ABSTRACT

Aims: Keratinophilic fungi include dermatophytes and a variety of other fungi. Information on their occurrence in several Caribbean countries is scarce. This study investigates occurrence of dermatophytes and other keratinophilic fungi in soils of Anguilla, British West Indies (BWI) and emphasizes its public health significance.

Study design: Place and Duration of Study: The study was done in the Department of Microbiology, Saint James School of Medicine, Anguilla (BWI). One hundred and ten samples of soils from habitats in several localities in Anguilla were examined.

Methodology: The samples were processed by Vanbreuseghem’s hair-bait technique. Bits of fungal growth on hair-bates were cultured on Mycobiotic agar to get pure cultures. Identification of the isolates was accomplished by a detailed study of colonial characters and microscopical features of the isolates.

Results: Eighty-five isolates of keratinophilic fungi were recovered. The identified species included two geophilic dermatophytes, _Micrurus fulvum_ and _M. gypseum_ and two species of _Chrysosporium_, namely _Chrysosporium keratinophilum_ and _C. indicum_, _M. fulvum_ was the
predominant species being recovered from 35 (31.8%) of the soil samples followed by C. keratinophilum being isolated from 15.5% of the samples. *M. gypseum* was present in 8 (7.3%) of the samples. The prevalence of other species was as follows: *C. indicum* (13.6%), *Chrysosporium* spp. (4.5%) and *Seztsdionium* spp. (4.5%).

**Conclusion:** The preponderance occurrence of *M. fulvum* over *M. gypseum* and that of *C. keratinophilum* over *C. indicum* in soils of Anguilla is a remarkable observation. The farmers, other workers, school children who come in contact with soil are likely to be infected with a dermatophyte and also the visitors to the beaches and the local population nearby.

**Keywords:** Anguilla; soils; *Microsporum fulvum*; *Microsporum gypseum*; *Chrysosporium keratinophilum.*

1. INTRODUCTION

Keratinophilic fungi are organisms that have ability to utilize keratin as a sole source of carbon and nitrogen. They are involved in the breakdown of keratinaceous substrates in environment and are ubiquitous in various countries worldwide. Most of them belong to hyphomycetes which include dermatophytes and a variety of non-dermatophytic keratinophilic fungi [1,2]. Most dermatophytic are anthropophilic (man loving) or zoophilic (animal loving) in their natural habitat, while some of them like *M. gypseum* complex and *Trichophyton terrestre* occur in soil as saprophytes and are termed geophilic [3]. Some of the dermatophytes may be associated with more than one ecological niche. For instance, *T. simii*, *M. persicolor*, *M. nanum* and *M. vanbreuseghemii* are primarily zoophilic but may also occur as geophilic [4-9]. Non-dermatophytic keratinophilic fungi, represented predominantly by species of *Chrysosporium* and other genera, occur as saprobes in soil [2,10-12]; some of them are potential pathogens for humans and animals [2,12].

Studies carried out in several countries in different parts of the world have demonstrated the occurrence of a variety of keratinophilic fungi, such as *Chrysosporium* species and dermatophytes including *M. gypseum* species complex, *Trichophyton terrestre* and *T. ajelloi* in soil [3,10,11]. Other dermatophytes known to occur infrequently or sporadically in soil in some countries are *T. mentagrophytes*, *M. cookei*, *M. vanbreuseghemii* and *T. gloriae* [8,11,12]. This study reports the occurrence of dermatophytes and other keratinophilic fungi in soils of Anguilla, British West Indies (BWI) and compares it with the other such studies in the West Indies and some other countries, viz. India and Brazil. The public health significance of the findings is discussed.

2. MATERIALS AND METHODS

The study was conducted in the Department of Microbiology, Saint James School of Medicine, Anguilla (BWI) May to October 2016. A total of 110 soil samples was collected between May to September 2016 from different types of habitats from several localities in sterile zip polythene bags from a depth of 3-5 cm, with the help of a stainless steel spatula disinfected with 70% isopropyl alcohol each time before and after use. The locality-wise distribution of samples was as follows: Crocus, George and North Hills Valley, The Quarter, and Sandy Ground-14, Beaches (Shoal Bay, Meads Bay, Rendezvous Bay, Blowing Point and Stone Bay)-9, Harbours (National Park Harbour and Island Harbour)-9, Cultivated fields-12, Gardens-10, Under trees- 9, School playgrounds -9, Sugarcane field-8, Grasslands-8, Poultry habitats-8, Hospital surroundings-7, and Offices surroundings-6. The samples were transported to the laboratory and processed within 18-36 hours of collection and processed by the well-known technique of Vanbreuseghem [13]. For this, pieces of mixed adult human (male) and child (male and female) hair 0.5- 1.5 cm long, sterilized by autoclaving were spread on the soil samples (about 25 g each) in disposable sterile Petri dishes (9 x 1.5 cm), using two Petri dishes for each sample. Small quantities of sterile distilled water (10-15 ml) were poured on the hair-baited plates. All the plates were incubated at room temperature (23-30°C). Sterile water was poured into the dishes periodically to provide moisture needed for fungal growth. Fungal growths appearing on hair baits after 2-4 weeks of incubation were microscopically examined and transferred to plates of Mycobiotic agar (Oxoid). The prepared Mycobiotic agar contained 0.05 mg/ml of chloramphenicol, and 0.4 mg/ml of cycloheximide to cut down contamination due to bacteria and saprophytic molds respectively. The cultures were microscopically examined to check for
purity and sub-cultured to get pure cultures of the isolates. Identification of the isolates was accomplished by studying in detail the colonial characters of the isolates such as color, texture and formation of pigment on reverse and microscopic features and comparing with descriptions of suspected fungi in standard books and manuals [3,12,14,15]. The microscopic characteristics including presence of distinctive morphological structures such as spores were studied by staining small portions of growth in mounts of Lactophenol blue (Sigma-Aldrich).

