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ORIGINAL PAPER

Modifications to the bottomless lift net for sampling nekton in tidal mangrove forests

Carole C. McIvor · Noah L. Silverman

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Abstract Sampling fishes in vegetated intertidal wetlands is logistically challenging. We modified the $2 \times 3\text{-m}^2$ bottomless lift net developed for sampling nekton (fish and decapod crustaceans) on the surface of salt marshes for use in tidal mangrove forests with a woody (as opposed to herbaceous) underground root system. As originally designed (Rozas, Mar Ecol Prog Ser 89:287–292, 1992), the lift net was buried directly in the marsh substrate. The net was raised at slack high tide thereby encircling nekton within the enclosed area. A chain-line on the net bottom prevented escape under the net once deployed. However, when we used this same design in tidal mangrove forests, the extensive woody roots and occasional slumping sediments resulted in uneven trenches that could not be cleared effectively during sample recovery. We made 3 modifications to the original net design: (i) lined the peat trenches with aluminum channels of uniform width and depth; (ii) replaced the previous chain-line with Velcro closures that directly attached the net to the inner face of the outer wall of the aluminum channel; and (iii) removed the subtidal pan previously used for concentrating the enclosed nekton at low tide, and filled in those depressions with on-site peat. In the modified version, the aluminum trench became the only subtidal refuge available to nekton, and it was from here that we collected the sample after the forest drained. These modifications permitted high clearing efficiency (93–100%) of fin-clipped individuals of two common species of estuarine resident fishes, *Kryptolebias marmoratus* (mangrove rivulus) and *Bathygobius soporator* (frillfin goby). Additionally, the density estimates of grass shrimp (*Palaemonetes* spp.) increased 10-fold post-modification.

Keywords Clearing efficiency · Everglades National Park · Modified lift net · Nekton · Riverine mangroves · Vegetated intertidal zone

Introduction

Flooded mangrove forests and tidal streams often contain high standing stocks of resident and transient nekton (fish and decapod crustaceans), some of which are of recreational or commercial importance (Odum et al. 1982; Robertson and Duke 1990). Intertidal mangrove forests present considerable challenges to

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C. C. McIvor (✉)
US Geological Survey, Southeastern Ecological Science
Center, 600 Fourth Street South, St. Petersburg,
FL 33715, USA
e-mail: carole_mcivor@usgs.gov

N. L. Silverman
ETI Professionals, 4902 Eisenhower Blvd, Suite 150,
Tampa, FL 33634, USA

Present Address:
N. L. Silverman
National Marine Fisheries Service, Southeast Regional
Office, 263 13th Avenue South, St. Petersburg,
FL 33701, USA

obtaining quantitative estimates of nekton density and biomass. Woody trunks, prop or buttress roots, pneumatophores, and subsurface roots make net deployment in all but the most open "park-like" mangroves extremely difficult. In southwest Florida, USA, riverine and fringing mangroves are generally flooded to at least a few tens of centimeters by twice-daily mixed semi-diurnal tides. In the dwarf mangroves of the south Florida mainland adjacent to Florida Bay in contrast, water levels are driven primarily by rainfall-driven water cycles coupled with seasonal changes in mean sea level (Provost 1974; McIvor et al. 1994). Thus, these two portions of south Florida impose different requirements for sampling within the respective mangrove forests. A 9-m^2 drop net, actively scooped following application of the fish toxicant rotenone, is both appropriate and effective in dwarf forests with poorly consolidated marl sediments (Lorenz et al. 1997), but the same drop net performs poorly in the forests of southwest Florida (McIvor unpublished).

Meter-squared throw traps have been routinely used to estimate small-fish (<10-cm SL) density in both upstream herbaceous freshwater marshes (Loftus and Eklund 1994) and in downstream shallow seagrass beds (Browder et al. 1999) within the Greater Everglades Ecosystem (GEE). However, it is impossible to use throw traps in dense mangrove forests with a closed canopy: there are too many impediments to its falling properly. We needed a quantitative method to develop estimates of nekton density in the extensive closed-canopy tidal mangrove forests lining Shark River, the estuarine portion of the major drainage system in the GEE. Such estimates are required to establish baseline conditions with which to judge success following the Everglades restoration efforts currently underway (http://www.evergladesplan.org). To meet this sampling need, we tested and modified the 2 × 3-m^2 bottomless lift net (Rozas 1992), originally developed for use on the surface of intertidal salt marshes, for use in the riverine mangrove forests along Shark River.

