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Microbial Bioremediation Strategies for Degradation of Persistent Organic Pollutants: Synergistic Approach Involving Microbes to Address the Global Concern

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Abstract

Persistent organic pollutants (POPs) are synthetic chemicals that do not degrade easily in the environment, resist chemical, biological and photolytic degradation, have a strong affinity towards fats (lipophilic) and can either be absorbed in atmospheric particles or occur in vapor phase. Polychlorinated biphenyls (PCBs), organochlorine pesticides (OCPs), dioxins, dibenzofurans and polycyclic aromatic hydrocarbons (PAHs) are few examples of POPs. Microbial degradation technology such as bioremediation is emerging as a suitable and cost-effective tool when compared to the traditional technologies for treating POPs. This technique involves three important components: microbe, contaminant and nutrient, forming the bioremediation triangle for degradation of pollutant. This article is a brief description of microbes mediated bioremediation of POPs including bacterial, fungal and algal degradation and enzymes involved in the process.

1. Introduction

Persistent organic pollutants (POPs) are chemicals of global concern. POPs are bio-accumulative and possess biomagnification properties. POPs are synthetic chemicals that do not degrade easily in the environment, resist chemical, biological and photolytic degradation, have a strong affinity towards fats (lipophilic) and can either be absorbed in atmospheric particles or occur in vapor phase. Because of their semi-volatile nature, these pollutants can be transported to long distances in the atmosphere. Major organic chemical compounds that cause contamination in environment are polychlorinated biphenyls (PCBs), organochlorine pesticides (OCPs), dioxins, dibenzofurans and polycyclic aromatic hydrocarbons (PAHs) (Ersekova et al., 2014) (Table 1). On the basis of their use POPs can be classified into two types - organochlorine and industrial chemicals. According to the United States

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Table 1: Characteristic and structural properties of persistent organic pollutants (Boudh et al., 2019)

Persistent organic pollutants (POPs)	Characteristic property	Use and negative impacts
Pure aldrin	White	Used for termite control. Toxic to human but much less toxic to plants
Dieldrin	Colourless to light tan crystals	Control wood borers, termites, and textile pests. Highly toxic for fish
Endrin	White crystalline solid	Used as insecticide and rodenticide. Half-life in soil up to 12 years. It is highly toxic for fish, aquatic invertebrates, and phytoplankton
Chlordane	Water insoluble, melting point is 104°C	Half-life of 4 years and detected in arctic air, water and organisms
Pure heptachlor	White crystalline solid, insoluble in water	Persistent dermal insecticide with some fumigant action
(DDT)	Odourless, colourless crystalline, or white powder form	Used to control mosquito vectors of malaria in numerous countries. DDT is highly persistent in soil and its half-life extends up to 15 years. DDT is highly toxic to fish, birds and affect their behaviour. The International Agency for Research on Cancer (IARC) has classified DDT as a possible human carcinogen
Toxaphene	Waxy solid, yellow to amber colour with a chlorine/ turpentine - like odour	Most heavily used OCPs. Toxaphene is highly toxic for fish and shows carcinogenic effect on rats and mice but it is nontoxic to plants
HCB	Chlorinated monocyclic aromatic compound, white crystalline solid	Used as fungicide. Wood preservatives. It shows toxicity in aquatic animals and is a possible human carcinogen
PCBs	Clear to yellow	Possible human carcinogens
Hexachlorocyclohexane	Consisting of a six-carbon ring	Possible human carcinogens
Polychlorinated dibenzo-p-dioxins (PCDDs)	Contains a dibenzo-1,4-dioxin skeletal structure	In some cases, toxic for humans
Polychlorinated dibenzofurans (PCDFs)	Tricyclic aromatic compounds	In some cases, toxic for humans

Environmental Protection Agency (USEPA) about 16 PAHs are considered as hazardous and carcinogenic to humans.

