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Ecology of the sea squirt *Ecteinascidia thurstoni* herdman, 1890 (ascidiacea: perophoridae) along Suez Canal and Egyptian Red Sea coasts

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ABSTRACT

The tunicate *Ecteinascidia thurstoni* is a colonial sea squirt. It has a seasonal rhythm, usually present during summer months with tropical and subtropical distribution pattern. It synthesizes a group of molecules called ecteinascidines. One of which is ET-743, a compound that has a most original anti-tumoral activity and is considered today one of the most promising substances effective against various solid-type tumors (currently in the market under the commercial denomination of Yondelis for treatment of sarcoma and related tumors, or in clinical trails, phase II/III, for other kinds of tumors). Allover the world, species of *Ecteinascidia* represent the only available source for this bioactive compound, which was first discovered in *E. turbinata*. During the present study, the ecology of *E. thurstoni* along Suez Canal and Red Sea was investigated. Its populations were observed to be highly gregarious due in part to their low larval dispersal, which is very localized, and hence, the larvae tend to settle close to their parent colonies. It is only recorded in shallow waters (0.5-1.5m) at mangrove habitats of Red Sea, epiphytic on their pneumatophores; or on pilings of jetties, and metal or cement banks of Suez Canal. Along Red Sea, a total of 29 mangrove sites were surveyed. From El-Monkata'a at Nabq Protected Marine Park (north) to Halayeb, at Egyptian-Sudanese border (south), four sites of them, namely, El-Rawisia, Wadi El Qala'an, Wadi Rawad El-Adaiah, and Wadi El-Ra'ada were found to have *E. thurstoni.* However, along Suez Canal, it is more frequent and has higher bicmass than Red Sea. It was recorded at southern sector of Suez Canal from Lake Timsah farther south to Bitter Lakes. Suez Canal's population has significantly different morphometric characteristics (zooid length, zooid weight, colony weight) than those of Red Sea. Studying the distribution of this species and locating its different populations along Suez Canal and Red Sea could help in characterizing their genetics, their chemistry and bacterial community at different isolated locations, which ultimately will help define the source of ET-743 and hence promotes its biosynthesis on commercial scale.

Key words: Distribution, tunicate, Sea squirt, *Ecteinascidia thurstoni*, Red Sea, Suez Canal.

INTRODUCTION

Many marine invertebrates such as sponges and ascidians (Rinehart, 2000) that have not been traditionally exploited as fisheries resources, have recently received increased attention, in this sense, due to their great economic potential for the production of compounds with pharmaceutical properties (Carballo *et al.*, 2000; Rinehart, 2000).

Species of *Ecteinascidia* which are colonial tunicates from the family Perophoridae, represent an important component of the benthic fauna of the mangroves (Carballo, 2000b; Hernández-Zanuy *et al.*, 2007). *E. turbinata* has been the subject of various studies examining its settlement, species succession, and larval behavior (Young and Bingham, 1987; Bingham and young, 1991, 1995). In the 1960s, interest in this species was high lightened when an extract of the animal was found to have potent cytotoxic properties, the ecteinascidins, that were identified and characterized (Rinehart *et al.*, 1990). Little is known about the production of these secondary metabolites or what function, if any, they play in the animal (Moss *et al.*, 2003). In this sense, *E. turbinata* synthesizes a group of compounds called ecteinascidines, one of which is ET-743, a compound that has a most original anti-tumoral activity and is considered today one of the most promising substances effective against various solid-type tumors (Rinehart *et al.*, 1990; Garcia-Rocha *et al.*, 1996; Jimeno *et al.*, 1996). Recently, it is currently in the market under the commercial denomination of Yondelis for treatment of sarcoma and related tumors, or in clinical trails (phases II/III) for other kinds of tumors (Pharmamar, 2008).

The supply of ectainaicidins for clinical development was also maintained during several years by harvesting of *E. turbinata* on a sustainable basis, which is an example of the production of metabolites while protecting natural populations (Carballo *et al.*, 1999). Harvesting of this tunicate in large quantities was possible with the establishment of a rational extraction program in diverse habitats, as is the case with a lot of shellfish for human consumption. A research program was carried out in order to find out the recovery capacity of *Ecteinascidia turbinata* populations, after carrying out harvesting experiments in the Caribbean and Mediterranean Sea (Carballo *et al.*, 1999). During the last few years, PharmaMar (Spanish pharmaceutical company) produced ecteinascidins for its clinical studies from aquaculture in a natural environment: more than 70 tons/year until 2000 (Carballo *et al.* 2000, Naranjo, 2001). Now, production through recollection or aquaculture is not already necessary and expensive. Moreover, currently, the production of Yondelis is completely synthetic, neither from natural resources, nor aquaculture activities (Cuevas *et al.*, 2000). The synthesis of this compound is a multi-step process, and the cost of reagents is affordable on a small scale, but still too expensive for large-scale synthesis for commercial purposes.

