Report on the status of U.S. Caribbean queen triggerfish *Balistes vetula* age, growth, and reproductive biology for the SEDAR80 Stock Assessment

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Introduction

Queen triggerfish *Balistes vetula* is a widely distributed member of the family Balistidae, found in tropical and sub-tropical waters of the western Atlantic; the geographical range in which it occurs with some abundance extends from northern waters off the coasts of North Carolina, throughout the Caribbean Sea, and as far south as Atlantic waters of southern Brazil (Fig 1). A study on the genetic population structure of queen triggerfish across its range throughout the Caribbean and south Florida documented a homogenous distribution of genetic variants indicating high connectivity across the region with no isolation detected for fish sampled from waters throughout the U.S. Caribbean or beyond (Antoni 2017).

Queen triggerfish occurs at depths up to 275 m and is associated with reef, rubble, and adjacent sandy habitats (Robertson and Van Tassell 2019). Several studies from north Caribbean waters have documented habitat attributes for areas where queen triggerfish are most commonly found. Diver surveys in Saba Bank documented adults associated with the whole spectrum of coral reef ecosystem strata, but most abundant in the outer reef flat zone characterized by hard bottom/pavement and submerged inner reef flat zone with low relief pavement and scattered rubble (Toller 2007; Debrot et al. 2020). Several studies from west Puerto Rico noted adult queen triggerfish associated with slope wall habitat at depths greater than 30 m (García-Sais et al. 2007; Garcia-Sais 2010). On-going video survey research of deepwater Puerto Rico coral habitats has documented queen triggerfish in association with hard bottom reef areas up to a maximum depth of 115 m (K. Overly, NMFS, personal communication). Preliminary analysis of queen triggerfish mean size at depth recorded for fish from visual surveys conducted as part of the National Coral Reef Monitoring Program in waters of the U.S. Caribbean indicated a significant correlation between depth and size; mean size of queen triggerfish increased with increasing depth of observations (L. J. Grove/J. Blondeau, Reef Fish Ecology Unit, NOAA, unpublished data).

Little is currently known about habitat associations and needs for post-hatch and small juveniles of queen triggerfish. In spite of a lack of scientific documentation, many sources assume queen triggerfish individuals in these smallest life stages are pelagic and may associate with sargassum because several studies from temperature waters of the Atlantic documented high abundances of small gray triggerfish *Balistes capriscus* associated with drifting sargassum mats (Wells and Rooker 2004; Hoffmayer et al. 2005; Casazza and Ross 2008). However, no extensive reports exist in the peer-reviewed literature on fishes associated with sargassum mats of tropical waters in the Caribbean. One study from Panama noted a personal observation of several small queen triggerfish (56-59 mm FL) in the stomach of a 460 mm FL dolphinfish *Coryphaena hippurus* caught in Caribbean waters as evidence supporting that queen triggerfish remain in pelagic habitat until they settle into benthic habitat around 50-70 mm FL (Robertson 1988). Larger juveniles (50+ mm FL) have been observed in shallow back reef sites (1-3 m) characterized by a mosaic of sand, sparse seagrass, scattered rock/rubble, and pavement with an assortment of crevices that triggerfish used for refuge along the coast of Panama (Robertson 1988). During sampling efforts in the current study, we have observed small triggerfish (50-80 mm FL) in shallow (< 5 m) nearshore waters of St. Croix (Fig 2) associated with similar habitat as described for Panama. We have also observed and collected small fish (60-80 mm FL) in Puerto Rico from shallow seagrass sites and rock/rubble pavement areas interspersed with seaweed.

Queen triggerfish is a sexually dimorphic gonochorist characterized by a medium size at sexual maturity (Rivera Hernández et al. 2019). It is a nesting benthic spawner that exhibits group-synchronous oogenesis and indeterminate fecundity (Rivera Hernández et al. 2019) and may form spawning aggregations during at least some months of its spawning season in associations with the full moon (Bryan et al. 2019; Rivera Hernández et al. 2019). The annual spawning season for queen triggerfish, determined through histological analysis of female gonad samples, starts as early as December and extends to August in waters of the north Caribbean (Rivera Hernández et al. 2019).

No published investigation exists yet on nesting strategies and associated spawning behaviors of queen triggerfish. Several researchers in north Caribbean waters, often investigating spawning aggregations of red hind *Epinephelus guttatus* around the full moon, have noted that queen triggerfish appear to occupy benthic nests at the same time (Figs 3-4). One of the red hind aggregation sites where queen triggerfish nests occurred is Lang Bank, St. Croix (Kobara et al. 2013). Fishers from Puerto Rico identified multiple sites where they have observed nesting queen triggerfish (Ojeda-Serrano 2007) (Fig 4). Bryan et al. (2019) investigated the spatial and temporal movement patterns of queen triggerfish via surgically implanting acoustic transmitters in 55 queen triggerfish caught in Buck Island Reef National Monument (BIRNM), St. Croix, and reported that during the full moon periods of January to

March, 12 individuals undertook repeated migrations to Lang Bank. In the same study, several other tagged fish larger than the size of 100% sexual maturity remained within BIRNM during the same-season spawning periods which may indicate a local resident nesting area for queen triggerfish (Bryan et al. 2019).

Triggerfish species are mostly aged using the first dorsal spine, mainly due the ease of obtaining the spines relative to extracting triggerfish otoliths which are small, fragile, and take more effort to extract (Manooch and Drennon 1987; Barroso-Soto et al. 2007; Aggrey-Fynn 2009; Kelly-Stormer et al. 2017; Allman et al. 2018; Shervette et al. 2021). However, otoliths are considered to provide more accurate and precise age estimates when compared to alternative ageing structures, like spines, scales, and fin rays, which can significantly underestimate the true age of a fish compared to otoliths (Buckmeier et al. 2002; Buckmeier et al. 2012; Lozano et al. 2014; Sinkus et al. 2017). Prior to our investigations on otolith-based age estimation in *Balistes* species, both gray triggerfish and queen triggerfish were thought to be relatively short lived with maximum spine-based age estimates of 15 y (Burton et al. 2015; Allman et al. 2018) and 14 y (Albuquerque et al. 2011), respectively. Shervette and Rivera Hernández (2022) utilized radiocarbon analysis of eye lens cores to validate the otolith-based age estimation for *Balistes* triggerfish species and documented that queen triggerfish is a relatively long-lived reef fish attaining a maximum age of 40 y. Otolith-based age estimation of gray triggerfish also extended longevity to a maximum age estimate of 20 y (Rivera Hernández and Shervette 2022).

In waters of the Atlantic and Caribbean, queen triggerfish is considered a data-deficient/poor species due to lack of information on population demographics. In the U.S. Caribbean, no current information was available on queen triggerfish population age structure and sex-specific and combined growth rates, despite its importance as one of the top commercially landed reef fish species in all U.S. Caribbean management platforms: Puerto Rico (PR), St. Thomas/St. John (STT/J) and St. Croix (STX) (SEDAR 2016; Matos-Caraballo 2018; Rivera Hernández et al. 2019). To fill in critical life history information gaps for queen triggerfish, the main goal of this study was to document age, growth, and reproductive biology of this species in U.S. Caribbean waters.

