BIOREMEDIATION OF CHLORPYRIFOS CONTAMINATED SOIL USING BIOSTIMULATION AND PHYTOREMEDIATION TECHNIQUES

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by

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Submitted

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Dedicated to my loved ones

CERTIFICATE

This is to certify that the thesis entitled “**Bioremediation of Chlorpyrifos Contaminated Soil Using Biostimulation and Phytoremediation Techniques**” being submitted by **Ms. Neeru Kadian** to the Indian Institute of Technology Delhi for the award of the degree of “Doctor of Philosophy” is a record of bonafide research work carried by her. She has worked under our guidance and supervision, and has fulfilled the requirements for the submission of this thesis. To the best of our knowledge the results contained in this thesis have not been submitted in part or full to any other University or Institute for award of any degree or Diploma.

(Dr. Anushree Malik) (Prof. Santosh Satya)

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ABSTRACT

Present study is an attempt to develop a suitable strategy for bioremediation of chlorpyrifos (CPF) contaminated soil by integrating the biostimulation and phytoremediation routes. To begin with, the biostimulation potential of four different organic amendments (farmyard manure, biogas slurry, mushroom spent compost and vermicompost) at 1% dose (designated as FM, BS, MS and VC, respectively) in chlorpyrifos (10 ppm) spiked soil was evaluated at 37°C for 74 days. Organic amendments (except farmyard manure) enhanced the microbial activity (dehydrogenase activity, DHA) over unamended CPF contaminated soil. A new GC method for simultaneous quantification of CPF and its major metabolites (TCP and oxon) has been developed. Substantial enhancement in chlorpyrifos degradation over the unamended spiked soil (SP) was observed in VC (30%) and MS (18%), which also depicted higher DHA. Similar results were obtained under ambient conditions also but increase in vermicompost dose (3% and 5%) did not induce shortening of the half life of CPF indicating that its utilization at 1% dose (normal agronomical practice) could be recommended.

Next, to evaluate the phytoremediation potential, four common plants {marigold (Tagetes erecta), canna (Canna indica), cactus (Cereus spp.), and berseem (Trifolium alexandrinum)} were chosen for pot experiments at 7 ppm chlorpyrifos concentration. In the canna (C) and marigold (M) setup, at 90 days the degradation of CPF was accelerated from 55.5% (SP) to 77.9% (M2) and 82.4% (C2), respectively. Planted soil also depicted overall higher microbial activity as compared to unplanted soil, once the plants were properly established. The overall DHA and chlorpyrifos degradation trends were analogous i.e. C2>M2>SP. In the next set of cactus and berseem experiments, cactus brought about maximum CPF degradation (44.5%) in the initial 2 days as compared to just 20% in case of berseem. However, in the latter phase, berseem performed better and overall trend of CPF degradation and DHA was observed as: berseem > cactus > unplanted soil.

In the integrated phytoremediation design, field experiments (3m × 3m plots) were conducted incorporating vermicompost and canna plant along with the knol khol vegetable crop to accommodate soil sampling at different intervals with minimum disturbance. Except T0 (soil spiked with 10 ppm chlorpyrifos, control), all the treatments contained 1% vermicompost and designated plants i.e. T1 (knol khol), T2 (canna), and T3 (canna and knol khol). Treatments T2 and T3 showed comparatively higher (99.3%) chlorpyrifos degradation

on 65th day with almost same residual pesticide (0.07 ppm) whereas T0 (control) and T1 (knol khol) showed ≈ 2.5 and 1.8 times more residual CPF in soil, respectively. Studies on the fate of major metabolites revealed that the major route of CPF degradation in soil was via TCP with correlation coefficients as T2 (0.84) > T1 (0.71) > T3 (0.68) > T0 (0.34). In case of oxon, an erratic pattern was observed characterized by frequent appearance and disappearance. Eventually, TCP and oxon declined in all the treatments incorporating canna plants. Special characteristic of canna to harbor more microbial flora as well as to regulate desirable microbes in each phase could probably facilitate breakdown of chlorpyrifos as well as TCP and oxon quite fast as compared to knol khol plants.

GC-MS analysis revealed that knol khol fruit body had higher number of metabolites as compared to canna. GC analysis of the harvested knol khol fruit revealed that oxon and TCP were present in produce from both the sole plot (T1) as well as mixed plot (T3) whereas canna plants (T2) contained only TCP. Finally, the integration of canna in knol khol (T3) reduced the oxon and TCP concentration by ≈ 10% in both the cases. Considering the plant density in T3 close to normal practices, the yield of main crop comes out to be statistically at par with sole vegetable crop plot (T1). Hence such an integration of phytoremediators with vegetable crop proves to be a positive step in decreasing the residual pesticide and their metabolites in the final produce. Based on the research findings of field experiments for vegetable crop (knol khol), a remediation package for bioremediation of CPF contaminated soil has been developed by integrating vermicompost and canna plant with knol khol crop. As indicated in the newly developed remediation package, with judicious planning and management such an integrated phytoremediation design would be quite feasible without compromising on the plant density.

Academically, the results constitute a pioneering effort on remediation of chlorpyrifos using organic amendments and phytoremediators as well as testify the integration of the above with vegetable crop in actual field condition. Also, it represents a novel insight on fate of chlorpyrifos in terms of its metabolites during bioremediation. In fact the study provides guidelines for further work addressing complex system under actual field practices where successive sprays of single/multiple pesticides are generally done. On the practical grounds, the suggested technology package is an eco-friendly and compatible approach for detoxification of pesticide contaminated soil for ensuring food safety in the long run.

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