurring only in the lower reaches of the estuary, while those species capable of spawning in the estuary (e.g. soley estuarine species) are largely restricted to the more upstream regions (Potter et al. 2015a; Loneragan et al. 1989). Furthermore, it also reflects differences in water phyisco-chemistry, sediment composition and the presence of macrophyte beds (Gill and Potter 1993; Prince et al. 1982; Humphries et al. 1992). Although the spatial distributions of confamilial species overlap, in these cases, the diet of these species differs when they co-occur, presumably in an effort to partition resources and reduce competition (Humphries and Potter 1993; Prince et al. 1982; Gill and Potter 1993; Poh et al. 2018).

Given the rapid proliferation of *A. pflaumii* in Port Phillip Bay and the marked spatial pattern of distribution exhibited by native species in south-western Australian estuaries, the objectives of the current study were to: (i) compare the patterns of abundance and distribution of *A. pflaumii* with native goby species in the shallow, nearshore and deeper, offshore waters of the Swan-Canning Estuary, and (ii) determine if there is a significant relationship between the abundance of key goby species and gobiid composition to salinity and water temperature. As *A. pflaumii* is thought to have colonised the Swan-Canning Estuary from Cockburn Sound and is found in other marine embayments in Australia, it is hypothesised that this species will be most abundant in the lower reaches of the estuary and exhibit a positive relationship with salinity. Patterns of abundance of native gobiid species in nearshore waters are also broadly compared with those recorded thirty years earlier in the same system by Gill and Potter (1993).

Materials and methods

Study site

The Swan-Canning Estuary, located in south-western Australia, is ~50 km long, has a surface area of ~55 km^2 and remains permanently-open to the Indian Ocean (Fig. 1). The estuary comprises a narrow entrance channel that opens into two basins (Melville and Perth Waters) and the tidal portions of the Swan and Canning Rivers, which extend ~ 29 and 13 km upstream from their entry points in to Melville Water, respectively. Although more than half of the estuary is shallow, i.e. < 2 m in depth, it reaches a maximum depth of ~ 20 m in the entrance channel (Fig. 1). Sediment composition differs markedly between the shallow, nearshore (< 2 m deep) and the deeper, offshore waters (> 2 m deep). Nearshore substrates contain a greater proportion of medium sand than the offshore sediments, which comprise higher percentages of particulate organic matter and finer inorganic particles (silt and fine sand; Tweedley et al. 2017). Regardless of water depth, the substrates of the downstream reaches contain the greatest proportions of fine sand, which is likely transported in from the nearshore marine environment (Wildsmith et al. 2005). The proportion of larger grain sizes was greater in upstream areas, as these terrestrially derived particles were likely deposited first as water velocity decreased progressively downstream (Gill and Potter 1993). The seagrass *Zostera muelleri* is present in the entrance channel, *Halophila ovalis* forms extensive beds in shallow parts of Melville Water and *Ruppia megacarpa* has also been recorded (Valesini et al. 2009; Department of Water 2014). Light penetration, which is reduced by tannins and particulate matter, generally limit these seagrass species to water depths < 2 m, except in the entrance channel, where they can grow in waters up to 4 m deep (Valesini et al. 2009; Department of Water 2014).

South-western Australia experiences a Mediterranean climate (Hallett et al. 2018), with hot, dry summers and cool, wet winters with ~80% of annual rainfall occurring between June and September (Hodgkin and Hesp 1998). This highly seasonal rainfall pattern, combined with the microtidal tidal regime (tidal range = ~0.6-0.8 m) results in marked seasonal variations in physical-chemical conditions of the water column in this salt-wedge estuary (J.R. Tweedley et al. 2016). Salinities are typically stable and relatively high throughout much of the estuary during the austral summer (December to February), but during winter, may vary markedly

along the estuary following substantial freshwater discharge (Crisp et al. 2018), leading to marked stratification of the water column and occasionally hypoxia in the upper most reaches (J.R. Tweedley et al. 2016).

Sampling regime

Data on the abundance of *Acentrogobius pflaumii* and other gobiid species were obtained from an established sampling regime designed to determine the biology and spatial and temporal distribution of the Western School Prawn (*Metapenaeus dalli*) in the Swan-Canning Estuary (Poh et al. 2019). The sampling methodology was identical to that described in Broadley et al. (2017), except that it was conducted every lunar month between October 2013 and March 2016, spanning two and a half years. In brief, sampling was conducted at 20 shallow, nearshore sites (< 2 m deep) and at 16 deeper, offshore sites (2-16 m deep) spread across five and four regions, respectively (Fig. 1). Two replicate trawls were conducted at night at each site over three days of the new moon phase of each lunar month. Nearshore sites were sampled using a hand trawl net, pulled by two people, that was 4 m wide and constructed from 9 mm mesh and dragged for 200 m. The net was, on average, 2.85 m wide during trawling, so each trawl covered an area of ~570 m^2. Offshore waters were sampled using an otter trawl, which was 2.6 m wide (4.4 m headrope width) and constructed from 25 mm mesh in the body, with a 9 mm cod end. The net was towed by boat for five minutes at a speed of ~1.6 knots, covering a distance of ~250 m and thus an area of ~650 m^2. Due to a boat engine malfunction in December 2014, catch data were not obtained from all offshore sites and thus data from this month were removed from all analyses. The mesh size of the otter trawl was successful in capturing gobies in the same estuary as small as 36 mm total length in previous studies of the fish fauna (Loneragan et al., 1989).

The catch of fish and prawns was sorted at the end of each hand or otter trawl, and each individual gobiid was identified to species and counted. All native species were returned to the water as quickly as possible, whereas all non-indigenous species were euthanised in an ice slurry, as per the conditions in Murdoch University Animal Ethics Committee permit #RW2566. Specimens of *A. pflaumii* were retained for detailed biological analyses to

determine size composition, age, growth and reproduction (Hogan-West et al., submitted). A total of 1,240 hand trawls and 960 otter trawls were conducted over the course of the study. Salinity, water temperature and dissolved oxygen concentration at the surface and bottom of the water column were recorded at each offshore site, and in the surface waters of nearshore sites in the Upper Canning Estuary, on each sampling occasion, using a Yellow Springs International Model 556 water quality meter.

Each gobiid species was assigned to one of the following life cycle categories, i.e. marine, estuarine, freshwater and diadromous, and their component guilds for fishes in estuaries (Potter et al., 2015). Among gobiids in south-western Australia, the marine category is represented by stragglers (MS), i.e. species typically entering estuaries irregularly and in low numbers. The estuarine category comprises species that complete their life cycle in estuaries, and are either essentially found only in estuaries (E), or are also represented by populations in marine waters (EM) or freshwater (EF). No freshwater or diadromous gobiids were recorded.

Statistical analyses
Univariate analysis of gobiid richness and diversity

The abundance of each gobiid species in each replicate sample was converted to a density (individuals 500 m^{-2}), and the gobiid richness (i.e. number of goby species) and total density of gobiids calculated using the DIVERSE routine in PRIMER v7 (Clarke and Gorley 2015). The data for each of the two dependent variables (*i.e.* gobiid richness and gobiid density) were examined separately in nearshore and offshore waters, as was the density of *A. pflaumii* in offshore waters, to determine whether transformations were required to meet the assumptions of homogenous dispersion among *a priori* groups. This was achieved by calculating the extent of the linear relationship (slope) between the $\log_e$ (mean) and $\log_e$ (standard deviation) of each variable among all groups and comparing them to the criteria in Clarke and Warwick (2001). These analyses demonstrated that gobiid density in nearshore waters required a fourth-root transformation and that gobiid density and that of *A. pflaumii* in offshore waters needed a square-root transformation.

The transformed data for each variable in each water depth were used to construct separate Euclidean distance matrices and subjected to a two-way Permutational Analysis of Variance (PERMANOVA; Anderson et al. 2008) tests. These analyses aimed to determine whether the values for each variable differed amongst Region (four or five levels: Lower Melville Water, Upper Melville Water, Middle Swan Estuary, Lower Canning Estuary and Upper Canning Estuary) and Lunar month (31 sampling occasions between October 2013 and March 2016). The null hypothesis of no significant differences among *a priori* groups was rejected if the significance level (*P*) was < 0.05, and the relative influence of each term in the model was quantified using the percentage contribution of the mean squares of that term to the total mean squares.

Multivariate analyses of gobiid composition

The abundance of each gobiid species recorded in each replicate sample was subjected to dispersion weighting to down-weight the effects of those species whose numbers exhibited erratic differences among replicate samples due to schooling (Clarke et al. 2006). The data were then square-root transformed to down-weight the contributions of species with consistently high values (across replicates within each Region × Lunar month combination) in relation to those with consistently low values. The resultant pre-treated data were used to construct a Bray-Curtis resemblance matrix, which was subjected to the same two-way PERMANOVA design described above, although now using multivariate data. Pairwise tests for selected influential terms in the model were also conducted using a pairwise PERMANOVA test.

As Region was identified as by far the most influential term in the model (see Results), the extent of the differences between individual regions detected by PERMANOVA in each water depth and the species responsible were explored visually using non-metric Multi-dimensional Scaling (nMDS) and shade plots (Clarke et al. 2014; Clarke and Gorley 2015). Each of the two Bray-Curtis resemblance matrices, derived from the replicate samples in a water depth, were subjected to the Bootstrap Averages Routine (Clarke and Gorley 2015). The averages of repeated bootstrap samples (bootstrapped averages) for each region were used to

construct an nMDS ordination plot. Superimposed on the plot was i) a point representing the group average (i.e. the average of the bootstrapped averages) and ii) the associated, smoothed and marginally bias-corrected 95% bootstrap region, in which 95% of the bootstrapped averages fall.

The transformed abundance of each species in each sample from each water depth separately were averaged for each region and used to produce a shade plot. The resultant plot is a simple visualisation of the frequency matrix, where a white space for a species demonstrates that the gobiid species was never collected, while the intensity of grey-scale shading is linearly proportional to the abundance of that species (Clarke et al. 2014). The order of the both the gobiid species (*y* axis) and regions (*x* axis) were determined by independent seriation. Thus, species exhibiting similar patterns of abundance across the regions were ordered together and *vice versa* for regions. These re-orderings are purely to aid visualisation, and are irrelevant to multivariate analyses of those samples (Tweedley et al. 2015).

Relationships between gobiids and salinity and water temperature

Pearson's correlations were used to determine whether trends in salinity and water temperature were significantly related ($P < 0.05$) to the spatial pattern of abundance of key gobiid species (*i.e.* those that represented >10% of the total number of gobiids). The density of each gobiid species and surface salinity and water temperature in the nearshore waters was averaged across lunar months to provide a single value for each of the 20 sites. The same approach was undertaken for the 16 offshore sites using only salinity and temperature data from the bottom rather than surface of the water column. Draftsman plots of the values for each pair of variables (biotic and environmental) at each site, in each water depth, were examined visually to assess whether the values for each variable were heavily skewed and, if so, which type of transformation would satisfy the assumption of homogeneity of variances. These plots demonstrated that each of the biotic variables required a square-root transformation. These pre-treated data were then subjected to two-tailed Pearson's correlations in SPSS 24. Note that depth was not investigated as a potential influencing factor for the abundance of each key gobiid species, as depth can vary widely within a region (Fig. 1) and both surface and bottom

salinity were strongly positively correlated with depth of the sites in offshore waters ($r = 0.968$; $P = < 0.001$ and $r = 0.982$; $P = <0.001$, for surface and bottom salinity respectively).

The BEST test (Clarke and Gorley, 2015) was used to determine whether gobiid composition in each water depth was related to salinity and/or water temperature ($P < 0.05$). The faunal data were dispersion-weighted and square-root transformed, while the two environmental variables were normalized to place them on a common scale. The pre-treated gobiid composition data for each site were used to construct a Bray-Curtis resemblance matrix, which was subjected to nMDS ordination. The mean salinity and water temperature at that site, represented by bubbles of different sizes, were then superimposed on the nMDS plot.

Results

Environmental conditions

Surface salinity ranged from 1 in the Upper Canning Estuary during October 2014 to 38.4 in that same region in March 2014 (Fig. 2a). In all lunar months except October 2013 (13), salinities in Lower Melville Water were > 20, whereas in all other regions they declined to ≤ 10. The ranges of surface salinity varied markedly among regions, from 16 in Lower Melville Water to as high as 36 in the Upper Canning Estuary. Differences in salinities among regions were smallest between January and April (< 5), but reached ~ 25 in May and June 2014.

