

Reproductive biology of *Lethrinus harak* on Guam

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TABLE OF CONTENTS

SUMMARY	3
INTRODUCTION.....	4
METHODS	5
Processing of gonads	5
Maturation and sex reversal	6
Reproductive seasonality	6
Reproductive development	6
RESULTS.....	6
Ovarian stages.....	7
Seasonal variability	8
Developmental ontogeny	8
DISCUSSION	26
CONCLUSIONS AND RECOMMENDATIONS	28

SUMMARY

The thumbprint emperor or Mafute (*Lethrinus harak*) is an important member of Indo-Pacific coral reef ecosystems and a primary component of the local recreational fishery. To assess reproductive seasonality and size and age at sexual maturity, 414 *L. harak* were sampled from local reefs, both inside and outside the Marine Preserves in 2007 and 2008. Histological examination of the gonads collected from numerous sites around the island indicates that *L. harak* is a protogynous hermaphrodite. Size and age at female maturation (20.8 cm and 4 yrs respectively) did not differ according to protection status, but there was some indication that age at sex change occurred earlier in fished populations. Little insight into the seasonality of reproduction was achieved because of a low number of mature females in monthly samples. We make several management recommendations.

INTRODUCTION

Reproductive patterns in coral reef fish have been shown to be complex as >50% of exploited species are now recognized as hermaphroditic (Sadovy 1996). Fish that change sex at some point in their ontogeny can exhibit protandry (functional males switch to functional females), protogyny (functional females switch to functional males), or simultaneous hermaphroditism (reproductive material of both sexes exists simultaneously). Protogyny has been established as the dominant sexual pattern in such families as Serranidae, Pomacanthidae, Lethrinidae, Labridae, and Scaridae, among others. Sex ratios for protogynous species typically change from female-dominated to male-dominated as size increases, though many factors can alter sex ratios so they should not be used to diagnose sexual patterns. Histological examination of gonads is required to identify key criteria which indicate protogynous hermaphroditism. Sadovy & Shapiro (1987) outline these criteria as follows: 1) membrane-lined central cavities in testes; 2) presence of transitional individuals; 3) atretic bodies of yolked oocytes within testes; and 4) sperm sinuses in the gonadal wall. Protogynous species can be monandric (exhibiting one developmental pathway; e.g., all males derived from females) or diandric (two male pathways; e.g., primary males develop from an immature stage and secondary males develop from mature females) (Sadovy & Shapiro 1987). Few studies have investigated developmental pathways in detail, though pathways appear to be diverse within and among families (Liu & Sadovy 2004). Liu & Sadovy (2004) highlight the importance of sampling all possible size classes, including juvenile stages, to understand male developmental pathways, as they can influence the way a species or population responds to fishing pressure.

Fishing alters the size and age structure of a population non-randomly as larger fish are more heavily targeted. For protogynids, this non-random selectivity leads to reduction in males, which typically constitute a small proportion of the total population (Bannerot et al. 1987; Jennings & Lock 1996; Sadovy 1996). This becomes problematic in fisheries biology because it often leads to sperm limitation which greatly reduces the reproductive capacity of the population and could cause recruitment failure (Coleman et al. 1996). Most current measures of spawning potential consider reproductive capacity to be highly correlated with spawner biomass (the total biomass of sexually mature fish; Coleman et al. 2000). While this relationship is accurate for gonochorists, Coleman et al. (2000) proposed that a 90% reduction of males (potentially constituting only a small proportion of a protogynous population) would result in a 90% reduction in the reproductive capacity of the population. In addition, assuming that male and female reproductive capacity increases with fish size (which has been demonstrated), the magnitude of reproductive decline would be even greater as both males and females are size-selectively targeted.

Exploitation has been demonstrated to alter the size or age at sex reversal. Mechanisms causing sexual transition in reef fish can be either endogenous (strict internal schedule

such as absolute size or age) or exogenous (social characteristics such as sex ratio or relative size of other fish) (Armsworth 2001). The latter often function as compensatory mechanisms for heavy fishing pressure. Such mechanisms include accelerated maturation, accelerated sex reversal, accelerated growth, and combinations thereof (Huntsman & Schaaf 1994; Huntsman et al. 1999). When sex reversal patterns and compensatory mechanisms are not accounted for, the applicability of stock assessment models becomes problematic, which further complicates the management of protogynous reef fish populations (Buxton 1992; Sadovy & Figuerola 1992).

In the genus *Lethrinus*, protogyny appears to be the dominant sexual pattern. Protogyny has been confirmed for six species (*L. atkinsoni*, *L. genivittatus*, *L. lentjan*, *L. miniatus*, *L. rubrioperculatus*, and *L. variegatus*) and has been suggested for many others, including *L. harak*, while gonochorism has been suggested for *L. atkinsoni*, *L. nebulosus*, and *L. obsoletus* (Young & Martin 1982; Ebisawa 1990, 1997, 1999; Bean et al. 2003; Sumpton & Brown 2004; Ebisawa 2006; Sadovy de Mitcheson & Liu 2008). Lethrinid sexual patterns are briefly reviewed in Sadovy de Mitcheson & Liu (2008). The reproductive biology of *L. harak* has been studied at least four separate times prior to the present study. Hilomen (1997) and Lassi (2003) studied the size and age at sexual maturity from the GBR and Fiji, respectively, and both suggested protogyny while Kulmiye et al. (2002) studied *L. harak* from Kenya and suggested protandry based on size-frequency distributions. Ebisawa (2006) provided the most comprehensive investigation of *L. harak* reproductive biology from Japan and suggested protogyny based on histological evidence.

In this chapter, I investigate the reproductive biology of *L. harak* on Guam with respect to size and age at maturation and sex reversal, spawning periodicity, and sexual pattern. Parameters are also compared between populations from protected and unprotected sites to determine whether protection status has an impact on reproductive biology. Such information has important management implications for insuring a healthy and reproductively viable population.

METHODS

Processing of gonads

Specimens of *L. harak* were sampled from various sites around Guam from June 2007 to June 2008 as in Chapter 3. Whole gonads removed from each individual were blotted dry, weighed to the nearest 0.001 g, sexed and staged macroscopically, and immediately preserved in FAACC (formaldehyde 4%, acetic acid 5%, calcium chloride 1.3%; Samoilys & Roelefs 2000) for histological processing. Sections of gonads embedded with paraffin wax were mounted on glass slides and stained with Haematoxylin and Eosin. These sections were viewed under a high powered microscope to confirm macroscopic sexes and stages and to investigate reproductive ontogeny. Individual female oocytes were classified into five stages following West (1990): chromatin nucleolar, perinucleolar,

cortical alveoli, vitellogenic, and ripe (Figure 1). To better understand the relationship between female body size, age, and reproductive contribution, ovary weight was used as a proxy for reproductive capacity and plotted with the VBGF for immature and active females (excluding resting and spent individuals) following Pears et al. (2006).

Maturation and sex reversal

Age and size of female maturation and age and size at sex reversal were explored by plotting the proportion of mature individuals or males over the various size or age classes, respectively. A sigmoidal curve was fitted to the data by minimizing the sums of squares using the variables a , b , and x_0 , where

$$y = \frac{a}{1 + e^{-((x-x_0)/b)}}.$$

Ages and sizes of maturation and sex reversal were compared graphically between populations within and outside of marine preserves. Sex ratios by age were compared between these populations using a replicated G test of goodness of fit.

Reproductive seasonality

Seasonality in the reproductive biology of *L. harak* was investigated using monthly gonadosomatic indices (GSI). GSI plots represent the proportional relationship between gonad weight and body weight and follow the assumption that gonad weight increases during times of spawning as a result of the swelling and ripening of oocytes. The frequency and occurrence of transitional individuals, spent individuals, and resting individuals were related to the GSI and to the mean monthly SST during the months fish were sampled. Transitional individuals contain proliferating reproductive material of one sex and degenerative material of the other sex. Sexual transition in some species of tropical reef fishes has been demonstrated to occur after spawning periods and is generally a quick process (Reinboth 1962, Chan & Phillips 1967, Kawaguchi & Marumo 1967, Bruslé & Bruslé 1975, Fishelson 1975, Warner 1975, Dipper & Pullin 1979, Jones 1980, Clavijo 1982). Hence, it is considered in this study as potential insight to spawning seasonality.

Reproductive development

Histology sections of *L. harak* gonads over a wide range of body sizes and ages were examined to confirm whether or not *L. harak* is a functional hermaphrodite. Evidence indicative of protogyny, protandry, and sequential hermaphroditism is outlined in Sadovy & Shapiro (1987), and these criteria were used to infer the sexual ontogeny of the species.

RESULTS

Gonads of 414 *L. harak* individuals were examined. These individuals ranged in size from 83 to 327 mm as in Chapter 3. The overall sex ratio was approximately M:F = 1:3.80, though this varied by size and age (Tables 1 and 2). Females were the dominant

sex in lower size and age classes whereas males were dominant in the greater size and age classes. However, the three largest specimens sampled were all female. High variability in sex ratios among monthly samples indicates that for some months, an inadequate sample size was obtained (Table 3).

