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Reproductive biology of the sea cucumber *Holothuria scabra* (Jaeger 1883) in Mahout Bay, Arabian Sea, Oman

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Abstract
The first official records of the exploitation of *Holothuria scabra* in Oman were reported around 10 years ago. Unfortunately, the population of this exploited stock followed the same rapid decline as most exploited population of this sea cucumber elsewhere. *H. scabra* has always been part of the traditional exploitation of the benthos in Ghubbat Hashish of Mahout Bay but the foreign demand for the product overseas has put increased pressure on the resource and led to overfishing. To fill the need for a management strategy as well as to establish the background studies necessary to assess the aquaculture potential of this species in Oman, it was thus necessary to assess its reproductive biology. The reproductive cycle of *H. scabra* population in Mahout was determined from monthly variations of the gonad somatic index based on dry weight relationships. Its reproductive status was examined from October 2008 to May 2010 with 15-30 individuals collected monthly. An annual period of reproduction was observed during late spring (pre-monsoon period) when a peak of GI index was recorded at 12% and 9% in April 2009 and 2010, respectively. The size at first maturity was estimated at 16.5 cm which corresponds to approximately 600 g fresh weight. The spawning appeared to be correlated to high temperature and precipitation which both may serve as exogenous cues for *H. scabra* gamete release. The application of this study in fisheries management and aquaculture is discussed.

Keywords: sea cucumber, *Holothuria scabra*, reproductive biology, spawning season, Oman

1. Introduction
Sea cucumbers are one of the high marine products demanded both for foodstuffs and medicine [15, 22, 43]. They are mostly exported in a dried form mainly under two trading names; trepang or bechedemer, but small quantities of both fresh and frozen sea cucumbers also enters the international market in particular to Japan [14, 45]. Nearly 90% of this trade takes place in the Far East where China, Hong Kong and Singapore dominate the business and China remains the main ultimate consumer [55, 45]. The price per dry kg of high premium *H. scabra* bechedemer in Hong Kong has jumped over USD 1000 [45]. Therefore, over-exploitation is witnessed in almost all *H. scabra* producing countries [11, 15, 33] Over 21 holothurian species have been recently recorded in the shallow waters of Oman [9]. Of these only *Holothuria scabra* is exploited commercially and has a value of about OMR 1.5 per individual [3]. This minor fishery is limited on the western side of Masirah Island and Mahout Bay which is characterized by large, shallow seagrass beds with fine sand in sheltered flats and lagoons [1, 2]. The first official records of the exploitation of *H. scabra* in Oman were reported around 10 years ago [3]. Unfortunately, this exploited stock followed the same rapid decline as most exploited population of this sea cucumber elsewhere [1, 12, 43, 57]. Overfishing evidences include a rapid decrease in population density which has been critically reduced to less than one individual per hectare as reported not only in Oman [1], but in many parts of the world, including [55] Milne Bay Province, [55] Papua New Guinea, [10] the Red Sea and [20] Sri Lanka. The fast overfishing characteristic of this species has been attributed to low recruitment, slow growth rate, late maturity and density-dependent reproductive success [13, 58] Indeed it has gone beyond overfishing and has been identified as distinctive and endangered species listed under international union for the conservation of nature (IUCN) red list [43, 57]. One of the global strategies applied to regulate the depleted stock and develop successful aquaculture programs is to study the reproductive biology which has been already investigated in almost all of its geographical range [47].

The reproductive biology of the Omani *H. scabra* population, which corresponds perhaps to northern limits of its Indian Ocean distribution, has not yet been described. Therefore, the present work aims to assess the reproductive biology of Omani *H. scabra* using gonado-somatic indices (GSI) method and estimate the size at first sexual maturity: both of capital importance for management strategy and aquaculture development.

2. Materials and Methods

2.1 Collection area

The study was carried out on *H. scabra* individuals collected in several seagrass beds of Mahout Bay (Ghubbat Hashish Bay; 20°27' N 58°10' E) of the Arabian Sea which is the only known location for holothurians (*H. scabra*) and shrimp (*Penaeus indicus*) fisheries in the Sultanate of Oman (Figure 1). Adults were collected almost within two years period from October 2008 to May 2010. The semi-sheltered Bay covers approximately 320 km^2 in Eastern Oman (Figure 1) and has a maximum depth of about 10 m in the southern part but most of the Bay is less than 5 m in depth. Tidal amplitude is about 1.6 m during spring tides and the Bay remains relatively protected from the severe wave climate generated by the South East monsoon winds during the summer. Dense stands of the mangrove *Avicennia marina* fringe the northern and western sides of Mahout [31] and the sandy bottom of the Bay is covered with sparse to dense sea grass beds dominated by *Halodule uninervis* and *Halophila ovalis* [2], making it an important habitat for several species including green turtles and shrimps.