3. RESULTS AND DISCUSSION

The distribution of keratinophilic fungi recovered from different types/habitats of soils in several locations across the island of Anguilla is indicated in Table 1. Out of 110 samples of soil examined, 78 samples were positive for keratinophilic fungi, with 7 of them yielding mixed growth of two fungi; thus 85 isolates of keratinophilic fungi were obtained. *M. gypseum*, a well-known geophilic dermatophyte was the commonest species, being recovered from 8 (7.3%) of the soil samples representing several soil type/habits, the other dermatophyte *M. fulvum* was isolated from a much larger number of samples-35 (31.8%) of the samples examined from a variety of soil types/habitats (Table 1). Other keratinophilic fungi were represented by species of *Chrysosporium* with *C. keratinophilum* being predominant species followed by *C. indicum*. The isolates of *M. fulvum* originated from a variety of soil types/habitats, predominantly cultivated fields, poultry farmers, school playgrounds and beaches.

The dermatophytes, *M. gypseum* and *M. fulvum* recovered from soils of Anguilla have been isolated from soils in other Caribbean countries, Bahamas [16], St Kitts & Nevis [17], Bonaire [18] and Jamaica [19]. In a study from Brazil, isolates of *M. gypseum* complex ("M. gypseum" and "M. fulvum") were recovered from 19.2% of the soil samples [20]. *M gypseum* has been recorded as one of the agents of tinea capitis in Cuba [21], Haiti [22], and Trinidad [23]. So far there are no reports of dermatophytic human infections from Anguilla. Nevertheless, the occurrence of *M. fulvum* in a variety of habitats, especially the cultivated fields, school playgrounds, and beaches suggests the likelihood of workers in these places and school children being infected and also the visitors to the beaches, and the nearby local inhabitants.

Fig. 1. Left side-Macroconidia of *M. gypseum* with slightly rounded terminal ends, and truncated proximal ends. Right side-Macrocondia of *M. fulvum*, longish and bullet shaped in lactophenol blue mounts.
Table 1. Distribution of keratinophilic fungi in different types/habitat of soils in Anguilla (BWI)

| Type/Habitat                | No. Samp. Exam. | No. (%) positive for different species of keratinophilic fungi |
|-----------------------------|-----------------|---------------------------------------------------------------|
|                             |                 | M. fulvum          | M. gypseum      | C. kerat.   | C. indicum | Chrys. spp. | Seped. spp. | Total |
| Croccus, George and North Hills | 14              | 5                  | 1               | 2           | 2          | 1           | 1           | 12    |
| Cultivated fields           | 12              | 6                  | 2               | 2           | 1          | -           | -           | 11    |
| Gardens                     | 10              | 4                  | 1               | 1           | 1          | 1           | 1           | 7     |
| Under trees                 | 10              | 2                  | 1               | 1           | 1          | 1           | 1           | 7     |
| Beaches                     | 9               | 3                  | 1               | 1           | 2          | -           | 1           | 8     |
| Harbours                    | 9               | 3                  | 1               | 1           | 1          | -           | -           | 6     |
| School playgrounds          | 9               | 4                  | -               | 2           | 1          | 1           | -           | 8     |
| Sugarcane field             | 8               | 2                  | 1               | 2           | 2          | -           | -           | 7     |
| Grasslands                  | 8               | 2                  | -               | 1           | 1          | 1           | -           | 5     |
| Poultry habitats            | 8               | 3                  | -               | 2           | 1          | 3, 11, 14, | 1           | 7     |
| Hospital surroundings       | 7               | 1                  | -               | 1           | 1          | 1           | -           | 4     |
| Offices surroundings        | 6               | -                  | -               | 1           | 2          | -           | -           | 3     |
| **Total**                   | **110**         | **35 (31.8)**      | **8 (7.3)**     | **17 (15.5)**| **15 (13.6)**| **5 (4.5)** | **5 (4.5)** | **85** |

Abbreviations: Samp.: Samples, Exam: Examined, M.: Microsporum, C., Chrysosporium Chrys.: Chrysosporium, kerat.: Keratinophilum, Seped.: Sepedonium
C. indicum has been reported as the dominant keratinophilic species of Chrysosporium world-wide [2,11,15,24]. The relatively more frequent occurrence of C. keratinophilum in the present study is noteworthy, a similar observation has been made in two of the studies on keratinophilic fungi from several parts of India [25,26]. It may be mentioned that C. keratinophilum has been identified as the etiological agent of a few cases of human skin and nail infection [27].

4. CONCLUSION

This communication represents the first study of keratinophilic fungi in soils in Anguilla. The preponderance occurrence of M. fulvum over M. gypseum in soils of Anguilla, (BWI) in the present investigation is a remarkable finding.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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