

Identification of sponges of inter tidal zone in North of Hengam Island, Persian Gulf

*¹S. Eisapor; ²Sh. Safaeian

¹ Young Researchers and Elite Club, Miyaneh Branch, Islamic Azad University, Miyaneh, Iran

² Faculty of Marine Science and Technology, North Tehran Branch, Islamic Azad University

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ABSTRACT: In this study marine sponges of inter tidal zone from Hengam Island Persian Gulf were collected by wading at low tides in Eskeleh station at locations 55° 53' 40" E & 26° 40' 53" N. For identifying identification of sponge samples, acid digestion method as a method of early detection and microtome section to identify the skeletal structure were used. All the identified sponges were from class Demospongia, two orders (Hadromerida, Haplosclerida) and four families (Clionidae, Callyspongiidae, Chalinidae, Niphatidae) and seven species *Amphimedon viridis*, *Haliclona rosea*, *Haliclona cinerea*, *Siphonochalina sp.*, *Callyspongia fallax*, *Callyspongia sp.*², *Cliona dioryssa*. This Research is the exact study to the identification sponges in basis of microtome sectioning to observe skeletal structures in Iranian Island.

Keywords: Marine sponges; Demospongia; Hengam Island; Persian Gulf

INTRODUCTION

The phylum porifera or pore-bearers, embraces a group of strange creature that were not placed definitely in the Animal Kingdom until the first half of the nineteenth century (Kotpal, 1996). They are the most primitive of multicellular animals, metazoan (Hooper, 2000; Hickman *et al.*, 2001). The porifera may be characterized as sedentary, aquatic, mostly marine, solitary or colonial, radially symmetrical or asymmetrical, multicellular organisms; without definite organs, systems, mouth and nervous tissue; with a porous body permeated with pores, canals and chambers (Kotpal, 1996). Sponges are often among the most important ecological groups of coastal marine ecosystems, in terms of number of species and biomass. Demospongiae, in particular, can be found in a wide range of depths, from shallow waters to the abyssal zone (Bergquist, 1978). Identification of sponges is important because we must know the species of sponge from which the "magic molecule" was discovered. Good taxonomy underpins every other branch of biological and biochemical science (Hooper, 2000).

In the literature there are about 7,000 "valid" species published worldwide, but estimated that there are at least 15,000 living species in all the world's seas and lakes. In Australia there are about 1,400 species

described in the scientific literature, but estimated that there are probably at least 5,000 species living in continental and territorial waters. Documenting and describing biodiversity is a long, time consuming process that requires accuracy and patience, but it is an essential prerequisite to conservation and management of our marine resources. There are approximately 1200 described species of sponges known from the South China Sea region (Hooper, 2000).

Skeletal structures of sponges are spicules and Spongin fibres. Spicules are formed by carbonates of lime or silica in the form of needle like pieces. Spongin fibres are composed of a silk-like scleroprotein. On the basis of the material they are formed of, spicules are of two types: Calcareous, made of calcium carbonate and Siliceous. According to the size, spicules are classified into two major types: Megascleres, which are large-sized and constitute main supporting framework of sponge body and Microscleres, which are smaller in size and occur in the mesenchyme. Based on the number of axis present in the rays spicules may be of three types: monoaxon, triaxon and polyaxon (Chandra, 2013).

Monaxon: These spicules grow along a single axis. These may be straight needle-like or rod-like or may be curved. Their ends may be pointed, knobbed or

*Corresponding Author Email: m_kerdgar@yahoo.com

hooked. If spicule has only one pointed end it is called Monoactinal. If there are two points it is called Diactinal. Amphidisc are spicules which have disc at both ends (Chandra, 2013). Demospongiae is the largest and most diverse class of the Porifera. It unites sponges with siliceous spicules (Hooper, 2000).