Recently, the investigation of secondary metabolites in the tunicate, *Ecteinascidia thurstoni*, pretreated with potassium cyanide, highlighted its pharmaceutical and medicinal importance, where ecteinascidines. ET-743, ET-770, Et-729, and Et-759 were present in samples collected from Suez Canal and Red Sea in 2005 (Youssef, 2007). A group of alkaloids, the Ecteinascidins (ET), including ET- 770 and ET-786, have been isolated with very high yields from *E. thurstoni* (Phuket Province, southern Thailand, on the Andaman Sea coast) pretreated with potassium cyanide. These extracts exhibited potent cytotoxic activity against cancer cells of breast, lung, colorectal, and nasopharyngeal tissues (Suwanborirux *et al.*, 2002). The finding of Suwanborirux *et al.* (2002) means that there are other types of ecteinascidines, (not ET-743) which could have a potential therapeutic value for treatment of other diseases.

The aim of the present work is to study the ecology of the tunicate *E. thurstoni* along Suez Canal and Red Sea, describing its pattern of distribution at different habitats, locating different populations along the shore, also, measuring the morphometric characteristics of these populations. This information will be helpful for studying genetics, chemistry and bacterial community of different *Ecteinascidia* populations along Suez Canal and Red Sea. Ultimately, this will aid in further development of the compound in the lab and hence its biosynthesis on an economic commercial scale.

MATERIALS AND METHODS

The present study was carried out along the coasts of Suez Canal and the Egyptian Red Sea. The coasts of Suez Canal are sedimentary, while Red Sea shores are mainly rocky. A pilot survey carried out in summer 2005, during which different habitats were surveyed by snorkeling. These habitats included pilings of jetties, metal and cement banks, and seagrasses along Suez Canal; and coral reefs, seagrasses and mangroves and rocky shores of Red Sea. This pilot survey showed that cement and metal banks and pilings of jetties along Suez Canal; and Mangroves along Red Sea represent suitable habitats for the tunicate *Ecteinascidia thurstoni*, to be present and grew. This usually occurs during summer months from end of May to the beginning of October. The species usually disappears during autumn and winter seasons, where ecteinascidians are species characteristics to warm tropical waters, in environmental conditions (especially temperature and salinity) adequate for its survival in natural shallow water.

After the pilot survey in summer 2005, during the following summers of 2006 and 2007, an intensive survey to study the ecology of this species by snorkeling at different habitats along Suez Canal and Red Sea was achieved. The study sites, their GPS position, were presented in Figure 1 and Tables 1& 2.

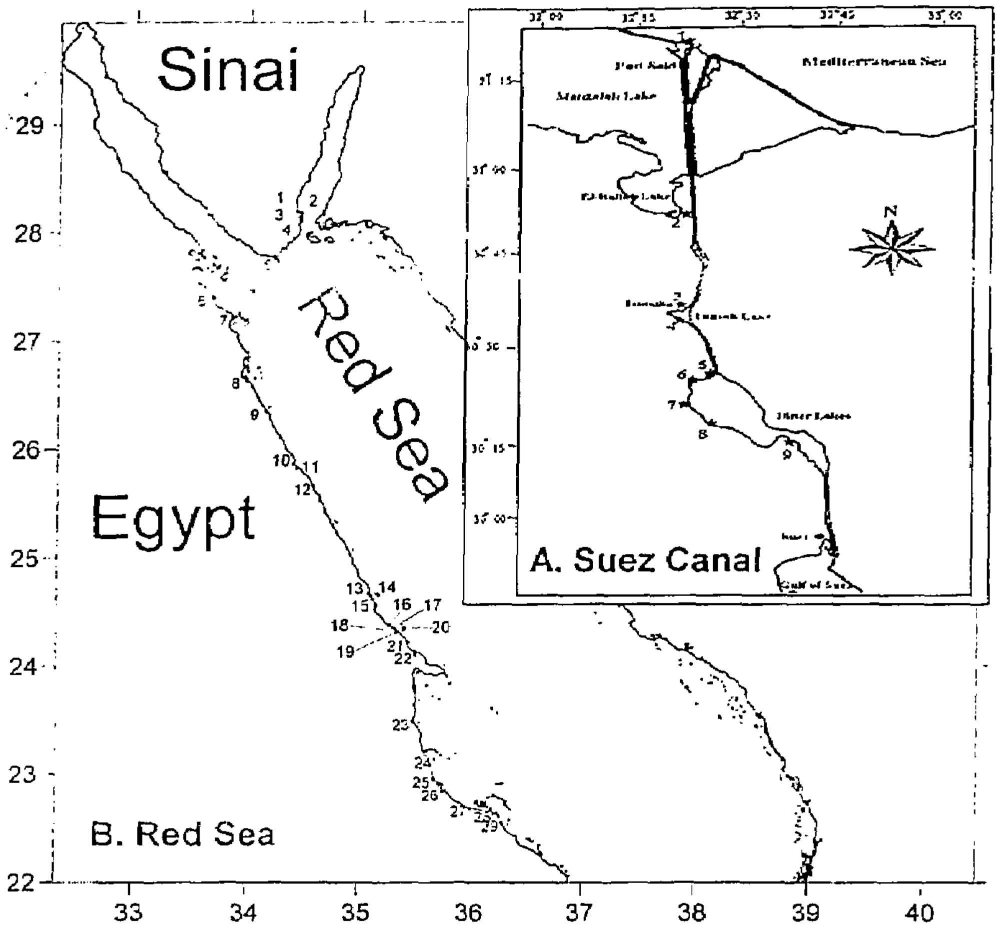

Fig. 1. Different surveyed sites along Suez Canal (A) and Red Sea (B).

