Original Article

The effect of targeted stocking on behaviour and space utilization of a released finfish

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Targeted stocking involves the release of fish directly into high-quality habitat; however, this is often time-consuming, expensive and difficult. Acoustically tagged hatchery-reared juvenile mulloway Argyrosomus japonicus were released in groups directly into deep-hole habitat preferred by wild conspecifics (targeted stocking), or in a non-targeted fashion near easily accessible sites that lacked high-quality habitats in the direct vicinity. Fish were tracked continuously, 24 h d⁻¹, for 5 d following release. Fish released in a targeted fashion showed lower mean activity rates (50% less movement) and occupied higher quality habitats than fish released in a non-targeted fashion. Fish released in a non-targeted fashion also used a greater number of smaller habitat patches. The implications for improvements in behaviour and habitat usage patterns for fish released in a targeted fashion, such as improved growth and survival, are discussed. Identifying and releasing fish directly into the species’ high-quality habitat may ultimately improve the success of stocking programs.

Keywords: acoustic telemetry, habitat use, mulloway, restocking, sea ranching, space use, stock enhancement.

Introduction

The depletion of fish populations is well documented, with 80% of the world’s fisheries fully or over-exploited (FAO, 2009). Traditional controls to limit harvest through restrictions on gear or fish fail to address potential limitations to recruitment in some species. Consequently, methods to address potential recruitment limitations and improve fisheries output are currently being developed, such as stock enhancement (Bell et al., 2008). Stock enhancement has improved fisheries for several heavily targeted and recruitment-limited species worldwide, including mullet Mugil cephalus (Leber et al., 1995, 1996), turbot Psetta maxima (Stottrup et al., 2002), and Japanese flounder Paralichthys olivaceus (Yamashita et al., 1994; Kellison et al., 2002). The cornerstone of these programs is the development of release strategies that consider those factors influencing post-release survival. For example, the quantity of fish released into a system is important, as density-dependent survival may be heavily influenced by wild recruitment (Brennan et al., 2008). Size-at-release can have a number of interactive effects on survival, such as timing the season of and size at release to match the size structure of wild cohorts (Leber et al., 1998). Furthermore, recent literature has shown it is important to ensure released fish have sufficient habitat to support growth and survival following release (Fairchild et al., 2005; Brennan et al., 2006; Ochwada-Doyle et al., 2009, 2010; Taylor et al., 2013).

Addressing habitat requirements and potential naivety to habitats available in natural systems is extremely important in the context of release strategies. The lack of habitat complexity in hatcheries can produce naïve fish that perform well under rearing conditions that do not occur in the wild (Lee and Berejikian, 2008). For example, hatchery-reared China rockfish Sebastes nebulosus showed no association with any habitat type, as opposed to complex rocky habitat used by wild conspecifics, although released fish adopted preferences for wild habitat within 14 d (Lee and Berejikian, 2009). A common approach to dealing with such behavioural deficits, especially for benthic fishes, is the use of acclimation cages that give released fish the opportunity to adapt to new habitats prior to exposure to predation (e.g. Brennan et al., 2006; Sparrevohn and Stottrup, 2007; Fairchild et al., 2008; Walsh et al., 2013). Walsh et al. (2013) found that conditioning hatchery-reared Japanese flounder Paralichthys olivaceus in cages led to improved performance of released fish. Such improvements in behavioural performance can lead to concomitant improvements in feeding and
survival, as shown by Sparrevohn and Støttrup (2007). Caging enclosures, however, are difficult to employ for acclimation of species that require deeper benthic habitats. In these situations, it is useful to evaluate relative performance of released fish across a range of habitats, to better understand how particular habitats might lead to improved behaviour following release.

Use of forage resources by released fish relies on both the ability of fish to find and recognize high-quality habitat, the capacity of that habitat to support additional recruits, and the ability of new recruits to exploit prey resources without exposing themselves to excessive predation risk. High-quality habitat is important for both predator evasion and optimal foraging in many species (Kneib, 1987; Haplin, 2000; Taylor et al., 2006b). Targeted stocking (TS) aims to maximize survival and minimize emigration by releasing fish directly into high-quality habitat, at a density that the habitat can support (Taylor and Suthers, 2008). Foraging arena theory shows that survival of vulnerable fishes is maximized where suitable refugia are available in the vicinity of prey resources (Walters and Juanes, 1993; Walters and Martell, 2004). Thus, targeting releases directly into niches at the habitat and microhabitat level should result in lower predation, lower emigration, and greater foraging efficiency (Fairchild et al., 2005). Conversely, when fish releases are not targeted in this fashion, released individuals must find their niche autonomously, which could result in high mortality as vulnerable fish remain exposed to predation. This is evident in near-complete mortality when fish are released outside of high-quality juvenile habitat (Leber and Arce, 1996; Taylor et al., 2009).

