



# Study of Isolation and Identification of root zone soil Fungi of *Parthenium* *hysterophorus* and *Cassia tora* in College Campus of Ahmednagar College, Maharashtra

Dr. Sangita Horo, Dr. A.K kulkarni

## Abstract

Generally *Parthenium hysterophorus* as a weed plant introduced with wheat grain from native of Mexico, America and Argentina variety. They grow abundant in roadside as well as crop fields as weeds and they dominated to the other plants. This weed is considered to be a cause of allergic respiratory problems, contact dermatitis, mutagenicity in human and livestock. Crop production is drastically reduced owing to its allelopathy. Also aggressive dominance of this weed threatens biodiversity. *Parthenium hysterophorus* also known as allergic and toxic plant but it has been observed that if there *Cassia tora* plants have been grown they cannot grow and they suppress the any growth of *Parthenium hysterophorus*. This growth suppression may be due to their microbial activity in root zone of soil. In this investigation soil sample has been taken from the root zone of *Parthenium and Cassia tora* and cultured into the in-vitro condition by preparation of Potato- dextrose -Agar medium with soil sample dilution method. There were different 30 species of fungi has been isolated but only 13 has been identified by the most updated keys for the most updated keys. The identified fungi confirmed with microbial expert.

**Key words** – *Parthenium hysterophorus*.and *Cassia tora*, microbial activity, soil dilution method

## 1. Introduction

Soil is the uppermost layers of the earth surface and occurs all the microbial activities are in it. The microorganisms plays major role in soil ecosystem. The soil serves as a reservoir for many microbial communities of plants and herbs which can be producing, co2 and nitrogen cycle<sup>1</sup>. Soil is an oligotrophic medium for the growth of fungi because the fungal growths are extremely limited for most of the time and readily available are present for short periods in a limited zone.<sup>2</sup> For most of the time, fungi are either dormant, or they metabolize and grow very slowly utilizing arrange of organic molecules. Fungi are an important part of the microbial ecology<sup>3</sup>. The majority of fungi decompose the lignin and the hard-to-digest soil organic matter, but some fungi consume simple sugars<sup>4</sup>. Fungi dominate in low pH or slightly acidic soils where soils tend to be undisturbed<sup>5</sup>. Fungi break down the organic residues so that many different types of microbes can start to decompose and process the residues into usable products. The microorganisms plays major role in soil ecosystem. Microbial composition and functioning changes the soil quality through decomposition of organic matter, recycling of nutrients and biological control<sup>6</sup>. The microbes and plants together regulate many soil processes including the carbon cycle and nutrient recycling. The microbial species diversity and the total microbial population determine the ability of plants to obtain soil nutrients like nitrogen, phosphorus and micronutrients. Plant diversity and abundance may change the entire soil ecosystem through the release of root exudates that attract or inhibit the growth of specific organisms<sup>7</sup>. These carbon-rich substances can range from less than 10% to as much as 20% a plant's total carbon production.<sup>4</sup> Hyphae must be in close contact with living or dead organic soil residues to absorb

nutrients, so they usually grow in association with other soil microorganisms<sup>8</sup>. They compete aggressively for scarce nutrients, and competition usually results in a succession or change in microbial populations as nutrients are absorbed or depleted. Initial colonizers absorb simple sugars, amino acids, and vitamins from plant parts such as fruits, seeds and vegetables, and are classified as “sugar fungus.” The dominance of these fungi is short-lived because waste products accumulate quickly<sup>9</sup>. The present studies were identified fungal community from soil samples collected from different locations of root zone of *P. hysterophorus* and *Cassia tora* weeds plant of Ahmednagar college campus and it was identified with microbial expert. This Investigation brought the finding reduced growth of *P. hysterophorus* where the *Cassia tora* grown.

## 2. Method and Materials

### 2 a. Preparation of culture medium

The Soil samples (approximately 5gm) were collected from our college campus under the root zone of *P. hysterophorus* and *Cassia tora* in clean dry and sterile polythene bags along with sterile spatula. The collected samples brought to the laboratory and preserved for further studies. Soil sample were placed in test tube for preparation of dilution series and culture done in different pH Medium like pH2, pH4, pH8, and pH9. The standard culture medium Potato –Dextrose Agar (PDA) has been prepared by taking of

Potato- 200gm (peeled)

Dextrose -20gm

Distilled water -1000ml.