<u>Overall goal</u>: The purpose of this study was to utilize a combination of fishery-dependent (FD) and fishery-independent (FI) samples to assess the size/age structure, growth,

reproductive seasonality, and spawning frequency for queen triggerfish from U.S. Caribbean waters.

Objectives: 1) describe length and age structure for males and females; 2) calculate growth overall for queen triggerfish and separately for females and males; 3) evaluate gear selectivity by examining differences in gear-specific means size and mean age; 4) update information on reproductive biology of queen triggerfish previously reported in Rivera Hernandez et al. (2019); and 5) report on age at sexual maturity for queen triggerfish from the north Caribbean.

Methods

Study region and management

The U.S. Caribbean is located in the western portion of the Caribbean archipelago and includes the territories of Puerto Rico (PR) and the U.S. Virgin Islands (USVI). It is divided into three fisheries management island platforms that reflect ecological, cultural (e.g., traditional fishing practices), and market preference distinctions specific to each platform area (CFMC 2018). The largest island management platform is PR in the west. Waters of PR are characterized by extensive coral reef ecosystems that extend across approximately 3370 km^2 within 3 nautical miles of the main island of PR and its associated smaller islands. In terms of average annual landings (lb), queen triggerfish ranks in the top five commercially targeted reef-associated fishes (Matos-Caraballo 2018). Queen triggerfish are caught commercially in PR mostly via trap (42% of catch) and spearfishing (45% of catch). Annual reported landings of queen triggerfish for PR from 2012-2018 ranged from 40,000 – 72,000 lb (Matos-Caraballo 2018).

USVI is composed of two management platforms: in the north is STT/J and STX in the south. STT/J occurs on a large shelf platform containing a fishable area of 1500 km^2 and consists of a complex network of reef ecosystems (Kadison et al. 2017). Queen triggerfish is mainly caught commercially in STT/J by traps (98% of catch), is the top reef fish landed (in terms of total mass of catch), and is primarily fished in shelf waters at depths > 25 m (CFMC 2018). Reported annual commercial landings of queen triggerfish in STT/J ranged from 43,000 – 50,000 lb for 2012-2016 (CFMC 2018).

The management platform of STX is located approximately 60 km south of STT/J, separated from the northern platform by the deep waters of the Anegada Pass. STX sits on a narrow platform surrounded by approximately 300 km^2 of shelf waters containing a network of coral reefs and associated ecosystems. Over 85% of the fishable area is at depths < 25 m (Kadison et al. 2017). Commercial fishers in STX target queen triggerfish using hook-and-line gear, traps, and spearfishing. Queen triggerfish ranks in the top five reef fish species landed (in terms of total mass). Reported annual commercial landings for this species in STX ranged from 12,000-19,000 lb for 2012-2016 (CFMC 2018).

Field collection and processing

Fish samples were collected through (1) fishery-dependent (FD) sampling via purchase of fish from local fishers (2013-2020); (2) the fishery-independent (FI) Southeast Area Monitoring and Assessment Program – Caribbean (SEAMAP-C); and (3) opportunistic FI sampling via collaboration with local fishers (2014-2021). Fishery-dependent samples were obtained directly from fishers by randomly selecting all triggerfish from one side of a cooler containing the day's catch or by purchasing all queen triggerfish landed by a fisher on the day of sampling. Fishery-independent SEAMAP-C samples were caught as part of the reef fish annual hook-and-line surveys conducted from 2014-2020. To ensure that small, immature fish were included in the study, researchers and fishers worked collaboratively to opportunistically obtain FI samples using a combination of fine mesh gillnets (in PR) and spearfishing (PR, STT/J, STX). Additional FI samples were obtained from fishers that were non-randomly selected from daily catches due to their large sizes to ensure that larger size samples were represented in age estimation. Sample numbers are presented in Table 1.

All fish were kept on ice until processing occurred. Fish samples were measured for length (SL, FL, TL to nearest mm then converted for summary table to 0.1 cm) and whole weight (g then converted for summary tables to kg). Gonads were removed, weighed (to the nearest 0.01 g), then preserved for sex determination and reproductive phase via histological processing in the lab (Rivera Hernández et al. 2019). Queen triggerfish otoliths were extracted and stored according to the protocol described in Rivera Hernandez and Shervette (2022). Size-related regression equations used to convert from one length to another and between lengths and weight are presented in Table 2.

Age estimation and growth

Sagittal otoliths (Fig 5) were read whole for age estimation as described in Rivera Hernandez and Shervette (2022). Sagittal otoliths from each fish were read blind (with no knowledge of fish size, date-of-collection, or sex) by a primary reader (VRS) with 10+ years of experience ageing tropical reef fishes and reading triggerfish otoliths. A subset of otoliths was read independently and blind by a secondary reader (JMRH) with 7 y of experience ageing tropical reef fishes. Percent agreement was calculated between readers. Average Percent Error (APE) was calculated to assess between reader precision and reported in Shervette and Rivera Hernandez (2022) along with a detailed accounting of the otolith-based age estimation method validation results via bomb radiocarbon. Samples for which reader disagreement of opaque zone counts occurred were re-examined simultaneously by both readers and a consensus age estimate was obtained. Edge analysis of otoliths reported in Shervette and Rivera Hernandez (2022) indicated that peak opaque zone formation occurred December through February. We combined this information with the month of peak spawning to establish a birthdate of 1 February and then calculated fractional ages from otolith opaque zone counts. A two factor ANOVA was used to test for significant differences in mean age by sample source (FI versus FD) and sex (female versus male). For size-at-age data, using fractional ages, separate von Bertalanffy growth functions (VBGF) were fit to estimated ages for the following groups: all samples combined, males, and females. A two-factor ANOVA was used to test the effect of sex on estimated size-at-age for ages 6-12, the most prevalent age classes present in the two datasets. The dependent variable for this was FL; the independent variables were age class and ageing structure.

Reproductive biology

Gonads were removed from each queen triggerfish sample; either the whole gonad or the posterior portion of each gonad was fixed in 11% seawater-buffered formalin, Davidson's fixative (Howard et al. 2004), or polyethylene glycol–ethyl alcohol–glycerol–acetic acid (PAGA) fixative (Zanini et al. 2012) for up to 2 weeks and then transferred to 70% isopropanol. Gonad samples were processed using standard histological procedures for triggerfish species (Kelly-Stormer et al. 2017; Rivera Hernández et al. 2019). The tissue samples were vacuum-infiltrated and embedded in paraffin wax. At least three transverse sections (~7 µm thick) were cut using a rotary microtome, mounted on glass slides, stained with double strength Gill hematoxylin, and counter-stained with eosin-y. Stained sections were viewed using a compound microscope to determine sex and reproductive phase, assessed according to the histological criteria described in Rivera Hernandez et al. (2019). Two readers independently assigned sex and reproductive state without knowledge of the capture date, specimen length, or specimen age. If differences in the assignment of reproductive phases occurred, readers examined the slide simultaneously to obtain a consensus phase assignment. If no consensus was reached, then that specimen was eliminated from the analyses. A two factor ANOVA was used to test for significant differences in mean size by sample source (FI versus FD) and sex (female versus male).