Nine species of marine sponges were reported from Kuwait (Soest, 2008) and about hundred species were reported from coasts of Oman (Soest and Beglinger, 2002). There is very limited information for the sponge fauna growing in the coastal and deeper waters of the Iranian coast of Persian Gulf, only from western and eastern coasts of Hengam Island ten species and four genera of sponges have been reported (Sadeghi *et al.*, 2007). Also six species of sponges have been identified from intertidal zone of Nayband bay (Safaeian *et al.*, 2009).

MATERIALS AND METHODS

In this study marine sponges were collected from intertidal zone Hengam Island for the purpose of improving information about sponges of Iranian coast of Persian Gulf and discovering biological and chemical resources as well as matters of ecology and conservation management.

Hengam Island located in the Hormoze strait mouth, Iran; which is between latitudes 26° 36' 43"- 26° 41' 15" N and longitudes 55° 54' 40"- 55° 54' 55" E.

Sponge specimens were collected by wading at low tides in Eskehleh station along the Hengam Island in intertidal zone from August 2011 (Fig. 1). Specimens were photographed in situ. After that they were rinsed in seawater and transferred to 70% ethanol, subsamples were collected for biological activity studies. Usually sponge identifications require two forms of histological preparation: one a spicule preparation (for those species with a mineral skeleton); and second, a perpendicular section through the sponge tissue, to determine the structure of the skeleton, the water-canal system, and other aspect of histology (Hooper, 2000).

Acid digestion

For microscopical studies of spicules, acid digestion method was carried out (Hooper, 2000; Soest *et al.*, 2006). Fragments of sponges were placed in centrifuge tubes. Several drops of acid nitric poured or pipetted into tubes till it covered the small fragment in the tube. They were heated gently over a flame until bubbling and all organic matter is digested, this procedure continued until liquid became clear and pale yellow. Then tubes were put in centrifuge and spined 3-5 min at 3000 rpm. After five times repetition spicules pipetted on to warming slides and allowed to dry. After that slides studied and photographed under microscope.

Section preparations

Fragments of preserved sponges were passed through a dehydration series, cleared in toluene, and wax embedded for at least 2 hours. Alternatively, fixed samples can be processed directly in paraffin embedding, on 2 hour. Sections cut from trimmed wax blocks, cutting from the center of the block to the exterior so as to include both the outer surface and inner skeleton relatively intact. For most species relatively thick sections are required (>50µm) Sponges' thinner sections are preferable. Cut sections are placed in clearing agent for an adequate period to dissolve wax and clear the 'tissue', then soaked in - ethanol (perhaps clearing and dehydrating several times until perfectly clear, and/or dewaxing on a hot plate), floated onto slides, orientated and flattened, and mounted (Hooper, 2000; Soest *et al.*, 2006). After that slides studied and photographed under microscope. After major characters about Sponges samples (spicules type and size, skeleton) were collected and studied in the lab, samples were identified by using "Guide to Sponge Collection and Identification" (Hooper, 2000). Then samples were sent to Zoological Museum of Amsterdam and confirmed by Dr. Soest.

Description Order and Family of the Sponges of Hengam Island, Iran:

Definition of Order Haplosclerida:

Main skeleton is partially or entirely composed of an isodictyal reticulation of sponging fibers and/or spicules, with uni- to multispicular tracts of diactinal spicules forming triangular, rectangular or polygonal meshes; megascleres are exclusively oxete or strongylote, bonded together with collagenous spongin or enclosed within spongin fibers; microscleres, if present, may include sigmas (frequently centrangulate), smooth toxas or microxeas. Nine families of sponges are included, seven of which are viviparous, with parenchymella bearing various patterns of ciliation, one oviparous group (Petrosiidae), and one uncertain (Lubomirskiidae) (Hooper, 2000).

