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Investigation on the Isolation of Soil Fungi from Different Soil in Dawei Township

Mar Lar Aung¹, Thi Thi Moore² and Tin Tin Aye³

Abstract

Different fungi were isolated from different soil samples collected at Dawei Township. In the course of the studies of fungi, the soil samples were collected at different depths of 2", 4" and 6". Soil type and pH were measured and investigated varieties of fungi in different soil samples. Chemical treatment dilution method and feeding method were utilized for the isolation of fungi with the culture medium LCA. Seven isolated fungi were identified and described to the generic level based on keys of Hanlin(2001) and Inaba (2004). The depth and pH of the soil investigated in the present work showed no influence on the fungal occurrence. Only a slightly variation was observed on account of climatic differences.

Introduction

Microorganisms are found in various ecosystems and many microorganisms are reflected by a variety of forms and physiological and biochemical properties. Numerous varieties of microorganisms are immensely diverse with respect to their habitats, living on earth and deeply involved with human life. Some microorganisms cause no harm at all and instead are actually beneficial to human society.

Life would have been impossible without microorganisms in nature since the types of life-threatening fungal and bacterial infections are increasing recently, the need of new and effective metabolites are desired. Man has taken advantages of the varieties of microorganisms to his benefit. The success of screening program depends upon selection of appropriate tests and suitable sources. The typical materials for microbial sources are soil, living and fallen leaves, leaf litters, dung, insect, fresh water, marine water and so on. The soil sample is the most effective and popular materials for especially isolating a number of microorganisms. Many of natural, semi-synthesized and synthesized antibacterial and antifungal metabolites have been reported in clinical and agricultural uses. Since some pathogenic microbes are resistant to antibiotics,

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antimicrobial metabolites today are required to have potent activity and be safe to animals, human and ecosystems.

The present work has been undertaken as an initial step toward the study of soil fungi present in the soil of Dawei Township. These isolated fungi were hoped for further investigations such as fermentation for the production, extraction and purification of antimicrobial metabolites.

A. Isolation of Fungi from soil samples

Five different kinds of soil samples were utilized for the isolation of microorganisms especially fungi. The soil samples were collected at Dawei Township, Taninthari Division. (Table 1)

Table 1. Soil sample employed for the isolation of microorganisms

Sample No	Soil Type	pH	Collected Place
S.1	Silt –sand	5.8	Dawei Township
S.2	Silt-clay	5.4	Dawei Township
S.3	Silt-sand	6.2	Dawei Township
S.4	Silt –clay	5.4	Dawei Township
S.5	Silt	5.6	Dawei Township

B. Methods for isolation of microorganisms were referred by the following method

1. Chemical treatment dilution method. (Figure 1)
2. Feeding method (Figure 2)

Chemical treatment dilution method

Nyunt Phay & Yamamura, 2005

1. Soil sample was air-dried at room temperature
2. Treatment with 1.5% phenol (means 0.1 g of soil + 10 ml of 1.5% phenol)
3. Dilution with water
4. Culture on LCA agar with Penicillin
G(0.1%)

