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ROLE OF MICROBES IN WASTE WATER TREATMENT: AN OVERVIEW

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ABSTRACT

The micro-organisms have their own properties during waste water treatment and mainly focus on the process. Human activities and their unbalanced development and unhygienic living processes resulting a threat to environment and all living organism such as waste water. Huge amount of waste discharge in water through various resources either industries or domestic sewage is rising concern in environment. There has been few waste-water treatment technology discovered. Chemical treatment has major negative side effect so treatment through biological treatment is comparatively far better than others as easy to operate and low maintenance. Numerous microorganism such as microalgae, fungi, bacteria, decomposers and protozoans etc accumulate dissolved organic substance or degrade/ catalyse them into smaller compound such as water, CO_2 or methane, known as term- biodegradation, this natural phenomena through techniques used to clean environment called bioremediation. Even some species of algae as- *Chlorella,Staurodesmus* etc have a beneficiary impact on sewage treatment as they absorb harmful dissolved elements, bacteria and other toxins. Moreover, fungi with help of their filamentous structure destroys sludges. In local waste water treatment plants, species of bacteria as proteobacteria has appeared predominant (21-62%) and specifically beta proteobacteria class majorly involved in organic and nutrient removal. Therefore, in short presence of microorganism decreases the level of pollution in sewer waste/ effluent.

Keywords- Biological treatment, microbes, environmental pollution, wastewater. breakdown, Biochemical oxygen demand.

Introduction

Clean water is an essential need of human kind. Though world is making progress in all around economy growing, increase in global quality of life, innovation etc, but still there's major population millions of people deprived from pure/ clean water, which is fundamental human right to provide /have access to clear water. Availability of pure water is more or less under 3% of all water on earth that has large impact on human quality of life, growth and development. As per recent year results, population will grow rapidly in upcoming years too and expected to be twice of current world population by the 2030. As per population demand for clean water will rise and if we did not deal properly with water pollution crisis now, there will soon another pandemic knocking on door. Even now half of population is growing through impure/ poor water, no sanitation and unhygienic environment (Singh et al. 2018). So, there should be a sanitation infrastructure in affected region/ countries by economic sectors (protection and restoration of water) it is global concern now huge amount waste of industries discharged in natural water resource (Singh et al. 2014). These detailed should be transparent and headlines by global media to spread awareness and authorities to make effort and take strict action against pollution. As per global reports, heavy metals contamination in fresh water causing major health issues is matter of concern various heavy metals in water are toxic for human and other living organism. As, Cd, Hg etc.

It is essential to have vast knowledge of waste water source to test their different aspects as – physical, chemical, biological etc to provide treatment strategy and hygiene education. Current research on waste water source can be classified into biological, physical and chemical treatment techniques, moreover in-situ or ex-situ technologies.

In-situ actions stand for- treatment conducted at specific/particular-site. Meanwhile ex-situ involves in elimination of pollutant at a remote place. For example- in-situ – flocculation, used as in situ method of treatment for surface and ground water (Yadav et al. 2021). It can be defined as process, either spontaneously or addition of clarifying agent leads colloidal particle came out into sediment in form of flake. Though physical water treatment is not much considered due to its negative sight effect. There must be precautions as large amount of used chemical could be lethal for user and it can cost more for large scale water diversion.

Biological waste water treatment technology has been recognised as the best treatment technique (Verma and Mohanty 1994). It is also known as biodegradation- a process by which organic substances broken down into smaller compounds as co_2 and water or methane by living microbial organism as- fungi, bacteria, protozoans etc decomposers, moreover micro-fauna. Microorganism maintain the stabilization of biological system by degrading pollutant (Andersson et al.). The biological waste water treatment has relatively cheaper cost, minimal excretion and lower damaging effect on environment, it is preferred in contrast to other methods. Moreover, it holds an economic advantage against both physical and chemical technology in terms of cost and investments. This review article has show`s that the application of biological waste water treatment and elaborate microbial aspects in waste water treatment and advantage to human kind.

Biological waste water treatment:

Biological processing is the most efficient way of removing organic matter from municipal wastewaters. These living systems rely on mixed microbial cultures to decompose and remove colloidal and dissolved organic substances from solution. The treatment chamber holding the microorganisms provide a controlled environment; for example, activated sludge is supplied with sufficient oxygen to maintain an aerobic condition. Wastewater contains the biological food, growth nutrients, and inoculum of microorganism. Persons who are not familiar with wastewater operations often ask where the "special" biological cultures are obtained. The answer is the wide variety of bacteria and protozoa present in domestic waste seeds the treatment units. Then, by careful control of wastewater flows, recirculation of settled microorganisms, oxygen supply, and other factors, the desirable biological cultures are generated and retained to process the pollutants. The slims layer on the surface of the media in a trickling filter is developed by spreading wastewater over the bed. Within a few weeks, the filter is operational, removing organic matter from the liquid trickling through the bed. Activated sludge in mechanical, or diffused-air, system is started by turning on the aerators and feeding the wastewater.