The lowest bottom salinity was 9.7 in the Middle Swan Estuary in October 2013, while the highest was 37.7 in Lower Melville Water during March 2014 (Fig. 2b). Salinities in the bottom waters ranged far less than the corresponding surface waters, e.g. bottom salinities in Lower Melville Water differed by 7 compared with 16 in the surface waters over the two years (Figs 2a,b).

Water temperature in each of the four regions in the Swan-Canning Estuary underwent a pronounced sinusoidal pattern, with values in surface waters typically ranging from a minimum of ~15 °C in June/July to a maximum of ~26 °C in January/February (Fig. 2c). Seasonal differences were greatest in the Upper Canning Estuary and lowest in Upper and Lower Melville Water and the Lower Canning Estuary. Surface water temperatures were mostly > 20 °C between October and April and < 20 °C during May-September (Fig. 2c).

Temporal patterns in bottom water temperature mirrored those in the surface waters, albeit the values from offshore waters were typically greater than those in the corresponding nearshore waters between May and September, whereas the reverse was true between October and March (cf Fig. 2c,d).

Mean dissolved oxygen concentrations in the surface waters in each lunar month exceeded 6.5 mg L^{-1} (average 7.2 mg L^{-1}) and ranged from 4.6 to 13.7 mg L^{-1}. Although concentrations were lower in the corresponding bottom waters, typically they were > 4 mg L^{-1} (average 5.2 mg L^{-1}) and thus well above the threshold for hypoxia of 2 mg L^{-1}. Mean oxygen concentrations also varied little among sites, particularly in the surface waters, i.e. 6.6 to 7.6 mg L^{-1}, with those at the bottom of the water coloum ranging from 3.2 to 6.3 mg L^{-1}, but were typically > 4.5 mg L^{-1}.

Contribution of gobiids in nearshore and offshore waters

A total of 319,636 fish, representing 45 species, were recorded at the 20 sites sampled in the nearshore waters of the Swan-Canning Estuary every lunar month between October 2013 and March 2016. Members of the Gobiidae were represented by five species and constituted ~ 2% of the total numbers of fish caught. Although at least three of the five gobiid species were present in each of the five regions, the mean density of gobiids differed markedly. Thus, more than 5 individuals 500 m^{-2} were recorded in the Lower and Upper Canning Estuary and Middle Swan Estuary, respectively, but only 1.1 and 0.3 individuals 500 m^{-2} were caught at the sites in Lower and Upper Melville Water, respectively (Table 1a). The most abundant gobiid species recorded in nearshore waters were *Arenigobius bifrenatus* and *Favonigobius punctatus*, comprising 46 and 34% of the total number of gobies respectively, with *Pseudogobius olorum* also making a significant contribution to the gobiid fauna (14%; Table 1a). Despite conducting 1,240 hand trawls in nearshore waters, no *Acentrogobius pflaumii* or any other non-indigenous gobies were recorded in these shallow areas of the Swan-Canning Estuary.

The percentage contributions of gobiid species differed considerably amongst the regions in nearshore waters. For example, *Favonigobius lateralis* comprised 90% of the gobiid fauna of Lower Melville Water, but declined in abundance in an upstream direction,

representing only 1.2% of the gobies in the Upper Melville Water and was not recorded in the Middle Swan Estuary or Upper Canning Estuary (Table 1a). In contrast, *A. bifrenatus* was by far the most abundant goby in the Upper and Lower Canning estuaries, representing 70 and 62% of all the gobies in these two regions, respectively, yet it made a relatively low contribution (< 10%) in the Middle Swan Estuary and was largely absent in Melville Water. *Favonigobius punctatus* was the dominant goby species in Upper Melville Water and the Middle Swan Estuary, representing 89 and 61% of all gobies, respectively, in those two regions (Table 1a).

A total of 22,682 fish from 47 species were recorded at the 16 sites in offshore waters, of which ~10% of the individuals and ~17% of the species (8) were gobiids. As in nearshore waters, the total number of gobiid species was relatively consistent among all regions, ranging between six and eight, except the Middle Swan (4 species), but their total density differed markedly among regions (Table 1b). Densities were greatest in Lower Melville Water (~ 3 individuals 500 m^{-2}) and least in Upper Melville Water (< 1 individuals 500 m^{-2}) with intermediate densities (~ 1.5-2 individuals 500 m^{-2}) in the other two regions (Table 1b).

The gobiid fauna in offshore waters was dominated by three species that collectively contributed 96% to the total number of gobies, namely *A. pflaumii* (47%), *A. bifrenatus* (29%) and *P. olorum* (20%), with none of the remaining five species representing > 2% of the total catch (Table 1b). The most abundant gobiid overall was *A. pflaumiii,* particularly in Lower Melville Water, where it represented 96% of all gobiids (Table 1b). The contribution of this species to the total fauna declined progressively in an upstream direction to 28, 18 and 0.4% of the individuals in Upper Melville Water, Lower Canning Estuary and Middle Swan Estuary, respectively. Instead, those three regions, and particularly the latter two, were dominated by *A. bifrenatus* and *P. olorum* (Table 1b). The remaining five species, including another non-indigenous species, *Tridentiger trigonocephalus,* were found in between one and four of the regions, but cumulatively, made relatively small contributions to the total gobiid fauna in all cases, i.e. < 10% and usually < 5% (Table 1b).

In total, more gobiid species were recorded in offshore than nearshore waters, with all five of the species found in shallower waters also being recorded in deeper waters. Three of

the species were recorded only in offshore waters, namely *A. pflaumii*, *T. trigonocephalus* and *Callogobius depressus*, although only *A. pflaumii* was caught in substantial numbers. The total density of gobiids in the offshore waters of all regions, except Lower Melville Water, was half of that in the corresponding nearshore waters (Table 1). Among the species that occurred in both water depths, the densities of all species, and particularly *F. punctatus* and *F. lateralis*, were greater in nearshore than offshore waters (Table 1).

Spatial distribution of **Acentrogobius pflaumii**

Over the study, *A. pflaumii* was recorded at 12 of the 16 offshore sites. Mean abundances were greatest at the most downstream sites in Lower Melville Water (> 5.5 *A. pflaumii* 500 m^{-2}), with substantial numbers recorded at the other two sites in this region (~ 2 *A. pflaumii* 500 m^{-2}; Fig. 3). Abundances declined in an upstream direction, with densities of < 0.5 recorded at all bar one site in each of the Lower Canning Estuary and Upper Melville Water. With the exception of one site in Upper Melville Water, *A. pflaumii* was recorded at each of the sites in these regions, albeit in low densities, but in only one of the four sites in the most upstream region (Middle Swan Estuary; Fig. 3).

Gobiid richness and density

The mean number of goby species and density of gobiids in nearshore waters of the Swan-Canning Estuary differed significantly among regions, lunar months and the Region × Month interaction was also significant (Table 2a, b). The percentage mean squares associated with Region, however, accounted for $\sim 90\%$ of the total mean squares for both gobiid richness and density, with the corresponding values for Lunar month and the interaction being only ~ 6 and $\sim 3\%$, respectively. Gobiid richness was the greatest in the two most upstream regions (Middle Swan and Upper Canning estuaries; ~ 0.9), lowest in Upper Melville Water (0.1) with intermediate values (~ 0.4) in the remaining regions (Fig. 4a). Mean densities of gobies in the two Melville Water regions were considerably lower than in the three more upstream regions (i.e. < 1.5 vs 5.5 to 10.7 individuals 500 m^{-2}, respectively; Fig. 4b).

Significant differences in gobiid richness and density were detected in offshore waters with all main effects and their interactions being significant (Table 2c, d). In both cases, Region was the most influential factor (> 50% of the total mean squares), followed by Lunar Month (~30%) and lastly the interaction (~11%). The mean number of goby species in Upper and Lower Melville waters (~ 0.4) was significantly less than in the Lower Canning and Middle Swan estuaries (~ 0.6; Fig. 5a). Trends for richness were less marked among lunar months than those for regions, but richness increased in a relatively sequential manner from October 2013 (0.28) until November 2014 (1.19), then declined to low of 0.19 in Feb 2015. However, richness did not follow this pattern in 2015/16 when it was greatest in July/August (0.6) and least in December 2015 (0.01, Fig. 5b). Mean densities of gobiids in the offshore waters of Lower and Upper Melville Waters were significantly greater and lesser, respectively than in all other regions, with the Middle Swan and Lower Canning estuaries having similar values to each other (Fig. 5c). Densities were highest during the austral winter, peaking in August of both 2014 and 2015, albeit to a lesser extent in the latter year (Fig. 5d).

Although the densities of *A. pflaumii* differed among regions, lunar months and their interaction was significant, Region was the most influential of these terms, accounting for 85% of the total mean squares, with Lunar month and the Lunar month × Region interaction contributing only 8 and 6%, respectively (Table 2e). *Acentrogobius pflaumii* was far more abundant in Lower Melville Water than in any other region, particularly the Middle Swan Estuary, where very low densities were recorded (Fig. 5e). Trends in the abundance of *A. pflaumii* were similar to those of total gobiid density (Figs 5d, f).

Gobiid faunal composition

Two-way PERMANOVA demonstrated that gobiid faunal composition in nearshore waters differed significantly among regions, lunar months and the interaction term was significant (Table 3a). Region accounted for 89% of the total mean squares, with only minor contributions made by Lunar month and the one-way interaction, *i.e.* 6 and 4%, respectively. Pairwise PERMANOVA demonstrated that gobiid composition differed significantly between each of the ten combinations of regions (Table 3b). Among the comparisons, the largest

differences were detected from regions at opposite ends of the estuary (i.e. Lower and Upper Melville Waters vs Upper Canning and Middle Swan estuaries) and the lowest between adjacent regions (i.e. Upper Canning vs Middle Swan estuaries; Table 3b). This is illustrated on the nMDS plot where there is a progression from left to right on the ordination from most downstream to most upstream (Figs 6a). The gobiid fauna of Lower Melville Water was dominated by *F. lateralis,* with relatively low densities of *F. punctatus* and *P. olorum* (Fig. 6b). These latter two species were the two main contributors to the depauperate gobiid fauna of Upper Melville Water with their densities increasing progressively upstream, being greatest in the Upper Canning and Middle Swan estuaries. Four of the five goby species (i.e. all except *C. mucosus*) were present in the Lower Canning Estuary, all in moderate abundances. There were no clear trends in the abundance of gobiid species among lunar months (data not shown).

The composition of the gobiid fauna in offshore waters differed significantly between both main effects and their interaction was also significant, but Region explained 77% of the total mean squares, followed by Lunar months (13%) and the interaction (6%; Table 3c). As in the nearshore waters, each region contained a statistically distinct gobiid composition, with the most pronounced differences in pairwise comparisons involving Lower Melville Water (Table 3d). Samples from this region formed a distinct group on the top left corner of the ordination plot, with those in the other regions lying progressively to the right as their distance upstream increased (i.e. Upper Melville Water, Lower Canning and Middle Swan estuaries; Figs 7a). While *A. pflaumii* was very abundant in Lower Melville Water, its abundance decreased substantially further upstream, in contrast to *A. bifrenatus* and to a lesser extent *P. olorum*, which increased in abundance upstream (Fig. 7b). Once again, there were no clear trends in the composition of gobiids among lunar months (data not shown).

Relationships between gobiid abundance and composition and salinity and temperature

The densities of *F. punctatus*, *A. bifrenatus* and *P. olorum* in nearshore waters were all very strongly negatively correlated with salinity ($r = -0.844$ to -0.953; $P = < 0.001$) and positively related to water temperature ($r = 0.707$ to 0.809; $P = < 0.001$; Table 4a). In offshore waters, the densities of *A. pflaumii* were positively correlated with salinity and negatively

correlated with temperature, which was the reverse of *A. bifrenatus* and, to a lesser extent, *P. olorum* (Table 4a).

The BEST test demonstrated that the spatial patterns in gobiid composition in nearshore waters were related most closely to salinity ($\rho = 0.614$, $P = 0.001$) and that, while also related to water temperature ($\rho = 0.483$, $P = 0.01$), a combination of the data for these two variables did not improve the extent of the correlation ($\rho = 0.601$). This is shown on the bubble plots where points for sites in Lower Melville Water had a far greater average salinity than those in the Upper Canning and Middle Swan estuaries (i.e. 31-35 vs typically 18-24; Figs 8a, b). Gobiid composition in offshore waters was best matched to a combination of salinity and water temperature ($\rho = 0.530$, $P = 0.001$), closely followed by salinity alone ($\rho = 0.524$, $P = 0.001$) and then water temperature ($\rho = 0.485$, $P = 0.001$). The gobiid fauna was most distinct in Lower Melville Water, which had higher salinities and lower water temperatures (Fig. 8d, e, f).