2. Historical Outlook of POPs

Organochlorine pesticides, such as benzene hexachloride (BHC) and dichlorodiphenyltrichloroethane (DDT), heavily contributed towards consistent supply of crop production post-World War II, but soon after mention of these pesticides as toxic persistent compounds in Rachel Carson's book "Silent Spring", they were banned in many developed countries in the mid-1970s. India continues is one of the major contributors of persistent organic pollutants globally. Use of DDT and HCH, persist in agricultural and pharmaceutical industries. In India the first pesticide production plant was established in 1952, at Rishra, West Bengal. After that, several pesticide manufacturing plants were established leading to several fold increase of technical grade pesticides. Taking notice of the problems that arose due to POPs, world scientific, technical and political energy came forward to implement the Aarhus protocol and Stockholm Treaty on POPs. On 24th June 1998, the Executive Body adopted the Convention on Long-range Transboundary Air Pollution (CLRTAP) in Aarhus (Denmark). The basic aim of this protocol was to eliminate any emission, discharge and losses of POPs. Figure 1 shows the 12 dirty dozen listed in three categories, according to Stockholm treaty. The Ministry of Environment and Forest, GOI, has also submitted the National Implementation Plan (NIP)

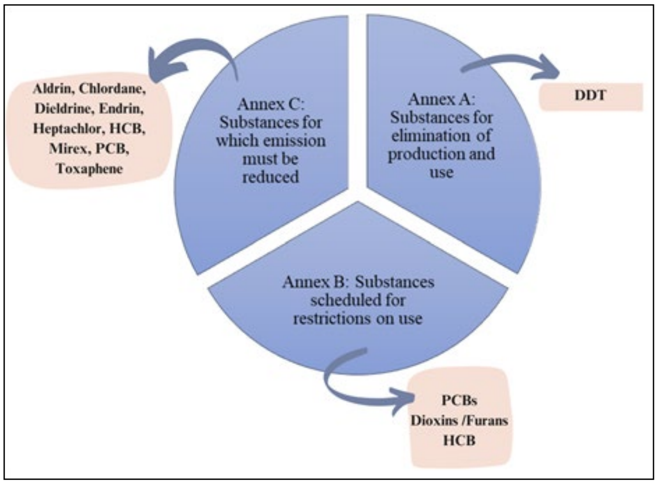

Figure 1: Chemicals listed in annexes of Stockholm convection

of the Stockholm Convention, in order to undertake activities concerning this global issue.

3. Microbial Degradation – a Constructive Tool for Bioremediation of Persistent Organic Pollutants

Traditional technologies for treatment of POPs including chemical reduction, land filling, incineration, solidification alkali metal reduction, solvent extraction and stabilization since long have been utilized by many industries. But due to high cost, requirement sophisticated instruments and manpower, such methods are not economical. Hence to over these problems of conventional technologies, microbial degradation technology is emerging as a suitable and cost-effective tool, especially the field of bioremediation (Figure 2).

