

Correlation of otolith to fish size of the invasive lionfish *Pterois volitans* near St. Kitts and Nevis, West Indies. An initial bio-survey and pilot study

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Abstract. In the first attempt to develop a local lionfish (*Pterois volitans*) population model for St. Kitts and Nevis, 105 specimens were harvested to inform and guide future management, disease events, environmental incidents, and planned targeted culling of this invasive species. Lionfish have a well-documented association with destruction of marine biodiversity and therefore these data were the logical first step toward a better understanding of lionfish on a regional level. Through the process of gathering this pilot study data, it was determined that the mass and length of lionfish otoliths was strongly correlated to the length and mass of the lionfish they came from. Otoliths are resistant to the processes of digestion in the cases of predation and consequently these conclusions will be useful for future marine food-web dynamics studies in the region.

Key Words: Caribbean, fisheries, marine conservation.

Introduction. Since its initial sighting off the coast of Florida in 1985 (Morris Jr. et al 2012), the invasion of the Pacific lionfish (*Pterois volitans*) has been one of the most rapid (Albins 2013) and important threats to biodiversity in the region's history (Sutherland et al 2017). Lionfish are ravenous carnivores, eating small reef fish and crustaceans at a rate approximately three times that of an equally sized native predator (Albins 2013), resulting in loss of local fish populations (Green et al 2012), destruction of marine biodiversity (Albins 2013; Malpica-Cruz et al 2021), and threatening local fisheries (Chagaris et al 2015). This invader's superior efficiency over that of native predators is likely accounted for by the early age of maturity, and fast growth rate (Bustos-Montes et al 2020). Lionfish can become sexually mature in less than one year, and spawn without adherence to seasonality, resulting in up to two million eggs produced per female per year (Morris Jr. & Whitfield 2009). Furthermore, lionfish thrive in an impressive and diverse water column ranging in depths of a few meters to over 300 meters (Morris Jr. et al 2012), and seem to be resistant to many native parasites and infections (Morris Jr. & Whitfield 2009). These facts make the eradication of lionfish virtually impossible, and management even more vital.

The first step in controlling any invasive species is to gain understanding of its population density and dynamics, so that its impact on local ecology and fishery economics can be determined, and population control methods can be informed (Switzer et al 2015). Attaining current population and distribution records is vital to understanding specific population thresholds, beneath which, lionfish damages might be mitigated (Green et al 2012).

There is limited evidence for the correlation between the predation of lionfish and slowed or reduced lionfish invasion of Caribbean waters (Hackerott et al 2013). Nonetheless, predator prey relationships are valuable to understanding lionfish impact on local marine ecology. In a theory previously described as "biotic resistance", a healthier native predator population may offer a superior defense to lionfish colonization and

overpopulation (Levine et al 2004; Richardson & Pyšek 2007; Mumby et al 2011; Beaury et al 2020). Predators including some sharks (Handwerk 2012; Diller et al 2014), eels (Jud et al 2011), and groupers (Maljković et al 2008; Diller et al 2014) are known to eat lionfish based on evidence of postmortem stomach contents.

Unfortunately, prey fish are often difficult to study once consumed due to the obvious destructive processes of digestion. However, the sagittal otoliths of fish are relatively tolerant to digestion (Battaglia et al 2010). Fish have 3 sets of these structures which are vital to hearing and balance (Schulz-Mirbach et al 2019). They grow in size by the addition of minerals in annular ring formations and therefore record a life history for each fish (Belchier et al 2004; Hoffman & Harding 2018). These characteristics make otoliths one of the most utilized tissues for studying prey fish and understanding predator diets (especially the sagitta which is the largest of the three otoliths) (Granadeiro & Silva 2000; Kasapoglu & Duzgunes 2013; Zan et al 2015; Hanson & Stafford 2017). Like for many other oceanic fish (Battaglia et al 2010; Zan et al 2015), the size of lionfish can be estimated from otolith length and weight (Perera & Puerto 2016). Therefore, studies of potential predators of lionfish may elucidate vital information about lionfish as prey, along with their energy contribution to the predator's diet (Hernandez-Milian et al 2015). These data are the foundation of food-web dynamics research in marine ecology (Koen-Alonso & Yodzis 2005; Battaglia et al 2015).