Materials and methods

Study sites

We sampled nekton from the flooded intertidal mangrove-forest floor at three locations along a salinity gradient on Shark River, Everglades National Park Florida, USA, from March 2001–April 2007. The Shark River arises as diffuse sheetflow in the grassy freshwater marshes of the GEE. Flow first becomes channelized about 15 km upstream of Tarpon Bay, a wide shallow embayment near the upstream limit of tidal influence. From upstream to down, our sampling locations were an unnamed first-order tributary to Tarpon Bay (at N25°24.563 W80°57.868, 19 km from the river mouth), mid-way on the Harney River (at N25°25.405 W81°03.597, 11.2 km from the mouth), and 6 km upstream of Ponce de Leon Bay, the mouth of Shark River (N25°21.895 W81°04.633). (Our upstream and downstream locations correspond to sites SRS-4 and SRS-6 respectively of the Florida Coastal Everglades Long Term Ecological Research network. Available at http://fce.lternet.edu/.)

Red mangroves (*Rhizophora mangle*) visually dominate the riverine forests along the entire portion of the river we sampled. Whereas *Rhizophora* was the primary mangrove species downriver (*Avicennia germinans* (black mangrove) was also present), forests at the upriver location also contained *Laguncularia racemosa (white mangrove)*, *Conocarpus erectus (buttonwood),* and *Cladium jamaicense (sawgrass)*. At the mid-river site, *Juncus* spp. formed a sparse groundcover under a mangrove canopy that included both *Laguncularia* and *Rhizophora*. During the 7 years of sampling, salinity averaged six psu upriver, 17 psu mid-river, and 26 psu downriver. Flooding depths at net set (slack high on spring tides) averaged 10 cm at the up- and mid-river locations and 23 cm at the downriver location. Tidal range at the mouth of Shark River is approximately 1.5 m. The width of riverine forest that routinely flooded narrowed with distance upstream as the tidal range was attenuated. We adjusted the placement of the bottomless lift nets to the different flooding pattern at each site; thus, lift nets were located at varying distances from the channel. Nets were within 1 m of the channel upstream, 9 m mid-river, and 16 m downriver.

Bottomless lift net as originally designed

Rozas (1992) designed a 2 × 3-m^2 bottomless lift net for sampling nekton within the *vegetated intertidal zone* of herbaceous salt marshes. The net was deployed at slack high tide by lifting the net remotely from its buried position within a shallow trench in the substrate. Burial in the substrate is necessary to avoid artifacts due to the presence of the net within the flooded wetland. If placed immediately on top of the forest floor, the net's presence would interfere with normal movements of the nekton and lead to significant sampling biases. This passive sampling device was reliant on the ebbing tide to concentrate nekton entrained by the net walls into a plastic pan buried flush with the substrate. This pan acted as a subtidal reservoir. The pan was removed when the marsh had drained, and the sample was poured into a collection net. Rozas (1992) also cautioned to inspect the unlined trenches for additional nekton before collapsing the net into the trenches for storage in the marsh between sampling events.

The bottomless lift net was constructed of 3-mm Delta mesh netting with two sleeves, the top one containing PVC pipe and nylon line for raising the net, and the bottom one containing galvanized chain to prevent nekton escaping under the net. This fixed-site, passive-enclosure net has been used in herbaceous marshes spanning the salinity range (Osgood et al. 2003), in a shallow intertidal unvegetated sand flat immediately adjacent to mangroves (Ellis and Bell 2004), and within a riverine mangrove forest in subtropical Australia (Meager et al. 2003). After installing and sampling nine such nets in intertidal mangrove forests along the Shark River in southwest Florida, we found that use of the net in densely forested habitats necessitated design modifications for optimal recovery of entrained nekton.