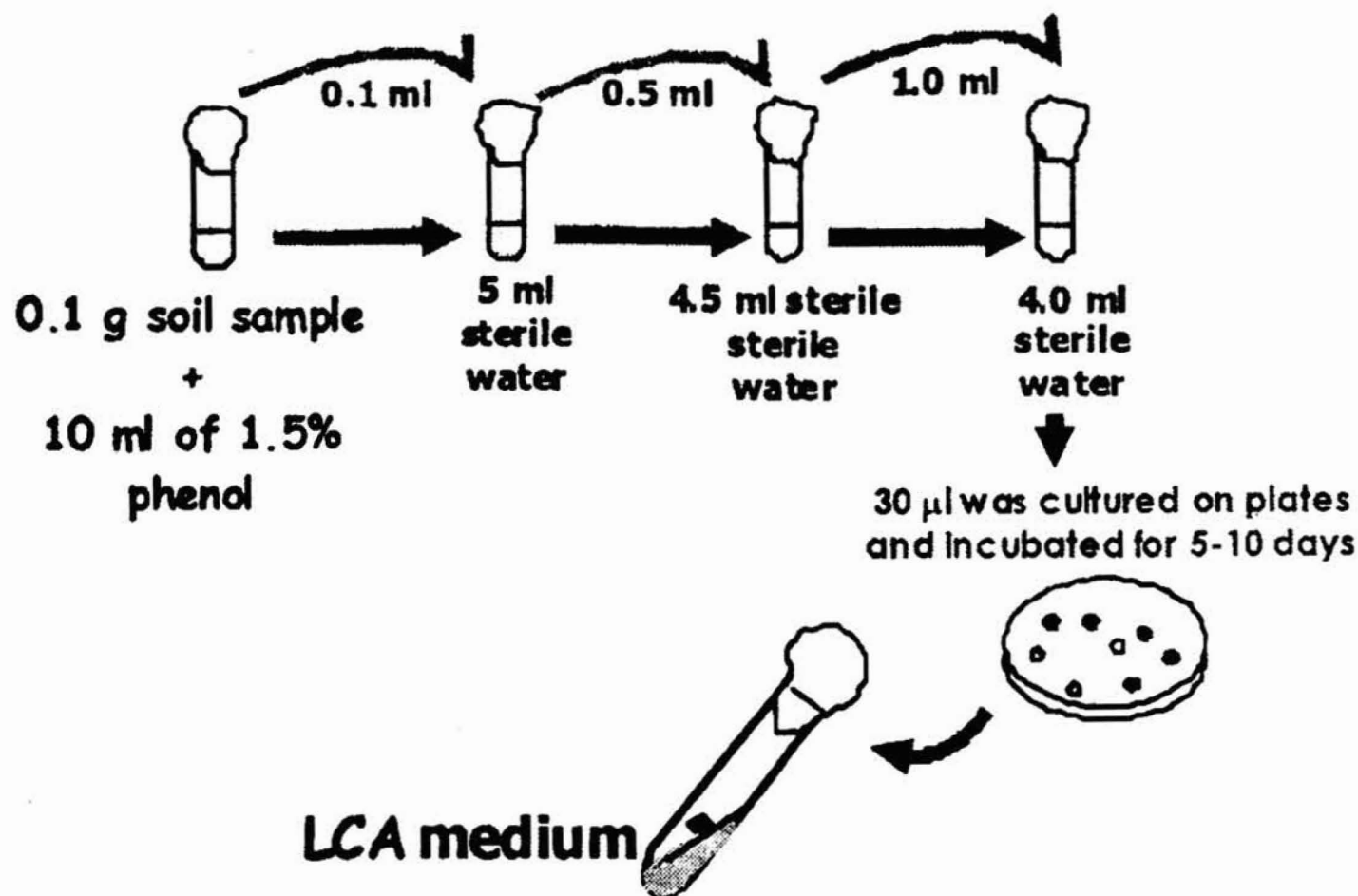


Figure 1. Chemical treatment dilution method

Feeding method (PBCC, 2002)