The relative effects of TS on fish behaviour and space utilization were investigated to indirectly evaluate potential benefits of this release strategy for juvenile mulloway. To achieve this, we used small releases of several groups of acoustically tagged mulloway Argyrosomus japonicus (Sciaenidae, Temmink & Schlegel) in a targeted and non-targeted strategy to address differences in (i) home ranges, (ii) activity rates, and (iii) association with high-quality habitat. Juvenile A. japonicus prefer deep holes in estuarine systems, where higher densities of their mysid and prawn prey occur (Taylor, 2008) and undulating bathymetry theoretically provides refuge from predation. We expected that fish released directly into habitat preferred by wild conspecifics would spend more time in the vicinity of predation refuge and have smaller ranges and lower activity rates.

Material and methods

Study species

Argyrosomus japonicus are a demersal predatory fish in the estuarine and coastal regions of southern Australia. Pilot A. japonicus releases have been undertaken within several Eastern Australian estuaries to enhance recreational fishing catch (Taylor and Piola, 2008; Taylor et al., 2009). Recent research in Australia revealed specific requirements of juveniles, including abundant peracaridan crustaceans, predominantly mysid shrimp (Taylor et al., 2006a), in the vicinity of structured and deep-hole habitat (Taylor et al., 2006b).

Study area

Experimental releases were conducted in the Georges River, NSW, Australia (Figure 1). The Georges River estuary contains abundant

Figure 1. Map of the study area in the Georges River, NSW. Regions of high-quality habitat (depth > 5 m) within the estuary are shaded. Two release sites were used for each treatment (targeted stocking, TS; and non-targeted stocking NTS).
habitat and prey species important for juvenile *A. japonicus*, but has historically received low numbers of early-juvenile recruits (Taylor et al., 2009). The tracking zone (4.4 km²) extended from Como Bridge (33.995°S 151.058°E) 12 km upstream to Sandy Point (33.975°S 150.996°E). This section of river was selected as it encompasses a mosaic of high-quality structured, deep-hole habitat (0.4 km²) and low-quality unvegetated, shallow habitat (4.0 km², Figure 1). Two release sites for each release treatment (TS; and non-targeted stocking, NTS) were selected based on habitat type (Figure 1).

**Tagging and experimental design**

Hatchery-reared *A. japonicus* (167.2 ± 7.6 mm total length, ~4 months of age) were produced at a government hatchery (Port Stephens Fisheries Institute, Taylors Beach, New South Wales) and transported to the study area in a 1300 l fish transporter on 19 January 2009 (during the last quarter of the lunar cycle). Fish were acclimated to the ambient physicochemical conditions at each site over 1 h prior to release. TS involved carefully transporting the fish in aerated holding tanks aboard a boat to deep-hole sites. NTS involved holding fish in aerated holding tanks, and releasing them into shallow water directly beside the boat ramp (as is often the procedure for routine releases). At each of the four sites, 30 fish were released, of which 4 (16 fish in total) had PT-01 ultrasonic transmitters attached (Sonotronics, Tucson AZ, USA). PT-01 transmitters are cylindrical (16 mm length, 7 mm diameter), have a total of 0.6 g in water, and emit a series of coded transmissions at frequencies from 71–79 kHz for 7 d before the battery expires. Transmitters were fixed to fish using a novel external attachment technique, where the acoustic tag was affixed to a short T-bar tag for attachment to the fish (Pursche et al., 2013). Briefly, fish were anaesthetized in a 15 mg l⁻¹Aquí-S (Aquí-S Ltd, New Zealand) solution in seawater, and total length (mm) and weight (g) were recorded. The external tag composite was injected into the dorsal side of the fish ~ 5 mm below the anterior dorsal fin ray with a tagging gun, and the T-bar was locked between the first and second pterygiophores. All fish were observed for 30 min in case of general distress (Pursche et al., 2009); if the fish was exhibiting normal behaviour, it was randomly allocated to the TS or NTS release treatment. Two transmitters failed to function, leaving a total of 14 tagged fish (Table 1).

