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Size at maturity for yellow tang (*Zebrasoma flavescens*) from the Oahu, HI, aquarium fishery

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Abstract While prior research has examined age, growth, and reproduction of yellow tang (*Zebrasoma flavescens*), size at maturity was only preliminarily assessed and is a very important biological metric to fully understand yellow tang biology and population status. We had a unique opportunity to sample from the Oahu, Hawai‘i, aquarium fishery and across a size range from 55- to 110-mm total length (TL). We found a smaller than expected size at maturity for females (L_{50} = 63.4-mm TL (CI: 62.7 – 65.5-mm TL)) and males (L_{50} = 67.4-mm TL (CI: 66.4 – 70.3-mm TL)). Females as small as 65-mm TL had ovaries that contained hydrated oocytes, suggesting that spawning can occur at a size that was previously considered within the juvenile size range. However, very low gonad weights (< 0.26 g) limit any significant egg production from these small, young individuals. These results provide additional insight into the biology of *Z. flavescens,* but further research is needed to determine if size at maturity varies spatially and under different levels of fishing pressure.

Keywords Size at reproductive maturity · Ornamental fish · Size at first spawning · Coral reefs

Significance statement Size at maturity for yellow tang, *Zebrasoma flavescens*, has never been assessed anywhere in its native range. Yellow tang are important herbivores on coral reefs and also highly valued in the ornamental (aquarium) fish markets. This study provides the first estimate of size at maturity from the aquarium fishery which is important to sustainable fisheries management and ornamental aquaculture.

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Introduction

A majority of yellow tang (*Zebrasoma flavescens*) in the global ornamental fish market come from wild collections from coral reefs in Hawai‘i (Wood 2001). The aquarium fishery is the most lucrative nearshore fishery in Hawai‘i, with *Z. flavescens* comprising the majority of the individuals collected, targeting sizes ranging from 30 to 128 mm (Walsh et al. 2005; Stevenson et al. 2011). The majority of the aquarium fishery catch comes from the west coast of Hawai‘i Island but aquarium fishing also occurs on Maui and Oahu (Walsh et al. 2004). Prior management for the aquarium fishery in Hawai‘i consists of mandatory reporting of commercial catch, commercial bag limits, and closed areas (Fishery Replenishment Areas (FRAs)) (Walsh et al. 2005). However, in 2019,

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aquarium permits were suspended and the use of fine-mesh net (the prominent gear type for aquarium collecting) was banned pending a court review of an environmental impact statement (EIS) (Senate Thirtieth Legislature 2019). As of July 2021, after multiple hearings and a revised EIS was presented to the State of Hawai'i Board of Land and Natural Resources, only West Hawai'i aquarium fishers were found to be compliant with Hawai'i Environmental Policy Act but no aquarium fish collecting permits have been issued. Oahu aquarium fishers may have to go through a similar process. The Hawai'i aquarium fishery remains controversial and its future is uncertain. Furthering the available biological information for yellow tang is important for future fisheries management and to help inform ornamental aquaculture efforts for this species (Callan et al. 2018).

Zebrasoma flavescens is a long-lived surgeonfish (Acanthuridae) that inhabits coral reefs. Settlement occurs at around 30-mm total length (TL) at approximately 62 days old to deep coral reef areas (10 – 25 m) and longevity was estimated at 41 years (Claisse et al. 2009a, b). In Hawai'i, they exhibit a size-based ontogenetic shift in habitat use from primarily deep coral-rich areas to both deep coral areas and shallow reef flats (Claisse et al. 2009b). This habitat shift from the deeper areas where the aquarium fishery operates to shallower water occurs at around 140-mm TL and 165-mm TL for females and males, respectively (Claisse et al. 2009b). The shift in habitat use results in a change in diet and behavior, including diurnal migration patterns to and from deep coral reef habitats that are used for refuge to shallow coral reef flats used primarily for feeding (Walsh 1984; Claisse et al 2011). Pair and group spawning occur along these migration routes around sunset, at specific drop-off locations (Walsh 1984).