Rivera Hernández et al. (2019) described the methods used in the current study to determine the reproductive season, spawning interval, and spawning frequency for queen triggerfish in U.S. Caribbean waters. Since that work was published, we have collected and processed additional samples for reproductive biology and report on the updated results. We also added the determination of age at sexual maturity for all fishes combined and separately for males and females. To do this we utilized logistic regression.

Results

A total 2,164 queen triggerfish samples were collected and processed for this study (Table 1). Across the 2013-2021 sampling period, a total of 711 fish were obtained from waters of PR with monthly totals ranging from a minimum of 27 for November to a maximum of 91 for August (Table 3). For STT, we collected a total of 719 queen triggerfish with a monthly total range of 36 fish representing October and 36 representing November to 107 fish collected for the month of May (Table 3). In STX, we collected 734 queen triggerfish with a month total range of 39 fish for May and 39 fish for August to 93 fish for July (Table 3). Sample numbers, mean size (FL), and size ranges by sex, source, and gear type are summarized in Table 1. Mean size differed significantly between males and females and between FI and FD collections; males were significantly larger than females and FD fish were significantly larger than FI fish (Table 4).

Shervette and Rivera Hernández (2022) utilized bomb radiocarbon to validate the otolith age estimation method for this species and demonstrated that spines do not provide accurate ages for triggerfish and should not be used for age-related parameters used in quantitative stock assessments. For queen triggerfish from U.S. Caribbean waters, a total of 2045 samples were aged using the validated otolith age estimation method. Queen triggerfish ages ranged from 0-23 y with a mean age overall of 8.4 y (Table 1). Mean age did not differ significantly between males and females, but did differ between FD and FI samples; FD samples had significanly older ages compared to FI samples (Table 4). The youngest fish aged was 0 y (n = 5, CV for mean size at age 0 was 16.226%). The oldest fish aged from the U.S. Caribbean was 23 y (n = 1); CV for mean size at ages 17+ y was 6.591% (n = 18) and CV for mean size at ages 18+ y (n = 12) was 5.257%. Results from the two factor ANOVA testing for differences in mean size at age between males and females indicated that females were significantly smaller than males for each of age classes included in the analysis (6-12 y; Table 5). A summary of mean size and size range at age for all fish combined, males, and females is provided in Table 6.

Parameter estimates for growth curves fitted to all size-at-age data combined, males, and females are provided in Table 7. Males attained a larger asymptotic length than females; K was 0.15 for both sexes and overall. Growth curves for the combined samples and for females were computed with t0 fixed to the value naturally generated for males (Fig 6). Also note that the the size-at-age data for the 23 NC/SC queen triggerfish samples, provided for visual comparison, appear to trend larger at age compared to the overall trends for the U.S. Caribbean fish (Fig 6). A similar latitudinal gradient was noted for gray triggerfish (Shervette et al. 2021), red porgy *Pagrus pagrus* (Potts and Manooch 2002), and queen snapper *Etelis oculatus* (Overly 2021).

In total, 2,067 gonads from queen triggerfish were collected. Sex and reproductive phase were assigned to 2,044 individuals. The smallest mature male was 12.2 cm FL, and the largest immature male was 24.2 cm FL; the L_{50} of males was 20.6 cm FL (Table 8; Fig 7). The smallest mature female was 19.6 cm FL, and the largest immature female was 25.7 cm FL. The L_{50} of females was 22.7 cm FL (Table 8; Fig 8). The combined L_{50} was 21.2 cm FL (Table 8; Fig 9).

Actively spawning females occurred from December through August in relatively low proportions relative to the total number of mature females samples each month; peak spawning for females was from December through February (Fig 10). Male queen triggerfish that were in the spawning capable phase occurred in all months of the year at relatively high proportions relative to all mature males sampled (Fig 11).

As aggregating nesting benthic spawners, queen triggerfish females exhibited group-synchronous oogenesis and indeterminate fecundity over the spawning season that started as early as the week after the full moon in December and extended until August. Spawning interval, defined as the number of days between spawning events in a female, was 20-60 days, indicating that a female could spawn 3-12 times over the estimated ~270-day spawning season.   We suspect this means that larger/older females can spawn at least once every month (timed to the moon cycle) for each month of the December – August season and younger/smaller females spawn less frequently. However, the need exists to conduct hypothesis-driven FI sampling to better understanding spawning frequency and spawning behavior for this species.

Literature Cited

Aggrey-Fynn J (2009). Distribution and Growth of Grey Triggerfish, *Balistes capriscus* (Family: Balistidae), in Western Gulf of Guinea. West African Journal of Applied Ecology 15.

Albuquerque C, Silva Martins A, de Oliveira Leite Junior N, Neves de Araujo J, Marques Ribeiro A (2011). Age and growth of the queen triggerfish *Balistes vetula* (Tetraodontiformes, Balistidae) of the central coast of Brazil. Brazilian Journal of Oceanography 59: 231-239.

Allman R, Patterson WI, Fioramonti C, Pacicco A (2018). Factors affecting estimates of size at age and growth in grey triggerfish Balistes capriscus from the northern Gulf of Mexico. Journal of fish biology 92(2): 386-398.

Antoni L (2017). Population structure, connectivity, and phylogeography of two balistidae with high potential for larval dispersal Balistes capriscus and Balistes vetula: The University of Southern Mississippi.

Barroso-Soto I, Castillo-Gallardo E, Quiñonez-Velázquez C, Morán-Angulo RE (2007). Age and growth of the finescale triggerfish, *Balistes polylepis* (Teleostei: Balistidae), on the coast of Mazatlán, Sinaloa, Mexico. Pacific Science 61(1): 121-127.

Bryan DR, Feeley MW, Nemeth RS, Pollock C, Ault JS (2019). Home range and spawning migration patterns of queen triggerfish Balistes vetula in St. Croix, US Virgin Islands. Marine Ecology Progress Series 616: 123-139.

Buckmeier DL, Irwin ER, Betsill RK, Prentice JA (2002). Validity of otoliths and pectoral spines for estimating ages of channel catfish. North American Journal of Fisheries Management 22(3): 934-942.

Buckmeier DL, Smith NG, Reeves KS (2012). Utility of Alligator Gar Age Estimates from Otoliths, Pectoral Fin Rays, and Scales. Transactions of the American Fisheries Society 141(6): 1510-1519, doi:10.1080/00028487.2012.717520.

Burton ML, Potts JC, Carr DR, Cooper M, Lewis J (2015). Age, growth, and mortality of gray triggerfish (Balistes capriscus) from the southeastern United States. [Journal Article]. Fishery Bulletin 113(1), doi:http://dx.doi.org/10.7755/fb.113.1.3.

Casazza TL, Ross SW (2008). Fishes associated with pelagic Sargassum and open water lacking Sargassum in the Gulf Stream off North Carolina. Fishery Bulletin 106(4): 348-363.

CFMC (2018). Caribbean Fishery Management Council Draft island based Fisheries Management Plans (FMPs); Available at https://bit.ly/37BYfnk.

Debrot AO, Brunel T, Schop J, Kuramae A, Bakkers Y (2020). Assessing effectiveness of the seasonal closure of the Moonfish Bank of the Saba Bank for two species of concern, the Red Hind and the Queen Triggerfish: the first five years. Wageningen Marine Research.