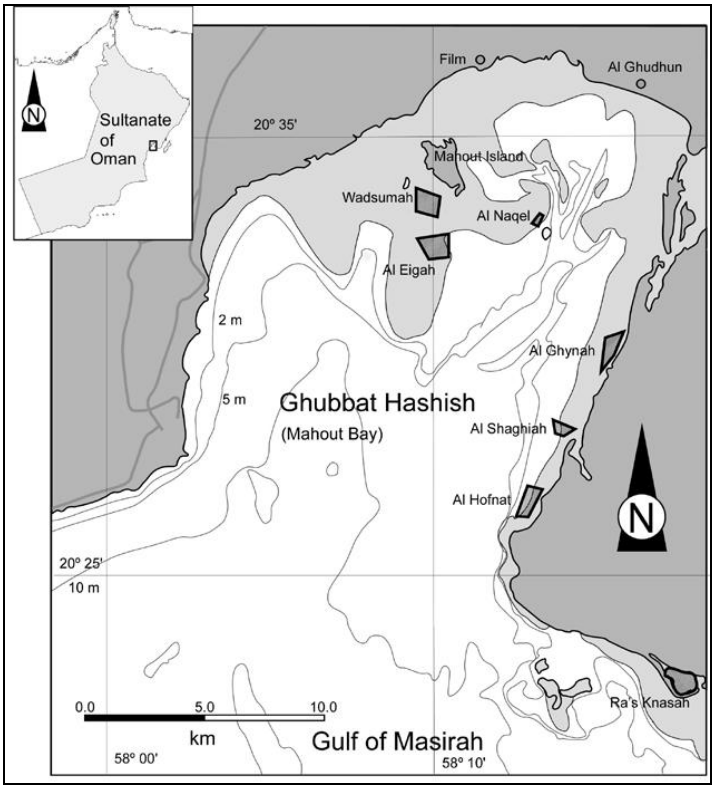

Fig 1: Map of Mahout Bay showing collection sites.

2.2 Sex ratio &Length-weight relationship

The chi-squared test was used to test whether there was a significant difference in the number of males and females sampled in each sampling period. Length measurements of Holothuroids are known to be difficult as they contract and retract their body constantly. Length was measured from scaled photographs of the bivium of relaxed individuals, using an ad-hoc software (*Holometer*) that allowed the measuring line to follow the middle row of tube feet (Figure 2). The length-weight relationship was modelled using a power relationship of the form:

W = a. Length ^b

Where W is the weight of the body wall and Length is the "curved length" of the body.

Because gonads were very variable in size (up to 15% of the dry weight), only the weight of the body wall was used as an indicator of size.

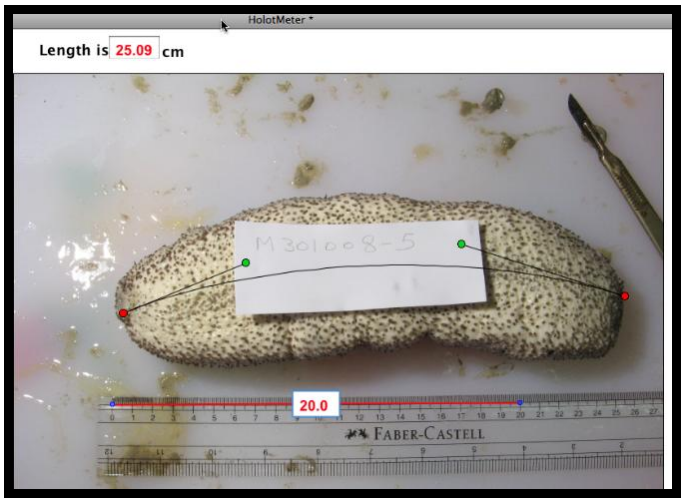

Fig 2: The ad-hoc software Holometer was used to estimate the total length based on a sea cucumber ventral image from mouth to anus tips.

