Final Report: Essential Fish Habitat and Life History Assessment of Commercially Important Herbivores in Pohnpei, Micronesia, to Enhance MPA Effectiveness

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Summary of Progress

A total of 516 and 423 *N. lituratus* were sampled to examine spatial habitat use and characterize the reproductive biology and age and growth, respectively, for this species. Similarly, 22 *N. unicornis* were taken from the field and 302 from markets to estimate these same parameters. Based on findings, both *N. lituratus* and *N. unicornis* are gonochostic species that spawn monthly throughout the year. For *N. lituratus*, a peak in reproductive activity was indicated from January-July, suggesting that optimal times for temporal management to protect reproductive adults be instituted within this period. For *N. unicornis*, GSI values did not provide a clear indication of spawning seasonality. Nonetheless, female with hydrated oocytes were observed from May-November. Reproductive activities for either species did not appear to conform to a specific lunar period. For both species, there was a significant sex-specific difference in age, with females older than males. Male *N. lituratus* matured at 1 yr old (range=1-13 years), while mature females appeared at Age 2 (range=1-13 years). For *N. unicornis*, females ranged from 1-16 years, while males were 1-14 years old, with no sex-specific difference in age. Mature *N. unicornis* appeared in samples at Age 1 for both sexes. For *N. unicornis*, samples appeared in markets well below their reported maximum lengths. For the acoustic tagging component, some vriability was noted in movement patterns (as receiver detections) that sugest differences in their vulnerable to fishing. Three of the 13 *N. unicornis* were recaptured by the fishery, while none of the 10 *N. lituratus* were taken. Patterns of movement showed most *N. lituratus* with reliable detection patters to reside primarily within the MPA boundaries where they were first samples, whereas *N. unicornis* were shown to move up to at least 4 km outside the MPA, with evidence of fisheries mortality. Based on these combined assessments, both species could benefit from management improvements. For both species, temporal fishing restrictions within the spawning season are recommended, with a recommended period from June-August for both species. Such a ban would fall outside the current grouper sales ban period. In addition, *N. unicornis* could benefit from a combination of a size limit for the fishery and an expanded marine protected area.

I. Acoustic and conventional tag and recapture

Conventional

Naso lituratus

A total of 516 *Naso lituratus* were captured and tagged between 2/25/10 and 3/21/10 including 10 acoustically tagged fish at or near the Kehpara Marine Sanctuary (***Figure 1***). Tagged *N. lituratus* ranged in size from 105-255 mm FL ($Range_F$=113-231 mm FL, mean±SD=173.1±24.6 mm FL; $Range_M$=105-255 mm FL, mean±SD=184.6±20.8 mm FL). Sex-specific sizes for taged fish were significantly different (t-test, $p<<0.00$). Of the 516 fish, 85 individuals were recaptured by the fishery (16.5%). These included 51 (of 315 females=16%) and 34 (of 199 males=17%). For recaptured *N. lituratus*, the time at liberty (as days) ranged from 2 to 261 d (mean±SD=71±81 d). No acoustically tagged *N. lituratus* were recovered by the fishery.

Naso unicornis

For *Naso unicornis*, a total of 22 individuls were captured and tagged, including 13 acoustically tagged fish (Table 1). Sizes ranged from 172-362 mm FL and averaged 273.5±58.2 mm FL. Of the 22 fish, only 3 were males. Similar to *N. lituratus*, data that could be used to determine residency and movement at and away from the capture and release site was limited. Nonetheless, 3 of 11 acoustically tagged *N. unicornis* were recaptured (=27%), with a total distance of movement for all fish of 8 km. None of the *N. unicornis* tagged only with conventional tags were re-taken during the study. Time at liberty for *N. unicornis* was 52±25 d. For *N. unicornis*, recapture information was insufficient to determine catchment area owing to the small sample size. Currently, maximum distance estimates are around 7-8 km from the point of tagging, with most fish remaining in close proximity to tag locations.

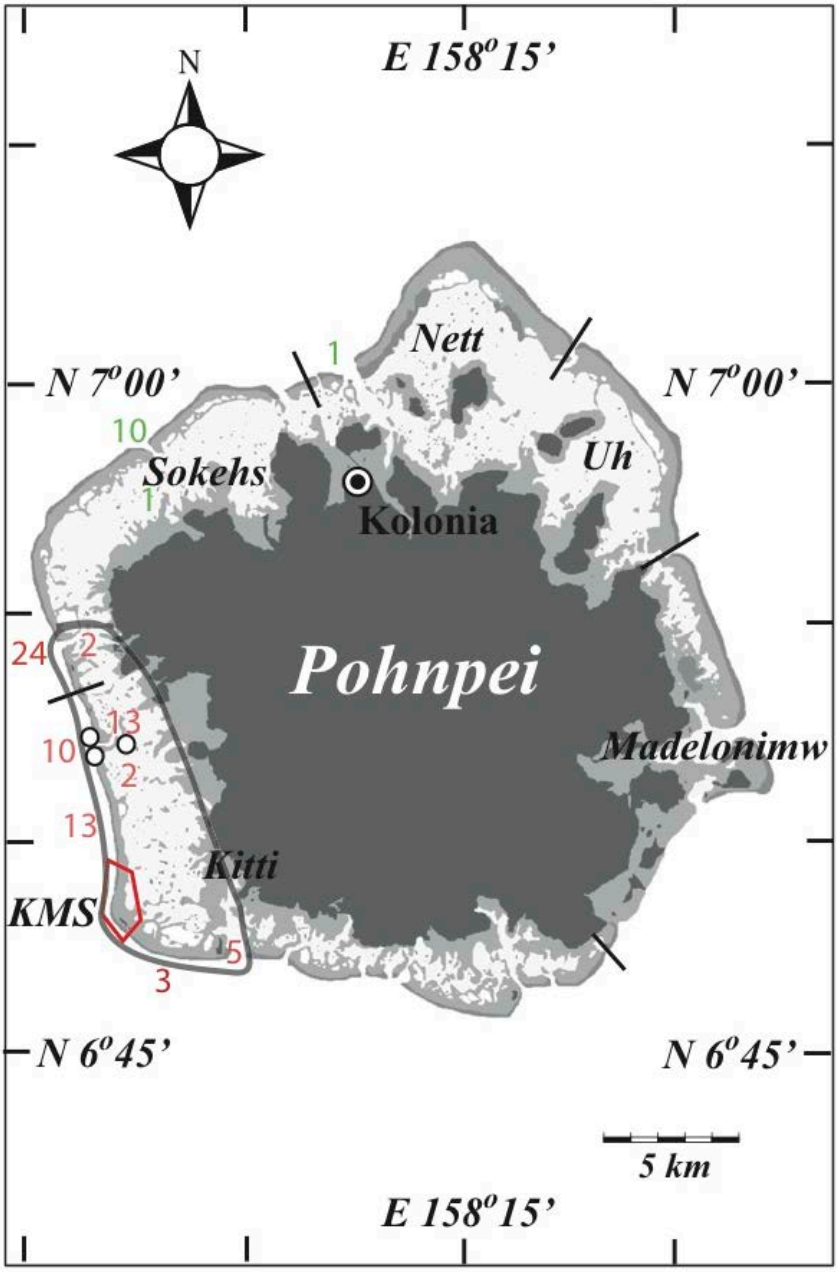

Figure 1*: Map of Pohnpei showing the capture location (KMS-Kehpara Marine Sanctuary, red polygon) and number of recaptures by location (red). Numbers shown in green are reported capture locations that are questionable, based on expected travel distances for Naso lituratus (also see Acoustic section below). The area circumscribed is the likely catchment area for tagged individuals. Approximate recapture locations of N. unicornis are represented by open circles.*

Table 1: Tagging summary of *Naso lituratus* (NL) and *Naso unicornis* (NU). BW=body weight; FL=fork length and SL=standard length; ID codes conform to acoustic tag IDs.

Date	Recapture date	Species	Sex	ID Code	Floy	BW (g)	FL (mm)	SL (mm)
3/2/10		NL	F	65051	0326	184.0	198	170
3/2/10		NL	F	65052	0327	180.0	202	172
3/2/10		NL	F	65053	0328	143.0	184	157
3/2/10		NL	F	65054	0329	216.0	206	175
3/2/10		NL	F	65055	0330	145.0	180	151
3/2/10		NL	M	65056	0331	177.0	200	170
3/2/10		NL	M	65057	0332	134.0	185	157
3/3/10		NL	M	65059	0382	nd	197	167
3/3/10		NL	M	65060	0385	202.0	207	176
3/3/10		NL	M	65061	0372	178.0	196	165
3/4/10	3/22/10	NU	F	65040	0386	441.0	285	235
3/4/10	5/12/10	NU	F	65041	0412	850.0	351	291
3/4/10		NU	F	65042	0414	1048.0	362	302
3/26/10		NU	F	65044	0576	nd	266	211
3/4/10		NU	F	65045	0413	409.0	280	232
3/26/10		NU	M	65046	0526	nd	322	265
3/26/10		NU	M	65047	0527	nd	295	243
3/4/10	3/17/10	NU	F	65048	0415	640.0	320	267
3/26/10		NU	F	65048	0529	nd	255	210
3/27/10		NU	F	65049	0533	nd	247	205
3/27/10		NU	F	65050	0532	nd	285	237
3/27/10		NU	F	64946	0530	nd	292	260
3/27/10		NU	F	64913	0531	nd	355	211

Recaptured locations for all recaptured fish are shown in ***Figure 1.*** For *N. unicornis* all recaptured fish were taken near Peleng Pass, either inside or outside the reef. For *N. lituratus*, all reliable information showed the fish to move and reside between Nahlap Channel and Dawak Channel, or *ca.* 19 km of linear distance along the reef. Recaptured occurred both inside and outside the reef. and suggest a relatively small catchment area of *ca.* 80 km^2 for *N. lituratus*. These findings confirm that the use of small-scale MPAs may be insufficient for the conservation and management of adult *Naso lituratus*.

