

**Reproductive biology of yellowfin tuna (*Thunnus albacares*) in  
the Western and Central Indian Ocean.**

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*Abstract*

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*This study assesses the reproductive biology of the yellowfin, *Thunnus albacares*, of the Western Indian Ocean. A total of 1561 yellowfin were sampled from commercial catches on-board of a purse-seiner in three different areas of the Western Indian Ocean and Seychelles cannery (from January 2009 to May 2010).*

*Besides the description of different oocytes developmental stages, some preliminary results that could be compared with previous studies have been obtained. In the present work the size at first maturity was estimated at 77,8 cm, the female-ratio was found to be 1:0.9, mean batch fecundity of 2.5 million oocytes and the mean relative batch fecundity of 61.9 oocytes per gram of body weight was calculated. Moreover, the area between 0° north and 10 ° south has been identified as the most active spawning ground and January, February and June have been the months when most developed ovaries were found corresponding the highest GSI values (over 1.5 GSI value).*

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## INTRODUCTION

Yellowfin tuna (*Thunnus albacares*) is a large epipelagic species widely distributed in the tropical and subtropical waters of the major oceans (Collette and Nauen, 1983). It is an important component of tuna fisheries worldwide and the major target species for tuna fishery in the Indian Ocean (Somvanshi, 2002). Due to its high demand, yellowfin is harvested by different fishing gears. Contrary to the situation in other oceans, the artisanal fishery component in the Indian Ocean (mainly using pole and line, driftnet and hand line) is substantial, taking an estimated 35 % of the total YFT catches during recent years (2000-2008). Total annual catches have increased steadily since the start of the fishery in the late 50s, reaching the 100,000 t level in 1984, the 200,000 t level in 1989 and peaking at around 400,000 t in 1993. Total annual catches averaged 345,000 t over the period 1993 to 2002. Yellowfin catches in the Indian Ocean during 2003, 2004, 2005 and 2006 were much higher than in previous years (an average catch of 466,000t) but have returned to a lower level in 2007-2008 (318,000t.) (IOTC, 2009). The purse seiners are the dominant fishing method harvesting 37 % of yellowfin tuna total catches (IOTC, 2009). Although during last years the problem of increasing piracy activity has modified the harvested areas, PS operate mainly in the western equatorial region of the Indian Ocean where 47 % of yellowfin tuna total catches occurred (Langley et al,2008).

The knowledge on reproductive strategy of yellowfin tuna is important for a comprehensive understanding of the population dynamics and for predicting the effect of fishing on the reproductive potential of a stock, which is essential for effective management decisions and for the sustainability of the resource. (Scheafer, 2001; Murua & Motos, 2006). For this propose, the use of accurate techniques is necessary to avoid biases at the estimation of these parameters (sex ratio, size at first maturity, spawning season, annual fecundity, etc.) that improve the ability to manage the stock (Nootmorn et al, 2005).

Yellowfin tuna (oviparous and with no sexual dimorphism in external morphological characters) have an asynchronous oocyte development (Schaefer, 2001). However, there is no published information about the recruitment process of oocytes to regulate fecundity, i.e. fecundity is determinate or indeterminate (Hunter and Maciewicz; 1985;

Murua and Saborido-Rey, 2003; Kjesbu 2009). It was described that yellowfin tuna is a multiple egg batch-spawner, spawning over a vast areas of the tropical zone throughout the year (Schaefer, 2001) and beyond this area the spawning seems to be related to the sea-surface temperature (SST). It was also showed that SST could regulate the spawning activity of yellowfin tuna; for example spawning activity is limited in temperatures below 24 °C (Schaefer, 1998; Itano, 2000). In the Indian Ocean, the spawning seems to occur mainly in the equatorial area (0-10°S) from December to March, with the main spawning grounds west of 75°E (IOTC, 2003). There are works that extend this period between January to June (Zhu et al, 2008) and Stequer et al (2001) described two reproductive season related to north monsoon (main spawning period) and south monsoon (less reproductive activity). Different sizes at first maturity were presented for yellowfin tuna by previous authors showing deviance among them; IOTC (2009) and Zhu et al (2008) estimated size at first maturity at around 100 cm in the Indian Ocean, while McPherson (1991) estimated it at 108 cm in the Western Pacific Ocean, Schaefer (1998) estimated this parameter at 92 cm in the Eastern Pacific Ocean, and Itano (2000) obtained a size at first maturity at 104 cm for equatorial West Pacific. However, the information of the reproductive biology of yellowfin available in the literature is scarce and few studies are dealing with the estimation of fecundity parameters in the Indian Ocean

Thus, the objectives of this work are to evaluate the reproductive strategy of yellowfin in the central and western Indian Ocean as well as to estimate different reproductive parameters, such as sex-ratio, size at first maturity and batch fecundity; which, in turn, will contribute to the knowledge of those basic parameters in the study of population dynamics.

## MATERIAL AND METHODS

### *Field sampling*

For the collection of the samples 3 surveys were carried out in 2009 (2 surveys) and in 2010 (1 survey) onboard a commercial purse seiner covering the area of the west coast of the Indian Ocean, Somalia, East South Seychelles, North West Seychelles, Chagos and the Mozambique Channel (Fig. 1).

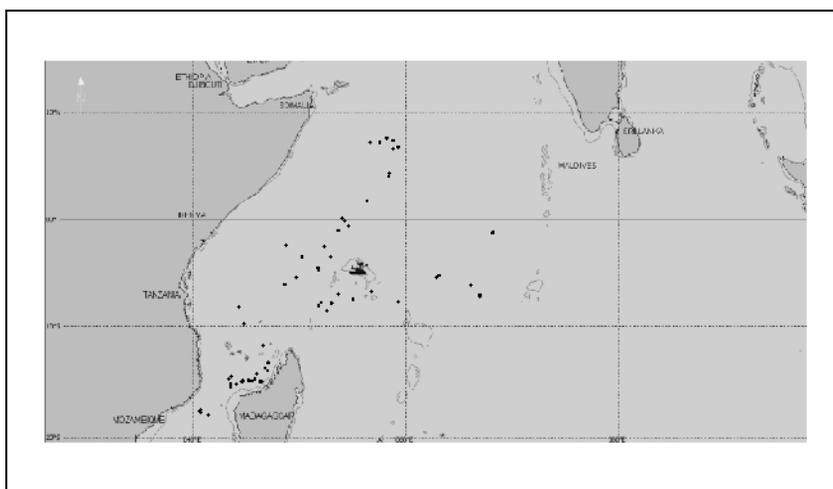


Fig 1. Location of points where yellowfin tuna were sampled in the Indian Ocean.

