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Larval development and post-larval growth of *Branchiomma bairdi* (Annelida: Sabellidae) from a Mediterranean population

Michela Del Pasqua,^a Marco Lezzi, Margherita Licciano, and Adriana Giangrande

Department of Biological and Environmental Sciences and Technologies (DiSTeBA), University of Salento, CoNISMa Unit, 73100 Lecce, Italy

Abstract. Branchiomma bairdi is a Caribbean fan worm introduced in several localities worldwide, including the Mediterranean Sea, where the species' range has rapidly expanded. Reproduction in *B. bairdi* was previously investigated in both extra-Mediterranean and Mediterranean areas, but no information is available on larval development and post-larval growth. In the present article, we examined these features for a population from the Mar Grande of Taranto (Ionian Sea). The species is hermaphrodite, and fertilization occurs *in situ*. Mucus seems to play an important role in fertilization, and also in preserving eggs before fertilization. The trochophore stage develops within the mucus and after hatching, larvae swim for about 3 d before settlement. The trochophore showed a distinct prototroch and two red dorsolateral larval eyes. The pelagic stage takes only 96 h even though prototroch is maintained after settlement, disappearing at 5 d, when larvae showed three chaetigers and branchial crown consisted of four radioles. Some interesting observations concerning changes in the morphology of chaetae and in the number of uncini during growth are also reported, together with discussion of the development of stylodes, an important diagnostic feature in *Branchiomma* species identification.

Additional key words: alien species, Branchiomma, reproduction, Sabellida, spermatozoa

Within Annelida, the members of the family Sabellidae are one of the most widely studied groups in terms of modes of reproduction and development (McEuen et al. 1983; Rouse & Fitzhugh 1994; Bick 1996; Giangrande 1997; Hsieh 1997; Rouse & Gambi 1998a,b; Gambi et al. 2000, 2001; Giangrande et al. 2000). Sabellids show a large diversity of reproductive strategies including broadcasting of gametes, deposition of egg masses, and brooding outside or inside the parental tube (McEuen et al. 1983; Knight-Jones & Bowden 1984; Rouse 2001). However, they all have lecithotrophic development, with varying length of pelagic life, from 15 d as in Sabella spallanzanii (GMELIN 1791) (Giangrande et al. 2000), to a few hours as in Branchiomma luctuosum GRUBE 1869 (Licciano et al. 2002). Direct development with brooding can also be present as in Perkinsiana littoralis (HARTMAN 1967) (Gambi et al. 2000). Reproductive mode is significantly correlated with body size, with small species that are brooders and large species that are free-spawning (Rouse &

Fitzhugh 1994). According to Rouse & Jamieson (1987), sperm morphology is often associated with reproductive strategy, with ect-aquasperm found in broadcasting species with pelagic development, and ent-aquasperm generally observed in intratubular brooders. However, the correlation between the sperm morphology and fertilization type still needs to be clarified (Rouse 1999). Moreover, in this family, the morphology of the acrosome is highly variable and indicative of phylogenetic relationships (Patti et al. 2003).

Branchiomma bairdi (MCINTOSH 1885) is a sabellid species originally described from Bermuda and recently introduced in several localities, where the species range has rapidly expanded as a consequence of the high capacity to colonize different substrates, especially in confined environments and impacted areas, and as a consequence of the occurrence of both sexual and asexual reproductive strategies (Tovar-Hernández & Yánez-Rivera 2012; Arias et al. 2013; Giangrande et al. 2014; Keppel et al. 2015). Notwithstanding its recent introduction in the Mediterranean Sea, this species has reached high densities especially in the Gulf of Taranto, a

^aAuthor for correspondence.

E-mail: michela.delpasqua@unisalento.it

eutrophic area of the Ionian Sea (Italy), where the species has been deeply investigated since 2012 (Giangrande et al. 2014; Lezzi et al. 2014, 2016; Stabili et al. 2014; Lezzi 2016). Two years of study on its population dynamics revealed that this species is an annual form, reproducing from July to October, and with an early maturity that can occur just 3 months after settlement (Lezzi 2016; Lezzi et al. 2016).

The reproductive biology of *B. bairdi* has been studied by Tovar-Hernández et al. (2009a, 2011), Tovar-Hernández & Yánez-Rivera (2012), and Arias et al. (2013) in a Pacific and in a Mediterranean population. These authors reported the presence of simultaneous hermaphroditism, intratubular fertilization, and brooding of eggs and embryos inside the parental tube attached to the body wall by means of mucus. At present, however, no data concerning the larval and post-larval growth are available.