The overarching goal of collecting lionfish specimens in this study was to create the first ever population survey of lionfish near the islands of St. Kitts and Nevis. However, the focus of this manuscript is on a subset of data aiming to better understand the correlation between lionfish otolith size (length and weight), and fish size (length and weight). Although data associated with otoliths of fish have been utilized in the study of many other marine species (Kasapoglu & Duzgunes 2013; Giménez et al 2016), to the authors' knowledge, data specific to lionfish otoliths of this nature has been documented in only one other population near the Alacranes reef in the southern Gulf of Mexico (Perera & Puerto 2016), and has never been substantiated elsewhere in Caribbean or Atlantic waters.

Material and Method. Data was collected from 105 lionfish, over an 8-month timeframe (between November 2019 and July 2021), across 5 geographic locations of the Caribbean waters southwest of St. Kitts and Nevis (Figure 1).

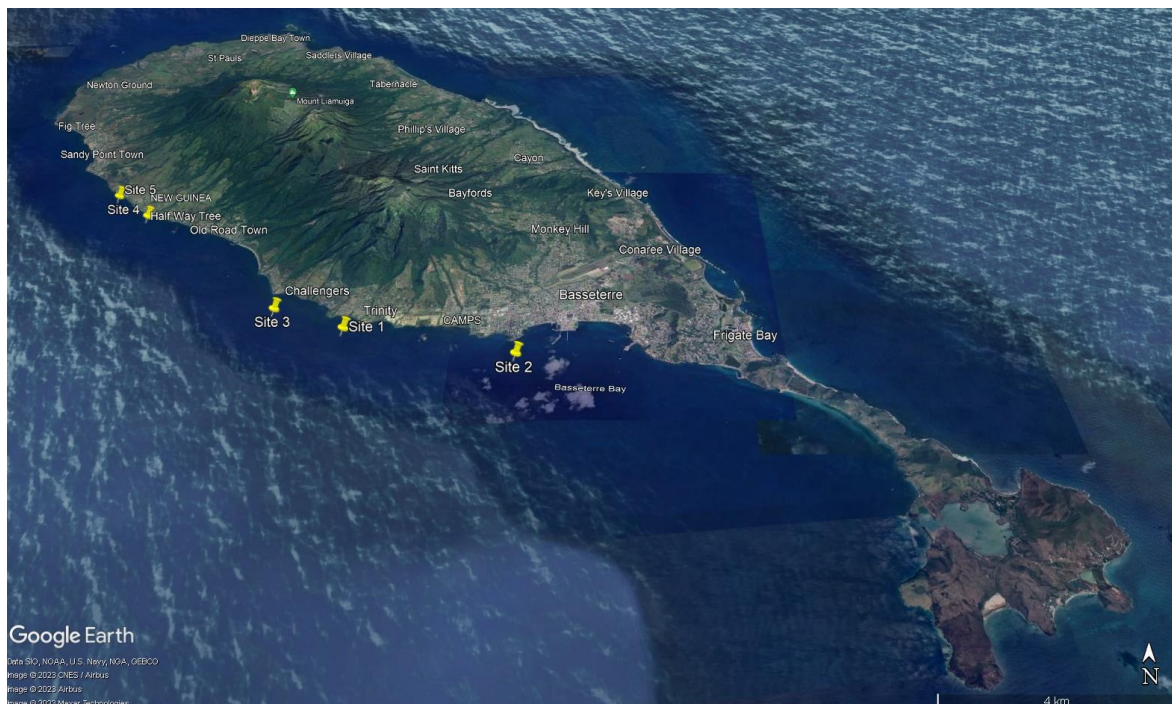


Figure 1. Map of dive site (Google Earth®).