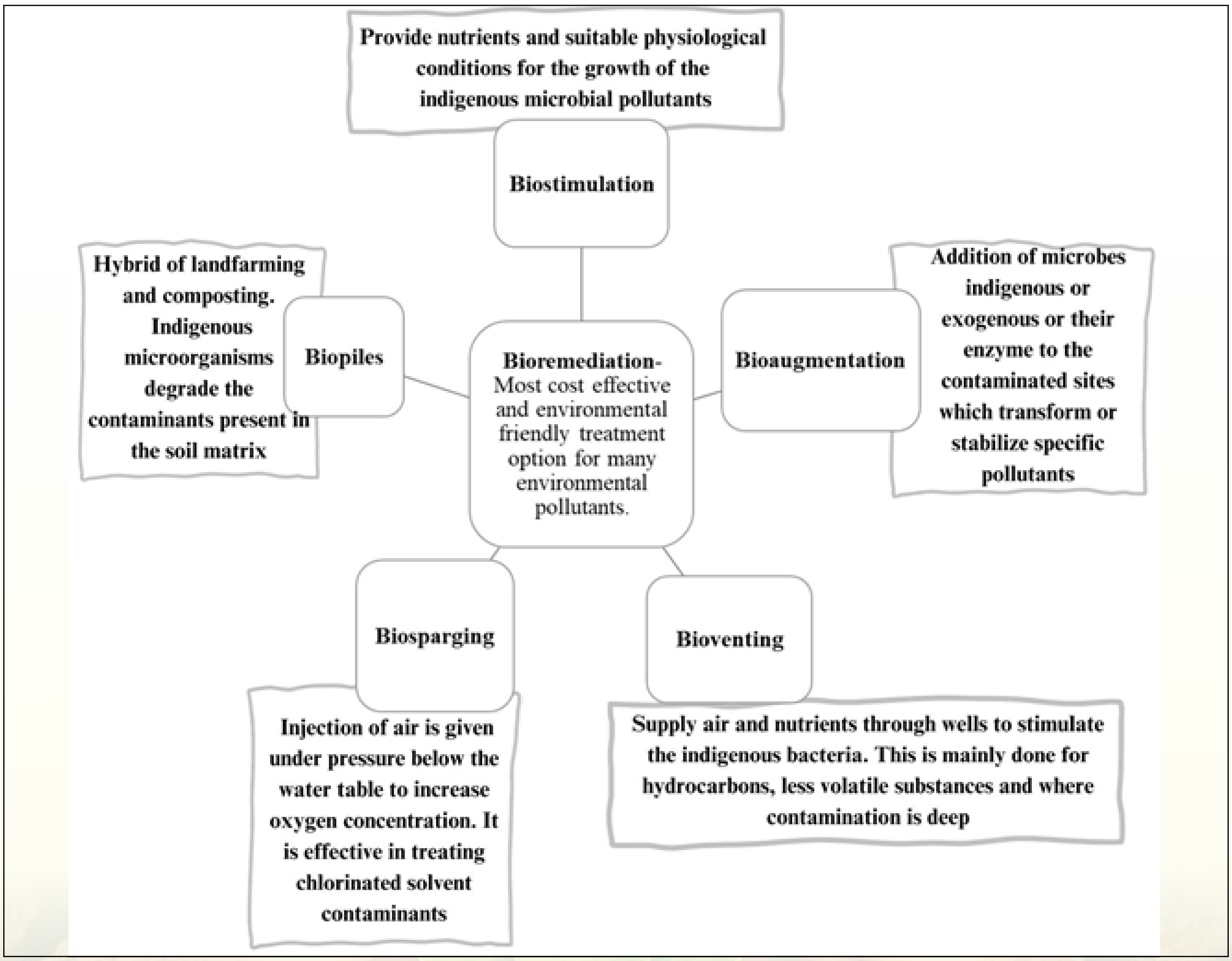

Figure 2: Bioremediation methods of POPs

Microbes found in nature have the ability to utilize hydrocarbons, minerals, pesticides, metals and lindane by changing their physical and chemical characteristics into simpler and less toxic compounds, further using them for nutrition, growth and other metabolic activity purpose (figure 3). Degradation by any kind of microorganism involves three important components: microbes, contaminant as source of food for these microorganisms and nutrient (nitrogen, phosphorus, electron accepter, etc.). These three components form the "bioremediation triangle" (figure 4).

Enzymes produced from bacterial, fungal and algal sources like laccase, oxidoreductases, manganese peroxidases (MnP), hydrolases, lignin peroxidases (LiP) and glutathione S-transferases (GST) are reported to degrade and detoxify POPs by catalyzing various enzymatic reaction of, oxidation of aromatic compounds, reduction of nitro and amino groups, ring cleavage and hydrolysis, addition of functional groups, dichlorination, etc. to change their chemical structure (Boudh et al., 2019).