Table 1. Different surveyed sites and their GPS position along Suez Canal.

No.	Site	Position	
1	Port Said	31° 13' N	32 ° 21' E
2	El-Kantara	31° 51' N	32 ° 18' E
3	El-Danvah Club	30° 33' N	32 ° 18' E
4	El-Ta'awen	30° 24' N	22° 18' E
5	Deversoir	30° 24' N	32 ° 21' E
6	Abu Sultan	30° 22' N	32 ° 18' E
7	Fayed	30° 19' N	32 ° 19' E
8	Fanara	30° 17' N	32 ° 18' E
9	Kabreet	30° 15' N	32 ° 28' E

Table2. Different surveyed sites and their GPS position along Red Sea.

No.	Site	Position	
1	El-Monkara'a	28° 12' N	34° 25' E
2	El-Rawisia	28° 11' N	34° 27' E
3	Marsa Abu-Zabad	28° 09' N	34° 27' E
4	El-Ghargana	28° 07' N	34° 27' E
5	Ras Mohamed	27° 44' N	34° 15' E
6	Al-Gonah	27° 24' N	33° 41' E
7	Abu-Monkar	27° 13' N	33° 52' E
8	South Safaga	25° 38' N	33° 59' E
9	Wadi Abu-Hamrah	26° 21' N	34° 09' E
10	Sharm El-Bahari	25° 52' N	34° 24' E
11	Sharm El-Qebly	25° 50' N	34° 26' E
12	Marsa Sagara	25° 40' N	34° 35' E
13	Wadi El-Gemal	24° 40' N	35° 05' E
14	Wadi El-Gemal Island	24° 40' N	35° 10' E
15	Ras Baghdadi	24° 39' N	35° 06' E
16	Wadi Masturah	24° 23' N	35° 16' E
17	Wadi Qala'an	24° 21' N	35° 19' E
18	Wadi Rawad El-Adaiah	24° 20' N	35° 20' E
19	Wadi El-Ra'da	24° 19' N	35° 20' E
20	Shawareet Island	24° 21' N	35° 24' E
21	Wadi Lahmy	24° 13' N	35° 26' E
22	Qora'at Hartway	24° 06' N	35° 30' E
23	El-Hamirah Mangrove	23° 29' N	35° 29' E
24	Shalateen Island	23° 08' N	35° 41' E
25	Sharm El-Madfa'a	22° 57' N	35° 40' E
26	Marsa Sha'ab	22° 50' N	35° 45' E
27	Marsa Abu-Fasi	22° 41' N	36° 00' E
28	El-Hoor	22° 38' N	36° 13' E
29	Adal Deep	22° 33' N	36° 17' E

At different surveyed sites, ecological data including depth, salinity (‰), temperature (°C), pH, and oxygen concentration (mg/l), and the presence or absence of *E. thurstoni* were recorded *in situ*. At sites, wherever *E. thurstoni* was present, samples have been collected and kept fresh in containers with sea water. On returning to the laboratory (Marine Sciences Department at Ismailia; Sharm El-Sheikh Environmental School; and Marine Parks at Hurghada), other biological parameters were measured, which include maximum colony diameter(mm), maximum number of zooids per colony, maximum weight of colony (g), maximum length of zooid (mm), and maximum weight of zooid (g). These data were used to compare which sites have a better population of *E. thurstoni*.

For data analysis, means and standard deviations were calculated for replicate determinations of various parameters investigated. Analysis of variance (One-way ANOVA) test was used to evaluate the significance of differences between groups of measured parameters at different stations with a level of significance set at $p \leq 0.05$.

RESULTS

Recorded environmental conditions along Suez Canal (Table 3) and Red Sea coast (Table 4) during the investigations period was more or less typically normal with temperature not less than 27±2°C and 27±1°C at both investigated habitats, respectively, while salinity was not less than 42±1‰, and 42=2‰ at all sites of Suez Canal or Red Sea, respectively. Also, *in situ* measurements showed all surveyed sites well aerated with good content of oxygen being 5.2±0.5mg/l; and 5.9±0.3mg/l at sites of Red Sea and Suez Canal, respectively. pH showed normal readings above the neutral limits, being 7.2±0.4; and 7.5±0.3 at sites of Red Sea and Suez Canal, respectively. Statistical analysis (One-way ANOVA) showed no significant difference between different sites for different parameters (temperature, salinity, pH, oxygen content) investigated (Table 7).

Table3. Occurrence of *E. thurstoni* at different sampling sites along Suez Canal during summer 2006, 2007.