Initially, a high rate of recirculation from the bottom of the final clarifier is necessary to retain sufficient biological culture. However, within a short period of time a settleable biological floc matures that efficiently flocculates the waste organics. The biological treatment technology is divided into two ways – (i) Aerobic and (ii) Anaerobic. In aerobic method, the pond contains algae and bacteria under aerobic condition.

Aerobic process- aerobic waste water treatment is a biological process that takes place in the presence of oxygen. It is the rapid and most efficient biological waste treatment which remove up to 98% of organic pollutants and yield a cleaner water effluent than anaerobic treatment.

Activated sludge process-The activated sludge process is the most widely used biological waste treatment in secondary stage of wastewater treatment. An activated sludge process refers to a multi-chamber reactor unit that makes use of highly concentrated microorganisms to degrade organics and remove nutrients from wastewater to produce a high-quality effluent. In this method, the sewage containing organic matter with the microorganisms is aerated (by a mechanical aerator) in an aeration tank (Singh 2015). This process speeds up waste decomposition. Aeration in an activated sludge process is based on pumping air into a tank, which promotes the microbial growth in the wastewater. The effluent from the aeration tank containing the flocculent microbial mass, known as sludge, is separated in a settling tank, sometimes called a secondary settler or a clarifier.

Anaerobic treatment-anaerobic treatment is proven and energy-efficient method for treating industrial wastewater (Li, et al. 2019). This treatment process is effectively utilized to treat high strength waste water and it employs organisms that function in the absence of oxygen and it will typically treat high-strength waste water to a level that will permit discharge to a municipal sewer system. Here, the amount of sludge produced is very small when we compared to aerobic treatment. Anaerobic treatment is a slow process and it occurs in many different stages. Anaerobic digestion is biological process which is used in wastewater treatment plants for sludge degradation and stabilization (Qadri et al. 2020). Once the process is completed, the wastewater can undergo

many additional treatments. This process is accepted because it is able to stabilize the water with little biomass production (QU X et al.2016).

Biogas is produced as the bacteria feed off the biodegradable material in the anaerobic process. Overall, the process converts about 40% to 60% of the organic solids to methane (CH_4) and carbon dioxide (CO_2).

Ecology of biological waste water

These microbes hold their own specific virtue, and during treatment technology they interact or co-operate on the basis of mechanism, either aerobic or anaerobic. Fungus can survive even in adverse condition as- reduced pH and less nitrogen, it is appropriate for water treatment. It has web like filamentous structure so have capability to deteriorate the sludge settle. Moreover, rotifers in initial of waste water treatment is a good sign, they can absorb organic matter, dispersed bacteria and harmful dissolved elements too.

The bacteria reproduce exponentially by binary fission, rapidly increasing the number of viable cells and biomass in the medium. The presence of excess substrate promotes the maximum rate of growth possible, limited only by the ability of the bacteria to reproduce. In this exponential growth phase., the increase in both the number of visible cell and the accumulation of biomass is represented by

$$R_g = \mu X$$

Where r_g = Biomass growth rate, mg/l per day

μ = specific growth rate (rate of growth per unit of biomass), per day

X = concentration of biomass, mg/l.

The declining growth phase is the result of diminishing substrate, limiting bacterial growth. the rate of reproduction decreases and some cells die so that the total biomass exceeds the mass of viable cells. When the substrate is depleted at the end of declining growth, the number of viable bacteria and biomass remain relatively constant, resulting in a stationary phase. Rate of growth in the declining growth is described by the Monod equation

$$\mu = \mu_{\max} \frac{[S]}{K_s + [S]}$$

Where μ = maximum specific growth rate, per day

S = concentration of growth -limiting substrate, mg/l.

K_s = saturation constant (equal to the limiting substrate concentration at half the maximum growth rate) mg/l.

Importance of microalgae in waste water treatment

Microalgae have capability of accumulating nutrient, toxin, heavy metal etc in their cells for growth and development, so they absorb them and remove N, P, heavy metal, bacteria and other substances from water body in short time period, and moreover they reduce BOD. Some species of microalgae reported in pond stabilization are- *micrractinium, chlorella*, *golenkinia*, *ankistrodesmus*. (Choudhary et al. 2015). These microalgae can be helpful in wide range sewage system and count as tertiary treatment. biological treatment is way more promising and reliable than chemical treatment.