Egg production follows a lunar pattern and peaks in spring and summer (Bushnell et al. 2010). Maximum batch fecundity was found to be around 24,000 eggs and no relationship was found between size and batch fecundity (Bushnell et al. 2010). However, females under 120-mm SL produced limited numbers of eggs (Bushnell et al. 2010). The previous estimate of size at maturity is 80–90-mm (TL) at 8 to 9 months post-settlement; however, maturity criteria were not provided (Walsh 1984). Additionally, *Z. flavescens* females have been found to spawn as small as ~ 90-mm TL (Bushnell et al. 2010).

Prior studies of this species in Hawai'i provided much of what is known about the biology and life history of *Z. flavescens* (Claisse et al. 2009a, b; Bushnell et al. 2010). However, working directly with fishers provides unique opportunities to discover aspects of fish biology that have never been captured before (Schemmel and Friedlander 2017). This study was initiated by conversations with a local aquarium fisher on egg releases from *Z. flavescens* during his collections on Oahu, Hawai'i. These accounts prompted this research on the assessment of size at reproductive maturity and spawning in *Z. flavescens* collected from the aquarium fishery. Here, we examine the reproductive biology of *Z. flavescens* from 55- to 110-mm TL, within the commonly collected size range for the aquarium fishery.

Materials and methods

Fish were donated from a local aquarium fisher from collection locations on the north and south shores of Oahu. A total of 166 *Z. flavescens* were collected on scuba with the aid of a barrier net from April to June 2016 (Table 1; Pacific Islands Science Center, 2020). Some of the individuals were kept alive for up to 2 days and euthanized by an ice bath before being donated to science.

Fish total length (TL) was measured to the nearest millimeter and fish weight (FW) and gonad weight (GW) were assessed to the nearest 0.01 g. Entire gonad samples were preserved in buffered formalin for a minimum of 3 days. Gonad samples were rinsed overnight in fresh water and dehydrated through a series of ethanol dilutions (30%, 50%, 70%). Gonad samples were processed at the University of Hawai'i John Burns Medical School; embedded in paraffin, sectioned at 5 µm, and stained with hematoxylin and eosin counter staining. Reproductive state was diagnosed with modified criteria by Brown-Peterson et al. (2011) (Table 2). Reproductive investment with fish size was assessed using gonadosomatic index (GSI) calculated as

$$\mathrm{GSI} = \mathrm{GW}/\mathrm{GFW} \times 100$$

where *GFW* is the gonad free fish weight.

Table 1 Reproductive state of *Zebrasoma flavescens* collected from the aquarium fishery from April to June 2016. Moon phase listed as name and radian, with new moon = 0, first quarter = $\pi/2$, full = π, and last quarter = $3\pi/2$

Date	Moon	Male		Female				
		Immature	Mature	Immature	Developing	Spawning capable	Actively spawning	Resting
4/26/16	Waning (4.0)	1	13	1		2		5
4/27/16	Waning (4.2)	4	6		1	8	1	
5/3/16	Waning (5.5)		15		2		2	
5/11/16	Waxing (0.9)		15		2	2	6	1
5/21/16	Full (3.0)	2	7	2		3	5	1
6/2/16	New (5.6)		14		6	5	8	
6/7/16	New (0.4)		3			1	3	3
6/28/16	Waning (4.9)		7	1	1	2	3	2
Total		7	80	4	12	23	28	12

Table 2 Characterization of reproductive state of assessed *Zebrasoma flavescens* females modified from Brown-Peterson et al. (2011)