2.3 Gonado-Somatic Index

On 20 occasions between October 2008 and May 2010 and on an approximately monthly basis, 15-30 individuals of *H. scabra* were collected from shallow water at low tides or purchased from fishers. Each specimen was measured to the nearest 0.5 cm and weighed to the nearest gram (TW) then photographed with a standard ruler for accurate length measurement (TL) using *Holometer* (Figure 2). The animal where then dissected to separate the main components: body wall (including longitudinal muscles), digestive tract, respiratory tree and gonads. The digestive tract was discarded because of its very high and variable content in sand and its negligible weight in comparison to the other components of the body. These components were weighed fresh, then after 96 h drying period in a laboratory drier at 60-70 ºC. Because echinoderm fluids are similar to sea water, it was necessary to correct the dry weights for salt content, particularly for highly branched organs such as the gonads or the respiratory tree, that always include a large amount of water. We assumed a 36 ppt salinity for this correction.

The corrected dry weight was calculated as:

Dry Weigh Corrected = Dry weight – (Wet Weight – Dry Weight) * 0.036

The Gonado-somatic Index was calculated as $GSI = \frac{\text{Gonad dry weight}}{\text{Total dry weight}}$,

Monthly average surface seawater temperature (SST), air temperature (AT), and rainfall were obtained from Oman's meteorological station in head office in Muscat for both 2009 and 2010.

To preserve the resource, the sampling focused on relatively small samples of large individuals (in order to optimize GSI estimates). It was thus impossible to calculate size at first maturity using the traditional % mature per size class method. Clearly, very small individuals are all immature, whereas all very large individuals were all mature. After sorting all collected animals by increasing total length, the smallest mature individuals as well as the largest immature individuals can easily be determined. By plotting the cumulative proportion of mature individuals in this interval (100% at 19 cm and 0% at 10cm) individuals in this maturation range, the size at 50% maturity corresponds to the median of this cumulative distribution.

3. Results

3.1 Sex-ratio

Over the 20 months of sampling for reproduction investigation, 324 *H. scabra* were dissected, 39% were female, 51% were male and 10% were indeterminate. The observed sex ratio of *H. scabra* at Mahout Bay was 1.3:1.0. A chi-squared test revealed that the relative overabundance of males was significant (χ^2= 8.19, DF = 1; P = 0.004).

3.2 Length-weight relationship

The overall length, measured on the live animals was used as an indicator of length. The power function linking body wall weight and length was highly significant (p< 0.001) with an allometric exponent of 2.70 in females and 2.5 in males (Figure 3).

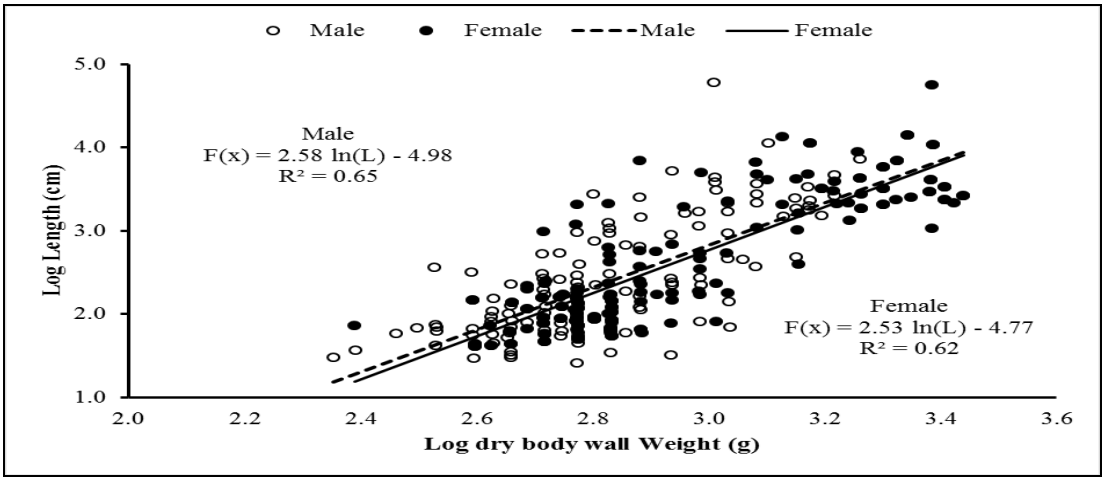

Fig 3: Length-Weight relationships in male and female *Holothuria scabra* collected during the 20 monthly samples near Mahout

Because of the relative imprecision in the length measurements, the coefficients of determination (R^2) for the Length-Weight relationships were relatively low (0.62 in females and 0.65 in males) and the difference in slope of the regressions was found non-significant (Table 1).

