

**ROLE OF PAECILOMYCES IN RAPID COMPOSTING AND
PLANT DISEASE MANAGEMENT**

by

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CERTIFICATE

This is to certify that the thesis entitled “*Role of Paecilomyces in Rapid Composting and Plant Disease Management*” submitted by Ms. Kalpana Arora has been prepared under my guidance with the rules and regulations of Indian Institute of Technology Delhi, India. The research report and results presented in this thesis have not been submitted for any degree or diploma in any other institute or university.

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ABSTRACT

*There has been a rapid industrialization and urbanization in India in the last few decades, which has led to improvement in the living standards and change in the production and consumption patterns. Consequently, the nature and volume of the Municipal solid waste has changed considerably. Thus demands a suitable and cost effective technology for the environmentally safe treatment and disposal of MSW. Composting is a promising method of waste management to stabilize solid waste not only to produce compost but also keep environment pollution free. Bioinoculants play an important role in nutrient recycling, rapid composting and also act as biocontrol agent in crop production. Since, modern farming practices involve extensive usage of chemical fertilizers and pesticides which result in land and water pollution and also affect the human health and other life forms severely., these practices should be substituted by alternative agricultural practices, which vitally reliant upon the proper and regular addition of organic manure and utilizing the biofertilizers and biopesticides. So it was considered of interest to explore the possibility of using lignocellulolytic fungus *Paecilomyces variotii*, nitrogen fixing and phosphate solubilizing bacteria along with the earthworms to fasten the rate of MSW stabilization and improve the quality of compost also having biopesticidal value.*

Therefore, in search of an integrated system for rapid composting of MSW and plant disease management, present study was taken up with following specific objectives:

- To screen and identify cheaper and easily available C and N sources for the multiplication of *P.variotii*.*
- To assess the potential of *P. variotii* along with efficient micro organisms (*Pseudomonas* and *Azotobacter*) and earthworms in rapid composting.*
- To study the role of *P.variotii* as biocontrol agent against *Fusarium* and *Verticillium* causing wilt diseases in tomato.*
- To prepare the formulations for rapid composting and controlling the selected pathogen.*

*Prior to the composting experiments, the growth parameters of the *P. variotii* were studied and interaction among all the bioinoculants (Efficient microbes and earthworms) and selected pathogens was tested. The optimum temperature, pH and relative humidity (RH) for *P. variotii*'s*

best growth on MSW were identified as 30°C, 5 and 70% respectively. The existence of 'functional compatibilities' among all the three microbes (*P. variotii*, *A. chroococcum* and *P. fluorescence*) and earthworms (*Eisenia fetida*) made them suitable to employ in rapid composting of MSW. Also, the antagonistic activity of *P. variotii* against *F. oxysporum* and *V. dahliae* (visible by inhibition zone) provided the possibility to use it as biocontrol agent against *Fusarium* and *verticillium* wilt of tomato.

P. variotii alongwith *Pseudomonas* and *Azotobacter* with earthworms improved the quality and quantity of compost over other treatments and decreased ~50% time of composting. The reduction in percentage of C:N ratio (25.48 to 7.01), cellulose (up to 6.38%), hemicelluloses (18.46%) and lignin content (16.46%) and increase in percentage of TKN (up to 2.5 %), phosphorus (up to 1.7%) and potassium (up to 1.8%), were noted within 60 days of composting. FTIR studies revealed the decrease in TOC content by showing the decrease in peaks of different C- moieties present in the compost.

The doubling time of *P. variotii* was found to be 8.2h and 90mg/L of bioactive metabolite, Dipicolinic acid (DPA) was produced. The growth analysis of fungus *P. variotii* on nitrogen rich non edible oil cakes (*Jatropha*, *Karanja*, *Neem* and *Mahua*) showed better results than Potato Dextrose Agar (PDA), the growth media commonly used. The growth rate of *P. variotii* was 0.42 cm/ day on *Jatropha* cake while on PDA it was 0.26 cm/day. Similarly, spore count was also enhanced from 2.5×10^9 of control to 5.1×10^{10} of *Jatropha* cake. In this context, it may be noted that *Jatropha*, *Karanja* and *Neem* oil cakes when amended with dextrose were found superior to PDA. However, the performance of *Mahua* cake was not found satisfactory. The mass cultivation of the *P. variotii* on the non edible oil cakes with their various combinations with saw dust proved the feasibility of these oil cakes as a substrate in SMF. The best treatment was *Karanja* oil cake (70%) with saw dust producing the spores up to 7.2×10^{12} per gram of substrate. The pot trials to control *F.oxysporum* infection in tomato plants with the formulated product were efficient.

Possibilities were explored to increase the DPA production by utilizing these non edible oil cakes and different carbon sources by experimental designing for medium optimization with Response Surface Methodology (RSM). The DPA production has increased by 2.7 times on

optimized the medium consisted of non edible oil cakes as N sources (*Jatropha* and *Karanja* cakes optimized at 20g/L and 45g/L respectively) and dextrose (10g/L).