Mangrove-associated constraints on use of the bottomless lift net

The extensive subsurface woody roots of mangrove trees made initial trench preparation difficult and time consuming but not impossible. A team installed on average 3 nets/day not including boardwalks. Cut ends of tree roots in the trenches, however, constituted a major impediment at clearing, causing our clearing device (standard aquarium dip net) to bind up, skip, or bend. Nekton abundance in trenches often exceeded that in subtidal pans at our sites; thus, repetitively scooping trenches became part of the routine clearing procedure. With monthly sampling in year 1, we also observed "investigator effects," i.e., trampling immediately adjacent to the net that resulted in slumping of peat and sediment into the sampling trench. Initial limited efficiency estimates for recapture of fin-clipped *Kryptolebias marmoratus* averaged 50% (five of 10 fish over three nets), a relatively low recovery rate when compared to that achieved with the same net design in salt marshes: 39% for *Palaemonetes pugio* Holthuis, 58–93% for four fish species (Rozas 1992). We knew our total clearing procedure was inadequate, and suspected nekton loss at clearing within the uneven trenches, and secondarily under the subtidal pan. We thus chose to capitalize on a design weakness (necessity of a trench) and turn it into an asset. This modification was a three-part process that involved; (i) lining the existing sediment trenches with aluminum channel; (ii) physically attaching the bottom of the net to the inner face of the outer wall of the aluminum channel (henceforth termed aluminum trenches); and (iii) dispensing with the chain line and subtidal pan altogether thereby making the aluminum trenches the *only* subtidal refuge.

Net construction

We made our aluminum trenches in a machine shop from rectangular aluminum channel (6063-T52) with 1/8-inch (3-mm) walls (available from Eastern Metals, 3 by 4 inch OD (7.6 by 10 cm)). We cut this aluminum (shipped in 24-ft lengths) on a 45-degree angle into 2- and 3-m lengths, the dimensions of our bottomless lift nets. We removed the tops (one of the 10-cm sides) of the cut aluminum channel with a table saw and saved this portion to make removable lids for the fabricated trenches (see below). We applied 2-inch (5-cm) Velcro to the inner face of the outermost side of each piece of aluminum channel and reinforced the adhesive backing with pop rivets. We also haphazardly drilled 3-mm holes in the bottoms of the aluminum channels to permit drainage on low tides and to prevent hydraulic pressure within the sediments from raising the trenches above substrate level. The only modification to the actual net was to replace the bottom sleeve containing galvanized steel chain with the matching half of the Velcro.

In the field, we assembled the four sides of the aluminum trenches with custom aluminum brackets attached on the *outside bottom of the frame* to retain the smooth interior surface, and we set the entire frame into the existing sediment trench with the top of the aluminum trench flush with the forest floor. Next the lift net (chain-line previously removed and replaced with Velcro) was carefully affixed by joining the two Velcro haves at the inner face of the outer wall of the aluminum trench to ensure no gaps or pockets existed. We removed the subtidal pans and filled in the resultant depression with peat cut from elsewhere in the forest. We now had perfectly even trench bottom and sides from which nekton could be efficiently cleared. To minimize "ghost-fishing" between sampling events, we used lids fabricated by affixing 1- by 4-inch (2.5- by 10-cm) pressure-treated wood to the matching aluminum tops that were removed during construction in the metal-working shop. Lids of aluminum alone failed to stay in place between sampling events, whereas addition of wood blocks to the underside of the aluminum lids permitted one to press the wooden portion of the lid securely into the aluminum trench.

Sampling protocol

We reached sampling sites by raised plank boardwalks, as per Rozas (1992). Boardwalks minimized habitat degradation from walking in the wetland, and provided safer transit for personnel. We visited lift nets on the low tide prior to the designated spring tide to be sampled for routine maintenance and to clear the trenches of organisms, detritus, and sediments that had settled there since the last trip. We saved all material scooped from the trenches and returned it to the forest well away from the lift nets. This step was necessary to avoid artificially elevated abundance estimates from organisms that recruited to the trenches between sampling events. We often found small *Uca* spp. and *K. marmoratus* (formerly *Rivulus marmoratus*) in trenches. [Newly recruited individuals of these two taxa had likely gained entry to the trenches through very small (<3 mm) crevices between abutting lids.]