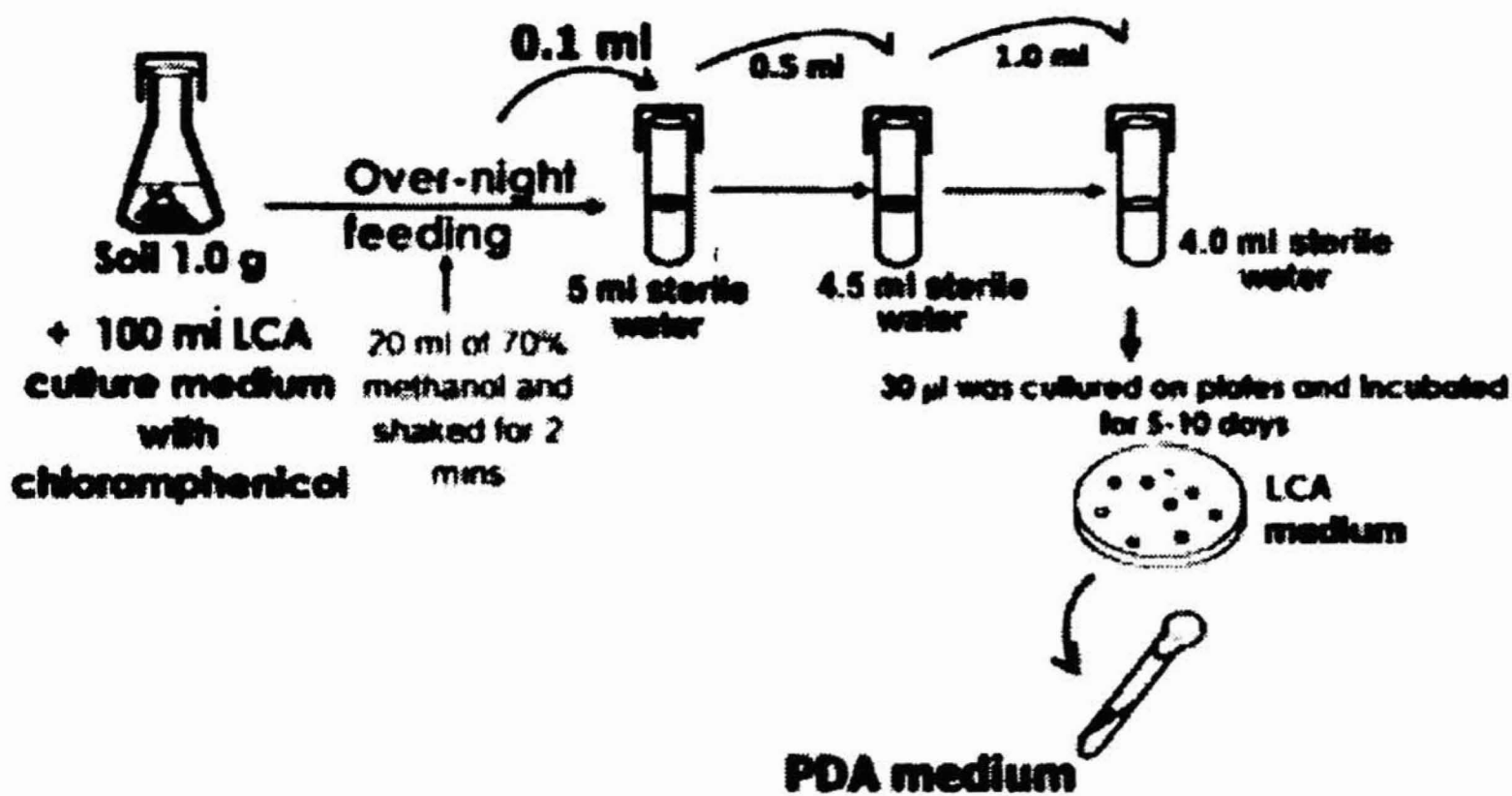


Figure 2. Feeding method

Medium used for the isolation of fungi

LCA medium (Ando , 2004)

Glucose	0.2 g
Sucrose	0.2 g
K ₂ HPO ₄	0.1 g
MgSO ₄ .7H ₂ O	0.05 g
KNO ₃	0.1 g
KCl	0.05 g
Agar	1.8 g
DW	100 ml
pH	6.5

(after autoclaving Penicillin-G 8 mg were added to the medium.)

