Title
Preliminary Surveys of the Commensal Amphipod, Leucothoe Spinicarpa (Abildgaard, 1789), in the colonial tunicate, Ecteinascidia thurstoni Herdman, 1891, in the Andaman Sea, Thailand

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Abstract Amphipods identified as, *Leucothoe spinicarpa* (Abildgaard, 1789), were found in the colonial tunicate, *Ecteinascidia thurstoni* Herdman, 1891, at 1-3 m depth, within a single coral reef area on the coast of the Andaman Sea of Phuket Province in southern Thailand. This represents the first record of commensalism between this amphipod and tunicate in Thai waters. Host-symbiont occurrences were low, with only 2.2% of all tunicate zooids harboring *L. spinicarpa*, and always with a single amphipod per zooid. All *L. spinicarpa* occurred in the branchial chamber of the tunicate and included female and male specimens. Amphipods found in the tunicates ranged between 0.4-2.1 mm in length.

Key words: amphipod, tunicate, *Leucothoe spinicarpa*, *Ecteinascidia thurstoni*, commensal

Introduction

Amphipods can be found in association with macroalgae and marine invertebrates, using them as habitat, food sources, and as protection from predators (Buschmann, 1990; Chavanich, 2006; Chavanich and Wilson, 2000; Duffy and Hay, 1991; Hacker and Steneck, 1990). Some amphipods living inside host organisms such as tunicates, sponges, and sea anemones are considered commensal symbionts (Thomas, 1979; Vader, 1970; Vader, 1984; Vader and Krapp-Schickel, 1996). They receive benefits from their hosts through the provision of microhabitats and food sources, while not harming their hosts (Jaramillo et al., 1981; Thiel, 1999; Vader, 1985; Vader and Beehler, 1983).

As a group, leucothoids are commensal amphipods usually found inside sponges and ascidians (Barnard and Karaman, 1991; Ortiz, 1975). To date, *Leucothoe spinicarpa* (Abildgaard, 1789) is the only species found inside the Thai colonial tunicate, *Ecteinascidia thurstoni* Herdman, 1891. This tunicate has attracted attention in Thailand recently as a new potential source of anti-cancer compounds (Chavanich et al., 2005). A group of alkaloids, the Ecteinascidins (Et), including Et 770 and Et 786, have been isolated with very high yields from *E. thurstoni* pretreated with potassium cyanide. These extracts exhibit potent cytotoxic activity against cancer cells of breast, lung, colorectal, and nasopharyngeal tissues (Suwanborirux et al., 2002).

The purpose of the present study was to investigate the host-symbiont association of *Leucothoe spinicarpa* and *Ecteinascidia thurstoni* in Thai waters, to discover preliminary details of the commensal amphipod populations and ascertain specifics such as the positions inside the tunicates where the amphipods were found.

Materials and Methods

During the 2nd Annual JSPS-NaGISA workshop on the taxonomy of marine amphipods in Nha Trang, Vietnam from September 30-October 3, 2004, the presence of an amphipod inside the tunicate,
Ecteinascidia thurstoni was noted. Subsequent identification during the workshop lead by Dr. Manuel Ortiz Touzet showed this amphipod to be Leucothoe spinicarpa (Abildgaard, 1789).

Specimens of the tunicate, E. thurstoni, were collected at 1-3 m depth along the coast of Phuket Province, southern Thailand. Tunicates were dissected, and amphipods found inside host specimens were collected and fixed in 5% buffered formalin for later identification. A total of 1500 tunicate zooids were collected between the months of September 2004 and April 2006. In addition, notes on the sex and position of amphipods in the tunicates collected, were recorded. Amphipod specimens were also measured from the anterior edge of the first pereion segment to the posterior edge of the fifth pereion segment.

**Results**

Leucothoe spinicarpa was the only amphipod found inside the tunicate Ecteinascidia thurstoni (Figures 1, 2). 2.2% of the tunicate zooids acted as hosts, with most zooids containing no amphipods
Table 1. Percentage of amphipods found inside the tunicate *Ecteinascidia thurstoni* Herdman, 1891.

<table>
<thead>
<tr>
<th>Tunicates</th>
<th>Percent of amphipods</th>
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</thead>
<tbody>
<tr>
<td>without amphipods</td>
<td>97.8%</td>
</tr>
<tr>
<td>with amphipods</td>
<td>2.2%</td>
</tr>
<tr>
<td>with female amphipods</td>
<td>0.67%</td>
</tr>
<tr>
<td>with male amphipods</td>
<td>1.06%</td>
</tr>
</tbody>
</table>

Fig. 3. Size classes of the amphipod *Leucothoe spinicarpa*.

All zooids hosted only a single individual *L. spinicarpa* amphipod. Both female and male amphipods were found, and all *L. spinicarpa* occurred in the branchial chamber part of the tunicate (Table 1). Amphipods found in the tunicates ranged between 0.4-2.1 mm in length (Figure 3).

Discussion

A previous study found three commensal amphipod species in four sponges and one solitary tunicate in Phuket, Thailand (Bahrdorff and Lofstedt, 2004). However, *L. spinicarpa* was not among the amphipods recorded. The present study is the first record of the commensal amphipod *L. spinicarpa* found associated with the colonial tunicate *E. thurstoni* in Thai water. *L. spinicarpa* has also been found inhabiting other tunicate species: *Ecteinascidia turbinata*, *Styela plicata*, *Ascidia nigra*, *Microcosmos exasperatus*, and *Clavelina oblonga* (Thiel, 1999).

The amphipod specimens obtained from inside the tunicates spanned a large size and age range (both juveniles and adults were present). This finding is contrary to Thiel’s (1999) report that only small specimens of *L. spinicarpa* live in colonial tunicates while larger ones live in solitary tunicates. He suggested that juveniles first lived with their parents in solitary tunicates, and when mature, sought colonial tunicates as their host. Our results do not match the suggested pattern. Another study by Bahrdorff and Lofstedt (2004) also seems to contradict Thiel (1999) in that it found no *L. spinicarpa* in solitary tunicates. In agreement with other studies (Thiel, 1999; Thomas and Klebba, 2006), we found *L. spinicarpa* occurring in the branchial chamber of the tunicate. This tunicate organ seems to facilitate commensal amphipods in finding food among the materials filtered by the host (Thiel, 1999).
Results on the population of commensal amphipods showed that only 2.2% of all tunicate zooiids were host to *L. spinicarpa* (Table 1). Previous studies have reported that high percentages of *L. spinicarpa* were found inside solitary tunicates (77-95% of their hosts) and colonial tunicates (9.4% of their hosts) (Thiel, 1999). Several factors such as choice of preferred hosts and abundance of hosts and other commensal amphipod species might influence the numbers and reproduction of *L. spinicarpa* populations. It appears that more studies are needed to further elucidate the factors involved and to determine the relationship between *L. spinicarpa* and its hosts.

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