Reproductive state	Oocyte stages present	Mature	Diagnostics
Immature	Chromatin nucleolar, perinucleolar	No	Immature individual with chromatin nucleolar (large nucleus (germinal vesicle) surrounded by a thin layer of cytoplasm, perinucleolar (germinal vesicle increases in size and nuclei appear at its periphery). Thin ovary wall
Developing	Chromatin nucleolar, perinucleolar, cortical alveolar, and early vitellogenic (VTI, VTII)	Yes	Developing individuals with cortical alveolar oocytes (appearance of cortical alveoli (yolk vesicles) in the cytoplasm and formation of the vitelline membrane) and/or early vitellogenic oocytes, VTI and/or VTII, present
Spawning Capable	Chromatin nucleolar, perinucleolar, late vitellogenic (VT III)	Yes	Presence of late stage vitellogenic oocytes (VT IIII), identified by dramatic increases in oocyte size and uniform distribution of yolk
Actively spawning	Chromatin nucleolar, perinucleolar, vitellogenic, hydrated, POF	Yes	Contains fully hydrated oocytes and or recent postovulatory follicles (POFs)
Resting	Chromatic nucleolar, perinucleolar, vitellogenic (VTI, VTII, VTIII)	Yes	Ovary dominated by atretic oocytes (regressing) or ovary only contains chromatin nucleolar and/or perinucleolar oocytes (regenerating). May contain some (non-atretic) cortical alveolar or vitellogenic oocytes. The ovary wall is thick and the ovary may contain unabsorbed material from past spawning events (atretic oocytes and POFs). Atretic oocytes were identified by their loss of spherical appearance and theca membrane breakdown

Male and female size distributions were compared using a Komogorov-Smirnov test. *T*-tests were used to determine if there were differences in mean size between males and females at each location (north and south shore of Oahu). Size at sexual maturity (L_{50}) were reported as the size at which 50% of individuals of a given sex are mature utilizing a logistic regression model with binomial family and logit link function (Chen and Paloheimo 1994). Estimates of L_{50} were generated using 1000 bootstrapped replicates of the model coefficients. Analyses were conducted in R (Development Core Team R 2013) using the statistical packages *boot* and *nls*.

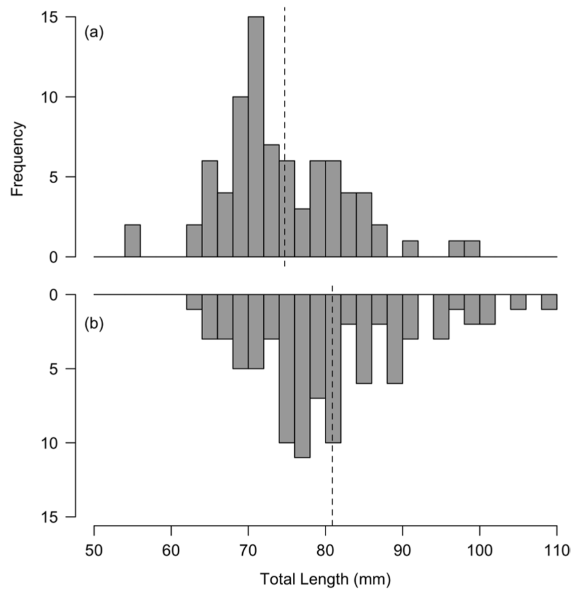

Fig. 1 Length frequency distribution for *Zebrasoma flavescens* **a** females and **b** males collected from the aquarium fishery off of Oahu, Hawai'i. Dashed vertical lines indicate mean total length (mm)

Results

A total of 166 *Z. flavescens* (87 males and 79 females) were sampled from the aquarium fishery on Oahu (Table 1). The results of Kolmogorov–Smirnov test indicated a significant difference between female and male size compositions ($D = 0.347$, $P < 0.001$; Fig. 1). Females ranged in size from 55- to 99-mm TL and males ranged in size from 64- to 110-mm TL. We found that within the size range sampled (55–110-mm TL), males were on average 6.2 ± 1.4 mm larger than females (t-value = 4.445, $P < 0.01$). Combined sex mean size was 77.9-mm TL.

The smallest sized spawning capable individual was very similar between females and males (female = 65-mm TL (Fig. 2D), male = 66-mm TL (Fig. 3B, C and D)). Four immature females were observed, ranging in size from 55- to 65-mm TL and 7 immature males, ranging in size from 64- to 75-mm TL. The estimated female L_{50} was 63.4-mm TL (CI: 62.7 – 65.5 mm TL) and the male L_{50} was estimated at 67.4-mm TL (CI: 66.5–70.2 mm TL) (Fig. 4).