As an understanding of reproductive strategies is useful to understand the invasiveness of alien species, the present study is focused on reproduction under laboratory conditions of Mediterranean specimens of *B. bairdi* collected from Mar Grande of Taranto, with the purpose of completing the previous observations on the reproductive features of this sabellid. Furthermore, it provides the first description of larval development, settlement, and post-larval growth in this species.

Methods

Specimens of *Branchiomma bairdi* were collected by SCUBA diving at 5 m depth along a dock wall in the "Mar Grande" of Taranto (Gulf of Taranto, Ionian Sea, Italy) in October 2015. We sampled individuals from naturally occurring large aggregations. Soon after collection, the worms were transferred to the laboratory and kept in an aquarium placed in an environmental chamber set at a temperature of 20°C, similar to that recorded at the sampling site.

In order to determine the presence or absence of gametes in the specimens, worms were removed from their tubes and a small quantity of coelomic fluid (several microliters) was withdrawn from the body wall at the abdominal level using an analytical microsyringe and examined under the microscope. The analysis of the coelom content enabled the separation of individuals whose coelomic cavity was found full of mature gametes from those devoid of germinal products. Egg diameter was measured with an ocular micrometer in 20 specimens (20 eggs for each specimen). In order to stimulate the spawning of gametes and fertilization, ripe specimens were removed from their tubes and manipulated by applying gentle pressure with finger tips. After this treatment, worms were housed in five beakers (four individuals per beaker) placed in a temperature-controlled room at 20°C. Each beaker contained 500 mL of constantly aerated seawater which was changed daily. At each water change, liquid food for filter-feeding marine invertebrates (SERA Coraliquid, SER-Germany, Heinsberg, Germany) was added at the concentration recommended by the manufacturer (25 μ L L⁻¹).

After worms had spawned in the experimental beakers, fertilized eggs were maintained in the original jars with adults as well as the other developmental stages, as eggs collected and transferred to beakers containing freshly filtered seawater collapsed. Subsequent stages of larval development were maintained in the same experimental conditions. Juveniles were fed by adding a cultured microalga (*Isochrysis galbana* PARKE 1949) to the 0.22-µm pre-filtered seawater at a concentration of ~2000 cells mL⁻¹. The algae were maintained in nutritive medium containing 0.22-µm filtered seawater enriched with the F/2 medium of Guillard & Ryther (1962). Algae were used during their exponential phase of growth.

Larval stages and post-larval growth were monitored daily using a stereomicroscope equipped with a digital camera (Carl Zeiss AxioCam ERc 5s, Zen 2012, Carl Zeiss Microscopy GmbH, Jena, Germany). Juveniles (*n*=20) were also fixed in a solution of 4% formaldehyde in seawater to allow further observations and measurements, such as body length, number of chaetigers, number of uncini for each torus, length and width of chaetigers 2 and 10, and number of radioles and stylodes. The same observations were also carried out on adults. Regression and Pearson's correlation analyses were performed on the dataset of both juveniles and adults to assess the relationships among the biometrical measurements taken.

Results

Fertilization and larval development

All the examined worms were found to be hermaphrodites. Unfertilized eggs drawn from the coelomic cavity were $150\pm63 \ \mu\text{m}$ (mean \pm SD; Fig. 1A) in diameter. After worms spawned, spermatozoa were found in the water column, while eggs embedded in mucus sank to the bottom of the jars. Worms removed from their tubes had bodies covered by a thick layer of mucus. When analyzed



Fig. 1. Gametes and early developmental stages of *Branchiomma bairdi*. A. Unfertilized egg taken from the coelomic cavity. B. Fertilized egg showing fertilization envelope. C. Mature spermatozoa with ect-aquasperm morphology. D. Developing embryos. E. Blastula stage. F. Early trochophore still within fertilization envelope.

under the microscope, the mucus was found to contain fertilized eggs with spermatozoa inside. Eggs were brownish in color, appeared enclosed in a fertilization envelope (100% fertilized), and measured a mean diameter of $250\pm17.74 \ \mu m$ (Fig. 1B). Spermatozoa were ect-aquasperm type, with long flagella and spherical heads (Fig. 1C). Larvae developed only from fertilized eggs embedded in the mucus and maintained in the original jars with the adults (Fig. 1D).