Table 1: ANCOVA table for the analysis of length-weight relationships between male and female *H. scabra* near Mahout.

Source	Df	Sums of Squares	Mean Square	F-ratio	Prob
lL	1	78.2359	78.2359	438.61	≤ 0.0001
Gender	1	0.086935	0.086935	0.48738	0.4857
lL*Gender	1	0.071511	0.071511	0.40091	0.5271
Error	288	51.3709	0.178371		
Total	291	136.281			

3.3 Gonado-somatic Index

Gonad growth was very rapid and took place in March and April with maximum GSI observed twice at 12% and 9% on April of 2009 and 2010, respectively (Figure 4).

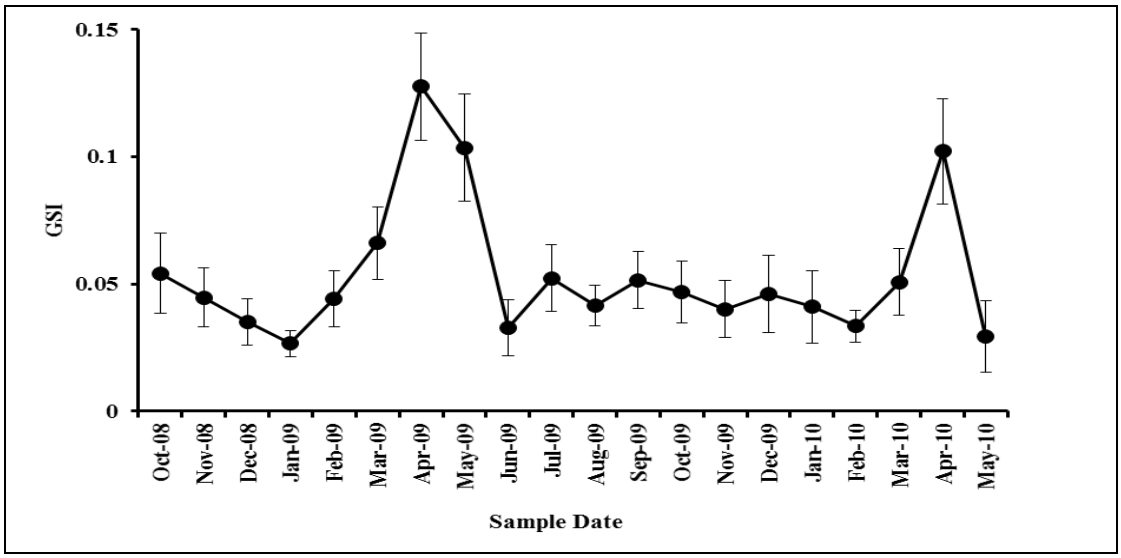

Fig 4: Evolution of the Gonad Somatic Index (GSI) based on dry weight of the gonad and the sum of the dry weight of all body components. Error bars represent variances of the data.

Gonad growth ended abruptly at the end of April by an apparently synchronous spawning event between the end of April and mid of May in 2009 and at the end of April in 2010. At the peak of the GSI, the gonad represented between 9-12 % of the total body mass of the sea cucumber (in dry weight). Before and after spawning, the average GSI was around 4-5 % of the total dry weight. Both in 2009 and 2010 spawning took place just prior to the maximum sea surface temperature 28 ºC (Figure 5) and precipitation 10 mm (Figure 6).