C. Results for the isolation of fungi

In the course of the isolation of microorganisms, seven fungi were isolated from different kinds of soil samples. (Table 2)

Table 2. Isolated fungi from five different soil samples

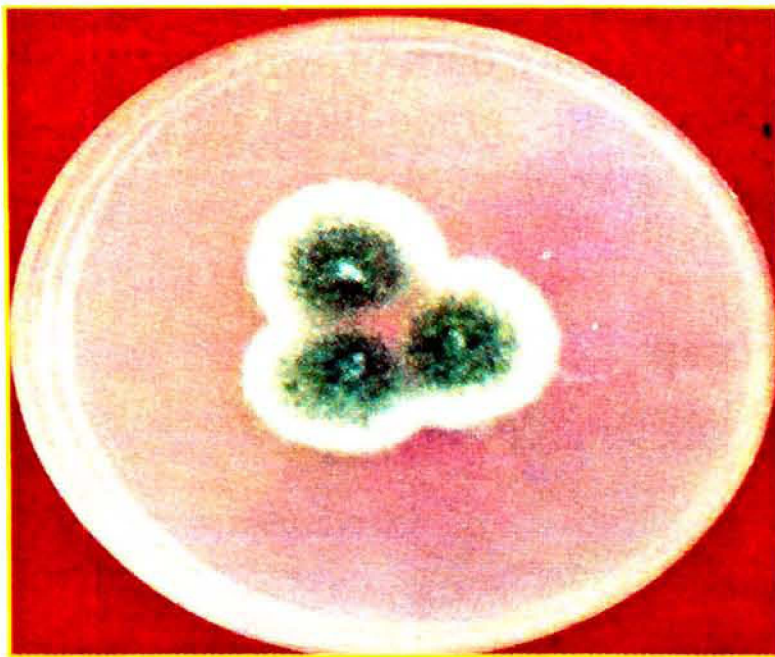
Soil type	PH	Isolation method	Isolated strain (Genera)
Silt - sand	5.8	Chemical Treatment method	<i>Ideriella sp</i>
Silt – clay	5.4	Feeding method	<i>Anthrinium sp</i>
		Feeding method	<i>Sporothrix sp</i>
Silt – sand	6.2	Feeding method	<i>Ramichloridium sp</i>
Silt – clay	5.4	Feeding method	<i>Botrytis sp</i>
		Chemical Treatment method	<i>Verticillium sp</i>
Silt	5.6	Feeding method	<i>Tretospeira sp</i>

D. Identification of fungi

Identification of the producing fungi by macroscopic and microscopic characters, For the study of macroscopic characters, three media such as Czapeck agar, Malt extract agar and Potato dextrose agar were employed and incubated for 7 days at 25 C. Good growth occurs on PDA, the optimum pH range is 5.5- 7.5; sporulation is best between pH 6.0-6.5; below pH 5.0 sporulation are reduced.

According to the macroscopical and microscopical characteristic features and based on the reference key of Ando and Inaba (2004), Hanlin (2001), Domsch (1993), Barnett (1956) and Corlett (1995), producing fungi were keyed out and grouped as the fungi imperfecti. (Figure 3-6)

Morphology and Micrograph of *Ideriella* sp



Morphology of *Ideriella* sp

(7 days old culture) Whitish green colonies, effuse, 2.8 cm diam on PDA.