The first survey was performed from 21 January to 23 March 2009, the second survey was conducted from 5 June to 25 July 2009, and the third survey comprised from 3 April to 21 May. A total of 869 yellowfin tuna were sampled, 320 female, 284 males and 265 indeterminate (Table 1). Onboard purse seiner, each individual was measured (fork length and thorax perimeter) to the nearest centimetre and weighed to nearest 0.1 kg. Moreover, gutted weight, maturity, sex were measured. The sampled lengths ranged from 30 to 161 cm. The gonads were removed, sex determined and weighed to the nearest gram and a 4-5 cm cross-section was cut from the right or left lobes and preserved in a solution of 4 % buffered formaldehyde (Hunter, 1985).

Moreover, to complete the sampling of yellowfin tunas for reproductive studies, samples were obtained from Seychelles' cannery. A total of 692 female of yellowfin tuna, caught by the purse seiner fleet operating in the Indian Ocean, were sampled in the cannery. At the cannery, only females of yellowfin tuna were sampled. Each individual,

was measured (fork length and thorax perimeter) to the nearest centimetre and weighed to nearest 0.1 kg. The gonads were removed, sex determined and weighed to the nearest gram and a 4-5 cm length section was sectioned and preserved in a solution of 4 % buffered formaldehyde (Hunter, 1985).

To study the effect of the sampling area in the parameters studies, three areas were defined for the analyses: Area 1, defined as Seychelles covered the zone between 0° North to 10° South, Area 2 defined as Somalia covered the zone between 0° North to 10° North and the Area 3, defined as Mozambique falls between 10° South to 20° South.

Sampling	Area of sampling	Data	N° of samples	Length (cm)
1 <sup>st</sup> survey	1 and 2	22/01/09-13/03/09	282	37 – 158
2 <sup>nd</sup> survey	1 and 2	11/06/09-17/07/09	260	30 – 161
3 <sup>rd</sup> survey	1 and 3	03/04/10-21/05/10	327	31 – 157
Cannery	-----	12/01/10-13/04/10	692	61 - 147

The collected gonad provided information for histological oocyte development description, size at first maturity, oocyte size frequency distribution and estimation of batch fecundity.

#### *Gonosomatic index (GSI)*

The gonosomatic index was calculated as following:

$$GSI = (W_g / W) \times 10^2$$

Where  $W_g$  was the weight of gonads in grams and  $W$  was the gonad free weight of the fish.

#### *Histological analysis*

A cross section from the ovary was dehydrated in ascending solutions of alcohol and embedded in ascending solutions of resin. Posterior, 3-5  $\mu$ m section was cut using the microtome followed by Harri's Haematoxylin and Eosin staining process.

The histological classification of yellowfin tuna ovaries followed the criteria of Wallace and Selman (1981) taken into account the modification made by Scheafer (1996, 1998) on the system of Hunter and Macewicz (1985). Each ovary was classified according to the most advanced oocyte stage present in the ovary (Murua & Motos, 2000). The different stages of atresia (resorption of oocytes and its follicle) were classified following the Hunter and Macewicz (1985) criteria and modified for yellowfin tuna (Table 2). The presence of postovulatory follicles (POF) was recorded. Taken into account the asynchronous development of yellowfin tuna ovary which means that oocytes of all stages are present without dominant populations (Wallace & Selman, 1981); females of yellowfin were classified as *immature* when all oocytes in the ovary present primary growth stage. In the other hand, ovaries with presence of oocytes at cortical alveoli stage or further developed stages were classified as *mature*.

**Table 2.** Atresia categories applied for the analysis following the description of Hunter and Macewicz (1985) and Murua and Motos (2006)

Atretic state	Atretic condition	Presence of POF	Spawning condition
0	$\alpha$ -atresia does not appear in cortical alveoli oocytes or in vitellogenic oocytes	NO	Prespawning active
1	$\alpha$ - atresia does not appear in cortical alveoli oocytes or in vitellogenic oocytes.	YES	Spawning
2	Incident of $\alpha$ - atresia is < 50% in yolked oocytes	YES	Spawning
3	Incident of $\alpha$ - atresia is > 50% in yolked oocytes	YES/NO	Inactive /Postspawning
4	Incidence of $\alpha$ - atresia in cortical alveoli or yolked oocytes is 100% or $\beta$ - atresia is present in all oocytes	---	Postspawning

### *Length at first maturity ( $L_{50}$ )*

The length at which 50 percent of females are mature was calculated using the proportion of mature females by 5 cm length classes, analysis histologically, fitted to a logistic equation as described by Ashton (1972) and applied to other species (Saborido & Junquera, 1998):

$$P = \frac{e^{\alpha+\beta L}}{1+e^{\alpha+\beta L}}$$

Where P is the predicted mature females' proportion and L is the fork length in cm;  $\alpha$  and  $\beta$  are the coefficients of the logistic equation. A nonlinear regression (the Marquardt

method without restrictions; Marquardt, 1963) was used to fit the logistic equation to the data.

### *Oocyte diameter distribution*

The selection of gonad for oocyte size frequency distribution analysis covered all oocytes stages by size range and sampling time. From each preserved gonad 0.04g ( $\pm$  0.0001g) of sample was collected and stained with Rose Bengel stain during at least 24h. Dyed sample was turned into a 125  $\mu$ m sieve and pulverized with fresh water in order to disaggregate the oocytes from the tissue. Oocytes were collected in a Petri dish. Photographs of each oocytes group were taken by a digital camera. Subsequently, each image was analyzed by imageJ, which is an image analysis software automatically counting and measuring all the oocytes of the image.

### *Batch fecundity*

Batch fecundity (i.e total number of oocytes that will be released per batch) were estimated using the gravimetric method (Hunter et al, 1989) by counting the oocytes in the most advance stage of maturation (migration and hydration phase) where the gap in size diameter between the standing stock of oocytes for next batches is developed. For the analyses 3 subsamples of 0.1 g ( $\pm$ 0.01) of each ovary were immersed in glycerine (Schaefer, 1987). The counting of the most advanced stage oocytes was done under a stereomicroscope. Batch fecundity was calculated as the weighted mean density of three subsamples multiplied by the total weight of the ovary. Relative batch fecundity was estimated dividing the batch fecundity and the gonad-free female weight.

## RESULTS

### *Sampled area, length frequency distribution and sex-ratio.*

.As shown in Fig. 2 the size distribution of fishes sampled at sea goes from 30 cm to 161 cm, and at the cannery goes from 61 cm to 147 cm.