Garcia-Sais JR (2010). Reef habitats and associated sessile-benthic and fish assemblages across a euphotic-mesophotic depth gradient in Isla Desecheo, Puerto Rico. Coral Reefs 29(2): 277-288, doi:10.1007/s00338-009-0582-9.

García-Sais JR, Castro R, Sabater-Clavell J, Carlo M, Esteves-Amador RF, Bruckner AW, Fairey L (2007). Characterization of benthic habitats and associated reef communities at Bajo de Sico seamount, Mona Passage, Puerto Rico final report.

Hoffmayer ER, Franks JS, Comyns BH, Hendon JR, Waller RS (2005). Larval and juvenile fishes associated with pelagic Sargassum in the north-central Gulf of Mexico.

Kadison E, Brandt M, Nemeth R, Martens J, Blondeau J, Smith T (2017). Abundance of commercially important reef fish indicates different levels of over-exploitation across shelves of the U.S. Virgin Islands. PLOS ONE 12(7): e0180063, doi:10.1371/journal.pone.0180063.

Kelly-Stormer A, Shervette V, Kolmos K, Wyanski D, Smart T, McDonough C, Reichert MJ (2017). Gray triggerfish reproductive biology, age, and growth off the Atlantic Coast of the Southeastern USA. Transactions of the American Fisheries Society 146(3): 523-538.

Kobara S, Heyman WD, Pittman SJ, Nemeth RS (2013). Biogeography of transient reef fish spawning aggregations in the Caribbean: a synthesis for future research and management. Oceanogr Mar Biol Annu Rev 51: 281-326.

Lozano IE, Llamazares Vegh S, Dománico AA, Espinach Ros A (2014). Comparison of scale and otolith age readings for trahira, *Hoplias malabaricus* (Bloch, 1794), from Paraná River, Argentina. Journal of Applied Ichthyology 30(1): 130-134, doi:10.1111/jai.12317.

Manooch C, Drennon C (1987). Age and growth of yellowtail snapper and queen triggerfish collected from the U.S. Virgin Islands and Puerto Rico. Fisheries Research 6: 53-68.

Matos-Caraballo D (2018). Puerto Rico/NMFS Cooperative Fisheries Statistics Program April 2012 – March 2018 NA07NMF4340039. DRNA Fisheries Research Laboratory.

Ojeda-Serrano E (2007). CCRI-"Caribbean Coral Reef Institute""Reef fish Spawning Aggregations of the Puerto Rican Shelf".

Overly KE (2021). Age, growth and mortality estimates for queen snapper Etelis oculatus in the U.S. Caribbean and Gulf of Mexico. University of Florida,

Potts JC, Manooch CS (2002). Estimated ages of red porgy (*Pagrus pagrus*) from fishery-dependent and fishery-independent data and a comparison of growth parameters. Fishery Bulletin 100: 81-89.

Rivera Hernández JM, Peña Alvarado N, Correa Vélez K, Nemeth R, Appeldoorn R, Shervette V (2019). Queen Triggerfish Reproductive Biology in U.S. Caribbean Waters. Transactions of the American Fisheries Society 148(1): 134-147, doi:10.1002/tafs.10124.

Rivera Hernández JM, Shervette V (2022). Photographic guide to extracting, handling, and reading otoliths from *Balistes* triggerfish species SEDAR80-WP-03. North Charleston, SC.

Robertson DR (1988). Extreme variation in settlement of the Caribbean triggerfish Balistes vetula in Panama. Copeia: 698-703.

Robertson DR, Van Tassell J (2019). Fishes: Greater Caribbean Online information system. https://biogeodb.stri.si.edu/caribbean/en/pages.

SEDAR (2016). SEDAR 46 Stock Assessment Report Caribbean Data-Limited Species. North Charleston, SC.

Shervette VR, Rivera Hernández JM (2022). Queen triggerfish *Balistes vetula*: Validation of otolith-based age, growth, and longevity estimates via application of bomb radiocarbon. PLOS ONE 17(1): e0262281, doi:10.1371/journal.pone.0262281.

Shervette VR, Rivera Hernández JM, Nunoo FKE (2021). Age and growth of grey triggerfish *Balistes capriscus* from trans-Atlantic populations. Journal of Fish Biology 98(4): 1120-1136, doi:https://doi.org/10.1111/jfb.14644.

Sinkus W, Shervette V, Ballenger J, Reed LA, Plante C, White B (2017). Mercury bioaccumulation in offshore reef fishes from waters of the Southeastern USA. Environmental Pollution 228: 222-233.

Toller W (2007). Habitat surveys of Saba Bank, Netherlands Antilles: an assessment of benthic communities and fish assemblages. Saba Bank Project Report.

Wells RJD, Rooker JR (2004). Spatial and temporal patterns of habitat use by fishes associated with Sargassum mats in the northwestern Gulf of Mexico. Bulletin of Marine Science 74: 81-99.

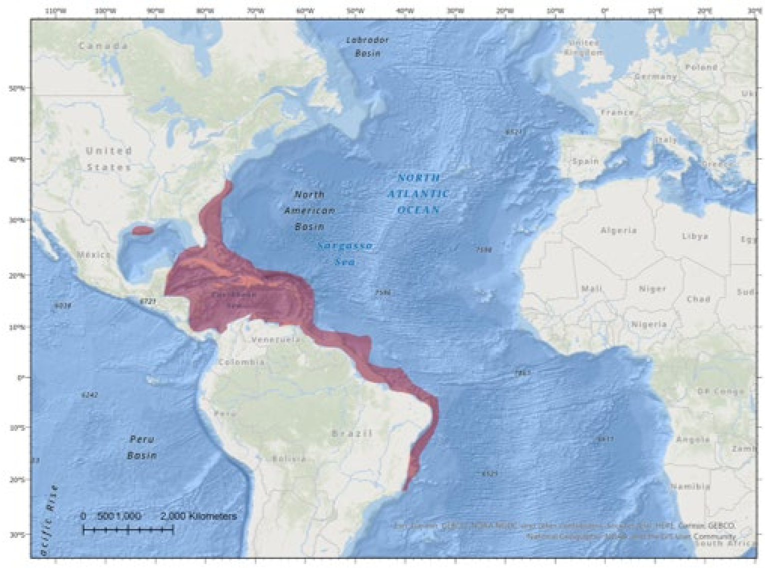

Fig 1. Red shaded area represents the general region where queen triggerfish occur with moderate to high abundance.

Fig 2. Image of a small (65 mm FL) queen triggerfish from St. Croix observed in nearshore, shallow water habitat characterized by a mosaic of sand, sparse seagrass, scattered rock/rubble, and pavement with an assortment of crevices. Photo by V Shervette.

Fig 3. Digital images of queen triggerfish on benthic nests in waters of west PR. The left image is from 21 Feb 2022 (five days after the full moon) by Veronica Seda of a queen triggerfish tending a nest Abrir La Sierra. Note that the female appears to be flashing white blotches; similar coloration signals have been noted in other triggerfishes when a female is guarding her nest and tending to the eggs. The right image is from 23 Apr 2021 (three days prior to a super full moon) by Michelle Schärer of a queen triggerfish tending a nest with eggs. Nests observed in this region were described as: "a pit about 25-50 cm deep in sandy depressions between the colonized substratum that could be between 1-3 m in diameter where one of the *B. vetula* would remain blowing head down near the substratum if egg masses were present."

