FUNGISTATIC FRONTIERS: THE IMPACT OF CHEMINOVA ON SOIL MICROBIAL DYNAMICS IN SUGARCANE CULTIVATION

Gangaiyammal A[1*], Manobharathi Vengaimaran[2], Surya C[3], Sangavai C[4], Ruffina D[5], & Umadevi Pongiya[6]

[1,5]Department of Microbiology, Dhanalakshmi Srinivasan College of Arts and Science for Women(A), Perambalur.*

[2,3,6]Department of Biochemistry, Dhanalakshmi Srinivasan College of Arts and Science for Women(A), Perambalur.

[4]Department of Biotechnology, Dhanalakshmi Srinivasan College of Arts and Science for Women(A), Perambalur

ABSTRACT

Bioremediation involves using living organisms to reduce or eliminate environmental hazards caused by the accumulation of toxic chemicals and hazardous waste. It presents a promising alternative to physical and chemical remediation methods, as it is often more cost-effective and can selectively achieve complete degradation of organic pollutants. In this study, the growth of fungi—specifically *A. flavus, A. terreus, F. solani, A. fumigatus, and A. niger*—was examined in PD broth with various concentrations of Cheminova (10, 20, 30, 40, and 50 ppm, along with a control). The fungi exhibited growth up to the 50-ppm concentration, though biomass varied significantly. At 60 ppm, growth was entirely inhibited. Notably, at 10 ppm, fungal growth decreased with increasing pesticide concentrations. *A. flavus* showed the highest growth at 53.1 (94.36), yielding a mycelial biomass of 5.1 g at pH 8, while *A. terreus* yielded 6.8 g, *Fusarium solani* produced 5.4 g, A. fumigatus yielded 6.8 g, and *A. niger* generated 6.3 g, all at pH 8. In India, sugarcane is a major commercial crop cultivated across many states. To protect these plants from various pests and diseases, numerous pesticides and fungicides are applied. This investigation analyzes the effects of Cheminova on the soil fungal community.

INTRODUCTION

Utilization of pesticides has become an integral component of modern agriculture, with millions of tons applied annually to enhance crop production and protect plants from a variety of harmful organisms. These target organisms include insects, fungi, bacteria, viruses, and competitive weeds that threaten the viability and yield of economically important crops. Despite the significant investment in pesticide applications, research indicates that less than 5% of these chemicals effectively reach their intended targets, leading to environmental accumulation and health risks. This inefficiency raises concerns about the long-term impact of pesticides not only on ecosystems but also on human health [1].

The detrimental effects of pesticide exposure are well-documented, ranging from acute poisoning to chronic health issues. Long-term exposure, even at low doses, has been associated with severe health consequences, including immune suppression, hormonal disruption, cognitive impairment, reproductive abnormalities, and an elevated risk of carcinogenesis. As a result, the reliance on chemical pesticides has sparked a growing demand for safer, more sustainable agricultural practices [2].

In response to these challenges, the use of microorganisms for the degradation and detoxification of toxic xenobiotics, particularly pesticides, has emerged as a promising bioremediation strategy. This biological approach involves harnessing the natural metabolic processes of microorganisms to break down harmful compounds, thereby mitigating pollution in contaminated environments. Bioremediation methods targeting pesticide contamination in soil have gained considerable attention for their ecological benefits and effectiveness, demonstrating significant success in various countries around the world [3].

Microbial biodegradation is a key mechanism for breaking down pesticides in soil and water, and it plays a crucial role in detoxifying these hazardous substances. The ability of specific microorganisms to metabolize and eliminate toxic chemicals highlights the potential for bioremediation to serve as a viable alternative to traditional remediation techniques. Conventional methods such as landfilling, recycling, pyrolysis, and incineration are often inefficient, costly, and may generate harmful by-products that further exacerbate environmental problems [4].

In contrast, biological decontamination methods, which leverage the capabilities of microorganisms, tend to be more effective in degrading a wide range of environmental pollutants without producing toxic intermediates. This characteristic makes bioremediation an attractive option for restoring contaminated sites and promoting environmental sustainability. By utilizing the natural abilities of microorganisms, we can develop innovative solutions that not only address pesticide pollution but also contribute to the health of ecosystems and communities [5].