Specimens were collected by SCUBA diving and pole-spearing, as is predominantly done for research and harvest of lionfish due to their almost invulnerability to conventional hook-and-line fishing (Morris Jr. & Whitfield 2009). Upon arriving to the surface, harvested fish were immersed into an ice/saltwater slurry in accordance with guidelines for use of fish in research and AVMA recommendations (AVMA 2020). For each specimen, total length, weight, sex, and reproductive staging was recorded. A metric ruler was utilized to measure the total length from tip of the snout to the longest point of the caudal fin, with a precision of 1 mm. Weight was determined by first blotting excess water from each fish with a paper towel prior to measuring on a scale (model Kern EW820-2NM©). Gonad staging was assessed where possible based on maturity as described in the Lionfish Dissection Guide (Green et al 2012). The gastrointestinal content and fecal samples for bacterial culture were collected, multiple organs were immediately fixed in formalin (for histologic study), and sagittal otoliths were retrieved, weighed, and measured for length. Weight of otolith was attained via a VWR® scale model #123-P. Otolith lengths were documented with an Olympus® dissection microscope and CellSens® imaging software. Where both sagittal otoliths could be successfully dissected free (n=68), the mean of the two was used in both weight and length otolith measurements. Data was collected and stored in an MS Excel database, with descriptive statistic performed in MS Excel and SAS 9.4. Further statistical comparisons were performed to assess the relationship between variables, using parametric or non-parametric tests, depending on the nature of the data. This research was approved by the Ross University School of Veterinary Medicine (RUSVM) Institutional Animal Care and Use Committee (IACUC # 19.11.32).

Results. Descriptive statistics were performed on fish and otolith variables, for the whole population and by fish sex. All four variables proved to be normally distributed after statistical assessment using the Shapiro-Wilk test and a graphical evaluation. Descriptive statistics are provided in Table 1.

Table 1

Descriptive statistics for lionfish (*Pterois volitans*)

<i>Variable</i>	<i>n</i>	<i>Mean</i>	<i>Standard deviation</i>	<i>Minimum</i>	<i>Maximum</i>
Otolith length (µm)	105	5523.3	838.47	3206.9	7501.3
Otolith weight (ug)	105	10.4	3.89	3.5	24
Fish total length (cm)	118	27.5	4.58	17	37.4
Fish mass (g)	118	312.4	160.27	57	752

Initial correlations were performed to evaluate the relationship between otolith length and weight. This correlation proved to be strong (correlation coefficient 0.81) and statistically significant ($p < 0.0001$). Fish mass and length were also obviously highly correlated (correlation coefficient 0.9; $p < 0.0001$) (Figure 2).

After the expected high correlations between both otolith measurements and both fish measurements were effectively demonstrated, the relationship between otolith length and fish mass was evaluated. The result indicated a strong statistically significant correlation of 0.74, $n=105$, $p < 0.0001$ (Figure 1). With further investigations, this correlation stratified by sex, and it was discovered that the pattern persists with correlation coefficients of 0.77 ($n=56$, $p < 0.0001$) and 0.79 ($n=46$, $p < 0.0001$) for females and males, respectively. When comparing otolith mass between lionfish sex, no statistically significant differences were found ($p > 0.05$).