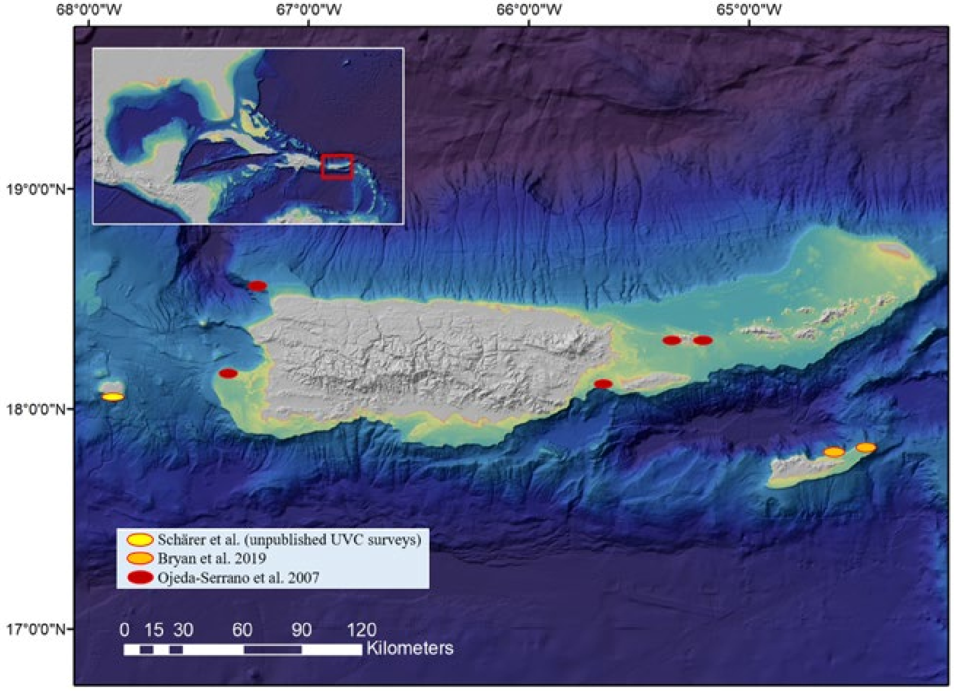

Fig 4. Map of U.S. Caribbean noting various locations identified in the literature where queen triggerfish benthic nesting activity has been noted.

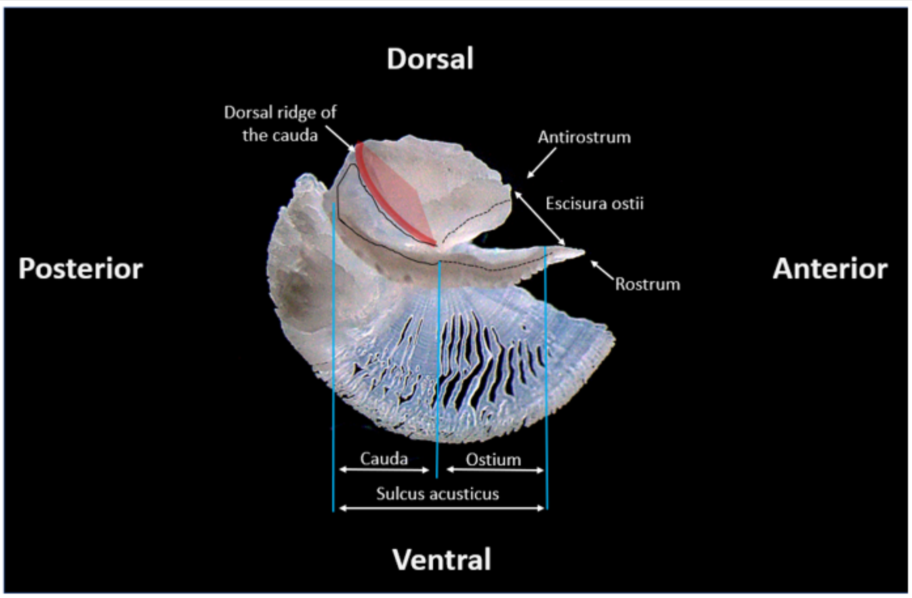

Fig 5. Digital image of the left sagittal otolith of a queen triggerfish. Sagittal otoliths are read using a stereo microscope with reflected light, against a black background while submerged in water at a magnification of 20-32x. Typically, we enumerate the opaque zones along the dorsal ridge of the cauda as indicated in the image by the red zone.

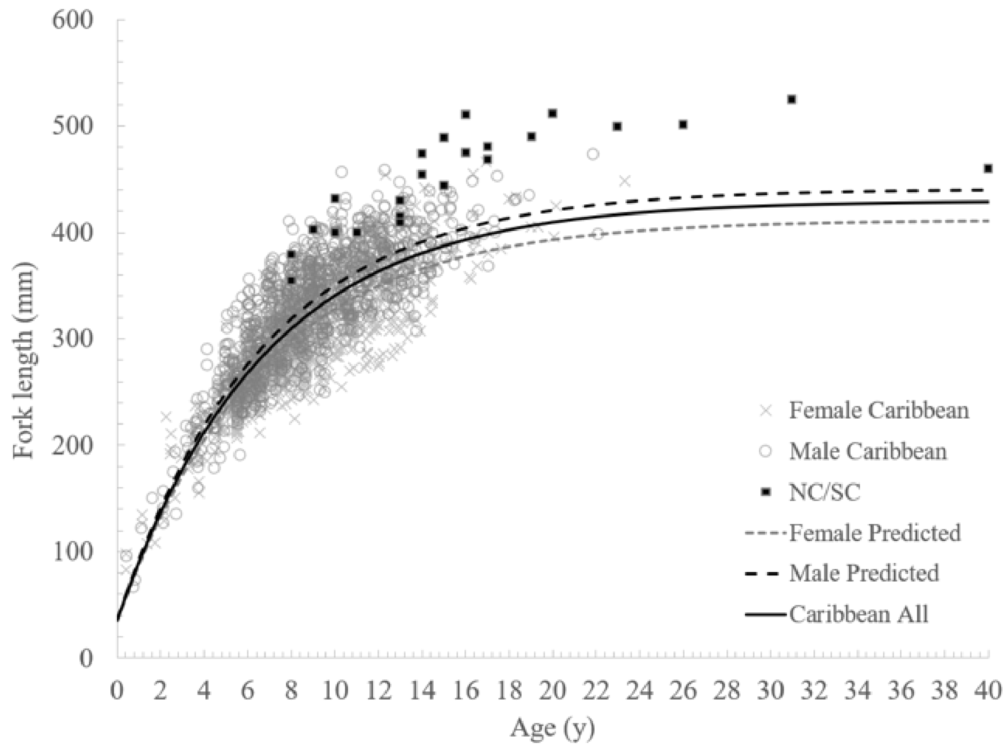

Fig 6. Length-at-age and von Bertalanffy growth curves for all Caribbean samples combined, female Caribbean samples, and male Caribbean samples. Length-at-age is also plotted for the 23 samples from NC/SC for visual comparison of overall trends.