As agricultural practices continue to evolve, the integration of bioremediation into pest management strategies offers a pathway toward more sustainable farming. This approach not only mitigates the adverse effects of chemical pesticide use but also aligns with broader environmental conservation efforts. In this context, the present investigation seeks to explore the effectiveness of microbial bioremediation techniques in the degradation of pesticides, emphasizing the importance of developing sustainable agricultural practices that prioritize both crop protection and environmental health. Through this research, we aim to contribute to a deeper understanding of the role of

microorganisms in detoxifying pesticide-laden soils and fostering a healthier ecosystem for future generations [6].

MATERIALS AND METHODS

Perambalur is one of the dry land agriculture districts of Tamil Nadu, located from 55 km of Trichy. For the present investigation soil samples were collected from different sugarcane Keelapuliyur Village.

Sampling schedule

For the enumeration of fungi, soil sample were collected in a aseptic manner at a depth of 5-10cm by making V shaped pit. From each sampling site, four samples were collected and pooled together. The soil sample were brought to the laboratory in a sealed covers and kept in the refrigerator until the use.

Isolation of soil mycoflora

Dilution plating technique described by Zhong and Cai (2007) [7] was used to isolated the fungi from soil. Soil sample weighting 1 g was diluted in 10 ml of water. One ml of the diluted sample was poured and spread on petri plate containing sterilized PDA medium in replicates. One percent streptomycin solution was added to the medium before pouring in to petriplates for preventing bacterial growth. The inoculated plates were incubated in a dust- free cupboard at room temperature for 7 days.

Observations

The colonies growth on PDA plates with different morphology were counted separately. A portion of growing edge of the colony was picked up with the help of a pair of needle and mounted on a clean slide with lactophenol cotton blue stain. The slide was gently heated in a spirit lamp soas to facilitate the growing edge of the colony was picked up with the help ofa pair of needle and mounted on a clean slide with lactophenol cotton blue stain. The slide was gently heated in a spirit lamp so as to facilitate thestaining and remove air bubbles, if any. The excess stain was removed withthe help of tissue paper and then the cover slip was sealed with transparent nailpolish. The slide was observed under a compound microscope.

Identification

Colony color and morphology were noted besides hyphae structure, spore size, shapes and spore bearing structures. They were compared with the standard works of Von Arx (1974); Anishworth et *al*., (1973) among others for identification of the species [8,9].

Presentation of data

Number of species is referred as species diversity. Population Density is expressed in terms of colony forming unit (CFU) per gram of soil dilution factor.

In order to assess the dominance of individual species in each sitepercentage contribution was worked out as follows.

$$\% \text{ Contribution} = \frac{\text{No of colonies of fungus in a sample}}{\text{Total no all colonies of all species in a sample}} \times 100$$

Pesticide assay

The mycelial discs of different fungus were inoculated into liquid potato dextrose medium along with different concentration of pesticide. It was maintained at room temperature for 7 days .After incubation the fungal mycelium.

Growth of fungi in different pH

pH ranging from 6 to 9 at an interval of pH was altered in the PDF medium byadding I N NaOH (to increase the pH) and I N HCL (to reduce the pH). The fungi were inoculated separately on plates containing PDA medium with pH ranged from 6 to 9. The plates were incubated at room temperature and the growth measurements were taken.

RESULTS

Mycoflora Mean fungal population density

Population mean density of fungi varied from 5.5 to 13.5×10^2 CFU/gwith the samples collected from all sampling station. (Table.1; Fig 1; plate 1)

Species Diversity

Altogether 5 species were recorded from all the sample collected. They were assignable to 2 genera (*Aspergillus* 4 and *Fusarium* 1).

Percentage contribution

The percentage contribution was recorded maximum in *Aspergillusflavus* (31.9%) followed by *A. terreus* (25%), *A. niger* (19.4), *Fusarium solanin* (18.2) and *A. fumigatu* (5.7%).

