INTRODUCTION

"The fungi are progressive, ever changing and evolving rapidly in their own way, so that they are capable of becoming adapted to every condition of life. We may be rest assured that as green plants and animals disappear one by one from the face of the globe, some of the fungi will always be present to dispose of the last remains."

- B.O. Dodge (1872–1960)

Fungi are achlorophyllous, eukaryotic microorganisms which grow as single cells or multicellular filaments, acquiring nutrition by absorption from the surrounding organic substrates. None of the biological matters can be free of fungal association (Maheshwari, 2005). There are fungi in soil, plant litter, decaying woods, submerged plant remains in stream or ponds, driftwoods in seawaters, on insects, with nematodes, on herbivore dung and so on (Dix and Webster, 1995). Based on their associations, fungi are considered with distinctive roles as saprophytes, parasites, endophytes or mutualists (Kendrick, 1992).

Fungi grow everywhere and in all sorts of habits and habitats. The necrotrophic and biotrophic fungi cause diseases in plants, insects, birds and animals. A small percentage of the fungi live in symbiotic association with plants or animals (Kendrick, 1992). The saprotrophic fungi are the primary agents responsible for decomposition of organic matter. These fungi bring about spoilage of food and damage fabrics, paper, leather and other consumer goods. They also grow on dung of all kinds of animals (Dix and Webster, 1995). The fungi are important industrially as they produce enzymes, organic acids, vitamins and antibiotics (Pointing and Hyde, 2001). There are fungi growing in extreme environments such as saltpans or Antarctic soils. The cold and harsh environmental conditions in the polar region aid the growth of psychrophilic and psychrotolerant fungi (Singh et al., 2006). These extremophillic fungi also have biotechnological potential (Puja and Singh, 2011).
Yet, very few fungi have so far been described from such environments (Gunde-Cimerman, 2003).

There have been numerous studies on the diversity, ecology and activities of fungi, around the world (Dix and Webster, 1995; Pointing and Hyde, 2001). Yet, the fungi haven’t been explored all enough particularly in the tropical belt and much more remained to be studied. Over the last two decades, systematic efforts were made in this laboratory to explore the biology of saprophytic fungi found growing in different habitats such as decaying plant litter in soil (Jacob, 2000; D’Souza, 2002; Puja, 2008), submerged plant litter in freshwater streams (Nair, 2002), dead insects (Keshava Prasad, 2004), aerial region of plants (Jalmi, 2006) and internal plant parts (Jacob, 2000; D’Souza, 2002; Puja, 2008). With high humidity, warm temperature, varied vascular plants and dependent herbivore animals, numerous ecological niches and microhabitats, the tropical belt of the world is considered as major reservoir of majority of known and most of the unknown taxa of fungi (Bhat, 2010).

The number of fungi on earth surface has been projected as 1.5 million, roughly six times the estimated number of vascular plants (Hawksworth, 1991). Of these, a mere 5-7% of the fungi are presently described (Bhat, 2010; Hawksworth, 2004; Rossman, 1994). Using conventional microscopic methods and modern molecular techniques, many of the unknown fungi are now detected (Bhat, 2010; Pennisi, 2004; Horton and Bruns, 2001). Based on high-throughput sequencing methods, it has now been estimated that as many as 5 million species of fungi might be present on the earth’s surface (Blackwell, 2011).
Succession of fungi

In order to achieve complete decomposition, one of the major events taking place in any habitat, is succession of microorganisms - be it fungi or bacteria (McIntosh, 1980; Morin, 1999; Dix and Webster, 1995). Most studies so far carried on fungal succession were based on the kind of substrata, viz. terrestrial wood (Lange, 1992), herbivore dung (Richardson, 2001; Kuthubutheen and Webster, 1986; Nagy and Harrower, 1979), hay (Breton and Zwaenepoel, 1991), aquatic wood (Fryar et al., 2002), pine cones (Kasai et al., 1995), wool (Ghawana et al., 1997), sugarcane (Sandhu and Sidhu, 1980), straw (Harper and Lynch, 1985) and living leaves (Wildman and Parkinson, 1979). Species that are lost during the course of succession are often assumed to be victims of competitive exclusion by later occupying species (Farrell, 1991). Analyzing the fungal succession on herbivore dung, Holmer and Stenlid (1997) suggested that the species producing fruiting bodies at later stages being much stronger than those fruiting at early stages, inhibition could be the mechanism driving the successional processes.