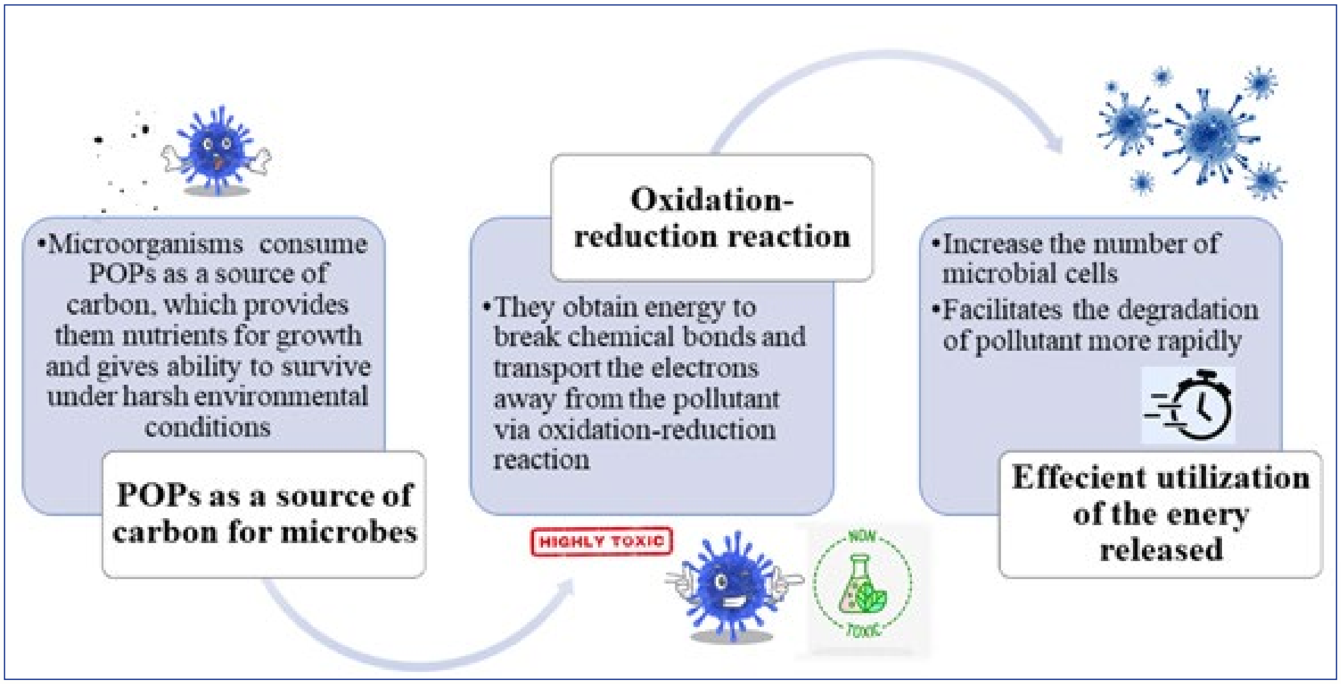

Figure 3: Principle of microbial degradation of persistent organic pollutants

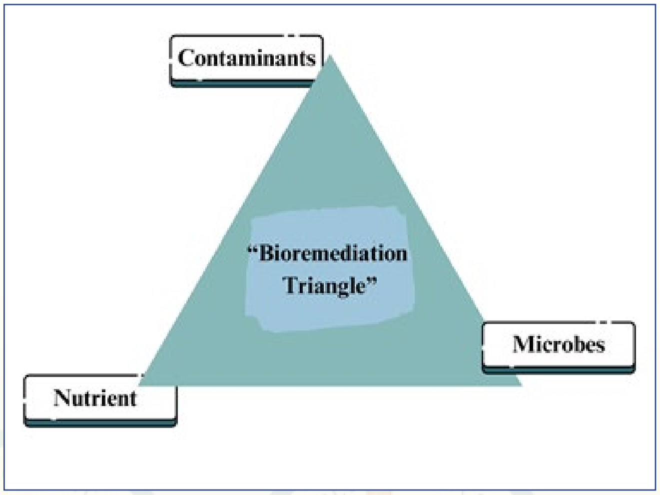

Figure 4: Three vital components for microbial degradation forming a bioremediation triangle

4. Microbial Strains and their Specificity in Degrading Persistent Organic Pollutants

Various microorganisms have potential for the significant remediation of POPs from the environment (figure 5). These include bacteria, fungi, algae, and actinomycetes, capable to degrade POPs through various mechanisms (table 2).

