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Assessment of volatiles organic compounds VOCs against thermophilic fungi

R.V. Shukla[1], Kiran Jain[2], Sammeksha Jain[3]

[1]HOD (Retd.) Dept. of Botany, C.M.D. College, Bilaspur, C.G.
[2]AP Dept. of Botany, Govt. Digvijay College, Rajnandgaon, C.G.
[3]AP, Sanskar City College of Pharmacy, Rajnandgaon, C.G.

ABSTRACT

The effect of eight volatile substance assessed on the mycelial growth of *Bergia papulaenera sp.*, *Myceliopthora fergusii* and *Scytalidium thermophilum var. Hetrospora.* Maximum growth inhibition in all the test fungi recorded in ethyl propionate saturated atmosphere, followed by n-amyle alcohol. The vapour of ethyl alcohol was found to be less toxic for the growth of all fungi tested. During study one of the isolate of thermophilic actinomycetes *Streptomyces sp.* showed strong antagonistic effect against thermophilic fungi.

Keyword: Thermophillic fungi, volatile compounds, antagonistic activity

INTRODUCTION

Temperature is probably the most important environmental decisive factor affecting the activities of living organisms. Most organisms that have been studied in depth are adapted to 'moderate' temperature and operated within a relatively narrow range of temperature. Presently there has been increasing interest in microorganisms, which are adapted to environment of extreme temperature of composting materials, hot springs coal mining area, bird's nest's etc.

Thermophilic fungi, one of the most economically important group of decomposers which create their own environment to survive where they live in warm habitats; geothermal soil, self heated coal waste piles, decomposing organic waste, storages etc ([15]Shukla 1985, [16]Shukla and Agrawal 1987, [17]Sigler et al. 1998). The majority of thermophilic fungi obtained play important part to process large quantities of agriculture, forest litter and municipal wastes.

In mushroom cultivation numbers of thermophilic fungi such as *Thermomyces lanuginosus, Myceliopthora thermophila, Sytalidium thermophilum* etc. used in preparation of suitable compost. Because in composting process the most vigorous thermophiles are up to 2 - 3 times faster in their growth than mesophilic group as the temperature usually exceed more than 50C. Many microorganisms colonized in composting material interact each other for their space and nutrition, where the antibiosis phenomenon process the microbes succession as many of them perish in the temperature maxima 60 - 70C.

The mycoflora, particularly thermophilic fungi remain involve in decomposition process live in an atmosphere of various volatile organic compounds (VOCs), which may be produced by themselves or by the decomposition of organic waste ([2]Hera & Baker, 1972; [1]Fries, 1973, [10]Satynarayan & Johri, 1974, [6]Lockwood 1977). Mostly, volatile compounds belong to higher series of alcohol, aldehyde and acetylenic products ([9]Robinson, et al, 1968) are inhibitory rather than stimulatory to the decomposing fungi ([8]Pathak and Agrawal, 1977; [4]Kushwaha, 1976; [3]Jain and Agrawal, 1978).

The role of volatile inhibitors against some fungi has been evaluated by many researchers as some of which are potentially inhibitory to fungi include ammonia, ethylene and some other unidentified substances [2a]Hera and Baker (1975), [14]Smith (1973). In this concern the present study was planned to find out role of volatile emanations of some organic compounds in response to test fungi.

Since most of the actinomycetes produce some toxic metabolites in the atmosphere where they live and hence are creating a limiting factor for the growth and survival of other microorganisms, therefore an isolate of *Streptomyces sp* was tested for its antagonistic effect against thermophilic fungi.

MATERIAL AND METHODS

Erlenmeyer flask of 150ml. capacity with 30 ml. of YpSs broth (pH 7) were sterilized at 15 lbs. pressure for 15 minutes and inoculated with an inoculum disc of 6mm. grown on YpSs agar medium. For a single treatment, three flasks were inoculated with each of the test organism, i.e., *Bergua-Papulospore sp., Myceliopthora fergusii S. thermophilium Var. heterosorea,* cultured were incubated at 45 +, - , 1C. for 24 hrs and then exposed to an atmosphere of VOCs. Four ml. of each volatile compound was poured in each ampoule having a side hole near its neck and the it was kept hanging inside the flask ever growing fungal cultures (71 gm) with the help of a cork in which ampule was fitted in its centre.

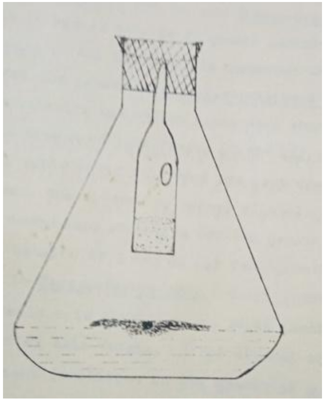

Fig 1.

The cork having ampule was tightly fixed over the month of flask. An ampule set of flask for each test organism was cultured without any treatment on control. The whole assembly was kept 45° C 5 days. After incubation period mycelial net was harvested from each flask initial on a pre weighed filter paper at 80 degree centigrade till content weight was obtained. The mycelial yield under each treatment was determined and the percentage of growth inhibition in each treatment was cultured as compare to control. To study the antagonist property of *Streptomyces sp*. the test strain was incubated in petridishes (by streak method) at one side after inoculation all the petridishes were kept for 48hrs incubation at 45 degree centigrade, after two days growth of *Streptomyces sp*. the plates were re-inoculated with the above mentioned test fungi. After 10 days incubation the results obtained are presented in plate.