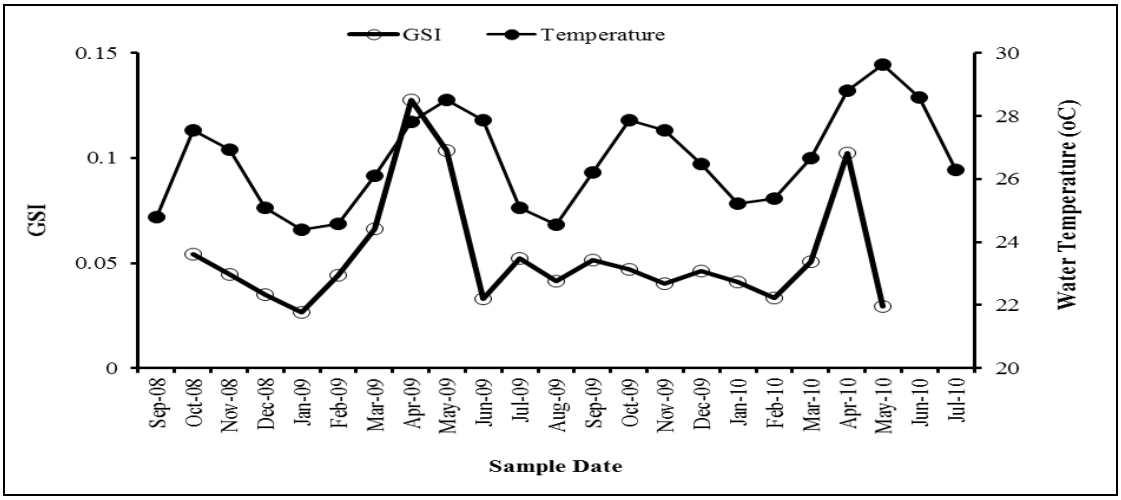

Fig 5: Monthly variation of GSI and Sea Surface Temperature (SST) near Mahout Bay. Monthly SST was obtained from Oman's meteorological station.

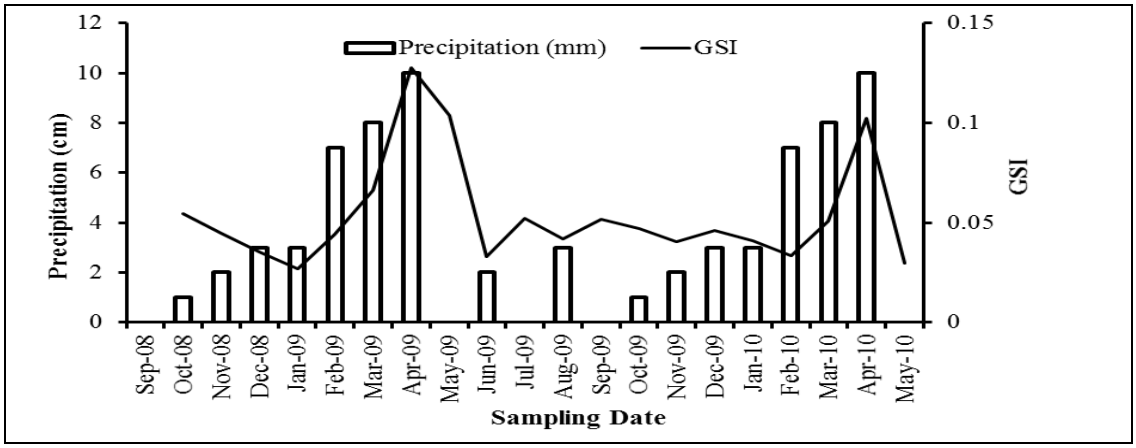

Fig 6: Monthly variation of GSI and monthly average precipitation (P) in Mahout Bay. Monthly precipitation data were obtained from Oman's meteorological station.

3.4 Size at first maturity

We found one 11 cm male individual and one 12 cm female individual with functional mature gonads but most individuals seem to reached maturity around 15 cm in the populations near Mahout. For the whole population, the size at 50% maturity was determined to be 16.5 cm (Figure 7) No specimens larger than 18cm were found immature.