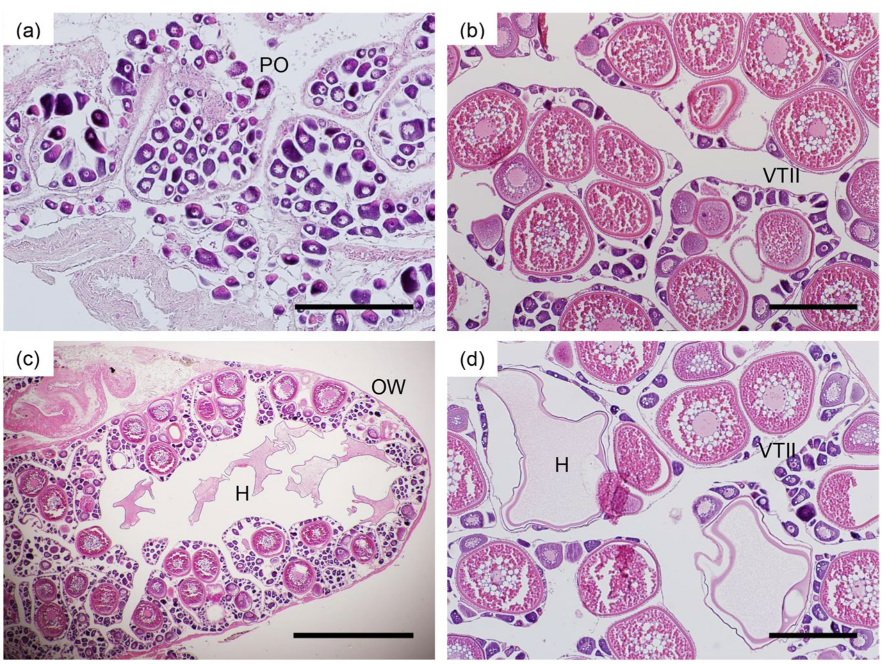

Fig. 2 Female reproductive states for *Zebrasoma flavescens* females. **a** Immature female with primary stage oocytes (PO) (63 TL; scale bar is 200 μm), **b** developing female with vitellogenic stage II oocytes (VTII) (78-mm TL; scale bar is 300 μm), **c** actively spawning female with hydrated oocytes (65 TL; scale bar 1000 μm), and **d** actively spawning female (81-mm TL; scale bar is 300 μm)

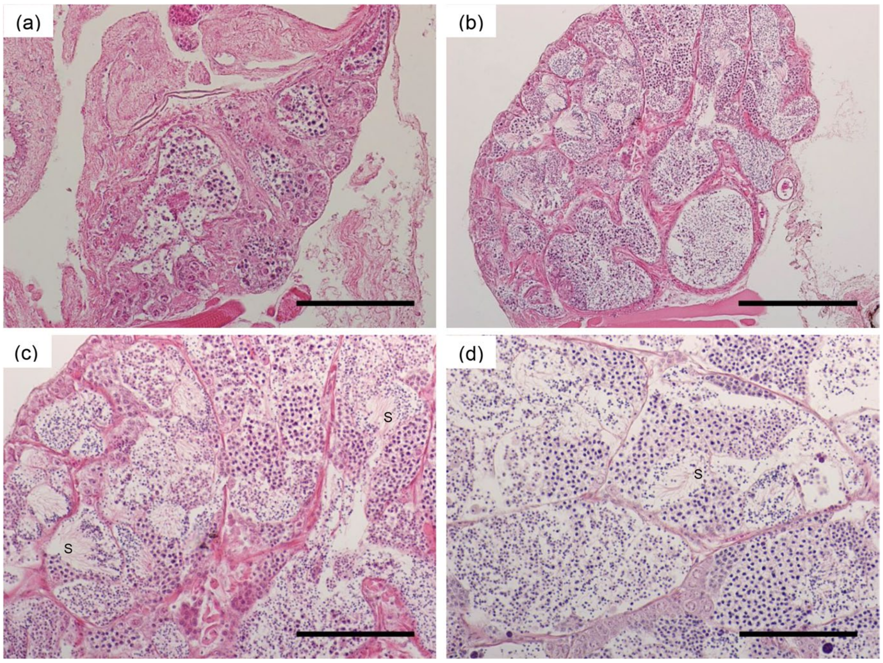

Fig. 3 Male reproductive states for *Zebrasoma flavescens.* **a** Immature male (64-mm TL; scale bar is 100 μ), **b** mature male with spermatozoa (S) (70 TL; scale bar is 200 μm), **c** increased magnification of **b** (scale bar is 100 μm), and **d** further increased magnification of a mature male with spermatozoa (89-mm TL; scale bar is 100 μm)