The non edible oil cake based formulation of *P. variotii* significantly reduced the % of disease incidence of tomato plants as compared to control. The treatment improved plant health by reducing wilt symptoms, vascular invasion and sporulation of the pathogen and decreasing the disease incidence (17.2% for *F. oxysporum* and 18.83% for *V. dahliae*). Minimum Inhibitory Concentrations (MIC) of the fungal extract (*P. variotii*) was observed with the 10^{-3} dilution for both *F. oxysporum* and *V. dahliae*. MIC of DPA was detected as 0.25 mg/mL for *F. oxysporum* and 0.21 mg/mL for *V. dahliae*.

Real time PCR studies provided an insight to the molecular mechanism involved in the biocontrol of *F. oxysporum*. As *P. variotii* produces certain bioactive compounds eg. DPA, which might have inhibited FOW 1 expression in *F. oxysporum*. Real Time analysis for the FOW1 gene which is responsible for the colonization of the *F. oxysporum* showed that it was down-regulated linearly on a time course. The attenuation of PGI gene (responsible for endopolygalactouronase) was also seen. Enhancement in the expression of CRE1 showed that *P. variotii* produce some bioactive substance (DPA) which may have inhibited FOW1 gene expression in *F. oxysporum*. Also, in concert with the over expression of CRE 1 gene and PGI gene is down regulated which further decrease the FOW1 expression.

The present study clearly reveals the possibilities to use the *P. variotii* with other bioinoculants successfully to enhance the rate of biodegradation and improve the quality of compost from MSW. Moreover, as far as the economics of this system is concerned it is obviously much cheaper as compared to the other composting processes which involve mechanical turning, aeration and so on. The experimental findings to exploit the antagonistic activity of *P. variotii* against *Fusarium* and *Verticillium* wilts proved it as promising biocontrol agent as an efficient, environmentally safe alternative.

CONTENTS

	Page no.
Certificate	i
Acknowledgements	ii
Abstract	iv
Contents	vii
List of figures	xiii
List of tables	xv
List of plates	xvii
Chapter I: Introduction	
1.1 Background	01
1.2 Waste generation: Global problem	02
1.3 MSW generation and its composition: The Indian scenario	03
1.3.1 Composition of MSW	05
1.4 Environmental impacts of MSW	05
1.4.1 Human health impacts of MSW	07
1.5 Solid waste management technologies	08
1.5.1 Rapid composting technologies	10
1.6 Compost for pathogen control	13
1.7 Biocontrol of Plant Diseases	13
1.8 Fusarium and Verticillium wilt	15
1.9 Sustainable Agriculture	16
1.10 Scope of work	18
1.10.1 Objectives	19
Chapter II: Review of Literature	
2.1 <i>Paecilomyces variotii</i> : Potentials and Applications	20
2.1.1 Rapid Composting	21
2.1.2 Enzyme Production for Industrial Purposes	23
2.1.3 Biocontrol Agent	23
2.1.4 In Bioremediation	24
2.1.5 As Phosphate Solubilizer	25

2.1.6	Other Applications	25
2.2	Solid waste generation and its potentials	26
2.2.1	Solid waste and its classification	26
2.2.2	Municipal Solid Waste	28
2.2.3	MSW characteristics and composition	29
2.2.4	Technologies for MSW management	31
	2.2.4.1 Landfilling	31
	2.2.4.2 Incineration	32
	2.2.4.3 Pyrolysis and pelletization	32
	2.2.4.4 Biomethanation (Biogas) technology	33
	2.2.4.5 Composting	33
2.3	Composting Technology	35
2.3.1	Rapid Composting	36
2.4	Quality characteristics of compost	41
2.4.1	FTIR analysis of compost	44
2.5	Non edible oil cakes as substrate for the fungal mass cultivation	46
2.5.1	Application of Experimental designing in medium optimization	48
2.6	Plant Disease Management	50
2.6.1	Types of Plant Diseases	50
2.6.2	Control measures	50
2.7	Biocontrol of Plant Diseases	52
2.7.1	Mechanism of Biocontrol	52
2.8	Fusarium wilt of tomato	56

2.8.1	Life cycle of <i>F. oxysporum</i>	57
2.8.2	Disease development	60
2.9	Verticillium wilt of tomato	60
2.9.1	Host specificity	60
2.9.2	Disease cycle and development	61

Chapter III: Materials and Methods

3.1	Selection of Microbial species (beneficial and pathogenic) for Rapid composting and plant disease management	63
3.2	Procurement of Microbial cultures	64
3.3	Culturing and subculturing of microbial strains	64
3.4	Growth of <i>P.variotii</i> on MSW as substrate under different temperature, moisture content and pH.	64
3.4.1	Effect of Temp.	64
3.4.2	Effect of moisture content	66
3.4.2	Effect of pH	66
3.5	Interaction between different selected microbes and bioinoculants	66
3.5.1	Microbe-microbe interaction	66
3.3.2	Microbe-earthworm interaction	66
3.6	Rapid composting of MSW employing bioinoculants	67
3.6.1	Effect of different combinations of bioinoculants on compost	67
3.7	Analysis of compost	68
3.7.1	Chemical analysis	68
3.7.2	FTIR	68

