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# Gonadal development and hermaphroditism of bluespotted seabream, *Pagrus caeruleostictus* (Valenciennes, 1830) from the Mediterranean Sea, Egypt

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ABSTRACT

The bluespotted seabream *Pagrus caeruleostictus* represents one of the commercially significant Sparidea fish; however, studies on its reproduction are scarce. Accordingly, histological examination of female and male gonads was carried out for the first time in Egypt with the aim of describing the main gonadal changes during the reproductive cycle. The frequency of the distribution of maturity stages revealed that male and female reproductive cycles are divided into five maturity stages. The GSI and egg diameter data together with the maturity distribution, explained that the *P. caeruleostictus* is a fractional spawner with extended spawning season (June-October). Sex ratio was found to be unbalanced, being in favor of the females (1 male: 1.4 female); both males and females have parallel size distributions, with males being predominant and larger sizes. The size at first maturity ($L_{50}$) was 27 cm and 29 cm total length (TL) for female and male fish, respectively. Absolute fecundity for ripe and spawning females ranged from 84,954 to 362,295 eggs, while the mean relative fecundity was 4970.864 ± 2398 egg/cm and 189 ± 54.38 egg/g to the total length and total weight, respectively. Gonadal histology distinguished rudimentary hermaphroditism for 83% of the young females, with evidence of low proportion of protogynous sex reversal for larger sizes. Results from the current research work provide essential basic information for the future of *P. caeruleostictus* fishery management and aquaculture plans.


## Introduction

Bluespotted seabream (*Pagrus caeruleostictus* Valenciennes, 1830) is included among Sparidae species (Akazaki, 1962; Orrell et al., 2002). It is commonly found in the Eastern Atlantic (Bauchot and Hureau, 1986) and in the majority of the Mediterranean Sea (Bauchot and Hureau, 1986; Fischer et al., 1987) at water depth ranging from 30 m to 200 m (Bauchot and Hureau, 1986; Schneider, 1990). Even though the bluespotted seabream is a highly valuable commercial species, it is an infrequent fish in the Egyptian market. It had not been mentioned or included in the statistical book of the General Authority of Fish Resource Development (GAFRD) in Egypt as there isn't any available data concerning the stock, reproduction and feeding. Only one recent study mentioned the bluespotted Seabream among phylogenetic study on Sparidae species in Egypt (Abbas et al., 2017).

Very little information is known about the reproduction of this species in the Eastern Mediterranean Sea; only one study discussed the reproductive aspects (Chakroun-Marzouk and Kartas, 1987). *P. caeruleostictus*is is a protogynous hermaphrodite (Chakroun-Marzouk and Kartas, 1987) that becomes sexually mature at the age of two years. Spawning time is recorded throughout the hot season (Bauchot and Hureau, 1986). The IUCN Red List reported that possible declines caused by fishing activities may threaten *P. caeruleostictus* (Russell, 2014).

Reproduction is considered the most vital phase in the life cycle of a species, as it determines its survival (Moslemi-aqdam et al., 2016). Teleost fishes display a variety of sexual strategies. Many families are known to have hermaphroditic species, according to Sadovy de Mitcheson and Liu (2008), 27 families display this feature. Sparidae are well recognized along with Teleosts for their diversification and the difficulty of their sexual patterns that in turn complicate the reproductive biology evaluation (Buxton, 1990; Sadovy de Mitcheson and Liu, 2008). In Sparidae, sexual

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patterns can differ even within the same genus, it can vary between functional sequential hermaphroditism or display bisexual gonadal progress during the juvenile phase, but it functionally acts as only one sex rudimentary hermaphroditism (Buxton, 1990).

Understanding the fish reproductive biology is a crucial factor that helps in discovering the species' life history and seasonal reproductive cycle. Additionally, it is critical for the sustainable management of the fish resources, particularly for establishing seasonal or space fishing prohibitions (Oso et al., 2013). Moreover, the identification and categorization of sexual patterns are important for phylogenic correlations (Sadovy and Domeier, 2005; Sadovy de Mitcheson and Liu, 2008). Histology is a potent technique for fish reproductive studies (Blazer, 2002), it is widely used for sex determination, reproductive stage, atresia assessment and hermaphroditism verification and classification (Blazer, 2002; Sadovy de Mitcheson and Liu, 2008).

This work aims to carry out, for the first time, a histological characterization of the maturity stages of *P. caeruleostictus* gonads. This will be done in order to investigate the species' reproductive strategy and spawning pattern, as well as to find out the size of sexual maturity in order to elucidate the reproductive structure of *P. caeruleostictus* populations from the Eastern Mediterranean. The study also targets an in-depth analysis of *P. caeruleostictus* reproductive annual cycle to provide the necessary information for fishery management and aquaculture.