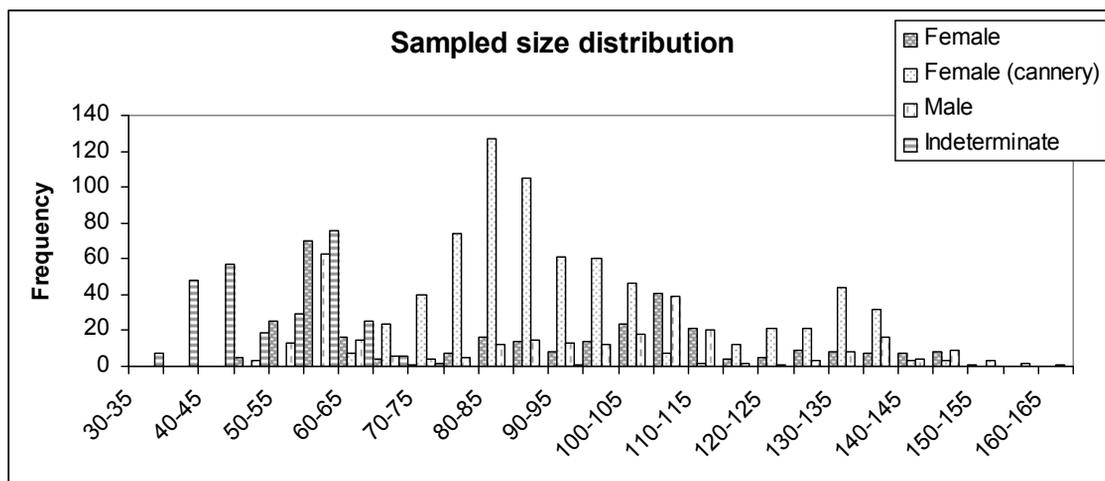


Fig 2. Yellowfin tuna size distribution sampled onboard purse seiner vessel. and cannery.

Sex determination of sampled fishes allowed analyzing the proportion of sex (male and female). The sex ratio over all yellowfin size was 1:0.9 (F:M), varied from 1:0.2 to 1:3. The Chi square values for each length classes are represented in the table.3. All length classes appeared not significantly different ( $p$  values  $> 0.05$ ) from the expected 1:1 with. Fig.3 shows the distribution of both sex proportions through 5 cm length classes where the proportion of females was slightly predominant at small and intermediate sizes. Nevertheless, proportion of males was significantly dominant at large size fishes ( $>135$ cm) where the female proportion disappeared over 155 cm fork length. This pattern has been described for yellowfin in different oceans (Fonteneau, 2005) and similar percentages were obtained for other tuna specie (Fonteneau, 2002; Sun et al, 2005; Marsac et al, 2006).

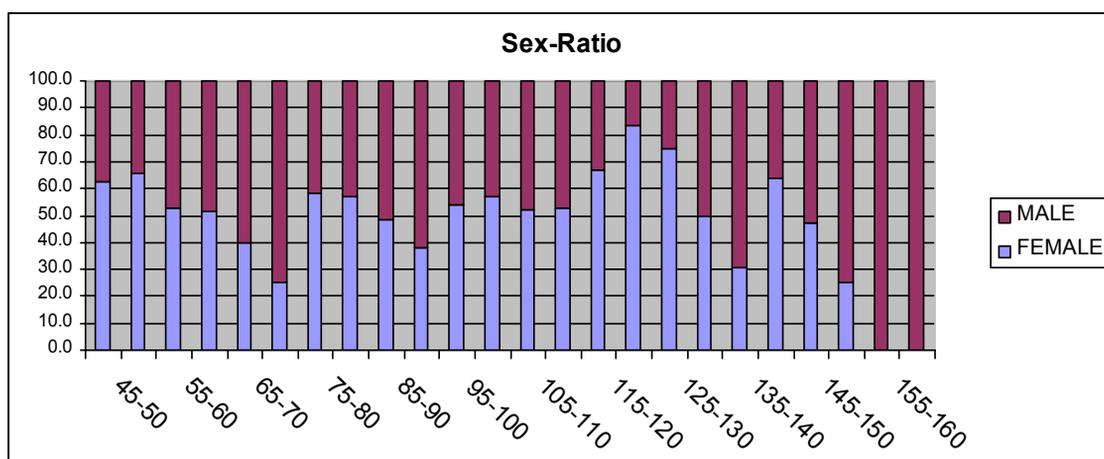


Fig 3. Proportion of males and females by 5 cm length classes.

Length Classes	Females	Males	Chi-square $\chi^2$
45-50	5	3	8
50-55	25	13	38
55-60	70	63	133
60-65	16	15	31
65-70	4	6	10
70-75	1	3	4
75-80	7	5	12
80-85	16	12	28
85-90	14	15	29
90-95	8	13	21
95-100	14	12	26
100-105	24	18	42
105-110	41	38	79
110-115	21	19	40
115-120	4	2	6
120-125	5	1	6
125-130	9	3	12
130-135	8	8	16
135-140	7	16	23
140-145	7	4	11
145-150	8	9	17
150-155	1	3	4
155-160	0	2	2
160-165	0	1	1

\* 0.05 > P > 0.01

\*\* P < 0.01

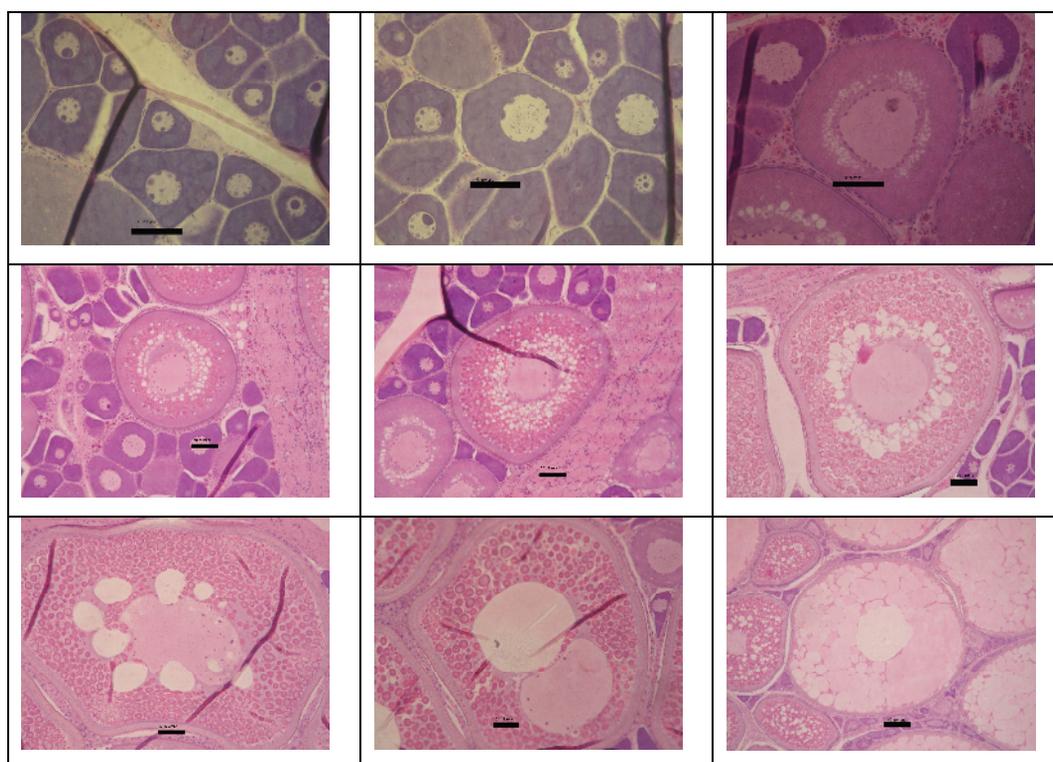
### *Stages of oocyte development*

From the total 423 females (292 from sea sampling and 131 from cannery) were analyzed histologically. The histological analysis of yellowfin tuna ovaries describes an asynchronous development where all oocyte stages are presented without a dominant