Definition of Family Niphatidae:

Encrusting, massive, fan-shaped, vase-shaped and branching growth forms, often with chimney-like oscular processes; ectosomal skeleton consists of a dense multispicular, three-dimensional, paratangential reticulation of diactinal spicules (oxeas or strongyles), usually more compact than the choanosomal skeleton; erect spicule brushes characteristically at the surface; choanosomal skeleton a reticulation of ascending and transverse-connecting spongin fibers, cored by multispicular tracts of oxeas; interstitial spicules also common;



Fig. 1: Location of sampling station on the coasts of Hengam Island

microscleres, if present, are sigmas or microxeas. Fourteen nominal genera, seven of which are probably valid (Hooper, 2000).

Definition of Family Chalinidae:

Encrusting, massive, cup-shaped, fan-shaped and branching growth forms, usually with spongy and delicate consistency; when present ectosomal skeleton consists of a special, tangential, unilayered, unispicular, isotrophic reticulation of oxeas bound by nodal spongin; choanosomal skeleton consists of an isodictyal reticulation of uni- or paucispicular primary tracts of oxeas, rarely multispicular, interconnected by uni- or paucispicular secondary tracts, and spicules are bonded together at their nodes of junction by small amounts of collagenous spongin, or they may be fully enclosed within light spongin fibers and form more robust reticulations; microscleres, if present, include only sigmas or toxas; parenchymella larvae are incubated and are completely and uniformly ciliated or have a bare posterior cap fringed by longer cilia. There are 26 nominal genera but only 13 of these are probably valid (Hooper, 2000).

Definition of Family Callyspongiidae:

Encrusting, massive, vase-shaped, tubular, fan-shaped and branching growth forms; surface characteristically sculptured with conules or ridges, and usually has an optically visible lace-like reticulation of spicules and/or fibres lying tangential to the surface; ectosomal skeleton a two dimensional tangential reticulation of close-set primary, secondary and sometimes tertiary sponging fibres, sparsely cored with small or vestigial oxeas or strongyles; choanosomal skeleton more widely spaced, composed of a reticulation of primary ascending (bi- or multispicular) and secondary connecting spongin fibres (uni- or aspicular), composed of well-developed fibers, cored by oxeas or strongyles;

spongin characteristically abundant; megascleres sometimes vestigial, with blackened axial canals, absent entirely or replaced by sand grains; microscleres, if present, include only toxas.

There are 21 nominal genera of Callyspongiidae, including many names created by Lendenfeld, but only five of these may be valid (Hooper, 2000).

Definition of Order Hadromerida:

Relatively cohesive order with uniform speculation of monaxonid megascleres; with radially arranged skeleton always obvious at surface if not within choanosome; spongin fibres poorly developed (if at all present); ectosomal spicules typically smaller than choanosomal spicules, usually standing perpendicular to surface and protruding through ectosome; microscleres, if present, euasters, streptasters- and derivatives, spirasters or spiraster-like spirules, or peculiar asterose-like discorhabds; all groups oviparous (where known), with development of parenchymella larva (in one case blastula larva) directly in seawater. Twelve families presently included (Hooper, 2000).

Definition of Family Clionidae:

Hadromerida displaying three different stages of growth: excavating (alpha), encrusting (beta) or massive (gamma). Tylostyles, styles or oxeas as megascleres and streptasters and/or microrhabds of different types as microscleres. The skeletal arrangement varies depending on the growth form of the species or specimens. The choanosomal skeleton is confused in specimens in the alpha stage and confused and arranged in 'ill-defined' tracts in the beta and gamma stages. The ectosomal skeleton consists of a cortex of megascleres arranged in palisade. In alpha-stage specimens the cortex is only present in the papillae. Microscleres can appear on the top of papillae, but they never constitute a well differentiated layer. Two functional types of papillae,

inhalant and exhalant, are present. All the species included in this family are able to excavate calcareous substrata, where they build galleries following a species-specific pattern. Some species in the alpha stage are able to spread over the substratum by lateral growth of the ectosome next to the papillae, reaching a beta or even a gamma stage of growth. Fifteen nominal genera of Clionids have been created, but only four of these may be valid (Hooper, 2000).