Ovarian stages

The ovarian development and maturation of female *L. harak* was classified into six stages: immature; maturing; mature, resting; mature active; ripe; and spent (Table 4). An additional stage, 'running ripe,' is often included in studies of reef fish reproductive biology, however, no individuals matching criteria common for this stage were encountered. It is highly likely, however, that *L. harak* undergoes a running ripe condition just prior to spawning. Of 327 females, 64 percent were classified as immature, 3.7% maturing, 5% resting mature, 17% mature active, 9.5% ripe, and 1% spent. Immature individuals comprised the majority of the female catch for 10 of the 13 months (Figure 2). Maturing females were rare and were found within the 185 to 235 mm size range (Figure 3). Macroscopic staging and sex determination proved to be inadequate as female staging agreed 76% of the time and sex determination agreed 92% of the time between macroscopic and histological techniques. Sexes and stages determined through histology were used in all plots and analyses.

There was a strong linear relationship between female gonad weight and fork length when gonad weight was log-transformed (Figure 4a). Similarly, there was a strong logarithmic relationship between female gonad weight and age when gonad weight was log-transformed (Figure 4b). Plotting ovary weight as a proxy for female reproductive capacity with the VBGF demonstrated the increase in reproductive potential for females with size and age (Figure 5). The largest female sampled had a whole gonad weight of over 45 grams and a GSI value which was 4.6 times greater than the mean GSI for all other active females.

Size and age at female maturation and sex reversal

Female *L. harak* mature over a relatively narrow size range where the length at 50% maturity (L_{50}) was 208 mm FL (Figure 6a). The age at 50% maturity (t_{50}) was approximately 3.8 years (Figure 6b). When compared between populations within and outside of marine preserves on Guam, size and age at maturity differed only slightly, which may be explained by variability in sample size (Figure 7). The length and age at 50% sex reversal was 241 mm FL and 5.38 years, respectively (Figure 8). The length at sex reversal differed only slightly between protected and unprotected populations (Figure 9a), but the difference for age at sex reversal was more pronounced. Male-female sex ratios were 1:1 at an estimated 5.2 years of age outside of marine preserves compared to an estimated 6.1 years within (Figure 9b). A replicated G test of goodness of fit determined that sex ratios by age were not significantly different between protection statuses as sample sizes for older ages were inadequate to detect differences ($G=4.963$, $df=7$, $p=0.664$).

Seasonal variability

Unfortunately, a low monthly sample size of reproductively active females hindered the efficacy of monthly gonadosomatic indices (GSI) in identifying the spawning season of *L. harak*. The minimum target sample size for mature females for this study was 15 per month. This was only achieved for two of thirteen months (Figure 10). For most months, the sample size was too low to obtain an adequate representation of the mean GSI. However, taken as it is, the monthly GSI plot indicates that the period of highest reproductive activity is between October and January (Figure 10). There is some correspondence between the monthly GSI values and the mean monthly sea surface temperature (SST) on Guam, indicating that the spawning season begins as the SST decreases from the annual high (Figure 11). In addition, resting individuals were encountered during months of low GSI values. The most convincing evidence of spawning periodicity is that transitional individuals were only encountered in seven months of the year, six of which were consecutive. One transitional individual was encountered in February and nine others were encountered from May to October. Except for the occurrence in February, the presence of transitional fish corresponded with the yearly increase in mean SST (Figure 12). If sex reversal occurs after spawning periods for *L. harak*, this may be evidence for a defined spawning season on Guam.

Developmental ontogeny

A total of ten transitional *L. harak* individuals were identified from the 414 gonads examined microscopically. Transitional individuals contained degenerative atretic oocytes in the presence of mature or maturing male reproductive material (Figure 13). All individuals were above the size and age at female maturation. Other features that were common in male *L. harak* gonads that are indicative of protogynous hermaphroditism included peripheral dorsal sperm sinuses and a remnant ovarian lumen (Figure 14). The presence of sperm crypts among degenerative female tissue suggests that *L. harak* male and female tissues are not delimited by connective tissue during sex reversal which is common in both serranids and scarids (Smith 1965; Choat and Robertson 1975; Figure 15).

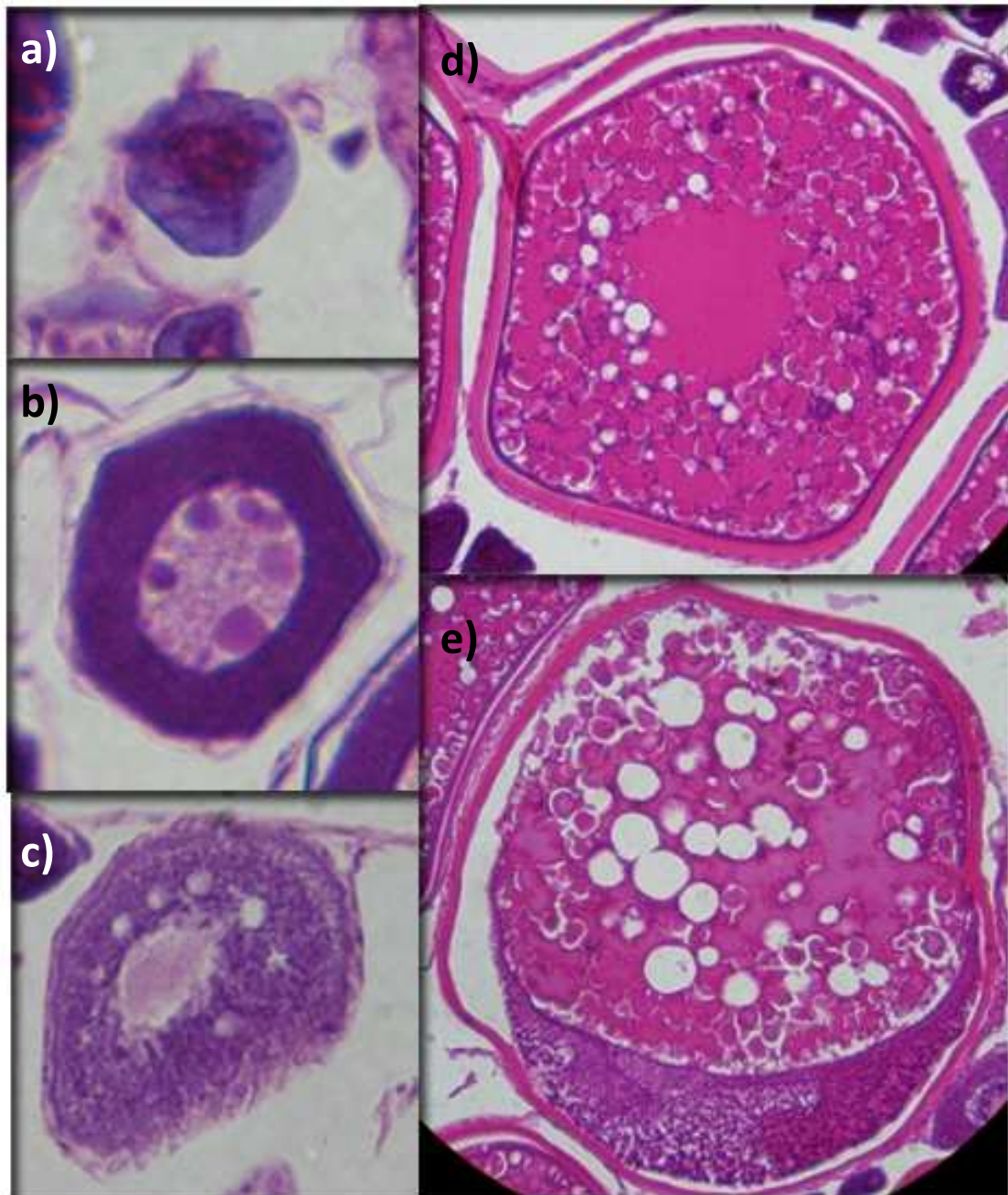


Figure 1: Stages of oogenesis in *Lethrinus harak* following West (1990), a) chromatin nucleolar, b) perinucleolar, c) cortical alveoli, d) vitellogenic, and e) ripe.

Table 1. Sex ratios and total sample sizes of *Lethrinus harak* by size class.

Size class FL (mm)	Total	Unsexed	Males	Females	% males	Sex ratio M:F
80-99	10	0	0	10	0.0	
100-119	12	0	0	12	0.0	
120-139	28	0	0	28	0.0	
140-159	33	0	0	33	0.0	
160-179	43	0	1	42	2.3	1 : 42.0
180-199	68	0	2	66	2.9	1 : 33.0
200-219	85	0	15	70	17.6	1 : 4.67
220-239	70	0	24	46	34.3	1 : 1.92
240-259	40	1	24	16	61.5	1 : 0.63
260-279	16	0	15	1	93.8	1 : 0.07
280-299	7	0	5	2	71.4	1 : 0.40
300-319	1	0	0	1	0.0	
320-339	1	0	0	1	0.0	
Total	414	1	86	328	20.8	1 : 3.80

Table 2. Sex ratios and total sample sizes of *Lethrinus harak* by age class.

