Developmental Lifecycle of *Augeneriella dubia* (Sabellidae: Fabriciinae) from the Kahuku Reef Flat, O’ahu, Hawai’i

Lael Prince

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Mentors: Dr. Goodwill and Dr. Bybee
Abstract:

*Augeneriella dubia* specimens were collected, preserved and photographed to determine early developmental life cycles. Turf algae was collected from Kahuku, Oahu, Hawaii from November 2009-November 2010, *Augeneriella dubia* were teased out and placed in either 10% formalin or 95% ethanol. Approximately 60 specimens were preserved every month. Light microscopy and scanning electron microscopy were utilized to document the different developmental stages: egg, intermediate stage (egg elongation and segmentation), juvenile (all the development stages of an adult but smaller), and adult. Possible evidence of spermathecae were also found in the radial crown. Developmental stages follow that of the subfamily Fabriciinae which lack a larval stage.

Introduction:

The Sabellid polychaete reproductive mode is correlated with body size (Gambi & Paolo 1999, Giangrande et al. 2000, Bybee et al. 2006, 2007). The family Sabellidae is divided into two subfamilies: Fabriciinae and Sabellinae. Thorson (1950) and Hermans (1975) found that most members of the subfamily Sabillinae are characterized by large body size, reproduce by broadcast spawning numerous small eggs (< 180 μm), and develop indirectly via a larval stage.

Thorson (1950) and Hermans (1975) noted that members of the subfamily Fabriciinae reproduce by brooding small numbers of large eggs (>180 μm). They found Fabriciinae eggs tend to develop directly without larval stages within the parental tube or in the radials. Rouse (1994, 2005) described the presence of a dorsal sperm duct in males which opens behind the radials for release of free sperm into the water. He also noted that some female Fabriciinae have a pair of pigmented spermathecae located in the radiolar crown. The presence of spermathecae would allow females to store sperm.
(Rouse 1994, Rouse et al. 2006). The objective of this study was to photograph early developmental stages and to verify the mode of reproduction of *Augeneriella dubia*.

**Materials and Methods:**

Between November 2009 to November 2010 algae were collected during a low tide of 0.6 or lower from the intertidal regions at Kahuku Reef (21°41’2.89”N and 157°56’40.74”W) Oahu, Hawaii. Algae were observed with a dissecting light microscope for presence of *A. dubia* and its tube. Adult *A. dubia* outside of tubes were collected and tubes with or without adults were separated. The tubes were incised and the sabellid developmental stages were photographed and preserved. Twenty-five adult specimens were fixed in 10% formalin and switched to 70% ethanol within two weeks for future histological studies; another 25 adult specimens were preserved in 95% ethanol for future DNA studies. Subadult stages were fixed in 10% formalin and switched to 70% ethanol.

Approximately 50 adults and 50 subadults of various developmental stages were preserved in SEM primary fixative consisting of 4% glutaraldehyde in 0.1M sodium cacodylate buffer with 0.35M sucrose, pH 7.4, for 1-2 hours. Specimens were washed twice with 0.1M sodium cacodylate buffer with 0.44M sucrose for a total of twenty to thirty minutes. Specimens were then post fixed with 1% OsO4 in 0.1M sodium cacodylate for one hour. The sabellids were then dehydrated through an ethanol series (30%, 50%, 70%, 85%, 95%) of 2-3 washes for 3-5 minutes in each dilution. The specimens were subsequently dehydrated with 100% ethanol for three washes of ten minutes each. The final step involved drying specimens in a Tousimis Samdri-795 critical point dryer overnight.
The *A. dubia* specimens were mounted on aluminum stubs with either double-stick carbon tape or with carbon paste, and coated with gold/palladium in a Hummer 6.2 sputter coater three times for two minutes each. Digital images were acquired with a Hitachi S-4800 Field Emission Scanning Electron Microscope at an accelerating voltage of 5kV. The samples were stored in a desiccator to prevent oxidizing.

**Results:**

*Augeneriella dubia* were found in a variety of turf algae in the intertidal region. Specimens were found in soft sandy tubes ranging in color from tan to light gray. The tubes averaged 4.5 mm in length.

The eggs averaged 150 µms in diameter (Figure 1A). Eggs were spherical, orange in color, with a defined membrane (Figure 2A). Eggs developed into oblong structures with the beginnings of segmentation classified as an intermediate stage (Figure 1B, Figure 1C, Figure 2B, and Figure 2C). No larval stage was documented. The juveniles were identified as having all of the adult structures but on a smaller scale (Figure 1D, Figure 1E, and Figure 2D). Adults averaged 3 mm in length (Figure 1F) and retained an orange color in the internal organs. Adults obtained pigmentation during development resulting in an external brown hue (Figure 2E).

Eggs could be seen through the translucent abdomen of some sabellids (Figure 3). Two pigmented cavities were found in the medial aspect of the radial crown, indicative of spermathecae (Figure 3).
Figure 1. – The developmental stages of *Augeneriella dubia* using scanning electron microscopy: (A) early egg, (B) late stage egg beginning to elongate, (C) early intermediate stage with segmentation and the beginnings of radials as small buds, (D) intermediate stage with chaetae and continued segmentation, (E) juvenile with all the anatomical features of an adult on a smaller scale, and (F) adult.

Figure 2. – The *Augeneriella dubia* lifecycle utilizing light microscopy: (A) an egg, (B) early intermediate stage showing segmentation, (C) intermediate stage of elongation and further segmentation of body, (D) juvenile stage with radials and cheatae present, (E) a full grown adult.
Figure 3.- The female *Augeneriella dubia* with the evidence of eggs in the abdomen and the possible presence of spermathecae in the radial crown (indicated by arrows).

**Conclusion:**

*Augeneriella dubia* develop directly from the egg into the juvenile inside the parental tube, lacking a defined larval stage. This is presumed to guarantee a high survival rate. The absence of a larval stage is characteristic of brooders of the subfamily Fabriccinea (Rouse et al. 2006, Thorson 1950, Hermans 1975). The possible presence of spermathecae would further indicate a brooding reproductive strategy. (Rouse 2005). Rouse (1994) states that all females of the subfamily Fabriciinae brood directly within their tubes. However, he also states that females of the *Augeneriella* genus lack spermatheceae or other specialized organs for sperm storage, but rather store sperm in the innersurface of the radial crown or in epidermal cells. *Augeneriella dubia* has pigmented cavities in the right location for spermatheceae, perhaps indicating that *A.*
Augeneriella dubia has been placed in the wrong genus. Histology and transmission electron microscopy is needed to confirm or reject the presence of spermatheceae.

All stages of development were found each month of the study, with the exception of June and July of 2010 due to insufficient collections. Augeneriella dubia are brooders. However, it cannot be stated they are continually brooding without additional collections during June and July in order to find subadults or histological studies looking for gametogenesis during the missed months.

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Literature Cited