Deployment remained unchanged from the method of Rozas (1992). Briefly, two people quietly approached the buried net via raised boardwalks and simultaneously pulled on nylon lines attached to the PVC frame to raise the net from a 3- to 5-m distance (Fig. 1a, b). Lines were conveniently tied off to trees, thereby holding the net upright. Clearing of aluminum trenches on the next slack low tide followed (Fig. 1c). Two people placed the dip nets back to back in the trench and steadily moved around the 10-m perimeter to meet on the opposite side. We repeated this procedure three times. Following preservation of the sample, we lowered the net into its aluminum trench and stored it in situ, capped by a lid, until the next sampling event.

Estimates of clearing efficiency and net effectiveness

We determined the clearing efficiency of our modified lift net from mark/recapture estimates of fin-clipped fish of two of the most common species captured over the course of this study, mangrove rivulus (*K. marmoratus)* and frillfin goby (*B. soporator)*. Sample size (N of fish) used in these tests was 85 *K. marmoratus* and nine *B. soporator.* As we had 9 lift nets (3 replicates in each of 3 locations along the river), we placed 9–10 previously-captured fin-clipped *K. marmoratus* in each net immediately following net deployment on a trip dedicated to estimating clearing efficiency. With fewer *B. soporator*, we placed 3 fish in each of 3 replicate nets at the downriver location. In all cases, we held fin-clipped fish (a small piece of dorsal portion of caudal fin removed) up to 24-h in aquaria or bait buckets prior to release, and used them only in the portion of the salinity gradient in which they had been captured. On the low ride following release of marked fish, we cleared the nets as usual and recorded the number of fin-clipped individuals recovered.

A second means of assessing net effectiveness was to compare density estimates from routine collections pre-and post-modification. Pre-modification we sampled 63 nets on 8 dates (Jan 2001–Dec 2002). We chose a comparable sample (63 nets in 8 sampling events) post-modification (Feb 2003–April 2005). For both time periods we calculated three metrics, the average density of the two most abundant taxa: (1) *K. marmoratus*; (2) *Palaemonetes* spp.; and (3) all nekton (excluding clupeids, engraulids and atherinids which are "strays" from an open-water habitat). We used samples from all three locations along the river.

Statistical analysis

For purposes of this paper (gear description), we report nekton counts and biomass summed across all locations, nets and times: variance due to spatial and temporal factors will be reported in a separate paper. Estimates of clearing efficiency (percentage of marked fish recovered in routine net clearing) are simple

Fig. 1 **a** Bottomless lift net buried in the peat flush with the forest floor; arrows indicate net perimeter; **b** Net rises from the trench through the water column as it is pulled from 3–5 m away with lines attached to the PVC-lined sleeves on top of the short (2-m) sides of the net; **c** View inside the deployed net. Note the even dimensions of the aluminum trench, indicated by an *arrow*. There is minimal chance of nekton escape under the net perimeter because Velcro holds the bottom of the net securely to the aluminum wall of the trench

Table 1 Abundance of nekton collected in modified 6-m^2 bottomless lift nets in riverine mangrove forests, Shark River, Everglades National Park, 2001–2007