Acoustic

For the acoustic tagging portion of the project 38 receivers were deployed within or adjacent to the MPA, including 10 in the back reef, 10 on the reef flat, 6 in three adjacent channels (one to the south and two to the north of the MPA, with the remaining receivers

deployed along the outer reef over a 15 km distance. Some of the receivers were concentrated within the MPA to assess residency patterns for this and other species (***Figure 2***).

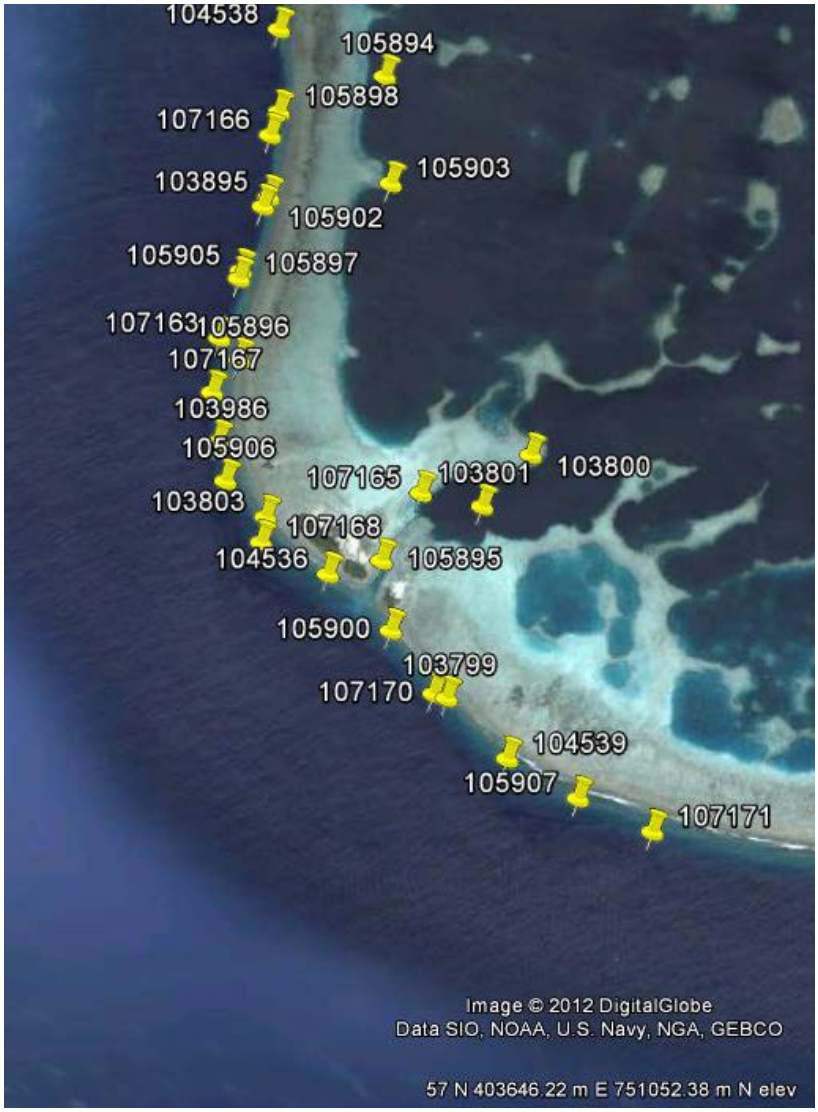

Figure 2*: Receiver placement along a portion of the outer reef in and adjacent to the Kehpara marine Sanctuary. The sanctuary boundaries are displayed in Figure 1.*

Naso lituratus

Inclusive within the 516 *N. lituratus* captured for conventionally tagging were 10 acoustically tagged fish, including 5 females and 5 males that ranged in size from 180 to

207 mm FL. Among those, several fish provided information useful to confirm spatial information provided from recaptures and to verify the effectiveness of the Kehpara Marine Sanctuary. Three individuals are highlighted herein, including two females (65051 and 65053) and one male (65061).

Female 65051.—Female 65051 (198 mm FL) was captured on 2 March and released along with the other tagged *N. lituratus* on 7 March within the MPA. This female was detected along approximately 3 linear km of the forereef, with 144 total detections made by 13 different receivers (18% of the total tag life). The last detection was for this fish was made on 40 April, for a total time at liberty of 41 days. The detection map (***Figure 3***) shows the percent detections for each of the receivers and shows that this female remained primary inside the MPA, with minimal detections outside. This fish was last detected approximately 5 km north of the core of the MPA, however, it was not returned by the fishery, suggesting possible Floy tag loss.

Female 65053.—Female 65053 (184 mm TL) was tagged on 2 March and realeased on 7 March 2012 inside the Kehpara Marine Sanctuary. The female was detected 1637 times over a period of 91 days (37% of total tag life) and along less than 3 km of reef. No detections were made to the south of the MPA, and few detections were made north of the sanctuary (***Figure 4***).

Male 65061.—This male (178 mm FL) resided exclusively inside the MPA (Figure 4, based on tag detections. In total, this male was detected 1815 times over a period of 7 months (194 days, or 79% of the tag life) (***Figure 5***).

Although all three individual shown above appeared to reside in the MPA, none was detected throughout the taglife of the transmitter, possibly suggesting that these fish were captured inside the MPA and went unreported by the fishery, or these individuals suffered natural mortality.

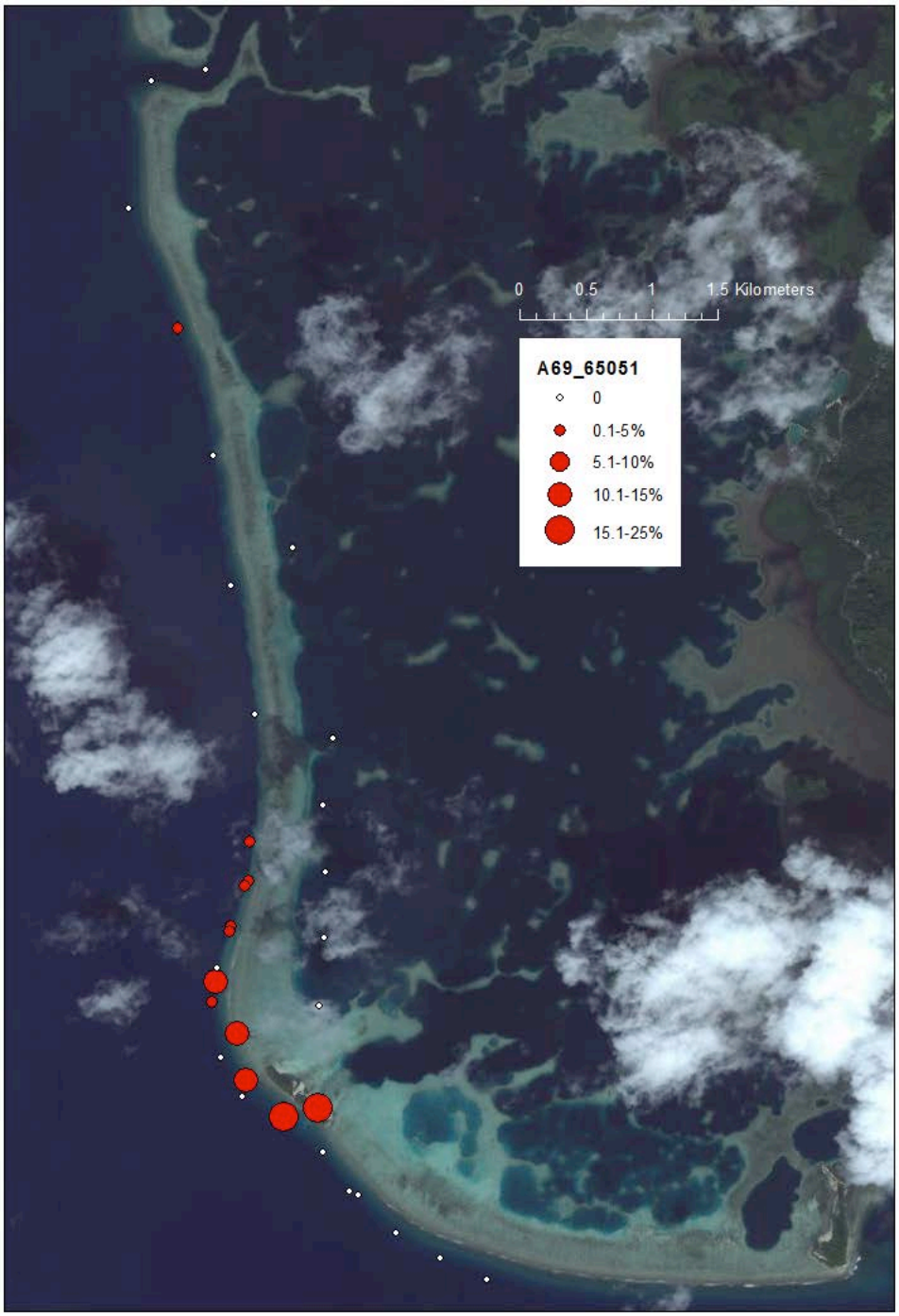

***Figure 3**: Percentage of detections by receiver location for N. lituratus female 65051. As shown by the fugure, this female spent the majority of time inside the MPA. No movement was detected to the south of the MPA or inside the lagoon.*