Discussion

As non-indigenous species are recognised as a pervasive and significant threat to aquatic environments globally, the capture of numerous individuals of the Striped Sandgoby *Acentrogobius pflaumii*, a species native to East Asia, in the Swan-Canning Estuary in south-western Australia, was a major concern. This prompted an investigation into the ecology of such species by detailed analysis of data collected by an established sampling regime. This information is vital to identify the native species that co-occur with this non-indigenous species and to assess *A. pflaumii*'s invasive potential and devise appropriate risk assessments and management plans.

***Abundance and distribution of* Acentrogobius pflaumii**

Acentrogobius pflaumii was not recorded in the 1,240 hand trawls conducted in the nearshore waters of the Swan-Canning Estuary over 31 consecutive lunar months, even though it represented almost 50% of the gobiids caught in adjacent offshore sites. This mirrors its distribution in its native range and in Port Phillip Bay, where it occurs in waters > 5 m deep (Yanagisawa 1978; Lockett and Gomon 2001; Horinouchi 2008). As gobies are benthic and

often inhabit burrows, the occurrence of *A. pflaumii* in the deeper, but not shallow waters of Lower Melville Water may be due to the sediment composition. As illustrated from the data provided in other studies, the substratum of the deeper waters of this region of the Swan-Canning Estuary and other south-western Australian estuaries are characterised by greater proportions of particulate organic matter and small inorganic particles (*i.e.* those with grain sizes <125 μm; Tweedley et al. 2012; 2017). The potential significance of sediment grain size is supported by that fact that *A. pflaumii* inhabits bare and vegetated soft sand, muddy sand and muddy sediments in Korea and Japan (Yanagisawa 1978; Horinouchi and Sano 2001, 2000; Horinouchi 2008).

Field observations have shown that *A. pflaumii* is able to partially bury itself and that it cohabits burrows of alpheid shrimps (Horinouchi 2008; Morgan et al. 2004), which are abundant in the sediments of Melville Water (B. Poh, Murdoch University, unpublished data). While, the mechanical capacity of *A. pflaumii* to bury itself is unknown, the coarser sediment in nearshore waters and absence of alpheid shrimp (Wildsmith et al. 2011; Tweedley et al. 2014) may reduce the potential for it to bury in these environments. It is thus relevant that most fishes exhibiting burying behaviours avoid sediments with larger grain sizes, due to the increased difficulty and energetic requirement of burial (Moles and Norcross 1995; Phelan et al. 2001; Stoner and Titgen 2003). Moreover, the gobiids that are abundant in the nearshore waters of the Swan-Canning Estuary, i.e. *F. lateralis* and *F. punctatus,* are well camouflaged against the course yellow substrates of the these waters. This contrasts with *A. bifrenatus* and *A. pflaumii,* whose darker colorations are more suited to muddier sediment. Thus, frequenting the shallow sandy habitats may increase the risk of predation from piscivorous fish and birds, especially as gobiids are less mobile than other fish species.

In addition to preferring deeper, offshore waters in the Swan-Canning Estuary, the abundance of *A. pflaumii* differed significantly among regions, with decreasing densities upstream in the estuary and its absence/very low abundance at sites upstream of Melville Water. This distribution indicates that *A. pflaumii* prefers areas where the salinity is close to that of full strength seawater. For example, salinities in Lower Melville Water were consistently the greatest of any region in the Swan-Canning Estuary, due to the close proximity

of this region to marine waters and only decreased to a minimum of ~33 at the bottom of the water column compared to ~20 at the surface. Furthermore, the density of *A. pflaumii* in this study was strongly positively correlated with bottom salinity. The tendency of *A. pflaumii* to prefer full strength seawater has also been recorded in Korea and Japan, where it inhabits coastal embayments and the lower reaches of estuaries (Horinouchi and Sano 2001; Kanou et al. 2004; Horinouchi 2008; Nan et al. 2009; Wang et al. 2015). When compared to native gobies, such as *P. olorum* and *F. lateralis,* that have been recorded in salinities as low as < 1 and as high as 76 and 50, respectively (Tweedley 2011; Potter et al. 2015b), *A. pflaumii* could be considered relatively stenohaline.

It is not known whether the lack of *A. pflaumii* in waters with a reduced salinity is solely due to a physiological inability to osmoregulate in salinities less than seawater or due to differences in sediment composition, food availability, habitat and/or biotic interactions with other species. Tweedley et al. (2017) demonstrated that sediment grain size increases in offshore areas upstream of Melville Water, which may explain why, although salinities in these areas of the Swan-Canning Estuary exceeded 32 in late summer and early autumn when the salt-wedge penetrated upstream, only three *A. pflaumii* were ever recorded in the Middle Swan Estuary. The penaeid *Metapenaeus dalli* occurred in greater densities at sites in Melville Water than further upstream and the results from laboratory experiments suggested that this species exhibited a preference for the finer sediment from Melville Water, where burying took half as long as in upstream sediments, thus reducing predation risk (Tweedley et al. 2017).

Gobiid diversity and composition

A total of eight gobiid species were recorded in the Swan-Canning Estuary during the current study, one more than thirty years earlier (Gill and Potter (1993). Two species, *A. pflaumii* and *Callogobius mucosus,* were caught only in the current survey, while *Afurcagobius suppositus* was found only in the previous study. The recent absence of *A. suppositus* is commensurate with its decline in abundance in the Middle Swan and Lower Canning Estuary between the 1970s and 2000s (Valesini et al. 2017). This likely reflects the upstream extent of sampling and the increasingly saline conditions in the estuary due to

declining rainfall and freshwater discharge (Hallett et al. 2018), as this species is known to inhabit upper, low salinity areas of estuaries and freshwater environments (Gill and Potter 1993; Morgan et al. 2014). In contrast, *C. mucosus* is a marine straggler species, that typically enters estuaries in low numbers, and its presence in the current study is likely caused by the increased salinities and greater number of samples collected.

Gobiid richness, in both water depths, and gobiid density in nearshore waters was greater in upstream areas (Canning Estuary and Middle Swan Estuary). This likely reflects the presence of estuarine gobiids, i.e. *A. bifrenatus, P. olorum* and *F. punctatus*, which are adapted to complete their life-cycle within the estuary and attain large populations in nearshore waters, increasing their likelihood of capture and thus gobiid richness in these regions. While the nearshore waters of Lower Melville Water contain a greater total number of gobiids, this included the marine straggler *C. mucosus,* an infrequently caught species and, when captured, occurred in low numbers. In contrast to nearshore waters, gobiid density in offshore waters was greatest in the most downstream regions, mainly due to the abundance of *A. pflaumii*, which, as discussed earlier, has a preference for salinities close to that of sea water (Horinouchi and Sano 2001; Kanou et al. 2004; Horinouchi 2008; Nan et al. 2009).

Although not compared statistically, mean gobiid density was far higher in the nearshore than offshore waters (i.e. 4.5 vs 1.8 individuals 500 m^{-2}), with the latter value declining to < 1 individual 500 m^{-2} excluding *A. pflaumii*. The low numbers of gobiids in offshore waters, was also recorded by previous studies (Loneragan et al. 1989; Gill and Potter 1993), with 26,232 individuals caught in nearshore waters in the latter study, but only 134 in corresponding offshore waters, of which 121 were *A. bifrenatus.* Each species found in both water depths exhibited a lower abundance in offshore waters, with the ratios of nearshore to offshore being particularly marked for *F. punctatus* 150:1 and *F. lateralis* 30:1. As mentioned earlier this could reflect the preference of these species for coarser sediments, which better suits their camouflage (Gill and Potter 1993; Gill 1996). Moreover, the burial of *F. lateralis* in silty substrates led to the accumulation of large amounts of fine particles on the gill lamellae and death (Gill and Potter 1993), an observation also made by Miller and Fouda (1986). However, even the densities of species such as *A. bifrenatus* and *P. olorum*, which prefer

muddy and silty sediments, like those in offshore waters (Gill and Potter 1993; Willis et al. 1999; Francis et al. 2003), declined by 74 and 39%, respectively in offshore than nearshore waters. For *P. olorum*, this decline may be due to the lack of aquatic macrophytes in offshore waters, which it is known to inhabit (Gill and Humphries 1995; Humphries and Potter 1993).

Gobiid species in the present study exhibited pronounced spatial segregation, presumably in an effort to partition resources and reduce competition (Gill and Potter 1993; Potter et al. 2015b). Lower Melville Water was the most distinct region in both water depths, with *F. lateralis* and *A. pflaumii* dominating the gobiid fauna of nearshore and offshore waters, respectively, and comprising > 90% of the individuals. These species attained their greatest densities in this region and their abundances were positively correlated with salinity, which, for *F. lateralis*, parallels the findings of Gill and Potter (1993). This reflects the fact that it is an Estuarine & Marine species and thus can spawn in marine waters and is known to occur in coastal waters and rock pools (Griffiths 2000; Valesini et al. 2004). Moreover, in the nearshore waters of four south-western Australian estuaries that become disconnected from the ocean at times, *F. lateralis* represented between 2 and 6% of all fish, but contributed as much as 35% of a nearby estuary whose connection to the ocean remained open (Hoeksema et al. 2009).

The overwhelming dominance of *A. pflaumii* in the offshore waters of Lower Melville Water is consistent with this species within its native range, e.g. the Jilong River Estuary in China (Nan et al. 2009) and in its non-native range, e.g. Port Phillip Bay in south-eastern Australia. In Port Phillip Bay, it was the most widely distributed and among the most abundant fish species recorded only one year after being first recorded (Lockett and Gomon 2001). This dominance in the Swan-Canning Estuary could be due to the fact that these 'open water' silty habitats in Melville Water were not previously colonised, to any substantial extent, by native gobiids. This hypothesis is supported by the fact that Maddern and Morrison (2009) found no other gobiid species utilising this habitat in the estuary and that both Loneragan et al. (1989) and Gill and Potter (1993) recorded few gobiids in offshore waters. Other gobies found typically only in the most downstream reaches of other estuaries include *Callogobius depressus, C. mucosus, Tridentiger trigonocephalus,* all of which are marine species (Griffiths 2003; Joon and Baek 2013). Moreover, *T. trigonocephalus* is typically associated with rocky

habitats (Akihito et al. 1984) and is known to lay eggs in dead oyster shells (Hoese 1973; Inui et al. 2009), which occur in the Swan-Canning Estuary in the most downstream parts of Melville Water.

Gobiid density was less in Upper Melville Water than in any other region, which is consistent with the findings of Gill and Potter (1993) and thought to be related to increased wave exposure in nearshore waters. In the corresponding offshore waters of this region, it may reflect the lower abundance of *A. pflaumii* compared to Lower Melville Water. *Favonigobius punctatus* dominated the nearshore waters of Upper Melville Water, comprising almost 90% of the gobiids, and contributing 33% to the gobiid fauna across the entire estuary. This is a marked increase on its contribution in the 1980s (Gill and Potter 1993).

The nearshore and offshore areas upstream of Melville Water, i.e. the Canning Estuary and Middle Swan Estuary, were typified by the presence of *A. bifrenatus* and *P. olorum,* with *F. punctatus* also being abundant in the shallows. The occurrence of these three native species in areas with lower salinities is responsible for their negative correlation with salinity found in both the current and earlier study (Gill and Potter 1993). These three species are able to spawn in the middle and upper regions of estuaries (Gill et al. 1996; Gill 1996), and their larvae have been caught in plankton nets in the Swan-Canning Estuary (Neira et al. 1992). Although not as abundant as in Lower Melville Water, *A. pflaumii* contributed 28 and 18% to the total number of gobiids in the offshore waters of Upper Melville Water and the Lower Canning Estuary, suggesting that, while there is spatial segregation amongst the gobiid species, some inter-specific competition may be occurring following the colonisation of the estuary by *A. pflaumii.* This finding contrasts with Francis et al. (2003), who found that although *A. bifrenatus* and *A. pflaumii* occupied similar habitats in two harbours in New Zealand, the populations of both species did not typically overlap spatially.