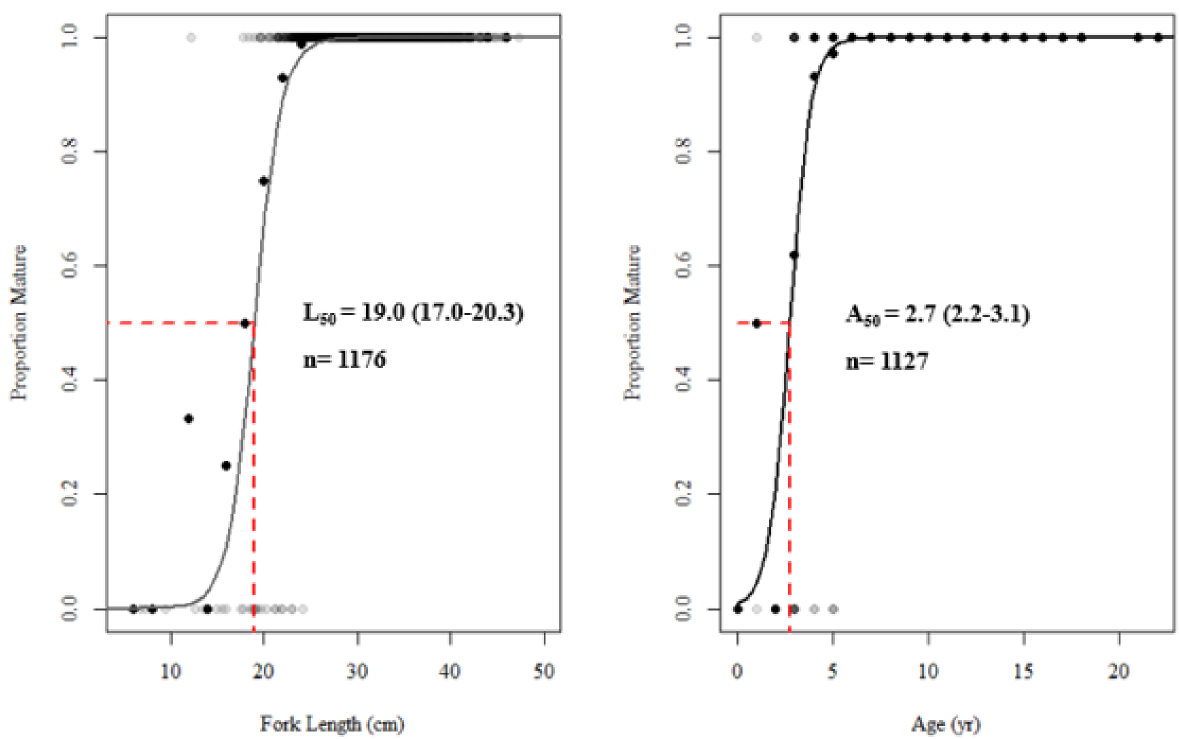

Fig 7. Male size (left) and age (right) at sexual maturity.

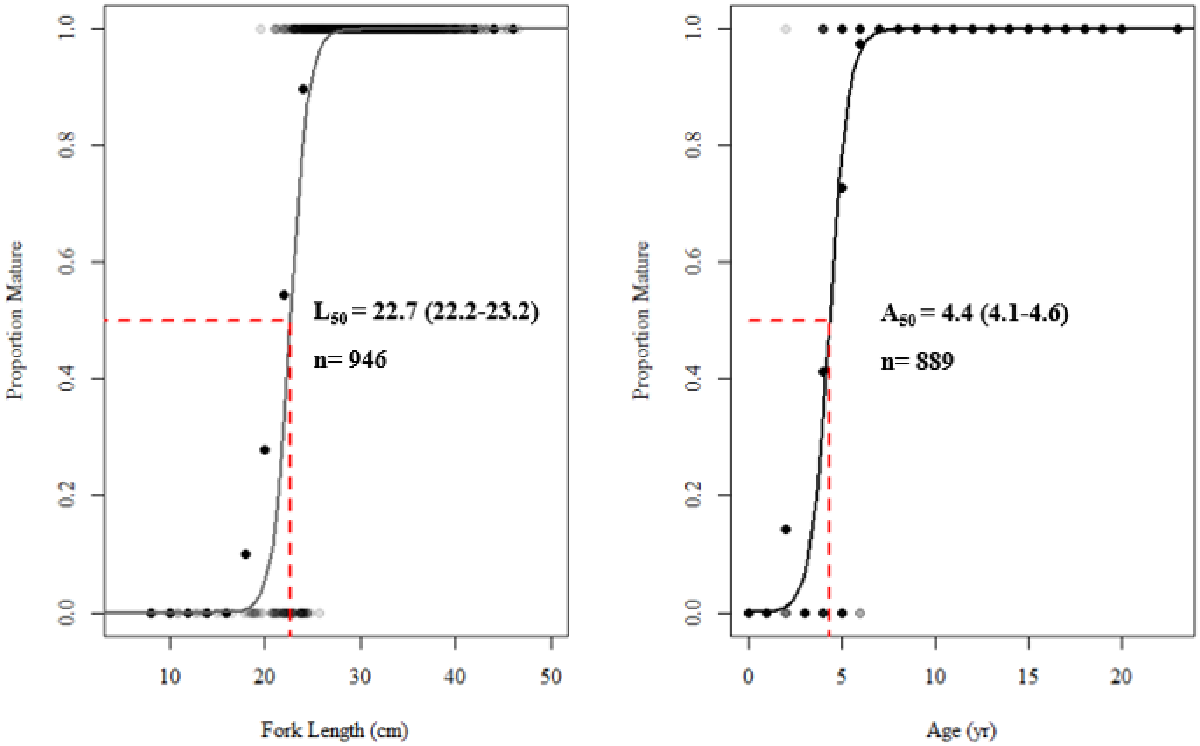

Fig 8. Female size (left) and age (right) at sexual maturity.