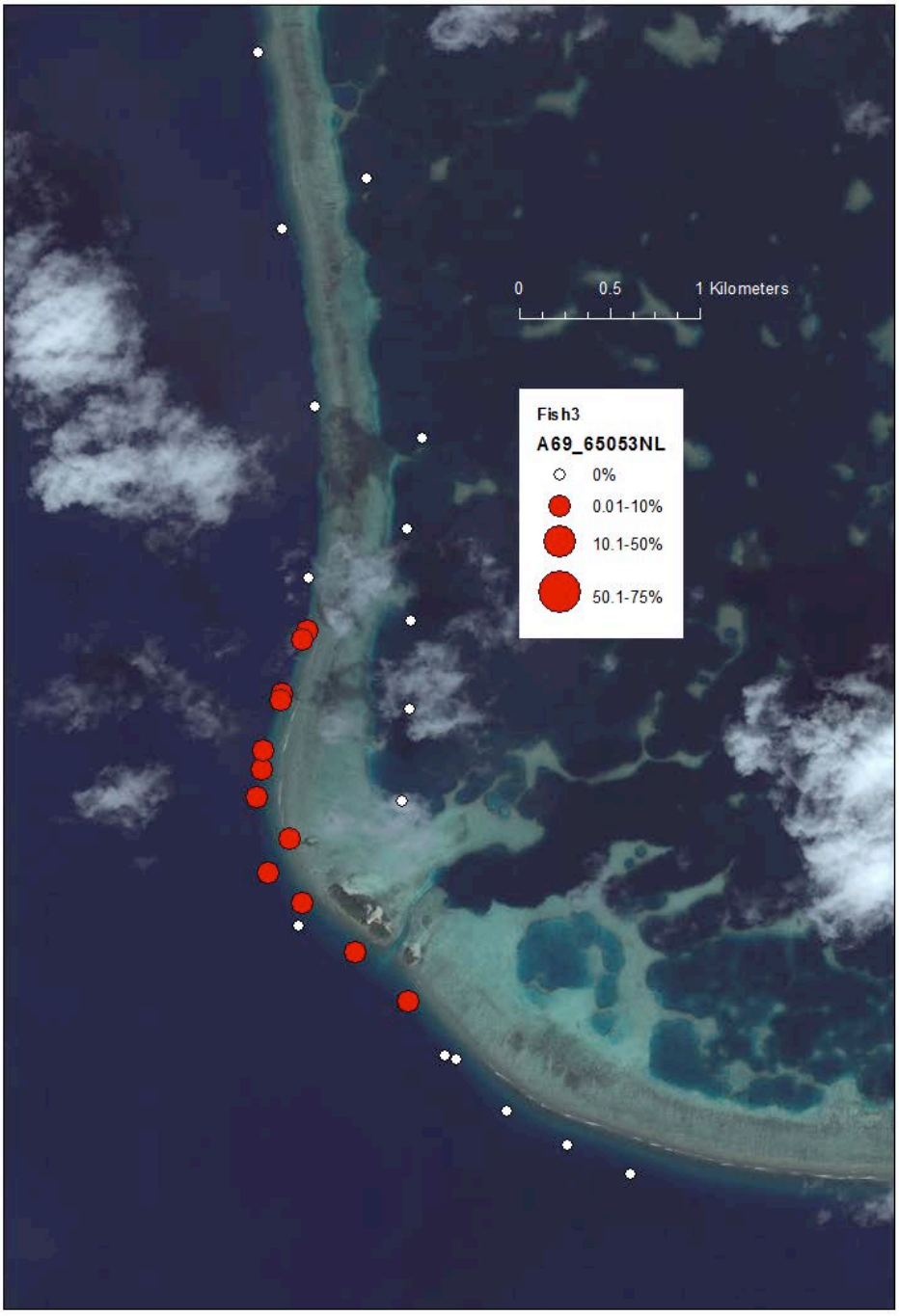

***Figure 4**: Detection patteron for Female 65053 showing residency primarily within the Kehpara Marine Sanctuary.*

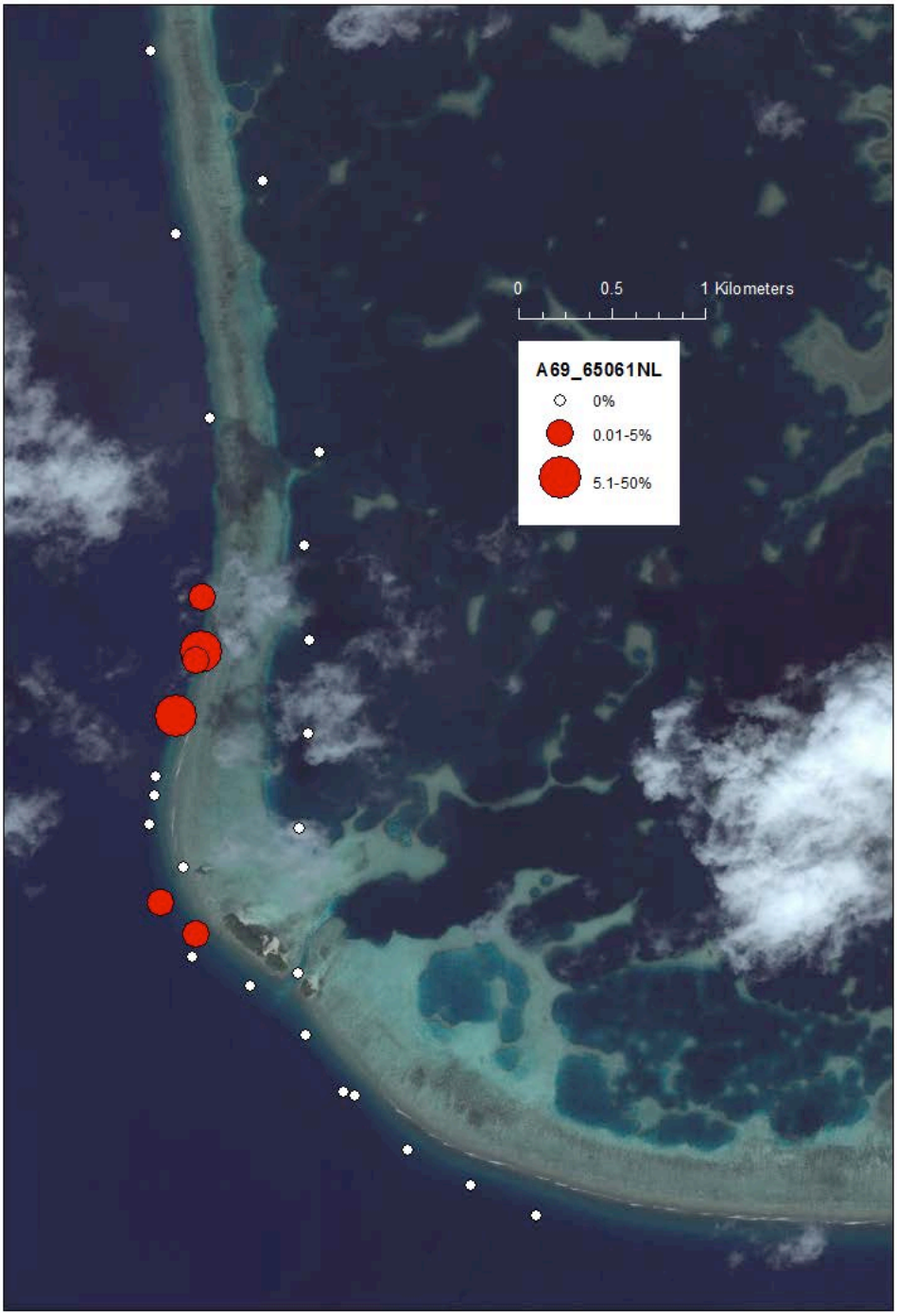

Figure 5*: Detection pattern of Female 65061, which was detected exclusively inside the MPA.*

Naso unicornis

A total of 13 *N. unicornis* were tagged between 4 and 27 March 2010, including 11 females (247-362 mm FL) and 2 males (295 and 322 mm FL). Of these, 3 individuals were receptured by the fishery between 13 and 69 days, suggesting a high fisheries mortality for the species. Three individuals (two females, one male) were chosen from the data to demonstrate detection patterns for the species.

Female 65040.—This female (285 mm FL) was tagged on 2 March and released on 5 March in the KMS. Following its release, it was detected 563 times over a 20 d period until its capture by the fishery approximately 5 km north of the MPA (***Figure 6***). The fish was detected greater than 50% of its time outside the MPA and showed a long distance migration a few days after release, residing or moving within or around the outer reef 3-5 km north of the MPA where it became a target of the fishery.

Female 65041.—Female 65041 (351 mm FL) was tagged on 2 March and released on 5 March inside the KMS. During its 12 days at liberty, it was detected by 15 receivers, with a total of 1316 detections. Although this individual was detected primarily within the MPA, it moved as far as 4 km N of the MPA and was last detected 1200 m north of the cental MPA receiver (***Figure 7***). The fish was reportedly captured inside the lagoon just south of Peleng Channel, approximately 6 km to the notrth of the MPA. It was not detected by any inner reef receiver.

Male 65047.—This male (295 mm FL) was detected over 62 d by 16 receivers a total of 3753 times (***Figure 8***). Its total linear distance of movement along the outer reef was approximately 5 km. Maximum distance of movement from the approximate release point was 4 km north of the MPA where it was last detected. The fish spent the majority of its time during the detection period inside the MPA, but appeared to be taken by the fishery or have suffered from natural mortality away from the array. The fish was not reportedly captured by the fishery, so there is no information on the fate of this animal.