Blastula stage could be detected at 1 h from fertilization (Fig. 1E). After 4 h, trochophore larvae were observed within the egg envelope and still embedded within the mucus (Fig. 1F). At 9 h, trochophores could freely swim and after 24 h, they elongated, exhibiting a mass of brownish material (yolk) in the middle of the body. At this stage, the larvae, which measured about 150±12.8 µm in length, showed a distinct prototroch, which separated the rounded episphere from the conical hyposphere, and two red dorsolateral larval eyes placed between the prototroch and anterior end (Fig. 2A); apical tuft and other ciliary bands were not detectable. At 48 h, body segmentation was observed as the formation of fine capillary chaetae protruding laterally from the first chaetiger. At 72 h, the larvae increased in size, the hyposphere elongated, and the prostomium became more prominent. At this stage, larvae measured a maximum length of ~200 µm, showed two

chaetigers, each with narrowly hooded chaetae (Fig. 2B.C), and started to swim near the bottom of the culture container to search for settlement sites. Although cilia of the prototroch continued to beat. settlement occurred at 96 h, and when the larvae stuck to the substratum, the anus opened, and they began to feed (Fig. 2D). At this stage, the prostomium showed a pair of extensions representing precursors of the branchial filaments and two small lobes appeared anteriorly on each side of the prostomium, probably representing the rudiments of the collar (Fig. 2E). Larval eyes were still present and the mass of yolk reduced in size. Three-chaetiger larvae, measuring 250±14.7 µm, were detected at 5 d when the disappearance of the prototroch could also be noticed. At this developmental stage, the branchial crown consisted of four radioles with actively beating cilia (Fig. 2F). Because there was 100% mortality at the three-chaetiger larval stage, observations of development in the experimental beakers were terminated at this stage. Post-larval growth, therefore, was described from juveniles already settled in the aquarium, where worms had spawned spontaneously.

Post-larval growth

After ~1 month from collection in the natural environment, juveniles were found attached to the

Fig. 2. Development in larvae of *Branchiomma bairdi*. **A.** Trochophore larva at 24 h after fertilization. **B.** Two-chaetiger larva (72 h). **C.** Detail of narrowly hooded chaetae (arrows). **D.** Recently settled larva (96 h). **E.** Detail of the prostomium showing rudiments of the collar and of the branchial filaments (arrows). **F.** Three-chaetiger larva (5 d).

aquarium wall where adults were maintained. These small worms measured 0.8 ± 0.06 mm in total length, had 12 segments, 3 pairs of radioles, 2 elongate dorsal lips, dorsolateral eye spots between the first chaetiger and the branchial crown, and interramal red spots already present throughout the length of the body, which ended with a bilobed pygidium, while ventral lips were not detected (Fig. 3A). At this stage, no finger-like projections (stylodes) were present. Thorax and abdomen were clearly defined by the chaetal inversion and the shift of the position of the fecal groove. Indeed, at this stage of development, juveniles had eight thoracic chaetigers with dorsal chaetae and ventral uncini, and four abdominal segments, of which three showed ventral chaetae and dorsal uncini (Fig. 3A). Thoracic and abdominal uncini appeared avicular with short handle, developed breast, and three rows of teeth above the main fang (Fig. 3B); avicular abdominal uncini were similar to the thoracic ones. Both superior and inferior thoracic notochaetae were paleate (Fig. 3C), while abdominal neurochaetae were broadly hooded (Fig. 3D).

At 42 d, juveniles were 1.4 ± 0.2 mm in total body length with 20 chaetigers in total. The branchial crown had five pairs of radioles with eight pinnules, four pairs of compound eyes, and a pair of stylodes at the base of the radioles (Fig. 4A). At 50 d, the reached total body length juveniles а of 2.46±0.7 mm and had 25 chaetigers (Fig. 5A). At this developmental stage, the branchial crown was formed by six pairs of radioles still bearing four pairs of compound eyes but having four pairs of developing stylodes, among which the basal one appeared longest (Fig. 4B). At this stage, the superior notochaetae appeared slender and narrowly hooded (Fig. 5B), while the inferior notochaetae were broadly hooded with knee up to twice as wide as shaft (Fig. 5C). Abdominal chaetigers exhibited only a few broadly hooded neurochaetae (Fig. 5D). After about 2 months, worms had 40 chaetigers and measured 5.35 ± 0.3 mm in total body length. At this stage, the branchial crown had from 10 to 13 pairs of radioles, each with five pairs of compound eyes and five pairs of stylodes of similar size (Fig. 4C). Macrostylodes, one pair on each radiole, appeared