4.1. Bacterial bioremediation

Many aerobic bacterial strains such as *Bacillus*,

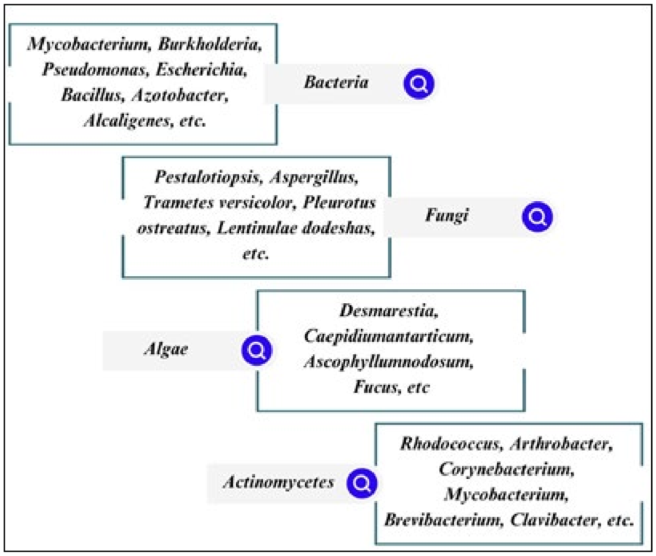

Figure 5: Microbes involved in degradation of POPs

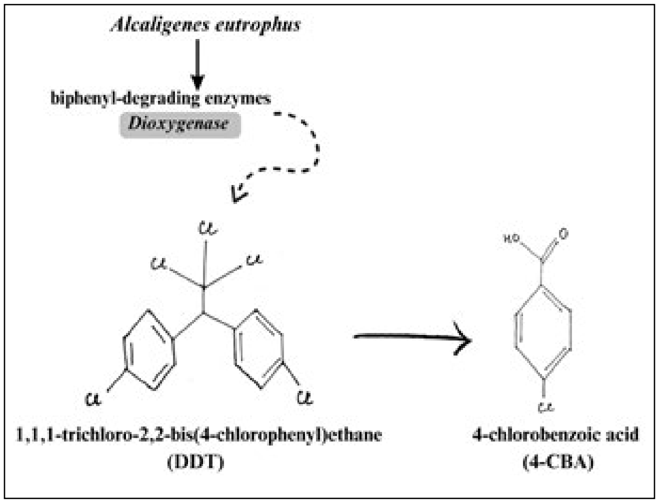

Figure 6: Aerobic degradative pathway of DDT by *A. eutrophus*

Pseudomonas, Micrococcus, and *Escherichia* are capable to bio-transform pollutant into carbon dioxide gas. Whereas, anaerobic bacteria like *Methanococcus, Methanospirillum, Desulfovibrio, Desulfotomaculum*, etc., can react with functional group of organic pollutants and degrade it, with removal of electron and lead to formation of methane and inorganic salts. Aerobic degradation of DDT by *Alcaligenes eutrophus* is done by dioxygenase enzyme that cause oxidation of DDT into 4-Chlorobenzoate (figure 6). A different strain, *Alcaligenes faecalis* is effective for the degradation of organochlorine-endosulfan residue (Boudh et al., 2019). Polycyclic aromatic hydrocarbons (PAHs) are well known to have high hydrophobicity, low solubility and are not easily eliminated from the atmosphere. *Alcaligenes, Mycobacterium* and *Pseudomonas,* can metabolize PAHs completely into CO_2 and other metabolic intermediates. *Pseudomonas mendocina* can degrade phenanthrene with the help of two active enzymes, PAH dioxygenase and catechol-2-3 oxygenase (Boudh et al., 2019). Polychlorinated biphenyls (PCBs) are another extremely toxic and hazardous pollutants. They can be aerobically and anaerobically degraded by bacteria like *Bacillus, Pseudomonas, Burkholderia* and *Rhodococcus* (Ahmed and Focht, 1973). These bacteria utilize biphenyl as a carbon source and bio-transform PCBs into trichloroacetic acid (TCA) and CBA via metacleavage pathway using types of biphenyl (Bph) dioxygenase (BphA, BphB, BphC, and BphD).