Growth of fungi in different pH with 10ppm of Cheminova

All the fungus showed a decreasing growth in pH 6, 7 and 8 then the pH 9 of the broth supplemented with pesticide.

A.flavus

A. flavus showed growth up to 50 ppm concentration of Cheminova but with varying biomass. At 60 ppm concentration the growth was completely arrested. At 10 ppm the growth was 53.1(94.36) which turned reducing towards increasing concentration the pesticides at 41g (73.39%) at 20ppm; 38.5g (68.42) at 30ppm; 37.5g (66.64%) at 40 ppm and 36.3g (64.51) at 50ppm. (Table 3; Plate 2).

A.terreus

A. terreus showed yield 47.1g (100%) of mycelium in the control. Among the treatment, the 60 ppm concentration of Cheminova arrested the growth completely. At 10ppm the growth was 41.5g (88.11) which further reduced as 36.8g (78.13) at 20ppm; 36.3g (77.07) at 30 ppm; 35.6g (75.58% at 40ppm and 27.1g (57.33) at 50ppm. (Table 3;plate 2).

F.solani

F.solani also showed growth up to 50ppm concentration of Cheminova but with varying biomass. At 10ppm the growth was 35.7g (87%) which turned reducing towards increasing concentration the pesticide as 32.9g (80.44%) at 20ppm; 28.3g (69.19) at 30ppm; 28.1g (68.70%) at 40ppm and and 23.5g (57.45%) at 50. At 60ppm concentration the growth was completelyarrested, when the control yielded 40.9g (100%) of wet mycelial biomass. (Table 2; Plate 2).

A.fumigatus

A.fumigatus showed yielded 48.7g (100%) of mycelium in the control. Among the treatments,

the 60ppm concentration of Cheminova arrested the growth completely. At 10ppm the growth was 47.7g (89.73%) which further reduced as 35.6g (73.10%) at 20ppm; 31.5g (64.68%) at 30ppm; 31.2g (64.06%) at 40ppm and 27.3g (56.05%) at 50ppm. (Table 3 ;Plate 2).

A. niger

A.niger exhibited growth up to 50ppm concentration of Cheminova but with varying biomass. At 10ppm the growth was 39.8g (90.04%) which turnedreducing towards increasing concentration of the pesticides as 37.8g (85.52%)at 20pm; 36.1g (81.67%) at 30ppm; 34.8g (78.73%) at 40ppm and 29.2g (66.06%) at 50. At 60ppm concentration the growth was completely arrested, when the control showed 44.2g (100%). (Table3; Plateb2).

Growth of fungi in different pH with 10ppm of Cheminova

All the fungus showed a decreasing growth in pH 6,7 and 9 then the pH 8 of broth supplemented with pesticide.

Aspergillus flavus yielded the mycelial biomass, 2.5g at pH 6, 3.5g at pH 7, 5.1 g at pH 8 and 2.1g at pH 9, while *A terreus* yielded 3.9g at pH 6, 1.2g at pH 7, 6.8g at pH 8 and 4.2g at pH 9, *Fusarium solani* yielded 2.8g at pH 6, 3.2g at pH 7, 5.4g at pH 8 and 3.4 at pH 9. 4. *fumigatus* yielded 2.8g at pH 6, 2.2g at pH 7, 6.8g at pH8 and 3.0g at pH9 and *A niger* yielded 2.7g at pH6, 1.5g at pH7, 6.3g at pH 8 and 4.0 at pH 9. (Table 4; Plate 3).

Table 1. Mean density (CFU) of fungi record in the soil collected in the soil collected from different sugarcane fields at Keelapuliyur, Perambalur (Dt).