Activities:

Fungi elaborate a variety of enzymes and other metabolites which affect the humans, plants and other living forms, in different ways (Barnett & Hunter, 1999). The beneficial aspects of fungal activities have been well recognised (Gupta and Soni, 2000; Mehta and Gupta, 1991; 1992; Nevalainen and Te’o, 2003; Bergquist et al., 2003; Henriksson et al., 2000). All these studies illustrate the potential of fungi in industrial and food fermentation processes (Asan, 2004). With high nutrient value, fruiting bodies of macro-fungi, viz. Lentinus edodes (shiitake), Pleurotus ostreatus
(oyster mushroom), and *Agaricus bisporus* (button mushroom) are now considered as excellent culinary delicacies (Hintz, 1999).

Fungal amylases are used in food, brewing, textile, detergent and pharmaceutical industries. Although alternate sources of amylase (porcine pancreas, human saliva, rat pancreas, malted grains, etc.) exist, a large proportion of amylase production is from fungi (Ali et al., 1989). Fungi have also been attributed for the production of cellulases and the crude enzymes produced by species of *Trichoderma* and *Aspergillus* are commercially available for agricultural practices (Wainwright, 1992). Cellulases find their application in textile industries (Kotchoni et al., 2003) and bio-waste degradation (Mandels and Reese, 1985; Hoffman and wood, 1985).

**Coprophilous fungi:**

The literary meaning of 'coprophilous' is 'dung-loving'. The undigested carbohydrates, hemicelluloses and lignin, along with amino acids, vitamins, growth factors and minerals in the herbivore dung, aid colonization and growth of diverse fungi (Dix and Webster, 1995). They represent a diverse community of morphologically and physiologically specialised mycota which provide a biological force for the decomposition and recycling of animal faeces (Richardson, 2008). The varying fungal components of animal dung are difficult to relate to a specific cause; many fungal conidia or spores are ingested by herbivorous animals while grazing (Larsen, 1971). Some of the ingested propagules might be endophytic, geophilic or phylloplane fungi. The intake of vegetation from a common source, may lead to the occurrence of several common fungi in both ruminant and non-ruminant dung. The composition of dung's mycobiota is influenced by various factors, such as antagonism, exploitation, competition, nutritional factors, and environmental conditions (Caretta,

**Succession of fungi on herbivore dung:**

Rayner and Todd (1979) defined fungal succession on herbivore dung as “the sequential occupation of the dung by thalli (usually mycelia) either of different fungi or of different associations of fungi”. The fungi replace one another as communities of mycelia get altered in space and time (Frankland, 1998). Generally, fungal succession is based on the sequence of sporulation on the substrate. The sequence of occurrence of fungi observed on dung after defecation has been, first by members of the zygomycetes, followed by ascomycetes and basidiomycetes. The zygomycetous fungi utilize simple sugars, starch, and protein (and subsequently sporulate); thereafter Ascomycetes use the hemicelluloses and celluloses (and sporulate), followed by the basidiomycetes using the lignin and cellulose (Fryar, 2002). The tenure required for fungi to fruit is independent of the nutritional status of the dung. Majority of coprophilous fungi start germinating within 6 h of defecation. Thereafter the mycelial growth starts, although the duration of fruiting varies. Thus, the fungal succession is said to be more of mycelial succession. Therefore, the sequence of fungal succession is more a succession of fungal fruiting bodies (Harper and Webster, 1964). The coprophilous fungi exhibit certain adaptive features for dung inhabitation. These included the following:

(i) **Phototrophic nature** and forcible discharge of spores towards light source. The spore-producing structures, viz. sporangiophore of mucorales, conidiophore of hyphomycetes and ascus or basidium of higher filamentous fungi, all get phototropically oriented and eject the spores to relatively long distances (Richardson,
This is an ecological adaptation acquired by majority of dung inhabiting fungi, with which, the spore is thrown high in air which enables the latter to fall on any vegetation. In the next feeding course of herbivores, the dispersed spores along with forage get into the stomach of animals.

(ii) **Adhesive projectiles**: Ascospores are armoured with gelatinous appendages as extension of their spores or a partial sheath. These projectiles enable attachment of spores to the herbages without being washed off by wind or water and retaining viability (Richardson, 2008; Dix and Webster, 1995).

(iii) **Pigmentation in exospores**: The exospores are often pigmented and provide protection against UV exposure (Richardson, 2008).

(iv) **Mucilaginous spores**: The gelatinous ascospores of coprophilous fungi, e.g. *Sordaria* and *Podospora*, favour adherence of the propagules to adjacent vegetation. These are later consumed by herbivores along with the vegetation (Caretta, 1998).

(v) **Resistance to digestive enzymes and acids in animal gut**: Passage of spores through the gut of herbivore animal leads stimulation to germinate, resulting with a vegetative stage, followed by sporulation. The spores get triggered for germination following exposure to the chemical and physical environment of the animal gut (Kuthubutheen and Webster, 1986).

**Significance of coprophilous fungi**

Coprophilous fungi are known to produce antifungal peptides and amino acid-derived metabolites (Gloer and Truckenbrod, 1988; Webber, 1981). Zaragozic acids, a group of potent, broad-spectrum, antifungal metabolites that inhibit squalene synthase were originally obtained from a coprophilous fungus, *Sporormiella intermedia* (Bills, 1994). Novel bioactive metabolites, decipinin A and decipienolides A and B, were
obtained from coprophilous fungus, *Podospora decipiens* (Che et al., 2002). Caretta (1998) observed that studies on coprophilous fungi might lead to the discovery of many more novel bioactive fungal metabolites.

Zygomycetes are considered as good sources of polyunsaturated fatty acids (PUFAs) and a good percentage of Mucorales are coprophilous (Bajpai et al., 1992). These fungi are known to accumulate the lipids intracellularly. In microorganisms, including bacteria, lipid content is usually less than 10% of the dry biomass. However, the mucoraceous fungi have the potential to accumulate lipid in their bodies equivalent to about 50% of dry biomass (Ratledge, 1992). Among the known microbial lipids, PUFAs have attracted great interest because of their nutritive value, especially in child health care. The first trial of PUFA production from mucoraceous fungi, with γ-linolenic acid as the target, was pioneered in the UK and Japan (Higashiyama et al., 2002).

**Present Work:**

While substantial work has been done on coprophilous fungi in other parts of the world, not much is known from India (Kirk et al. 2008) There is hardly any detailed study from the west-coast region of India (Bhat, 2010). Realizing this lacuna in our understanding of the diversity, ecology and prospecting of fungi in this part of the country, a modest beginning was initiated in this Department, on coprophilous fungi (Colgaonkar, 2001; Yadav, 2006). Encouraged with the preliminary results obtained, an elaborate plan was drawn on coprophilous fungi with following objectives:

- Isolation and culturing of fungi appearing on dung of various herbivore animals from different parts of Goa and neighbouring regions of Maharashtra and Karnataka.
- Taxonomic documentation of isolated coprophilous fungi.
- Analysis of pattern of appearance of fungi on cattle and rabbit dung over a period of time and their significance.
- Screening of some of the isolated coprophilous fungi for metabolites, significant from neutra- and pharma point of view.

Results of this study formed the subject matter of this thesis.