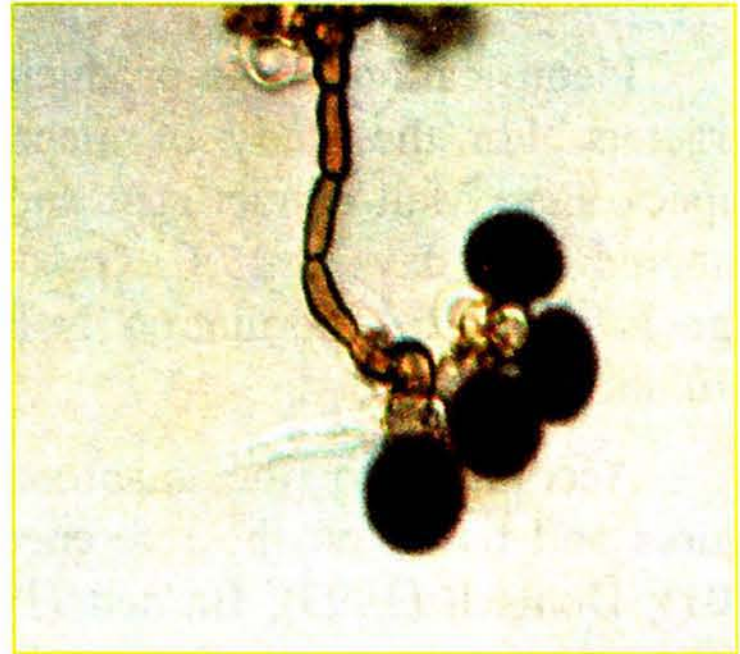
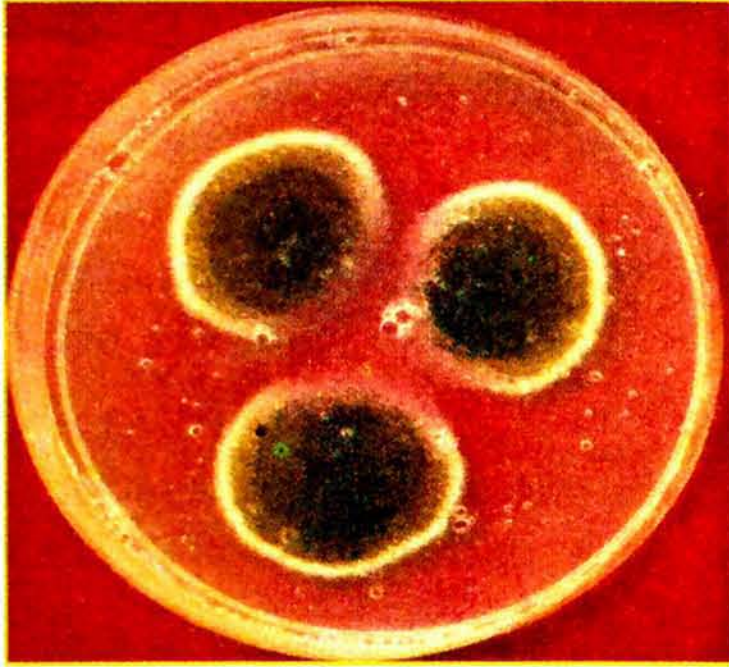


Micrograph (x 5000)

Conidia lacking septa, lunate with pointed ends produced in clusters near the apex of the conidiophore.