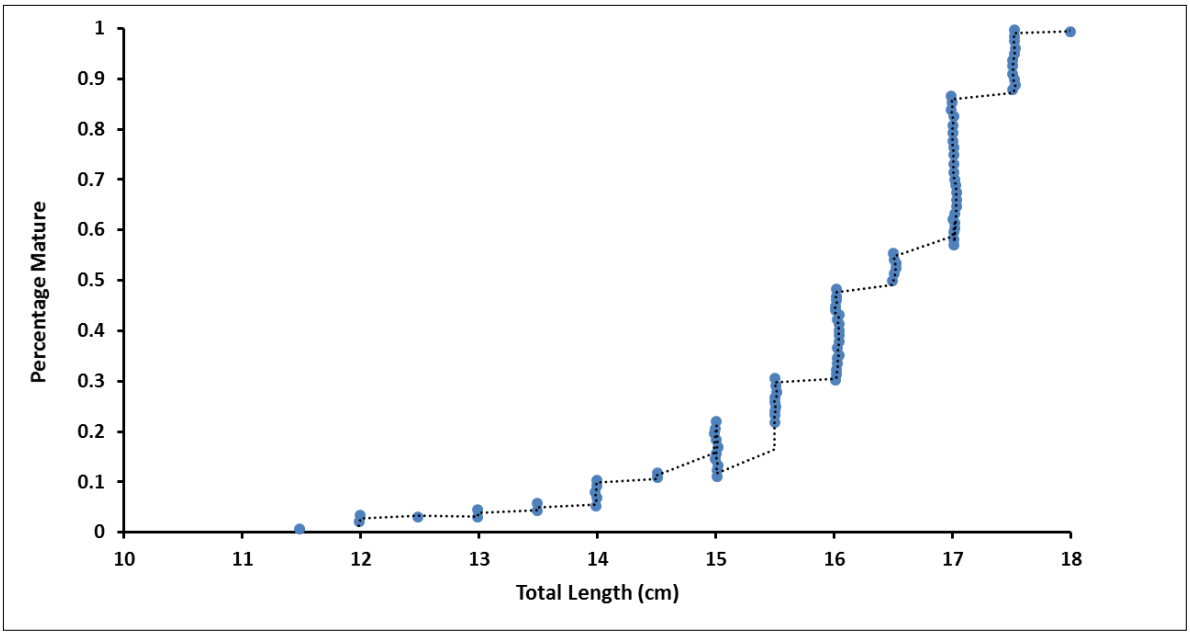

Fig 7: Size at first maturity. The curve represents the relative percentage of the population sexually mature as of function of the length of the animals. The steps result precision at which length measurements were collected (0.5 cm).

4. Discussion

Locally, Mahout Bay also known as Ghubbat Hashish (grass meadow) is one of the largest seagrass habitat in Oman [31] with all three species; *Halodule uninervis*, *Thalassoden dronciliatum* and *Halophila ovalis*, making it one of the marine biodiversity hot spot in the sultanate. It is also an important feeding ground for green turtle, *Chelonia mydas* (Linnaeus, 1758). It is the center of a significant penaeid prawn fishery as well mainly *P. indicus* and *P. semisulcatus* [38] and more recently has also seen the rapid development of *H. scabra* fishery [3]. Although typically classified as sub-tropical or tropical, the marine climate of the Arabian Sea sees large temperature variations, due to the development of a summer monsoon linked upwelling [49]. Because of this unique oceanography, sea water temperature ranges from 19 ºC during the summer southwest monsoon in the southern region of Oman and up to 30 ºC during the fall and late spring.

The scenario of a distinct annual reproductive season support the hypothesis that reproduction of widely distributed tropical marine invertebrates changes from continuous (perhaps monthly) to seasonal with increasing latitude [54]. The situation of Oman corresponds to a "false high latitude" due to the presence of the summer upwelling [49]. The sandfish *H. scabra* is largely distributed between 30°N and 30°S throughout the tropical Indo-West Pacific. The presence of the fishery of this species in the Arabian Sea countries was reported in Oman [3], India [30], Pakistan [29], Yemen [35], Iran [1], Maldives [29] and Somalia [56]. However, the fisheries in these countries are generally not successfully managed, partially due to lack of key ecological and socioeconomic information [17].

In this study, individuals with identifiable sexes were present in 18 monthly samples and only few individuals without gonads were encountered, which support findings in Tanzania and the Solomon and Cook Islands [21, 32, 46]. The sex ratio was not different from one in the 2008–2009 samples, which is also common for sea cucumber species [4, 36, 40, 46, 47, 53], however, the sex ratio shifted to significantly more males in the 2009–2010 sample. Such unbalanced sex ratio has been reported for other species: *Holothuria tubulosa* [51] and *Holothuria whitmaei* [50], suggesting that fishing pressure and high demand and thus overexploitation could have an impact on the sex ratio shifting.