No.	Site	Salinity (‰)	Temp. (°C)	pH	Oxygen (mg/l)	Habitat	*E. thurstoni* Present/ absent
1	Port Said	43 ± 1	27 ± 1	7.5 ± 0.3	6.5 ± 0.3	Cement banks of Suez Canal	-
2	El-Kantara	43 ± 1	27 ± 1	7.9 ± 0.3	6.9 ± 0.4	Cement banks of Suez Canal	-
3	El-Danva Club	42 ± 2	28 ± 2	8.2 ± 0.2	6.2 ± 0.4	Cement banks of Suez Canal	+
4	Tosun	43 ± 1	27 ± 1	7.9 ± 0.2	7.1 ± 0.3	Metal pilings of jetties	+
5	Deversoir	44 ± 2	28 ± 1	7.8 ± 0.2	7.5 ± 0.3	Metal pilings of jetties	+
6	Abu -Sultan	43 ± 1	27 ± 1	7.8 ± 0.3	6.9 ± 0.5	Metal pilings of jetties	++
7	Fayed	43 ± 1	28 ± 2	8.2 ± 0.3	6.3 ±0.3	Metal pilings of jetties	+++
8	Fanara	43 ± 1	28 ± 2	8.3 ± 0.3	5.9 ±0.3	Cement banks of Suez Canal	++
9	Kabreet	44 ± 2	29 ± 1	8.5 ± 0.2	7.2 ±0.2	Cement banks of Suez Canal	+

- Absent
– Rare
–+ Commom
–++ Abundant

Table4. Occurrence of *E. thurstoni* at different sampling sites along Red Sea during summer 2006, 2007.

No.	Site	Salinity (‰)	Temp. (°C)	pH	Oxygen (mg/l)	Habitat	*E. thurstoni* Present/ absent
Nabq & Ras Mohamed							
1	El-Monkara'a	44 ±2	28 ±1	7.5 ±0.2	6.1 ±0.3	*Avicennia marina*	-
2	El-Rawisia	44 ±2	28 ±1	7.5 ±0.3	6.2 ±0.2	*A. marina*	+
3	Marsa Abu -Zabad	44 ±1	28 ±1	7.7 ±0.3	6.1 ±0.1	*A. marina*	-
4	El-Ghargana	44 ±1	27 ±2	7.5 ±0.2	6.3 ±0.2	*A. marina*	-
5	Ras Mohamed	44 ±1	27 ±2	7.6 ±0.3	6.2 ±0.2	*A. marina*	-
Hurghada							
6	Al-Gonah	45 ±1	28 ±1	7.7 ±0.3	5.3 ±0.4	*A. marina*	-
7	Abu-Monkar	45 ±1	27 ±1	7.8 ±0.1	5.4 ±0.4	*A. marina*	-
8	South Safaga	44 ±1	27 ±1	7.8 ±0.1	5.3 ±0.3	*A. marina*	-
Quseir							
9	Wadi Abu-Hamrah	44 ±1	27 ±2	7.9 ±0.1	5.9 ±0.3	*A. marina*	-
10	Sharm El-Bahari	44 ±2	27 ±2	8.1 ±0.2	5.6 ±0.5	*A. marina*	-
11	Sharm El-Qebly	43 ±2	27 ±2	8.1 ±0.2	5.8 ±0.3	*A. marina*	-
12	Marsa Sagara	43 ±2	29 ±1	7.6 ±0.3	5.7 ±0.4	*A. marina*	-
13	Wadi El-Gemal	43 ±1	27 ±1	7.7 ±0.3	5.6 ±0.4	*A. marina*	-
14	Wadi El-Gemal Island	43 ±1	28 ±1	8.1 ±0.2	5.5 ±0.3	*A. marina*	-
15	Ras Baghdadi	44 ±2	28 ±1	8.2 ±0.3	6.1 ±0.2	*A. marina*	-
Hamata							
16	Wadi Masturah	42 ±1	27 ±2	7.2 ±0.4	5.9 ±0.3	*A. marina*	-
17	Wadi Qala'an	43 ±1	27 ±1	7.3 ±0.5	5.8 ±0.3	*A. marina*	+
18	Wadi Rawad El-Adaiah	43 ±1	28 ±1	7.5 ±0.5	6.3 ±0.2	*A. marina*	+
19	Wadi El-Ra'da	44 ±2	27 ±1	7.6 ±0.4	6.2 ±0.2	*A. marina*	++
20	Shawareet Island	43 ±1	28 ±1	7.7 ±0.4	5.7 ±0.3	*A. marina*	-
21	Wadi Lahmy	44 ±1	27 ±2	7.5 ±0.5	5.6 ±0.4	*A. marina*	-
22	Qora'at Hartway	44 ±1	28 ±2	7.8 ±0.3	6.0 ±0.3	*A. marina*	-
Shalateen							
23	El-Hamirah Mangrove	44 ±1	28±1	7.6 ±0.2	6.1 ±0.3	*A. marina*	-
24	Shalateen Island	43 ±1	27 ±1	7.7 ±0.4	5.8 ±0.4	*A. marina*	-
25	Sharm El-Madfa'a	44 ±2	27 ±1	7.8 ±0.4	5.7 ±0.4	*Rhizophora mucronata*	-
26	Marsa Sha'ah	43 ±1	28 ±1	7.7 ±0.3	6.2 ±0.3	*A. marina, R. mucronata*	-
Halayeeb							
27	Marsa Abu-Fasi	44 ±1	27 ±1	7.9 ±0.4	5.9 ±0.4	*A. marina, R. mucronata*	-
28	El-Hoor	43 ±1	28 ±1	8.1 ±0.2	5.2 ±0.5	*A. marina, R. mucronata*	-
29	Adal Deep	44 ±1	29 ±1	8.2 ±0.2	5.3 ±0.5	*A. marina, R. mucronata*	-