Conclusions, management implications and future research

The non-indigenous species *A. pflaumii*, while not present in nearshore waters of the Swan-Canning Estuary, was the most abundant gobiid species in the deeper, offshore waters, representing almost 50% of all gobiids recorded. The density and percentage contribution of

this species to the gobiid fauna in the offshore waters declined progressively in an upstream direction from 96% in Lower Melville Water to only 0.4% in the Middle Swan Estuary. Thus, *A. pflaumii* displays a preference for deeper waters with soft sediment substrates, and regions where salinities are stable and close to that of full strength seawater. The overwhelming contribution of *A. pflaumii* in the lower estuary, which historically contained few gobiids (Loneragan et al. 1989; Gill and Potter 1993), indicate that it has successfully exploited an unoccupied niche. Based on this spatial distribution and the fact that mature, spawning and depleted *A. pflaumii* gonads were recorded within the estuary (Hogan-West et al., submitted), this species is likely to belong to the Estuarine & Marine guild (sensu Potter et al. 2015a). Thus, like *F. lateralis*, another Estuarine & Marine species, *A. pflaumii* is a species that contains populations in which the individuals can complete their life cycles within the estuary (Hogan-West et al. submitted), but which are also represented by populations in marine environments, such as Cockburn Sound (Mead-Hunter 2005; Horinouchi and Sano 2000; Baeck et al. 2004).

Although the spatial distribution of *A. pflaumii* overlaps with *A. bifrenatus* and *P. olorum,* its preference for salinities close to seawater suggest that this species lacks the osmoregulatory abilities of co-occurring highly euryhaline native gobiids*,* which can penetrate the upper parts of the estuary. Determining the acute and gradual salinity preferences and tolerances of *A. pflaumii* and native species would be valuable. While, dietary studies on native gobiids show that there is little overlap in dietary preferences (Gill and Potter 1993; Humphries and Potter 1993), there are currently no complementary dietary data for *A. pflaumii*. It is relevant, however, that in its native waters, *A. pflaumii* consumes large quantities of polychaetes (Horinouchi and Sano 2000; Horinouchi 2008), similar to the diet of *A. bifrenatus* (Gill and Potter 1993)*.* Thus, studies of the diet and behaviour of *A. pflaumii* would be valuable for determining the impact of this non-indigenous species on trophic interactions and competition. As *A. pflaumii* exhibits rapid growth, an extended spawning period and is present in nearshore marine waters, it is recommended that this species is placed on a watch list and, if possible, presence/absence sampling be undertaken in nearby estuaries. Given that it is not found in shallow waters, surveys would need to sample offshore waters using a fine mesh otter trawl (as in this study) or using SCUBA (as in Maddern and Morrison 2009).

References

Akihito, Prince., M. Hayashi, and T. Yoshino. 1984. Suborder Gobioidei. In *The Fishes of the Japanese Archipelago*, eds. K. Masuda, C. Amaoka, C. Araga, T. Uyeno, and T. Yoshino, 236 289. Tokyo: Tokai University Press.

Anderson, M.J., R.N. Gorley, and K.R. Clarke. 2008. *PERMANOVA+ for PRIMER: guide to software and statistical methods*. Plymouth, UK: PRIMER-E.

Baeck, G.W., J. W. Kim, and S.H. Huh. 2004. Maturation and spawning of Striped Goby (*Acentrogobius pflaumi*) (Teleostei; Gobiidae) collected in the Gwangyang Bay, Korea. *Korean Journal of Fisheries and Aquatic Sciences* 37:226–231.

Bax, N., A. Williamson, M. Aguero, E. Gonzalez, and W. Geeves. 2003. Marine invasive alien species: a threat to global biodiversity. *Marine Policy* 27 (4):313-323.

Blackburn, T.M., F. Essl, T. Evans, P.E. Hulme, J.M. Jeschke, I. Kühn, S. Kumschick, Z. Marková, A. Mrugała, W. Nentwig, J. Pergl, P. Pyšek, W. Rabitsch, A. Ricciardi, D.M. Richardson, A. Sendek, M. Vilà, J.R.U. Wilson, M. Winter, P. Genovesi, and S. Bacher. 2014. A unified classification of alien species based on the magnitude of their environmental impacts. *PLOS Biology* 12 (5):e1001850.

Broadley, A.D., J.R. Tweedley, and N.R. Loneragan. 2017. Estimating biological parameters for penaeid restocking in a temperate Australian estuary. *Fisheries Research* 186 (2):488-501.

Carlton, J.T. 1999. The scale and ecological consequences of biological invasions in the world's oceans. In *Invasive Species and Biodiversity Management*, eds. P.J. Schei, and A. Viken, 195-212. Dordrecht: Kluwer Academic Publishers.

Clarke, K.R., M.G. Chapman, P.J. Somerfield, and H.R. Needham. 2006. Dispersion-based weighting of species counts in assemblage analyses. *Marine Ecology Progress Series* 320:11-27.

Clarke, K.R., and R.N. Gorley. 2015. *PRIMER v7: User Manual/Tutorial.* Plymouth: PRIMER-E.

Clarke, K.R., J.R. Tweedley, and F.J. Valesini. 2014. Simple shade plots aid better long-term choices of data pre-treatment in multivariate assemblage studies. *Journal of the Marine Biological Association of the United Kingdom* 94 (1):1-16.

Clarke, K.R., and R.M. Warwick. 2001. *Change in marine communities: An approach to statistical analysis and interpretation*. 2nd edition Aufl. Plymouth: PRIMER-E.

Cohen, A.N., and J.T. Carlton. 1998. Accelerating invasion rate in a highly invaded estuary. *Science* 279 (5350):555-558.

Crisp, J.A., N.R. Loneragan, J. R. Tweedley, F.M.L. D'Souza, and B. Poh. 2018. Environmental factors influencing the reproduction of an estuarine penaeid population and implications for management. *Fisheries Management and Ecology* 25 (3):203-219.

Crowder, L., and E. Norse. 2008. Essential ecological insights for marine ecosystem-based management and marine spatial planning. *Marine Policy* 32 (5):772-778.

Department of Water. 2014. Monitoring seagrass extent and distribution in the Swan-Canning Estuary. Perth, Western Australia: Department of Water.

Francis, M.P., C. Walsh, M.A. Morrison, and C. Middleton. 2003. Invasion of the Asian goby, *Acentrogobius pflaumii*, into New Zealand, with new locality records of the introduced bridled goby, *Arenigobius bifrenatus*. *New Zealand Journal of Marine and Freshwater Research* 37 (1):105-112.

Gill, H.S. 1996. Threatened fishes of the world: *Papillogobius punctatus* Gill & Miller, 1990 (Gobiidae). *Environmental Biology of Fishes* 47 (3):268-268.

Gill, H.S., and P. Humphries. 1995. An experimental evaluation of habitat choice in three species of goby (Pisces: Gobiidae). *Records of the Western Australian Museum* 17:231-233.

Gill, H.S., and I.C. Potter. 1993. Spatial segregation amongst goby species within an Australian estuary, with a comparison of the diets and salinity tolerance of the two most abundant species. *Marine Biology* 117 (3):515-526.

Gill, H.S., B.S. Wise, I.C. Potter, and J.A. Chaplin. 1996. Biannual spawning periods and resultant divergent patterns of growth in the estuarine goby *Pseudogobius olorum*: Temperature-induced? *Marine Biology* 125 (3):453-466.

Gomon, M.F., D.J. Bray, and R.H. Kuiter. 2008. *Fishes of Australia's Southern Coast*. Chatswood, Australia: New Holland.

Griffiths, S.P. 2000. The use of clove oil as an anaesthetic and method for sampling intertidal rockpool fishes. *Journal of Fish Biology* 57 (6):1453-1464.

Griffiths, S.P. 2003. Spatial and temporal dynamics of temperate Australian rockpool ichthyofaunas. *Marine and Freshwater Research* 54 (2):163-176.

Hallett, C.S., A.J. Hobday, J.R. Tweedley, P.A. Thompson, K. McMahon, and F.J. Valesini. 2018. Observed and predicted impacts of climate change on the estuaries of south-western Australia, a Mediterranean climate region. *Regional Environmental Change* 18 (5):1357-1373.

Hewitt, C.L., M.L. Campbell, R.E. Thresher, R.B. Martin, S. Boyd, B.F. Cohen, D.R. Currie, M.F. Gomon, M.J. Keough, J.A. Lewis, M.M. Lockett, N. Mays, M.A. McArthur, T.D. O'Hara, G.C.B. Poore, D.J. Ross, M.J. Storey, J.E. Watson, and R.S. Wilson. 2004. Introduced and cryptogenic species in Port Phillip Bay, Victoria, Australia. *Marine Biology* 144 (1):183-202.

Hodgkin, E.P., and P. Hesp. 1998. Estuaries to salt lakes: Holocene transformation of the estuarine ecosystems of south-western Australia. *Marine and Freshwater Research* 49 (3):183-201.

Hoeksema, S.D., B.M. Chuwen, and I.C. Potter. 2009. Comparisons between the characteristics of ichthyofaunas in nearshore waters of five estuaries with varying degrees of connectivity with the ocean. *Estuarine, Coastal and Shelf Science* 85 (1):22-35.

Hoese, D. 1973. The introduction of the gobiid fishes *Acanthogobius flavimanus* and *Tridentiger trigonocephalus* into Australia. *Koolewong* 2:3-5.

Hogan-West, K., P.G. Coulson, J.R. Tweedley, and N.R. Loneragan. submitted. Biological characteristics of the introduced gobiid *Acentrogobius pflaumii* in a warm-temperate estuary and thus very different from its typical environment. *Marine Biodiversity*.

Horinouchi, M. 2008. Patterns of food and microhabitat resource use by two benthic gobiid fishes. *Environmental Biology of Fishes* 82 (2):187-194.

Horinouchi, M., and M. Sano. 2000. Food habits of fishes in a *Zostera marina* bed at Aburatsubo, central Japan. *Ichthyological Research* 47:163–173.

Horinouchi, M., and M. Sano. 2001. Effects of changes in seagrass shoot density and leaf height on the abundance of juveniles of *Acentrogobius pflaumii* in a zostera marina bed. *Ichthyological Research* 48:179-185.

Hulme, P.E. 2009. Trade, transport and trouble: managing invasive species pathways in an era of globalization. *Journal of Applied Ecology* 46 (1):10-18.

Humphries, P., and I.C. Potter. 1993. Relationship between the habitat and diet of three species of atherinids and three species of gobies in a temperate Australian estuary. *Marine Biology* 116 (2):193-204.

Humphries, P., I.C. Potter, and N.R. Loneragan. 1992. The fish community in the shallows of a temperate Australian estuary: relationships with the aquatic macrophyte *Ruppia megacarpa* and environmental variables. *Estuarine, Coastal and Shelf Science* 34 (4):325-346.

Inui, R., N. Onikura, M. Kawagishi, M. Nakatani, Y. Tomiyama, and S. Oikawa. 2009. Selection of spawning habitat by several gobiid fishes in the subtidal zone of a small temperate estuary. *Fisheries Science* 76 (1):83.

Jackson, J.B.C., M.X. Kirby, W.H. Berger, K.A. Bjorndal, L.W. Botsford, B.J. Bourque, R.H. Bradbury, R. Cooke, E. Jon, J.A. Estes, T.P. Hughes, S. Kidwell, C.B. Lange, H.S. Lenihan, J.M. Pandolfi, C.H. Peterson, R.S. Steneck, M.J. Tegner, and R.R. Warner. 2001. Historical overfishing and the recent collapse of coastal ecosystems. *Science* 293 (5530):629-638.

Joon, H.I., and H. Baek. 2013. Reproductive cycle of Chameleon Goby, *Tridentiger trigonocephalus* in the southern coastal waters of Korea. *Development & Reproduction* 17:353-361.

Jude, D.J., R.H. Reider, and G.R. Smith. 1992. Establishment of Gobiidae in the Great Lakes Basin. *Canadian Journal of Fisheries and Aquatic Sciences* 49 (2):416-421.

Kanou, K., M. Sano, and H. Kohno. 2004. Food habits of fishes on unvegetated tidal mudflats in Tokyo Bay, central Japan. *Fisheries Science* 70:978-987.

Katsanevakis, S., M. Coll, C. Piroddi, J. Steenbeek, R.L.F. Ben, A. Zenetos, and A.C. Cardoso. 2014a. Invading the Mediterranean Sea: biodiversity patterns shaped by human activities. *Frontiers in Marine Science* 1:Article 32.

Katsanevakis, S., F. Tempera, and H. Teixeira. 2016. Mapping the impact of alien species on marine ecosystems: the Mediterranean Sea case study. *Diversity and Distributions* 22 (6):694-707.

Katsanevakis, S., I. Wallentinus, A. Zenetos, E. Leppäkoski, M.E. Çinar, B. Oztürk, M. Grabowski, D. Golani, and A.C. Cardoso. 2014b. Impacts of invasive alien marine species on ecosystem services and biodiversity: a pan-European review *Aquatic Invasions* 9 (4):391-423.