The female gonadosomatic index (GSI) ranged from 0.07 to 2.66, while the male GSI range was much smaller (0.03 – 0.84). However, the GSI was highly variable with both low and high GSI estimates for females and males larger than 60-mm TL (Fig. 5). Actively spawning females (classified as having recent post ovulatory follicles and/or hydrated oocytes) GSI ranged from 0.07 to 2.47. Maximum gonad weights were 0.26 g for females and 0.10 g for males.

Discussion

Our research found that *Zebrasoma flavescens* collected from the aquarium fishery on Oahu, Hawai'i, are capable of spawning around 65-mm TL. This was surprising because individuals this small were previously considered well within the juvenile stage of their life history. However, these small individuals are unlikely to contribute significant egg production.

We found a size difference between female and male size compositions and mean length, which was also observed by Claisse et al. (2009b). Claisse et al. (2009b) found that males had higher growth rates than females when in deeper water habitats (i.e., younger). This leads to sexual dimorphism, with males tending to be larger than females (Claisse et al. 2009b). Given that females mature and spawn at a very small size compared to males, their earlier shift in energy allocation from growth to reproduction results in the observed sexual dimorphism in size.

The size distribution and mean size of the *Z. flavescens* sampled for this research from the Oahu aquarium fishery (55–110-mm TL; mean 78-mm TL) were similar to those from the West Hawai'i aquarium fishery (35–149-mm TL; mean 71-mm TL) with the exception of our sampling missing the smallest and largest individuals (Stevenson et al. 2011). Effort was made to sample across the full size range from the aquarium fishery; however, the smallest and largest individuals present in the Hawai'i aquarium fishery were not observed in the aquarium fishing locations sampled on Oahu.

Prior *Z. flavescens* studies estimate size at maturity at 80–90-mm TL (Walsh 1984), and size at first spawning at 90.7 TL (Bushnell et al. 2010). This study's finding of *Z. flavescens* size at maturity at 63.4-mm TL is well below the expected size at maturity and prior estimates. Size at maturity is generally confined to life history constraints, often referred to as Beverton-Holt invariants, and is thought to be an evolutionary constraint to maximize reproductive output (Charnov 1993, 2008; Jensen 1996). For surgeon

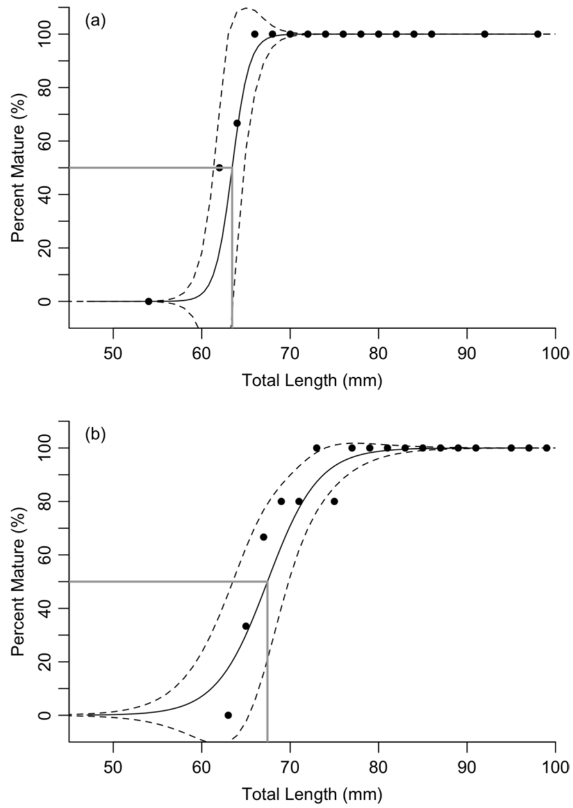

Fig. 4 Size at maturity for *Zebrasoma flavescens* **a** females (63.4 mm TL; CI: 62.7 – 65.5-mm TL), and **b** males (67.4-cm TL; CI: 66.4 – 70.3-mm TL) from the aquarium fishery. Dashed lines represent predicted 95% confidence intervals