3.8	Media preparation using non edible oil cakes	68
3.8.1	Solid medium	68
3.8.2	Broth preparation	69
3.9	Fungal biomass estimation	69
3.9.1	Assay of Fungal Growth and Sporulation	69
3.9.2	Dry Weight Determination of Fungus	69
3.10	Optimizing the non edible oil cake concentration by Completely Randomized Design (CRD)	70
3.11	Preparation of <i>P. variotii</i> filtrate samples for chemical analysis	70
3.11.1	Qualitative analysis	70
3.11.2	DPA estimation	71
3.12	Experiment design for increasing the DPA production	71
3.12.1	Identifying the significant variables using Plackett–Burman Design (PBD)	71
3.12.2	Optimization of the selected C and N source concentration by RSM	72
3.12.3	Software for experimental design	73
3.13	Solid-State Fermentation	73
3.14	Assessment of Phytotoxicity (PT)	74
3.15	Pot Experiments to check the biocontrol efficacy of SSF product	75
3.16	Determination of Pathogen infection	76
3.17	Biocontrol efficacy of <i>P. variotii</i> against <i>F. oxysporum</i> and <i>V. dahliae</i>	77
3.17.1	Plant Materials and Growth Conditions	77
3.17.2	Pathogenic <i>F. oxysporum</i> and <i>V. dahliae</i> inoculums preparation	77
3.17.3	Plant harvest and growth analysis	78
3.17.4	Plant Biomass	78
3.17.5	Biochemical analysis	78
3.18	Molecular mechanism of biocontrol	79
3.18.1	Determination of Minimum Inhibitory Concentrations (MICs) of <i>P. variotii</i> filtrate and DPA against plant pathogens	79
3.18.2	Determining the Mechanism of Infection and its Virulence gene attenuation by <i>P. variotii</i> through Real Time PCR	79

Chapter IV: Results and Discussion

4.1	Optimization of growth conditions for <i>P. variotii</i>	83
4.1.1	Effect of different temperature on the growth of <i>P. variotii</i>	83
4.1.2	Effect of different pH on the growth of <i>P. variotii</i>	85
4.1.3	Effect of different relative humidity on the growth of <i>P. variotii</i>	85
4.2	Lab level studies on the interaction between selected microbes, plant pathogens and earthworms	88
4.2.1	Interactions among beneficial organisms	88
4.2.1.1	Microbe-microbe interaction	89
4.2.1.2	Microbe-earthworm interaction	90
4.2.2	Interaction between <i>P. variotii</i> and plant pathogens	94
4.3	Role of <i>P. variotii</i> in process efficiency (composting and vermicomposting)	95
4.3.1	Physiochemical characteristics of the substrate used for rapid composting	95
4.3.2	pH and EC	96
4.3.3	Total organic carbon (TOC)	100
4.3.4	Total Kjeldhal Nitrogen (TKN)	100
4.3.5	Cellulose, hemicelluloses and Lignin	102
4.3.6	Phosphorous and Potassium	105
4.3.7	FTIR analysis of Vermicompost at different time intervals (Treatment T1: <i>P. variotii</i> + <i>Ps. fluorescence</i> + <i>A. choroococcu</i>)	109
4.3.8	Chemical Changes during rapid composting of MSW by treatment T1 (field level trial)	114
4.4	Growth kinetics of <i>P. variotii</i> during submerged fermentation	119
4.5	Culturing of <i>P. variotii</i> on non traditional substrates:	123

4.5.1	Growth of <i>P. variotii</i> on non-edible oil cakes	123
4.5.2	Medium optimization for biopesticidal DPA production by <i>Paecilomyces variotii</i> using response surface methodology (RSM)	135
4.5.2.1	Screening of important factors by Plackett-Burman design for DPA production	135
4.5.2.2	Optimization of selected C & N sources by RSM using BBD	136
4.5.2.3	Verification of the predicted concentration in the optimal medium	144
4.5.2.4	Morphological alterations of <i>P. variotii</i>	144
4.5.3	Mass Cultivation of <i>P. variotii</i> on various non edible oil cakes (in different combinations with sawdust) and its efficacy against <i>Fusarium</i> wilt of tomato	150
4.6	Plant disease management through <i>P. variotii</i>	161
4.6.1	Disease incidence	164
4.6.2	Change in growth parameter	165
4.6.3	Change in metabolic activities	166
4.7	Molecular approach to identify the mechanism of biocontrol	177
4.7.1	Minimum Inhibitory Concentration of <i>P. variotii</i> filtrate and DPA activity against <i>F. oxysporum</i>	177
4.7.2	Gene expression analysis by qRT-PCR	178
Chapter V: Summary and Conclusions		
5.1	Performance and efficacy of bioinoculants (<i>P. variotii</i> , <i>A. chroococcum</i> , <i>P. fluorescence</i> and <i>E. fetida</i>) for MSW treatment	191
5.2	Mass production of <i>P. variotii</i> and its application as biocontrol agent against <i>Fusarium</i> and <i>Verticillium</i> wilt.	192
References		196
Bio-data		222