RESULT AND DISCUSSION

The data showed that different volatile compounds have varied effects on test fungi. Ethyl propionate emanating vapours caused maximum inhibition in case of *Myceliopthora fergusii* (86.6%) and *Bergua-Papulaspora sp*. (86%) while it caused only 64.2% growth inhibition in case of *S. thermophillum* var. heterospores. The volatile compounds used were found active against the growth of *Bergua-Papulaspora.sp.* in most of the cases VOCs caused more than 75% inhibition in growth of test fungi (Table 1), however ethyl alcohol could cause only 62.3 % growth inhibition in these fungi.

Fig. 2. i. *Bergua papulaspora* sp.
ii. Myceliopthora fergusii

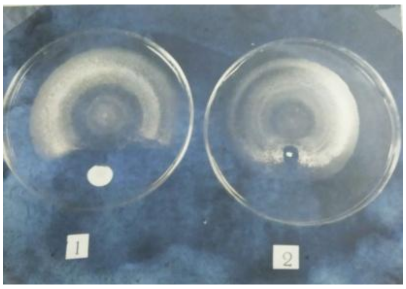

Fig. 3. i. *Humicola lanuginosa* strain I
ii. *Humicola lanuginosa* strain II

The vapours of n- amyl alcohol, n-butyl alcohol were found more affective against the growth of *S. thermophillum.* var. heterosporous causing 69.3% and 68% growth inhibition respectively. In *Bergua.papuluaspora sp.* ethyl formate, n-butyl acetate and acetic acid glacial caused 38.4 % inhibition in mycelial growth of this fungus. Ethyl alcohol could cause only 30.7% inhibition in the growth of *S. Thermphilum sp*. For the growth of *Myceliopthora fergusii* n-butyl acetate vapours caused 92.7% inhibition and are found next to ethyl propionate as far as its toxicity for this fungus is concerned.

The vapours emanating from ethyl butyrate, ethyl formate, n-amyl alcohol, n-butyl alcohol, glacial acitic acid and ethyl alcohol could cause 73.1, 63.3, 63.5, 52.0, 51.9, and 28.8 per cent growth inhibition, respectively in case of *Myceliopthora fergusii*. In the present study, vapours from different volatile compounds inhibit mycelial growth in all the test fungi, but, *S.thermophilum var. heterospore* was found comparatively somewhat resistant against emanating vapours of test VOCs, while *Bergua Papulaspora sp.* Showed higher sensitivity for these test compounds.[11]Satyanarayana and Balistic (1966) have reported some aromatic hydrocarbon in soil . [10]Satynarayam and Johri(1978) [15]Shukla (1985) reported the inhibition of mycelial growth as a result, interaction of volatile inhibitors produced by one organism against another. Similarly, [5]Lewish (1976) illustrated the evolution of volatiles from decomposing plant tissue when affected the mycelial growth and melanization of Rhizoptonia solanie . [7]Nerman (1930) have studied the production of volatile organics by the yeast fungus Diploccoccus aaggregatus and identified about 28 different organic compounds including ethyl alcohol, n-amyl alcohol, n-butyl alcohol, ethyl propionate, ethyle butyrate, ethyl formate, n-butyl acetate and acetic acid . the ethyl as cause of soil fungi statis has variously been reported by a number of workers ([14]Smith,1973; [2a]Hera and Baker,1975; [12]Smith and Reotall; 1971). It is also to be noted here that the vapours emanating from the test volatile compounds inhibit the mycelial growth in all the test fungi. [1]Fries (1973) suggested the possible mechanism in which volatile compounds act on the enzymes, membrane and intermediatary metabolism of the effected organism. [8]Pathak and Agrawal (1977) have reported the inhibitory effect of some organic volatiles on the growth of Coprophilous fungi. [3]Jain and Agarwal (1978) have reported the stimulatory effect of ethanol, n-butanol, on the bgrowth of *keritinophyton terreum.* Thus, as reported by above workers and as also noted in present study the volatile emanatiene of organic compounds cannot be generalized for their effect on different microorganism.

The present streptomysis sp. Showed antagonistic activity by inhibiting its growth of all the three fungi tested . the result shows that in the nature these strain might be playing an important role in 'antibiosis', hence it is not allowing these fungi to grow in its surrounding.

It can be assumed that due to antagonistic properties in these strain it might be playing a significant role during microbial succession in these habitats where a no. of microorganism are involved in the process of compositing degradation of organic matter.

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Table: Mycelial growth of some thermophilic fungi in volatile compounds

Volatile compounds	***Bergua- pepulaspora* sp Dry weight % in growth Inhibition (-)**		***Chrysosporium ampulliforeri.* Dryweight % growth (-)**		**Syta:day**
1.Ethyl alcohol	43	62.3	15	28.8	24
2. n-Amyl alcohol	23	80.0	19	63.5	24
3. n-Butyl alcohol	25	78.1	25	52.0	25
4. Ethyl propionate	16	86.0	7	86.6	28
5. Ethyl formate	16	86.0	14	73.1	32
6. Ethyl butyrate	18	84.3	19	63.5	30
7.Ethyl format	15	86.9	9	82.7	30
8.Acetic acid glacial	17	85.1	27	51.9	30
Central	114		52		78