Age class (years)	Total	Unsexed	Males	Females	% males	Sex ratio M:F
1	28	0	0	28	0.0	1 :
2	77	0	0	77	0.0	1 :
3	99	0	5	94	5.1	1 : 18.8
4	80	0	17	63	21.3	1 : 3.71
5	40	1	17	23	43.6	1 : 1.29
6	19	0	12	7	63.2	1 : 0.58
7	26	0	16	10	61.5	1 : 0.63
8	12	0	7	5	58.3	1 : 0.71
9	7	0	5	2	71.4	1 : 0.40
10	3	0	2	1	66.7	1 : 0.50
11	4	0	2	2	50.0	1 : 1.00
12	2	0	0	2	0.0	1 :
13	1	0	0	1	0.0	1 :
Total	398	1	83	315	20.9	1 : 3.78

Table 3. Sex ratios and total sample sizes from monthly samples of *Lethrinus harak* from Guam.

<i>Month</i>	<i>Total</i>	<i>Unsexed</i>	<i>Males</i>	<i>Females</i>	<i>% males</i>	<i>Sex ratio M:F</i>
June 07	30	0	4	26	13.3	1 : 6.50
July	20	0	9	11	45.0	1 : 1.22
Aug	24	0	7	17	29.2	1 : 2.43
Sept	33	0	7	26	21.2	1 : 3.71
Oct	33	0	5	28	15.2	1 : 5.60
Nov	17	0	3	14	17.6	1 : 4.67
Dec	36	0	2	34	5.6	1 : 17.0
Jan 08	17	0	2	15	11.8	1 : 7.50
Feb	20	0	4	16	20.0	1 : 4.00
Mar	13	0	5	8	38.5	1 : 1.60
Apr	96	0	17	79	17.7	1 : 4.65
May	35	1	10	24	29.4	1 : 2.40
June	31	0	10	21	32.3	1 : 2.10
Total	405	1	85	319	21.0	1 : 3.76

Table 4. Descriptions of stages of ovarian development in *Lethrinus harak*.

<i>Lethrinus harak</i>	Ovary	
	Macroscopic	Microscopic
Stage 1 - Immature	Gonad lobes thin and cylindrical, transparent and often with a pinkish tint. Lobes typically ≤ 2 cm in length. Oocytes and ovigerous folds not discernible.	Densely packed primary oocytes in the chromatin nucleolar and perinucleolar stages. Thin gonad wall and tight ovigerous folds.
Stage 2a - Maturing	Similar to stage 1 but more dense and usually with blood appearing. Slightly orange or yellow in color.	Ovaries tightly packed and dominated by perinucleolar stage oocytes that have multiple nucleoli easily visible at the periphery of the nucleus. Many cortical alveoli stage oocytes present which are ≥ 4 times larger than other previtellogenic oocytes. Thin gonad wall.
Stage 2b - Mature, resting	Long and slightly flaccid with blood vessels fading. Orange-brown in color and translucent towards edges.	Ovary dominated by previtellogenic primary oocytes (chromatin nucleolar and perinucleolar) but also with a large proportion of cortical alveoli stage oocytes. Gonad wall very thick and presence of brown bodies and atretic oocytes common.
Stage 3 - Mature active	Gonad lobes thick and peach in color. Blood vessels usually prominent. Oocytes and ovigerous folds visible through gonad wall.	Ovary dominated by vitellogenic yolk stage oocytes but with previtellogenic oocytes present in various proportions. Ovigerous lamellae disappearing or gone.
Stage 4 - Ripe	Similar to stage 3 only thicker and more tightly packed. Color can vary from brown-orange to light peach. Blood vessels often depressed but not always. Large oocytes visible through gonad wall but ovigerous folds not discernible.	Vast majority of oocytes in vitellogenic or ripe stages and densely packed. Gonad wall relatively thin and ovigerous lamellae not discernible. Large yolk proteins apparent in oocytes and nucleus often broken down.
Stage 5 - Spent	Gonad lobes completely flaccid and transparent. Often brown-orange in color. Blood vessels slightly visible.	Thick gonad wall that lacks definitive shape. Numerous previtellogenic stage oocytes scattered throughout and an abundance of brown bodies.

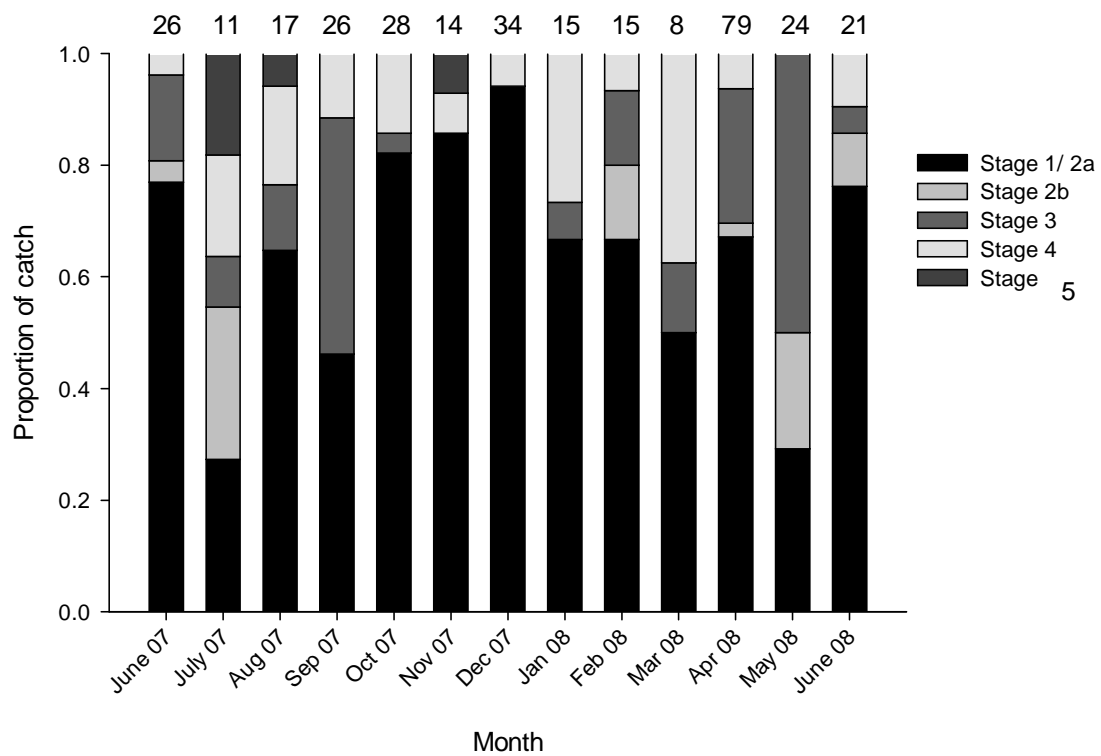


Figure 2: Proportions of ovarian stages of females in monthly samples of *Lethrinus harak* from Guam. Numbers over bars refer to sample size.

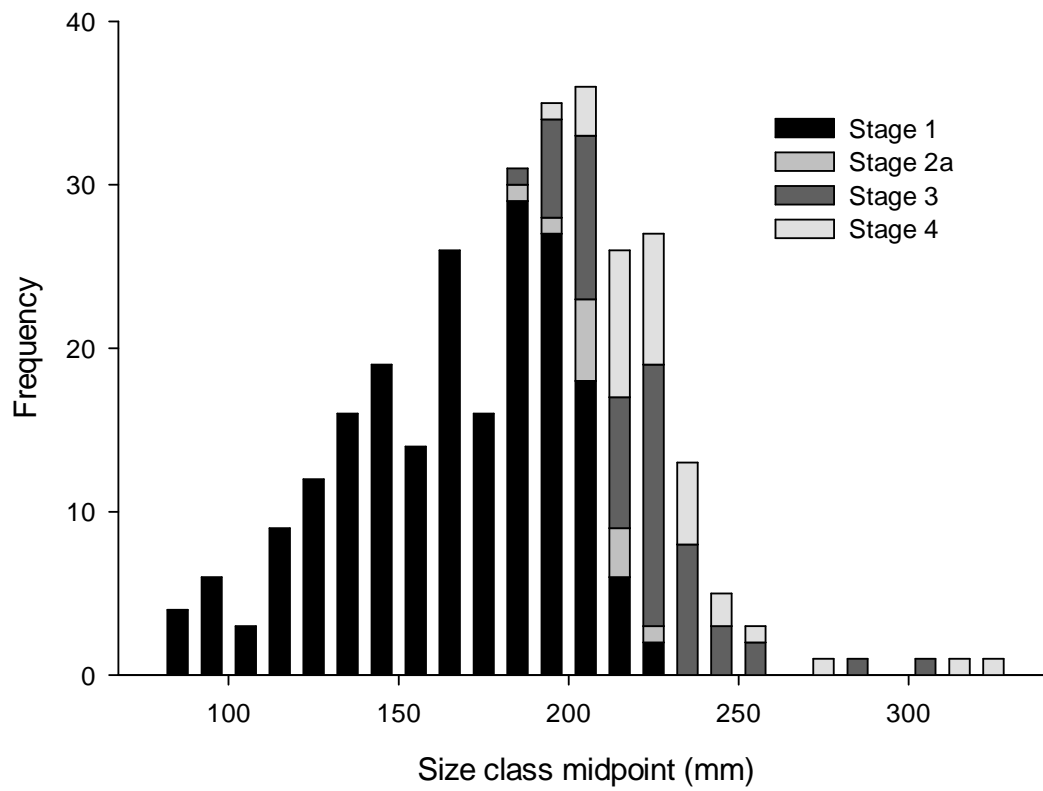


Figure 3: Size frequency distribution of female *Lethrinus harak* collected from June 2007 to June 2008 with proportions of ovarian stages by size class.