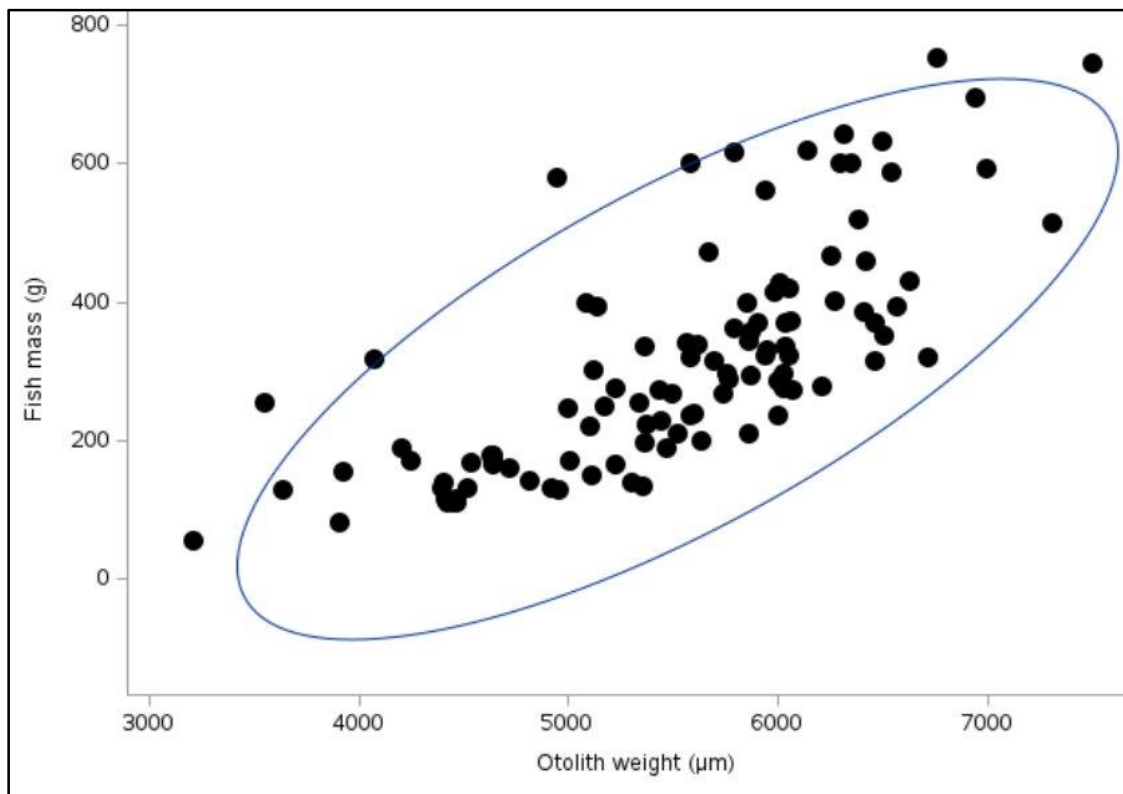


Figure 2. Scatterplot depicting the correlation between lionfish (*Pterois volitans*) mass (g) and otolith length (μm).

Discussion. Region-specific data gathered in this project is the first logical step in developing a current local lionfish population model, and for assessment of future management plans, disease events, environmental incidents, and planning targeted culling (Morris Jr. & Whitfield 2009; Johnson & Swenarton 2016; Dahl et al 2019; Fogg et al 2019). Furthermore, this is the first time lionfish population has been studied around the St. Kitts and Nevis region and is only the second set of data to document the association between lionfish size and otolith size. This data is similar to that of a study of lionfish in the southern Gulf of Mexico (Perera & Puerto 2016).

The authors acknowledge that attaining age measurements from each specimen in this study would have been ideal, as otolith annuli have been used successfully to determine age in other studies (Fogg et al 2019). Unfortunately, our attempts to measure age via otolith annuli was not productive. This is not the first time otolith annulus visualization has been reported as challenging with limited success. It is well documented that otoliths from tropical fish contain annuli which may be indistinguishable or difficult to read compared to those living in temperate regions. This is likely due to the lack of seasonality associated with minimal variation in water temperatures and therefore consistent growth of both the animal and its otoliths year-round (Hart & Pitcher 1983; Caldow & Wellington 2003; Marriott & Mapstone 2006; Green et al 2009; Fogg et al 2019). Age estimates attained from otoliths are typically accepted when multiple readers attain a minimum standard of variation, but this still does not assure that the ages attained are precise and accurate (Hanson & Stafford 2017). Furthermore, even when a measurement of age is gathered fruitfully, lack of within-reader agreement can result in many erroneous datapoints. For example, in one study, the measurements of 689 otoliths (58% of total samples) had to be eliminated (Giménez et al 2016), and only there was only 51% agreement between otolith readers in another (Blakeway et al 2022).

Overall, data in this study supports previous works indicating a strong correlation between otolith and fish size (Perera & Puerto 2016). Our data provides a baseline population database and provides insight to future research of predators of lionfish near St. Kitts and Nevis. This is especially important as a valuable complement to human

spearfishing because predator fish will naturally select smaller or wounded lionfish that are more easily captured, whereas human culling efforts focus heavily on larger, healthy specimens for harvest and consumption (De León et al 2013).

Conclusions. This study provides the first regional parameters for red lionfish near the islands of St. Kitts and Nevis and describes a direct correlation between fish size and otolith dimensions. This information will assist in future stock assessments, food-web dynamics research and management decision-making processes. Direct correlation of otolith size to fish size allows for interpretation of predator diet from stomach content study (Zan et al 2015). Continued recordkeeping of local lionfish population and distribution will be necessary to determine the accuracy of healthy ecology thresholds over time, as well as relative measures of culling and predation successes.

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Conflict of Interest. The authors declare that there is no conflict of interest.

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