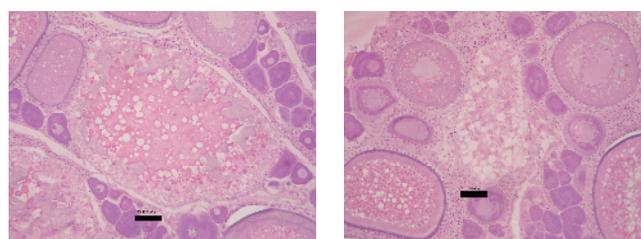
population of oocytes randomly mixed (Schaefer, Murua & Saborido,2003). The oocyte in its development process pass through different stages before they are spawned (Murua & Motos, 2000). The table.4 and figure 4 summarized the different oocytes development stages identified according to the classification described by Wallace and Selman (1981) and West (1990) and modified for yellowfin tuna.

Table 4. Summary of oocyte developmental stages in yellowfin tuna. The morphological characteristics and size ranges are given for each stage. Measurements are made in histological material.

Oocyte development stage	Characteristic	Oocyte diameter (µm)
<b>Primary growth</b>		
Chromatin nuclear	Oocyte is surrounded by a few squamous follicle cells. The nucleus is large and centrally –located, surrounded by a thin layer of cytoplasm and containing a large and single nucleolus.	
Perinucleolar	Nucleus increase in size and multiple nucleoli appear at its periphery. The “Balbiani bodies” migrated from the nucleus to the cytoplasm. At the end, some vacuoles appear in the cytoplasm and the chorion precursors material start to accumulate in patches.	
<b>Cortical alveoli formation</b>	Start to appear spherical vesicle at the periphery of the cytoplasm. They increase in size and number forming rows and giving rise to cortical alveoli. Oil drops begin to accumulate in the cytoplasm. At this stage chorion and follicle layers are apparent.	95 – 220
<b>Vitellogenic (yolked)</b>	This stage is characterized by the appearance of “true” yolk vesicle in the cytoplasm. Besides, the separation of the chorion in two different layers occurred: inner and outer zona radiate. This stage is subdivided in 3 different stages.	
VIT 1	Oil droplets occupy more cytoplasmic area than yolk granules.	220 – 275
VIT 2	Oil droplets occupy similar cytoplasmic area than yolk granules.	275 – 400
VIT 3	Oil droplets occupy less cytoplasmic area than yolk granules.	400 – 500
<b>Maturation</b>		
Initial migration	The nucleus (germinal vesicle) starts to migrate to the animal pole and the oil droplets fuse to coalescence into a unique oil globule.	500 – 550
Advanced migration	The nucleus completes its migration to the animal pole and the unique oil droplet is clearly evident at central part of the oocyte.	550 – 600
Hydration	Yolk granules fuse in yolk plates, and at late on in a homogeneous mass. The nucleus has disintegrated and the cortical alveoli and cytoplasm are restricted to a thin peripheral layer. The oocyte significantly increases in size due to the uptake of fluids. Hydrated oocyte has a translucent appearance.	600 – 750



**Figure 4.-** From left to right in the first line, chromatin nuclear, perinuclear and cortical alveoli stage are represented. From left to right in the second line, three stages of the vitellogenic phase are presented (VIT 1, VIT 2 and VIT 3 respectively). In the third line from left to right maturation phases are represented (early migration, late migration and hydration respectively).



**Figure 5.-** Pictures of alfa (left) and beta (right) atresia in yellowfin tuna oocytes.

### *Oocyte diameter distribution*

The oocyte diameter frequency distribution for each ovarian stage is described in Fig 6. The size frequency distribution at different development stages from cortical alveoli, early and late vitellogenesis stages (VIT 1, VIT 2 and VIT 3) is continuous without any gap in diameter between previtellogenic and vitellogenic oocytes; providing evidences for a continuous recruitment of oocytes during spawning season (i.e. indeterminate fecundity). The largest largest modal group of advanced oocytes starts to be separate in diameter from the advanced yolked stage during initial migration at around 490  $\mu\text{m}$ . At

hydration stage the oocytes increase the size above 700  $\mu\text{m}$  and the hiatus between the coming batch and reservoir of oocytes for next batches is evident.

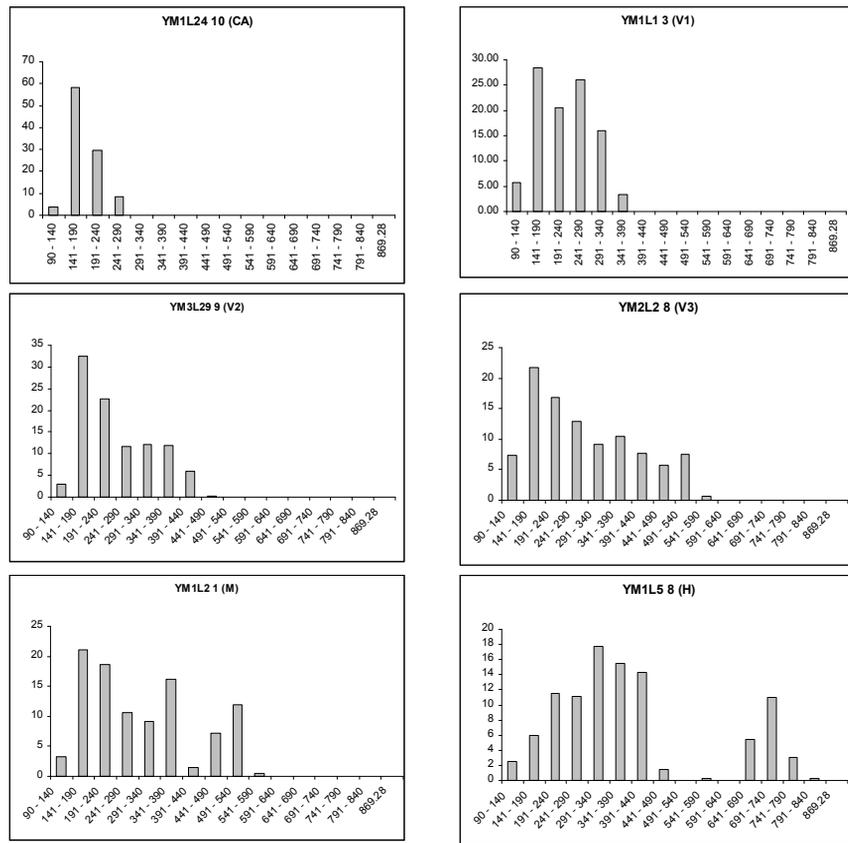


Figure 6.- The oocyte diameter frequency distribution for each ovarian stage.