Species	Common name	Count	Percent (%) abundance	Cumulative % abundance	Biomass (g)
Kryptolebias marmoratus Poey	Mangrove rivulus	450	55.1	55.1	126.3
Palaemonid spp.	Grass shrimp	165	20.2	75.4	16.1
Eucinostomus spp.	Mojarra	37	4.5	79.9	50.1
Bathygobius soporator (Valenciennes)	Frillfin goby	29	3.6	83.5	82.1
Poecilia latipinna (Lesueur)	Sailfin molly	25	3.1	86.5	15.8
Gambusia holbrooki Girard	Eastern mosquitofish	24	2.9	89.5	4
Lophogobius cyprinoides (Pallas)	Crested goby	15	1.8	91.3	29.6
Lucania parva (Baird & Girard)	Rainwater killifish	15	1.8	93.1	4.6
Fundulus grandis Baird & Girard	Gulf killifish	10	1.2	94.4	22.6
Anchoa mitchilli (Valenciennes)	Bay anchovy	8	1	95.3	2.1
Eucinostomus gula (Quoy & Gaimard)	Silver jenny	7	0.9	96.2	25.6
Callinectes sapidus Rathbun	Blue crab	6	0.7	96.9	63.1
Penaeid spp.	Penaeid shrimp	5	0.6	97.5	1.7
Fundulus confluentus Goode & Bean	Marsh killifish	4	0.5	98	1.6
Fundulus spp.	Killifish	4	0.5	98.5	0.4
Cichlasoma urophthalmus (Gunther)	Mayan cichlid	3	0.4	98.9	15.4
Belonesox belizanus Kner	Pike killifish	2	0.2	99.1	4.3
Eucinostomus harengulus Goode & Bean	Tidewater mojarra	2	0.2	99.4	8.5
Ctenogobius smaragdus (Valenciennes)	Emerald goby	1	0.1	99.5	0.3
Cyprinodon variegatus Lacepede	Sheepshead minnow	1	0.1	99.6	0.1
Gobiosoma bosc (Lacepede)	Naked goby	1	0.1	99.8	0.2
Procambarus fallax Fagen	Slough crayfish	1	0.1	99.9	1
Strongylura notata (Poey)	Redfin needlefish	1	0.1	100	2.6
Total nekton captured		816			478.1
N of nets		189			189
Average nekton/net		4.3			2.5

averages reported on a species basis with the individual fish considered as the unit of replication. To compare pre- and post-modification density estimates, we used repeated measures analysis of variance (RM ANOVA, SAS 9.1) with individual nets as the unit of replication, one analysis of each of three metrics. Density data failed to meet the assumption of normality despite log transformation. Given the relatively robust sample size (63 nets) within each of the two time periods and the robustness of ANOVA to departures from normality (Zar 1999), we feel this use of a parametric test is justified. Repeated measures analysis was appropriate because nets were fixed in space and sampled repeatedly over time (Winer 1971).

Results

We captured 816 individuals from 21 taxa of nekton in 189 net deployments over 7 years (Table 1). Dominance was high: six species made up 90% of the catch, and two species made up 75%. This forest assemblage was clearly dominated numerically by mangrove killifish (*K. marmoratus)* and grass shrimp (*Palaemonetes* spp.).

Recapture of fin-clipped *K. marmoratus* exceeded 93% across nine nets (79 of 85 fish recovered). This 93% recovery rate following net modification compares with a 50% pre-modification recovery estimate from a smaller effort (10 *K. marmoratus* in three nets). Following modification, recovery of

B. soporator was 100% (nine of nine recaptured): we have no pre-modification estimates for this species.

For matched sample sizes (63 nets) pre- and post-modification, there was no significant difference in either total nekton density (mean ± 1 SE) (RM ANOVA, 4.07 ± 0.65 vs. 5.76 ± 1.04/net, $P \geq 0.325$) or *K. marmoratus* density (RM ANOVA, 2.87 ± 0.59 vs. 2.83 ± 0.49/net, $P \geq 0.644$). However, the density estimates of *Palaemonetes* spp. increased post-modification by a factor of 10 (RM ANOVA, from 0.14 ± 0.06/net to 1.9 ± 0.76/net, $P \leq 0.020$), an indication that the modified net was an improvement over the original design for this species.

Discussion

Pronounced dominance by relatively few taxa is a common feature of many tidal wetland fish assemblages (e.g., Crona and Rönnbäck 2007; Kneib 2000; Robertson and Duke 1990; Rozas et al. 2007). The small-fish assemblage we sampled in riverine mangroves along the subtropical Shark River in south Florida provides a heightened example of this phenomenon. What is most unusual about this assemblage however is its dominance by *K. marmoratus*, a species of special concern not commonly captured in Everglades National Park (Taylor and Snelson 1992; Taylor 2000), nor sampled quantitatively elsewhere. This species is physiologically specialized for an intertidal habitat having the capability of emersion and cutaneous respiration (Abel et al. 1987; Taylor 2000). *K. marmoratus* has been observed in crab burrows as well as under damp leaves and logs, and remains in the forest over the low-tide cycle (Taylor 2000). Thus ours is the first published report of the systematic occurrence of *K. marmoratus* as part of a larger quantitative assemblage-level investigation. We found mangrove killifish to be common in intertidal forests along Shark River when the nekton assemblage was sampled with a method appropriate for this highly structured habitat (Fig. 1). (*K. marmoratus* also dominates the hydrologically-modified, mosquito-ditched mangrove forests of a preserve on Tampa Bay in west-central Florida (Richards et al. in revision).)