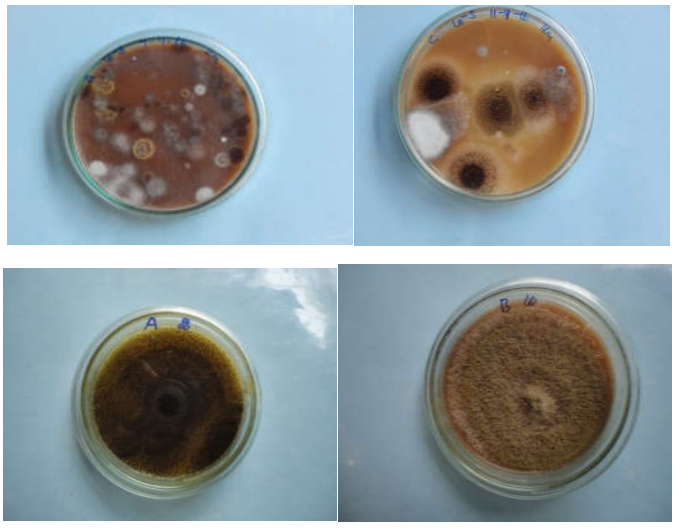
S.No	Organisms	Station-1		Station-1I		Station-1II		Station-1V		Station-V		Total no. of colonies	% Contribution
		TNC	MD	TNC	MD	TNC	MD	TNC	MD	TNC	MD		
1	*Aspergillus flavus*	3	1.5	6	3	6	3	7	3.5	6	3	28	31.9
2	*Aspergillu*	-	-	3	1.5	7	3.	12	6	-	-	22	25

	s terreus						5						
3	*Fusarium solani*	3	1.5	2	1.	4	2	4	2	3	1.5	16	18.2
4	*Aspergillus fumigatus*	3	1.5	-	-	-	-	1	0.5	1	0.5	5	5.7
5	*Aspergillus niger*	4	2	3	1.5	6	3	3	1.5	1	0.5	17	19.4
	Total no.of mean colonies	13	6.5	14	7	23	11.5	27	13.5	11	5.5	88	

TNC-Total number of colonies, MD-Mean density

Fig 1: Soil collected from sugarcane fields at Keelapuliyur, Perambalur (Dt).

Plate 1: Fungal population density- *A. niger and A. flavus*



A. Niger ***A. flavus***

Table 2. Physico-Chemical Characteristics of soil collected from

Parameters	**Station I**	**Station II**	**Station III**	**Station IV**	**Station V**
pH	7.89	7.56	7.96	7.48	7.49
Electrical conductivity	0.48	0.52	0.26	0.41	0.26
Organic carbon	0.12	0.16	0.15	0.18	0.16
Organic matter	0.24	0.32	0.30	0.36	0.32
Available Nitrogen	112.2	106.5	103.8	105.2	97.8
Available Phosphorus	3.75	4.25	4.75	4.50	4.00
Available Potassium	118	125	124	119	125
Available Zink	0.89	0.79	0.82	0.96	1.02
Available Copper	0.48	0.49	0.42	0.48	0.52
Available Iron	4.89	4.58	4.63	4.57	4.62
Available Manganese	2.16	2.48	1.59	1.89	1.84
Cat ion Exchange capacity	23.6	25.8	24.6	27.1	28.6
Calcium	10.6	11.2	10.7	10.5	11.3
Magnesium	6.8	6.9	6.8	6.4	6.5
Sodium	1.26	1.25	1.63	1.45	1.29
Potasium	0.24	0.28	0.26	0.21	0.23

Table 3. Mycelial biomass of different fungi in different ppm of Cheminova

Fungi	Cheminova (ppm)	Mycelial biomass(g)	% of growth
Aspergillus flavus	10	53.1	94.36
	20	41.3	73.39
	30	38.5	68.42
	40	37.5	66.64
	50	36.3	64.51
	60	-	-
	0	56.27	100
Aspergillus terreus	10	41.5	88.11

	20	36.8	78.13
	30	36.3	77.07
	40	35.6	75.58
	50	27.1	57.53
	60	-	-
	0	47.1	100
Fusarium solani	10	35.7	87.0
	20	32.9	80.44
	30	28.3	69.19
	40	28.1	68.70
	50	23.5	57.45
	60	-	-
	0	40.9	100
Aspergillus fumigatus	10	43.7	89.73
	20	35.6	73.10
	30	31.5	64.68
	40	31.2	64.06
	50	27.3	56.05
	60	-	-
	0	48.7	100
Aspergillus niger	10	39.8	90.04
	20	37.8	85.52
	30	36.1	81.67
	40	34.8	78.73
	50	29.2	66.06
	60	-	-
	0	44.2	100

