

Ovarian maturity, egg development, and offspring generation of the deepwater shrimp *Plesionika edwardsii* (Decapoda, Pandalidae) from three isolated populations in the eastern Atlantic

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ABSTARCT

For the first time, ovarian maturity, egg development and brood size were analysed in three isolated populations (the Madeira, Canary and Cape Verde Islands) of *Plesionika edwardsii* (Decapoda, Pandalidae) in the eastern Atlantic. Multiple different colour patterns were observed in the same ovarian maturity stage, which was verified histologically, invalidating the extensive use of ovarian colour as a maturity stage criterion. The physiological length at sexual maturity, based on the maturity of the ovaries, was higher in Madeira (carapace length of 19.73 mm) and decreased to Cape Verdes (16.39 mm). Synchronic ovarian maturation was observed during the embryo incubation process, and ovigerous females bearing embryos in the final stage of development were observed throughout the year. Females are multiple spawners during the reproductive season, after which the reproductive process ends and a rest period begins. The absence of females larger than the size—at-sexual-maturity with ovaries in stage I, the incubation of embryos in the final developmental stages, suggests that the resting period begins with a process that reabsorbs the energy located in the ovaries and that the resting period occurs asynchronously in females in each of the studied populations. A potential relationship was found between brood size and female body size, and the number of eggs increased significantly with female size in the three study areas. The same egg mass colour pattern can correspond with different embryo developmental stages, thus invalidating the use of egg mass colour as a criterion for developmental stage.

INTRODUCTION

Some pandalid species from deep habitats are the main targets of commercial fishing activities, and the deep-water shrimp species of the genus *Plesionika* Bate, 1888 occur and play an important ecological role in the benthic ecosystems of tropical and sub-tropical regions along the continental shelf and slope in the Atlantic and Mediterranean [1]. *Plesionika edwardsii* has a circumtropical distribution (except in the eastern Pacific) and inhabits a variety of bottoms including mud, sand, rocks, and corals at depths between 110 and 680 m but mainly at 250–380 m. In the eastern Atlantic, the species has been found from SW Spain to Angola, including the Macaronesian archipelagos, Cape Verde Islands and entire Mediterranean region [2].

Information about aspects such as ovarian maturation, egg development and brood size are scarce and are lacking for the Atlantic. Egg development plays a major role in the life history traits of invertebrates because the energy contained in the eggs as nutrients must ensure the survival and development of the embryos [3]. In pandalids, brood size is linked to female body size as a consequence of the eggs being stored within the cephalothorax and the average egg size [4]. Furthermore, from a fisheries perspective, knowledge of these characteristics is important for understanding the reproduction of the species, and in the

case of brood size, it is essential for the estimation of reproductive potential and recruitment, one of the main aspects of population sustainability. The goals of this manuscript are to study ovarian maturity and egg development, estimate the brood size of the species and establish a maturity scale for this species in the Atlantic region.

MATERIALS & METHODS

Individuals A total of 320 female *Plesionika edwardsii*, 45 ovigerous and 35 non ovigerous caught each quarter in 2012 around the Madeira (MA), Canary (CIS) and Cape Verde (CVS) archipelagos, were studied in the laboratory. The shrimp were collected using semi-floating shrimp traps deployed at approximately 2.5 m above the sea floor between 100 and 350 m in depth.

Carapace length (CL, from the posterior edge of the eye socket to the mid-dorsal rear edge of the carapace) was measured with a digital calliper to the nearest 0.01 mm, and total weight (TW) was measured to the nearest 0.01 g.

Individual shrimp were dissected, and their maturity stage was macroscopically visually assessed based on the structure, shape, position in the cephalothoracic cavity, and

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colour of the dorsal and ventral faces of the ovaries. The mature ovary visibly extended anteriorly to the orbital margin and occupied most of the dorsal part of the cephalothorax. Ovaries were fixed and preserved in 4% buffered formaldehyde and subsequently processed histologically to verify the macroscopic stage.

The percentage of mature females for each 10-mm CL class was determined, and the size at sexual maturity (the carapace length at which 50% of the ovaries are mature) was determined from the relationship between the percentage of mature ovaries and the carapace size. The number of mature ovaries by carapace size was fitted to a logistic equation by non-linear regression.

Ovigerous females from all size classes were used for the morphological study of the eggs and then to estimate brood size. Pleopods with attached eggs were removed from the females, and egg masses were then placed on a 100-µm mesh, washed and isolated from the pleopods. For each embryo developmental stage, a total of 12-15 females of different size classes were selected to estimate egg size. In each ovigerous female, the sizes of 65-75 randomly selected eggs were measured and then used to calculate the maximum diameter (Md in mm), minimum diameter (md in mm), mean diameter (xd in mm), and aspect ratio (Md/md).

The total number of stage III embryos attached to the pleopods was directly counted in 60 females of all size classes under a binocular microscope. The brood size, defined as the egg production by batch, was estimated by the gravimetric method and described fitting a power function to the number of embryos and the carapace length data

RESULTS & DISCUSSION

Many of the studies that have macroscopically assigned maturity stage to *Plesionika* species used maturity scales based on the size of the gonads and, primarily, the colour pattern of the carapace. In some cases, the same maturity scale has been used for different *Plesionika* species, including *Plesionika edwardsii*, although the ovarian coloration pattern differs among species. The maturity scale based on the morphology of the gonads was histologically confirmed. Multiple different colour patterns were observed in the same ovarian maturity stage, which was verified histologically, invalidating the extensive use of ovarian colour as a maturity stage criterion

The estimation of size at sexual maturity based on ovarian maturation (physiological maturity) instead of ovigerous condition was more reliable because a high fraction of non ovigerous females are mature but can be in the resting period, which can lead to an overestimation. The physiological length at sexual maturity, which was based on the maturity of the ovaries, was estimated to be 19.73 mm CL in MA, 18.56 mm CL in CIS, and 16.39 mm CL in CVS. The histological examination of the subsamples of 100 ovigerous females (larger than the size at sexual maturity) from each archipelago found females with ovaries in the resting stage (18-20%) bearing stage I or II eggs, females with advanced ovaries (32-36%) bearing stage II or III eggs, females with mature ovaries (30-36%)

bearing stage III or IV eggs, and post-spawning females (10-16%) only bearing eggs in stage I. No ovigerous females were observed with eggs in stages III or IV or with ovaries in stage I. In the analysed ovigerous females, all of the maturity stages and all of the egg developmental stages were recorded in the four quarters. The histological examination of the subsample of 100 non-ovigerous females (larger than the size at sexual maturity) found females with ovaries in the resting (12-38%), advanced (30-36%), and mature stages (32-52%) and all of the maturity stages were recorded in the four quarters. Females are multiple spawners during the reproductive season, after which the reproductive process ends and a rest period begins. The absence of females larger than the size-atsexual-maturity with ovaries in stage I, the incubation of embryos in the final developmental stages, suggests that the resting period begins with a process that reabsorbs the energy located in the ovaries and that the resting period occurs asynchronously in females in each of the studied populations.

The number of eggs increased significantly with female size in the three areas. When the sizes of the eggs are compared among the study populations, it can be observed that less energy is invested in reproduction (in terms of number of eggs and size of the eggs) in Cape Verdes than in the other regions. This pattern agrees with the general tendency of the production of larger eggs at higher latitudes. Female body size is the principal factor determining brood size in decapods and the reduction in the brood size and the higher maximum female body size from Madeira to Cape Verdes supports this relationship.

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