Figure 3 Morphology and Micrograph of fungi

Morphology and Micrograph of *Anthrinium* sp



Morphology of *Anthrinium* sp

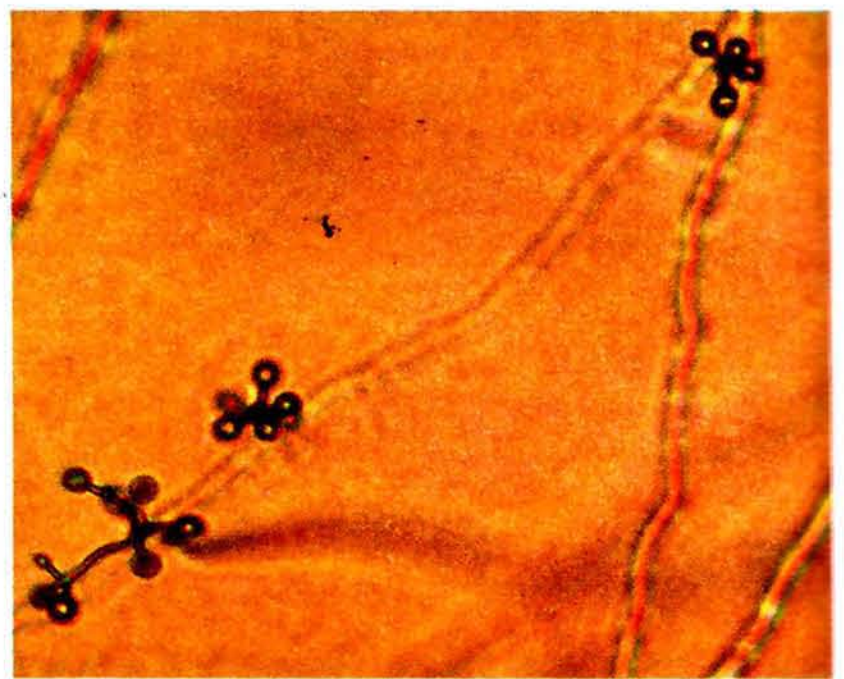
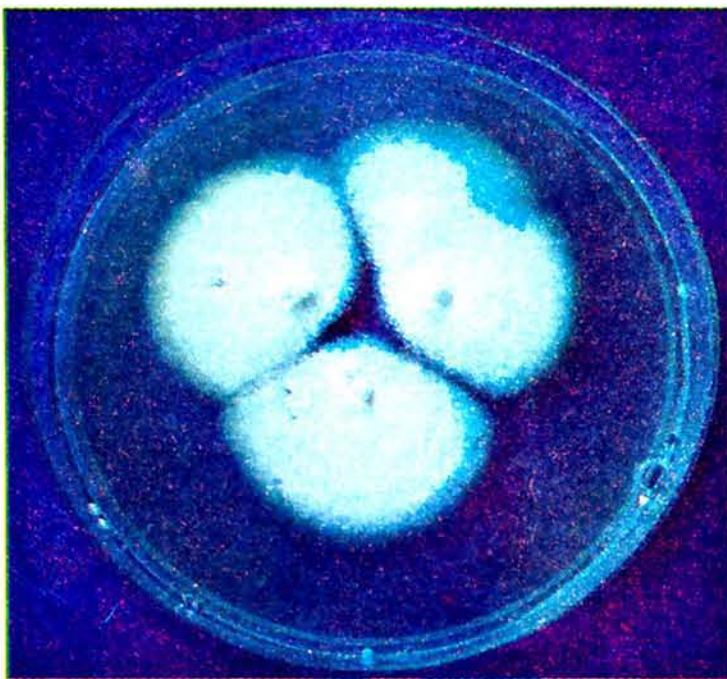
(7 days old culture)

Dark brown colonies, 3.5 cm diam on PDA.

Micrograph (x 5000)

Conidia are dark brown and usually occur in grape-like masses on white woolly colonies

Morphology and Micrograph of *Sporothrix* sp



Morphology of *Sporothrix* sp

(7 days old culture) Grayish colonies, finely floccose, velvety, 2.5 cm diam on PDA.

Micrograph (x 5000)

One-celled conidia are produced on short roughened or toothed-branches of the vegetative filament.

Figure 4 Morphology and Micrograph of fungi

Morphology and Micrograph of *Ramichloridium* sp



Morphology of *Ramichloridium* sp
(7 days old culture)
Light brown colonies,
2.6 cm diam spreading on PDA.