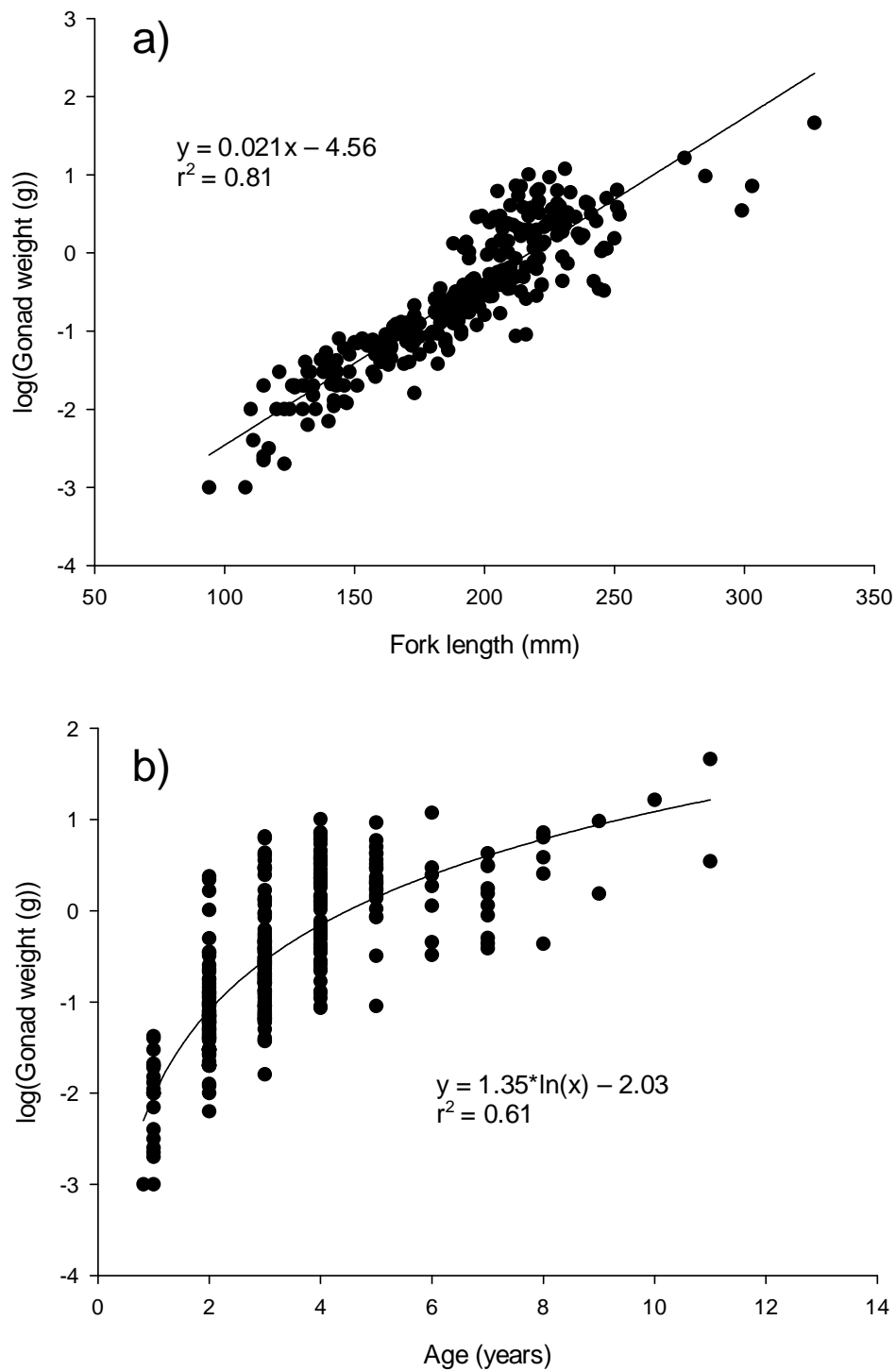


Figure 4: Relationship between a) fork length and gonad weight and b) age and gonad weight for reproductively mature *Lethrinus harak* females on Guam.

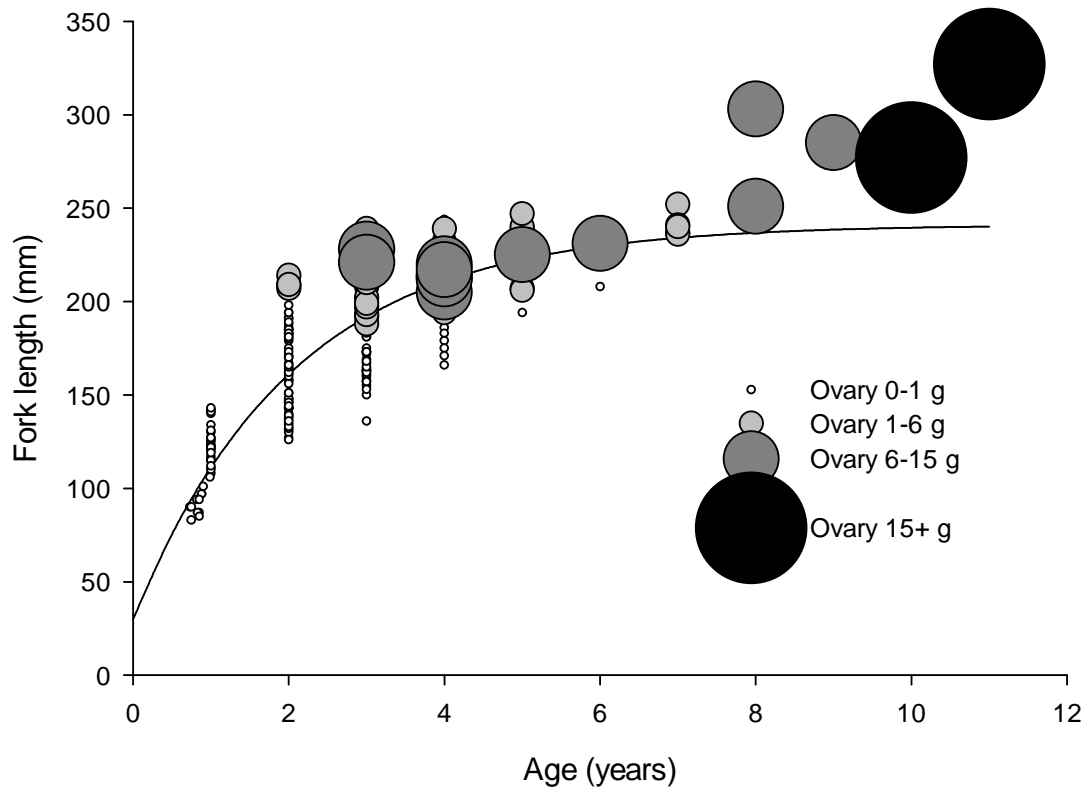


Figure 5: Ovary weight plotted over length-at-age data and VBGF for immature and active female *Lethrinus harak*.

