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BIODEGRADATION OF CHLORPYRIFOS BY BACTERIAL STRAIN ISOLATED FROM SOIL CONSORTIA

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ABSTRACT:

The main economy of the India depends on the agriculture. Farmer reliable on the agriculture as a source of income. In order to take more crops the vast variety of the pesticides are used. Pesticides have sold and used without restriction in developing country. Amongst that organophosphate (Ops) account for 38% of the total pesticide used globally. OPs have the broad spectrum activity against the wide range of the pest and insects. But the prolonged used of the Ops can cause nerve and muscular disorder and it also contaminate the ecosystem .To overcome this problem the bioremediation is the convenient and cheap solution. The present review discuss the biodegradation of the chlorpyrifos by the bacteria strain isolated from the soil consortia.

Key words: - Chlorpyrifos, Pesticide, Organophosphate, Biodegradation

USE OF PESTICIDE IN AGRICULTURE.

In order to meet the increasing demand of food in developing country, vast variety of the pesticides have been used to protect the plants, crops from the various pests, insect. Pesticides are such a chemical which protects the plants, crops from the detrimental effect of the pest by controlling, inhibiting or by killing them. Pests can reduce the quality and quantity of the crops, can affect the cost of farming. Pesticides are therefore used for to overcome such a situation. (Jayashree and Vasudevan 2007).

USE OF CHLORPYRIFOS IN AGRICULTURE.

Chlorpyrifos (O,O diethyl o-(3,5,6- trichloro-2 pyridyl)phosphorothioate is the organophosphate pesticide having the broad spectrum activity against the pest and insects, it is moderately toxic in nature (Xu et al. 2007) (Figure 1).CP widely used for the pest control on Wheat, Soyabean, cotton, Rice, Chickpea (Fang et al 2008).It is effective against a variety of chewing and sucking insects, flies and Mosquito on economically important crops such as cotton, wheat, rice, vegetables, citrus fruits etc (K.D. Racke 1993) Chlorpyrifos acts on pests primarily as a contact poison with some action as a stomach poison. It is available as a granules, wettable powder, dusting powder and emulsifiable concentrate (Mallick et al., 1999).

The physical and chemical properties of chlorpyrifos are given in the table 1.

PERSISTENCE NATURE OF THE CHLORPYRIFOS

CP have the high soil absorption coefficient and low water solubility (2 mgL-1) Racke 1993.It is therefore immobile in soil and unlikely to leach or to contaminate the ground water (Ajaz et al.,2005)CP can remain effective up to 5-17 years since it is resistance to biodegradation (Baskaran et al 1999) . The persistence studies of B.K.Singh and co-workers recognized the half-life of CP from 36-36 days in the soil.(B.K.Singh et al.,2002) This persistence is depends in the pH of the soil climate condition and other factors of the soil and may range from one week to one year (K.D. Racke 1993).The soil with slightly alkaline pH reported to degrade CP In 90 days.

Principle metabolites of the CP is TCP (3,5,6,-trichloro-2-pyridinol) has the antimicrobial

activity and prevents the growth of the CP degrading bacteria (Racke et al., 1990).

TOXIC EFFECT OF THE CHLORPYRIFOS

According to standard definition of the pesticide, it is the substance that are poisonous and affect to the target organism and it is safe for the non-target organisms and the environment.(Dhiraj Sud 2020). CP is not selective in nature, therefore the toxic nature of the Cp is not restricted to the targeted organisms butalso toxic to non-target organism including vertebrates. The accidental spillage of the chlorpyrifos to the aquatic environment could negatively impact on the aquatic animal. (Russel L Carr et al., 1997).

Chlorpyrifos is moderately toxic to humans. The main mode of action of the chlorpyrifos is the phosphorylation and inactivation of the acetylcholinesterase (ACGIH 1991) the enzyme required for proper nerve functioning. The poisoning from the chlorpyrifos can affect the nervous system, cardio vascular system and respiratory system. The acute exposure of the organophosphate or the cholinesterase inhibiting compounds may results into numbness, tingling sensation, incoordination, headache, dizziness, nausea, abdominal cramps, sweating, blurred vision, and slow heartbeat. Very high doses may results in unconsciousness, incontinence and convulsion or fatality (Eaton, 2008)

Incorporation of the chlorpyrifos in human may results in neural disorder, inhibition of DNA synthesis interference with gene transcription altered synaptic function (Lakshmi et al., 2009)

Chlorpyrifos readily absorbed into the bloodstream through gastrointestinal tract if it is ingested, through lungs if it is inhaled, or through the skin if there is direct exposure or skin contact. In human chlorpyrifos eliminated through the kidney. The principle metabolites are also eliminated rapidly. Chlorpyrifos is moderately toxic to the birds but it is highly toxic to the aquatic animals such as fresh water fish, aquatic invertebrates and marine organisms. The use of the chlorpyrifos can be hazardous to wildlife and honeybees (Cho et al., 2004)

BIODEGRADATION OF CHLORPYRIFOS

The degradation of pesticide can be carried out by the different mechanisms which include Physical, chemical, and biological agents which play significant roles in the transformation of insecticide, herbicide, and fungicide molecules to various degradation products.