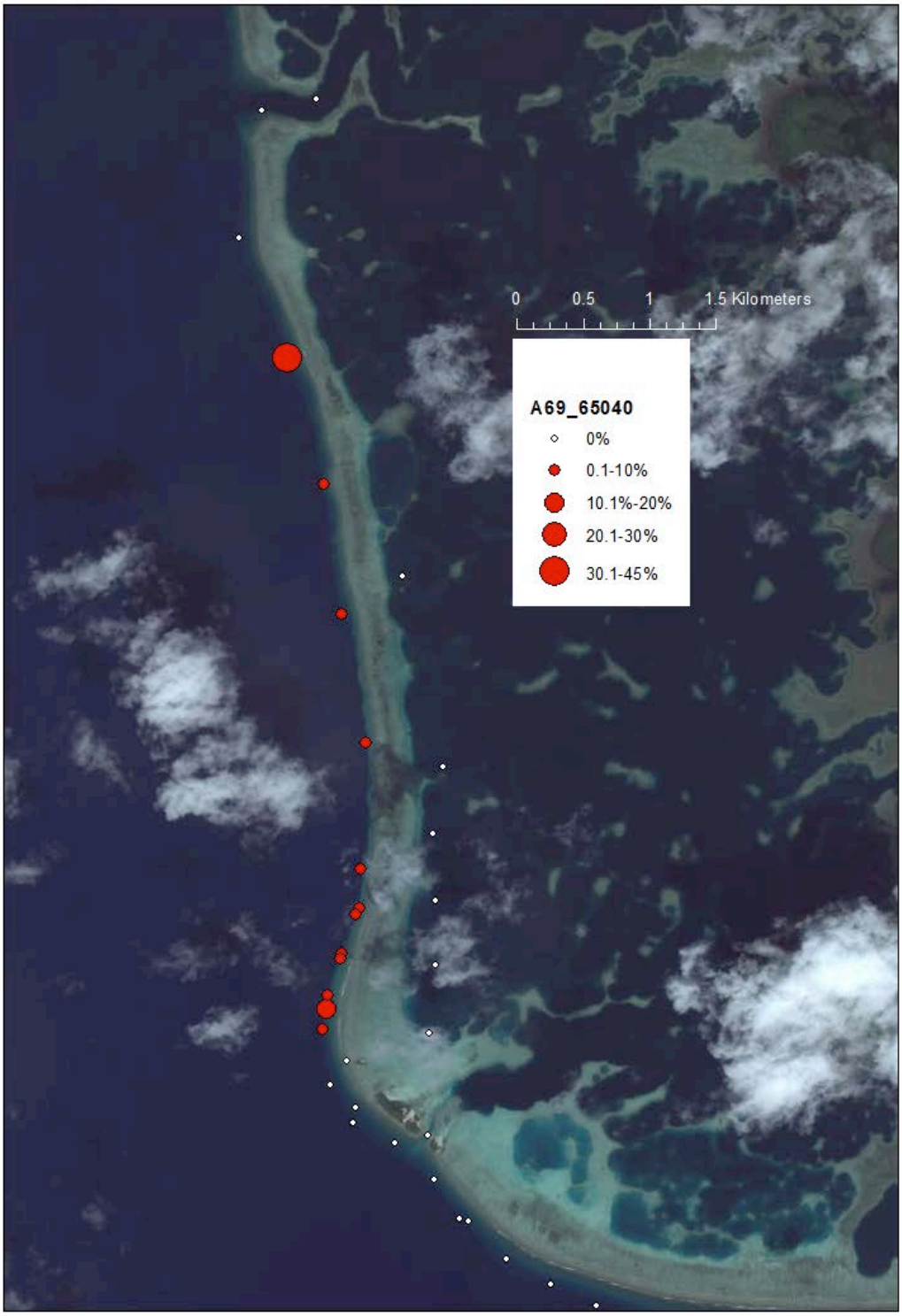

***Figure 6**: Detection pattern of Female 65040 showing movement to the north of the MPA where it was recaptured by the fishery approximately 20 d later.*

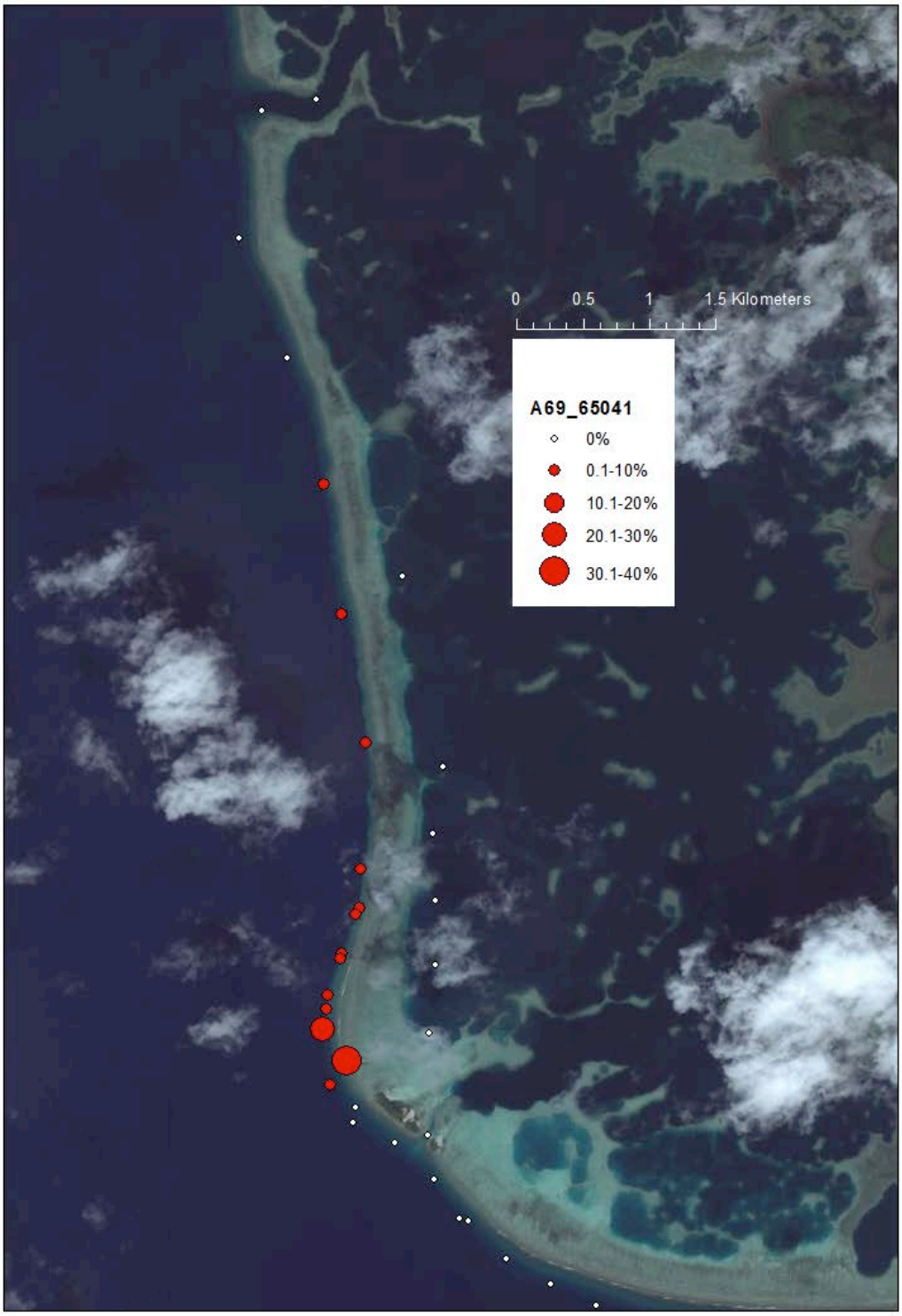

***Figure 7**: Detection pattern of 65041 showing primary residency during the detection period inside the MPA. The fish was captured by the fishery shorty after release.*