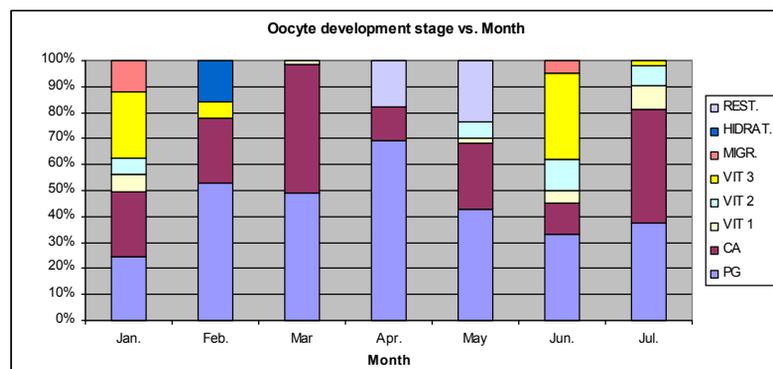
### Seasonal variation in occurrence of various oocyte stages

According to the classification summarized in the Table 1, 39.0 % of the individuals were at the primary stages, 28.6 % were at CA, 22.2 % were at vitellogenic stage (4.5 % VIT 1, 5.0 % VIT 2, 12.8 % VIT 3), 5.9 % at maturation (4.7 % Migratory, 1.2 % Hydrated) and 4.3 % of the fishes showed ovaries at resting stage (atretic stage 3 and 4) (Table 5). The 60 % of sampled females (258 up to 423 females) were mature, from which the 93 % were active and 7 % inactive at resting stage.

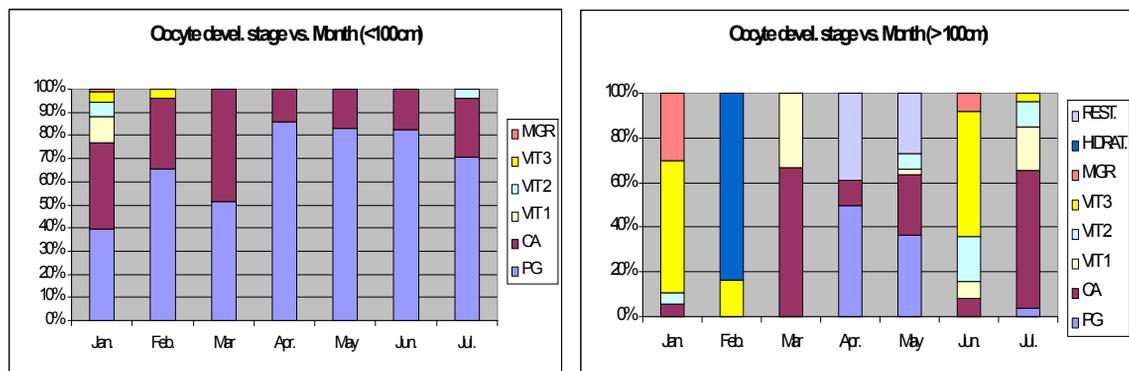
**Table 5.-** Summary of oocytes developmental stages by 5 cm length classes

Length (cm)	Immature	Mature						Total
		Active					Inactive	
		C.A.	VIT 1	VIT 2	VIT 3	MIGR.		
48-53	15	2						17
53-58	32	6						38
58-63	38	3						41
63-68	12	5						17
68-73	15	6	1	1				23
73-78	12	7	1	1				21
78-83	5	16	3	4	1			29
83-88	5	19	5	1	1	1		32
88-93	1	11			2			14
93-98	2	6			1			9
98-103	7	7	1				1	16
103-108	17	10	3	3	2		10	45
108-113	4	14	1		1		6	26
113-118		2	1	1	2			6
118-123		2			7		1	10
123-128				1	3	4	1	9
128-133			1	3	12	4	1	21
133-138			1	2	12	6		21
138-143		4		3	9	3	1	20
143-148		1		1	1	2	1	6
148-153			1				1	2
Total		121	19	21	54	20	5	18
Total	165				258			423

The proportion of oocytes developmental stages in relation with spatial and temporal factors are further analyzed in the Fig. 7. Regarding to temporal factor (month), January, February and June are the months where ovaries appear more developed with oocytes at advanced vitellogenic, migratory and hydrated stages. In contrast, March, April and May are the months where the ovaries are less developed, with high proportion of primary growth and cortical alveoli as the most advanced stage of development (more than 50 % of the specimens).

**Figure 7.-** Proportion of different oocyte developmental stages analyzed by month.

Moreover, the influence of seasonality upon the development of oocyte stages was also analyzed by fish size. The trend in oocyte development seasonality is similar to the overall pattern when fishes are divided in two groups: fish larger than 100 cm long (FL) and smaller than 100 cm (published length at first maturity). The smaller group of fishes (Figure 8) shows a considerable predominance of primary growth stage during all months with percentage of 50 % or higher; whereas in the case of larger fish the variability among months is more evident and January, February and June are again when the ovaries are more developed and closer to spawning.



**Figure 8.** - Proportion of different oocyte developmental stages analyzed by month and regrouping surveyed population between larger and smaller than 100 cm.

Same analyses were applied for each surveyed area. In Figure 9 can be observed that area 1 which was defined as Seychelles shows the more developed ovary stages with more than 60 % of mature females, from which more than 24 % was at vitellogenesis stage and 8 % at maturation. In the other two areas the proportion of primary growth and cortical alveoli developmental stages were higher than the rest of ovary stages. In Figure 10 the relation between the oocyte developmental stage and area is observed for different fish length classes (<100cm and >100cm). The comparison between areas could not be accurate as surveys were performed during different period of time in each area, with only short period of overlapping time between areas.

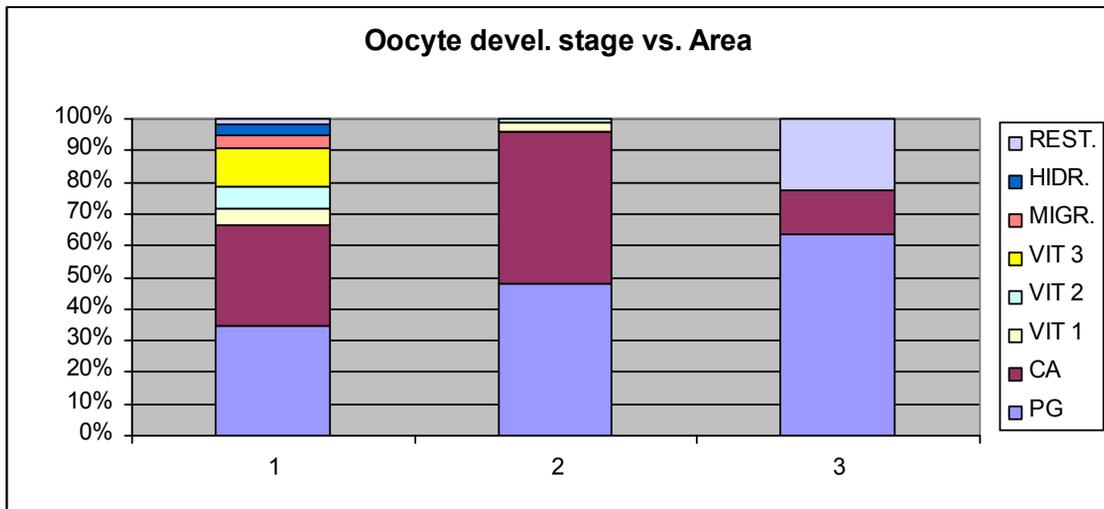


Figure 9.- Proportion of different oocyte developmental stages analyzed by area.