RESULTS AND DISCUSSION

Name of all species, of collection is presented in Table 1.

Table 1: Identified marine sponges in Hengam Island.

| Sample No. | Order | Family | Species | Key Features |
|------------|---------------|-----------------|-----------------------------|---|
| 1 | | Niphatidae | <i>Amphimedon viridis</i> | blue, Rocky substrate, Thumbnail growth, Oxeas, Strongyles |
| 2 | | Chalinidae | <i>Haliclona cinerea</i> | blue-cram, Rocky substrate, Oxeas, Sigmas |
| 3 | | | <i>Haliclona rosea</i> | Red, The coated substrate, three Oxeas |
| 4 | | | <i>Callyspongia sp.</i> | Cream, rocky bed, fan shape, Oxeas |
| 5 | | Callyspongiidae | <i>Siphonochalina sp.</i> | Light cream, rocky substrate, a cylindrical growth, Oxeas Microxeas |
| 6 | Haplosclerida | | <i>Callyspongia flallax</i> | Cream, Horizontal expansion, into plant tissue lattice, Oxeas, Strongyles |
| 7 | Hadromerida | Clionidae | <i>Cliona dioryssa</i> | Orange, bore into lime stone substrate, Tylostyles +Oxeas |



Fig. 2: *Amphimedon viridis*, size 12×8×4 cm.

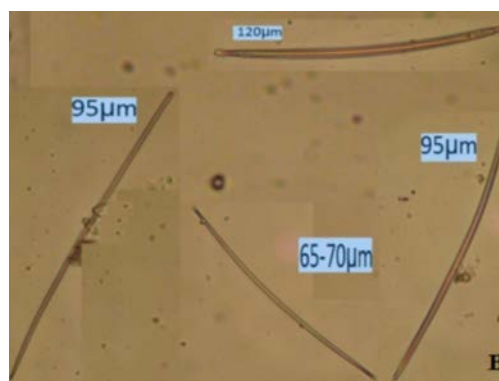


Fig. 3: A) Skeletal structure *Amphimedon viridis* Magnitude: ×400, B) Megascler, Monoaxon spicule, Oxea and Strongyles.

Sample NO.2

Class: Demospongiae

Order: Haplosclerida

Family: Chalinidae

Species: *Haliclona cinerea* (Grant, 1826)

Description of Species *Haliclona cinerea*:

It's a very variable species, forming blue-cram encrusting sheets with rounded lobes to tall chimney-

like growths with large terminal oscules. It is very soft and when torn has characteristic and diagnostic "slime strands". It occurs intertidally under stones. Sponge size was 15×8×7 cm. Encrusting form, with spongy and delicate consistency (Fig.4). Megascler short, rather thick oxea of two different lengths and microscler was simga. A regular structure of spicules, which are connected by spongin at the nodes (Fig. 5).



Fig. 4: *Haliclona cinerea*

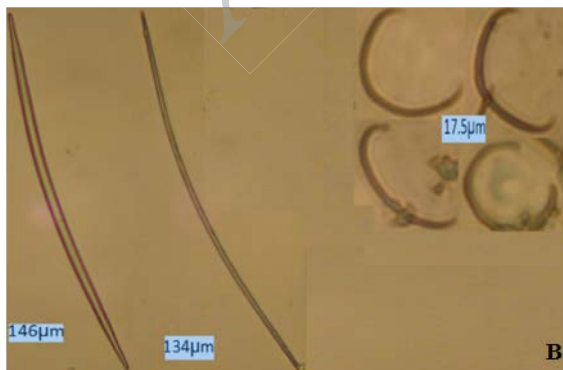
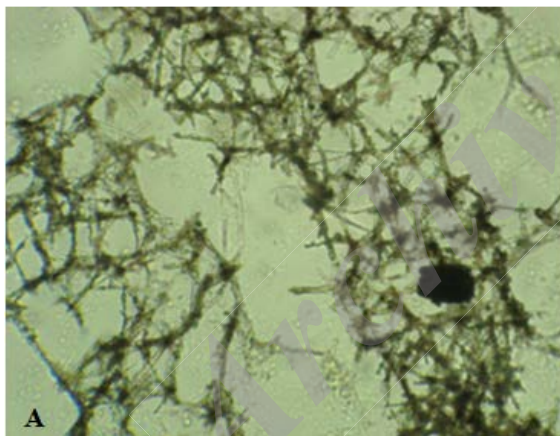