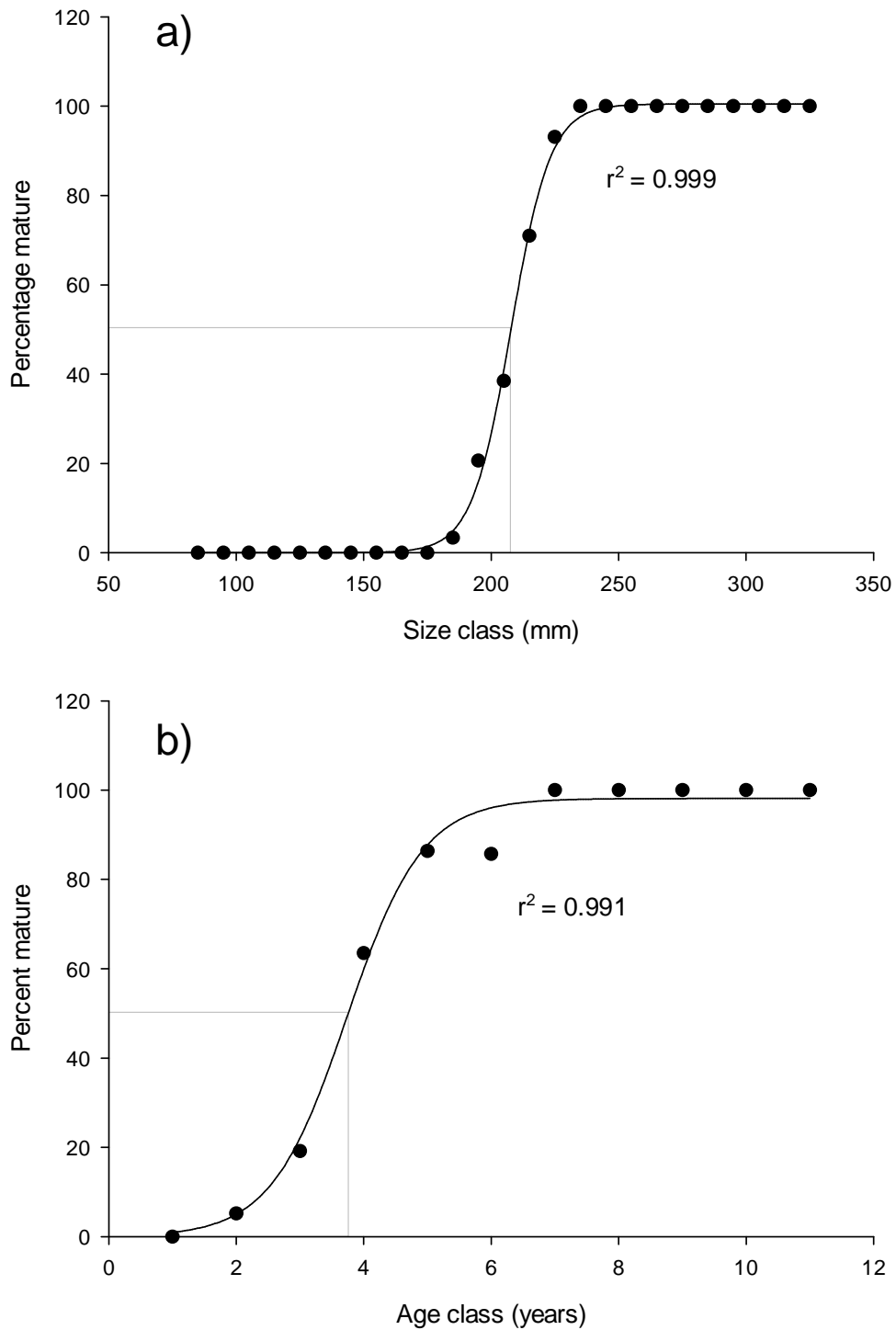


Figure 6: a) Length and b) age at female maturation represented by the proportion of mature individuals by size and age class.

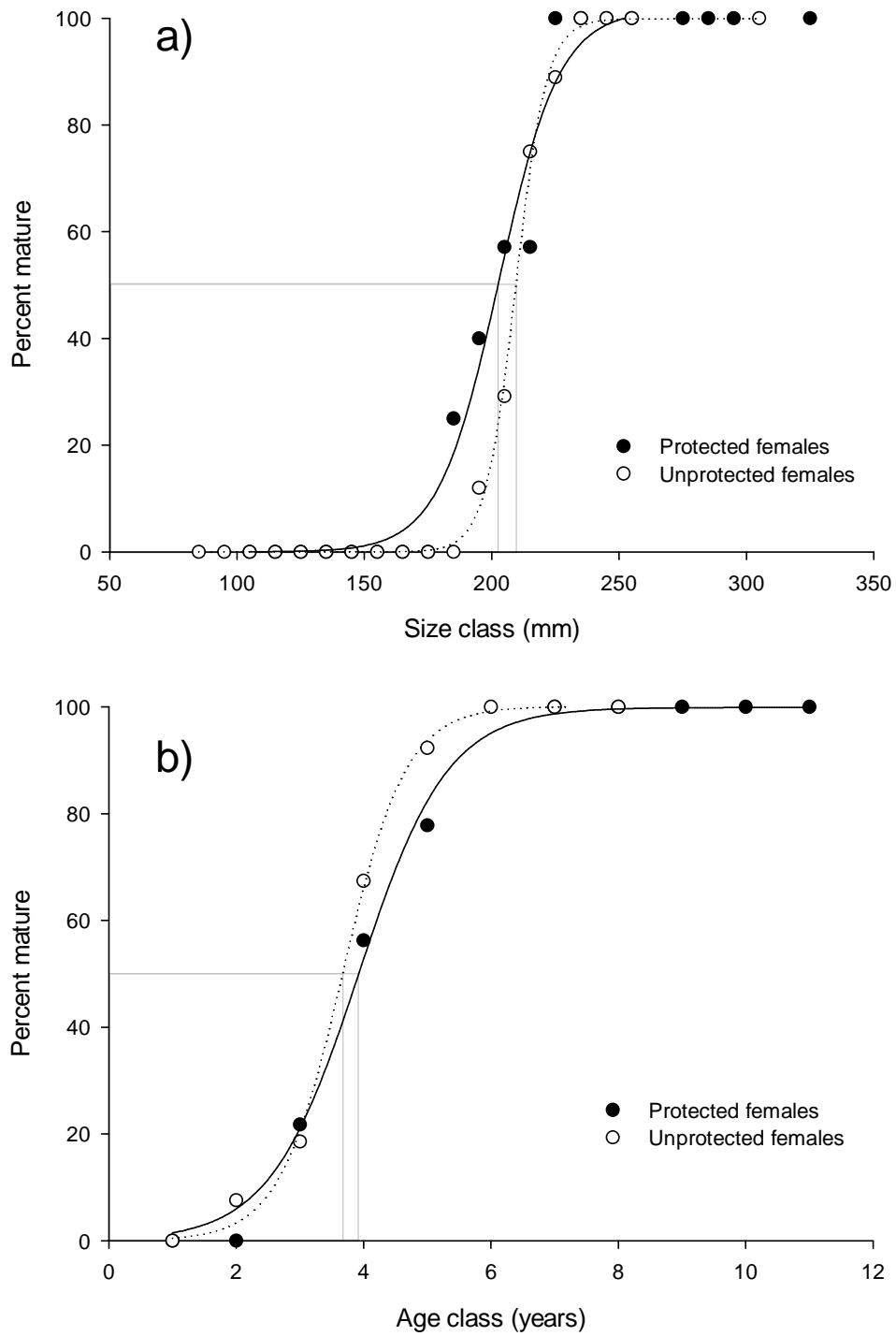


Figure 7: Comparisons of a) size and b) age of female maturation for *Lethrinus harak* between populations within and outside of marine preserves.

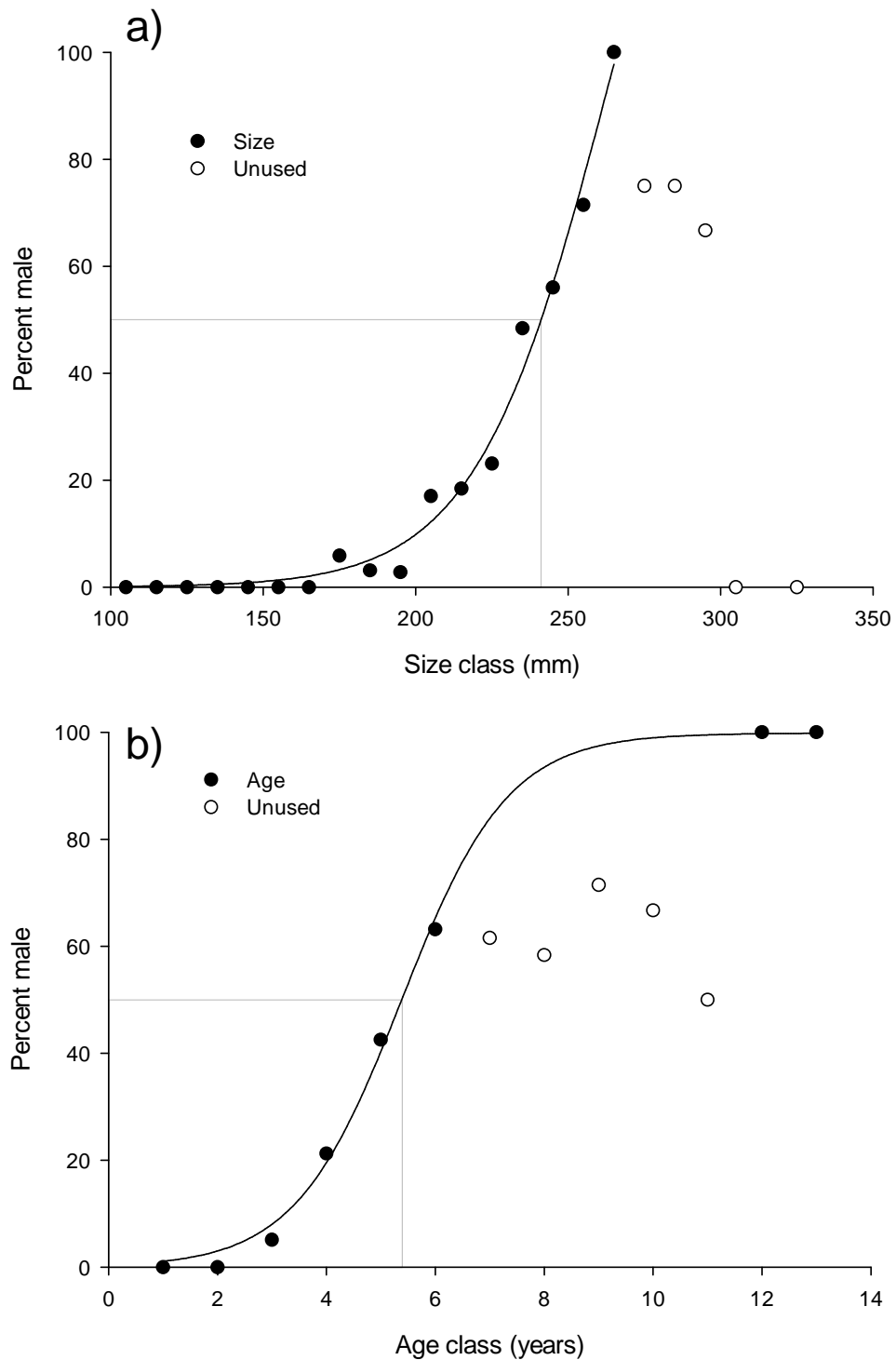


Figure 8: a) Length and b) age at sex reversal for *Lethrinus harak* represented by the proportion of males by size and age classes.