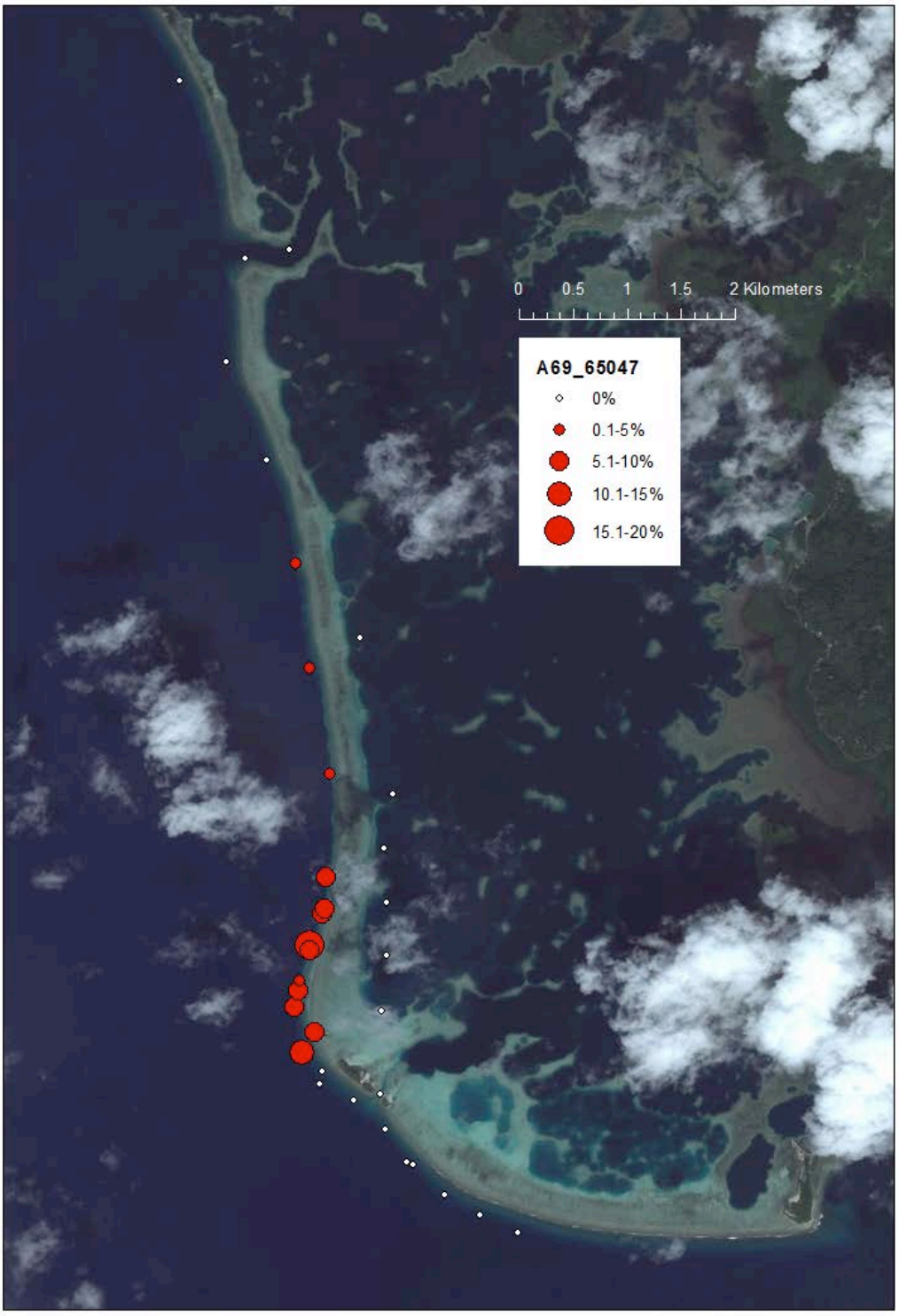

***Figure 8**: Detection pattern of Male 65047. The fish remained inside the MPA for most of its time at liberty, but disappeared from the array 4 km to the north 62 d after release.*

II. Age and growth

Naso lituratus

Of the 423 *N. lituratus* taken from markets or collected from recaptured, 236 individuals (230 markets; 6 recaptures) were used to provide sex-specific information on age and growth that included 129 females and 107 males. *N. lituratus* ranged from 1-13 years, with females 4.1±2.7 years (mean±Standard deviation, hereafter) (***Figure 9***) and males 3.4±2.6 years (***Figure 10***). A significant difference was shown between sexes for age (t-test, p=0.02). Otolith weight was demonstrated to be a good predictor of age for the species (***Figure 11***). The age-length relationship for combined sexes is shown in ***Figure 12***.

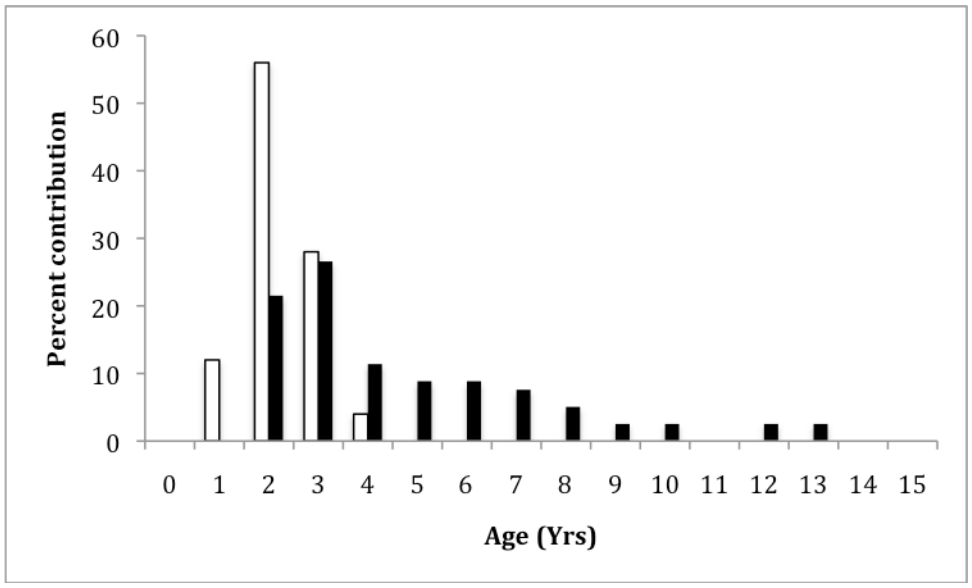

Figure 9*: Age distribution of female N. lituratus examined microscopically. White bars represent immature individuals, while mature fish are shown in black.*

***Figure 10**: Age distribution of male N. lituratus examined microscopically.White bars represent immature individuals, while mature fish are shown in black. Some 'immature' fish are shown in the largest age classes, reflecting the difficulty in differentiating immature from mature, resting males.*

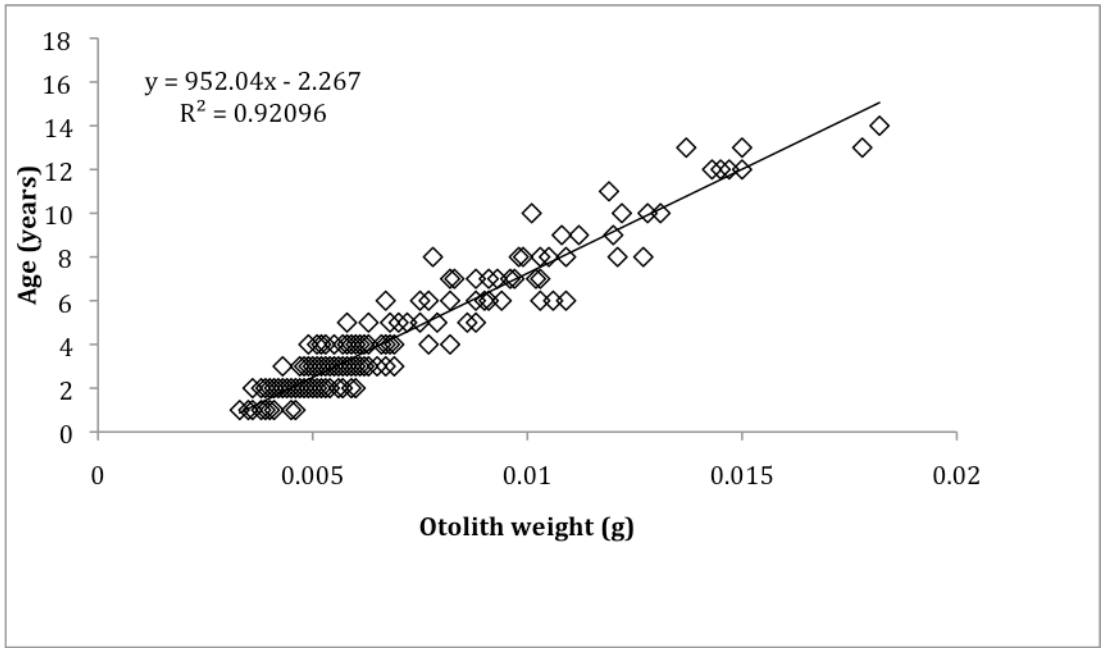

Figure 11: Otolith weight-age relationship for N. lituratus.

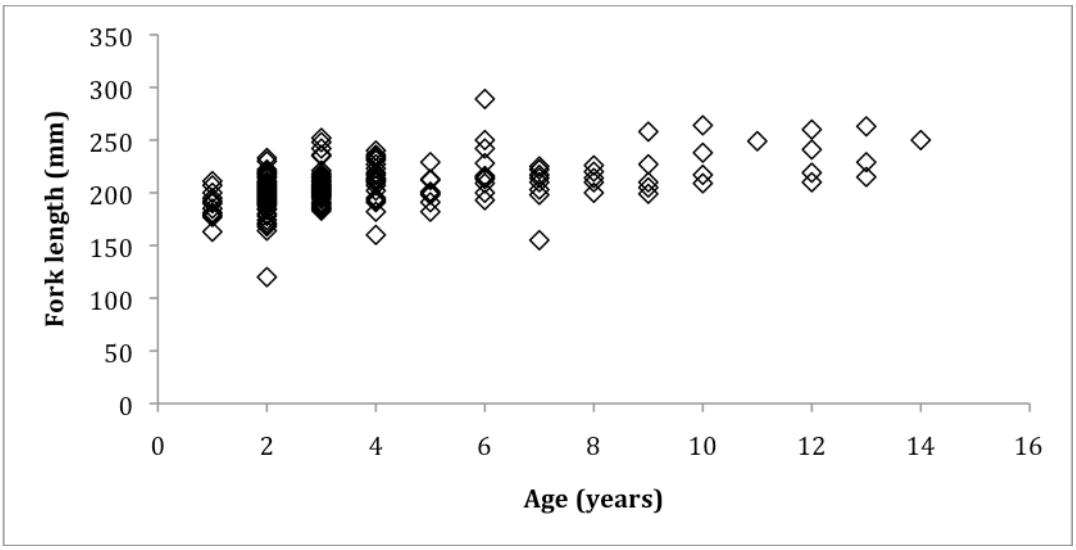

***Figure 12**: Age-length relationship for N. lituratus.*

Naso unicornis

A total of 232 *N. unicornis* were sampled for age and growth from markets. Of those, 216 fish (138 female; 78 male) were examined for age. Females ranged from 1-16 years old (***Figure 13***), while males were 1 to 14 years in age (***Figure 14***). Males age averaged 4.0±2.3 (SD) years, while females averaged 3.5±2.2 years. There was no significant difference shown between sexes for age (t-test, p=0.09). As with *N. lituratus*, *N. unicornis* otolith weight was a good indicator of age (***Figure 15***). The age-length relationship for the species in shown in ***Figure 16***. For males, the youngest fish (Age 1) were 222 and 228 mm FL and were both spent males.

