Reproduction and growth in captivity of the polychaete \textit{Nereis diversicolor} O. F. Müller, 1776, using two different kinds of sediment: Preliminary assays

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\textbf{ABSTRACT}

The polychaetes used as bait for sport and commercial fishing are a natural resource exploited in a large number of estuaries. One of the most common species captured is the ragworm \textit{Nereis diversicolor} O. F. Müller (1776). This species is also used in aquaculture, mainly because of its easy availability to local dealers, and plays an important role as a nutrient stimulating gonad maturation and spawning in hatcheries.

The high economic value of these worms, and the size of their catch in the main Portuguese estuaries, have led to their intense exploitation as commercial bait.

Accordingly, a small-scale reproduction study of this species was carried out under controlled conditions (temperature, salinity and luminosity) in two different sediments (sand and mud), following its development from the larval to the adult stages. Such elucidation of certain aspects of its life cycle is instrumental for the species’ future culture in artificial systems. The present study represents the starting point of large-scale production that will reduce the anthropogenic disturbance to biological estuary communities. The main objective of this study was to obtain preliminary results that would enable us to understand the period of growth necessary to reach a commercially valuable size. Such growth was rapid in both types of sediment, taking only a few months.

\textbf{Key words:} \textit{Nereis diversicolor}, reproduction, growth, sediments, culture.

\textbf{RESUMEN}

Reproducción y crecimiento en cautiverio del poliqueto \textit{Nereis diversicolor} O. F. Müller, 1776 en dos tipos de substrato: ensayos preliminares

Los gusanos de mar utilizados como cebo, tanto en la pesca profesional como en la deportiva, constituyen un recurso que es explotado en un gran número de estuarios. Uno de los gusanos más usados con esta finalidad es el nereido \textit{Nereis diversicolor} O. F. Müller, 1776. Además de ser normalmente utilizada como cebo fresco, es una especie cada vez más empleada en acuicultura debido, por un lado, a la facilidad de su obtención en el mercado y, por otro, al importante papel que parece desempeñar en la maduración de las gónadas e inducción de las puestas en especies cultivadas.

El elevado valor económico asociado a la captura de estos nereidos en los principales estuarios portugueses ha provocado la intensificación de su extracción con fines comerciales.

En este ámbito se inició el estudio de la reproducción de esta especie a pequeña escala bajo condiciones controladas (temperatura, salinidad y luminosidad) en dos tipos de substrato (arena y fango). Se estudió el desarrollo de la especie desde su fase larvaria hasta su estadio adulto, con la finalidad de conocer aquellos as-
INTRODUCTION

The ragworm *Nereis diversicolor* O. F. Müller (1776) is one of the species belonging to the Nereididae family that is used as commercial bait. There are several families (Arenicolidae, Eunicidae, Glyceridae, and Nephtyidae) exploited worldwide in sport and commercial fishing. The quest for these species is increasing rapidly, mostly due to their important role as a nutrient stimulating gonad maturation and spawning in hatchery-reared species, e.g. *Solea vulgaris* (Quensel, 1806), *Solea senegalensis* (Dinis, 1986), *Penaeus kerathurus* (Luis, 1989) and *Penaeus vannamei* (Lytle, Lytle and Ogle, 1990).

These features, along with artificial fertilisation (Ozoh and Jones, 1990), apparently made these species the easiest ones to reproduce in captivity. In addition, the absence of a planktonic larval stage made the culture process cheaper.

The other main point of the culture and commercial use of this bait is to reduce the substrate harvesting disturbance, and the great biogeochemical and benthic community impact (Gambi et al., 1994).

With the purpose of elucidating certain aspects of *N. diversicolor's* life cycle, especially the sediment type suitable to its culture, a small-scale experiment was conducted under controlled conditions of temperature, salinity and light using two different kinds of sediments (sand and mud). In this assay, the development was followed from the larval to adult stages.

Experiments by Dean and Mazurkiewicz (1975) on polychaetes with benthic development, such as *N. diversicolor*, showed that some species may develop simply without sediment, while others prefer the same sediment used by adults. Some larvae stop growing and die when the type of sediment changes, showing an extreme sensitivity to this parameter. The elucidation of this and other questions regarding to culture of this species will make it possible, in the near future, to develop large-scale artificial systems, like those already being developed with other species in the UK and the Netherlands (Olive, 1994). Large-scale culture provides not only an increasing market of live bait, but will also decrease the biological impact on estuarine communities, with lower environmental effects.

MATERIALS AND METHODS

Aquaria (38 × 45 × 45 cm³) with constant aeration were mounted, with a 15 cm sediment layer (sand and mud) from the estuary-lagoon systems of the southwest coast of Portugal (Aljezur and Carrapateira), locations where this species develops freely. Two different kinds of sediments were used (figure 1): medium sand with strong focus on classes 1 through 2 PHI (500-250 µm) proceeding from Carrapateira lagoon, and mud with high concentrations of silt and clay, but also mixed with some fine and very fine sand (3 and 4 PHI 125 and 63 µm) from the Aljezur estuary. The sediments of these aquaria were previously dried out in a heater (at 90 °C during 24 h) to eliminate other animals...