All these constituent mixed well in conical flasks (250ml capacity).The medium transferred in conical flasks.The medium is sterilized in autoclave at 15 Lbs. for 20 minutes. All glasswares, petriplates and pipettes were sterilized and The test tubes are stored in refrigerator. After cool down put 1 pinch of antibiotic(Benzathine penicillin injection IP) to get pure culture of fungi. All work has been conducted under the laminar airflow to avoid the contamination. All collected soil sample of both plants done first 1gm of soil is transferred to 9 ml of sterilized water in the test tube and next taken 1/100 diluted soil sample has been transferred to 9 ml sterilized water blank to get 1/1000 dilution. Now taken another 1 ml of 1/100 dilution and again transferred it to 9 ml water blank to have 1/1000. Such serial dilution technique has been done and dilution soil sample were spread on the culture medium of petridishes. From the collected soil samples of soil dilution method diluted with 1gm soil is in 10ml of sterilized distilled water<sup>10</sup>. 1ml of suspension was added to sterile petriplates in triplicates containing sterile Potato Dextrose Agar the plates were incubated at 28C<sup>0</sup> for 5-7 days. A greater number of species was isolated most of the fungus grown heavily. When inoculums were transferred into petriplates containing nutrient media cells are not separated from each other. Therefore, there develop mixed colonies. Therefore spread plate technique is employed for pure culture<sup>11</sup>.

### 2. b. Identification of Fungi

The isolated fungi were identified to the genus level and to the species when possible on the basis of macro morphological texture and micro morphological characteristics using suitable media, slide cultures and the most updated keys for identifications<sup>12</sup>. The identified fungi confirmed with microbial expert. Hence isolation of pure culture from mixed colonies is rather difficult. The hyaline mycelia /spores/conidia and cytoplasm can be stained by using Lactophenol and cotton blue was used<sup>13</sup>. Cotton blue were stained cytoplasm and results in light blue background. Lactophenol acts as a cleaning agent. The stained specimens was observed under the lightmicroscope (Magnus MLXi plus) for identification and microphotograph was taken under 10X × 40X magnification.

### 3. Result

Fungi are more abundant in cultivated soil. They are grown more in the upper layers of 'A' horizon Soil fungi can be functionally divided into saprophytic sugar fungi, lignin decomposers, coprophilous fungi, predaceous type and root inhibitor<sup>14</sup>. Soils represent a natural development of numerous fungi, most of which are saprophytes and facultative parasites<sup>14</sup>. Fungi are more capable to survive in acidic soil. Fungi reveal a more pronounced rhizosphere effect in relatively infertile soil, such differences suggested that exudates are more effective in microbial stimulates under condition of limited available substrates. *Mucor Sps. Rhizopus Sps. Aspergillus oryzae, Sterile mycelium, Penicillium sps, Aspergillus Sps. Mucor Sps. Arthospra Sps, Monilia Sps. Alternaria Sps. Penicillium, Sps., Fusarium Sps. Tortula Sps, Aspergillus Sps.* were grown the soil sample of *Cassia tora* ( Cultured on Plate no.- 1,2,4 & Shown on the table-1) While *Aspergillus sps., Mucor Sps., Rhizopus Sps. Fusarium Sps. Penicillium islandicum, Aspergillus niger, Aspergillus fumigates, Aspergillus luchensis, Fusarium dimerum, Mucor geopilus* (Cultured on Plate no.- 5,6,7,8 Shown on & table-1). This is to observed that the growth of the *Aspergillus* sps are more in soil sample of *Parthenium* but in the soil sample *Cassia tora* fungi were grown *Arthospra Sps, Monilia Sps. Alternaria Tortula Sps. Verticillium*. May be because of these growing differences to suppressed the growth of *Parthenium* plant by *cassia tora* due to their microbial activities in soil.