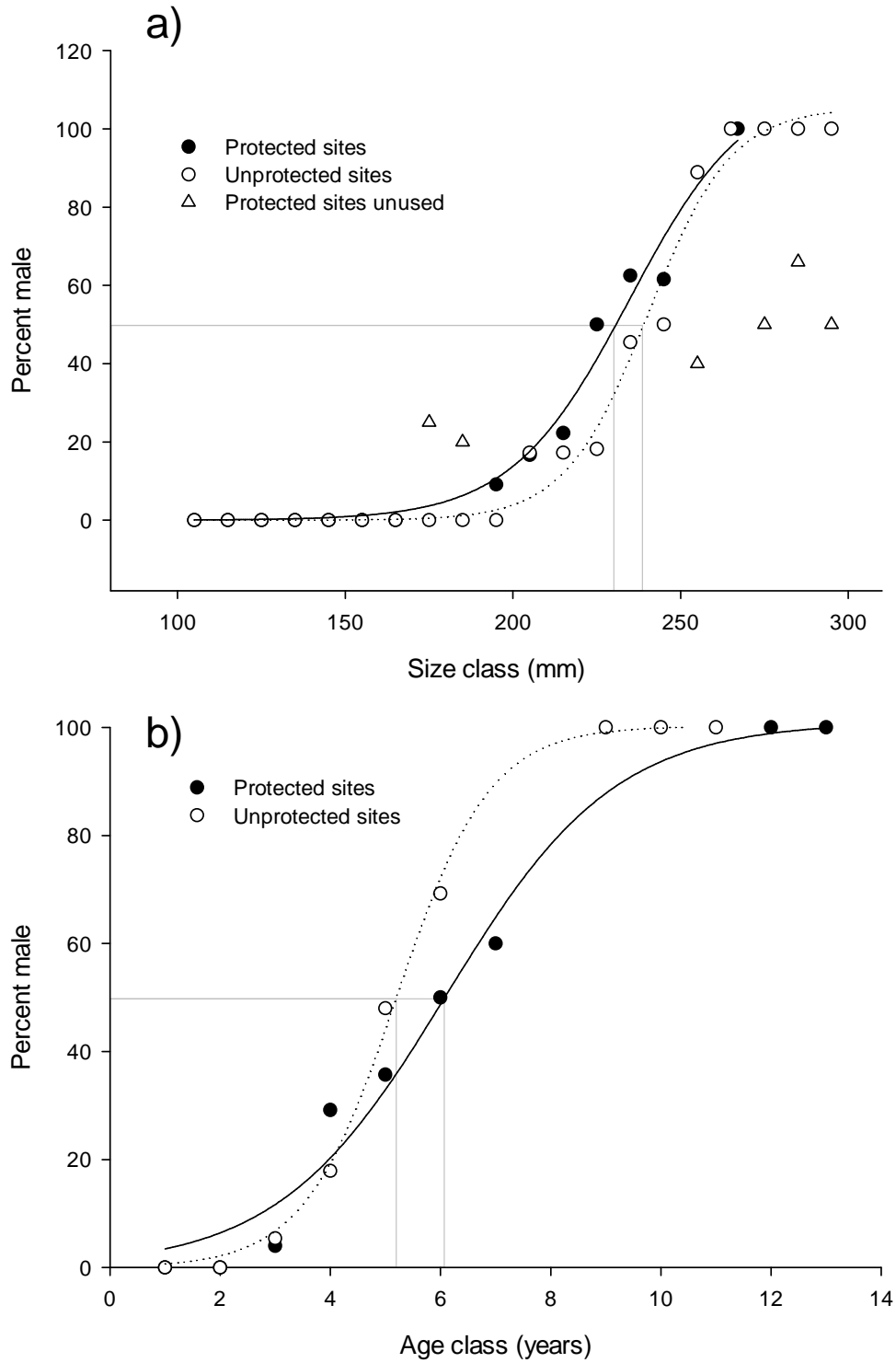


Figure 9: Comparisons of a) size and b) age of sex reversal for *Lethrinus harak* between populations within and outside of marine preserves.

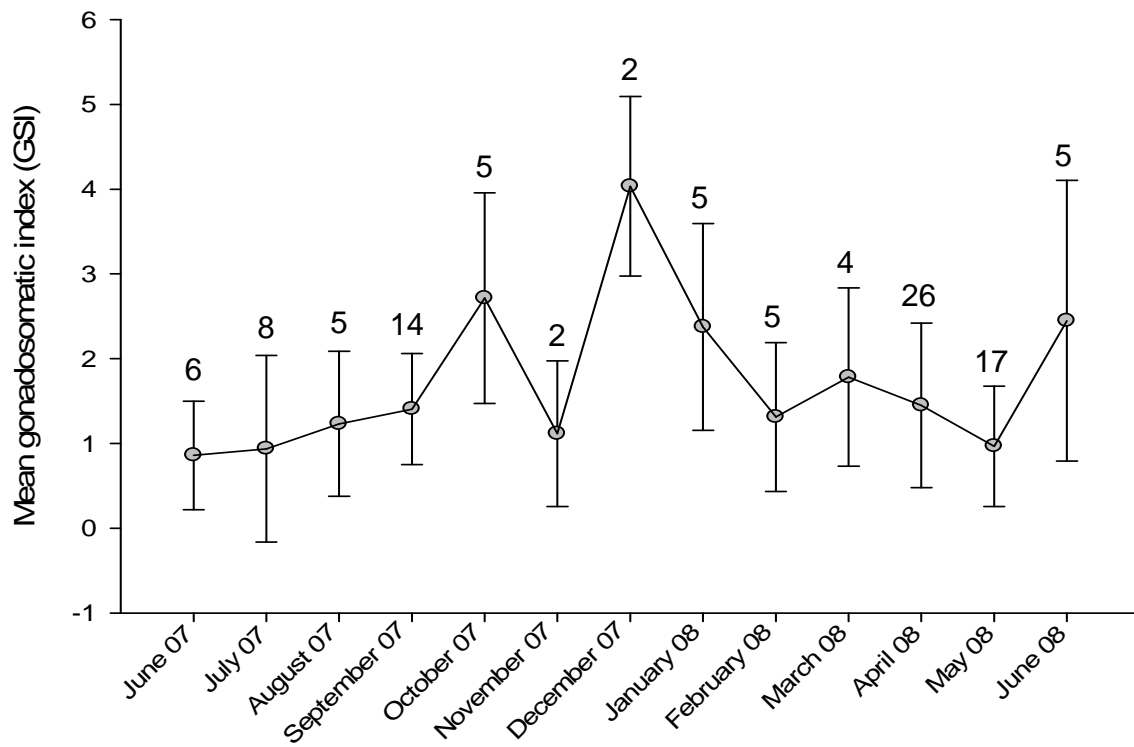


Figure 10: Mean monthly gonadosomatic indices for mature female *Lethrinus harak* from Guam. Error bars represent the standard error about the mean and numbers indicate sample size.

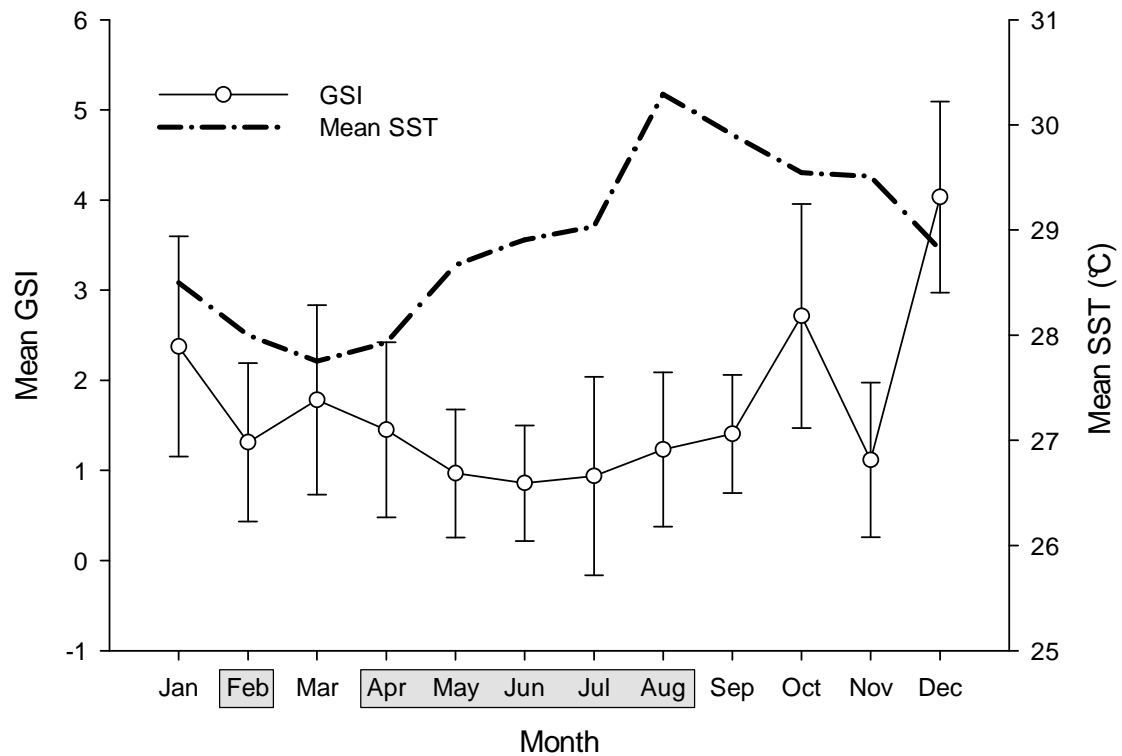


Figure 11: Plot of monthly gonadosomatic index values (+SE) for *Lethrinus harak* and mean sea surface temperatures over the period when sampling occurred. Grey boxes surround months in which resting mature females were encountered.