Fig. 3. Juvenile of *Branchiomma bairdi* at 12-chaetiger stage. A. Entire individual. B. Avicular thoracic uncini. C. Thoracic notochaetae. D. Abdominal neurochaetae.

after worms reached 50 chaetigers (8 ± 0.5 mm in total body length) and the radioles had completely formed, with 11 pairs of compound eyes and 11 pairs of stylodes (Fig. 4D). Starting from the base of each radiole, the following formula is recognizable: five pairs of microstylodes, one pair of macrostylodes, and four pairs of microstylodes. At 75 d (60 chaetigers, and 22 ± 1.1 mm in total body length), the adult morphology was almost complete with two pairs of microstylodes. Observations on juvenile growth stopped after about 3 months when the worms reached 28 ± 1.09 mm in total body length and had 70 chaetigers. At this stage, the peristomial eye spots disappeared.

The relationship between body length and number of chaetigers was logarithmic and the correlation between these variables was significant (r=0.77, p<0.05). The number of chaetigers reached a plateau at about 70 chaetigers when specimens were about 28 mm in total body length, after which they continued to grow without adding other segments (Fig. 6A). There was a linear relationship between body length and the width of the second and tenth chaetigers, respectively, with significantly high correlation values (r=0.96, p<0.05; r=0.95, p<0.05)



Fig. 4. Stylode development along the radiole in *Branchiomma bairdi*. **A.** 42 d after fertilization. **B.** 50 d after fertilization. **C.** 60 d after fertilization. **D.** 70 d after fertilization.



Fig. 5. Juvenile of *Branchiomma bairdi* 25-chaetiger stage. A. Entire individual **B.** Superior thoracic notochaetae. C. Inferior thoracic notochaetae. **D.** Abdominal neurochaetae.

(Fig. 6B). The number of thoracic and abdominal uncini increased exponentially with growth (Fig. 7). The first chaetiger (collar segment) developed only chaetae; the second chaetiger had 20 uncini when the worm had 45 segments, and reached a final number of about 90 uncini when the number of body segments increased to about 70. The number of uncini also decreased from the anterior-most chaetigers (thoracic) to the posterior end of the body (abdominal). A linear relationship was



Fig. 6. Relationships among biometrical measurements in *Branchiomma bairdi*. **A.** Body length–number of chaetigers relationship. **B.** Body length and width of the second and tenth chaetiger relationships (W, chaetiger width).



Fig. 7. Relationships between the number of chaetigers and the number of uncini per torus. T, thoracic chaetiger; A, abdominal chaetiger; Ai, abdominal chaetigers from the intermediate body region; Ap, abdominal chaetigers from the posterior body region.

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Fig. 8. Relationships between the number of uncini per torus and body width.

estimated between the number of uncini and the width of chaetigers 2 and 10 (Fig. 8), with a maximum of 90 per torus at the thorax and 30 at the abdomen at \sim 4 mm width.

Discussion

In accordance with previous observations by Tovar-Hernández et al. (2011) and Tovar-Hernández & Yánez-Rivera (2012) of an eastern Pacific population of *Branchiomma bairdi*, Mediterranean specimens from the Ionian Sea were observed to be simultaneous hermaphrodites, as also reported by Licciano et al. (2002) for the co-occurring congener *B. luctuosum*.

Tovar-Hernández et al. (2011) also reported intratubular fertilization, with brooding of eggs and embryos attached to the body wall by mucus inside the parental tube, although how individuals fertilize the eggs is still unknown. Based on our observations, the body of the worms removed from their tubes was covered by a thick layer of mucus containing fertilized eggs and spermatozoa. This suggests that in B. bairdi, fertilization does not occur in the water column, but instead takes place in the parental tube and probably within the mucus, as occurs in Sabella spallanzanii (Giangrande et al. 2000). We found that fertilized eggs continued their development only when they remained with adults, which continuously produced mucus, but not when transferred into other tanks containing freshly filtered seawater. Therefore, the presence of mucus seems to play a role in protecting eggs of B. bairdi. Other authors have noted that eggs of S. spallanzanii are also released and fertilized in mucus (Stabili et al. 2009), and in this species, all attempts to achieve in vitro fertilization failed when eggs and sperms were combined in seawater. Besides the importance of mucus for egg fertilization and development, however, it is worth mentioning that the presence of adults in the jar where embryos and larvae developed might also have favored larval survival and settlement. As suggested by several studies on reproductive biology and larval settlement behavior in gregarious sabellids (Scheltema et al. 1981; Jensen & Morse 1984; Pawlik 1988; Toonen & Pawlik 1996, 2001; Chan & Walker 1998; Murray et al. 2011), chemical cues released by conspecific adults are strongly involved in larval settlement and metamorphosis and are key factors triggering gregarious settlement in polychaete larvae (Callaway 2003). Moreover, gregarious settlement behavior by larvae often leads to the formation of conspecific adult aggregations (Burke 1986; Pawlik 1986; Amieva et al. 1987; Murray et al. 2011). Accordingly, in the Gulf of Taranto, B. bairdi is found in aggregations formed by 5-30 individuals including both juveniles and adults.