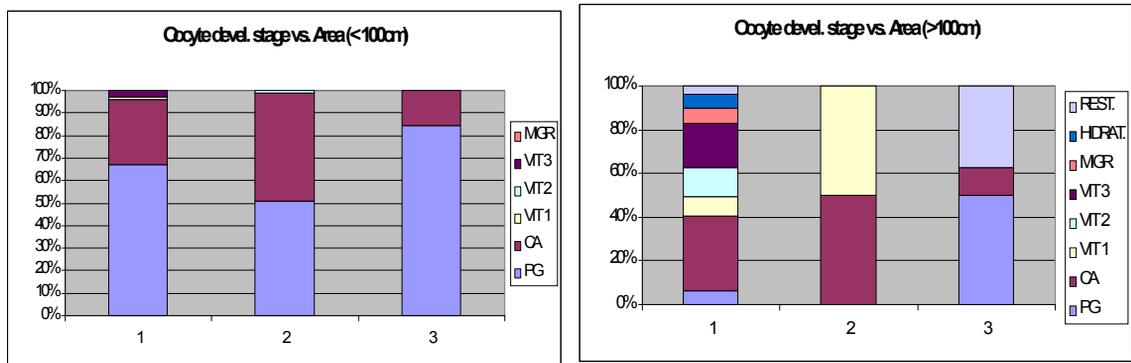


Figure 10.- Proportion of different oocyte developmental stages analyzed by area and regrouping surveyed population between larger and shorter than 100 cm and larger and shorter 78 cm.

Figure 11 shows the seasonality of oocyte developmental stages in area 1, where surveys were performed from January to February (2009), May (2010) and from June to July (2009). In this figure a pattern similar to the fig 7 can be observed being January, February, and June, July the months showing most developed ovaries.

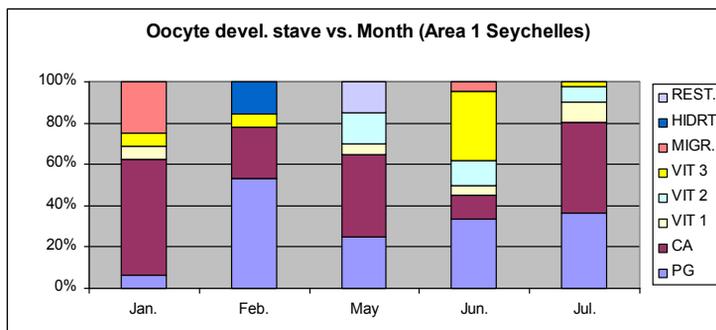


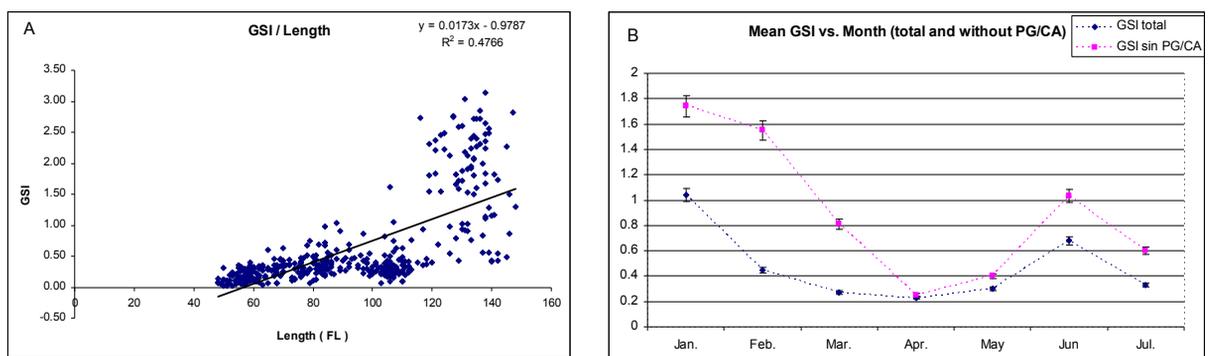
Figure 11.. Proportion of different oocyte developmental stages analyzed by month and for Seychelles area

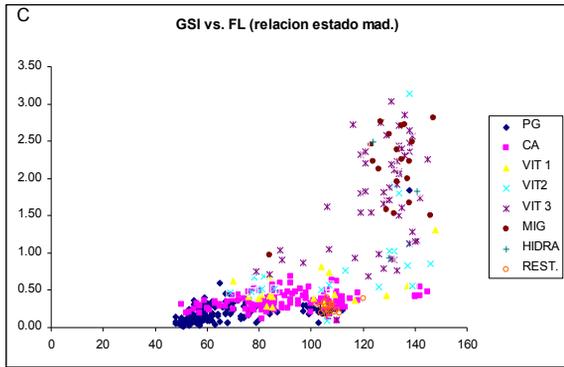
## *Gonosomatic indices (GSI)*

In Figure 12 two periods where the mean GSI was high can be observed; the first period corresponds to January and February (with 1.04 and 0.45 respectively) and the second period to June with values of 0.68. These results are similar to those described by Stéquert et al (2001), where two reproductive seasons were described; the first and main one during the north monsoon (November to March), and the second one and with lower percentage of spawning females during the south monsoon (June to August).

These two reproductive seasons are more appreciable if primary growth (PG) and cortical alveoli (CA) oocyte developmental stages are removed from the analysis. In this case (Figure 12), the mean GSI values increase and a main period corresponding to January, February and March is observed with a second peak on June and July. During April and May the mean GSI remains low with values similar to the analysis including PG and CA (Figure 12). However, it has to be taken into account that low values months correspond to 2010 survey and that difference in GSI could be driven by area/annual variation.