- Absent
+ Rare
++ Commom

Examination of the collected specimens showed that the colony of *Ecteinascidia thurstoni* (Fig. 2A) consists of thick clusters of individual zooids connected at their bases by a net work of stolons that adheres to the surface of the object, e.g. pneumatophores of mangroves, on which the colony grows. Figure 2(B) shows that the community of *E. thurstoni* in Suez Canal usually has big colonies, while the community of Red Sea has small colonies at most sites as shown in Figure 2(D). However, medium size or dense colonies were recorded at Wadi El-Ra'ada as shown in Figures 2(E); and (F), respectively.

During the field survey in summer 2005, *E. thurstoni* was found in mangrove habitats of the Red Sea, while other habitats which include coral reefs, seagrasses, artificial hard substrates e.g. jetties, cement and metal pilings did not have any individuals or colonies of such tunicate. On the other hand, along Suez Canal coasts, *E. thurstoni* were reported flourishing on pilings of jetties, metal and cement banks of the Canal especially at Bitter Lakes (Table 5), where the species colonies were more frequent than in the Red Sea.

Table 5. Occurrence and frequency of *E. thurstoni* at different habitats of Suez Canal and Red Sea (summer, 2005).

Habitat	Occurrence	Frequency
Red Sea		
Coral reef	absent	-
Seagrass	absent	-
Mangroves	Present	less frequent
Suez Canal		
Seagrass	absent	-
pilings of jetties	present	frequent
Metal & cement banks	present	frequent

During summer 2006 and 2007, along the Red Sea, the survey was concentrated mainly on mangroves, where 29 sites were investigated (Table 4). The survey showed that only at 4 sites out of 29, *E. thurstoni* was present. These sites were El-Rawisia at Nabq Protect Marine Park, Gulf of Aqaba (north); and the other three sites were Wadi El Qala'an, Wadi Rawad El-Adaiah, and Wadi El-Ra'ada (south). At the first three sites, *E. thurstoni* was found and represented by colonies that have very few, small zooids. These zooids were usually found on pneumatophores of the mangrove *Avicennia marina*, or on thallus of the seaweed *Laurancia obtusa* (at El-Rawisia). At the fourth site (Wadi El-Ra'ada), *E. thurstoni* was represented by colonies with relatively numerous zooids on the basal part of pneumatophores of *Avicennia marina*. This site (Wadi El-Ra'ada) was the most productive along the coast of Red Sea for collection of this tunicate. Also, the field survey has shown that roots of the mangrove *Rhizophora mucronata* along Shalateen – Halayeeb sector did not support any colonies of this tunicate (Table 4). On the other hand, along Suez Canal, investigation of nine sites during the same season has shown that the species usually flourishes on pilings of jetties especially at Fayed on Bitter Lakes (Table 3), where big colonies with large zooids were found and collected.

The maximum morphometric characteristics of *E. thurstoni* colonies at different sites were presented in Table 6. The Data showed that the population of Suez Canal is more productive in terms of biomass, where the colonies could reach a maximum weight of 300g at Fayed (Bitter Lakes) which usually grow at shallow calm water. The higher weight of colonies mainly related to colony diameter, zooid length, weight, and number per colony. From Table 4, it is clear that colonies with the maximum diameter (100mm) were collected from Fayed. Also, colonies with the maximum number of zooids were collected from the same site (250 zooid/colony) on the Suez Canal; and from Wadi El-Ra'ada (163 zooid/colony) on the Red Sea coast. Zooids that had a maximum length (20mm) were recorded at Fayed and Fanara on Suez Canal; and at mangrove of Wadi El-Ra'ada (11mm). Also, zooids with a maximum weight of 1.3g and 1.2g was recorded at Fayed and Fanara, respectively, while, they weighed 0.26g at Wadi El-Ra'ada.

Table 6. The morphometric characteristics of E. *thurstoni* along Suez Canal and Red Sea coasts (samples collected during summer 2006, 2007)

No.	Site	Max. diameter or length of colony (mm)	Max number of zooids/ colony	Max. weight of colony (g)	Max. length of zooid (mm)	Max. weight of zooid (g)
Suez Canal						
3	El-Danva Club	70	110	90	17	1.0
4	Tosun	70	150	120	17	1.0
5	Deversoir	80	155	140	18	1.1
6	Abu -Sultan	90	140	130	18	1.1
7	Fayed	100	250	300	20	1.3
8	Fanara	70	170	150	20	1.2
9	Kabreet	50	120	110	19	1.1
Red Sea						
2	El-Rawisia	Very patchy, separate, on thallus of *Laurancia obtusa*, growing on pneumatophores.	20	1.00	7	0.15
17	Wadi Qala'an	Very Patchy, separate, on basal part of pneumatophores.	5	1.00	6	0.25
18	W. Rawad El-Adaiah	Very patchy, separate, on basal part of pneumatophores.	6	1.00	7	0.25
19	Wadi El-Ra'ada	130 mm, basal part of pneumatophores.	163	30	11	0.26

One-way ANOVA showed highly significant difference between sites of Suez Canal and Red Sea for different morphometric characteristics (zooid length, weight, and number per colony, as well as colony weight) (Table 7).