Fig. 5: A) Skeletal structure *Haliclona cinerea*, Magnitude: ×400, B) Megascler, Oxea and 4 types of microscler sigmas.

Sample NO.3

Class: Demospongiae

Order: Haplosclerida

Family: Chalinidae

Species: *Haliclona rosea* (Bowerbank, 1866)

Description of Species Haliclona rosea:

It was rose-pink, thinly encrusting sponge with scattered, slightly elevated, with tubular oscules. It was megascler Oxea in different size (Fig. 6).

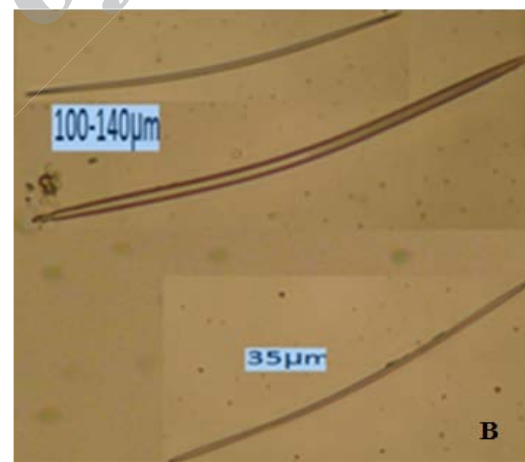


Fig. 6: A) *Haliclona rosea*, B) Oxea megascler

Sample NO.4

Class: Demospongiae

Order: Haplosclerida

Family: Callyspongiidae

Species: *Callyspongia sp.* (Duchassaing and Michelotti, 1864)

Description Species Callyspongia sp.:

Its live color was cream. Sponge size was 10×10×5 cm. fan-shaped growth form (Fig. 7). Megascler was oxeas in two short sizes. Skeletal structure composed of thick fibers lying tangential to the surface (Fig. 8).



Fig. 7: *Callyspongia sp2*.

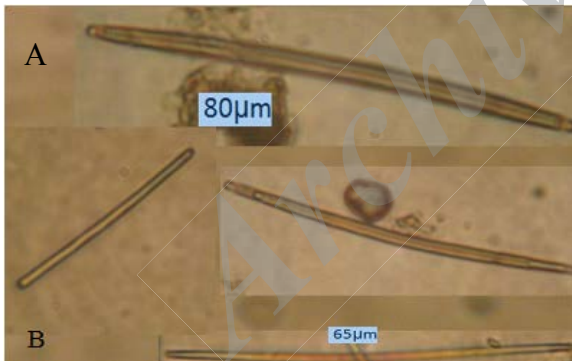


Fig. 8: A) Skeletal structure *Callyspongia sp2*, Magnitude: $\times 400$, B) Megascler, Oxea

Sample NO.5

Class: Demospongiae

Order: Haplosclerida

Family: Callyspongiidae

Species: *Siphonochalina sp.* (Schmidt, 1868)

Description Species Siphonochalina sp.:

Its live color was cream. Its size was 12 \times 5 \times 7 cm. tubular growth form (Fig. 9). Megascler was oxeas and microscler was microoxea. A microtome section of

skeletal has fiber networks with oval forms cored by oxeas. (Fig. 10)