Stabilization of pond

It is a kind of non-conventional system, also known as- biological treatment system, which is easy to operate and limited equipment maintenance enhance its preference or make it better. And with the help of appropriate architecture to cultivate bacteria and algae, it will completely remove organic waste in water (Sanda and Ibrahim 2020). As a result, it will decrease problem rising during wastewater treatment.

Bacteria and structural unit

Heterotrophic bacteria have efficient role in removing / disposal of waste, organic and inorganic matters. They work in clusters forms as floc, biofilm or granule.

Extracellular polymeric substances (EPS)

Microbial origin such as- protein, polysaccharides, DNA etc. The production of EPS placed by cell lysis, cell secretion biosorption from waste water treatment. It is important in determining the biomass structure and properties (Dwivedi AK. 2017). It sustains and protect it during treatment against toxin, forms a diffusion barrier to remove toxic compound, moreover help in sludge flocculation, settling, metal binding, dewatering etc.

Flocs

Floc is activated sludge or masses of bacteria held together by slime, it forms bacterial colony by the attachment of cells and organic pollutant by physiochemical process. Flocs component is mainly – bacteria and EPS. And their stability is facilitated by environmental stress and microorganism content. Biological floc is composed of bacteria, fungi and protozoa.

They have a crucial role in degradation of organic substance of waste water treatment (Oh et al. 2010). It is surface associated assemble of microbial cell enclosed in EPS matrix. Biofilm formed after microorganism is immobilized on a solid platform supported through interactions as- electrostatic interaction, hydrophobic interaction and covalent bond (Singh et al. 2022). The process leads by microorganism and their component- cilia, cell-wall, fimbriae and EPS.

Biofilm formation considered in 4 main stages-(i) bacterial attachment to surface. (ii) microbial formation (iii) biofilm maturation(iv) detachment/ dispersal of bacteria (Hamza et al. 2016). Transport in biofilm drive by diffusion, while its thickness depends on ability of substances and oxygen penetration. When channel and pores crossed into biofilm this cause resistance to toxic compound.

Aerobic granule

Activated sludge having bacteria can form aerobic granules. They are actually a type of sludge, that can self-immobilize microorganism into strong compact structure and provide settlement / stability. Granular sludge is able to remove nutrient and increase tolerance to toxicity.

Protozoans- protozoa also crucial role in removal of organic waste, they mainly feed on bacteria, and through predation, they keep maintain through predation, they keep maintain their density bacterial population in water body (Dwivedi AK. (2017). They promote their development and growth and eliminating mineral nutrients. Moreover, they even secrete some stimulatory substance to promote beneficial bacteria growth and activity and suspend other particles too.

Biological treatment with activated sludge

Water contamination is a major environmental issue. It has been a threat to human health, ecology and economic stability globally. At present time biological treatment is a best way for treating waste water due to its simple procedure, easily applicable and convenient low cost.

In biological treatment especially activated sludge is one of the most important waste water treatment technologies. 60% of the sewage treatment is based on this technology. Waste water is mixture of heavy metals, organic matter, nitrates, hardness, suspended solids, gases, carbonates, and chlorides (Ahmaruzzaman and Sharma 2005). The most dangerous parameter is organic matter. Activated sludge process is used to solve organic matter such as - oxidising carbonaceous biological matter, oxidising nitrogenous matter (mainly ammonium and nitrogen), by nitrate-reducing bacteria (NRB) and removing nutrients (nitrogen and phosphorus), oxidising sulphur biological matter by sulphide-oxidizing bacteria (SOB). Microorganism as- bacteria present compound to grow (Chaoua et al. 2019). For bio flocculation suspended microorganism aggregate through EPS secreted by themselves. Aeration tank, settling tank, and Storage tank are present in activated sludge. The function of the aeration tank is to supply oxygen to the gain/ liquor/ aperitive, and the settling tank allows biological flocs to settle, and the function of the storage tank is to receive the final liquid waste. Activated sludge is a biological indicator, they eradicate (stamp out) organic compounds through metabolic reactions of microorganism (Oh. et al. 2001). They oxidised and adsorb highly complex compounds such as – ammonia, and nitrogen instead of degrading them, and

microorganism growth and the adsorption capacity is representing the removal process of organic matter in the wastewater.