The described strategy of fertilization within the tubes of adults in B. bairdi cannot be inferred from the morphology of the spermatozoa, which have a spherical nucleus, rounded acrosome, four mitochondria, a long flagellum (Tovar-Hernández et al. 2009a,b), and correspond to the ect-aquasperm type of Jamieson & Rouse (1989). It has been hypothesized that in B. bairdi, spermatozoa could be released in the water column and then filtered by conspecifics. However, self-fertilization cannot be excluded (Tovar-Hernández et al. 2011). The ect-aquasperm type is often associated with external fertilization (Rouse & Fitzhugh 1994), and was also described for B. luctuosum (Licciano et al. 2002), Branchiomma lucullanum (DELLE CHIAJE 1828) (Dragesco-Kernéis 1980), Branchiomma bombyx (DALYELL 1853) (Rouse 1999), and Branchiomma nigromaculatum (BAIRD 1865) (Berrill 1977). Among them, only members of B. luctuosum and B. nigromaculatum are known to be free spawners with external fertilization (Berrill 1977; Licciano et al. 2002), while members of B. lucullanum, and probably also B. bombyx, are extratubular brooders (Dragesco-Kernéis 1980; Rouse 1999). Therefore, the ect-aquasperm morphology is characteristic in all investigated Branchiomma species, even in those with different fertilization strategies, thus supporting the suggestion by Rouse (2005) that the structure of sperm alone cannot be used to infer the fertilization biology of a taxon. The presence of ect-aquasperm type could be explained by the presence in the genus Branchiomma of simultaneous hermaphroditism and the absence of specialized organs for sperm storage.

It is well known that reproductive mode is correlated with worm body size, with small-bodied species tending to be intratubular brooders with direct development, whereas large worms are broadcast spawners with direct development (Schroeder & Hermans 1975; McEuen et al. 1983; Rouse & Fitzhugh 1994; Giangrande 1997). This trend is present within Sabellidae as well, even though they are constrained in having lecithotrophic development. Within the genus Branchiomma, individuals of B. luctuosum and B. nigromaculatum are large-bodied broadcast spawners with external fertilization and pelagic larval phase (Berrill 1977; Licciano et al. 2002), while individuals of B. lucullanum are smallbodied extratubular brooders, with developing larvae held in a jelly mass around the tube opening (Dragesco-Kernéis 1980). In comparison to the species mentioned above, B. bairdi can be defined as an intermediate- or large-sized sabellid, and it is reported as an intratubular brooder by Tovar-Hernández et al. (2011) and Arias et al. (2013). Therefore, the reproductive strategy in B. bairdi seems not to be correlated with size. We removed worms from their tubes for experimental observations and did not observe intratubular brooding, but it must be stressed that only the early stages of development remained embedded in the mucus, and the subsequent development included a very short pelagic larval phase (3 d). Moreover, Tovar-Hernández et al. (2011) and Arias et al. (2013) also reported that the earlier stages of development (until metatrochophora) were brooded and glued to the body segments by means of mucus, although they do not provide data on subsequent developmental stages. As suggested by Tovar-Hernández et al. (2011), brooding could have some advantages when compared to free-spawning, such as physical protection from a number of benthic predators and stable chemical environment created by the mucus. Thus, brooding in B. bairdi is devoted to the physical protection of the early developmental stages without excluding the presence of a dispersive stage, similar to B. luctuosum (Licciano et al. 2002).

