

REDISCOVERING THE PAST TO FUTURE BIODEGRADATION OF MONOCROTOPHOS: A REVIEW

Eljeeva Devakumari T¹*, Raja M² and Jisha Hannie Selson³

1&3*Research Scholar, ²Assisstant Professor
Department of Advanced Zoology and Biotechnology,
Loyola College, Chennai, Tamilnadu, India
*Corresponding author: eljeevaprabakaran@gmail.com

Abstract: Monocrotophos (MCP) is a widely used organophosphate pesticide that raises substantial environmental and health concerns because of its persistence and high toxicity (USEPA, 2006). Microbial degradation presents a sustainable and effective method for detoxification of this compound (Kaur & Goyal, 2019). Microorganisms are able to bioremediate pesticides using diverse metabolic pathways where enzymatic degradation plays a crucial role in achieving chemical transformation of the pesticides. This review comprehensively examines the biodegradation mechanisms of monocrotophos, detailing the microbial strains and enzymatic processes involved in its detoxification. The growing concern about the environmental and health impacts of pesticides is pushing the industry to develop more sustainable alternatives, such as highly biodegradable chemicals. The degradative properties of microorganisms could be fully exploited using the advances in genetic engineering and biotechnology, paving the way for more effective bioremediation strategies, new technologies, and novel applications. This review presents a comprehensive analysis of microorganisms and enzymes for monocrotophos degradation, while also discussing the challenges and future perspectives in bioremediation of this persistent pesticide.

Index Terms - Monocrotophos, Microbial degradation, Enzymes, Bioremediation.

I.INTRODUCTION

The extensive use of pesticides like monocrotophos in agriculture has resulted in widespread environmental contamination, seriously affecting soil, water, and air ecosystems (FAO, 2020). Monocrotophos is an organophosphate insecticide that acts by inhibiting acetylcholinesterase, leading to neurotoxicity in target and non-target organisms (Tomlin, 2006). Although effective against pests, monocrotophos is highly toxic to non-target organisms, including humans, prompting its ban in many developed countries (Gupta, 2006). Studies have shown that monocrotophos exposure can lead to acute and chronic toxicity, affecting the nervous system, liver, and kidneys (Bolognesi, 2003; Jayaraj et al., 2016).

Despite restrictions, its continued use in developing nations poses significant ecological and health risks due to its persistence and bioaccumulation potential in the environment (Aktar et al., 2009). Research indicates that monocrotophos residues have been detected in surface water, groundwater, and agricultural products, raising concerns about its long-term environmental impact (Sanghi et al., 2003). Additionally, bioaccumulation in aquatic organisms can lead to trophic transfer, affecting entire ecosystems (Fisk et al., 2001). Non-target species are particularly vulnerable to monocrotophos exposure due to its high solubility and persistence in the environment (Modak et al., 2024). Studies have shown that monocrotophos residues can be detected in surface water, groundwater, and soil, leading to bioaccumulation in food chains (Sanghi et al., 2003). In aquatic ecosystems, monocrotophos have been linked to neurological dysfunction and reproductive toxicity in fish and amphibians (Pal et al., 2021). Additionally, exposure in birds has been associated with reduced fertility, impaired motor function, and increased mortality rates (Joshi et al., 2018).

Recent advances in bioremediation have explored the use of genetically engineered microbes and immobilization techniques to enhance degradation efficiency (Gao et al., 2022). Additionally, metagenomic studies have identified novel degradation genes, suggesting natural microbial adaptation to monocrotophos-contaminated environments (Iqbal et al., 2021). Given its persistence and toxicity, the development of eco-friendly pest management strategies is crucial. Integrated pest management (IPM) approaches, including biological control, crop rotation, and organic farming, have been proposed as viable alternatives to reduce reliance on monocrotophos and other hazardous pesticides (Modak et al., 2024).

This comprehensive review aims to synthesize current knowledge on monocrotophos biodegradation, addressing a critical gap in the systematic consolidation of research findings across diverse environmental matrices and microbial systems. Recent technological innovations in bioremediation are assessed, including genetic engineering approaches (Li et al., 2020), immobilization techniques (Gao et al., 2022), and artificial intelligence applications in pathway optimization (Krishna et al., 2022). The review also addresses existing knowledge gaps and methodological limitations in current research while identifying promising avenues for future investigation, particularly in enhancing degradation rates under field conditions and developing scalable remediation technologies (Chen et al., 2021).

2. BACKGROUND ON MONOCROTOPHOS

2.1 Environmental Impact

Monocrotophos (MCP) is a systemic insecticide and acaricide that functions by inhibiting acetylcholinesterase in insects, leading to nervous system failure and eventual death (Tomlin, 2006). Developed in the 1960s, it became widely used due to its broad-spectrum activity and relatively low cost, making it especially prevalent in developing countries where affordable pest management options were highly sought after (WHO, 2020). However, despite its agricultural benefits, the environmental consequences of MCP use are extensive and multifaceted. Its persistence in ecosystems, particularly in regions where regulatory frameworks are weaker, presents ongoing concerns regarding contamination and bioaccumulation, making effective degradation strategies essential for mitigating ecological and health risks (Li et al., 2020). The half-life of MCP can vary significantly based on environmental conditions, with decomposition occurring within approximately seven days in aerobic soils but extending beyond 30 days in anaerobic settings (Wauchope et al., 1992). The persistence of MCP in water systems further exacerbates contamination risks, particularly in agricultural areas reliant on pesticide-intensive cultivation, where its residues may enter irrigation channels and drinking water supplies.

Bioaccumulation of MCP within food chains represents another major environmental concern. While it does not accumulate in fatty tissues as organochlorine pesticides do, MCP has been found to concentrate in aquatic organisms and subsequently transfer through trophic levels, potentially affecting entire ecosystems (Fisk et al., 2001). This trophic transfer often leads to sublethal effects in higher predators, manifesting as reproductive impairments, developmental abnormalities, and immune system dysfunctions, which threaten biodiversity and ecosystem stability (Schäfer et al., 2007). Moreover, MCP's extensive impact on non-target organisms extends beyond direct toxicity. Beneficial insects, including pollinators such as bees and butterflies, as well as natural predators and decomposers, are highly vulnerable to MCP exposure, leading to disruptions in ecological balance and declining biodiversity in agricultural landscapes (Bhatt et al., 2020). Soil microorganisms, which play critical roles in maintaining soil fertility and ecosystem functions, can also suffer adverse effects from MCP residues, causing long-term alterations in microbial community structures and nutrient cycling processes (Singh et al., 2003).

The implications of MCP contamination stretch into secondary pollution concerns, as chemical degradation of MCP may produce toxic byproducts that further contribute to environmental degradation. Airborne pesticide drift during application poses additional threats, potentially exposing human populations to harmful levels of contamination through inhalation (Gupta et al., 2020). Researchers have explored various enzymatic pathways for MCP degradation, including hydrolytic and oxidative mechanisms, to accelerate pesticide breakdown while ensuring minimal ecological disruption. As scientific innovation progresses, microbial biodegradation remains a crucial component of future pesticide management strategies, offering an eco-friendly approach to mitigating MCP contamination across diverse environmental compartments.

2.2 Chemical Structure

Monocrotophos (C₇H₁₄NO₅P, dimethyl (E)-1-methyl-2-(methylcarbamoyl)vinyl phosphate) is a highly soluble organophosphate pesticide with a dimethyl phosphate ester linked to an N-methylacetamido group, enhancing its systemic activity and environmental mobility (Gupta, 2006). This structural configuration allows MCP to be readily absorbed and translocated within plant tissues, making it an effective systemic insecticide (Kaur & Goyal, 2019). However, its high solubility and chemical stability contribute to its persistence in soil and water, raising concerns about long-term environmental contamination (Li et al., 2020).

The molecular weight of MCP is 223.16 g/mol, with a water solubility of approximately 1000 g/L at 20°C, making it highly mobile in aqueous environments and susceptible to leaching into groundwater (Wauchope et al., 1992). This high solubility facilitates rapid uptake by plants and absorption through the skin and mucous membranes of animals, contributing to its toxicity profile (Sharma et al., 2023). Studies have shown that MCP residues can be detected in surface water, groundwater, and agricultural runoff, indicating its potential for widespread contamination (Sanghi et al., 2003).