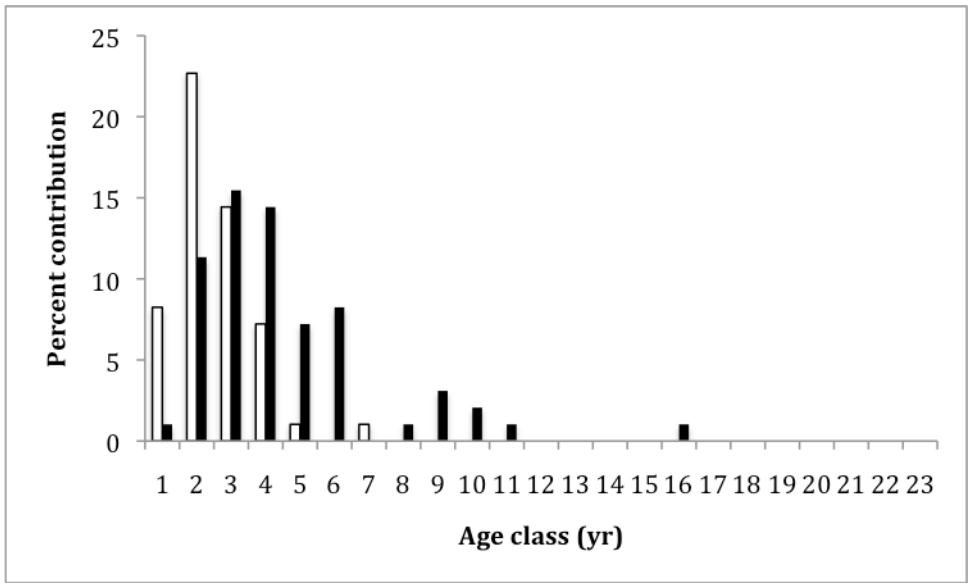

***Figure 13**: Age distribution of female N. unicornis sampled from Pohnpei January 2010-January 2011. Mature individuals dominated upper size classes and were represented in the 1-yr age category.*

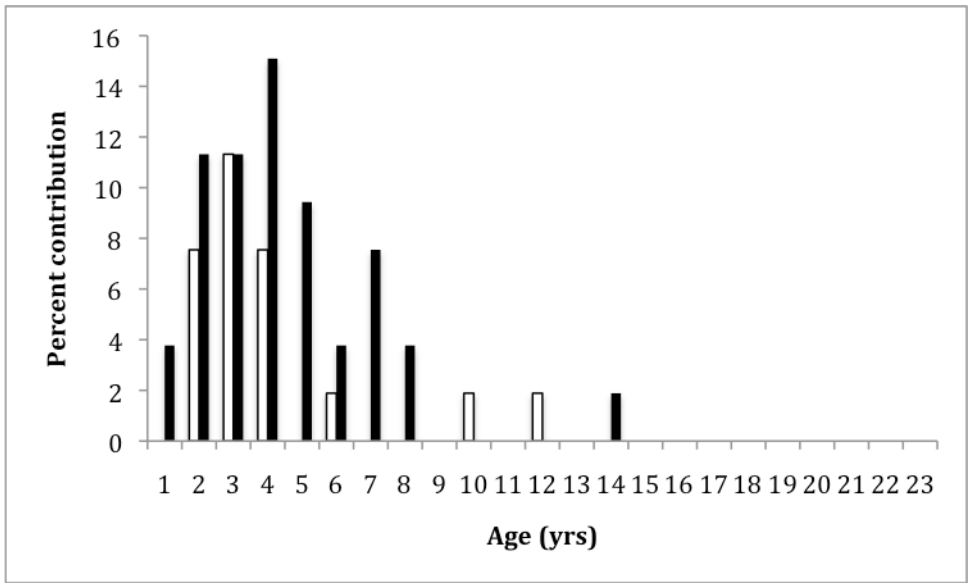

***Figure 14**: Age distribution of male N. unicornis sampled from Pohnpei January 2010-January 2011. Mature individuals dominated upper size classes and were represented in the 1-yr age category.*

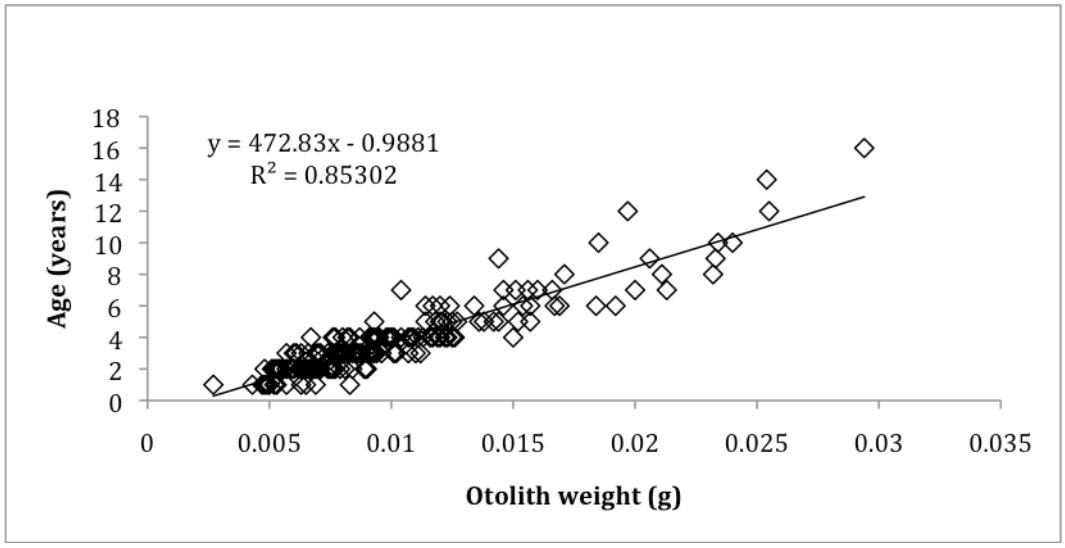

***Figure 15**: Otolith weight-age relationship for N. unicornis.*

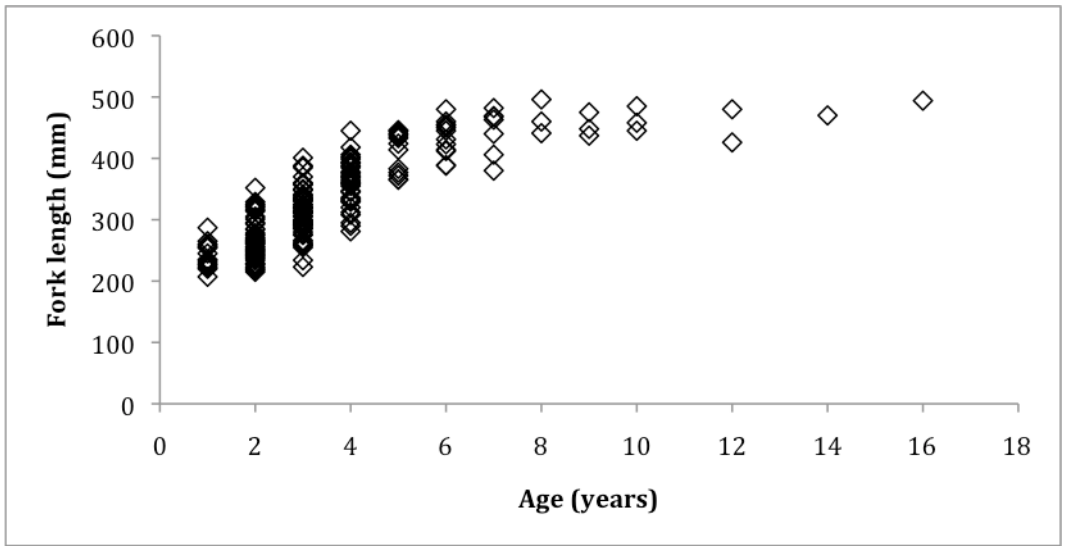

***Figure 16**: Age-length relationship for N. unicornis.*

III. Reproductive Seasonality and Sexual Pattern

N. lituratus

To examine reproductive life history for *N. lituratus* 314 samples were analyzed microscopically, using criteria from Appendix A . Among the *N. lituratus* 199 females and 115 males examined, no juveniles were found, suggesting that sexual development and maturity occur within the first year. Sizes of developing (F2D), mature, resting (F2R) and mature (F3, mature, active; F4, ripe; F5, spent) females are shown in ***Figure 17*** and indicate a wide overlap among fish in various development stages. ***Figure 18*** shows the size distribution of imature and mature male *N. lituratus*. ***Figure 19*** shows the monthly distribution of female (***top***) and males (***bottom***) *N. lituratus* gonad development stages, along with sample size for each stage. Based on the seasonal distribution of female development stages and the gonadosomatic index, *N. lituratus* spawns monthly througout the year, with a possible peak from January through August.