Fig. 9: *Siphonochalina sp.*

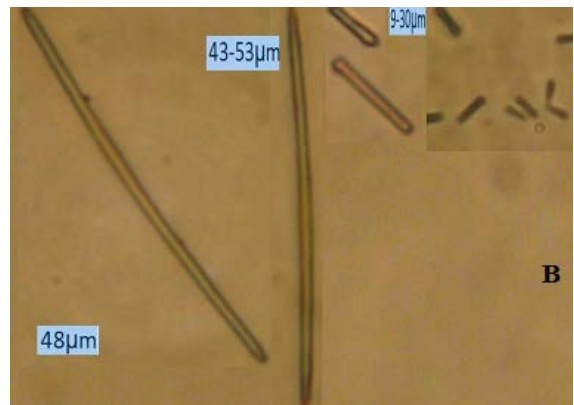


Fig. 10: A) Skeletal structure *Siphonochalina Sp.*, Magnitude: $\times 400$, B) Megascler, Oxea and microscler, microoxea

Sample NO.6

Class: Demospongiae

Order: Haplosclerida

Family: Callyspongiidae

Species; *Callyspongia flallax* (Duchassaing and Michelotti, 1864)

Description Species *Callyspongia fallax*:

Lobate, repent masses with a tendency to form upright, short, anastomosing tubes. Individual lobes 2-3 cm in diameter, tubes up to 4 cm high. Oscules and vents apical, up to 12 mm in diameter, flush thin collars, Surface smooth and Consistency firmly spongy. (Fig. 11, 12)

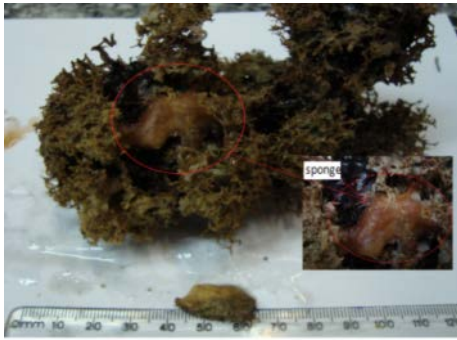


Fig. 11: *Callyspongia fallax*

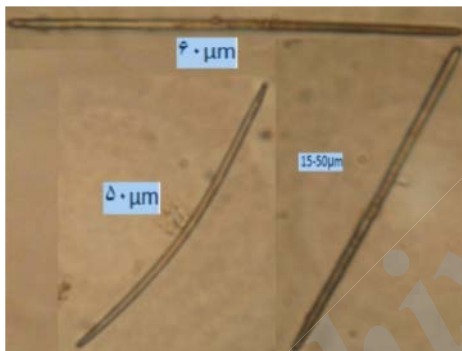


Fig. 12: *Callyspongia fallax*, Oxea Megascler

Sample NO.7

Class: Demospongiae

Order: Hadromerida

Family: Clionidae

Species: *Cliona dioryssa* (Topsent, 1925b)

Description of Species *Cliona dioryssa*:

It was orange, very small and size was 2×2×1 cm. This species is visible as scattered papillae, protruding through the surface of corals, or as incrustations, that actually bore into coral heads. A variety of different Tylostyles as megascleres (Fig.13). Microtome cutting was show a regular structure of megascleres (Fig.14).

In this study all identified sponges were from class Demospongia, two orders (Hadromerida, Haplosclerida) and four families (Clionidae, Callyspongiidae, Chalinidae, Niphatidae) and seven species from North Hengam Island consist of: *Amphimedon viridis*, *Siphonochalina sp.*, *Callyspongia sp.2*, *Callyspongia fallax*, *Haliclona rosea*, *Haliclona cinerea*, *Cliona dioryssa*.