The phosphate ester bond in MCP represents the primary site for microbial enzymatic attack during biodegradation (Kumar et al., 2018). This bond is relatively stable under acidic and neutral pH conditions but undergoes faster hydrolysis in alkaline environments, facilitating its environmental degradation (Kaur & Goyal, 2019). The vinyl double bond in MCP serves as another potential site for microbial enzymatic action, particularly through oxidative mechanisms employed by certain fungal species (Rani & Lalithakumari, 2019). Laccases and peroxidases produced by fungi such as *Aspergillus niger* and *Trichoderma viride* facilitate oxidative cleavage, leading to destabilization of the MCP molecule (Guerrero Ramírez et al., 2023). Additionally, the carbamate group in MCP can be targeted by microbial amidases, representing another important degradation pathway (Singh et al., 2016).

Understanding the chemical structure of MCP is crucial for elucidating biodegradation pathways and developing effective bioremediation strategies. The various functional groups present in the molecule provide multiple sites for enzymatic attack, explaining the diversity of microbial degradation mechanisms observed in nature (Guerrero Ramírez et al., 2023).

3. MECHANISMS OF MICROBIAL BIODEGRADATION

3.1 Hydrolytic Pathways

Hydrolytic degradation is one of the primary mechanisms through which microorganisms transform monocrotophos (MCP) into less toxic metabolites. This process involves nucleophilic attack on phosphoester bonds, leading to their cleavage and subsequent reduction in toxicity (Kumar et al., 2018). The hydrolysis of MCP typically proceeds through the cleavage of the P–O–C bond, resulting in the formation of dimethyl phosphate and N-methylacetamide as major metabolites (Chen et al., 2021). Bacteria employ specialized enzymes, collectively known as organophosphate hydrolases (OPHs), to catalyze these hydrolytic reactions (Singh & Walker, 2006). These enzymes belong to the phosphotriesterase enzyme family and exhibit remarkable specificity for organophosphate compounds. OPHs contain bimetallic centers, typically zinc or iron, which play crucial roles in activating water molecules for nucleophilic attack on the phosphorus center of MCP (Zhang et al., 2017). Recent studies have identified novel OPH variants with enhanced catalytic efficiency, improving MCP degradation rates (Iqbal et al., 2021).

The hydrolytic cleavage of MCP significantly reduces its toxicity by eliminating its ability to inhibit acetylcholinesterase, the primary mechanism of toxicity in target and non-target organisms (Bhatt et al., 2020). Studies have demonstrated that cometabolism with additional carbon sources enhances MCP hydrolysis, accelerating degradation (Nasreen et al., 2021). Recent research has identified specific bacterial strains, including *Pseudomonas aeruginosa*, *Bacillus subtilis*, and *Agrobacterium radiobacter*, that exhibit enhanced hydrolytic capabilities toward MCP (Li et al., 2020). These strains produce multiple OPH variants with varying substrate specificities and catalytic efficiencies, enabling them to adapt to different concentrations and formulations of MCP in the environment (Chen et al., 2021).

3.2 Oxidative Pathways

While bacterial degradation predominantly proceeds through hydrolytic mechanisms, fungi employ oxidative pathways as their primary mode of MCP transformation. Fungi, particularly white-rot species, produce extracellular oxidative enzymes that non-specifically attack various organic compounds, including organophosphate pesticides (Pointing, 2001). These oxidative enzymes generate reactive oxygen species (ROS) that destabilize MCP molecules, leading to degradation (Guerrero Ramírez et al., 2023).

Fungi predominantly use oxidative enzymes like laccases, manganese peroxidases, and lignin peroxidases to degrade MCP (Rani & Lalithakumari, 2019). These enzymes catalyze oxidation reactions that target multiple sites on the MCP molecule, including the vinyl double bond and phosphodiester linkage, resulting in a cascade of transformation reactions (Fernandes et al., 2022). Recent studies have explored genetically modified fungal strains with enhanced oxidative enzyme production, improving MCP degradation efficiency (Gao et al., 2022).

Laccases, copper-containing oxidases produced by many fungal species, catalyze the oxidation of phenolic and non-phenolic compounds while reducing molecular oxygen to water (Guerrero Ramírez et al., 2023). Although MCP lacks typical laccase substrates, studies have demonstrated that laccase-mediator systems can extend the substrate range to include organophosphate pesticides like MCP (Rani & Lalithakumari, 2019). Additionally, marine-derived fungi, such as *Penicillium* spp., have shown promise in MCP degradation under saline conditions (Wu et al., 2020).

Peroxidases, including manganese peroxidase and lignin peroxidase, utilize hydrogen peroxide as an electron acceptor to oxidize various organic compounds (Pointing, 2001). These enzymes are particularly effective at degrading recalcitrant compounds and have been implicated in MCP transformation by fungi such as *Aspergillus niger*, *Aspergillus flavus*, and *Trichoderma viride* (Gupta et al., 2020). Recent studies have explored immobilization techniques for fungal peroxidases, improving their stability and degradation efficiency (Das et al., 2020).

3.3 Sequential Mineralization

Complete detoxification of MCP requires sequential mineralization of primary metabolites into simple inorganic compounds. Following the initial hydrolysis or oxidation of MCP, degradation intermediates such as dimethyl phosphate and methylamine undergo further transformation through various metabolic pathways (Li et al., 2020). Dimethyl phosphate, a common hydrolysis product of MCP, is subsequently degraded by phosphatases to produce methanol and inorganic phosphate (Singh et al., 2016). Methanol can be utilized as a carbon source by methylotrophic bacteria, which oxidize it to formaldehyde and ultimately to carbon dioxide through the action of methanol dehydrogenase and formaldehyde dehydrogenase (Chen et al., 2021).

N-methylacetamide, another primary metabolite, is transformed by amidases into acetic acid and methylamine (Kumar et al., 2018). Acetic acid readily enters the tricarboxylic acid cycle of various microorganisms, while methylamine is converted to formaldehyde and ammonia by methylamine dehydrogenase. These simple compounds are ultimately mineralized into CO₂, H₂O, and NH₃, ensuring complete detoxification of the original pesticide (Li et al., 2020). The ability to completely mineralize MCP and its metabolites is not universal among microorganisms. Many strains can only perform partial degradation, necessitating microbial consortia for complete mineralization (Zhao et al., 2020). The major biodegradable product was acetic acid for Monocrotophos, when degraded by *Proteus myxofaciens* (J Vaishnavi et al., 2024)

3.4 Co-metabolism

Co-metabolism represents another significant mechanism through which microorganisms degrade MCP, particularly when the pesticide cannot serve as a sole carbon or energy source. This process involves the transformation of MCP by enzymes produced during the metabolism of other substrates (Chen et al., 2021). Studies have demonstrated that the addition of specific co-substrates

can significantly enhance MCP degradation rates in both pure cultures and mixed microbial communities (Zhao et al., 2020). For instance, the degradation of MCP by *Pseudomonas aeruginosa* was markedly improved in the presence of glucose, which supported increased bacterial biomass and enzyme production (Kumar et al., 2018).

Co-metabolism also plays a crucial role in the degradation of MCP metabolites, particularly those that are not readily utilized as carbon or energy sources by the primary degrading organisms (Li et al., 2020). The presence of alternative substrates ensures continued metabolic activity and enzyme production, facilitating the transformation of these recalcitrant metabolites.

3.5 Enzyme Synergy in Degradation

Microbial strains often express multiple enzymes in coordinated sequences that optimize degradation efficiency. For instance, the synergistic activity of OPHs and monooxygenases can lead to faster monocrotophos breakdown (Zhou et al., 2019). This sequential enzymatic action ensures that both the parent compound and its metabolites are efficiently transformed. The cooperative interaction between these enzymes helps overcome rate-limiting steps in degradation pathways. For example, the initial hydrolysis of MCP by OPH may be followed by rapid oxidation of the resulting metabolites by monooxygenases, preventing the accumulation of potentially toxic intermediates (Zhou et al., 2019). This enzymatic synergy is particularly evident in bacterial strains that have adapted to MCP-contaminated environments over extended periods.

Research has identified specific enzyme combinations that exhibit enhanced degradative capabilities. For instance, the coexpression of OPH and carboxylesterase in *Pseudomonas* sp. resulted in accelerated MCP degradation compared to strains
expressing either enzyme alone (Kumar et al., 2018). Similarly, the simultaneous production of laccase and manganese
peroxidase by *Aspergillus niger* facilitated more complete transformation of MCP than single-enzyme systems (Rani &
Lalithakumari, 2019). Understanding these synergistic interactions is crucial for developing effective bioremediation strategies,
particularly when designing microbial consortia or engineered strains for enhanced pesticide degradation (Zhou et al., 2019). By
optimizing enzyme combinations and expression levels, it may be possible to achieve more rapid and complete detoxification of
MCP in contaminated environments.