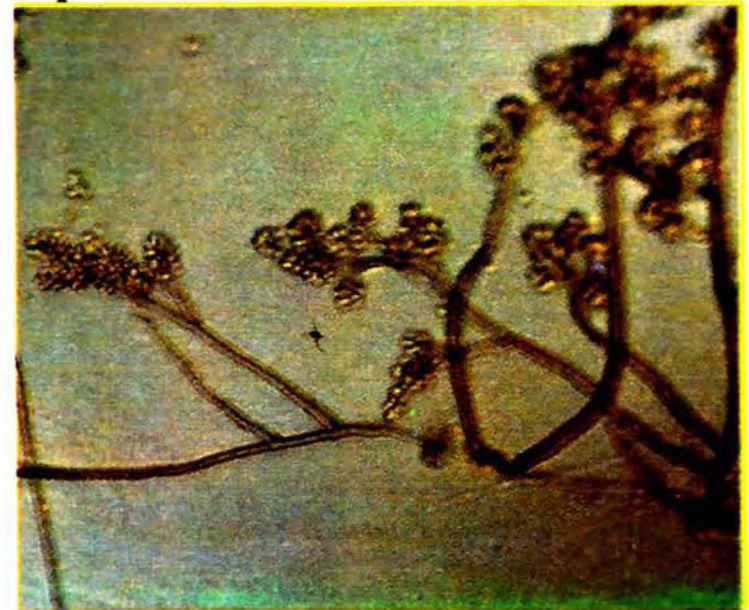


Micrograph (x 5000)
Conidia one-celled, slightly clavate
with acuminate bases, smooth-
walled. Conidiophore erect,
unbranched.

Morphology and Micrograph of *Botrytis* sp



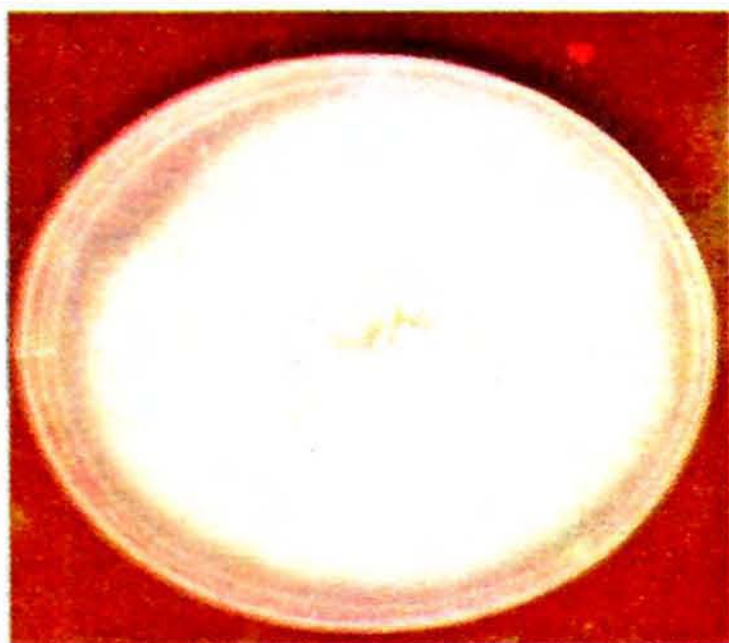
Morphology of *Botrytis* sp
(7 days old culture)
Colonies spreading broadly, Light-
grey to grayish brown and 4.3 cm
diam on PDA.



Micrograph (x 5000)
Conidia obovoid, smooth-walled.
Conidiophores with branched tops.
Spores cover the ultimate branches
are produced synchronously.

Figure 5 Morphology and Micrograph of fungi

Morphology and Micrograph of *Verticillium* sp



Morphology of *Verticillium* sp

(7 days old culture)

Whitish to cream colonies, flocculose, 5.6 cm diam on PDA.



Micrograph (x 5000)

Conidia ellipsoidal to short cylindrical, hyaline, 1 or 2-celled. Conidiophore slender, branched with several whorls of phialides.

Morphology and Micrograph of *Tretospeira* sp



Morphology of *Tretospeira* sp

(7 days old culture) Grayish colonies, spreading broadly, 5.9 cm diam on PDA.



Micrograph (x 5000)

Dictyospore, spore body subdivided by intersecting septa in more than one plane. Conidiophore with septa, monoconidigenous locus.

Figure 6 Morphology and Micrograph of fungi

Discussion and Conclusion

Many microorganisms especially fungi grow in soil. Therefore soil has probably been studied more extensively than any other natural habitats. Identification of soil fungi depend on soil sources, soil type and pH and different media and various isolation techniques.

Chemical treatment dilution method and feeding method employed for rare microorganisms. The results given in the present study meant for the soil fungi in the area, culture media and isolated techniques. If the area, media and techniques are changed the results may be different.

These results were hoped that further study on soil fungi may lead to the production of antimicrobial metabolites.

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