Transformation mechanisms include oxidation, hydrolysis, reduction, hydration, conjugation, isomerization, and cyclization. Resultant products are usually less bioactive than the parent pesticide molecule, but numerous cases have been documented of metabolites with greater bioactivity. The physical and chemical properties of the degradation products are also different from those of the parent compound, and their fate and significance in the environment also are altered with the structural changes. The concept of "environmental activation" is introduced, to describe the transformation of a pesticide to a degradation product that is of significance in the environment as a result of its environmental toxicology or chemistry.(Joel R. Coats 1991)

The above study reveals the toxic impact of CP on non-targeted organisms, even serious damage to human health and other organisms in an environment. Hence there is need to degrade CP and its metabolites from the environment. The degraded product or the metabolites of the CP should be nontoxic or less toxic than the chlorpyrifos. It was found that Chlorpyrifos- Oxon the major metabolite of Chlorpyrifos is much more toxic than the Chlorpyrifos itself (D.W.Sparling et al.,2007) So the analysis of the degraded product is also necessary.

CHLORPYRIFOS BIODEGRADATION BY NATURAL PROCESS (PHYSICAL PROCESS)

Chlorpyrifos in natural environment undergoes adsorption, hydrolysis, oxidation or photolysis.

The photolytic experiment involves direct treatment of CP in sunlight or ultraviolet light. It has been reported photolysis of CP at different pH of the soil and different temperature have the different fate. Alkaline soil/ condition pH 8 cause the higher degradation of the CP than the Acidic soil/condition pH (5.5). The degradation rate is higher at higher temperature (400 C) than that of the low temperature (220 C) (B.Maroune et al.,2015).

MICROBIAL DEGRADATION OF CHLORPYRIFOS

Microbial degradation is carried out by the Microorganisms such as bacteria and fungi. Most of the bacteria cannot survive due to toxic effect of the pesticide. But most of them are evolved such that they can use the pesticide as a soul carbon and phosphorus source.

BACTERIAL BIODEGRADATION OF CHLORPYRIFOS

It has been reported that cp is degraded co-metabolically by certain bacteria in liquid media but several attempts to isolate CP degrading bacteria is not successful (Racke et al., 1993; Mallick et al., 1999).

Several study have been made to isolate the chlorpyrifos degrading bacteria, the use of Microorganism is the efficient method for the degradation of the chlorpyrifos. The study shows that the pseudomonas sp. have the highest capacity to degrade chlorpyrifos and its metabolites(TCP and DEP) to nontoxic compound like CO2, H2O. Amongst the various Pseudomonas species, P.putida MAS -1 is reported to be more efficient.(R.A.Gilani et al., 2016). Singh et al first isolated the CP degrading bacterium Enterobacter B-14 which hydrolyze the CP to diehtylthiophosphate (DETP). This DETP then can be used as a source of energy. (Yang et al., 2006; Li et al., 2007).Alkaligenes faecalis is also capable of degrading chlorpyrifos and results into the formation of TCP (Yang et al., 2005).

Enterobacter strain B-14, Stenotrophomonas sp. G1, and Sphingomonas sp.DSP -2 have been studied. Sphingomonas sp.DSP -2 rapidly degraded CP in alkaline medium. Cp was degraded up to 98.7% in soil with pH8.7. However only 58.1% degradation was achieved in soil with pH 4.8 and 90% degradation was achieved in the neutral medium.(X Li et al.,2007).

Enterobacter strain B-14 utilizes Cp as a soul source of carbon and phosphorus. The degradation nearly complete in two days at temperature 350C in mineral salt medium inoculated with nitrogen. The addition of glucose and succinate delayed the degradation process.(B.K.Singhet al.,2004).Bacterial strain Stenotrophomonas sp. G1species degraded 63% of CP (50 mgL-1) at temperature 400C.This strain not only degrade CP but also other OPs like methyl parathion, diazinon, profenofos and triazophos, parathion , methyl paraoxon effectively.(S.Deng et al 2015)

Chlorpyrifos has been reported to be degraded co metabolically in liquid media by Flavobacterium sp. and also by an Escherichia coli with an opd gene (Singh et.al. 2004)Arthobacter sp. strain B-5 hydrolyzed chlorpyrifos depending on the concentration of the substrate. Chlorpyrifos (10mg/L) was completely degraded in the mineral salts medium by FlavobacteriumSp. ATCC 27551 and by Arthobacter sp. for 48 hr respectively. (Xu, 2007).