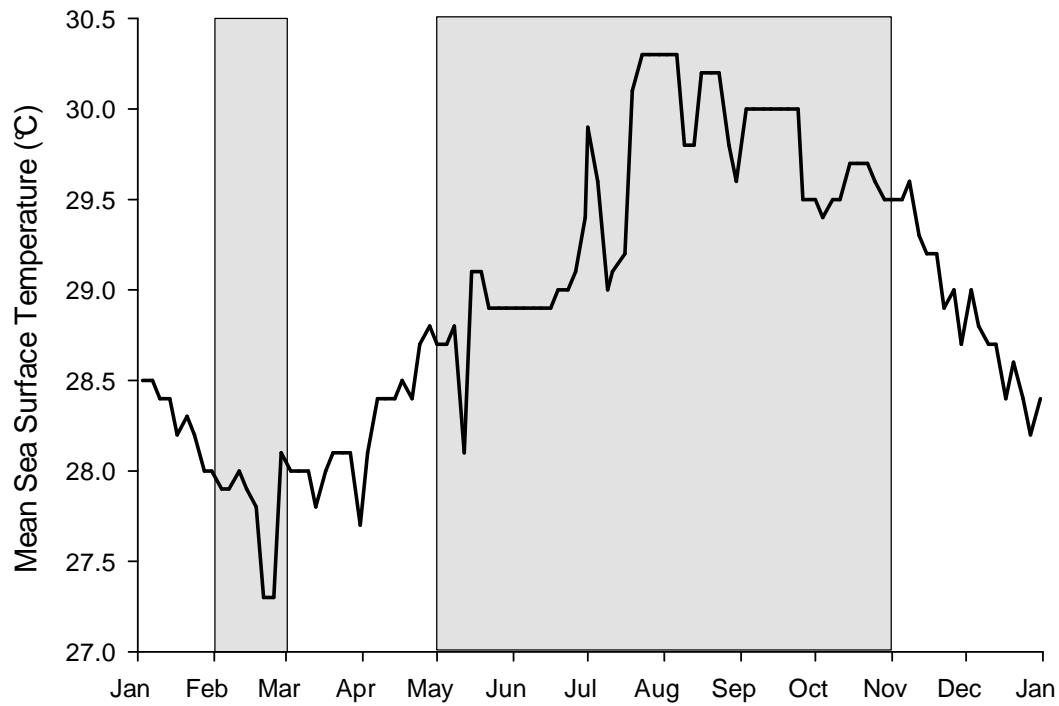


Figure 12: Mean sea surface temperature on Guam taken every three to four days and months in which transitional *Lethrinus harak* individuals were encountered (indicated by grey area).

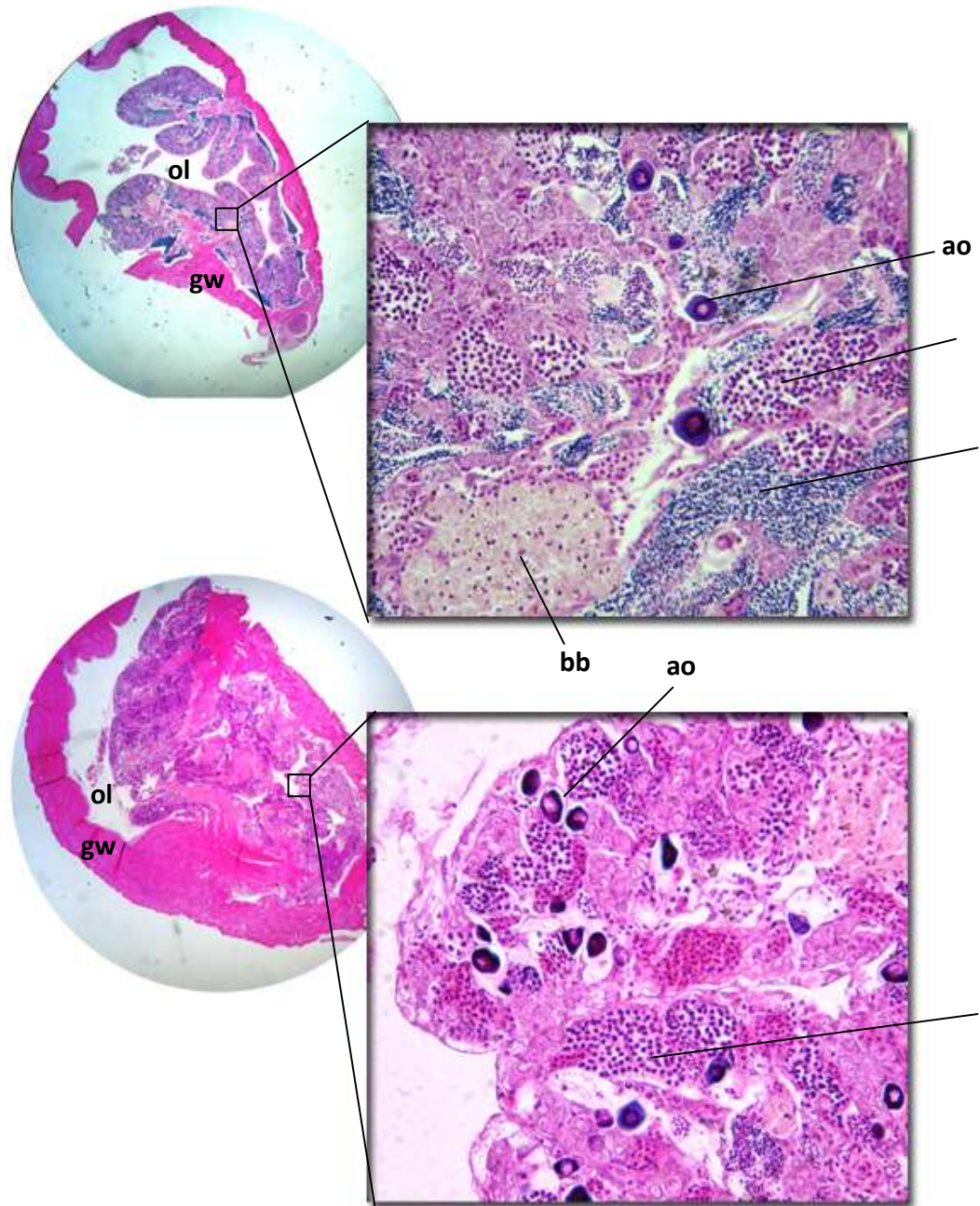


Figure 13: Pictures of two transitional *Lethrinus harak* (243 mm FL, 7 years old and 248 mm FL, 8 years old) at 4x (circle) and 40x (square) magnification. ao = atretic oocyte, bb = brown body, gw = gonad wall, ol = ovarian lumen.



Figure 14: Picture of a mature functional male *L. harak* (225 mm FL, 5 years old) displaying a peripheral dorsal sperm sinus filled with mature spermatogonia and a remnant ovarian lumen. bv = blood vessel, gw = gonad wall, ol = ovarian lumen, sg = spermatogonia, ss = sperm sinus.