Lockett, M.M., and M.F. Gomon. 1999. Occurrence and distribution of exotic fishes in Port Phillip Bay. In *Marine biological invasions of Port Phillip Bay, Victoria*, eds. C.L. Hewitt, M.L. Campbell, R.E. Thresher, and R.B. Martin, 178-192. CSIRO Marine Research, Hobart: CRIMP Technical Report 20.

Lockett, M.M., and M.F. Gomon. 2001. Ship mediated fish invasions in Australia: two new introductions and a consideration of two previous invasions. *Biological Invasions* 3 (2):187-192.

Loneragan, N.R., I.C. Potter, and R.C.J. Lenanton. 1989. Influence of site, season and year on contributions made by marine, estuarine, diadromous and freshwater species to the fish fauna of a temperate Australian estuary. *Marine Biology* 103 (4):461-479.

Maddern, M., and M. Morrison. 2009. Introduction of the Streaked Goby *Acentrogobius pflaumii* (Bleeker 1853) (Pisces: Gobiidae) into southwestern Australia. *Australian Zoologist* 35 (1):96-99.

Masuda, H., K. Amaoka, C. Araga, T. Uyeno, and T. Yoshino. 1984. *The Fishes of the Japanese Archipelago*. Tokyo, Japan.: Tokai University Press.

Mead-Hunter, D. 2005. Another introduced fish species for Western Australian waters. *The Western Australian Naturalist* 24:204.

Middleton, M.J. 1982. The Oriental Goby, *Acanthogobius flavimanus* (Temminck and Schlegel), an introduced fish in the coastal waters of New South Wales, Australia. *Journal of Fish Biology* 21 (5):513-523.

Miller, P.J., and M.M. Fouda. 1986. Notes on the biology of a red sea goby, *Silhouettea aegyptia* (Chabanaud, 1933) (Teleostei: Gobiidae). *Cybium* 10:395-409.

Moles, A., and B.L. Norcross. 1995. Sediment preference in juvenile pacific flatfishes. *Netherlands Journal of Sea Research* 34 (1):177-182.

Molnar, J.L., R.L. Gamboa, C. Revenga, and M.D. Spalding. 2008. Assessing the global threat of invasive species to marine biodiversity. *Frontiers in Ecology and the Environment* 6 (9):485-492.

Morgan, D.L., H.S. Gill, M.G. Maddern, and S.J. Beatty. 2004. Distribution and impacts of introduced freshwater fishes in Western Australia. *New Zealand Journal of Marine and Freshwater Research* 38 (511-523).

Morgan, D.L., P.J. Unmack, S.J. Beatty, B.C. Ebner, M.G. Allen, J.J. Keleher, J.A. Donadson, and J. Murphy. 2014. An overview of the 'freshwater fishes' of Western Australia. *Journal of the Royal Society of Western Australia* 97:263–278.

Nan, L.N., S. Changchun, and Z. Junsheng. 2009. Species composition and seasonal variation of fish larvae and juveniles in Jiulong River Estuary. *South China Fisheries Science* 5:1-8.

Neira, F.J., I.C. Potter, and J.S. Bradley. 1992. Seasonal and spatial changes in the larval fish fauna within a large temperate Australian estuary. *Marine Biology* 112 (1):1-16.

Ojaveer, H., B.S. Galil, D. Minchin, S. Olenin, A. Amorim, J. Canning-Clode, P. Chainho, G.H. Copp, S. Gollasch, A. Jelmert, M. Lehtiniemi, C. McKenzie, J. Mikuš, L. Miossec, A. Occhipinti-Ambrogi, M. Pećarević, J. Pederson, G. Quilez-Badia, J.W.M. Wijsman, and A. Zenetos. 2014. Ten recommendations for advancing the assessment and management of non-indigenous species in marine ecosystems. *Marine Policy* 44:160-165.

Phelan, B.A., J.P. Manderson, A.W. Stoner, and A.J. Bejda. 2001. Size-related shifts in the habitat associations of young-of-the-year winter flounder (*Pseudopleuronectes americanus*): field observations and laboratory experiments with sediments and prey. *Journal of Experimental Marine Biology and Ecology* 257 (2):297-315.

Poh, B., J.R. Tweedley, J.A. Chaplin, K.M. Trayler, Crisp J.A., and N.R. Loneragan. 2019. Influence of physico-chemical and biotic factors on the distribution of a penaeid in a temperate estuary. *Estuarine, Coastal and Shelf Science* 218:70-85.

Poh, B., J.R. Tweedley, J.A. Chaplin, K.M. Trayler, and N.R. Loneragan. 2018. Estimating predation rates of restocked individuals: The influence of timing-of-release on metapenaeid survival. *Fisheries Research* 198:165-179.

Potter, I.C., and G.A. Hyndes. 1999. Characteristics of the ichthyofaunas of southwestern Australian estuaries, including comparisons with holarctic estuaries and estuaries elsewhere in temperate Australia: a review. *Austral Ecology* 24 (4):395-421.

Potter, I.C., J.R. Tweedley, M. Elliott, and A.K. Whitfield. 2015a. The ways in which fish use estuaries: a refinement and expansion of the guild approach. *Fish and Fisheries* 16 (2):230-239.

Potter, I.C., L.J. Veale, J.R. Tweedley, and K.R. Clarke. 2016. Decadal changes in the ichthyofauna of a eutrophic estuary following a remedial engineering modification and subsequent environmental shifts. *Estuarine, Coastal and Shelf Science* 181:345-363.

Potter, I.C., R.M. Warwick, N.G. Hall, and J.R. Tweedley. 2015b. The physico-chemical characteristics, biota and fisheries of estuaries. In *Freshwater Fisheries Ecology*, ed. J. Craig, 48-79. Wiley-Blackwell.

Prince, J.D., I.C. Potter, R.C.J. Lenanton, and N.R. Loneragan. 1982. Segregation and feeding of atherinid species (Teleostei) in south-western Australian estuaries. *Marine and Freshwater Research* 33 (5):865-880.

Rowe, D.K., A. Moore, A. Giorgetti, C. Maclean, P. Grace, S. Wadhwa, and J. Cooke. 2008. Review of the impacts of gambusia, redfin perch, tench, roach, yellowfin goby and streaked goby in Australia. Canberra, Australia: Australian Government Department of the Environment, Water, Heritage and the Arts.

Shao, K.T., J.P. Chen, P.H. Kao, and C.Y. Wu. 1993. Fish fauna and their geographical distribution along the western coast of Taiwan. *Acta Zoologica Taiwanica* 4:113-140.

Simberloff, D. 2003. How much information on population biology is needed to manage introduced species? *Conservation Biology* 17 (1):83-92.

Stoner, A.W., and R.H. Titgen. 2003. Biological structures and bottom type influence habitat choices made by Alaska flatfishes. *Journal of Experimental Marine Biology and Ecology* 292 (1):43-59.

Thresher, R.E. 1999. Diversity, impacts and options for managing invasive marine species in Australian waters. *Australian Journal of Environmental Management* 6 (3):137-148.

Tweedley, J.R. 2011. The Relationships Between Habitat Types and Faunal Community Structure in Broke Inlet, Western Australia. PhD, Murdoch University, Australia, Perth.

Tweedley, J.R., R.M. Warwick, and I.C. Potter. 2016. The contrasting ecology of temperate macrotidal and microtidal estuaries. *Oceanography and Marine Biology: An Annual Review* 54:73-171.

Tweedley, J.R., C.S. Hallett, R.M. Warwick, K.R. Clarke, and I.C. Potter. 2016. The hypoxia that developed in a microtidal estuary following an extreme storm produced dramatic changes in the benthos. *Marine and Freshwater Research* 67 (3):327-341.

Tweedley, J.R., N.R. Loneragan, J.A. Crisp, B. Poh, A.D. Broadley, A.L. Bennett, K.P. Hodson, K.M. Trayler, G.I. Jenkins, and J.A. Chaplin. 2017. Stock enhancement of the Western School Prawn (*Metapenaeus dalli*) in the Swan-Canning Estuary; evaluating recruitment limitation, environment and release strategies. Murdoch University, Perth, Western Australia: Report for the Fisheries Research and Development Corporation.

Tweedley, J.R., R.M. Warwick, K.R. Clarke, and I.C. Potter. 2014. Family-level AMBI is valid for use in the north-eastern Atlantic but not for assessing the health of microtidal Australian estuaries. *Estuarine, Coastal and Shelf Science* 141:85-96.

Tweedley, J.R., R.M. Warwick, and I.C. Potter. 2015. Can biotic indicators distinguish between natural and anthropogenic environmental stress in estuaries? *Journal of Sea Research* 102 (0):10-21.

Tweedley, J.R., R.M. Warwick, F.J. Valesini, M.E. Platell, and I.C. Potter. 2012. The use of benthic macroinvertebrates to establish a benchmark for evaluating the environmental quality of microtidal, temperate southern hemisphere estuaries. *Marine Pollution Bulletin* 64 (6):1210-1221.

Valesini, F.J., N.J. Coen, M.D. Wildsmith, M. Hourston, J.R. Tweedley, C.S. Hallett, T.E. Linke, and I.C. Potter. 2009. Relationships between fish faunas and habitat type in south-western Australian estuaries. Project 2004/045. Final Report for Fisheries Research and Development Corporation. Perth: Murdoch University.

Valesini, F.J., A. Cottingham, C.S. Hallett, and K.R. Clarke. 2017. Interdecadal changes in the community, population and individual levels of the fish fauna of an extensively modified estuary. *Journal of Fish Biology* 90 (5):1734-1767.

Valesini, F.J., I.C. Potter, and K.R. Clarke. 2004. To what extent are the fish compositions at nearshore sites along a heterogeneous coast related to habitat type? *Estuarine, Coastal and Shelf Science* 60 (4):737-754.

Valesini, F.J., J.R. Tweedley, K.R. Clarke, and I.C. Potter. 2014. The importance of regional, system-wide and local spatial scales in structuring temperate estuarine fish communities. *Estuaries and Coasts* 37 (3):525-547.

Veale, L., J.R. Tweedley, K.R. Clarke, C.S. Hallett, and I.C. Potter. 2014. Characteristics of the ichthyofauna of a temperate microtidal estuary with a reverse salinity gradient, including inter-decadal comparisons. *Journal of Fish Biology* 85 (5):1320-1354.

Wang, L.J., F. You, Q.X. Wang, Z.H. Wu, and M.X. Liu. 2015. Length–weight and length–length relationships of 11 fish species from Zhimai River estuary, China. *Journal of Applied Ichthyology* 31 (2):435-436.

Warwick, R.M., J.R. Tweedley, and I.C. Potter. 2018. Microtidal estuaries warrant special management measures that recognise their critical vulnerability to pollution and climate change. *Marine Pollution Bulletin* 135:41-46.

Wildsmith, M.D., I.C. Potter, F.J. Valesini, and M.E. Platell. 2005. Do the assemblages of the benthic macroinvertebrates in nearshore waters of Western Australia vary among habitat types, zones and seasons? *Journal of the Marine Biological Association of the United Kingdom* 85:217-232.

Wildsmith, M.D., T.H. Rose, I.C. Potter, R.M. Warwick, and K.R. Clarke. 2011. Benthic macroinvertebrates as indicators of environmental deterioration in a large microtidal estuary. *Marine Pollution Bulletin* 62 (3):525-538.

Williams, S.L., and E.D. Grosholz. 2008. The invasive species challenge in estuarine and coastal environments: marrying management and science. *Estuaries and Coasts* 31 (1):3-20.

Willis, T.J., J.E.H. Saunders, D.L. Blackwood, and J.E. Archer. 1999. First New Zealand record of the Australian bridled goby, *Arenigobius bifrenatus* (Pisces: Gobiidae). *New Zealand Journal of Marine and Freshwater Research* 33 (2):189-192.

Wonham, M.J., J.T. Carlton, G.M. Ruiz, and L.D. Smith. 2000. Fish and ships: relating dispersal frequency to success in biological invasions. *Marine Biology* 136 (6):1111-1121.

Workman, M.L., and J.E. Merz. 2007. Introduced yellowfin goby, *Acanthogobius flavimanus*: diet and habitat use in the Lower Mokelumne River, California. *San Francisco Estuary and Watershed Science* 5:1-13.

Worm, B., E.B. Barbier, N. Beaumont, J.E. Duffy, C. Folke, B.S. Halpern, J.B.C. Jackson, H.K. Lotze, F. Micheli, S.R. Palumbi, E. Sala, K.A. Selkoe, J.J. Stachowicz, and R. Watson. 2006. Impacts of biodiversity loss on ocean ecosystem services. *Science* 314 (5800):787-790.