Table 7. One way analysis of variance for different ecological parameters and morphometric characteristics of *E. thurstoni* between different stations.

Source of variation	df	Sum of squares	Mean square	F. value	p. value
Salinity	5	216.658	43.332	0.319	0.898
Temperature	5	83.511	16.702	0.119	0.987
pH	17	946.500	55.676	0.307	0.991
O_2	25	3615.500	144.620	1.819	0.140
Weight of zooid	6	200.667	33.444	25.948	0.0001*
Weight of colony	10	217.250	21.725	86.900	0.0001*
Length of zooid	6	213.000	35.500	76.071	0.0001*
No. zooids/ colony	16	220.000	13.750	55.00	0.0001*

P value ≤ 0.05%

* *significantly different*

DISCUSSION

Results dealing with the distribution of the ascidian *Ecteinascidia thurstoni* along Suez Canal and Egyptian Red Sea showed that it is like other species of *Ecteinascidia*, has a seasonal pattern, usually present during summer months. Its distribution is mainly related to seasonal fluctuation in temperature, where, at low temperature, the colonies usually do regression to the stolons. Those stolons usually buds again in summer to form new zooids and hence colonies by asexual reproduction. A primary characteristic of *Ecteinascidia* species, to consider, is its stolon-generated asexual growth by budding (Satoh, 1994). Budding results in a rapid development of the colony and ensures the animal's survival during adverse environmental conditions in winter (Carballo, 2000a).

All over the world, the distribution of *Ecteinascidia* was described along Estany des Peix Lagoon on Formentera Island, Spain (Carballo *et al.*, 1997), mangels of Gulf of Mexico and the Yucatan Peninsula by Carballo (2000b), and in Cuba by Hernández-Zanuy *et al.* (2007), where it was found in shallow waters of 1-3m depth. Along Suez Canal and Egyptian Red Sea, the most interesting point regarding its distribution was its occurrence at the most north of the Red Sea (Gulf of Aqaba – El-Rawisia) and farther south at Hamata mangroves; the distance in between (600 km), couldn't support any colonies of the tunicate *Ecteinascidia*. This could be related to the settlement behavior of the larvae. *Ecteinascidia* populations are highly gregarious due in part to low larval dispersal, and so become very localized (Bingham and Young, 1991; Carballo, 2000a); the larvae tend to settle close to their parent colonies. The short distance dispersal of the tadepole larvae, due to short life span (few hours), and the location of the settlement structures (e.g. pneumatophores) close to the population nucleus provide some guarantee of success for larval capture and settlement and to prevent advection to inappropriate sites. So, this could partially explain the patchiness existence of *Ecteinascidia* species along the Red Sea. Other environmental conditions e.g. currents, tidal pattern, and predators could play additional important roles on their distribution. Long-distance exchange between isolated islands probably occurs through rafting of adults colonies on fragmented mangrove roots rather than through larval dispersal (Bingham and Young, 1991).

The other interesting feature is that the species has different morphometric characteristics (zooid's length, weight, and number per colony, in addition to colony weight) at different locations of Suez Canal and Red Sea. Suez Canal population had maximum morphometric characteristics than Red Sea population (Table 6). Zooid length (height) ranges from 6mm to 20mm. The minimum and medium zooid lengths were recorded at Red Sea sites, while the maximum zooid lengths or heights were recorded at Suez Canal sites. For Mediterranean population of *E. turbinata*, zooid height or length ranged from 12.3mm to 20.65mm at the Estany des Peix Lagoon on Formentera Island, Spain

(Carballo *et al.*, 1997). In Phuket Province, southern Thailand, this tunicate is only found in one reef area at 1–3 m depth, on the Andaman Sea coast. The zooid ranges between 8 and 12 mm in height (Chavanich *et al.*, 2005). These populations with different morphometric characteristics could be studied for their bacterial community, chemistry and genetics. The resulting information could be implemented for achieving the biosynthesis of ET-743. The development of this drug is hampered during last years due to the difficulty in obtaining adequate cheap supply of the compound. Margo Hygood, Oregon Health and Science University (Pers. Comm.) hypothesizes, that ET-743 is in fact microbial in origin. It shows remarkable similarity to other microbial secondary metabolites. So, characterizing the bacterial community as well as the chemistry of *Ecteinascidia* species from different locations will be helpful to identify which bacterial species are common to all tunicates containing the bioactive metabolites. Furthermore, identification of a non-bioactive population of *Ecteinascidia* would be extremely valuable in determining the true symbionts of the animal, as well as the potential source of the compound. Ultimately, this will aid in further development of the compound in the lab and hence its biosynthesis on an economic commercial scale.