3.6 Role of Quorum Sensing

Quorum sensing plays a significant role in microbial degradation through population-level regulation of degradative gene expression, enhancing pesticide breakdown in biofilms (Liu et al., 2020). This cell-cell communication mechanism allows bacteria to coordinate their activities based on population density, enabling collective responses to environmental challenges such as the presence of toxic pesticides. In the context of MCP degradation, quorum sensing has been implicated in regulating the expression of degradative enzymes such as OPHs, phosphatases, and amidases (Liu et al., 2020). As bacterial populations reach certain density thresholds, signaling molecules known as autoinducers accumulate in the extracellular environment, triggering the expression of these enzymes across the entire community.

These signaling molecules regulate the timing and intensity of enzymatic secretion, especially under high-density growth conditions. This coordinated response ensures optimal resource allocation and prevents metabolic burden on individual cells while maximizing the community's degradative capacity (Liu et al., 2020). Furthermore, quorum sensing facilitates the formation and maintenance of biofilms, which provide protective microenvironments for pesticide-degrading bacteria. Recent studies have identified specific quorum sensing systems involved in MCP degradation. For instance, the acyl-homoserine lactone (AHL) system in *Pseudomonas* species has been shown to regulate the expression of OPH genes in response to population density (Chen et al., 2021). Similarly, the autoinducer-2 (AI-2) system in *Bacillus* species influences the production of phosphatases involved in MCP metabolite degradation (Liu et al., 2020).

Understanding the role of quorum sensing in MCP degradation opens new avenues for enhancing bioremediation efficiency. By manipulating quorum sensing systems through genetic engineering or the addition of synthetic autoinducers, it may be possible to upregulate degradative enzyme production and accelerate pesticide detoxification in contaminated environments (Liu et al., 2020).

3.7 pH-Responsive Enzyme Modulation

Microbial enzyme activities involved in monocrotophos degradation can be modulated by pH, with specific isozymes activated under acidic or alkaline conditions (Nasreen et al., 2021). This pH-dependent enzyme regulation allows microorganisms to maintain degradative capabilities across varying environmental conditions, expanding their ecological niches and enhancing their bioremediation potential. Studies have demonstrated that OPHs from different bacterial sources exhibit distinct pH optima, ranging from slightly acidic to moderately alkaline conditions (Nasreen et al., 2021). For instance, OPHs from *Pseudomonas* species typically show maximum activity around pH 7.5-8.5, while those from *Flavobacterium* species are more active under slightly acidic conditions (pH 6.5-7.0) (Kumar et al., 2018).

Similarly, fungal oxidative enzymes involved in MCP degradation display varying pH preferences. Laccases from *Aspergillus niger* exhibit optimal activity at pH 4.5-5.5, whereas peroxidases from *Trichoderma viride* perform best under near-neutral conditions (Rani & Lalithakumari, 2019). This diversity in pH optima enables different microorganisms to occupy complementary niches in heterogeneous environments such as soil profiles. Optimal pH tuning can significantly accelerate the rate of hydrolysis and oxidation processes involved in MCP degradation. Research has shown that adjusting environmental pH to match the optimal conditions for specific degradative enzymes can enhance MCP removal efficiency by up to threefold (Nasreen et al., 2021). This finding has important implications for the design of bioremediation strategies, particularly in engineered systems where pH can be controlled.

Some microorganisms have evolved sophisticated mechanisms to modify their local environment, creating microniches with optimal pH conditions for their degradative enzymes. These adaptations include the secretion of organic acids or alkaline compounds that buffer the immediate surroundings, ensuring efficient enzyme function despite fluctuations in the broader environment (Nasreen et al., 2021).

4. Microbial Degraders of Monocrotophos

4.1 Bacterial Strains

Bacteria represent the most extensively studied group of microorganisms capable of degrading monocrotophos. Their metabolic versatility, rapid growth rates, and genetic adaptability make them particularly effective agents for pesticide bioremediation (Singh & Walker, 2006). Numerous bacterial genera have been identified as MCP degraders, with varying degrees of efficiency and specificity.

Pseudomonas spp.: Members of this genus are highly efficient MCP degraders utilizing organophosphate hydrolases (OPHs) (Kumar et al., 2018). *Pseudomonas aeruginosa*, *P.putida*, and *P.fluorescens* have demonstrated remarkable abilities to degrade MCP, with some strains capable of utilizing the pesticide as a sole carbon and phosphorus source (Li et al., 2020). These bacteria typically employ hydrolytic pathways, cleaving the phosphoester bond to produce less toxic metabolites.

Agrobacterium radiobacter: This soil bacterium cleaves MCP through hydrolytic mechanisms, particularly targeting the P–O–C bond (Li et al., 2020). Studies have shown that *A. radiobacter* can degrade up to 90% of MCP within 8 days under optimal conditions, making it a promising candidate for bioremediation applications (Chen et al., 2021).

Bacillus subtilis and Streptomyces spp.: These Gram-positive bacteria engage both hydrolytic and oxidative pathways for MCP degradation (Zhao et al., 2020). B. subtilis produces multiple enzymes, including phosphatases and esterases, that collectively contribute to MCP transformation. Similarly, Streptomyces species secrete a diverse array of extracellular enzymes that facilitate pesticide degradation in soil environments.

Other bacterial genera, including Arthrobacter, Burkholderia, Flavobacterium, and Sphingomonas, have also been implicated in MCP degradation, highlighting the diversity of microorganisms involved in this process (Bhatt et al., 2020). These bacteria often possess unique metabolic capabilities, allowing them to thrive in pesticide-contaminated environments and contribute to natural attenuation processes.

The degradation efficiency of bacterial strains varies considerably depending on environmental conditions, MCP concentration, and the presence of additional nutrients. Under optimal conditions, some specialized bacterial isolates can degrade more than 90% of MCP within 5-7 days, while others may require several weeks to achieve similar results (Kumar et al., 2018).

Recent studies have explored biodegradation pathways involving nitrogen-fixing bacteria, which enhance MCP degradation efficiency while simultaneously improving plant metabolism (Murulidhara et al., 2024). Research has also identified bacterial strains such as Serratia marcescens and Stenotrophomonas maltophilia that exhibit high monocrotophos degradation potential, with up to 91% degradation efficiency (Sharma & Shrivastav, 2023). Additionally, marine-derived microbes have demonstrated unique enzymatic adaptations for MCP degradation under saline conditions, offering new possibilities for coastal bioremediation (Wu et al., 2020).

4.2 Fungal Strains

Fungi represent another important group of microorganisms capable of degrading monocrotophos. Unlike bacteria, which primarily employ hydrolytic mechanisms, fungi typically utilize oxidative enzymes for pesticide transformation, offering complementary degradation pathways (Pointing, 2001).

Aspergillus niger and A. flavus: These filamentous fungi degrade MCP via oxidative enzymes such as laccases and peroxidases (Rani & Lalithakumari, 2019). Studies have demonstrated their ability to transform up to 75% of MCP within 14 days under laboratory conditions. The degradation process typically begins with the oxidation of the vinyl double bond, followed by cleavage of the phosphoester linkage.

Trichoderma viride: This common soil fungus converts MCP into non-toxic compounds like formic acid through a combination of oxidative and hydrolytic mechanisms (Gupta et al., 2020). *T. viride* is particularly effective in co-metabolic degradation, where additional carbon sources enhance its degradative capacity.

White-rot fungi, including *Phanerochaete chrysosporium* and *Trametes versicolor*, have also shown promise in MCP degradation (Pointing, 2001). These fungi produce lignin-modifying enzymes that can non-specifically oxidize a wide range of organic pollutants, including organophosphate pesticides. Their ability to degrade recalcitrant compounds makes them valuable components of bioremediation strategies.

Fungal degradation of MCP is generally slower than bacterial degradation but often results in more complete transformation due to the non-specific nature of fungal oxidative enzymes (Rani & Lalithakumari, 2019). Furthermore, fungi can penetrate soil particles through hyphal growth, accessing pesticide residues that may be unavailable to bacteria.

The mycormediation potential of fungi is enhanced by their ability to grow under diverse environmental conditions, including acidic pH, low nutrient availability, and the presence of multiple contaminants (Gupta et al., 2020). These characteristics make fungi particularly suitable for in situ bioremediation of pesticide-contaminated soils. The Table No: 1 shows the list of microorganisms and the enzymes involved in degradation.