Yanagisawa, Y. 1978. Studies on the interspecific relationship between gobiid fish and snapping shrimp. I. Gobiid fishes associated with snapping shrimps in Japan. *Publication of the Seto Marine Biological Laboratory* 24:269–325.

Figures
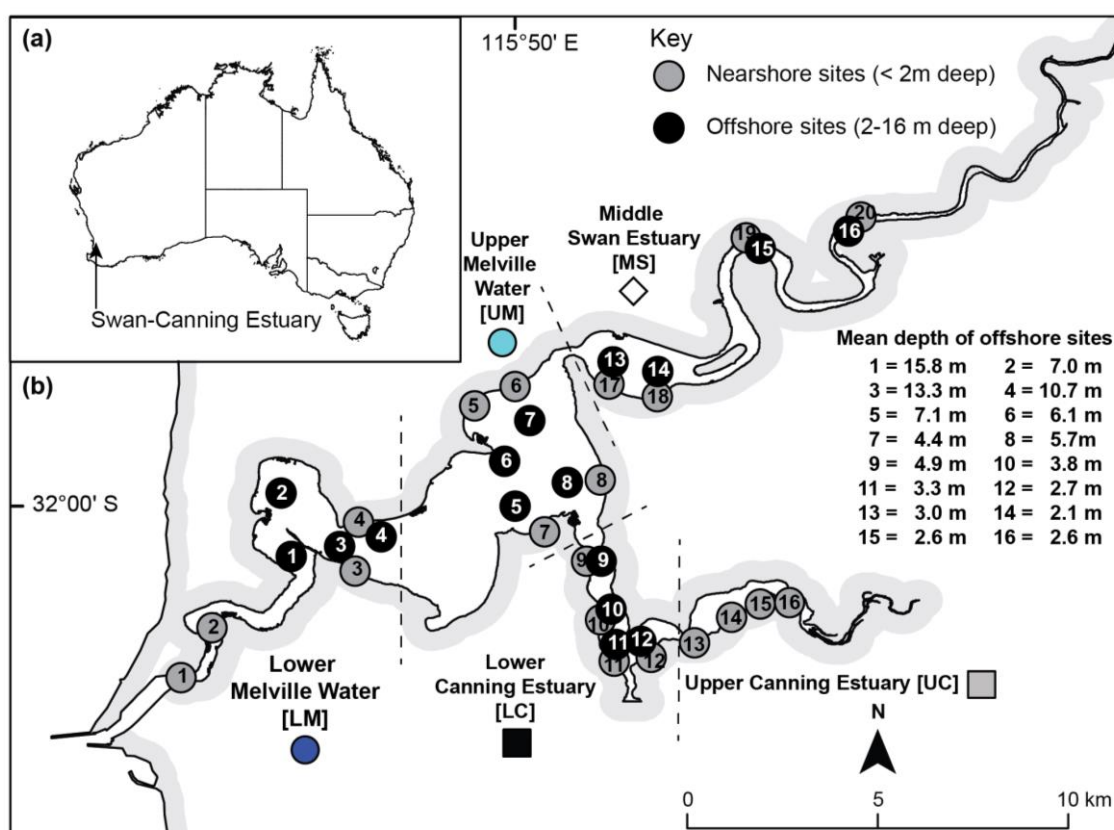
Fig. 1. Map showing the location of the Swan-Canning Estuary in south-western Australia and the 20 nearshore (< 2 m deep) and 16 offshore (> 2 m deep) sites in Swan-Canning Estuary sampled over 31 consecutive lunar months between October 2013 and March 2016. Dotted lines denote the separation among the five regions (bold face) of the estuary. The mean depth of each offshore site is provided. Seagrass (*i.e. Halophila ovalis, Ruppia megacarpa* and/or *Zostera muelleri*) was present at nearshore sites 1 to 7, 9, 12 and 14).

Fig. 2. Mean values for (a) surface and (b) bottom salinity and (c) surface and (d) bottom water temperature recorded in each of t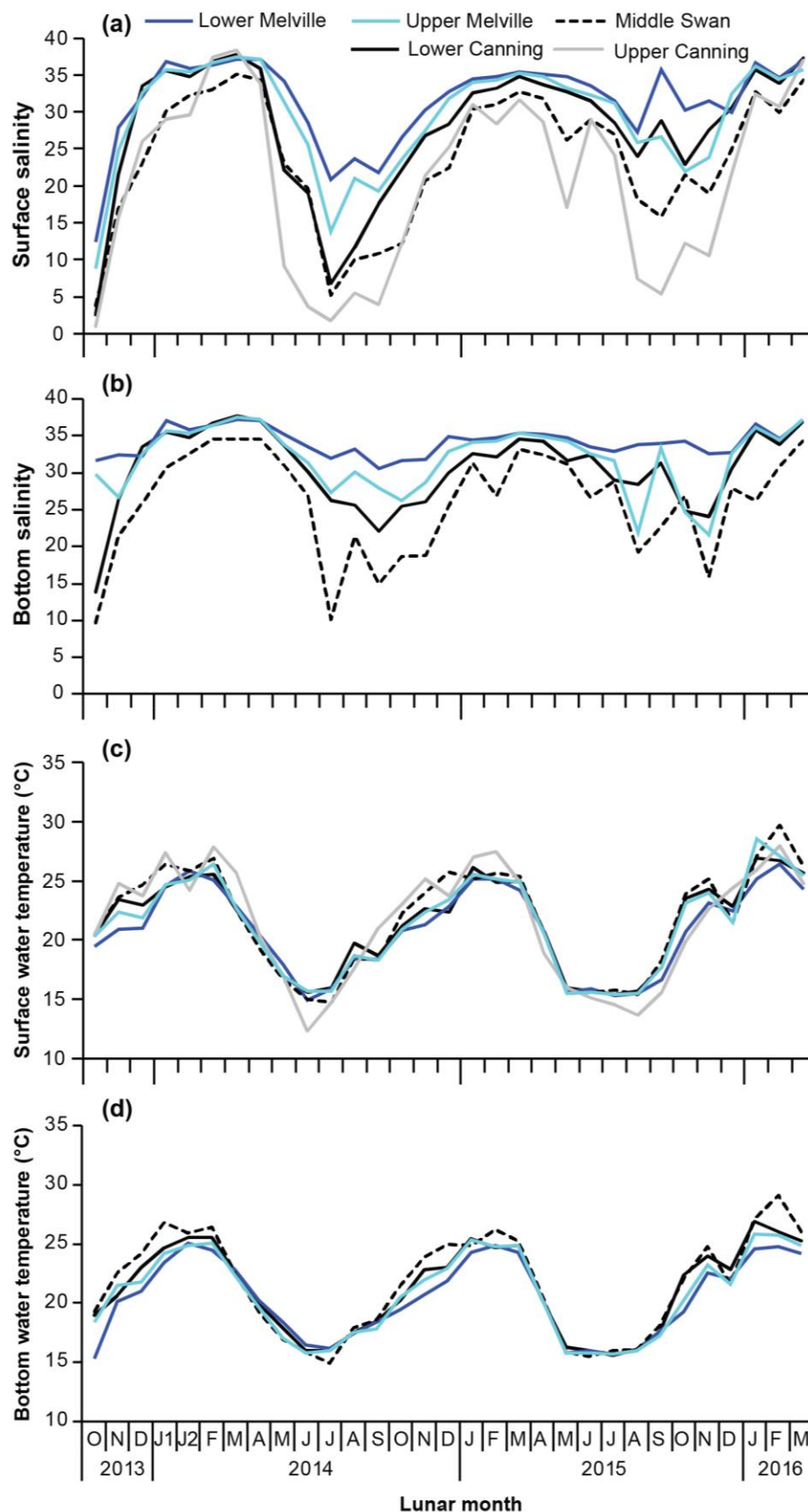he five regions of the Swan-Canning Estuary in each lunar month between October 2013 and March 2016. Note there were two lunar months in January 2014 and that due to shallow depths, only the surface waters of the Upper Canning Estuary were sampled.
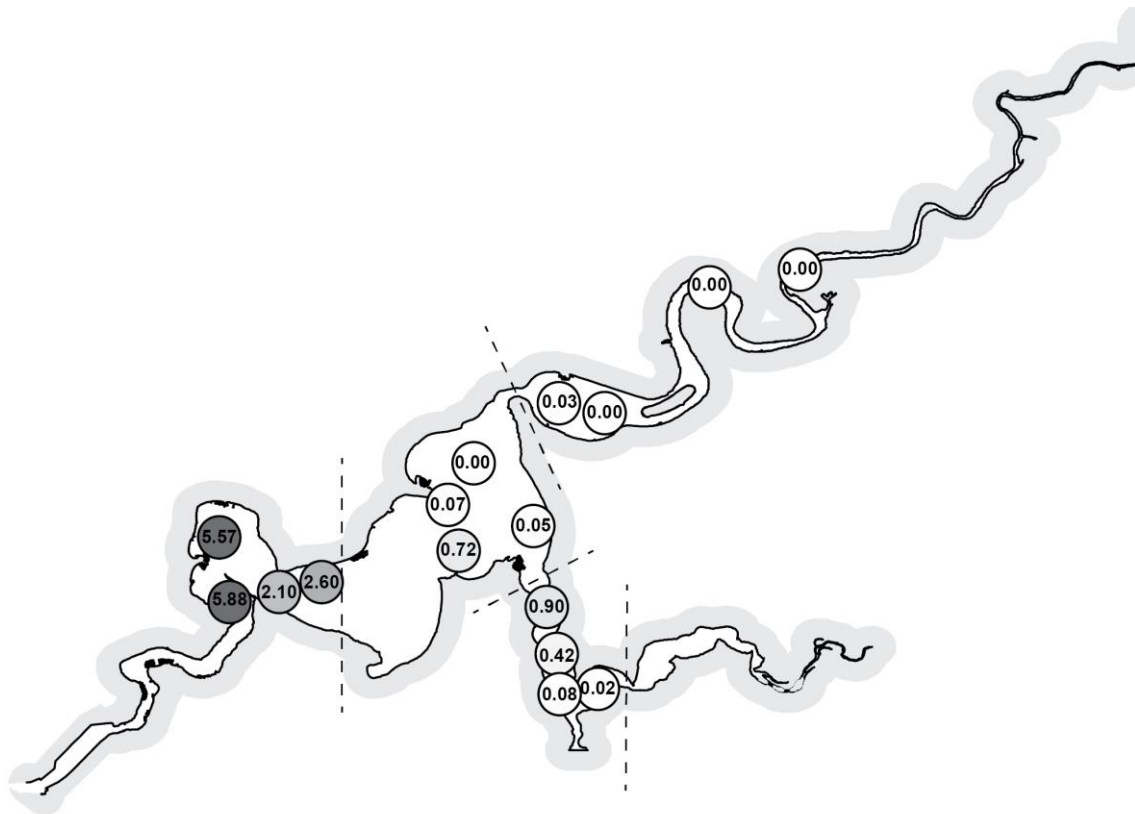
Fig. 3. Mean density 500 m^{-2} of *Acentrogobius pflaumii* at each of the 16 offshore sites in the Swan-Canning Estuary sampled every lunar month between October 2013 and March 2016. Shading intensity (white to dark grey) proportional to abundance.
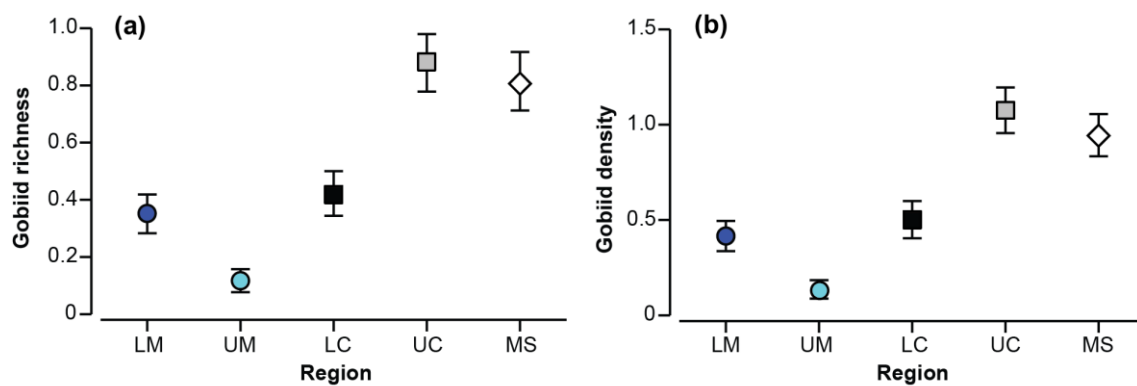
Fig. 4. Mean and 95% confidence intervals for (a) gobiid richness and (b) gobiid density (individuals 500 m^{-2}) among regions in nearshore waters of the Swan-Canning Estuary between October 2013 and March 2016. LM = Lower Melville Water, UM = Upper Melville Water, LC = Lower Canning Estuary, MS = Middle Swan Estuary and UC = Upper Canning Estuary.
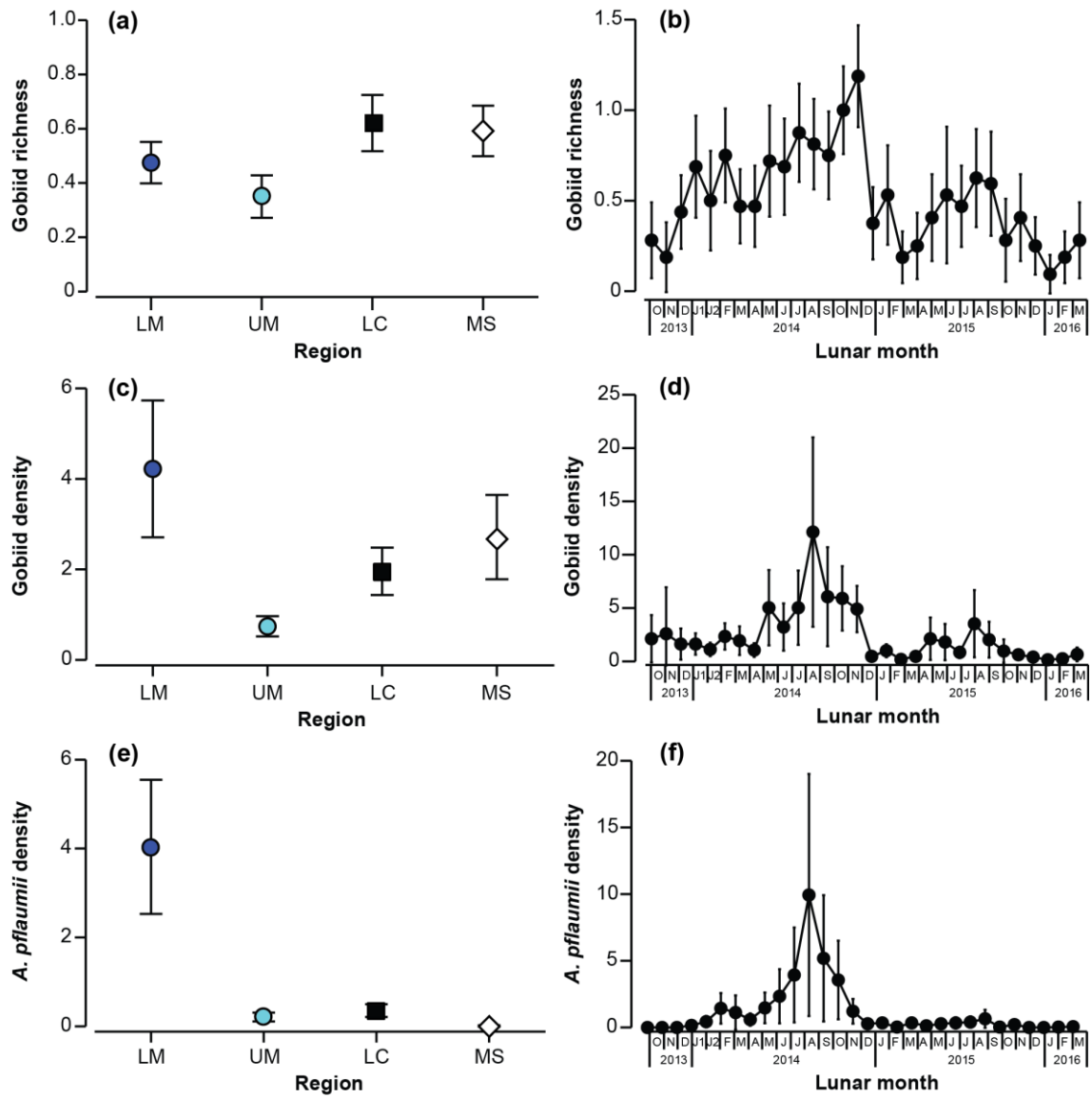
Fig. 5. Mean and 95% confidence intervals for gobiid richness among (a) regions and (b) lunar months, gobiid density (individuals 500 m^{-2}) among (c) regions and (d) lunar months and density of *Acentrogobius pflaumii* (individuals 500 m^{-2}) among (e) regions and (f) lunar months in offshore waters of the Swan-Canning Estuary between October 2013 and March 2016. LM = Lower Melville Water, UM = Upper Melville Water, LC = Lower Canning Estuary, MS = Middle Swan Estuary and UC = Upper Canning Estuary.