Another very important heterogenous group of bacteria belonging to diverse phylum of gram-positive bacteria is *Actinomycetes*. They are economically significant to soil ecosystem and are capable of degrading and detoxifying array of organic pollutants like hydrocarbons, pesticides like organophosphates, organochlorines, s-triazines, carbamates, etc., and many aromatic compounds. The species belonging to genus *Clavibacter, Mycobacterium, Corynebacterium, Arthrobacter, Brevibacterium, Streptomyces, Rhodococcus*, etc., have ability to degrade pesticide via mechanism of co-metabolism. Cytochrome P450s are pressing enzymes behind bioremediation by *Actinomycetes*.

4.2. Mycoremediation

Fungi are group of most primitive and diverse saprophytes. These are eukaryotic organisms capable of degrading substances consisting of lignin and cellulose. White rot fungus, *Phanerochaete chrysosporium* can break fluorine and benzopyrene into quinone by using certain intracellular as well as extracellular enzymes (Bhattacharya et al., 2012). Lignin peroxidase (LiP) is an extracellular enzyme belongs to the family of oxidoreductases, responsible for the oxidative cleavage in nonphenolic substrates, whereas manganese peroxidase (MnP) is a glycoprotein that oxidize Mn (II) to Mn (III) and catalyzes the degradation of phenolic substrates. Other potent enzymes responsible for degradation of PCBs are lipases, versatile peroxidase, cytochrome P450 and laccases (Bhattacharya et al., 2012). Ligninolytic enzymes found in many other strains of fungi, e.g., *Pestalotiopsis, Aspergillus, Pleurotus ostreatus, Trametes versicolor, Lentinula edodes* and mould can degrade PCBs through oxidation and reduction.

Table 2: Half-life, microbe involved in degradation and degradation pathway of different persistent organic pollutants (Pang et al., 2022; Fuentes et al., 2016; Qiu et al., 2018)

POP	Half-life (Years)	Microbes	Degradation pathway
Aldrin	1.5–5.2	*Pseudomonas fluorescens* (Bacteria), *Pleurotus ostreatus* (Fungus)	Oxidation, reduction and hydroxylation.
Dieldrin	5	*Burkholderia* sp., *Cupriavidus* sp. (Bacteria), *Penicillium miczynskii* (Fungus)	Oxidation, reduction, hydroxylation, and hydrolysis.
Endrin	12	*Cucurbita pepo* (Zucchini), *Cucumis sativus* (Cucumber)	Desorption, uptake and translocation though roots.
Chlordane	More than 20	*Streptomyces* sp. (Actinomycetes)	Dechlorination
Heptachlor	4.3	*Shigella* strain H	Transformation, hydrolysis, and reduction.
DDT	8	*Serratia marcescens* DT-1P, *Alcaligenes eutrophus*	Aerobic degradation
HCB	2.7 – 6	HCB-dechlorinating microorganisms	Dechlorination
PCBs	10	*Bacillus*, *Pseudomonas*, *Burkholderia* and *Rhodococcus*	Aerobic and anaerobic degradation
TNT	1.9 – 5.2	*Porphyra yezoensis* (red alga), *Acrosiphonia coalita* (green alga), *Portieria hornemannii* (red algae), *Desulfovibrio* and *Clostridium*	Mineralization or Reduction pathway through bacteria

4.3. Phytoremediation

Hydrocarbon-degrading enzymes produced by algal species like *Dermarestia sp.*, *Ascophyllum nodosum*, *Rhodococcus, Fucus sp.,* and *Caepidium antarcticum* can efficiently degrade hydrocarbons. Red and green algae (*Porphyra yezoensis*, *Acrosiphonia coalita*, *Portieria hornemannii*) under axenic conditions can degrade 2,4,6-trinitrotoluene (TNT) and 1,3,5-trinitrobenzene into 2-amino,4,6 2,4-Dinitrotoluene (DNT) and 4-amino, 2,6, dinitrotoluene (Cruz-Uribe et al., 2007).

5. Conclusion

Persistent organic pollutants pose a significant environmental threat, requiring prompt removal upon spillage or contamination. Existing physical and chemical methods for removal are not optimal. A recent breakthrough involves the use of microorganisms in biological methods, proving to be both efficient and cost-effective in degrading persistent organic pollutants. Nevertheless, additional research and intervention are needed to standardize the microbial-mediated breakdown of these pollutants.

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