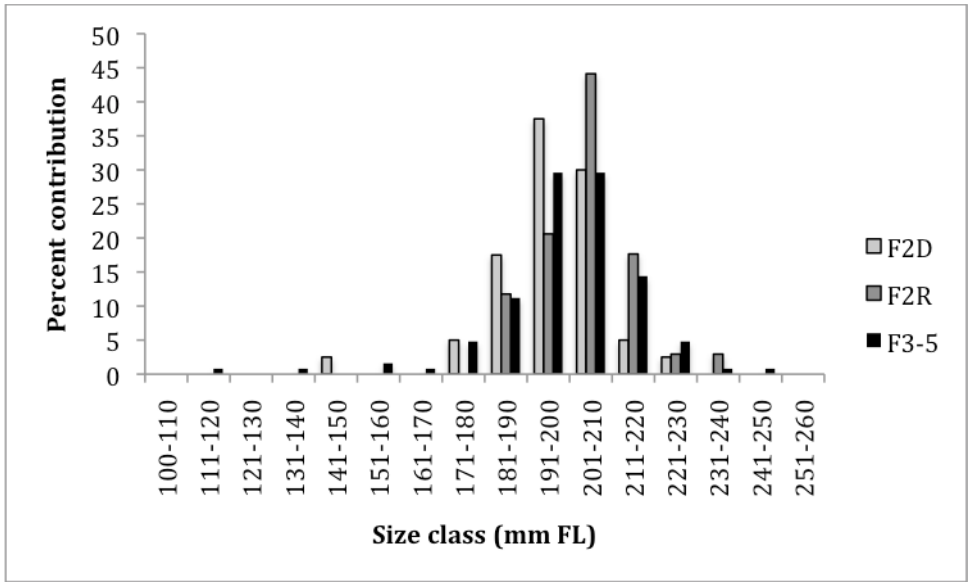

Figure 17*: Size frequency distribution of female N. lituratus examining microscopically No juveniles (F1) were observed. F2D=developing females; F2R=mature, resting females; F3-5 represents mature (F3), ripe (F4) and spent (F5) individuals.*

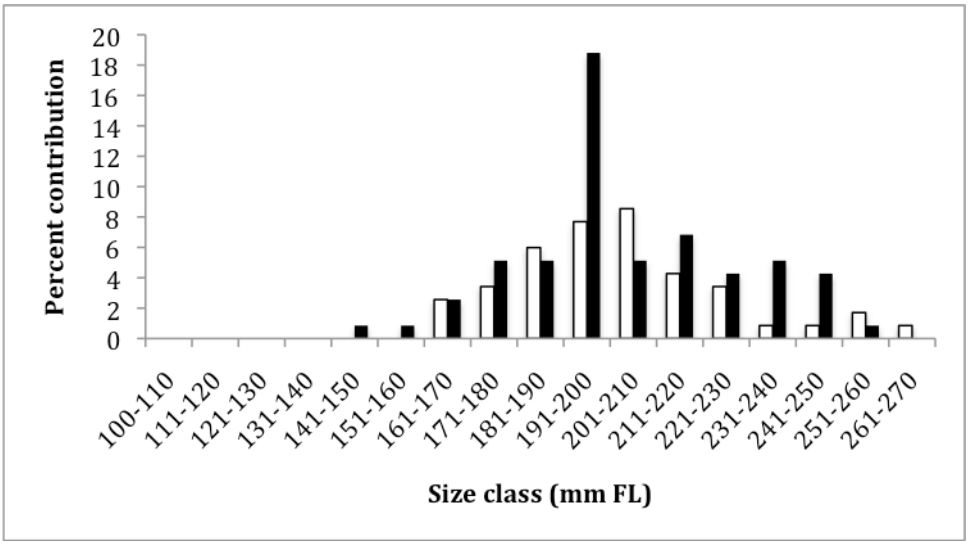

Figure 18*: Size frequency distribution of mature (black bars) and immature (white bars) male N. lituratus sampled in Pohnpei January 2010-January 2011.*

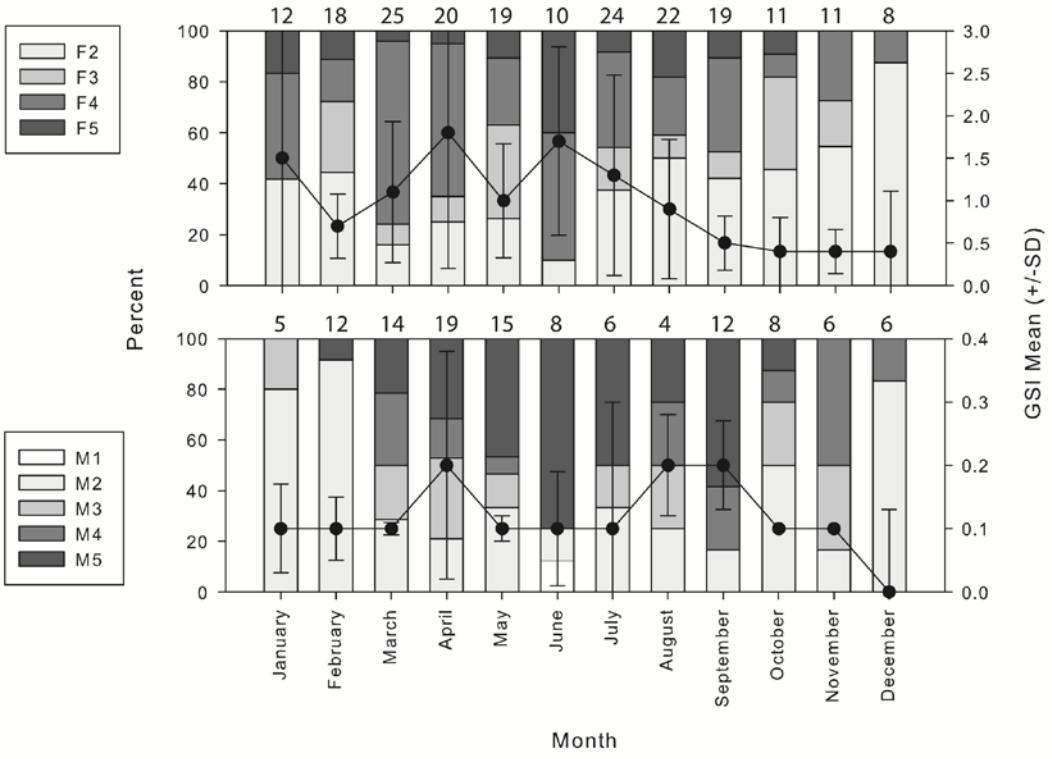

Figure 19*: Seasonal distribution of gonad development stages for N. lituratus. Monthly sample sizes are given above each bar. The mean (±SD) gonadosomatic index (GSI) is represented by the solid line. Female stages are as above. M1=immature or early stage mature, resting male; M2=developing male; M3=mature male; M4=ripe male; M5=spent male.*

N unicornis

A total of 302 individuals that included 123 males and 179 females were selected for microscopic gonad analysis. ***Figure 20*** shows the size distribution of females taken during sampling and shows mature (F3-F5) and mature, resting females (F2R) largely within the upper size classes. Females ranged from 203 to 494 mm FL (mean=330±70.6 mm FL), with the smallest mature female 249 mm FL (Age 2). The youngest mature female (Age 1) was 269 mm FL. Some immature females in the larger size categories suggests delayed maturation in some individuals or, alternatively, atresia of tissues typically used for assessing maturity (i.e. thickened gonad wall, muscle bundles, residual late stage oocytes or post-ovulatory follicles. The size distribution of males by gonad development stage (as mature versus immature fish) are shown in ***Figure 21***. In contrast to females, mature and immature male *N. unicornis* overlapped widely. Only one individual was sampled in the largest size class. Male size ranged from 197-665 mm FL and and averaged 331.0±80.7 mm FL. There was no sex-specific size difference between market-sampled *N. unicornis* (t-test, p=0.8).

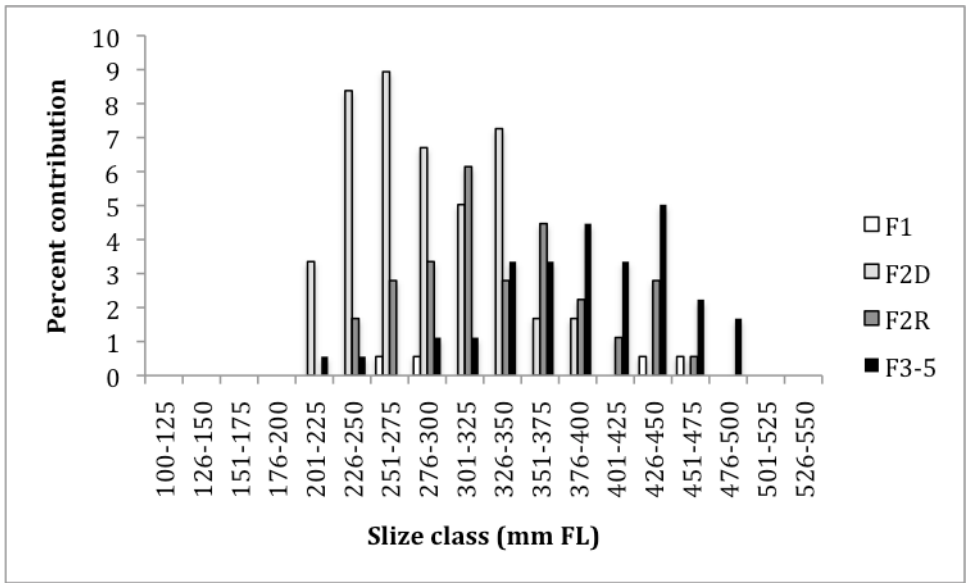

Figure 20*: Size frequency distribution of female N. unicornis sampled in Pohnpei January 2010-January 2011. Mature individuals (F3-5) dominate the upper size classes, with immature (F1) and developing females (F2D) primarily within lower size categories.*

