

# Filamentous Fungi Isolated from Clinical Samples Stored for a Long Time in the Sand

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Fungi in nature which are commonly present in variety of environments such as dust, soil, water, vegetation and rotting wastes, can spread large areas with different air movements [1]. Clinically relevant fungi include yeasts, filamentous fungi and dimorphic fungi. Yeasts are unicellular organisms and include the relatively common *Candida spp.* and *Cryptococcus spp.* The filamentous fungi (molds) are characterized by branching hyphae, which are either septate or aseptate and are further subdivided into the hyaline and the dematiaceous (darkly pigmented) groups. These classes include the more common *Aspergillus spp.* and the zygomycetes, including *Rhizopus*, *Rhizomucor*, *Mucor* and *Fusarium* species. Finally, the dimorphic, or so called 'endemic', fungi are filamentous at 25°C and yeasts or spherules in host tissue or when incubated at 35°C, and include *Blastomyces*, *Histoplasma*, *Coccidioides*, *Paracoccidioides* and *Penicillium marneffei*. With the exception of the endemic fungi and trauma, induced inoculation, invasive fungal infections are largely confined to immune compromised patients, with variations in patterns of susceptibility based on the degree and category of immune dysfunction. Frequently, cited risk factors for fungal infections are HIV/AIDS, hematopoietic stem cell transplant (HSCT), lymphoid malignancies, neutropenia, hereditary immune defects, immune suppressive medications transplant recipients receiving immune suppressive therapy (e.g. corticosteroids). Use of broad spectrum antibiotics, having a central venous catheter, cancer, diabetes mellitus, intravenous drug abuse, artificial pulmonary ventilation and mechanical breakdown of the blood brain barrier via surgery or trauma [2-4]. Invasive fungal infections have constituted an increasingly important cause of morbidity and mortality in immune compromised patients [4]. Furthermore, because risk-factors for these infections continue to increase in frequency, it is likely that the incidence of nosocomial fungal infections will continue to increase in the coming decades [3].

Moulds such as *Mucorales*, *Aspergillus spp.*, *Fusarium spp.*, *Paecilomyces spp.*, *Alternaria spp.* In addition, *Scedosporium spp.* are the main opportunistic filamentous fungi that can cause serious and rapidly fatal infections in immune compromised patients [3,5]. These infections are difficult to diagnose and cause high rates of morbidity and mortality, despite anti fungal therapy. Early initiation of effective anti fungal therapy and reversal of underlying host defects remain the corner stones of treatment for nosocomial fungal infections [3]. The genus *Aspergillus* contains approximately 175 species, but only a minority of them have been associated with human disease. Infections are caused mostly by *Aspergillus fumigatus*, followed by *Aspergillus flavus*, *Aspergillus terreus*, *Aspergillus niger* and *Aspergillus nidulans*.

The conidia (spores) are easily aerosolised [2,3,6]. *Aspergillus spp.* are opportunistic moulds that cause several clinical pictures of aspergillosis, such as allergic broncho pulmonary aspergillosis, chronic necrotizing aspergillosis, and invasive aspergillosis [2,3,6]. The mortality rate of invasive aspergillosis is as high as 50-100% [7]. Overall, the outcome of infection appears to depend more on host factors than on the virulence or pathogenicity of the individual *Aspergillus* species [3]. Immuno-

compromised patients are mainly infected through the irrespiratory tract by inhaling infectious fungal spores [8]. The construction, renovation, demolition and excavation activities have been shown to increase the amount of air borne fungal spores dramatically, and in consequence increase the risk for *Aspergillus* infection in susceptible patients [9].

The present study, aimed preservation for a long time of filamentous fungi isolated from clinical samples in sterile. We collected these from sea coast of Mersin province. After then the sand washed three for times, it was kept in water in room temperature for one day. This procedure repeated for two times for the sand. The sand dried in the Pasteur oven at 50-55°C. We have put about 10-15 g of dried sand into the vial. The vials were sterilized at 170°C for one hour. On the other hand rubber bottle caps were sterilized with ethylene oxide. Five different filamentous fungi species (*Aspergillus*, *Fusarium*, *Acremonium*, *Alternaria* and *Cladosporium*) produced as a pure culture in SDA medium. Breeding colonies of filamentous fungal hyphae and spore structures collected, enough with curve-ended loop were placed into vials and closed with caps. All vials were stored at room temperature. The stored and samples in vials at five different times (1, 2, 3, 6 and 12 months) were sub cultured in SDA medium.

Only two vials except, all of the samples which inoculated were grown in SDA medium (Table 1).

The filamentous fungi colonies growth is observed in SDA medium (Figure 1).

The reason for the absence of growth in two samples might be due to inadequate inoculation of sample into SDA medium from sand storage.

Mold/genus	1.month	2.month	3.month	6.month	12.month
<i>Aspergillus spp.</i>	+	+	+	+	+
<i>Fusarium spp.</i>	+	+	+	+	-
<i>Acremonium spp.</i>	+	+	+	+	+
<i>Alternaria spp.</i>	+	+	+	-	+
<i>Cladosporium spp.</i>	+	+	+	+	+

Note: (+):growth, (-):no growth

**Table 1:** Growth result of filamentous fungi in SDA medium.

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Figure 1: After storage in sand, *Acremonium spp.* growth in SDA medium.

## Conclusion

This procedure for long-term storage of filamentous fungi isolated from clinical samples was assessed cheap, easy and as a simple method.

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