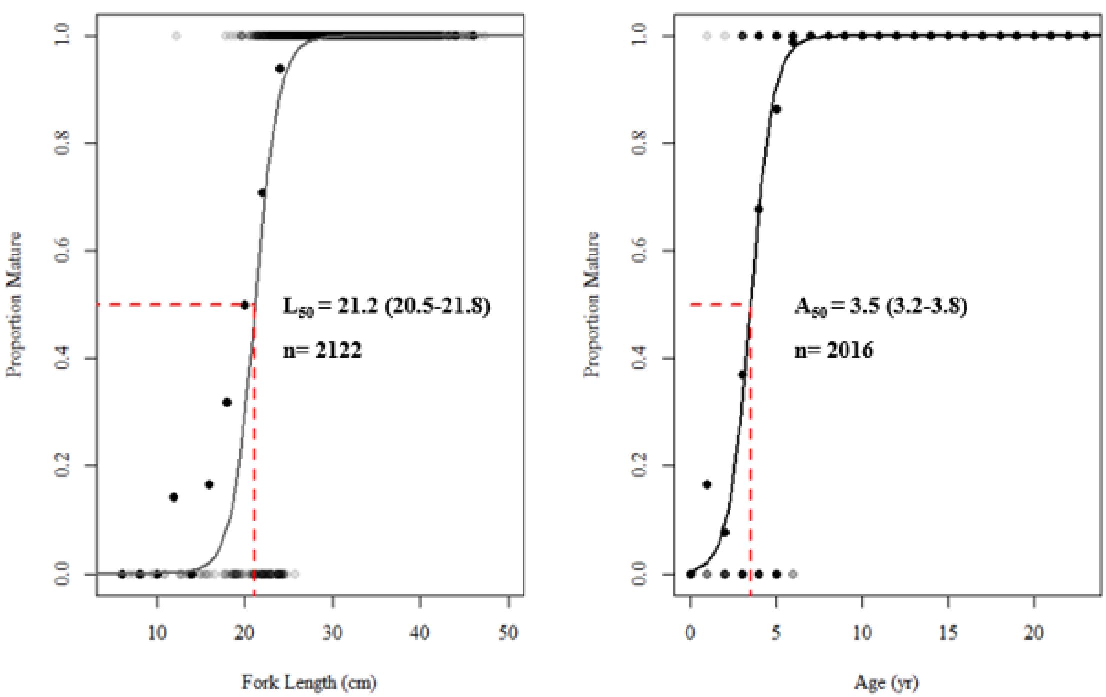

Fig 9. Overall size (left) and age (right) at sexual maturity.

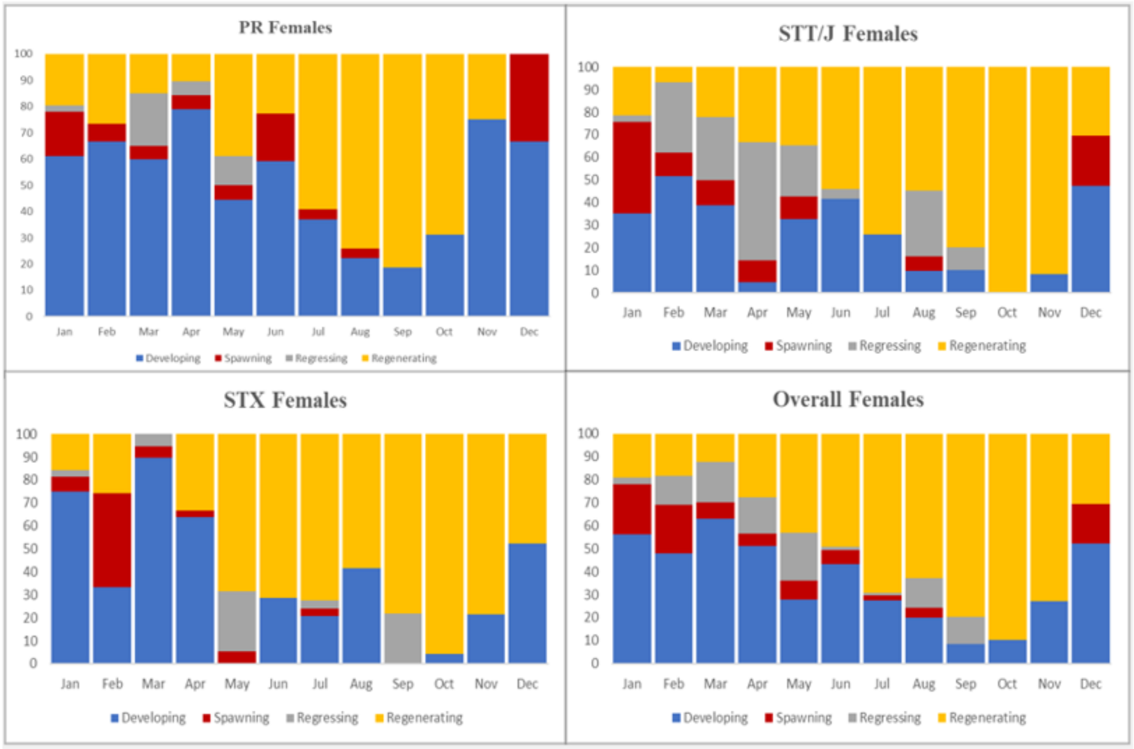

Fig 10. Reproductive seasonality of female queen triggerfish in the U.S. Caribbean. Monthly percentage of individual females in each reproductive phase are presented.

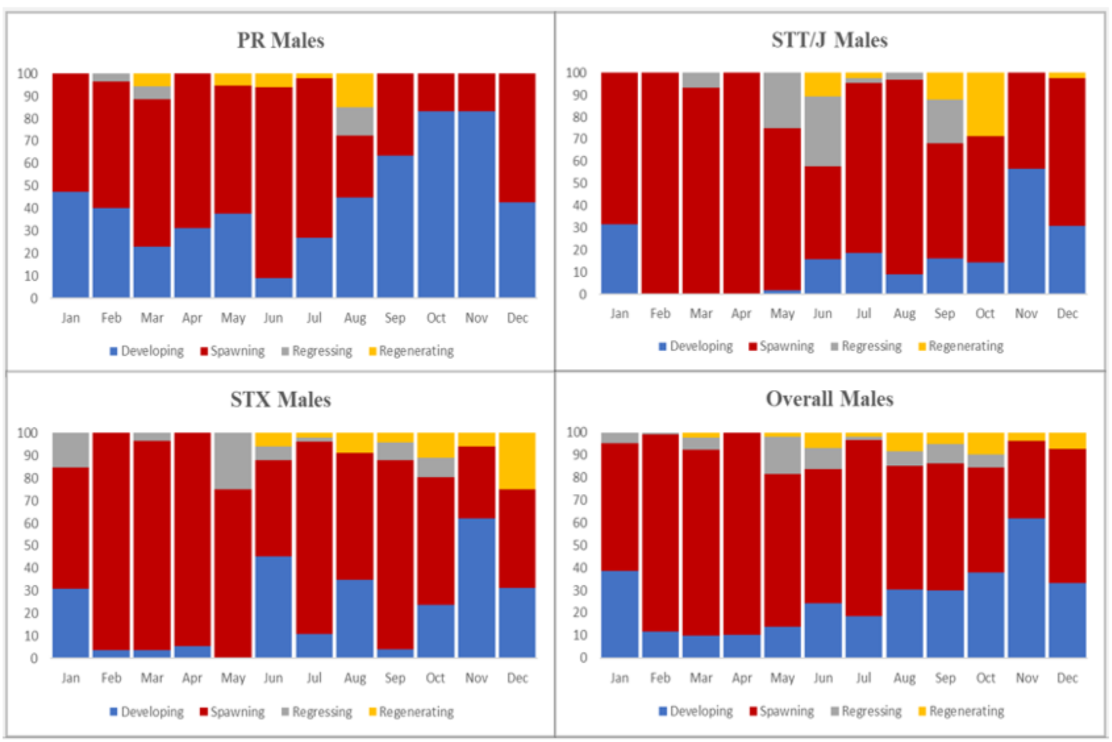

Fig 11. Reproductive seasonality of male queen triggerfish in the U.S. Caribbean. Monthly percentage of individual males in each reproductive phase are presented.

Table 1. Summary of samples caught by sex, gear type, source, and island management platform. Table is divided into summary values by size and age.