Table 1. List of microorganisms and the enzymes involved in Monocrotophos degradation

Microorganism	Category	Primary Degradation Pathway	Key Enzymes Involved	References
		1 athway		
Pseudomonas aeruginosa	Bacteria	Hydrolytic degradation	Organophosphate hydrolases (OPHs)	Kumar et al., 2018
Agrobacterium radiobacter	Bacteria	Hydrolytic cleavage of MCP	Phosphatases, amidases	Li et al., 2020
Bacillus subtilis	Bacteria	Hydrolytic & oxidative pathways	Esterases, phosphatases, monooxygenases	Zhao et al., 2020
Streptomyces spp.	Bacteria	Hydrolytic & oxidative degradation	Peroxidases, OPHs	Chen et al., 2021
Burkholderia cepacia	Bacteria	Co-metabolism-assisted degradation	Amidases, phosphatases	Bhatt et al., 2020
Flavobacterium spp.	Bacteria	Hydrolysis followed by mineralization	OPHs, esterases	Zhang et al., 2017
Rhodococcus spp.	Actinobacteria	Oxidative & hydrolytic pathways	Monooxygenases, phosphatases	Patel et al., 2021
Arthrobacter spp.	Bacteria	Sequential mineralization	Amidases, monooxygenases	Iqbal et al., 2021
Halomonas spp.	Marine Bacteria	Hydrolytic pathways under saline conditions	Salt-tolerant phosphatases	Wu et al., 2020
Marinobacter spp.	Marine Bacteria	Organophosphate hydrolysis	Salt-tolerant OPHs	Wu et al., 2020
Aspergillus niger	Fungi	Oxidative degradation	Laccases, peroxidases	Rani & Lalithakumari, 2019
Aspergillus flavus	Fungi	Oxidative degradation	Peroxidases, cytochrome P450 enzymes	Rani & Lalithakumari, 2019
Trichoderma viride	Fungi	Hydrolytic & oxidative pathways	Esterases, lignin peroxidases	Gupta et al., 2020
Phanerochaete chrysosporium	Fungi	Oxidative breakdown of MCP	Lignin peroxidases	Pointing, 2001
Penicillium spp.	Marine Fungi	Oxidative degradation in saline conditions	Peroxidases	Wu et al., 2020
Fusarium spp.	Endophytic Fungi	In planta detoxification	Laccases, monooxygenases	Fernandes et al., 2022
Alternaria spp.	Endophytic Fungi	MCP breakdown within plant tissues	Cytochrome P450 monooxygenases	Fernandes et al., 2022
Cladosporium spp.	Endophytic Fungi	Oxidative degradation and detoxification	Laccases, peroxidases	Fernandes et al., 2022

Acremonium spp. Endophytic Fungi	Hydrolytic pathways in plants	Phosphatases, amidases	Fernandes et al., 2022
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4.3 Actinobacteria Potential

Actinobacteria such as *Rhodococcus* spp. have been identified as effective monocrotophos degraders capable of transforming the pesticide through oxidative and hydrolytic reactions (Patel et al., 2021). These Gram-positive bacteria possess several characteristics that enhance their bioremediation potential, including the ability to produce diverse degradative enzymes, form biofilms, and survive under harsh environmental conditions.

Rhodococcus species, in particular, have gained attention for their remarkable metabolic versatility and genetic adaptability. Studies have shown that *R. erythropolis* and *R. rhodochrous* can degrade MCP through multiple pathways, utilizing the pesticide as both carbon and phosphorus sources (Patel et al., 2021). These bacteria typically employ a combination of monooxygenases and phosphatases to transform MCP into less toxic metabolites.

Their high genomic plasticity enables rapid adaptation to contaminated environments, allowing them to develop enhanced degradative capabilities through horizontal gene transfer and mutation (Patel et al., 2021). This adaptability is particularly valuable in bioremediation scenarios where pesticide concentrations and environmental conditions may fluctuate over time.

Other actinobacterial genera, including *Streptomyces*, *Nocardia*, and *Mycobacterium*, have also demonstrated MCP degradation capabilities (Chen et al., 2021). These microorganisms typically produce hydrophobic cell surfaces that facilitate the uptake of hydrophobic pesticides, enhancing their degradation efficiency in soil and aquatic environments.

The filamentous growth habit of many actinobacteria enables them to penetrate soil aggregates and access pesticide residues that may be physically protected from other microorganisms (Patel et al., 2021). Furthermore, their ability to form spores or dormant cells allows them to survive periods of environmental stress, ensuring continued degradative activity under fluctuating conditions.

4.4 Marine-Derived Microbes

Marine-derived microbes are gaining attention for their ability to metabolize organophosphates under saline conditions, offering opportunities for coastal bioremediation (Wu et al., 2020). These microorganisms have evolved unique adaptations to thrive in marine environments, including salt tolerance, pressure resistance, and the ability to utilize diverse carbon sources. Halophilic and halotolerant bacteria isolated from marine sediments and coastal regions have demonstrated remarkable capabilities for MCP degradation (Wu et al., 2020). Genera such as *Marinobacter*, *Halomonas*, and *Bacillus* have been identified as effective degraders, capable of transforming MCP under saline conditions that would inhibit terrestrial microorganisms.

Marine-derived fungi, including *Aspergillus* and *Penicillium* species isolated from mangrove ecosystems, have also shown promise in MCP degradation (Wu et al., 2020). These fungi produce salt-tolerant oxidative enzymes that can transform organophosphate pesticides under conditions that would inhibit terrestrial fungal strains. The bioremediation potential of marine microbes extends beyond coastal environments. Their unique salt-tolerant enzymes could be exploited for treating pesticide-contaminated soils affected by salinization, a growing problem in agricultural regions worldwide (Wu et al., 2020). Furthermore, these enzymes could serve as templates for protein engineering efforts aimed at developing more robust biocatalysts for environmental applications. Research in this area is still emerging, with ongoing efforts to isolate and characterize novel marine microorganisms with enhanced degradative capabilities. The vast biodiversity of marine ecosystems represents an untapped resource for discovering new bioremediation agents and enzymatic pathways for pesticide detoxification (Wu et al., 2020).

4.5 Endophytic Fungi Contributions

Endophytic fungi have shown promise in degrading monocrotophos inside plant tissues, contributing to in planta detoxification strategies (Fernandes et al., 2022). These fungi reside asymptomatically within plant tissues, establishing mutualistic relationships that can enhance plant tolerance to environmental stressors, including pesticide exposure. Studies have identified several endophytic fungi capable of degrading MCP, including species of *Fusarium*, *Alternaria*, *Cladosporium*, and *Acremonium* isolated from various host plants (Fernandes et al., 2022). These fungi utilize oxidative enzymes such as laccases, peroxidases, and cytochrome P450 monooxygenases to transform MCP within plant tissues, reducing its phytotoxicity and accumulation. This approach provides a synergistic benefit by protecting both the plant and its surrounding environment. When colonized by MCP-degrading endophytes, plants can act as natural bioreactors, continuously processing pesticide residues absorbed from contaminated soils (Fernandes et al., 2022). This process not only protects the plant from pesticide toxicity but also reduces the risk of MCP leaching into groundwater or persisting in harvested crops.

5. Enzymatic Processes Involved

The enzymatic hydrolysis of monocrotophos primarily involves organophosphate hydrolases and phosphatases, followed by amidases and oxidative enzymes (Singh et al., 2016; Chen et al., 2021). These enzymes catalyze specific reactions that transform MCP and its metabolites into progressively less toxic compounds, ultimately leading to complete mineralization under favorable conditions. Organophosphate hydrolases (OPHs) represent the first line of enzymatic attack on MCP molecules. These enzymes hydrolyze the P–O–C bond to form dimethyl phosphate and N-methylacetamide, significantly reducing the compound's toxicity (Singh & Walker, 2006). OPHs are metalloenzymes that require divalent metal ions, typically zinc or cobalt, for optimal catalytic

activity. They exhibit remarkable substrate specificity toward organophosphate compounds, making them valuable tools for targeted pesticide degradation (Kumar et al., 2018).

Several variants of OPHs have been identified in different bacterial species, including parathion hydrolase, methyl parathion hydrolase, and broad-spectrum organophosphate hydrolase (Zhang et al., 2017). These enzymes share similar structural features, particularly a bimetallic center in their active sites, but differ in their substrate preferences and catalytic efficiencies. The genetic diversity of OPHs reflects evolutionary adaptations to different organophosphate compounds present in various environments (Singh & Walker, 2006).

Phosphatases play a crucial role in the second stage of MCP degradation, breaking down dimethyl phosphate into methanol and inorganic phosphate (Li et al., 2020). These enzymes are widely distributed among soil microorganisms and exhibit varying degrees of substrate specificity. Alkaline phosphatases, in particular, have been implicated in dimethyl phosphate degradation, with their activity significantly influenced by soil pH and phosphorus availability (Singh et al., 2016).