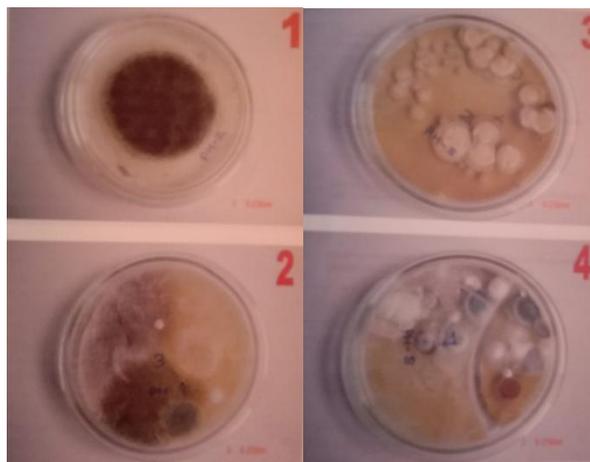


Plate no.- 1,2,3 and 4 soil sample of *Cassia tora*

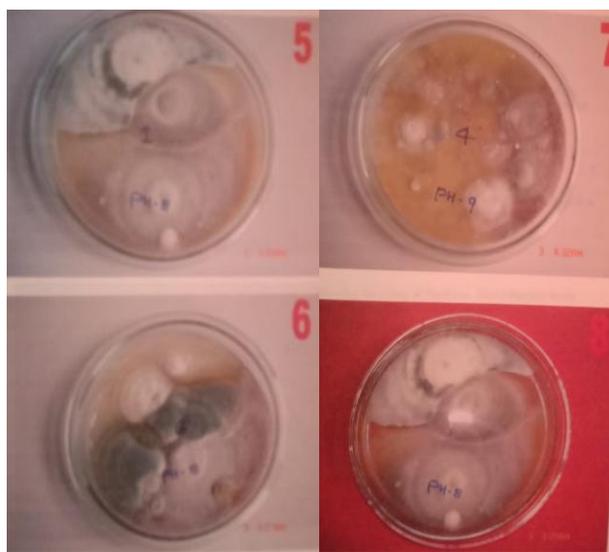


Plate no.- 5,6,7 and 8 soil sample of *P.hysterophorous*

Sr. No.	Dilution factors	Name of Identified Fungi (A)	Colour of colony	Name of Identified Fungi (B)	Colour of colony
1.	1/10	<i>Aspergillus sps.</i>	Yellow green	<i>Mucor Sps.</i>	Black
		<i>Penicillium sps.</i>	Yellow Brown	<i>Rhizopus Sps.</i>	Grey white
		<i>Mucor Sps.</i>	Black	<i>Aspergillus oryzae</i>	Yellow green
		<i>Rhizopus Sps.</i>	Grey white	<i>Sterile mycelium</i>	Brown
		<i>Fusarium Sps.</i>	Brown black	<i>Penicillium sps</i>	Yellow Brown
2.	1/100	<i>Penicillium islandicum</i>	Yellow	<i>Aspergillus Sps.</i>	Yellow green
		<i>Aspergillus niger</i>	Green	<i>Mucor Sps.</i>	Black
		<i>Sterile mycelium</i>	White	<i>Arthospra Sps.</i>	-
		<i>Rhizopus Sps.</i>	Grey white	<i>Monilia Sps.</i>	Brown
		<i>Sterile mycelium</i>	White	<i>Alternaria Sps.</i>	Brown Black
		<i>Aspergillus Sps.</i>	Yellow green	<i>Penicillium Sps.</i>	Yellow Brown
3.	1/1000	<i>Aspergillus fumigates</i>	Green	<i>Rhizopus Sps.</i>	Brown
		<i>Aspergillus luchensis</i>	Green	<i>Penicillium sps</i>	Yellow
		<i>Fusarium dimerum</i>	Brownish yellow	<i>Aspergillus Sps.</i>	Black green
		<i>Aspergillus niger</i>	Green	<i>Fusarium Sps.</i>	Pinkish White
		<i>Mucor geopilus</i>	Black brown	<i>Sterile mycelium</i>	Brown
4.	1/10000	<i>Aspergillus Sps.</i>	Green	<i>Penicillium sps.</i>	Yellow
		<i>Fusarium dimerum</i>	Brownish yellow	<i>Fusarium Sps.</i>	Black
		<i>Mucor geopilus</i>	Black brown	<i>Tortula Sps</i>	Yellow green
		<i>Penicillium islandicum</i>	Yellow	<i>Verticillium</i>	Grey white