Figure 1. Grain-size distribution (cumulative curves) of the sediments (sand and mud)
(crustaceans, molluscs and annelids) and possible contamination that could interfere with the experiment. Due to the difficulty in establishing the sex of mature animals (Dales, 1950), 10 N. diversicolor adults (around 10 cm long) bought from a commercial dealer were transferred to each aquarium. Since it was impossible to establish gender, this number was used in order to guarantee a male/female presence in each aquarium.

During the experiment, water salinity was kept between 14 and 17 ‰, values considered ideal for this species’s reproduction (Bartels-Hardege and Zeeck, 1990) and the temperature was 20 ± 2 °C (Duchon, 1971). To avoid the accumulation of catabolism products and consequent deterioration of the water quality, new water was added every week. The photoperiod was adjusted to a long photophase (16 h of daily light, LD 16:8) and a 100-lux intensity was supplied by a fluorescent light during the first month. Later on, a second lamp was installed that increased the luminosity to 175 lux.

Initially, each individual was fed every three days with commonly available commercial food used for tropical fish (Tetra Menu). This food type had been used successfully with this species (Garwood and Olive, 1981; Bartels-Hardege and Zeeck, 1990). After 22 days, they were fed daily, because some cannibalism was detected. To establish the beginning of the reproduction period, sediments sampled next to the gallery holes of the ‘reproducers’ were collected, since Bartels-Hardege and Zeeck (1990) stated that the eggs are fertilised there. Six individuals were collected 3 times a week from each aquarium and anaesthetised with MS-222, an anaesthetic commonly used on cold-blooded animals. Later they were measured (total length, from the antenna base to the anal cirrus insertion) using a binocular microscope and, when needed, a microscope. After measurements, the worms were stored in a plastic bag filled with nitrogen and lyophilised, being weighed afterwards to calculate biomass. The linear growth rate (mm/day) was calculated using the difference between the final average length and the initial one, divided by the total number of days of the experiment. The number of setae was recorded for each individual studied.

RESULTS

The first larval stages were not detected, which might be due to the fact that larvae stay inside the gallery tubes protected by the females and attached to the mucus. Dispersion usually happens after the female’s death (Bartels-Hardege and Zeeck, 1990), the larvae becoming more active when the tenth setae is fully developed (Clay, 1967). However, based on the number of setae of the first individuals found (8 parapodia and setae) the conclusion is that the hatching might have occurred about a month after the beginning of the experiment. The first eggs were observed in the samples taken after 4 weeks, so this was considered day zero, the beginning of the culture. The results (linear growth, weight growth and the increase in setae) are shown in figures 2 and 3; the linear growth rate obtained for both sand and mud are, respectively, 0.97 and 0.84 mm/day. Commercial size, according to observation at the local market, was obtained in 64 days (sand) and 62 days (mud). After 72 days, individuals were attained with a length of 7 cm (70 % of the initial reproducers’ size).

DISCUSSION

The high rate of growth observed in this experiment (18 setae in 14 days, compared with Dales’ (1950) result of individuals with 3 setae in 2 weeks) might be related to the fact that the culture temperature used was double the average one used previously. According to Neuhoff (1979), only at temperatures close to 15 °C does N. diversicolor present a high growth rate. Olive et al. (1991), reporting on Nereis virens (Sars, 1835) culture, found that the growth rate is a complex function between the temperature and culture density.

Specimens showed a steady development, measured in terms of setigerous increase (lower standard deviation) in both sediments, as opposed to the one obtained with the linear or weight growth, where the deviation values are higher (figures 2 and 3). This reflects a morphological difference between individuals of the same age. This could indicate a problem for the commercial culture of N. diversicolor, which could be overcome by selecting the reproduction stock among those individuals with higher growth. The observed linear growth is very similar using both sediments, although the linear growth rate was higher in sand. There was also a clear gradual increase of total length, and consequently, of the biomass. Its standard deviation also increases, which explains what was mentioned pre-
Previously about differences at an individual level. Bridges et al. (1996), working with Nereis (Neanthes) arenaceodentata (Moore, 1903), noted these differences with the start of competition for space and food, and also when the number of worms per square metre is still low, reflecting a positive influence on growth. When the density is high, growth declines. This increase (linear and weight growth) of some individuals becomes more and more evident over time, since the larger worms use their size for both food and territory control. When comparing the biomass graph with the one of setae numbers, in the last four days of culture, it is possible to infer that there is a strong weight difference among individuals, which cannot be verified if we compare it with the setae numbers, which are almost the same (figures 2 and 3).

It is noteworthy that there were 10 N. diversicolor in each aquarium, due to the lack of gender determination. Since it was impossible to previously determine the total number of males and females, consequently the number females that spawned was also unknowable.

Analysing the biological parameters (total length, biomass and number of setae) obtained using both substrates, and taking into account that the observation, handling and sampling of worms were easier in the aquaria using sand as substrate I suggest its use in future experiments.

Although preliminary, these results, combined with the high commercial value of this species, explain the importance that this resource may have in aquaculture, to both commercial trade (fishermen) and aquaculture professionals.
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