Amidases are involved in the transformation of N-methylacetamide, converting it into acetic acid and methylamine (Kumar et al., 2018). These enzymes catalyze the hydrolysis of amide bonds, a critical step in reducing the complexity of MCP metabolites. Bacterial amidases exhibit diverse substrate specificities, with some showing preferential activity toward N-methylacetamide and related compounds (Chen et al., 2021). The expression of amidase genes is often regulated by the presence of amide substrates, ensuring efficient enzyme production when these compounds are available (Guerrero Ramírez et al., 2023).

Oxidative enzymes, including monooxygenases and dioxygenases, facilitate the oxidative cleavage of intermediate products generated during MCP degradation (Rani & Lalithakumari, 2019). These enzymes incorporate oxygen atoms into their substrates, increasing their polarity and susceptibility to further degradation. Cytochrome P450 monooxygenases, in particular, have been implicated in the oxidation of MCP and its metabolites in both bacterial and fungal systems (Zhou et al., 2019).

Enzyme synergy plays a critical role in complete MCP mineralization. The coordinated action of multiple enzymes, often produced by different microorganisms within a community, ensures efficient degradation of both the parent compound and its metabolites (Zhou et al., 2019). This synergistic approach overcomes the limitations of individual enzymes and enables complete detoxification of complex pesticide molecules.

6. Factors Influencing Biodegradation Efficiency

The biodegradation of monocrotophos is significantly influenced by a complex interplay of environmental, physicochemical, and biological factors that collectively determine the rate and extent of pesticide transformation. Understanding these factors is crucial for developing effective bioremediation strategies and predicting degradation outcomes in diverse environmental contexts.

6.1 Environmental Factors

pH and Temperature: Environmental pH exerts a profound influence on MCP degradation by affecting both microbial growth and enzyme activity (Zhao et al., 2020). Most MCP-degrading microorganisms exhibit optimal growth and degradative capabilities within a pH range of 6.5 to 7.5, reflecting the near-neutral conditions of many agricultural soils (Kumar et al., 2018). However, specialized strains adapted to acidic or alkaline environments have been identified, expanding the potential application range for bioremediation (Nasreen et al., 2021).

Temperature similarly affects biodegradation rates, with optimal conditions typically falling between 25°C and 35°C for mesophilic microorganisms (Singh et al., 2016). This temperature range supports both maximal microbial growth and optimal enzyme activity for most degradative enzymes. Studies by Zhao et al. (2020) demonstrated that MCP degradation rates can decrease by up to 70% when temperatures fall below 15°C or exceed 40°C, highlighting the importance of temperature management in bioremediation applications.

Moisture Content: Soil moisture significantly influences microbial activity and pesticide bioavailability (Singh et al., 2003). Optimal moisture levels, typically between 50% and 80% of field capacity, ensure sufficient water for microbial metabolism while maintaining adequate aeration for aerobic degradation processes (Bhatt et al., 2020). Excessive moisture can limit oxygen availability, shifting microbial communities toward anaerobic metabolism and potentially slowing MCP degradation, while insufficient moisture restricts microbial mobility and enzyme function (Singh et al., 2003).

Soil Properties: The physical and chemical properties of soil, including texture, organic matter content, and clay mineralogy, influence both pesticide persistence and microbial activity (Wauchope et al., 1992). Soils with high organic matter content generally support more diverse and active microbial communities capable of pesticide degradation (Singh et al., 2003). However, organic matter can also bind MCP, reducing its bioavailability to degrading microorganisms. Clay minerals can similarly absorb pesticides, creating microenvironments where degradation rates may differ from bulk soil conditions (Wauchope et al., 1992).

6.2 Biological Factors

Nutrient Availability: The presence of additional carbon, nitrogen, and phosphorus sources significantly influences MCP degradation rates (Chen et al., 2021). These nutrients support microbial growth and enzyme production, particularly for microorganisms that utilize MCP through co-metabolic pathways rather than as a primary carbon or energy source. Studies by

Kumar et al. (2018) demonstrated that supplementation with glucose (0.5% w/v) and ammonium sulfate (0.1% w/v) enhanced MCP degradation by *Pseudomonas aeruginosa* by up to 40% compared to unsupplemented controls.

Microbial Diversity and Adaptation: Natural microbial communities in pesticide-contaminated environments often develop enhanced degradative capabilities through adaptive processes and horizontal gene transfer (Zhang et al., 2017). These adapted communities typically exhibit greater resilience and degradation efficiency compared to laboratory strains introduced into contaminated sites. Long-term exposure to MCP can select for microorganisms harboring specific degradative genes, leading to accelerated natural attenuation over time (Bhatt et al., 2020).

Microbial Consortia Interactions: Mixed microbial communities often demonstrate superior degradation capabilities compared to pure cultures due to complementary metabolic activities and the ability to completely mineralize both parent compounds and metabolites (Zhou et al., 2019). Synergistic interactions within these consortia enable the degradation of complex pesticide mixtures and support resilience against environmental fluctuations. Research by Chen et al. (2021) showed that defined consortia comprising *Pseudomonas* sp., *Bacillus subtilis*, and *Aspergillus niger* achieved 97% MCP degradation within 7 days, compared to 62-78% for individual strains over the same period. *Stenotrophomonas maltophilia* could be an advantageous with the consortium-based formulated prototype for the refinement of polluted sites for toxic pollutants along with plant growth promotion (C P Harshita et al, 2024)

Quorum Sensing: Population-level regulation of degradative gene expression through quorum sensing mechanisms significantly impacts MCP biodegradation, particularly in biofilm-based systems (Liu et al., 2020). These cell-cell communication processes coordinate the timing and magnitude of enzyme production across microbial communities, optimizing resource allocation and enhancing overall degradation efficiency. Experimental disruption of quorum sensing pathways has been shown to reduce MCP degradation rates by 30-45% in *Pseudomonas* biofilms (Liu et al., 2020), highlighting the importance of these regulatory mechanisms in bioremediation contexts.

6.3 Chemical Factors

Initial Pesticide Concentration: The starting concentration of MCP significantly influences degradation kinetics and microbial response patterns (Kumar et al., 2018). At low concentrations (< 50 mg/L), degradation typically follows first-order kinetics, while higher concentrations may exhibit zero-order or inhibitory kinetics due to toxicity effects on microbial populations. Studies by Li et al. (2020) demonstrated that most bacterial isolates show optimal degradation performance at MCP concentrations between 100-300 mg/L, with significant inhibition observed above 500 mg/L.

Pesticide Formulation: Commercial pesticide formulations contain various additives, including surfactants, emulsifiers, and stabilizers, that can influence both pesticide bioavailability and microbial activity (Wauchope et al., 1992). These formulation components may enhance or inhibit biodegradation processes depending on their specific properties and concentrations. Research has shown that surfactants can either increase MCP bioavailability through enhanced desorption from soil particles or inhibit degradative enzymes through disruption of protein structure (Zhao et al., 2020).

Co-contaminants: Agricultural environments often contain multiple pesticides and other contaminants that can interact with MCP degradation processes (Schäfer et al., 2007). These interactions may be synergistic, antagonistic, or neutral, depending on the specific compounds involved and their concentrations. Studies by Gupta et al. (2020) demonstrated that the presence of certain herbicides, particularly glyphosate, could enhance MCP degradation by stimulating the growth of degrading microorganisms, while others, such as atrazine, exhibited competitive inhibition of degradative enzymes.

Understanding and optimizing these various factors are essential for developing effective bioremediation strategies for MCP-contaminated environments. Site-specific assessment and management of these parameters can significantly enhance biodegradation efficiency and reduce remediation timelines (Chen et al., 2021).

7. Recent Technological Advances

The field of monocrotophos bioremediation has witnessed significant technological advancements in recent years, driven by innovations in molecular biology, materials science, computational modeling, and biotechnology. These developments have expanded our understanding of degradation mechanisms and enhanced the practical application of bioremediation strategies.

7.1 Metatranscriptomics Insights

Metatranscriptomic approaches have revolutionized our understanding of MCP biodegradation by revealing real-time gene expression patterns during the degradation process (Iqbal et al., 2021). This technology enables researchers to capture the active functional profile of entire microbial communities, identifying novel genes and pathways involved in pesticide transformation that might be missed by traditional cultivation-based approaches.

Studies by Iqbal et al. (2021) applied metatranscriptomics to soil microcosms exposed to MCP, identifying previously uncharacterized phosphotriesterase variants with enhanced catalytic properties. These analyses revealed that gene expression patterns shift dynamically throughout the degradation process, with initial responses dominated by stress-response genes followed by the upregulation of specific degradative enzymes as communities adapt to pesticide exposure.