Pseudomonas aeruginosa, Bacillus cereus, Klebsiella sp., and Serratiamarscecens obtained from consortia showed 84, 84, 81, and 80% degradation of chlorpyrifos (50 mg/L) in liquid medium after 20 days and 92, 60, 56, and 37% degradation of chlorpyrifos (50 mg/L) in soil after 30 days. Some recent reports indicate bacterial degradation of chlorpyrifos by Flavobacteriumsp. ATCC 27551 and Arthrobacter sp., isolated from contaminated sources, which degrade

chlorpyrifosco-metabolically, and Enterobacter strain B-14, Alcaligenes faecalis, and Klebsiella sp., which degrade and utilize chlorpyrifos as sole carbon source (Jilani S. and Khan, 2004). Bacillus sp. And Micrococcus sp. possess potential to degrade chlorpyrifos (Getzin, 1981; Gomez et al., 2007).

FUNGAL BIODEGRADATION OF THE CHLORPYRIFOS

Recent study shows that acremonium sp. strain (GFRC-1) enriched with carbon and nitrogen could degrade CP (300 mg L-1) with an ease of 84% degradation achieved in 20 days. (G. Kulshrestha et al., 2011). Certain soil fungi Trichoderma viride and Aspergilus niger has also been reported Chlorpyrifos degradation.(Hussain S. et al.,2007). The study shows that Phanerochaete chrysosporium, Aspergillus terreus, and Verticillium sp also have the chlorpyrifos degrading activity.(17) Fungal degradation of chlorpyrifos was reported by Verticillium sp. DSP in pure cultures and its use in bioremediation of contaminated soil (Xu, 2007).It has been reported that Verticillium sp. and Brassica chinensis can degrade the chlorpyrifos in culture medium ranging from 1-100mg L-1(Arisoy, 1998; Trejo and Quintero, 2000; Bhalerao and Puranik, 2007).

The chlorinated pyridinyl ring of chlorpyrifos undergoes cleavage during biodegradation by P. chrysosporium. But the degradation of chlorpyrifos proves more efficient by mixed populations than by pure cultures of fungi. Mixed population of fungi, such as Alternaria alternata,Cephalosporium sp., Cladosporium cladosporioides, Cladorrhinum brunnescens, Fusarium sp., Rhizoctonia solani, and Trichoderma viride, reveal the degradation of chlorpyrifos in liquid culture more efficiently (Singh et al., 2004)

ENZYMATIC BIODEGRADATION OF CHLORPYRIFOS.

Enzymatic degradation of the pesticide has gained a lot of interest. Large number of aquatic species produces enzymes that are capable of hydrolyzing many organophosphate pesticides. These enzymes are organophosphorus acid anhydrases. They are also referred as paraoxonase, easterase phsophotriesterase, diisopropyl fluorophosphatase, somanase, and parathion hydrolase (Liu et al., 2001).Organophosphorus acid anhydrases can hydrolyze a wide variety of organophosphorus acetylcholinesterase inhibitors (Liu et al., 2001).Various researchers highlighted the presence of oph gene in the cells of microbes that degrade organophosphorus compounds and hydrolase as the chief enzyme behind the process (Mulbry and Karns, 1989; Ohshiro et al., 1997; Cui et al., 2001; Liu et al., 2001; Gao et al., 2012).

CONCLUSION

The studies shown that Chlorpyrifos is effective against the target organisms as well as the non targeted organisms also it disturb the ecosystem through the accumulation. Therefore the degradation of the chlorpyrifos is necessary for the safety of human being and the environment. Bioremediation is the cheap and convenient method to get rid of toxic effect of the Chlorpyrifos but it take longer duration and controlled condition. The molecular characterization and the mechanism of biodegradation need to study at the genetic level. Plant growth promoting rhizobacterial can be used as an alternative which promotes the growth of the plants and the biodegradation of the Organophosphate pesticides. The synergistic activity and cometabolism need to be study with different bacteria. This may give the better results.

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Table 1.Physical and chemical properties of CP.

Property	Value
Chemical Name	O,O-diethyl O-(3,5,6-trichloropyridine-2-yl) thiophosphate
Molecular and Emperical formula	$C_9H_{11}C_{13}NO_3PS$
Molecular weight	350 a.m.u.
Physical State	Solid
Colour	White
Melting Point	41-42^0 C
Decomposition Temprature	160^0 C

Fig.1 Chemical structure of Chlorpyrifos

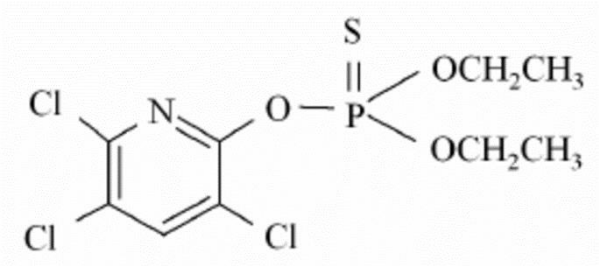

Figure 2 .Metabolic pathway for the bacterial degradation of chlorpyrifos