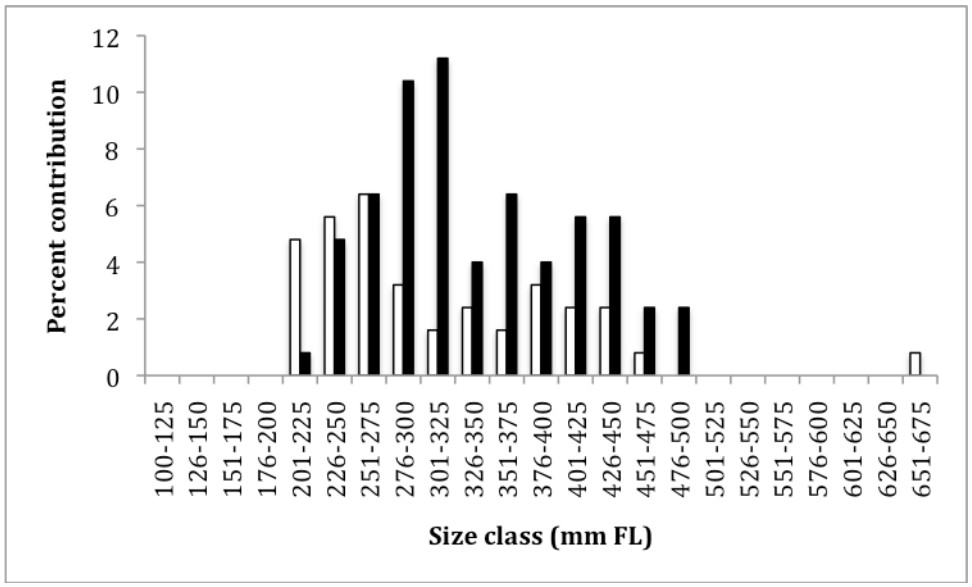

Figure 21*: Size frequency distribution of male N. unicornis sampled in Pohnpei January 2010-January 2011. Mature individuals (black bars) overlap widely with immature fish (white bars).*

Figure 22 shows the distribution of females (***top***) and males (***bottom***) by gonad development stage. Based on the presence and distribution of mature gonads, together with trends in the gonadosomatic index, *n. unicornis* appears to spwn monthly. There was no clear indication of a specific lunar cycle attached to spawning.

IV. Continuing efforts

We are currently developing the draft manuscript for age, growth and reproduction of these two species. The manuscript will be a comparative assessment of these two species in Pohnpei and Guam, based on this and previously collected data from Guam using Guam Department of Aquatic and Wildlife Resources (DAWR). A report on the findings will be provided to the Pohnpei State Department of Lands and Natural Resources in January 2013.

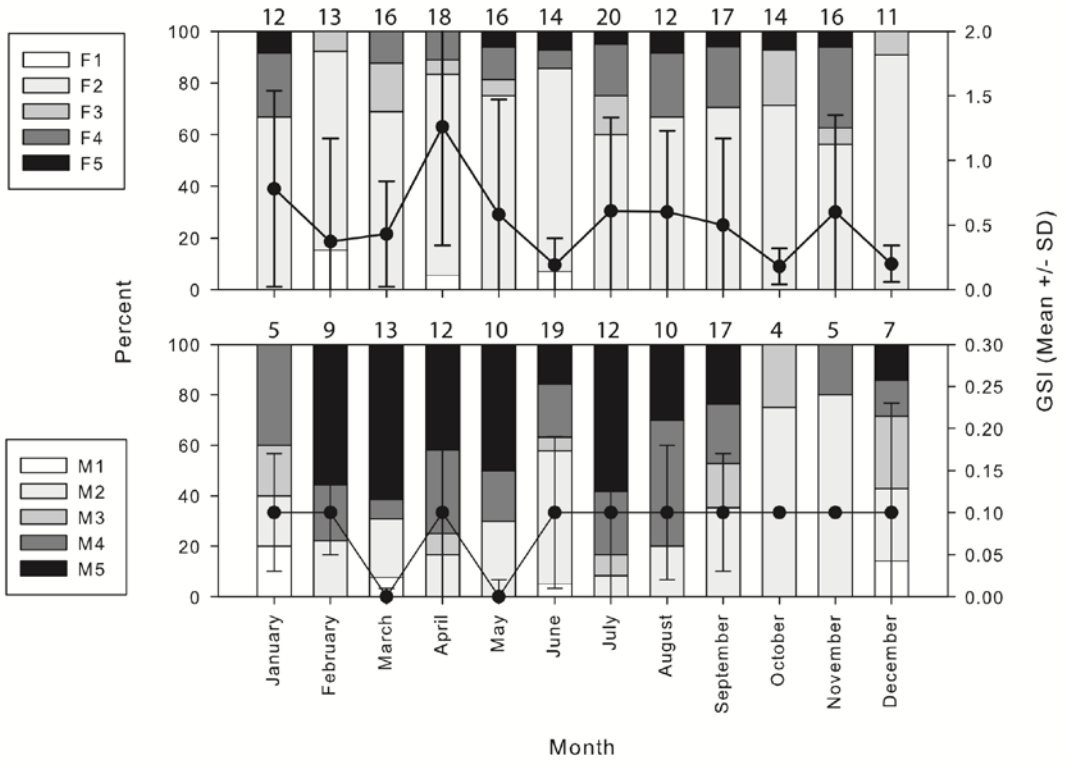

Figure 22*: Seasonal distribution of evelopmental stages of female (top) and male (bottom) N. unicornis sampled in Pohnpei January 2012-January 2011. Monthly sample size are shown above each bar. The mean (±SD) gonadosomatic index is shown by the solid line.*

Appendix A: Criteria used in the assessment of sexual development in orangespine unicornfish, *Naso lituratus*, and bluepsine unicornfish, *Nqso unicornis*.

Maturity stage	Macroscopic	Microscopic
Ovaries		
F1 (immature)	Small, strand-like tissue, compact, pink or cream; oocytes indiscernible; indistinguishable from M1 males	Gonad wall thin; tightly-packed previtellogenic Stage 1 (nucleolar) and 2 (perinucleolar) oocytes and gonia
F2 (developing; mature, resting)	Relatively small but rounded, greyish with thickened gonad wall; oocytes indiscernible and small (<0.4 mm); Indistinguishable from M2 prior to cortical avleolus stage when oocytes become discernible.	Stages 1 and 2 and cortical alveolus stage oocytes present. **Developing:** includes developing (maturing) and inactive females with actively advancing oocytes, but without signs of prior spawning. **Mature, resting:** individuals that are recovering from spawning with (1) distinct muscle bundles, (2) thick gonad wall, and/or (3) an absence of post-ovulatory follicles; lumen may be present and oocytes may be disorganized or largely atretic, including later stage oocytes.
F3 (mature, active)	Large and greyish with transparent gonad wall; large vitellogenic oocytes becoming clearly visible and tightly packed	Mainly vitellogenic (Stage 3) oocytes; gonad wall stretched and thin; Stage 1 and 2 oocytes relatively few; no large-scale atresia or post-ovulatory follicles
F4 (mature, ripe)	Ovary large, clear, hydrated oocytes visible through wall; typical of individuals just prior to spawning; egg release possible with application of light abdominal pressure	All stages of oocyte development, but dominated by Stage 4 (hydrated) and late Stage 3; yolk fusion and hydration are extensive
F5 (post-spawn)	Ovary flaccid with obvious capillaries; few oocytes visible	Post-ovulatory follicles numerous and large prominent muscle bundles scattered throughout the gonad; the gonad wall is thickened; few Stage 3 and/or 4 oocytes may be present and undergoing atresia

Maturity stage	Macroscopic	Microscopic
Testes		
M1 (immature/inactive)	Indistinguishable from F1 females (see the description of F1)	Gonad filled with varying amounts of stroma (muscle tissue), usually the dominant feature; 1^o and 2^o spermatocytes, and spermatids largely absent (< 10%); sperm sinus present or not; seminiferous tubules may appear as gonia-lined rings; some signs of prior spawning, such as contracted seminiferous tubules or thickened muscles bands among/between seminiferous tubules apparent; gonial lobules and brown bodies may be evident.
M2 (developing or resting, mature male)	Gonad greyish in appearance and flaccid; M2 individuals are indistinguishable from F2 until milt becomes evident in the sperm sinus	**Developing:** Gonad largely filled with stroma; 1^o and 2^o spermatocytes, and spermatids in minor proportions, usually < 25% of the volume; sperm sinus present or not; gonia-filled seminiferous tubules a major feature; gonial lobules and brown bodies may be evident; some Stage 1 previtellogenic oocytes present in varying amounts, but not the dominant feature. **Resting. mature:** Muscle tissue and reproductive tissue relatively equal, but muscle tissue prominent; fused seminiferous tubules suggesting prior spawning; sperm sinuses apparent; thickened tunica; 1^o spermatocytes and gonia usually the dominant tissue.
M3 (mature)	Gonad expanding and becoming rounded and large; greyish in appearance; milt may run from macroscopically sectioned gonad and some pockets of milt visible through the tunica	Sperm crypts and seminiferous lobules fused and primarily full of sperm/spermatids and 2^o spermatocytes; Sperm/spermatids and 2^o spermatocytes representing <50% of the total volume; sperm sinus present with thick or thickening tunica, but generally not filled or partially filled.
M4 (mature, active)	Testes large and white with sperm visible in sinuses; milt release with light abdominal pressure	Seminiferous lobules extensive, fused and filled primarily with sperm; sperm sinuses full and sperm not atretic; gonad volume > 50% sperm/spermatids, with relatively few gonia
M5 (post-spawn)	Testes flaccid and bloody; sperm release still possible on application of abdominal pressure	Sperm sinuses and seminiferous lobules largely empty or contracted with little or no sperm; muscle bundles abundant and gonad empty of early stages of spermatogenic tissue; thick tunica and some signs of early atresia of remaining sperm/spermatocytes.