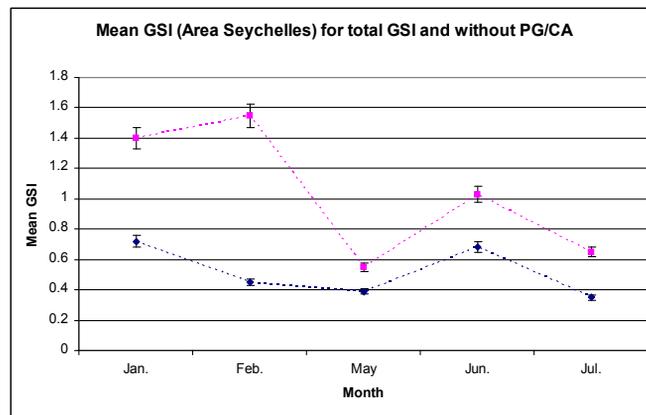
The relation among yellowfin tuna fork length and GSI values are shown in the Figure 12. An increase in GSI values is observed as the size of the fish increase. Moreover, in the fig.12 (c) can be observed that the higher values of GSI correspond to the more advanced oocyte development stage.





**Figure 12.-** (A) The variation of GSI values with the fork length (cm) in yellowfin tuna females. (B) Mean GSI values by month including all developmental stages and excluding PG and CA stages. (C) The GSI values variation regarding the oocyte developmental stages.

Moreover, the mean GSI values for the area of Seychelles is similar to the overall pattern (Figure 12); where a decrease of the mean GSI value in January is observed, however the values are in the range corresponding to spawning stage.



**Figure 13.-** Mean GSI values by month for the Seychelles area.

### *Stages of oocyte development and corresponded GSI range*

The variability of GSI values at each ovarian stage is shown in Figure 14 and Table 6. Range of values at most developed ovaries is wider and there is an increase of GSI values as the ovary develops. Advanced vitellogenesis (VIT 3), migratory nucleus and hydration stages shows mean GSI values over 1.5 which corresponds to stages related with spawning activity. The GSI values at resting stage were similar to PG stage, which is representative of the stage of the ovary.

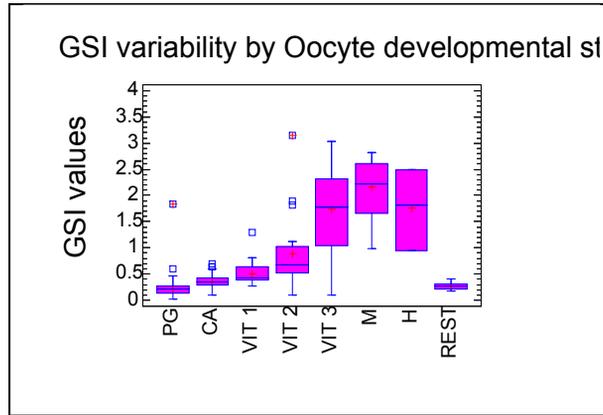


Figure 14.- Representation of GSI values range for each of the oocyte developmental stages.

Table.6. Max. and min. GSI values for each of oocyte developmental stages.

GSI values vs. Oocyte development stage								
	PG	C.A	VIT 1	VIT 2	VIT 3	MIG.	HIDR.	
Min.		0.02	0.1	0.28	0.46	0.69	1.51	0.94
Mean		0.22	0.36	0.51	0.68	1.72	2.22	1.75
Max.		0.46	0.69	0.81	1.12	2.75	2.82	2.49

*Length at first maturity (L<sub>50</sub>)*

Table 7 shows the parameters obtained by the fitted logit model that were used to obtain the maturity curve showed in the fig. 15. The Length at first maturity was estimated at 77.8 cm fork length.

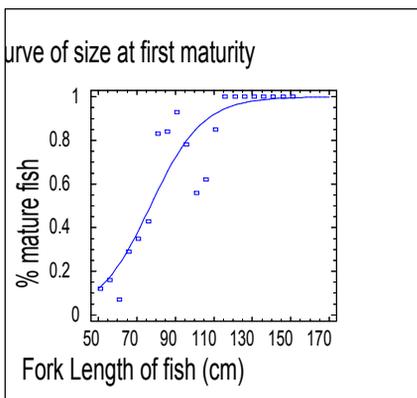


Table 7. Summary of logit parameters for yellowfin tuna females maturity curve.

Parameters	$\alpha$	$\beta$
Estimate	-5.68885	0.0737604
Standard error	1.42676	0.0183982
N° of females = 423		
L50(- $\alpha$ / $\beta$ )= 77.8 cm		
R squared = 80 %		

Figure 15.-

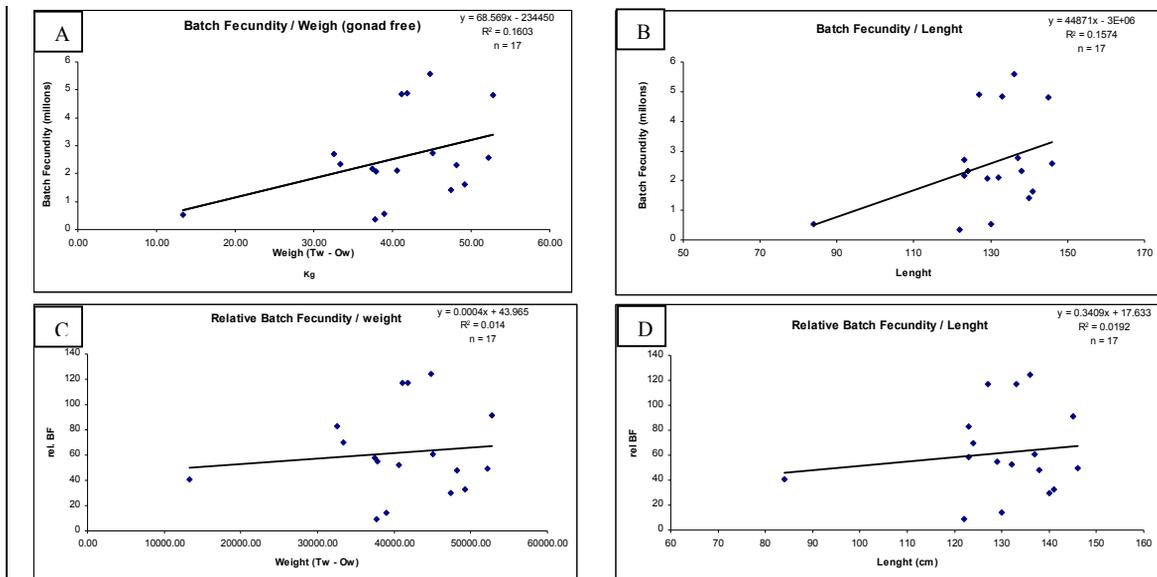
Curve of size at first maturity

### Batch fecundity

For the batch fecundity estimation 17 ovaries with advanced matured oocytes (migratory nucleus and hydrated stages) were analyzed by gravimetric method. All of them present a fully developed hiatus and the oocytes were easily distinguished. Selected ovaries do not present new postovulatory follicles that could indicate that part of the batch was already ovulated and bias the batch fecundity estimation.

The estimated mean batch fecundity was 2.56 millions and it varied from 0.34 millions to 5.57 millions of oocytes. BF was related to gonad-free female weight and length positively with a linear regression, although the relationship only explain little variability observed ( $R^2 = 0.16$  and  $R^2 = 0.157$  respectively) (Figure 15).