	Size			Age		
Category	**n**	**Mean FL mm**	**Range**	**n**	**Mean age y**	**Range**
Overall	2164	314	67-473	2045	8.4	0-23
Male	1177	324	67-473	1128	8.4	0-22
Female	947	301	83-466	890	8.3	0-23
Unknown	40	315	129-430	27	8.5	2-16
Source						
FD	1851	319	204-458	1764	8.6	2-23
FI	313	283	67-473	281	7.1	0-21
Trap ALL	1023	336	150-473	990	9.2	2-23
Spear ALL	893	296	67-433	851	7.8	0-22
Spear FD	702	304	220-433	670	8.1	4-22
Spear FI	191	260	67-415	181	6.6	0-16
Hook ALL	117	336	190-410	78	8.7	3-15
Hook FD	42	351	247-410	24	9.6	6-14
Hook FI	75	327	190-405	54	8.3	3-15
Net ALL	131	256	83-434	126	5.9	0-18
Net FD	118	262	211-434	114	6.3	3-18
Net FI	13	188	83-284	12	2.6	0-6
Island						
Puerto Rico	711	296	67-434	662	7.9	0-20
St. Thomas/John	719	345	150-473	683	9.6	2-23
St. Croix	734	300	190-414	700	7.7	2-22

Table 2. Regression equations for length-length and length-weight relationships of queen triggerfish samples from the current study. Length measurements used were mm; whole weight was measured in g.

Size Conversion Relationship	Equation	R-squared
SL-FL	$y = 1.104x + 9.7855$	$R^2 = 0.9865$
SL-TL	$y = 1.6369x - 23.951$	$R^2 = 0.8999$
SL-Wt	$y = 0.0001x^{2.7702}$	$R^2 = 0.9722$
FL-SL	$y = 0.8935x - 5.0213$	$R^2 = 0.9865$
FL-TL	$y = 1.4751x - 35.546$	$R^2 = 0.8991$
FL-Wt	$y = 5E\text{-}05x^{2.8708}$	$R^2 = 0.9792$
TL-SL	$y = 0.5498x + 40.659$	$R^2 = 0.8999$
TL-FL	$y = 0.6095x + 53.289$	$R^2 = 0.8991$
TL-Wt	$y = 0.0004x^{2.3894}$	$R^2 = 0.9206$

Table 3. Monthly queen triggerfish sample collections among the three island-based management platforms.

Month	PR	STT	STX	Grand Total
January	82	58	62	202
February	47	66	83	196
March	70	53	53	176
April	46	47	56	149
May	59	107	39	205
June	63	44	58	165
July	88	79	93	260
August	91	67	39	197
September	61	47	55	163
October	47	36	74	157
November	27	36	80	143
December	30	79	42	151
Grand Total	**711**	**719**	**734**	**2164**

Table 4. Results from ANOVAs testing for significant differences in mean size and mean age.

Source	df	Sum of Squares	Mean Square	*F*	*P*
Length (FL mm)					
Sex	1	143,721	143,721	47.8	< 0.001
Source	1	319,015	319,015	106.1	< 0.001
Sex x Source	1	1816	1816	0.6	0.437
Error	2120	6,373,933	3007		
Age					
Sex	1	21	21	2.4	0.124
Source	1	474	474	54.0	< 0.001
Sex x Source	1	25	25	2.9	0.089
Error	2014	17,677			

Table 5. Results from ANOVA testing for significant differences in mean size at age.

Source	df	Sum of Squares	Mean Square	*F*	*P*
Length (FL mm)					
Age (6-12 y)	6	1,185,621	197,603	209.9	< 0.001
Sex	1	210,826	210,826	223.9	< 0.001
Age x Structure	6	8477	1413	1.5	0.174
Error	1494	942			

Table 6. Length-age matrix for Caribbean queen triggerfish samples. Number of samples (n) within each age group, mean size (mm FL), and size range (mm FL) are indicated for all samples combined and then separately for males and females.

	Overall				Males				Females		
Age class	n	Mean size	Size range		n	Mean size	Size range		n	Mean size	Size range
0	5	83	67-98		3	78	67-95		2	91	83-98
1	6	125	108-150		2	136	122-150		4	120	108-135
2	13	167	127-227		6	155	127-193		7	177	129-227
3	35	202	155-243		21	205	160-243		14	199	155-237
4	59	229	178-290		30	233	178-290		29	226	185-257
5	198	255	190-339		110	260	190-339		88	250	207-319
6	260	277	211-360		140	284	231-359		120	269	211-360
7	286	300	237-410		154	312	239-410		132	286	237-373
8	293	317	225-409		168	330	247-409		125	300	225-388
9	209	332	243-419		120	342	250-419		89	319	243-380
10	182	346	255-456		101	358	298-456		81	331	255-415
11	153	360	273-420		98	370	292-420		55	342	273-416
12	125	365	275-458		67	378	321-458		58	352	275-454
13	91	373	292-447		51	383	307-447		40	359	292-431
14	49	382	307-442		27	384	343-439		22	379	307-442
15	20	402	370-439		15	406	370-439		5	390	379-414
16	16	404	356-466		6	412	394-448		10	399	356-466
17	6	400	368-453		4	408	368-453		2	383	381-385
18	5	432	430-435		3	432	430-435		2	433	432-434
19	2	405	405-405						2	405	405-405
20	2	411	396-425						2	411	396-425
21	1	473			1	473					
22	1	398			1	398					
23	1	449							1	449	

Table 7. von Bertalanffy growth parameters for queen triggerfish. So that parameter estimates can be easily compared among the models, the value for t_0 in these models was fit to 0.585 which was the naturally generated value for the "males" model. Fractional ages used to compute growth parameters. Data ranges below parameter estimates are 95% confidence intervals (CI).

Model	n	L_∞ (FL cm)	*K*	t_0	R^2	P-value
All Caribbean	2045	44.4 43.1-46.0	0.13 0.12-0.14	-1.12 -1.57- -0.74	0.69	< 0.001
All Caribbean (t_0 fixed)	2045	43.0 42.3-43.8	0.15 0.14-0.16	-0.585	0.69	< 0.001
Females (t_0 fixed)	890	41.2 40.2-42.3	0.15 0.14-0.16	-0.585	0.70	< 0.001
Males	1128	44.1 42.8-45.8	0.15 0.13-0.17	-0.585 -1.07- -0.18	0.72	< 0.001

Table 8. Summary of population-related reproductive characteristics of queen triggerfish from the current study and from other *Balistes* studies for comparison.

	Size (mm FL)					Age (y)				
Group	n	FL_{50} 95% CI	Slope; Intercept	Smallest Mature	Largest Immature	n	A_{50} 95% CI	Slope; Intercept	Youngest Mature	Oldest Immature
Combined	2122	21.2 20.5-21.8	0.75 -15.80	12.2	25.7	2016	3.5 3.2-3.8	1.51 -5.32	1	6
Male	1176	19.0 17.2-20.3	0.69 -13.3	12.2	24.2	1127	2.7 2.2-3.1	1.77 -4.84	1	5
Female	946	22.7 22.2-23.2	1.09 -24.71	19.6	25.7	889	4.4 4.1-4.6	1.97 -8.57	2	6