**Table -1: Isolated fungi from the root soil of (A) *Parthenium hysterophorous* and (B) *Cassia tora***

#### 4. Discussion

The studies on rhizosphere of there microflora of different plants have received considerable attention, hence data on microbial ecology of rhizophere soil of weeds like *P. hysterophorous* and *Cassia tora* were included. The rhizophere of these plant shows 14 form of diff. fungus associated with the root zone. Such studies of this locality will definitely help in better understanding of microbial ecology of roots. Rhizophere is such region extending a few mm from surface of each root, where the microbial population of soil influenced by the chemical activities of plant. Protease has been reported to be found in several *Aspergillus* species<sup>16</sup>. The exudates contain amino acid for microorganisms in rhizophere effect beneficial to the plant<sup>17</sup>. Generally *P. hysterophorous* plant introduced with the wheat grain from Maxico, America and Argentina variety. They grown as weeds and dominated to other crop plants<sup>18</sup>. I have observed that where *Cassia tora* are grown the *P.hysterophorous* couldn't able to grow due to the soil microbial effect. Olutiola and Nwaogwugwu<sup>19</sup> reported that *A. aculeatus* is able to produce protease with optimum activity expressed at pH 7 several other *Aspergillus* species that can produce<sup>17</sup>.In general rich microflora around the root may be due to richness of food added sloughed of protein of root tissue and exudates. The exo enzymes for example proteases and peptidase released by some fungi can break down organic matter and subsequently capture nitrogen-containing compounds, thus providing a direct path from organically bound nitrogen in the soil to the plant<sup>19</sup>.

#### 5. Acknowledgment

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#### 6. Conclusion

*Cassia tora* and *P. hysterophorous* both are weed plants but *Cassia tora* consumed as a fodder for animals and it is consume by indigenous people it used also for medicinal purpose while *P. hysterophorous* is considered to be a cause of allergic respiratory problems, contact dermatitis, mutagenicity in human and livestock and animals also doesn't consume for fodder purpose. The present study of isolation and identification of soil fungi from *Cassia tora* and *P.hysterophorous* show the maximum colonies in 1/100 dilution and minimum fungi are present in 'A' horizon. The result in general reveal that the peculiar of *Cassia tora* *Mucor Sps. Rhizopus Sps. Aspergillus oryzae, Sterile mycelium, Penicillium sps, Aspergillus Sps. Mucor Sps. Arthospra Sps, Monilia Sps. Alternaria Sps. Penicillium, Sps.,Fusarium Sps. Tortula Sps, Aspergillus Sps* while the *P.hysterophorous* *Aspergillus sps., Mucor Sps., Rhizopus Sps. Fusarium Sps. Penicillium islandicum, Aspergillus niger, Aspergillus fumigates, Aspergillus luchensis, Fusarium dimerum, and Mucor geopilus. Aspergillus oryzae, Arthospra Sps, Monilia Sps. Alternaria Tortula Sps. Verticillium sps.*are secretes some alpha-amylase that might cause the luxuriant grows of *cassia tora* which in turn suppress growth of *P.hysterophorous*. It is Maxico exotic weed plant which has entered in Indian agricultural farm since long time and the suppress the crops. But where there is maximum growth of *Cassia tora* the *P. hysterophorous* growth is suppressed. Fungi secrets of secondary metabolites by the roots<sup>19</sup>. It may be due to secretion of secondary metabolites by the roots of *Cassia tora* that suppressed the germination of *P. hysterophorous* seeds and therefore they can't found in where *cassia tora* grows.

#### 7. References

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