Finally, it could be recommended to study the distribution of *E. thrustoni* and locating its different populations along Suez Canal and Red Sea that could help in studying their genetics, as well as their chemistry and bacterial community at different isolated locations, which will accurately help to define the source of ET-743 and hence its biosynthesis on an economic commercial scale becomes more cheaper than chemical synthesis.

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REFERENCES

Bingham, B. L. and Young, C. M. (1991). Larval behavior of the ascidian *Ecteinascidia turbinata* Herdman; An in situ experimental study of the effects of swimming on dispersal. J. Experimental Marine Biology and Ecology, 145: 189-204.

Bingham, B. L. and Young, C. M. (1995). Stochastic events and dynamics of a mangrove root epifaunal community. Marine Ecology P.S.Z.N., 16: 145–163.

Carballo, J. L. (2000a). Larval ecology of an ascidian tropical population in a Mediterranean enclosed ecosystem. Marine Ecology Progress Series, 195: 159–167.

Carballo J. L. (2000b). Distribución de *Ecteinascidia turbinata* (Ascidiacea: Perophoridae) en los manglares del la Península de Yucatán (México). Revista de Biología Tropical, 48(2/3): 365-369.

Carballo, J. L.: Naranjo, S.; Kukurtzii, B. and Hernandez, A. (1997). Estudio de la poblacion de *Ecteinascidia turbinata* Herdman 1880 (Ascidiacea: Perophoridae) en la Isla de Formentera (Mar Mediterraneo. Espana): Distribucion, densidad y crecimiento. Ciencias del Mar, UAS, 15: 7-15.

Carballo, J. L.: Hernández-Zanuy, A.; Naranjo, S. A.; Kükürtzü, B. and García, A. (1999). Recovery of *Ecteinascidia turbinata* Herman 1880 (Ascidiacea: Perophoridae) populations after different levels of harvesting on a sustainable basis. Bulletin of Marine Sci., 65: 755-760.

Carballo, J. L.; Naranjo, S.; Kukurtzü, B.; De La Calle, F. and Hernández-Zanuy, A. (2000). Production of *Ecteinascidia turbinata* (Ascidiacea: Perophoridae) for obtaining anticancer compounds. Journal of the World Aquaculture Society, 31(4): 481-490.

Chavanich, S.; Koeysin, P.; Viyakarn, V., Piyatiratitivorakul, S.; Menasveta, P.; Suwanborirux, K. and Poovachiranon, S. (2005). A tunicate from a Thai coral reef: a potential source of new anticancer compounds. Coral Reefs, 24: 621.

Cuevas, C.; Pérez, M.; Martín, M. J.; Chicharro, J. L.; Rivas, C. F.; Flores, M.; Francesch, A.; Gallego, P.; Zarzuelo, M.; de la Calle, F.; García, J.; Polanco, C.; Rodríguez, I. and Manzanares, I. (2000). Synthesis of Ecteinascidin ET-743 and Phthalascidin Pt-650 from Cyanosafracin. Bulletin of Organic Letters, 2 (16): 2545-2548.

Garcia-Rocha, M.; Garcia Gravalos, M. D. and Avila, J. (1996). Characterization of antimitotic products from marine organisms that disorganize the microtubule network: Ecteinascidin 743, isohomohalichondrin-B and LL-15. British J. of Cancer, 73: 875–883.

Hernández-Zanuy, A. C.; Carballo J. L.; García-Cagide, A.; Naranjo, S. and Esquivel, M., 2007. Distribución y abundancia de *Ecteinascidia turbinata* Herdman (Ascidiacea-Perophoridae) en Cuba. Revista de Biología Tropical, 55 (1): 247-254.

Jimeno, J. M.; Faircloth, G.: Cameron, L.; Meely, K.; Vega, E.; Gomez, A.; Fernandez-Sousa J. M. and Rinehart, K. (1996). Progress in the acquisition of new marine-derived anticancer compounds: Development of Ecteinascidin-743 (ET-743). Drugs of the Future, 21: 1155–1165.

Moss, C.; Green, D. H.; and Perez, B. (2003). Intracellular bacteria associated with ascidian *Ecteinascidia turbinata*: phylogenetic and in situ hybridization analysis. Marine biology, 143: 99-110.

Naranjo S. A.; Kukurtçu, H. B.; Barbero, C.; Martin, S. and Carballo, J. L. (2001). Aquaculture of *Ecteinascidia turbinata* Herdman, 1880 as source of marine anticancer agents. In: Biology of Ascidians, L. Charles & Y. Hideyoshi (eds.). Springer-Verlag.

Pharmamar (2008). "http//www.pharmamar.com"

Rinehart, K. H. (2000). Antitumor compounds from tunicates. Medicinal Research Review, 20(1): 1-27.

