Biodiversity is fundamental to both eukaryote and prokaryote ecology (Hughes et al., 2001; Ward, 2002). Coral reefs are tropical, shallow water ecosystems known for its exceptionally diverse flora and fauna, complex food web and trophic organization. It occupies 0.2% of the ocean area (Kleypas, 1997). They are the most biologically productive and diverse of all natural ecosystems, the high productivity stemming from their efficient biological recycling, high retention of nutrients, protection of coastlines from erosion and their structure which provides habitat for a vast array of other organisms. The greatest significance of reefs lies in the fact that they generate and maintain a substantial proportion of tropical marine biodiversity. Thus their influence is global and multifaceted (Opdyke, 1992). India has a wide geographical distribution of oceanic atolls and fringing coral reefs. Lakshadweep island reefs and Andaman and Nicobar Islands are oceanic while that of Gulf of Kutch, Gulf of Mannar, and Palk Bay are fringing shelf reefs. Coral reefs form the habitat of a vast array of animal phyla including porifera, coelenterates, helminthes, annelids, arthropods, molluscs, echinoderms and chordates (Roberts, 2002). Among the different phyla, Porifera (sponges) represent a significant component of the coral reef ecosystem. About 486 species of sponges have been described in the Indian waters (Thomas, 1998). The Gulf of Mannar and Palk Bay have the highest diversity (319 species) followed by Andaman and Nicobar islands (95 species), Lakshadweep (82 species) and Gulf of Kutch (25 species) (Venkataraman and Wafar, 2005). The distribution of the sponges in the Indian coral reefs is given in Figure 1.
Figure 1. Distribution of sponges in the coral reefs of India (Venkataraman and Wafar, 2005)
Sponge (phylum Porifera), a sessile benthic invertebrate, represent a significant component of the deep water and shallow water communities especially of coral reefs (Dayton, 1974; 1989). They are the simplest and evolutionarily ancient metazoans lacking specialized tissues and organs that had first appeared in the Precambrian (Li et al, 1998). They are called as the “pore bearers” and include three extant classes or sublineages namely, Hexactinellida, Demospongiae and Calcarea and one completely extinct class, Archaeocyatha (Borchiellini et al, 2001). The phylum encompasses 6000 taxonomically validated species. Sponges have been excellent source for natural products that are bioactive compounds and have been reviewed by Faulkner (2001). Sponges are active filter feeders. About 24,000 liters of water can be pumped through a one kilogram sponge in a single day (Vogel, 1977). This remarkable filter feeding ability enables it to efficiently take up nutrients like organic particles and microorganisms from the sea water (Reiswig, 1974; Pile, 1997; Wehri et al, 2007). The plasticity of sponge trophic ecology and intimate associations of the sponges with the symbiotic microbes may be among the few factors contributing to the worldwide ecological success despite large spatial and temporal variations in food sources (Taylor et al, 2007).

Sponges represent an ecological niche that harbours a largely uncharacterized microbial diversity which includes heterotrophic bacteria, archaea, cyanobacteria, green algae, red algae, diatoms and dinoflagellates (Garson et al, 1998; Cerrano et al, 2003; Hentschel et al, 2006). Earliest studies of sponge-bacterial associations date back to the 1970s where bacterial populations were observed using electron microscopy (Vacelet and Donadey, 1977; Wilkinson, 1978; Friedrich et al, 2001; Webster and Hill, 2001). These studies recognized three broad types of microbial associates in the sponges namely (i) abundant populations of sponge-specific microbes in the sponge mesohyl (ii) small populations of specific bacteria occurring intracellularly and (iii) populations of non-specific bacteria resembling those
in the surrounding sea water. Bacteria form about 40-60% (10^8 to 10^{10} bacteria g^{-1}) of sponge biomass. The advent of molecular techniques over the past decades has revolutionized our understanding of microbial diversity and function. It is apparent that sponge-associated communities are highly diverse, with a range of different microorganisms consistently associated with the same host species and can be evolutionarily ancient or recently initiated relationship involving microorganisms which are present in the surrounding sea water. Several bacteria belonging to Actinobacteria, Bacteroidetes, Cyanobacteria, Firmicutes, Planctomycetes, Proteobacteria and Verrucomicrobia have been isolated in pure culture from marine sponges (Burja and Hill, 2001; Hentschel et al, 2001; Pimental Elardo et al, 2003; Kim et al, 2005; Sfanos et al, 2005). Studies based on 16S rRNA gene sequence analysis revealed information on the enormous phylogenetic diversity of the sponge-associated bacteria (Hentschel et al, 2001, 2002, 2006; Webster et al, 2001; Olson and McCarthy, 2005). Several studies reported that the sponge-associated bacteria were assigned to different bacterial phyla (Althoff et al, 1998; Lopez et al, 1999; Hentschel et al, 2003, 2006; Taylor et al, 2005; Enticknap et al, 2006; Thiel et al, 2007). Host-associated bacterial communities are potentially critical components of marine microbial diversity, yet our understanding of bacterial distribution on living surfaces lags behind that of planktonic communities.

Although bacteria are a food source for sponges, not all bacteria are broken down for food and some bacteria form symbiotic relationships. These relationships are involved in nutrient acquisition, stabilization of the skeleton, metabolic waste processing and production of secondary metabolites. Also there is increasing evidence that these microorganisms might be involved in the secondary metabolism which was originally attributed to the host (Haygood et al, 1999). Numerous studies have been carried out on the antimicrobial activity of sponge and sponge-associated bacteria from different regions (Kelman et al, 2001; Pabel et al, 2003; Kelly et al, 2005) for biotechnological applications.
This close association of the sponges with microorganisms has been one of the rationales for these primitive metazoans being the focus of recent interest in addition to being an excellent resource of biologically active metabolites (Taylor et al, 2007). However, limited studies have been carried out on functional aspects of the associated microbes. Hence an attempt was made to study the role of hydrolytic enzymes in the nutrition of the host (Feby and Nair, 2010). Despite, the importance of sponges in a number of ecosystems and the vast array of novel bioactive compounds that have been isolated from them, we still lack a clear picture of the microbial diversity and the factors which influence its hosts.

Of all the described classes of sponges, the Class Demospongiae is the most abundant, forming 85% of the sponges (Hooper and van Soest, 2002) and their association with large microbial consortia is well recognized (Hentschel et al, 2003, 2006; Hill, 2004; Imhoff and Stohr, 2003). Similarly, Demosponges are reported as the most abundant sponges in the Indian waters (Venkataraman and Wafar, 2005). Most of the studies were to explore microbes for bioactive compounds and enzymes (Thakur and Anil, 2000; Mohapatra et al; 2003; Selvin et al, 2004, 2009a; Thakur et al, 2005; Anand et al, 2006, Feby and Nair, 2010). Studies on the diversity of sponge-associated microbes from Indian waters are very limited (Selvin et al, 2009b). Besides, very little is known about the spatial and temporal variations in diversity and functional aspects of sponge-associated microorganisms.

Therefore, a study was carried out to estimate the bacterial diversity associated with two species of Demosponges - *Sigmadocia fibulata* and *Dysidea granulosa*, common and widely distributed tropical sponges of the shallow waters in the coral reef ecosystems of Lakshadweep and Gulf of Mannar. In the present study, a combination of traditional cultivation and molecular approaches has been used to emphasize similarities and differences in the composition of the bacterial community of the two
sponges. In addition, the metabolic versatility and antimicrobial activities of the sponge-associated bacteria were also studied to understand the functional role of these bacteria. This study is the first to incorporate both the culture-dependent and culture-independent investigation of the bacterial assemblages associated with sponges in Indian waters.