Addition of aluminum trenches was the primary modification to the bottomless lift net design. These trenches: (i) added structural integrity to sediments around the net perimeter thereby reducing investigator effects of trampling from repeat visits; (ii) acted as a barrier to growth of mangrove roots into the trenches; and (iii) prevented slumping of peat and sediments into the trench area that was crucial for both net storage and provision of a subtidal refuge for nekton pending sample collection.

We attribute the high post-modification recovery rates of fin-clipped fish to this addition of aluminum trenches to the original net design. A custom scoop net with a sturdy frame of 6-mm solid aluminum stock was highly effective in the smooth trenches with constant cross-sections, much more so than an aquarium dip net in the former bare-sediment trenches. Further, physically attaching the net bottom to the aluminum trench with Velcro prevented escape of nekton through gaps where the net and chain-line formerly met the substratum. We thus perfected an existing enclosure net design (Rozas 1992) for use within tidal mangrove forests and demonstrated that it could be cleared with high efficiency.

It is possible that the presence of aluminum trenches (and buried net) could either artificially attract or deter nekton from the immediate area. We have no direct test of net bias but offer the following observations. Prior to sampling, we scooped trenches and returned the mix of detritus, sediment, and entrained organisms to the mangrove forest well away from the net. We have observed that both small fiddler crabs (*Uca* spp.), which we do not sample, and *K. marmoratus* occur in the trenches following one or more month's hiatus in sampling despite having lids covering the trenches. By removing *K. marmoratus* from the trench portion of the 6-m^2 enclosed area of the net prior to sampling, we likely underestimated their abundance: thus, our estimates of *K. marmoratus* density are possibly conservative. Deterrence of nekton by aluminum trenches is unlikely given that neither of the two most abundant taxa (*K. marmoratus, Palaemonetes* spp.) declined in density post-modification.

Because ours is an enclosure net, the device is quantitative and yields density estimates as opposed to simply catch-per-unit-effort (Rozas and Minello 1997). The sample area, however, is relatively small (6 m^2) because swiftly lifting a larger water-saturated net through the water column would be logistically difficult. Thus using this device that samples 6 m^2

requires nekton densities adequate to generate sufficient non-zero samples for statistical analysis.

This modified lift net is somewhat expensive to install as pressure-treated wood (for raised boardwalks) and rectangular aluminum channels can both be costly. Construction of aluminum trenches also requires access to a machine shop. When a sampling project is known to be long term, however, the cost-benefit ratio would likely be favorable. Post-installation, little maintenance is required. Nine sites with aluminum trenches and attached netting have been in the field for 6 years in the forests along Shark River with no maintenance to trenches and only minor mending of other net parts.

The fixed-site requirement of this net constrains experimental designs to repeated measures analyses and does not permit stratified random designs. As a passive device, its use is dependent on predictable water movement, in this case a falling lunar tide, to concentrate nekton into a subtidal refuge. This is a requirement common to all passive-enclosure nets or traps used in intertidal habitats. Thus, use of this modified lift net is appropriate for tidal mangroves but not for non-tidal mangroves, e.g., the dwarf mangroves of the south Florida mainland adjacent to Florida Bay. Water levels in these latter forests are driven primarily by rainfall-driven water cycles coupled with seasonal changes in mean sea level (McIvor et al. 1994). Synoptic weather events that unpredictably decrease or elevate the level of the predicted tide such that the forest either does not flood or does not drain can also disrupt planned sampling when the investigator is solely dependent on tide tables and without real-time tide data.

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