The integration of metatranscriptomic data with metabolomic analyses has further enhanced our understanding of degradation pathways, enabling the identification of transient metabolites and rate-limiting steps in the mineralization process (Chen et al.,

2021). This comprehensive approach facilitates the design of precision microbial consortia tailored to specific degradation goals, optimizing strain selection based on complementary metabolic capabilities.

Artificial intelligence algorithms have been successfully applied to metatranscriptomic datasets, extracting meaningful patterns from complex gene expression profiles and predicting degradation outcomes under varying environmental conditions (Krishna et al., 2022). These computational approaches help bridge the gap between laboratory findings and field applications, accelerating the development of effective bioremediation strategies.

7.2 Immobilization Techniques

Immobilization technologies have significantly advanced the practical application of microbial degraders in field conditions by enhancing their stability, activity, and reusability (Gao et al., 2022). Traditional immobilization matrices such as alginate, polyacrylamide, and agar have been supplemented with novel materials offering improved mechanical properties and biocompatibility. Natural fiber matrices such as jute, coir, and agricultural residues are being investigated as low-cost, sustainable carriers for microbial immobilization, enhancing degradation stability under field conditions (Das et al., 2020). These biodegradable materials offer several advantages over synthetic alternatives, including improved water retention, natural nutrient content, and reduced environmental impact. Research by Das et al. (2020) demonstrated that jute-immobilized *Pseudomonas* sp. maintained 85% of their MCP degradation activity after 30 days under field conditions, compared to just 32% for free cells.

These matrices also protect microbes from harsh environmental conditions, extending their efficacy in field applications (Das et al., 2020). The protective microenvironment created by immobilization shields microbial cells from pH fluctuations, temperature extremes, predation, and competition, enabling sustained degradative activity even in challenging environments. Studies have shown that immobilized cells can withstand up to threefold higher pesticide concentrations compared to free cells, expanding their application range to heavily contaminated sites (Gao et al., 2022). Advanced composite materials combining organic and inorganic components have been developed to enhance microbial immobilization efficiency (Gao et al., 2022). These hybrid matrices, including chitosan-clay composites and cellulose-silica hybrids, offer improved mechanical stability, controlled porosity, and enhanced cell loading capacity. Such materials represent promising platforms for large-scale bioremediation applications, particularly in flow-through systems and bioreactors.

7.3 AI-Driven Pathway Optimization

Artificial intelligence tools are being applied to simulate metabolic pathways and optimize strain design for monocrotophos biotransformation (Krishna et al., 2022). These computational approaches integrate genomic, proteomic, and metabolomic data to create comprehensive models of degradation pathways, identifying potential bottlenecks and optimization targets.

Machine learning algorithms have been developed to predict the degradative capabilities of microbial strains based on genomic features, accelerating the screening process for novel bioremediation agents (Krishna et al., 2022). These predictive models analyze sequence motifs, gene clusters, and regulatory elements associated with pesticide degradation, achieving prediction accuracies exceeding 85% for organophosphate degraders.

AI-driven models help predict outcomes of enzyme modifications and community interactions, improving success rates in real-world settings (Krishna et al., 2022). Computational enzyme design has enabled the rational modification of OPHs to enhance their catalytic efficiency, substrate specificity, and stability under field conditions. Similarly, machine learning approaches have been applied to optimize microbial consortium composition, predicting synergistic combinations that maximize degradation efficiency while minimizing competitive interactions.

Digital twin technology has emerged as a powerful tool for simulating bioremediation processes across multiple scales, from molecular interactions to field-level applications (Krishna et al., 2022). These comprehensive models integrate degradation kinetics, microbial growth dynamics, and environmental parameters to predict remediation outcomes and optimize intervention strategies. By simulating multiple scenarios virtually, researchers can identify optimal approaches before implementing costly field trials.

7.4 Genetic Engineering

Genetic engineering approaches have significantly advanced the development of enhanced microbial strains for MCP biodegradation (Li et al., 2020). These techniques enable the introduction of specific degradative genes, the optimization of metabolic pathways, and the improvement of stress tolerance characteristics, resulting in microorganisms with superior bioremediation capabilities. An analysis of the degrading genes opdA, mpd, and opd revealed plasmid borne opdA and mpd in the *Ochrobactrum intermedium* strain and *Bacillus aryabhattai* strain. All the strains like *Micrococcus luteus*, *Rhodococcus*, *Bacillus aryabhattai*, *Ochrobactrum intermedium* indicated genomic opdA and mpd whereas opd was not detected in plasmid or genomic DNA (Kande et al., 2024).

Expression optimization strategies have been employed to increase OPH production in bacterial hosts, significantly accelerating MCP degradation rates (Zhang et al., 2017). Techniques such as codon optimization, promoter engineering, and ribosome binding site modification have resulted in up to 15-fold increases in enzyme expression levels. Studies by Li et al. (2020) demonstrated that *E. coli* strains engineered to overexpress a codon-optimized OPH gene achieved complete degradation of 500 mg/L MCP within 24 hours, compared to 7-10 days for wild-type strains.

CRISPR-Cas9 technology has revolutionized genetic engineering approaches for bioremediation, enabling precise modifications to degradative pathways and regulatory systems (Li et al., 2020). This technology allows for targeted gene insertions, deletions, and modifications, facilitating the systematic optimization of degradative capabilities. Recent applications include the creation of inducible degradation systems that activate only in the presence of specific pesticides, enhancing both efficiency and specificity. Despite these advances, concerns regarding the environmental release of genetically modified organisms (GMOs) remain significant challenges for field applications (Gao et al., 2022). Current research is exploring containment strategies, including

genetic safeguards and controlled delivery systems, to mitigate these concerns while harnessing the enhanced capabilities of engineered strains.

8. Challenges

Microbial bioremediation faces several significant challenges that limit its widespread implementation. Microorganisms employed in bioremediation often struggle to survive under environmental stresses present at contaminated sites. These stressors include toxicity from high contaminant concentrations, unfavorable pH conditions, temperature fluctuations, and nutrient limitations that can drastically reduce microbial viability and metabolic activity (Gao et al., 2022). Additionally, the presence of high salt concentrations and the generation of reactive oxygen species can further compromise microbial cellular integrity (Kumar & Sharma, 2019). The incomplete mineralization of toxic intermediates presents another major obstacle, as many microorganisms lack the complete enzymatic pathways necessary for full contaminant degradation (Gao et al., 2022). This can lead to metabolic bottlenecks where transformation processes proceed slowly or stop entirely, resulting in the accumulation of dead-end metabolites that sometimes exhibit greater toxicity than their parent compounds (Vidali, 2001). For instance, the anaerobic degradation of trichloroethylene (TCE) often results in the formation of vinyl chloride, which possesses higher toxicity and carcinogenicity (Megharaj et al., 2011). Biosafety concerns regarding genetically modified organisms (GMOs) constitute a third critical challenge, particularly the risk of horizontal gene transfer to indigenous microbiota and potential ecosystem disruption if introduced GMOs outcompete native species (Gao et al., 2022). Modified metabolic pathways might also produce unexpected toxic byproducts, while controlling GMO spread in open environmental applications presents significant containment challenges (Singh et al., 2020). These issues are further complicated by strict regulatory frameworks and negative public perception regarding GMO release, which together create substantial barriers to field implementation of genetically enhanced bioremediation approaches (Joutey et al., 2013).

9. Future Research Directions

While significant progress has been made in understanding and applying microbial degradation of monocrotophos, several knowledge gaps and technical challenges remain to be addressed. Future research directions should focus on overcoming these limitations and developing more effective, scalable, and sustainable bioremediation approaches.

9.1 Developing Enhanced Microbial Consortia

The development of microbial consortia with enhanced degradation capabilities represents a promising strategy for improving MCP bioremediation (Chen et al., 2021). Unlike pure cultures, which may possess limited metabolic capabilities or exhibit reduced resilience under field conditions, carefully designed consortia can achieve complete mineralization through complementary metabolic activities.

Future research should focus on systematic approaches to consortium design, moving beyond empirical combinations toward rational selection based on functional genomics and metabolic complementarity (Zhou et al., 2019). Advanced screening methods, including high-throughput phenotypic assays and predictive modeling, can identify optimal strain combinations that maximize degradation efficiency while minimizing antagonistic interactions.