Fusarium solani ***A. fumigatus***

A.flavus ***Aspergillus terreus***

A.niger

Plate 2: Growth of fungi in PD broth with different concentration (10, 20, 30, 40, 50 ppm and control) of cheminova

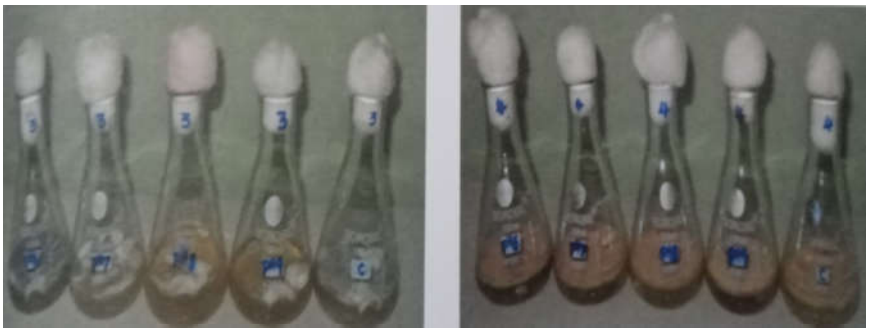
Table. 4. Growth of fungi in different pH

Organism	**pH6**	**pH7**	**pH8**	**pH9**
Aspergillus flavus	2.5	3.5	5.1	2.1
Aspergillus terreus	3.9	1.2	6.8	4.2

Fusarium solani	2.8	3.2	5.4	3.4
Aspergillus fumigatus	2.8	2.2	6.8	3.0
Aspergillus niger	2.7	1.5	6.3	4.0

Plate 3 : Growth of fungi in PD broth supplement with Cheminova and different level of pH 6, 7, 8 and 9.

Fusarium solani ***A.flavus***



A. fumigatus ***Aspergillus terreus***

Aspergillus niger

DISCUSSION

Pesticides play a crucial role in modern agriculture, serving as indispensable tools for safeguarding economically significant crops from various threats posed by weeds, insects, fungi, and nematodes. In the United States alone, approximately 600 different pesticides were registered for use

in 1989, with millions of kilograms applied to crops annually. This industry represents a global market valued at over $20 billion each year [10]. Most pesticides are synthetic organic compounds classified based on the specific pests they target. The four primary categories of pesticides include herbicides, insecticides, fungicides, and nematicides, each designed to control weeds, insects, plant pathogenic fungi, and nematodes, respectively [11].

Understanding the degradation rates of these pesticides in soil is vital for predicting their environmental fate and potential impact. Research by Ou et al. 1982 identified several physical, chemical, and biological factors that influence the degradation rates of pesticides in soils. Key factors include soil type, temperature, pH, moisture content, organic matter content, and the presence of microbial populations [12]. Among these, soil temperature and moisture are considered particularly significant environmental variables affecting pesticide degradation [13].

The physical and chemical characteristics of the soil are essential for assessing pesticide degradation. For example, optimal conditions such as soil texture, pH levels, and moisture content can enhance the breakdown of pesticides, reducing their persistence in the environment. Research has indicated that variations in these factors can lead to differing degradation rates across various crop fields, including paddy fields and groundnut fields [14].

In discussing pesticide degradation, it is important to consider not only the inherent properties of the pesticides themselves but also the ecological context in which they are applied. The interactions between pesticides and soil microorganisms play a critical role in the bioremediation processes, where specific microbial communities can facilitate the breakdown of these chemicals into less harmful substances. Understanding these dynamics is crucial for developing sustainable agricultural practices that minimize the environmental impact of pesticide use. Furthermore, as agricultural practices evolve, the push for integrated pest management (IPM) strategies emphasizes the need for reducing reliance on chemical pesticides. By incorporating biological control methods and optimizing pesticide application practices, farmers can enhance crop protection while mitigating the adverse effects on soil health and biodiversity. This holistic approach not only helps maintain crop yields but also fosters a healthier ecosystem, underscoring the importance of balancing agricultural productivity with environmental stewardship. In conclusion, while pesticides remain vital for protecting crops in modern agriculture, understanding their degradation in the soil and the environmental factors that

influence this process is essential. Continued research into the interactions between pesticides and soil ecosystems will contribute to more sustainable agricultural practices, ensuring that crop protection measures do not compromise environmental health.