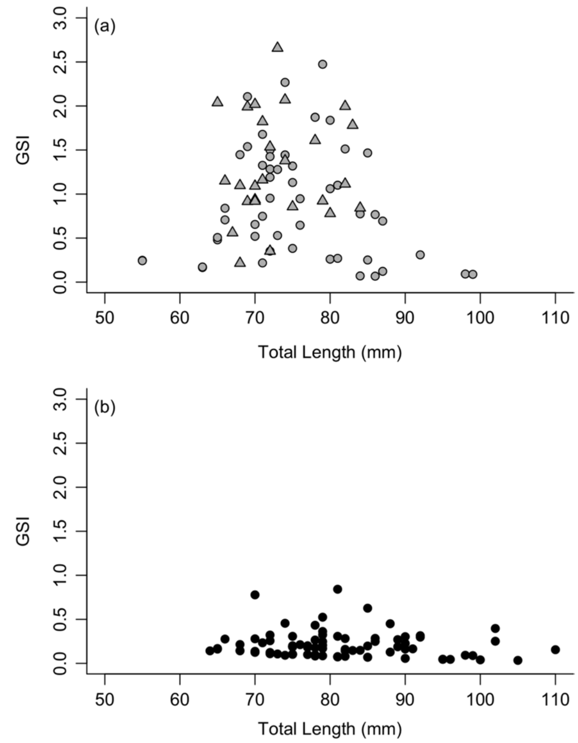

Fig. 5 Gonadosomatic index for *Zebrasoma flavescens* **a** females and **b** males assessed from the aquarium fishery. Actively spawning females are represented by triangles

fishes such as *Z. flavescens*, predicted size at maturity is estimated at 0.79 of asymptotic size (Nadon and Ault 2016). For *Z. flavescens* females with an asymptotic size of 156-mm TL (Claisse et al 2009b), the Beverton-Holt invariants ratio for surgeon fish would predict a size at maturity around 123-mm TL. This estimate is nearly double the size at maturity found in this study.

Zebrasoma flavescens ages at maturity, based on the published growth curve (Claisse et al. 2009b), are estimated at 6.5 months for females and 7 months for males. This estimated age at maturity for *Z. flavescens* is young compared to that of other Acanthurids (Taylor et al. 2014). While age at maturity can vary widely across species, it is not unusual for acanthurid species to reach maturity fairly rapidly. In Hawai'i, *A. blochii*, *A. dussumieri*, *A. olivaceus*, and *A. xanthopterus* reach maturity between 1 and 3.0 years (Pardee et al. in review). Furthermore, in Hawai'i, *Acanthurus triostegus* females mature around 440 days and males mature around 168 days (Longenecker et al 2008). Macro-increments (annual deposition) have been validated for several acanthurids (Choat and Axe 1996), while micro-increments which are assumed to be deposited daily have not been validated for most species and discrepancies have been found between micro-increments and macro-increments (Langston et al 2009). Given these discrepancies, we have chosen not to include an estimate of age at maturity in this study. Further research is needed to validate the daily formation of increments to determine age at maturity.