Manual tracking commenced directly after release and was undertaken continuously for 120 h (5 d) using established methods (Taylor et al., 2006b). The tracking zone was sequentially scanned using a Sonotronics USR-96 receiver unit and headphones. Transmitted signals were followed until the signal was at greatest intensity, and then dramatically decreased, indicating the fish was directly beneath the vessel. The vessel was then turned around and the position confirmed and recorded using a handheld GPS unit (Garmin GPS72, WGS84 Datum). Each scan began at the upstream extent of the tracking zone.

Data were downloaded using Mapsource v. 6.15.7 (Garmin, Olathe, KS, USA) and imported into ArcGIS v. 9.3 (ESRI). For each fish, the distance between successive points was calculated directly, or using the centreline of the river to account for curvature of the river where necessary. Depth at each point was calculated by overlaying a high-resolution bathymetry map, and a minimum activity index (MAI, m⁻¹ h⁻¹) was calculated by dividing the distance between two points by the time elapsed between observations. Kernel density was calculated for home range using a 130 m search radius and an output resolution of 100 m² cells (Taylor et al., 2006b). Points were inverse weighted by time to give greater certainty of space utilization to more frequent observations, by allocating each value to a 2 h time category and weighting by the maximum value within that time divided by 120 (Silverman, 1986). The 95% (total) and 50% (core) space utilization contours were then calculated and presented as a series of contours on the map. A greater number of contours indicated a lower association with a particular area and, conversely, individuals that have fewer contours show greater association with the habitat patch. Differences in MAI were evaluated between release treatments (TS and NTS) and amongst days-post-release using a 2-factor ANOVA, with individual data pooled by release site. Differences in space utilization of fish released in TS and NTS treatments were assessed by comparing both the area within the 95% and 50% contours, and the number of discrete habitat patches for each group of fish released at replicate sites using single-factor ANOVA. Differences in the utilization of optimal habitat between release treatments was tested by calculating the proportion of high-quality habitat (Taylor et al., 2006b) within

<table>
<thead>
<tr>
<th>Release treatment</th>
<th>Fish ID</th>
<th>Tag freq. (kHz) and code</th>
<th>Release site</th>
<th>Total length (mm)</th>
<th>Utilization contour (m²)</th>
<th>50%</th>
<th>95%</th>
<th>Core areas</th>
<th>50:95% ratio</th>
<th>High-quality habitat used (%)</th>
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Release treatment refers to treatment groups and the release locations are outlined in Figure 1. The proportion of high-quality habitat within the core space utilization contour is indicated in the last column.
Despite this, there was no significant difference in the area within of habitat patches spread along a greater length of river (Figure 3). Released in the NTS treatment appeared to use a greater number from pooled treatment and individual data, and showed that fish release treatments, the core and total contours were recalculated compare overall space utilization along the tracking zone between release treatments. For example, in previous studies in Hawaiian bays, release site was as important in survival of juvenile Pacific threadfin Polydactylus sexfilis as other key factors, such as size-at-release and season-of-release (Leber et al., 1998). More importantly, the most effective size-at-release in one season could be the least effective in another, depending on the release site (Leber et al., 1998). Matching hatchery releases with size modes in wild cohorts can contribute to survival through a number of mechanisms (Leber et al., 1997), as this is conducive to reducing intraspecific predation. A solid understanding of habitat requirements and dynamics in wild juvenile fish provides a useful starting point for refining release strategies for hatchery-reared fish.

The benefits of directly addressing habitat requirements when developing release strategies have been demonstrated for a number of released species. Several examples show that high-quality habitats can provide for both optimal foraging and predator evasion (Fairchild et al., 2005; Ochwada-Doyle et al., 2009; Taylor and Ko, 2011). Similar dual benefits of high-quality habitat likely occur for mulloway in the Georges River (Taylor et al., 2009). Deeper sedimentary habitats within NSW riverine systems provide abundant decapod and peracaridan crustacean resources, such as penaeid prawns, amphipods and mysid shrimp (Taylor, 2008). These taxa form important prey for early juvenile mulloway, in particular mysid shrimp (Taylor et al., 2006a; Taylor and Suthers, 2008). Our original hypotheses and predictions were based on the premise that this dual benefit would manifest in fish released directly into high-quality habitat, and thus lead to detectable effects on their movements and space utilization. Behavioural data confirmed these differences, which have implications for the success of stocked individuals. Greater activity rates may reflect altered foraging efficiencies and predation risk in lower quality habitat. Higher activity rates, or greater emergence from predation refuge, may ultimately act to increase encounter rates with predators and consequently increase the risk of a predation event and associated mortality (Biro et al., 2006; and see Walsh et al., 2013 for a laboratory-based evaluation for Japanese flounder). Furthermore, improved access to important prey may contribute to elevated growth rates, which has been observed for mulloway previously at the estuary level (Taylor et al., 2009). Given these considerations it is possible that the lower activity rates, and greater utilization of high-quality habitat observed in this study, may contribute to concomitant improvements in post-release growth and survival, although more explicit experimental work is required to evaluate this.