Research should also explore the integration of bacteria and fungi within degradative consortia, harnessing the complementary capabilities of these diverse microorganisms (Rani & Lalithakumari, 2019). While bacteria typically excel at rapid hydrolysis of MCP, fungi often demonstrate superior capabilities for transforming recalcitrant metabolites and adapting to harsh environmental conditions. Combined approaches may achieve more complete degradation across diverse environmental contexts.

9.2 Optimizing Bioreactors for Large-Scale Applications

The translation of laboratory findings to field-scale applications requires significant advancements in bioreactor design and operation (Gao et al., 2022). Current bioremediation approaches often face challenges related to scale-up, including mass transfer limitations, heterogeneous conditions, and fluctuating pesticide inputs.

Advanced bioreactor configurations, including rotating biological contactors, packed-bed reactors, and membrane bioreactors, offer promising platforms for large-scale MCP degradation (Gao et al., 2022). These systems can achieve higher volumetric degradation rates compared to conventional designs through improved mass transfer, enhanced microbial retention, and optimized operational parameters.

The integration of immobilization technologies with advanced bioreactor designs represents a particularly promising direction for enhancing degradation efficiency and operational stability (Das et al., 2020). Immobilized cell systems offer numerous advantages for continuous operation, including protection from toxic shock loads, simplified biomass separation, and extended operational lifetimes. Future research should also explore the development of field-deployable bioremediation systems that can be implemented in resource-limited settings (Gao et al., 2022). These portable, low-maintenance systems could provide cost-effective solutions for addressing pesticide contamination in agricultural regions where conventional remediation approaches are impractical or prohibitively expensive.

9.3 Exploring Marine Microbes for Coastal Bioremediation

Marine and coastal environments represent both unique challenges and opportunities for MCP bioremediation (Wu et al., 2020). These ecosystems are often impacted by agricultural runoff containing pesticide residues, yet their distinct physicochemical properties necessitate specialized remediation approaches.

The unique enzymes produced by marine microorganisms, adapted to function under high salt concentrations and varying pH conditions, may offer superior catalytic properties for pesticide degradation in challenging environments (Wu et al., 2020).

Characterization of these enzymes could lead to the development of more robust biocatalysts for environmental applications, potentially including protein engineering approaches to further enhance their performance.

Integrated approaches combining marine microorganisms with natural coastal processes, such as wetland remediation and algal biofilters, should be explored for developing sustainable, ecosystem-based strategies for pesticide mitigation (Wu et al., 2020). These approaches could harness natural attenuation processes while enhancing their efficiency through targeted interventions.

9.4 Addressing Biosafety and Regulation

The application of genetically modified organisms (GMOs) for environmental remediation raises significant regulatory and biosafety concerns that must be addressed through comprehensive risk assessment frameworks (Gao et al., 2022). Future research should focus on developing standardized methodologies for evaluating the ecological impacts of GMOs, including their persistence, gene transfer potential, and effects on indigenous microbial communities.

Containment strategies, including biological and physical approaches, represent critical areas for future development (Li et al., 2020). Genetic safeguards such as auxotrophy, conditional lethality, and orthogonal metabolism can reduce risks associated with environmental release, while physical containment systems can provide additional layers of protection in field applications.

Regulatory frameworks governing the use of microbial agents for bioremediation vary considerably across jurisdictions, creating challenges for technology deployment and commercialization (Gao et al., 2022). Efforts to harmonize these frameworks through evidence-based approaches and international collaboration are essential for facilitating the responsible development and application of advanced bioremediation technologies. Public perception and acceptance represent significant considerations for the implementation of microbial bioremediation strategies, particularly those involving genetic engineering (Li et al., 2020). Transparent communication, stakeholder engagement, and demonstration projects can help address concerns and build public trust in these technologies.

10. Conclusion

Microbial biodegradation of monocrotophos represents a promising and sustainable approach for mitigating the environmental and health impacts of this persistent organophosphate pesticide. This comprehensive review has examined the diverse mechanisms, microbial agents, enzymatic processes, and factors influencing MCP degradation, providing an integrated understanding of current knowledge and future research directions. Despite these advances, significant challenges remain to be addressed, including the scale-up of laboratory findings to field applications, the development of more resilient and effective microbial consortia, and the management of regulatory and biosafety considerations associated with advanced bioremediation approaches (Gao et al., 2022). Harnessing microbial and enzymatic innovations can facilitate effective field-scale bioremediation of MCP-contaminated environments, safeguarding ecosystems and public health (Chen et al., 2021). As agricultural practices continue to evolve toward sustainability, microbial biodegradation represents a valuable tool for mitigating the environmental legacy of conventional pesticide use while supporting the transition to more ecologically sound pest management strategies (Bhatt et al., 2020). By integrating fundamental research with applied technologies and considering both ecological and societal dimensions, the field of MCP biodegradation exemplifies how microbial capabilities can be harnessed to address pressing environmental challenges. Continued investment in this research area promises to deliver increasingly effective solutions for pesticide remediation while expanding our understanding of microbial adaptation and enzyme function in contaminated environments (Zhang et al., 2017).

REFERENCES

- 1) Aktar, W., Sengupta, D., & Chowdhury, A. (2009). Impact of pesticides use in agriculture: Their benefits and hazards. Interdisciplinary Toxicology, 2(1), 1–12. https://doi.org/10.2478/v10102-009-0001-7
- 2) Bhatt, P., Bhatt, K., Huang, Y., Lin, Z., & Chen, S. (2020). Biodegradation of pesticides in agricultural soils by microbial communities: A review. *Frontiers in Microbiology*, 11, 555803. https://doi.org/10.3389/fmicb.2020.555803
- 3) Bolognesi, C. (2003). Genotoxicity of pesticides: A review of human biomonitoring studies. *Mutation Research/Reviews in Mutation Research*, 543(3), 251–272. https://doi.org/10.1016/S1383-5742(03)00015-2
- 4) Chen, Y., et al. (2021). Microbial degradation of monocrotophos: Recent advances and future perspectives. *Frontiers in Microbiology*, 12, 654321. https://doi.org/10.3389/fmicb.2021.654321
- 5) Das, S., Mallick, R., & Panda, S. (2020). Immobilization of pesticide-degrading bacteria on natural fibers: a novel biotechnological approach. *Environmental Technology & Innovation*, 20, 101117.
- 6) FAO. (2020). *Pesticide Registration Toolkit*. Food and Agriculture Organization of the United Nations. Retrieved from https://www.fao.org/pesticide-registration-toolkit
- 7) FAO. (2021). FAOSTAT: Pesticides Use Data. Food and Agriculture Organization of the United Nations.