Reproductive output from the *Z. flavescens* size range targeted by the aquarium fishery is likely very low. *Zebrasoma flavescens* spawning activity occurs throughout the year with peaks in spawning during late spring with monthly peaks in reproductive output following a lunar cycle (Bushnell et al. 2010). Our sampling occurred across multiple different moon phases during the peak spawning season (March

– July) (Bushnell 2007). Maximum reproductive output occurs during the full moon, but spawning occurs throughout the month with the possible exception of new moon (Bushnell et al. 2010). We observed a relatively high occurrence of hydrated oocytes, indicating spawning within 12 h (Bushnell 2007), with no apparent lunar pattern. *Zebrasoma flavescens* GSI during peak spawning can be as high as 8–9 (Bushnell 2007); however, the GSI for actively spawning females sampled from the aquarium fishery (this study) is on the lower end for spawning females (< 2.7). There is a strong relationship between GSI and fecundity for *Z. flavescens*, with a range of 0–5000 eggs for females with a GSI at or below 3.0 and up to 28,000 eggs for females with a GSI around 8.0–9.0 (Bushnell 2007). The smallest female sampled for fecundity was 103-mm SL with a batch fecundity of 919 eggs (Bushnell 2007). Significant egg production (~5000 eggs per batch) occurs in females > 118-mm SL (Bushnell 2007). Furthermore, we found that female gonad weights for *Z. flavescens* from the Oahu aquarium fishery were extremely low (< 0.27 g), limiting any significant egg production.

It remains unknown if the small size at maturity is representative of populations across Hawai‘i. Fish life history, including size at maturity, can vary across small scales, including among islands within the Hawaiian Islands (Depczynski and Bellwood 2006; Taylor and Choat 2014; Schemmel and Friedlander 2017). This research was based on populations from Oahu (North and West Oahu), while prior life history assessments for Z. *flavescens* in Hawai‘i have focused on West Hawai‘i (Claisse et al. 2009a, b; Bushnell et al. 2010). Coral reef habitats vary between Oahu and West Hawai’i, with Oahu having lower overall percent coral cover and lower percent cover of *Porites compressa* (Franklin et al 2013; Donovan et al 2018; Asner et al 2020), the coral species associated with *Z. flavescens* (Walsh 1984). Furthermore, differences in reef regimes, measured by differing states of benthic cover and fish assemblages, likely influence growth and thus size at reproductive maturity. Oahu is predominately regime 1, low coral cover and low fish biomass, whereas West Hawaii regime 5, characterized by high coral cover and moderate fish biomass (Donovan et al 2018). The differences in coral reef habitat and fish biomass (including conspecifics) between the islands can lead to differences in key habitats for refugia and feeding and density dependent on survival and growth (Hayes et al. 1996; Lorenzen and Enberg 2002; Claisse et al. 2009a).

Historical and recent fishing pressure also differs between Oahu and West Hawai‘i. The aquarium fishery began in the 1970s and expanded rapidly (Walsh et al. 2004). Historically, most of the aquarium fishing was centered on Oahu; however, the majority of the catch in the later years came from West Hawai‘i (Walsh et al. 2004). It is speculated that local depletion and hurricane impacts resulted in the shift in effort and catch of aquarium fish between the regions (Walsh et al. 2004).

Understanding if the small size at maturity is natural or was influenced by high fishing pressure and selectivity is an important question to address as small size at maturity may constrain the reproductive output of the individual. Increased mortality and fishery selectivity of small individuals have been shown to reduce size and age at maturity in a variety of animals (Allendorf and Hard 2009), including many fisheries species (reviewed in Kuparinen and Merilä 2007). Alternatively, the aquarium fishery may remove the slower growing individuals, resulting in faster growing individuals remaining in the population which may allocate more energy to growth and thus reach a larger size at maturity (Rochet 1998). Therefore, further research is needed on the potential variability in size at maturity between locations and across varying levels of fishing intensity. This will enable understanding of the variability in reproductive characteristics for this species and possibly determine the impact, if any, that the aquarium fishery had on the life history of *Z. flavescens.*

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Data availability Data and metadata is available at Pacific Islands Fisheries Science Center. 2020. *Life History Program Life History* Estimates, https://inport.nmfs.noaa.gov/inport/item/59002.

Declarations

Ethics approval Fish handling methods were in accordance with principles detailed in Guidelines for the Use of Fishes in Research (American Fisheries Society), and in the U.S. Government Principles for the Utilization and Care of Vertebrate Animals Used in Testing, Research, and because fish were donated, a formal protocol was waived by the regulating agency University of Hawai‘i Animal Care and Use Committee.

Conflict of interest The author declares no competing interests.

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