The estimated mean relative batch fecundity for 17 specimens was 61.9 oocytes per gram of body weight. The relative batch fecundity ranged from 9.23 to 124.56 oocytes per gram of fish.



**Figure.16.-** (A) Batch fecundity and female gonad free weight relationship. (B) Batch fecundity and yellowfin female length relationship. (C) Relative batch fecundity and female gonad free weight relationship. (D) Relative batch fecundity and yellowfin female length relationship.

## DISCUSSION

Changes of sex ratio by fish length are an important parameter which may be directly related with differences at growth rate, and/or natural and fishing mortalities by sex (Fonteneau, 2002). The high proportion of males at large size has been observed in the main three oceans (Capisano, 1991; Schaefer, 1998; Timochina, 1992). In the Indian Ocean the size at which males start to be dominant was reported at 154 cm (Fonteneau, 2002), and more recent works of Nootmorn et al (2005), Marsac et al (2006) and Zhu et al (2008) have noticed that this size could be around 145 cm. According to Marsac et al (2006) this could be representative of high fishing mortalities at large fishes in the last decade. The females dominant pattern described for the Atlantic Ocean between 125 and 140 cm was not observed by Fonteneau (2002) in the Indian Ocean; however, Nootmorn et al (2005) noticed a different range where females proportion shows a dominant pattern between 95 and 135 cm. In the present study, the proportion of males was significantly dominant at large size when fork length was larger than 150 cm and female proportion disappeared over 155 cm fork length. These estimate is higher than those obtained by Marsac et al (2006), Nootmorn et al (2005) and Zhu et al (2008) and is lower than those obtained by Fonteneau (2002). Moreover, it was observed that females proportion was higher than males at fork length between 115 and 130 cm fork length which falls into the range described by Nootmorn (2005). However, the sample size is not enough to accurately described sex-ratio specially for larger fishes.

The size at first maturation for yellowfin tuna in the Indian Ocean was described by Stequer and Marsac (1989), between 120 cm and 140 cm. Bashmaker et al (1991) found that yellowfin tuna females were matured at 120 cm in the western part of the Indian Ocean; and Zhu et al. (2008) estimated a value of 100 cm for yellowfin females. According to Romena (2000) the maturity of yellowfin tuna females ranged from 75.9 cm to 134.5 cm. Others authors have described the length at first maturity for yellowfin females in other oceans; for example, McPherson (1991) estimated that 50 percent of yellowfin females reach the maturity at 108 cm in the Western Pacific Ocean. Schaefer (1998) estimated the length at first maturity for yellowfin tuna females at 92 cm in the Eastern Pacific Ocean. In the present study, the length at first maturity ( $L_{50}$ ) for yellowfin tuna females in the Western Indian Ocean was estimated at 77.8 cm fork length. This value is significantly smaller than the length determined in previous studies

by Stequer and Marsac (1989), Bashmaker et al (1991) and Zhu et al. (2008); and it is more similar to the values obtained by Romena (2000).

The differences on the estimates of above studies can really in the fact that different oocyte stages were used to define a mature female. In the present work, ovaries with the most advanced oocytes in the cortical alveoli stage were defined as mature (Murua and Motos, 2000; Lowerre-Barbieri et al., 2009; Brown-Peterson et al., in press). This was because, the cortical alveoli stage, which is dependent of gonadotropin for their formation (Wallace and Selman 1981, Luckenbach et al. 2008; Lubzens et al. 2010), is the precursor of vitellogenesis indicating the onset of oocyte development and, hence, gonad maturation (Murua and Motos, 2000). However, different criteria were applied for the definition of mature females in previous work of McPherson (1991), Schaefer (1996, 1998), Itano (2000) and Zhu et al (2008) where mature stage of female fishes was corresponded to an advanced vitellogenesis phase or further developed ovaries. Mackie and Lewis (2001) put forward the discussion of defining cortical alveoli stage as mature fish, since it is uncertain what fractions of the females that begin vitellogenesis actually complete it.

Moreover, the fishing method used for obtaining the fish could also explain the differences in maturity ogive between studies (Koido and Szuki, 1989; Schaefer, 1998). Depending on the fishing methods, it could exist a limitation in the availability of some size of fish and in turn, affect in the estimation of size at first maturity.

In the Indian Ocean, the spawning was reported to occur mainly in the equatorial area (0-10°S) from December to March, with the main spawning grounds west of 75°E (IOTC, 2003). Stequer and Marsac (1989) reported a spawning intensity between January and March in the central Indian Ocean and Stequer et al (2001) also reported a main reproductive season during the north monsoon (November to March), and the second one and with lower percentage of spawning females during the south monsoon (June to August). Nootmorn et al (2005) estimated the spawning season for females and males between November and April in the eastern Indian Ocean. Zhu et al (2008) reported a spawning season for females between January and June in the West Central Indian Ocean. In the present work, the mean GSI values estimated by season showed two reproductive seasons; the first period with the highest values, January and February,

with 1.74 and 1.55 respectively, and the second period on June with values of 1.03. These results are consistent with estimated by Stéquert et al (2001), where two reproductive season were described; the first and main one during the north monsoon (November to March), and the second one and with lower percentage of spawning females during the south monsoon (June to August). The GSI values observed during January, February and June are over 1.5 and according to Kiodo et al (1989), Stequert & Marsac (1989) and Nootmorn et al (2005), the values of GSI between 1.5 and 2.5 correspond to fish capable to reproduce. In this sense, the relation between oocyte developmental stages and GSI observed in this study agrees with the results presented by other authors. The lower values of GSI corresponded to samples with high proportion of primary growth and cortical alveoli as the most advanced stage of oocyte development (more than 50 % of the specimens) and periods with low reproductive activity with mean GSI values ranging between 0.25 and 0.80. The area of west of Seychelles, corresponding to the limits between 0° north and 10° south, is where more reproductive activity was observed in yellowfin females. This area is included into the zone defined as the main spawning ground for yellowfin tuna, at west of 75° East (Langley et al, 2009, IOTC, 2003).

The values of batch fecundity and relative batch fecundity obtained in this study are similar to those described by Schaefer (1996, 1998) for Eastern Pacific Ocean yellowfin and Sun et al (2005) for the Western Pacific Ocean where 68.0, 67.3 and 62.1 oocytes per gram of body weight were estimated respectively. However, it is higher than the 54.7 oocytes per body gram reported by Itano (2000) in the Western Pacific. The estimated mean batch fecundity at present work was 2.56 million oocyte, similar to the value observed by Sun et al (2005) who reported a batch fecundity of 2.71 million oocyte, whereas it was significantly higher than the value reported by Schaefer (1996, 1998) who reported 1.57 million and 1.55 million respectively. Itano (2000) obtained a significantly higher value of 3.45 million oocytes.

Although the sampling through a complete year was not possible and, hence, the different areas were surveyed at different period of time, the spatio-temporal influence on the reproductive biology analysed here can be of valuable interest to compare the spawning activity along the western Indian Ocean region.

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