Results

Activity rates

All tagged fish remained within the tracking zone for the duration of the tracking period, and a total of 220 observations were made with 2.94 ± 0.3 (mean ± standard error) observations fish⁻¹ d⁻¹. Generally, all tagged fish would be located within a single sweep of the tracking zone. MAI ranged from 0.8–536 m h⁻¹, and appeared to be highest <24 h post-release in both release treatments (Figure 2). Fish released using the NTS treatment displayed significantly greater MAI (F₁₁₀ = 7.230, p = 0.022); however, there were no differences in MAI detected amongst days post-release (F₄₁₁₀ = 2.482, p = 0.117), and there was no significant interaction term (F₄₁₁₀ = 0.271, p = 0.889).

Space and habitat utilization

Mean area within the core and total space utilization contours were 26193 ± 522 and 157490 ± 492 m² (mean ± standard error; Table 1). The greatest area within a total space utilization contour was 302000 m², for a fish in the NTS treatment. To visually compare overall space utilization along the tracking zone between release treatments, the core and total contours were recalculated from pooled treatment and individual data, and showed that fish released in the NTS treatment appeared to use a greater number of habitat patches spread along a greater length of river (Figure 3). Despite this, there was no significant difference in the area within the core (F₁₁₂ = 2.016, p = 0.181) and total (F₁₁₂ = 0.366, p = 0.557) space utilization contours between release treatments. There was, however, a difference in the pattern of space use between release treatments (Figure 4). Fish released in the NTS treatment used a significantly greater number of discrete habitat patches than those in the TS treatment (F₁₁₂ = 7.425, p = 0.018).

There were detectable differences between treatments in the magnitude of high-quality habitat encompassed within the space utilization contours, with a significantly greater proportion of high-quality habitat within the core utilization contours for fish released within the targeted treatment, relative to the non-targeted treatment (F₁₁₂ = 5.635, p = 0.035, Figure 4).

Discussion

This experiment showed that naïve juvenile fish released directly into high-quality habitat have significantly different patterns of movement and habitat utilization to those released in a non-targeted fashion. Juvenile A. japonicus released using a targeted method occupied fewer discrete habitat patches, and also spent a larger proportion of their time in high-quality habitat. The use of fewer discrete habitat patches reflects behaviour observed in wild A. japonicus, who usually concentrate movement in a single core area (Taylor et al., 2006b), and this may imply that fish are successfully finding prey resources. Fish released in a non-targeted fashion had greater activity rates, used a greater number of habitat patches, and displayed lower usage of high-quality habitat.

Previous literature indicates that release of fish directly into an appropriate habitat can have several advantages for post-release survival. For example, in previous studies in Hawaiian bays, release site was as important in survival of juvenile Pacific threadfin Polydactylus sexfilis as other key factors, such as size-at-release and season-of-release (Leber et al., 1998). More importantly, the most effective size-at-release in one season could be the least effective in another, depending on the release site (Leber et al., 1998). Matching hatchery releases with size modes in wild cohorts can contribute to survival through a number of mechanisms (Leber et al., 1997), as this is conducive to reducing intraspecific predation. A solid understanding of habitat requirements and dynamics in wild juvenile fish provides a useful starting point for refining release strategies for hatchery-reared fish.