## Material and methods

### Sampling

A total of 298 specimens of bluespotted seabream, *P. caeruleostictus*, was collected monthly from Alexandria landing centers during the period of December 2015 to November 2016. All individuals sampled were sexed, measured to the nearest millimeter (total length, TL) and weighed to the nearest gram (total weight and gutted weight). The maturity stage was determined macroscopically following a maturity key designed for fishery (Nikolsky, 1963; Núñez and Duponchelle, 2009) with some modifications (stages are described in Table 1).

Gonads were cut into smaller pieces and placed in neutral buffered 10% formalin for fecundity assessment and histological examinations. The length at first sexual maturity was considered as the length at which 50% of all individuals were sexually mature (Maturing stage II). The proportion of mature individuals by their size's class was determined, and size at first maturity ($L_{50}$) was derived by fitting a logistic curve to the proportion of sexually mature individuals by length (King, 1995):

$$P = 1/\left(1 + e^{-r(L - L_m 50)}\right)$$

The equation was transformed to: $L_n((1-P)/P) = r_1 L_m - r_2 L$where *P*: is the proportion of sexually mature individuals, $r_1$: is the slope of the curve, $r_2$: is the intercept, *L*: is the total length of the fish in cm. $L_m$: is length at sexual maturity ($\mathbf{L_{50}}$).

$L_{50}$ was estimated by plotting $L_n$ [(l − P)/P] against L then $L_m$ ($L_{50}$) = the intercept divided by the slop.

Gonadosomatic Index (GSI) was calculated using the following formula GSI = gonad Weight/gutted Weight × 100.

### Fecundity and egg diameter

A total of 19 ovaries which were macroscopically identified as ripe and spawning during spawning months, were used to investigate the fecundity and oocyte size distribution of *P. caeruleostictus*. Three sub-samples of eggs were taken from different sites of the ovarian right lobe and weighed to the nearest 0.01 g. Subsequently, oocytes' average number was calculated, as the total number of oocytes (all stages of oocytes) in the fully ripe ovary during the spawning period (Bagenal, 1978). The absolute fecundity was derived according to the equation of Nikolsky (1963): **F = GW/w * X**where **F** is the absolute fecundity, **GW** is the total gonad weight (g), **w** is the mean weight of sub-samples and **X**: the counted number of eggs in the sub-sample. The relative fecundity was estimated as the absolute fecundity according to length and weight on the corresponding length and weight, respectively (Number of eggs per unit gram body weight and cm of the body length) as recommended by Bagenal (1978).

Differences in oocyte sizes were detected by shaking the ovarian sample and pouring three sub-samples into a small Petri dish, oocytes diameter from different stages were measured to the nearest μm using a sensitive ocular micrometer (at 1 μm sensitivity) set in the eye piece of a light microscope. Mean egg diameters were calculated according to Murua et al. (2003) and Jakobsen et al. (2009).

### Histological examination

The fixed gonadal samples were rinsed in ethyl alcohol (70%) for two days prior to dehydration, then cleared followed by paraffin wax embedding and sectioned at 4–8 μm and then stained with hematoxylin and eosin (H&E) and examined microscopically.

## Results

The gonads were elongated and hanging by a dorsal mesentery in the posterior region of the visceral cavity, both lobes of gonad had evenly developed. In immature and maturing females elongated tissues from the abdominal side were observed in the number of investigated samples and considered as a sign of rudimentary hermaphroditism. Of the total samples examined, 130 (43.6%) were females, 92 (30.8%) males, and the sex of the remaining 76 (25.6%) fish could not be macroscopically recognized, as they were very thin and translucent gonads and were recorded as thread. For sex identified individuals, females represented 58%, while males represented 42% with sex ratio of 1: 1.4 (M: F); therefore, the sex ratio skewed towards females. The unidentified sex samples ranged in length from 14.9 to 17.6 cm and in total weight from 100.54 to 282.3 g, while females ranged from 16.9 to 56 cm and 192.77 to 1585 g, the identified males ranged from 22.7 to 59.6 cm and 301.18 to 3200 g.

**Table 1**
Morphological characteristics for different maturity stages for female and male of *P. caeruleostictus*.

| Maturity stage | Morphological characteristics | |
|---|---|---|
| | Female | Male |
| Immature (I) | Ovaries were very thin semi-transparent tubes | Testes were thin transparent but wider than ovaries |
| Maturing (II) | Ovaries as semi-transparent pale orange or yellowish wider tubes with no visible eggs | Testes appear as white thinner tubes |
| Nearly ripe (III) | Ovaries are larger in size and orange in color with visible oocytes as small granules | Testes are larger, deeper with creamy-white colour |
| Ripe and running (IV) | Ovaries reach the maximum diameter with larger oocytes, deep orange color with plentiful veins and arteries | Testes are very large and fragile when handled and sperm can extrude if the testis break |
| Spent (V) | Ovaries' size decrease, reddish in color, and flaccid | Testes are pinkish white in colour, shrunken in size |

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