Fig. 6. (a) Two-dimensional nMDS ordination plots constructed from bootstrap averages for the average dispersion-weighted and square-transformed densities of the various goby species recorded in the nearshore waters of each of the five regions of the Swan-Canning Estuary in each of the 31 lunar months between October 2013 and March 2016. (b) Shade plot of the dispersion-weighted square-root transformed densities (fish 500 m^{-2}) of the five native gobiid species recorded in the nearshore waters each of the five regions of the Swan-Canning Estuary averaged, across the 31 lunar months between October 2013 and March 2016. The order of the regions (x axis) and species (y axis) determined by seriation.

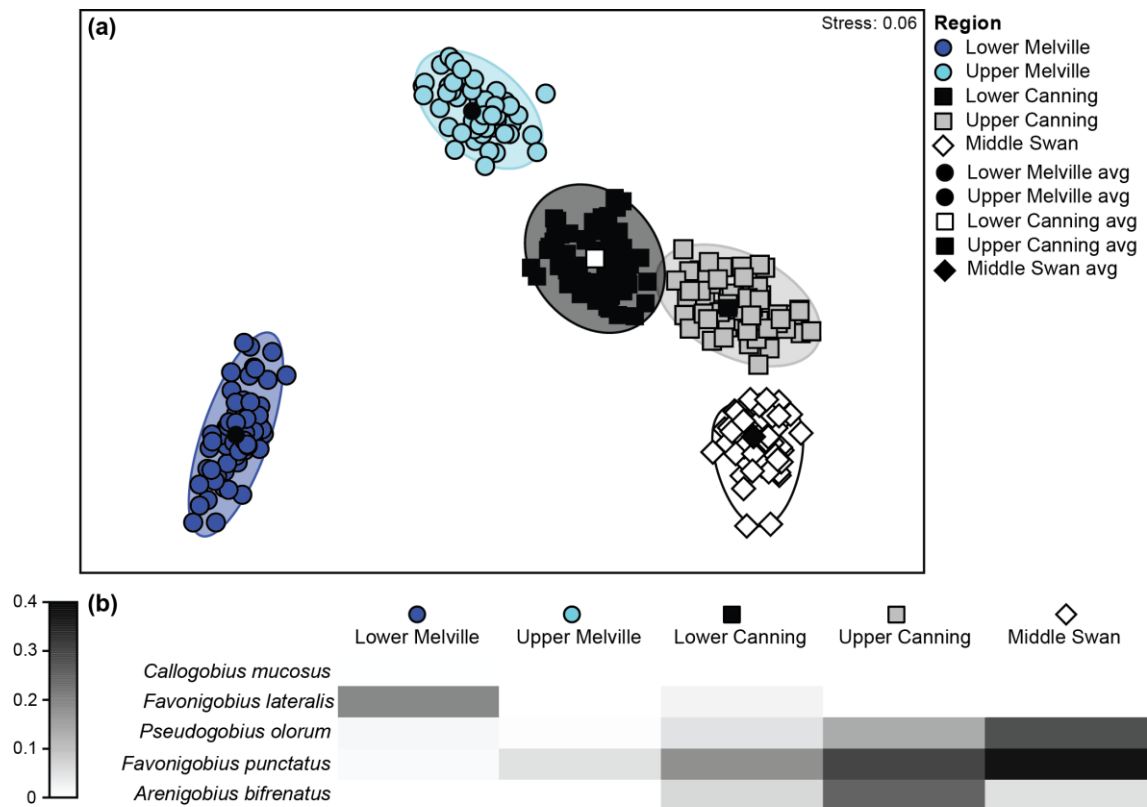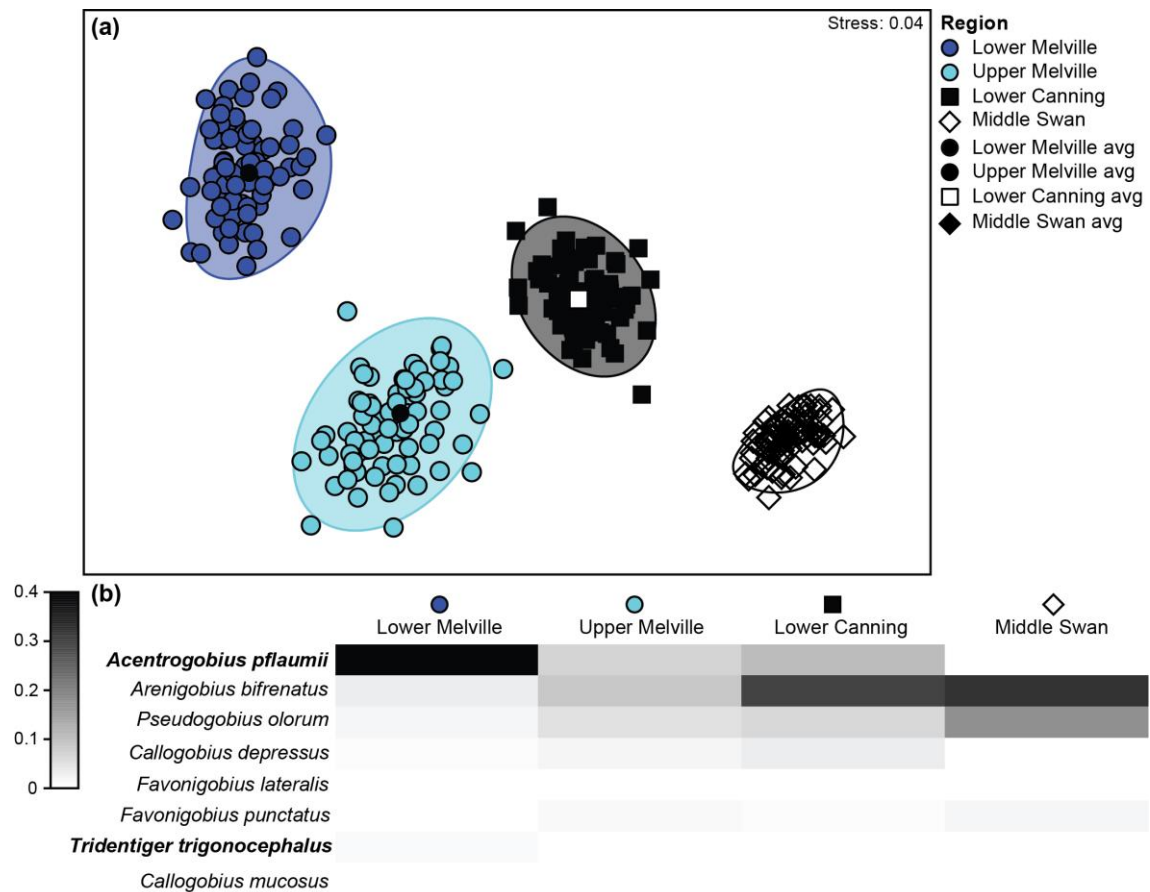**Fig. 7.** (a) Two-dimensional nMDS ordination plots constructed from bootstrap averages for the average dispersion-weighted and square-root transformed densities of the various goby species recorded in the offshore waters of each of the four regions of the Swan-Canning Estuary in each of the 31 lunar months between October 2013 and March 2016. (b) Shade plot of the dispersion-weighted and square-root transformed densities (fish 500 m^{-2}) of the eight native and non-indigenous gobiid species recorded in the offshore waters each of the four regions of the Swan-Canning Estuary, averaged across the 31 lunar months between October 2013 and March 2016, excluding December 2014. The order of the regions (x axis) and species (y axis) determined by seriation. Non-indigenous species highlighted in bold.
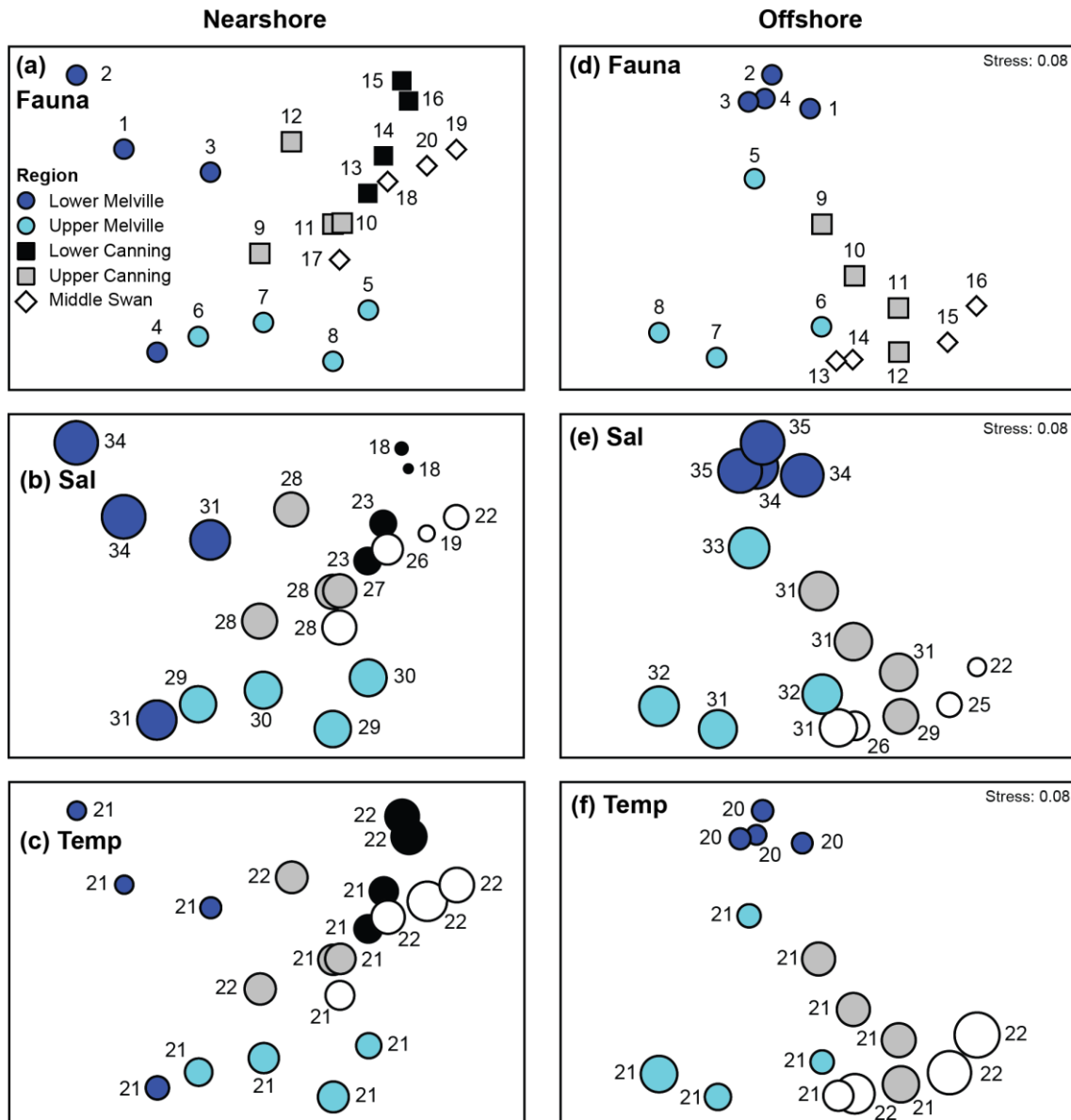
Fig. 8. nMDS ordination plots of the mean gobiid composition (a, d; fauna) recorded at each of the 20 nearshore and 16 offshore sites and the magnitude of the mean salinity (b, e; Sal) and water temperature (c, f; Temp) at each site overlaid as circles of proportionate size.