For the size at first maturity, males and females were grouped because towards the end of the study, the small number of individuals collected did not allow the use of separate curves. As for other sea cucumbers, and in the same species elsewhere, the reproductive cycle is most likely to be synchronous [39]**.** Analysis of size showed that around Mahout Bay *H. scabra* attains maturity at the length of 16.5 cmwhich corresponds to approximately 600 g fresh weight which also corresponds to15 to 18 cm which that reported for sexual maturity of *H. scabra* [33]. The present finding is also comparable with those of earlier investigations for instance 16.8 cm reported in Dar-es-Salaam [32] and 16 cm in New

Caledonia [16] and Egypt [26]. The corresponding wet weight reported here is found comparable with Okinawa *H. scabra* population in Japan [37]. There seems to be little variation across this species geographical range; 17.4 cm was reported in Malaysia [4], Lokani [34] in Papua New Guinea reported the size of 14 cm, and 15 cm was reported in Torres Strait [52].

Conand (1993) [16] reported the difficulty in determining intraspecific differences in the reproductive state of sea cucumbers because of the flexible form of this animal. This make the measurement of the gonad index in holothurians difficult because of their soft body (precluding the use of length) and variable water content [25] (precluding the use of wet mass), suggesting that standardized parameters such as body wall, dry and ash-free dry weight would a more accurate estimate of reproductive state [39]. Therefore, all samples used here for GI investigation were based on a dry mass (after correction for salt content).

Reproductive cycle in holothurians can be annual [5, 7, 8, 41, 42, 47], biannual [20, 27] or even continuous pattern which can occur usually in tropical regions [24, 28, 46, 48].

From the gonad index results, it is apparent that the Omani population of *H. scabra* exhibits an annual reproductive season, with a single "spring" spawning towards the end of the inter-monsoon period after the rapid spring warming of the water from 24 ºC in January-February to 28 ºC in early May.The same tendency towards annual reproductive pattern of shrimp *P. indicus* in the area was noted by Mohan and Siddeek (1995) [38]. Similarly, the early growth of seagrasses at the same habitat was reported by Jupp *et al*. (1996) [31] during the same period. Within the framework of this research, a well-marked spent period was seen in September and October during collection of broodstock for spawning induction and fertilization; the gonad tubules regress in length and diameter.

A short period of maturation and spawning appears to take place in Mahout Population during April and May. Similar tendency of short period was found in Moreton Bay population in Australia for the same species [39], although a prolonged period of six months was reported in Toliara population in Madagascar [47]. This pattern could be related to the environmental cues affected the reproduction. The GI peak at 2010 was shown higher (12 %) than (9 %) that in 2009, if this reduction continues in the following sampling years in parallel with no fishery management then this suggest as evidence that the reproduction of *H. scabra* may probably be on the way of collapse. However, further sampling will be required in the future to confirm such tendency. Various environmental factors may influence gametogenesis and time of spawning [25]. In several sites located in tropical or subtropical areas, *H. sabra* seems to show a continuous reproductive cycle [24, 28, 46, 48]. However, in most locations, including near Mahout in Oman, the reproductions follows a clear annual cycle [38]. This is likely the results of the strong seasonal signal impressed on the marine system by the annual monsoon cycle. During the South West monsoon (June - September), the water temperature drop by as much as 5ºC because of a coastal upwelling, whereas during the North-East monsoon (December-March), both a decrease in insolation and evaporative cooling contribute to a second decrease in SST.

The effect of such an intermittent upwelling has been shown to affect population of *Holothuria fuscocinerea* in Mexico [7] and other echinoderm species in the same area [6]. Because the gonad maturation coincides with the spring increase in SST in Mahout, temperature appears to be involved in initiating gonad development in *H. scabra*. Although, precipitation is highly irregular and generally low, average rainfall shows also a maximum during these few months of gonad growth and could possibly affect reproduction through changes in salinity or primary production. This primary production can be used immediately by the meroplankton including the sea cucumber larvae [5, 25, 47] and after sedimentation to juveniles and adult organisms.

5. Conclusion

The present study gives an overview of two the reproductive cycle of a population of *H. scabra* in Western Oman. Size at first maturity is 16.5 cm and spawning coincided both in 2009 and 2010 with the end of the inter-monsoon period (April-May) and the rapid spring warming of the water from 24ºC in January-February to 28ºC in early May.

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