Based on our results, the mature eggs from individuals of *B. bairdi* from the Mediterranean had a mean diameter (150 μ m) greater than eggs recorded from both Pacific and Maltese populations (110 μ m) (Tovar-Hernández et al. 2011; Arias et al. 2013). As previous available information on *B. bairdi* deals only with the earlier larval stages until metatrochophora, no comparison of our developmental data with those from Pacific and Maltese populations is possible. Among other sabellids, larvae of *B. bairdi* and *B. luctuosum* have the shortest pelagic period, as a pelagic period of 15 d has been reported for *S. spallanzanii*, 9 d for *Megalomma vesiculosum* (MONTAGU 1815) (Wilson 1936), 7 d for *Myxicola infundibulum* (RENIER, in Meneghini 1847) (Dean et al. 1987), and 6 d for *B. nigromaculatum* (Berrill 1977).

Some taxonomic considerations can be inferred from post-larval growth observations. The examination of the thoracic chaetae in juveniles allowed to describe a different morphology from the adult ones. This is of particular interest and corroborates the transformation series of chaetal forms hypothesized by Fitzhugh (1989) in his study on sabellid phylogeny. According to this author, the broadly hooded inferior thoracic notochaetae present in the Branchiomma clade appear as the apomorphic condition derived from the paleate chatae. The presence of thoracic paleate notochatae in B. bairdi juveniles reflects the phylogenetic relationship existing between the distinct taxa of the family Sabellidae.

The morphology of abdominal and thoracic uncini (avicular uncini with three rows of teeth above the main fang) is similar in juveniles and adults. This feature is particularly useful as it may prevent possible misidentifications with Branchiomma boholense (GRUBE 1878), where only a single large tooth is present over the main fang, even at early developmental stages. Indeed, as stated by several authors, in the past. B. bairdi could have been misidentified as B. boholense (Giangrande et al. 2012; Arias et al. 2013), and according to Tovar-Hernández et al. (2009b, 2011), the shape and number of teeth in thoracic uncini are one of the main diagnostic features to differentiate these two related species. Another important diagnostic feature useful to distinguish between B. bairdi and B. boholense is the shape of the macrostylodes (Tovar-Hernández & Knight-Jones 2006; Tovar-Hernández et al. 2009b, 2011), which in B. bairdi are strap-like (also reported by Arias [2013] for the Maltese population), while in *B. boholense* they are tongue-like. Moreover, in B. boholense, macrostylodes differ significantly from all the other stylodes, contrary to what is observed in *B. bairdi*. The morphology of stylodes in B. bairdi remained quite similar during the juveniles' growth. Stylodes first appeared associated with compound eyes at the base of the radioles when juveniles were at the 20chaetiger stage, and measured 1.4±0.2 mm in total length. When four pairs of stylodes were detected (25-chaetiger stage), the most basal one was longer, probably because the others were still growing. Indeed, with increasing body length and number of chaetigers, all the stylodes showed a similar length. Macrostylodes (one pair on each radiole) were not recognizable until about the 50-chaetiger stage $(8\pm0.5 \text{ mm in body length})$. Our observations stopped at the 60-chaetiger stage (22 ± 1.1 mm in total length)

The number of uncini per torus increases with worm growth; this feature is also correlated with width of the worms that further increases even when posterior segment addition drastically slowed. Indeed, as also observed by Parapar et al. (1993) in *Eunice vittata* (DELLE CHIAJE 1828), in *B. bairdi* initial growth is the result of an increase in number of chaetigers, but afterwards growth is mainly due to the enlargement of the chaetigers in length and width, with increase in width prevailing in final stages.

In conclusion, the information on the life cycle and reproduction of B. bairdi can explain the invasive potential of this species. In Mediterranean areas, B. bairdi colonizes unstable environments, where population size may fluctuate dramatically from year to year. The life-history traits of B. bairdi such as fast growth, early maturity, hermaphroditic habit, and short larval period may allow this species to build up very dense populations rapidly, even from a few settling larvae, as already suggested for B. luctuosum (Licciano et al. 2002). Moreover, the short larval period could also be a winning strategy in the competition for space with co-occurring sabellids, including other alien species such as B. luctuosum and the autochthonous S. spallanzanii (Giangrande et al. 2014). Tovar-Hernández et al. (2009a,b) mentioned shipping and aquaculture as vectors in the Gulf of California but did not clarify whether introduction occurs as adults or as larvae. However, as the pelagic larval phase in individuals of B. bairdi is very short (3 d), it could be hypothesized that the species' dispersion, as for other species, should be mainly linked to the accidental transport of adults as hull fouling rather than larvae in ballast water. Guidelines and effective regulations for cleaning and maintenance practices of vessels and movable immersed parts in harbors and marinas should be promoted, as well as sanitary prescriptions for the commerce of shellfish and other restocking shipments in mariculture trade.

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