Table 1. Life cycle guilds (LC), rankings by abundance (R), mean densities of each gobiid species (individuals 500 m^{-2}; X), standard deviation (SD) and percentage contribution to total gobiid catch (%) in the five and four different regions, respectively, and pooled regions of the (a) nearshore and (b) offshore waters of the Swan-Canning Estuary between January 2013 and March 2016. Total number of samples and species, individuals and density (individuals 500 m^{-2}) are also provided. Non-indigenous species are highlighted are in bold and species contributing >10% of total goby catch in each region are shaded grey. Life cycle guilds for gobiid species are allocated according to the criteria in Potter et al (2015a), i.e. solely estuarine (E), estuarine & marine (EM), estuarine & freshwater (EF) and marine straggler (MS). * On the basis of the data in this study and Hogan-West et al. (submitted) *Acentrogobius pflaumii* was assigned to the EM guild (see Discussion).

(a) Nearshore

Species	LC	Whole estuary R	X	SD	%	Lower Melville R	X	SD	%	Upper Melville R	X	SD	%	Lower Canning R	X	SD	%	Middle Swan R	X	SD	%	Upper Canning R	X	SD	%
Arenigobius bifrenatus	E	1	2.05	10.88	46.04					3	0.01	0.08	2.35	1	3.02	28.60	61.75	3	0.62	3.92	9.58	1	6.59	21.80	70.10
Favonigobius punctatus	E	2	1.50	5.14	33.68	3	0.04	0.30	3.40	1	0.27	1.58	89.41	2	1.24	4.59	25.40	1	3.97	13.43	61.02	2	1.98	5.79	21.01
Pseudogobius olorum	EF	3	0.61	2.18	13.63	2	0.07	0.51	6.17	2	0.02	0.24	7.06	4	0.19	1.09	3.93	2	1.91	5.76	29.40	3	0.84	3.29	8.89
Favonigobius lateralis	EM	4	0.30	1.92	6.63	1	1.03	2.93	90.12	4	<0.01	0.06	1.18	3	0.44	6.58	8.91								
Callogobius mucosus	MS	5	<0.01	0.01	0.02	4	<0.01	0.06	0.31																

	Whole estuary	Lower Melville	Upper Melville	Lower Canning	Middle Swan	Upper Canning
Total no. samples	1,240	248	248	248	248	248
Total no. species	5	4	4	4	3	3
Total density	4.45	1.15	0.30	4.88	6.50	9.41

(b) Offshore

Species	LC	Whole estuary R	X	SD	%	Lower Melville R	X	SD	%	Upper Melville R	X	SD	%	Lower Canning R	X	SD	%	Middle Swan R	X	SD	%
Acentrogobius pflaumii	EM*	**1**	**0.86**	**2.64**	**47.35**	**1**	**3.01**	**9.00**	**95.48**	**2**	**0.16**	**0.60**	**28.00**	**2**	**0.26**	**0.86**	**17.89**	**4**	**0.01**	**0.10**	**0.37**
Arenigobius bifrenatus	E	2	0.52	1.55	28.71	2	0.07	0.39	2.20	1	0.18	0.67	30.67	1	0.88	2.27	60.53	2	0.95	2.87	46.08
Pseudogobius olorum	EF	3	0.37	1.61	20.44	3	0.05	0.36	1.71	3	0.15	0.66	26.67	3	0.19	0.85	13.16	1	1.08	4.55	52.40
Callogobius depressus	MS	4	0.04	0.30	2.01	5	0.01	0.08	0.24	4	0.05	0.30	8.00	4	0.09	0.83	6.32				
Favonigobius punctatus	E	5	0.01	0.12	0.74	6	<0.01	0.05	<0.01	6	0.02	0.15	2.67	5	0.02	0.14	1.05	3	0.02	0.16	1.11
Favonigobius lateralis	EM	6	0.01	0.13	0.53	6	<0.01	0.05	<0.01	5	0.02	0.30	4.00	5	0.02	0.19	1.05				
Tridentiger trigonocephalus	MS	**7**	**<0.01**	**0.05**	**0.21**	**4**	**0.02**	**0.14**	**0.49**					**7**	**<0.01**	**0.05**	**<0.01**				
Callogobius mucosus	MS	8	<0.01	0.01	<0.01	6	<0.01	0.05	<0.01												

	Whole estuary	Lower Melville	Upper Melville	Lower Canning	Middle Swan
Total no. samples	960	240	240	240	240
Total no. species	8	8	6	7	4
Total density	1.82	3.14	0.58	1.46	2.08

Table 2. Degrees of freedom (*df*), mean squares (MS), percentage mean squares (%MS), Pseudo-*F* value (pF) and significance level (*P*) for two-way PERMANOVA tests on (a,c) gobiid richness and (b,d) gobiid density (individuals 500 m^{-2}), in nearshore and offshore waters, respectively and (e) density of *Acentrogobius pflaumii* (individuals 500 m^{-2}) in offshore waters. Significant differences highlighted in bold and those terms in the model responsible for > 25% of the total mean squares are shaded in grey.

Term	*df*	(a) Nearshore gobiid richness				(b) Nearshore gobiid density			
		MS	%MS	pF	*P*	MS	%MS	pF	*P*
Lunar month	30	1.73	5.91	5.18	0.001	3.00	6.88	7.08	0.001
Region	4	26.37	90.16	78.97	0.001	38.98	89.42	91.96	0.001
Lunar Month × Region	119	0.82	2.79	2.44	0.001	1.19	2.73	2.80	0.001
Residual	1086	0.33	1.14			0.42	0.97		

Term	*df*	(c) Offshore gobiid richness				(d) Offshore gobiid density				(e) Offshore density of *A. pflaumii*			
		MS	%MS	pF	*P*	MS	%MS	pF	*P*	MS	%MS	pF	*P*
Lunar month	29	2.20	31.51	5.50	0.001	9.44	28.24	7.81	0.001	4.39	7.86	8.70	0.001
Region	3	3.66	52.43	9.15	0.001	18.91	56.54	15.63	0.001	47.51	85.02	94.06	0.001
Lunar Month × Region	87	0.72	10.32	1.80	0.001	3.88	11.60	3.21	0.001	3.48	6.22	6.88	0.001
Residual	840	0.40	5.73			1.21	3.62			.51	0.90		

Table 3. Degrees of freedom (df), mean squares (MS), percentage mean squares (%MS), Pseudo-*F* value (pF) and significance level (*P*) for a two-way PERMANOVA test on the densities of the various goby species recorded in the regions in (a) nearshore and (c) offshore waters of the Swan-Canning Estuary every lunar month between October 2013 and March 2016. Significant differences highlighted in bold and those terms in the model responsible for > 25% of the total mean squares are shaded in grey. PERMANOVA *t* values for pairwise comparison of the gobiid faunal composition between the various regions in the (b) nearshore and (d) offshore waters. Note that a significant difference was detected for each pairwise comparison (*P* < 0.05). LM = Lower Melville Water, UM = Upper Melville Water, LC = Lower Canning Estuary, MS = Middle Swan Estuary and UC = Upper Canning Estuary.

Terms	(a) Nearshore gobiid fauna					(c) Offshore gobiid fauna				
	df	MS	%MS	pF	*P*	df	MS	%MS	pF	*P*
Lunar month	30	8683	6.35	4.48	0.001	29	9876	13.33	4.66	0.001
Region	4	121330	88.70	62.56	0.001	3	57365	77.41	27.09	0.001
Lunar month × Region	119	4827	3.53	2.49	0.001	87	4751	6.41	2.24	0.001
Residual	1086	1939	1.42			840	2118	2.86		

	(b) Nearshore gobiid fauna					(d) Offshore gobiid fauna			
	LM	UM	LC	MS			LM	UM	LC
UM	7.14					UM	5.41		
LC	5.79	5.06				LC	5.61	3.86	
MS	8.86	10.60	5.50			MS	7.64	5.03	2.36
UC	10.09	12.40	7.10	3.39					

Table 4. Pearson's correlations coefficients (*r*) and significant values (*p*) between the density (individuals 500 m^{-2}) of abundant gobiid species and salinity and water temperature at sites in the (a) nearshore (*n* = 20) and (b) offshore waters (*n* = 16) of the Swan-Canning Estuary. Non-indigenous species are highlighted are in bold.

Species	Salinity		Temperature	
(a) Nearshore	*r*	*P*	*r*	*P*
Arenigobius bifrenatus	-0.953	< 0.001	0.718	< 0.001
Favonigobius punctatus	-0.933	< 0.001	0.809	< 0.001
Pseudogobius olorum	-0.844	< 0.001	0.707	< 0.001
(b) Offshore				
Acentrogobius pflaumii	**0.856**	**< 0.001**	**-0.800**	**< 0.001**
Arenigobius bifrenatus	-0.868	< 0.001	0.738	0.001
Pseudogobius olorum	-0.624	0.001	0.512	0.043