CONCLUSION

In a nutshell, this investigation reveals the remarkable resilience of specific fungi in the face of Cheminova exposure, underscoring their potential as agents of bioremediation. *A. flavus* stands out as the most vigorous species, exhibiting notable growth and biomass production across the tested concentrations, suggesting its promising role in detoxifying contaminated environments. However, the significant reduction in fungal viability at 60 ppm highlights a critical toxicity threshold that warrants attention in agricultural practices. This study not only deepens our understanding of the interactions between pesticides and soil fungal communities but also emphasizes the necessity for sustainable approaches in managing agricultural chemicals, particularly in regions like India where sugarcane cultivation is prevalent. Ultimately, these findings advocate for further exploration into harnessing fungal capabilities to mitigate environmental hazards linked to pesticide usage.

REFERENCE

1. Greaves MP, Davies HA, Marsh JA, Wingfield GI, Wright SJ. Herbicides and soil microorganisms. CRC critical reviews in microbiology. 1976 Jan 1;5(1):1-38.
2. Domsch KH. Effects of pesticides and heavy metals on biological processes in soil. Plant and Soil. 1984 Feb;76:367-78.
3. Wainwright M. Effects of fungicides on the microbiology and biochemistry of soils—a review. Zeitschrift für Pflanzenernährung und Bodenkunde. 1977;140(5):587-603.
4. Schuster E, Schröder D. Side-effects of sequentially-applied pesticides on non-target soil microorganisms: field experiments. Soil Biology and Biochemistry. 1990 Jan 1;22(3):367-73.
5. Lewis JA, Papavizas GC, Hora TS. Effect of some herbicides on microbial activity in soil. Soil Biology and Biochemistry. 1978 Jan 1;10(2):137-41.
6. Martin JP. Influence of pesticides on soil microbes and soil properties. Pesticides and their effects on soils and water. 1966 Jan 1;8:95-108.
7. Zhong, W.H. and Cai, Z.C., 2007. Long-term effects of inorganic fertilizers on microbial

biomass and community functional diversity in a paddy soil derived from quaternary red clay, applied soil ecology, 36: 84-91.

8. Von Arx, J.A., 1974. The Genera of Fungi Sporulating in Pure Culture 2nd edn., Vaduz Germany: A.R. Ganter, Verlag, KG. FL-9490, 375 pp.
9. Anisworth, G.C., Sparrow, F.K. and Sussman, A.S., 1973. "The fungi - An advanced treatise: A taxonomic review with keys." In: *Ascomycetes and Fungi Imperfecti*. New York: Academic Press, **4**(4): 621.
10. McDougall, J and Woodburn, J., Agrochemical review, county Natwest Woodmac, London, UK (1989). Moorrnan TB. (1989). A review of pesticide effects on microorganisms and microbial process related to soil fertility. *Journal Prod. Agric* **2**(1):14-23.
11. Hassal, K., The chemistry of pesticides, The Macmillan Press, Ltd., London, UK (1989)
12. Ou, L. T., Gancarz, D.H., Whwwler, W.B., Rao, P.S.C., and Davidson, J.M., "influence of soil temperature and soil moisture of degradation and metabolism of carbofuran in soil,*J. Environ*. **11**, 293-298 (1982).
13. Smith, A.E and Walker, A. "A quantitative study of asulam persistence in soil," Pestic. Sci., 8, 449-456.
14. Srinivasan, R (2012) Integrating biopesticides in pest management strategies for tropical vegetable production. J Biopest, 5 (Supplementary): 36-45.