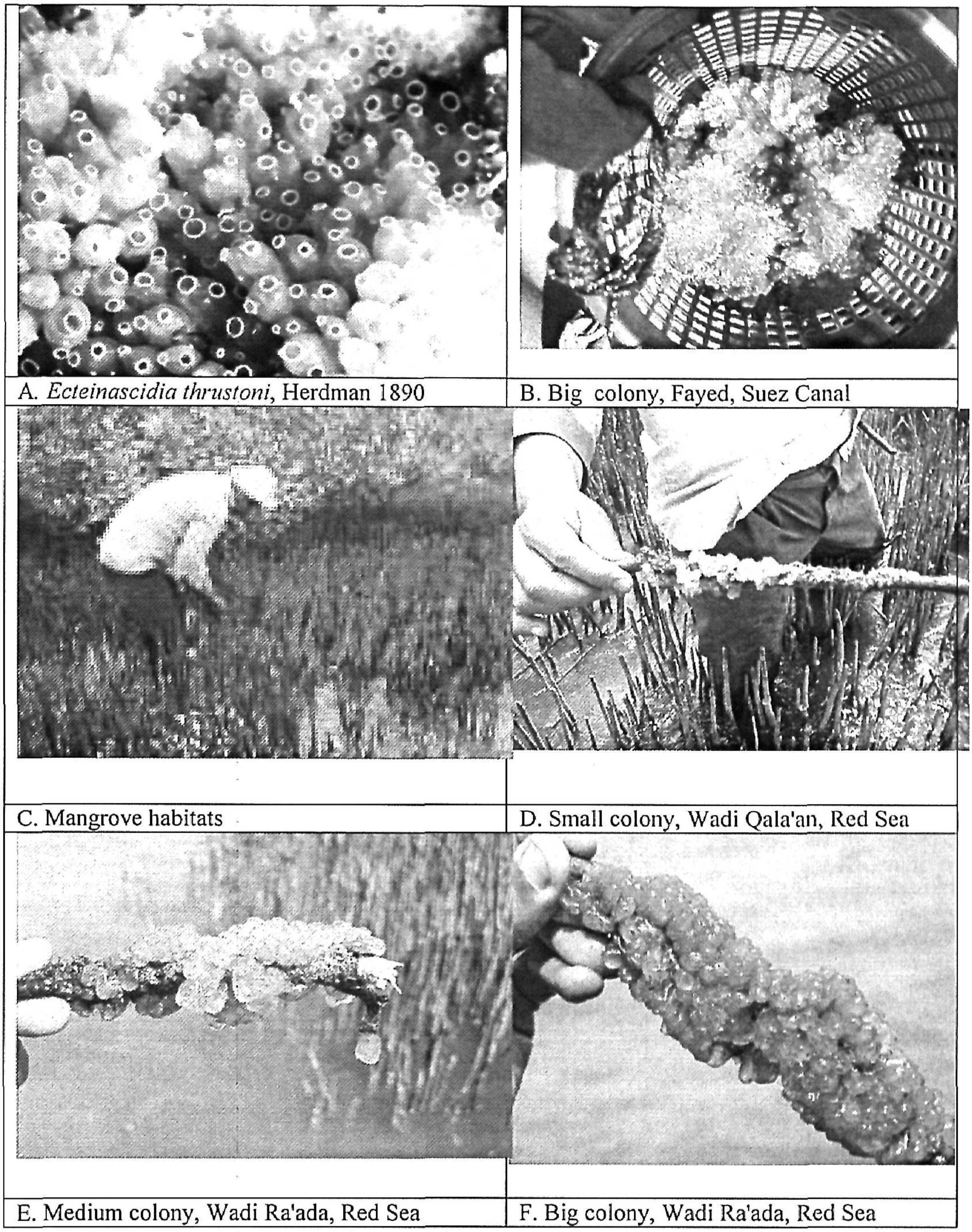
Rinehart, K. H.; Holt, T. G.; Fregeau, N. L.; Stroh, J. G.; Keiper, P. A.; Sun, F.; Li, L. H. and Martin, D. G. (1990). Ecteinascidins 729, 743, 745, 579A, 579B and 770. Potent antitumer agents from the Caribbean tunicate *Ecteinascidia turbinata*. J. Organic Chemistry, 55: 425–455.

Satoh, N. (1994). Developmental biology of ascidians developmental and cell biology series. Cambridge University Press. The Edinbburgh Building, Cambridge, UK.

Suwanborirux, K.; Charupant, K.; Amnuoypol, S.; Pummangura, S.; Kubo, A. and Saito, N. (2002). Ecteinascidins 770 and 786 from the Thai Tunicate *Ecteinascidia thurstoni*. Natural Product, 65 (6): 935-937.

Young C. M. and Bingham, B. L. (1987). Chemical defence and aposematic coloration in larvae of the ascidian *Ecteinascidia turbinata*. Marine Biology, 96: 539–544.

Youssef, D. T. A. (2007). Chemistry part. In: Symbionts and chemistry of the ET-743 producing tunicate *Ecteinascidia turbinata*. Annual progress Report #2 (unpublished), M. G. Haygood, A. A-F. Gab-Alla (eds.), presented to US-Egypt Science and Technology Joint Fund, Grant Number, BIO8–002–006.



A. *Ecteinascidia thrustoni*, Herdman 1890

B. Big colony, Fayed, Suez Canal

C. Mangrove habitats

D. Small colony, Wadi Qala'an, Red Sea

E. Medium colony, Wadi Ra'ada, Red Sea

F. Big colony, Wadi Ra'ada, Red Sea

Fig. 2. Colonies of *E. thurstoni*, collected from Suez Canal and Red Sea.