Until now there is only one report of marine sponges in the east and west borders of Hengam Island, Iranian water of Persian Gulf at 5 to 20 meters depths (Sadeghi, et al., 2007). Among mentioned marine sponges in our article and report from Hengam Island some similarities are obvious. Two orders (Hadromerida, Haplosclerida), three families (Callyspongiidae, Chalinidae, Clionidae) and three genera (*Callyspongia*, *Cliona* and *Haliclona*). Only one species of *Haliclona cinerea* are similar. Also, research in the North Persian Gulf from Nay Band Bay and Bustaneh was performed (Safaeian, S, et. al, 2009); Samples of the two orders (Hadromerida, Haplosclerida) and three families (Clionidae, Chalinidae, Niphatidae) and genera (*Haliclona*) are similar. But the species are different, species of our study were, *Haliclona rosea* and *Haliclona cinerea*.

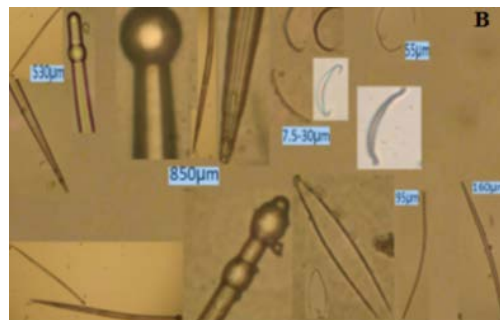


Fig. 13: A) *Cliona dioryssa*, B) Megasclers monoaxon spicule, Tylostyle.



Fig. 14: Skeletal structure of *Cliona dioryssa* Magnitude ×200.

These differences are most likely due to the depth and differences in localities and beds. Then species and genera of this study are reported for the first time in the Iranian coast of Persian Gulf.

But according to the above two studies and the current study, we can conclude that most sponges in the North waters of the Persian Gulf has belonged to the class Demospongia. Probably two orders of Haplosclerida and Hadromerida are common in Persian Gulf coasts and Islands. Also we can say that two families Clonidae and Chalinidae are adaption with temperature and salinity of Persian Gulf and have been able to expand their Species. Each three research are similar in *Haliclona* species and this resemblance indicative acclimation *Haliclona* species with Persian Gulf Conditions. There is not report on the presence of the 7 species identified in the Persian Gulf and Oman Sea.

Callyspongia flallax, *Cliona dioryssa* and *Amphimedon viridis* are reported from coastal waters of Florida, Bahamas and Caribbean. (Soest, 2008)

Haliclona cinerea is reported from coastal waters of British Isles: wide-spread in Ireland, West coast of Scotland, Channel Islands, France, reaching south to the Mediterranean, Azores, Madeira and West Africa (Senegal) (Hooper and Soest, 2002).

Haliclona rosea is reported from coastal waters of Davis Strait, south-west and east coast Greenland, Iceland, Spitsbergen, Murmansk, Norway, Denmark, Swedish west coast, the Netherlands, British Isles, France. (Hooper and Soest, 2002).

Since from the Persian Gulf Waters *Haliclona* (*Reniera*) *toxius* is reported from coastal waters of Nay Band Bay (Safaeian, et al., 2009) and *H. oculata*, *H. sp.*, *H. cinerea*, *H. simulans* and *Cliona lobata* are reported from east and west boarders of Hengam Island (Sadeghi, et al., 2007).

As one hundred species of sponges have been recorded only from coasts of Oman (Soest and Beglinger, 2002), and considering these primary steps in Iranian marine sponge identification, Iranian scientists can expect many sponge species in Persian Gulf. It seems very important to investigate Iranian coast of Persian Gulf in an organized project. The last but not the least important benefit in sponge studies is looking for the secondary metabolites from Iranian marine sponges which can be considered as natural marine drugs.

It seems to *Haliclona* Adopted of Persian Gulf condition. *Siphonpchalina sp.*, *Haliclona rosea*,

Cliona dioryssa, *Haliclona rosea* and *Callyspongia flallax* are reported for the first time of Persian Gulf and Hengam Island inter-tidal zone.

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