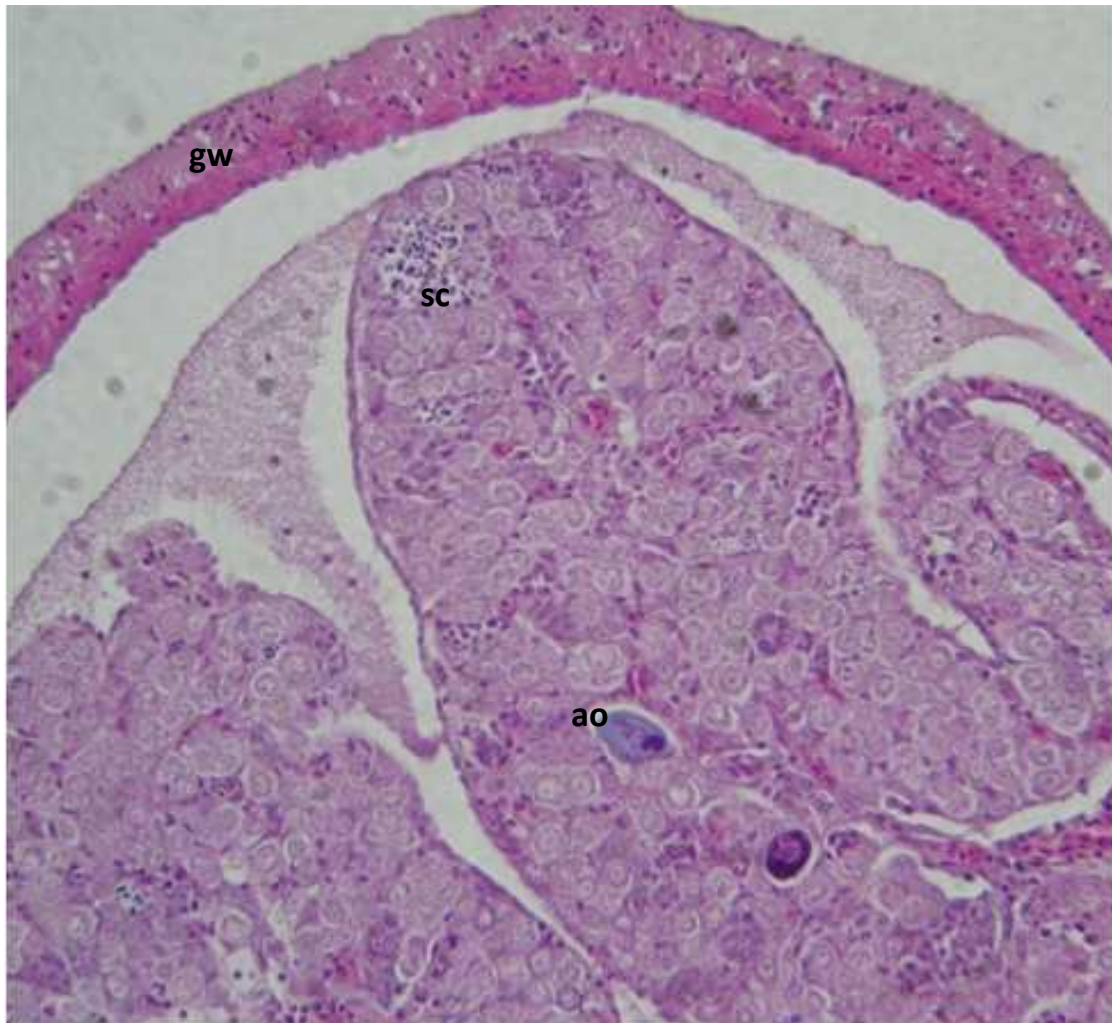


Figure 15: Picture of an immature male *L. harak* (186 mm FL, 3 years old) with atretic oocytes and developing sperm crypts. ao = atretic oocyte, gw = gonad wall, sc = sperm crypt.

DISCUSSION

Evidence from this study indicates that *L. harak* is a protogynous hermaphrodite, which is consistent with conclusions from Ebisawa (2006) and with other species in the family Lethrinidae. Ten transitional individuals were identified, all of which were larger than L_{50} and four were within the size range at which 100% of the female population was mature. Other features evident in *L. harak* which suggested protogyny were the presence of peripheral dorsal sperm sinuses and a remnant ovarian lumen in mature male testes. In addition, sex-specific length and age frequency distributions follow the pattern typical of protogynous hermaphrodites in which males dominate the higher length and age classes and females dominate the lower. Sadovy de Mitcheson & Liu (2008) noted that many lethrinid species show size-specific sex ratios indicative of

monandric protogynes but primary males may exist having male testes with the structure of an ovarian lumen. Evidence of this for *L. harak* exists in that five male individuals were identified below the size of 50% female maturation. All five had the structure of an ovarian lumen, three of these contained mature spermatozoa, and another contained sperm crypts in the presence of atretic oocytes. This merits a more detailed investigation of the early gonadal development of the species.

The data set adequately supports conclusions concerning sexual ontogeny, female maturation, and sex reversal. However, the sample size of larger, older fish was inadequate for exploring questions regarding seasonality in reproduction and comparisons of size/age at sex reversal. The vast majority of *L. harak* specimens collected were immature. Obtaining an adequate number of large individuals in monthly samples proved to be difficult as populations outside of marine preserves were comprised of smaller, younger, and less abundant individuals (see Chapter 2). Permits to collect specimens in marine preserves were not issued until the last months of the study period. The greater abundance of mature female *L. harak* in Guam's marine preserves is reflected in the monthly sample sizes for the GSI plot, in which April and May had the largest sample sizes. The study would have benefited if marine preserve collection permits had been obtained earlier and sampling from these sites was spaced out over the duration of the study period to ensure an adequate monthly sample of functionally mature females.

There was little discrepancy in size and age at female maturation between protected and unprotected *L. harak* populations. However, there was a distinct difference in the age at sex reversal between protection statuses which was driven by a higher abundance of older females within protected areas. Fishing non-randomly selects for larger and, indirectly, older individuals in a population and therefore truncates size and age structures. This affects a population's reproductive potential and if fishing persists at unsustainable levels, the stock will collapse. Reef fish populations have been shown to have a decreased size at maturity in response to heavy fishing pressure (McGovern et al. 1998) and accelerated sex reversal has been demonstrated by manipulating social structures (Ross 1981; Shapiro 1981; Nemtzov 1985; Liu & Sadovy 2004). Such changes in population processes can act as compensatory mechanisms when faced with a declining reproductive potential (Huntsman & Schaff 1994). Despite vast differences in sex ratios between protected and unprotected sites for ages 9 through 11 in the present study, these differences were not significant because of low sample sizes of old individuals. With increased sampling, it is likely that these differences would be significant, indicating that females from protected sites are delaying sex reversal until a later age because of a greater abundance and longevity of males within the protected population.

The small proportion of resting, inactive *L. harak* females in this study (~15% of mature females) coupled with their discrete seasonal occurrence creates confusion regarding the reproductive seasonality for the species on Guam. Anecdotal reports from Palau

insist that individuals are seen forming spawning aggregations in lagoons throughout the year (Johannes 1981). Other studies have produced equally unclear GSI plots as the present study. From Fiji, Lassi (2003) suggests that *L. harak* spawns year-round with the strongest GSI peaks occurring from August to October. In Kenya, the spawning season has been reported to be from October to February (Kulmiye et al. 2002). In the Ryukyu Islands of Japan, it has been reported to spawn from April to July (Ebisawa 2006). It seems most likely that *L. harak* spawns throughout the year across its distribution with varying seasonal intensity depending on location. Data suggests that smaller *Lethrinus* species, similar to *L. harak*, have longer spawning seasons whereas larger species tend to have shorter seasons (Loubens 1980; Brown et al. 1994; Sadovy 1996). Furthermore, the difference in mean GSI values among ovarian stages in *L. harak* is not very pronounced and active individuals are encountered throughout the year, so it would require a very large monthly sample size of active females to get a clear definitive picture of spawning seasonality.

Like many marine teleosts, *L. harak* exhibits a strong pattern of increased female reproductive potential with size and age. Reproductive capacity increases throughout an individual's lifetime as less energy is used for growth and more is allocated to reproduction (Roff 1984). Therefore, the importance of larger, older individuals in a population is obvious. It is clear from previous chapters that Guam's marine preserve network is effectively building up older age classes for both males and females which, in turn, has a positive effect on the total reproductive potential. For protogynous populations in which reproductive capacity is heavily influenced by social structure, marine protected areas provide a unique form of protection from fishing pressure. Wherever fishing occurs, large females, which are disproportionately important to reproductive processes, become rare and the mean size, age, and proportion of males is generally reduced, leading to sperm limitation (Smith 1982).

Currently, the only management regulation protecting *L. harak* on Guam is the network of marine preserves. This and previous chapters have highlighted the efficacy of the preserves in increasing the reproductive biomass and creating larger and older population structures within protected sites. The vast differences emphasize that protected sites on Guam possibly contribute disproportionately to the total reproductive capacity of the island-wide *L. harak* population. The reproductive and demographic data collected in this study would lend itself conveniently to assess the reproductive contribution of Guam's marine preserves.

CONCLUSIONS AND RECOMMENDATIONS

- For *L. harak* we recommend DAWR introduce a minimum size limit of 21 cm or 8.5 in (snout to fork length) which is equal to the length at 50% maturity (L_{50}) for all female fish combined (20.8 cm). Introducing a size limit for this species would,

in the long-term, guarantee the sustainability of the fishery without having to establish additional Marine Preserves (Sladek Nowlis 2000)

- This size limit concurs with the 23 cm recommended size limit by Taylor and McIlwain (2010) which, if enforced, would lead to a significant increase in spawning biomass for populations outside Guam's Marine Preserves. Furthermore, an introduction of a size limit would have very little impact on overall yield available for exploitation by local fishermen.
- A minimum size limit based on the L_{50} but several cm less than the size at sex change (24 cm) would have the added benefit of ensuring enough of the female population transition to males. One of the biggest challenges with managing protogynous (female to male sex change) populations is the significant reduction in the number of males through size selective fishing mortality.
- Because the largest and oldest *L. harak* females make a disproportionate contribution to spawning output we recommend introducing a maximum size limit, similar to the approach taken in Hawaii for nearshore species including surgeonfish, parrotfish and jacks. This limit would be 31 cm, or the size at which the relationship between ovary weight and fork length becomes curvilinear (see Fig. 9 in Taylor and McIlwain 2010).
- Minimum size limits, however, would offer little or no protection to small, immature *L. harak* unless unselective fishing methods, such as small-mesh monofilament nets, are severely regulated or totally banned.
- As there were few or no peaks in the GSI over time, more research is being conducted to determine the exact time of month/day these species spawn.

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