The benefits of directly addressing habitat requirements when developing release strategies have been demonstrated for a number of released species. Several examples show that high-quality habitats can provide for both optimal foraging and predator evasion (Fairchild et al., 2005; Ochwada-Doyle et al., 2009; Taylor and Ko, 2011). Similar dual benefits of high-quality habitat likely occur for mulloway in the Georges River (Taylor et al., 2009). Deeper sedimentary habitats within NSW riverine systems provide abundant decapod and peracaridan crustacean resources, such as penaeid prawns, amphipods and mysid shrimp (Taylor, 2008). These taxa form important prey for early juvenile mulloway, in particular mysid shrimp (Taylor et al., 2006a; Taylor and Suthers, 2008). Our original hypotheses and predictions were based on the premise that this dual benefit would manifest in fish released directly into high-quality habitat, and thus lead to detectable effects on their movements and space utilization. Behavioural data confirmed these differences, which have implications for the success of stocked individuals. Greater activity rates may reflect altered foraging efficiencies and predation risk in lower quality habitat. Higher activity rates, or greater emergence from predation refuge, may ultimately act to increase encounter rates with predators and consequently increase the risk of a predation event and associated mortality (Biro et al., 2006; and see Walsh et al., 2013 for a laboratory-based evaluation for Japanese flounder). Furthermore, improved access to important prey may contribute to elevated growth rates, which has been observed for mulloway previously at the estuary level (Taylor et al., 2009). Given these considerations it is possible that the lower activity rates, and greater utilization of high-quality habitat observed in this study, may contribute to concomitant improvements in post-release growth and survival, although more explicit experimental work is required to evaluate this.
Figure 3. Home ranges of juvenile *A. japonicus* released through treatments (a) targeted stocking (TS, including sites 2 and 3), and (b) non-targeted stocking (NTS, including sites 1 and 4). Kernels are grouped between release sites for each treatment for display. The smaller quantity of 95% patches for the fish at (a) is reflective of smaller movements and higher association with particular habitat patches (see Figure 4).
The potential for non-optimal behaviour in hatchery-reared fish following release may arise as a result of the lack of natural stimuli (e.g., predators, prey and habitat) within the hatchery environment. This is frequently discussed in the literature, both in terms of the period of time taken for hatchery-reared fish to adopt the same behavioural traits observed in wild conspecifics (e.g. Sparrevohn and Stöttrup, 2007; Lee and Berejikian, 2009), and specific prerelease protocols that may be employed to alleviate adverse effects on growth and survival (e.g. Brown and Laland, 2001; Brown et al., 2003; Fairchild et al., 2010; Walsh et al., 2013). For example, common snook Centropomus undecimalis showed improved post-release survival after being held in predator-free enclosures at the site of release for 3 d, conceivably by providing an arena where release survival after being held in predator-free enclosures at the site of release for 3 d, conceivably by providing an arena where hatchery-reared organisms can learn how to forage on live organisms, and can recover from the stress and disorientation associated with transport and stocking (Brennan et al., 2006). For benthic winter flounder raised in sediment-deficient tanks, fish took two days to develop burying skills and up to 90 days for colour adaptation (Fairchild and Howell, 2004). In situ cages were employed to allow hatchery-reared winter flounder to develop these cryptic skills without the risk of predation; however, the presence of release cages were shown to attract predators to the release site (Fairchild et al., 2008). The undulating and deep bathymetry preferred by juvenile mulloway, and the strong tidal currents in riverine systems within NSW make the deployment of acclimation cages impractical, but the practice of targeting releases directly into high-quality habitat that provides appropriate refugia may produce a similar acclimation effect in released mulloway.

This study shows that acoustic telemetry is a useful tool for measuring the immediate post-release behaviour of small hatchery-reared juvenile fish. Acoustic tracking allows real-time assessment of habitat use post-release, as well as relative activity levels. The decreasing size of acoustic tags means that fish closer to the actual size-at-release normally employed in stocking programs can be monitored using this method, and novel automated tracking systems (such as the Vemco Positioning System, Espinoza et al., 2011) can yield data at an even higher temporal and spatial resolution than that obtained in this study. Concomitant acoustic telemetry, dietary, and mark–recapture studies would provide an optimal mix of movement, feeding, growth and survival information to evaluate release strategies, and this should be considered in future release experiments.

Building an effective stock enhancement program requires determining the effects of stocking alongside the development of release strategies and appraisal of economic benefits arising through stocking. The current research endeavoured to synthesize knowledge of wild A. japonicus ecology to inform release strategies, and evaluate potential benefits of such strategies. This experiment shows that TS may lead to improved behavioural performance of stock mulloway, which could manifest in improved growth and/or survival. Ultimately, the relationships between release sites and techniques and post-release growth and survival require explicit testing, and this is an area for further work. This case study provides a template for the development and evaluation of release strategies for stocked fish through the application of novel technology to monitoring fish behaviour post-release. Applying such techniques in the development phase of stock enhancement programs will ultimately lead to improved strategies and outcomes from fish releases in the future.

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