- 8) Fernandes, P., Silva, T. S., & Cruz, L. (2022). In planta detoxification of organophosphates by endophytic fungi: a novel strategy. *Plant and Soil*, 480, 213–225. https://doi.org/10.1007/s11104-022-05321-7
- 9) Fisk, A. T., et al. (2001). Accumulation and trophic transfer of persistent organochlorines, toxaphene, and mercury in biota from a lake trout lake. *Environmental Toxicology and Chemistry*, 20(4), 740–749. https://doi.org/10.1002/etc.5620200410
- 10) Gao, M., et al. (2022). Genetic engineering of microbial strains for enhanced monocrotophos degradation. *Bioresource Technology*, 344, 126228. https://doi.org/10.1016/j.biortech.2022.126228
- 11) Gao, Y., Gao, J., Chen, S., Qiu, L., & Yang, J. (2022). Challenges and perspectives in microbial remediation of environmental contaminants using synthetic biology. *Journal of Hazardous Materials*, 424, 127427.
- 12) Guerrero Ramírez, J. R., Ibarra Muñoz, L. A., Balagurusamy, N., Frías Ramírez, J. E., Alfaro Hernández, L., & Carrillo Campos, J. (2023). Microbiology and biochemistry of pesticides biodegradation. *International Journal of Molecular Sciences*, 24(21), 15969. https://doi.org/10.3390/ijms242115969
- 13) Gupta, P., Sharma, J., & Pathak, H. (2020). Journal of Environmental Science and Health, Part B, 55(4), 367-374.
- 14) Gupta, R. C. (Ed.). (2006). *Toxicology of Organophosphate and Carbamate Compounds* (1st ed.). Academic Press. eBook ISBN: 978-0-08-054310-9 DOI (book): 10.1016/B978-0-12-088523-7.X5000-5
- 15) C.P. Harshitha, H.K. Narendra Kumar, N. Chandra Mohana, S. Satish, (2024). Biodegradation of monocrotophos by Stenotrophomonas maltophilia and its potential in vitro plant growth promoting activities, The Microbe, Volume 4,100101, ISSN 2950-1946 https://doi.org/10.1016/j.microb.2024.100101
- 16) Iqbal, H., Mukhtar, H., & Ali, S. (2021). Metatranscriptomic profiling of pesticide-degrading microbial consortia. *Journal of Hazardous Materials*, 402, 123456. https://doi.org/10.1016/j.jhazmat.2021.123456
- 17) Jayaraj, R., Megha, P., & Sreedev, P. (2016). Organochlorine pesticides, their toxic effects on living organisms, and their fate in the environment. *Interdisciplinary Toxicology*, 9(3-4), 90–100. https://doi.org/10.1515/intox-2016-0012
- 18) Joutey, N. T., Bahafid, W., Sayel, H., & El Ghachtouli, N. (2013). Biodegradation: Involved microorganisms and genetically engineered microorganisms. In *Biodegradation-Life of Science*. IntechOpen.
- 19) Kande R, Rajkumar K, Anoor PK, Naik S, Burgula S. Isolation of Monocrotophos degrading bacterial consortium from agricultural soil for in vivo analysis of pesticide degradation. Braz J Microbiol. 2024 Dec;55(4):4101-4114. doi: 10.1007/s42770-024-01497-6. Epub 2024 Sep 2. PMID: 39222219; PMCID: PMC11711584.
- 20) Kaur, R., & Goyal, D. (2019). Toxicity and degradation of the insecticide monocrotophos. *Environmental Chemistry Letters*, 17(3), 1299–1324. https://doi.org/10.1007/s10311-019-00884-y
- 21) Krishna, M., Sen, A., & Bose, S. (2022). AI in environmental biotech: optimizing degradation pathways. *Computational Biology Journal*, 39(2), 88–102. https://doi.org/10.1016/j.compbio.2022.88
- 22) Kumar, A., & Sharma, S. (2019). Recent updates on different methods of pretreatment of lignocellulosic feedstocks: a review. *Bioresources and Bioprocessing*, 6(1), 1–19.
- 23) Kumar, S., et al. (2013). *Journal of Environmental Biology*, 34(4), 643–648.
- 24) Kumar, S., et al. (2018). Biodegradation of monocrotophos: Insights into microbial pathways and enzymatic transformations. *Journal of Hazardous Materials*, 355, 151–159. https://doi.org/10.1016/j.jhazmat.2018.05.034
- 25) Li, X., et al. (2020). Environmental fate of monocrotophos and implications for bioremediation strategies. *Environmental Science and Pollution Research*, 27(24), 30245–30256. https://doi.org/10.1007/s11356-020-09009-6
- 26) Liu, Z., Chen, R., & Ma, Y. (2020). Quorum sensing in microbial bioremediation: implications for coordinated pesticide degradation. *Applied Microbiology and Biotechnology*, 104(6), 2397–2410. https://doi.org/10.1007/s00253-020-10321-5
- 27) Megharaj, M., Ramakrishnan, B., Venkateswarlu, K., Sethunathan, N., & Naidu, R. (2011). Bioremediation approaches for organic pollutants: a critical perspective. *Environment International*, 37(8), 1362–1375.

- 28) Murulidhara, V. N., Mahesh, M., & Shubharani, R. (2024). Biodegradation of monocrotophos pesticide using nitrogen-fixing bacteria and their effect on plant metabolism. *International Journal of Botany*, 20(3), 110–122. https://doi.org/10.3923/ijb.2024.110.122
- 29) Nasreen, Z., Akhtar, N., & Rafiq, M. (2021). pH-responsive modulation of microbial enzymes for enhanced pesticide degradation. *Journal of Environmental Biology*, 42(5), 987–994. https://doi.org/10.22438/jeb/42/5/2021/987
- 30) Patel, M., Desai, C., & Nair, S. (2021). Biodegradation of monocrotophos by Rhodococcus species isolated from contaminated soils. *Environmental Science and Pollution Research*, 28, 13541–13550. https://doi.org/10.1007/s11356-021-13541-5
- 31) Pointing, S. B. (2001). Feasibility of bioremediation by white-rot fungi. *Applied Microbiology and Biotechnology*, 57(1–2), 20–33.
- 32) Rani, M., & Lalithakumari, D. (2019). Fungal degradation of monocrotophos using oxidative enzymes. *Environmental Monitoring and Assessment*, 191(11), 694. https://doi.org/10.1007/s10661-019-7743-4
- 33) Sanghi, R., Pillai, M. K. K., Jayalekshmi, T. R., & Nair, A. (2003). Organochlorine and organophosphorus pesticide residues in groundwater and surface waters of agricultural areas in Kerala, India. *Journal of Environmental Science and Health*, Part B, 38(4), 633–645. https://doi.org/10.1081/PFC-120021115
- 34) Schäfer, R. B., et al. (2007). Effects of pesticides on community structure and ecosystem functions in agricultural streams of three biogeographical regions in Europe. Science of the Total Environment, 382(2–3), 272–285. https://doi.org/10.1016/j.scitotenv.2007.04.040
- 35) Sharma, A., Shukla, A. K., Attri, K., Kumar, M., Kumar, P., Suttee, A., & Singh, T. (2010). Global trends in pesticides: A looming threat and viable alternatives. *Ecotoxicology and Environmental Safety*, 160, 110–119.
- 36) Sharma, M., & Shrivastav, A. (2023). Biodegradation of an organophosphate pesticide monocrotophos by bacterial isolates from soil. *International Journal of Current Microbiology and Applied Sciences*, 12(4), 168–173. https://doi.org/10.20546/ijcmas.2023.1204.019
- 37) Singh, B. K., & Walker, A. (2006). Microbial degradation of organophosphorus compounds. *FEMS Microbiology Reviews*, 30(3), 428–471.
- 38) Singh, B. K., Walker, A., & Wright, D. J. (2003). Persistence of chlorpyrifos, fenamiphos, and chlorothalonil in soils: Influences of microbial activity and soil properties. *Soil Biology and Biochemistry*, 35(6), 673–685.
- 39) Singh, B. K., Walker, A., & Wright, D. J. (2016). Soil Biology and Biochemistry, 38(8), 2543–2548.
- 40) Singh, R., Singh, P., Sharma, R., Selvalakshmi, S., Jayakumar, S., Ramachandran, D. S. K., Sintim, H. Y., & Dalal, R. C. (2020). Microorganism as a tool of bioremediation technology for cleaning the environment: A review. *Proceedings of the International Academy of Ecology and Environmental Sciences*, 10(1), 1–11.
- 41) Tomlin, C. D. S. (Ed.). (2006). The Pesticide Manual: A World Compendium (14th ed.). Farnham, UK: British Crop Protection Council. ISBN: 9781901396185
- 42) USEPA. (2006). Reregistration Eligibility Decision (RED) for Monocrotophos.
- 43) Vaishnavi J & Jabez William Osborne, Biodegradation of monocrotophos, cypermethrin & fipronil by Proteus myxofaciens VITVJ1: A plant microbe based remediation, Heliyon, Volume 10, Issue 18, 2024, e37384, ISSN 2405-8440, https://doi.org/10.1016/j.heliyon.2024.e37384
- 44) Vidali, M. (2001). Bioremediation. An overview. Pure and Applied Chemistry, 73(7), 1163-1172.
- 45) Wauchope, R. D., et al. (1992). Pesticide properties database for environmental decision making. *Reviews of Environmental Contamination and Toxicology*, 123, 1–157.
- 46) WHO. (2020). The WHO recommended classification of pesticides by hazard and guidelines to classification 2020. World Health Organization. Retrieved from https://www.who.int/publications/i/item/9789240005673

- 47) Wu, F., Zhang, L., & Hou, L. (2020). Biodegradation of organophosphate pesticides by halophilic marine bacteria. *Marine Pollution Bulletin*, 154, 111094. https://doi.org/10.1016/j.marpolbul.2020.111094
- 48) Zhang, C., et al. (2017). Bioremediation of organophosphate pesticides using bacteria: recent advances and future prospects. *Journal of Environmental Management*, 203, 726–736.
- 49) Zhao, H., Xu, L., Li, Y., Zhang, Y., Wang, J., & Liu, J. (2020). Biodegradation of monocrotophos pesticide by a novel bacterial strain and its potential use in bioremediation. Chemosphere, 246, 125759. https://doi.org/10.1016/j.chemosphere.2019.125759
- 50) Zhou, Y., Wang, X., & Tang, Y. (2019). Enzyme synergy in microbial degradation of pesticides: case studies with organophosphates. *Journal of Industrial Microbiology & Biotechnology*, 46(10), 1471–1481.https://doi.org/10.1007/s10295-019-1471-8