Besides, EPS is a basic fundamental element for sludge floc matrices. Therefore, it is necessary to banish (eliminate) EPS by using surfactants like sodium dodecyl sulphate (SDS) during pre-treatment to increase the removal rate of sludge degradation. Because formation of EPS enzyme complex may reduce the reactivity of enzymes on the sludge. Removal of EPS provide substrate to microorganism, and improves lytic activity for anaerobic biodegradability, thus biogas yield production will increase (Rocca et al. 2007). In addition, the morphology of activated sludge can be damaged or affected by ionic surfactants such as linear alkylbenzene

sulphonate, for example, the sodium salt of dodecylbenzene sulphonate. Which causes fragmentation of the flocs and lysis of the protozoa.

Composition in waste- water treatment

Waste water treatment plant has abundance of biodegrading microorganism they are well organised in species-rich structure. As- bacteria, protozoa, metazoan, microalgae, fungi filamentous bacteria etc. In biological treatment, these microbes are mostly in the forms of masses as – biofilms, floc and granules (Dubey and Raichaudhary 2019). It is identified through various electron microscopic methods, that there is presence of highly complex extracellular polymeric substance in culture media activated sludge, granulated sedge and biofilm.

EPS actually binds these microbes in clumps on surface through physiochemical interaction and keep them aggregated in 3d gel – like hydrated matrix (Cornwell et al. (2019). These interactions are weak in nature, include hydrogen bonding, electrostatic, hydrophobic, Vander walls forces. Meanwhile EPS are by product of metabolic activities on the surface of bacterial cell. EPS generated by microbes in bioreactor, who consume organic material of waste-water. Accumulation of EPS lead by various mechanism as – excretion, secretion, lysis of cell, absorption of component. EPS has biodegradability and biosorption protein, nucleic acid, phospholipase and other component, moreover, some humic substance and uric acid reported in EPS.

Microbial community in waste-water treatment plants

In waste-water treatment**,** exposure of microbes and their community having a role will be beneficial to work on causes (Kumari et al. (2023). There is various treatment technique utilized in biological method- as-denaturing gradient gel electrophoresis terminal restriction fragment length polymorphism, 16S rRNA gene-clone library, fluorescence in-situ hybridization analysis etc. They are leading it to an evaluation of microbial community present in waste-water treatment plant. it is more like culturing method (maintaining condition for growth) for microbial community characterization.

This method for analysis is direct and effective, in a natural environment culture setup has most of bacterial exposure, so cannot be cultured in synthetic media or in laboratory, so molecular based techniques is considered (Shah and Ahmed 2023). In this regard- phylum proteobacteria is reported predominant approx. 21-62% in local waste water treatment plant, though class- betaproteobacteria, largely involve in organic waste and nutrient removal. Moreover, *Acidobacteria*, *Bacteriodetes* and *chloroflex* are subdominant in treatment techniques. It is reported in textile industry during microbial community assessment of textile waste-water treatment plant in comparison with municipal waste-water treatment, there is specific key microbes that involve in purification of textile waste. The study discloses that there is generous amount of nitrifying and denitrifying bacteria, those were involved in phosphate accumulation too, where as in textile industry sulphur reducing bacteria were identified (Mittal. A 2011). In coalmine water treatment system to study presence of microbes, an illumine throughput- sequencing method was setup. The result demonstrates the occurrence of major genera as-*azoarcus*, *comamonas*, *thiobacillus, pseudomonas*, *ohtaekwangia, thauera*, and some nitrifying bacteria as-*Nitrosomonas, nitrospira*. results show difference in coalmine in compare to municipal waste treatment plant.

study of microbial coal- mine water treatment and their communities will be helpful in treatment approaches and further development.

Microbial electro- remediation technology

Human activities and rapid industrialization leading us to polluted environment and generating waste water including metal pollutants. Heavy metal in waste water is lethal to health and increase environmental hazards (Amir et al. 2001).

To avoid toxicity and cancerous impact of different metals, there's strict regulation and guidelines on disposal or transport of metals are adapted. For example- American EPA priority listed- on the basis of pollutants, naturally occurring arsenic (As) is highly toxic and disastrous to human kind (Das and Veziroğlu 2001). Even around the world, developing nations and millions of people are affected with metal toxicity (arsenic toxicity). It ended up in having neurogenic disease or if get worse than death. An ideal solution for it would be store them during treatment and regain instead to get rid of them. Though there's some new techniques and methods exists to eradicate them or arsenic treatment from waste water, recent innovative researches and thoughtful techniques leads towards development and growth. They are practical, at cheaper cost and noted ecofriendly. Still several researches, more development and advanced technology should be considered in the direction of pollution remediation. It requires broader scope of management, applications and conventional techniques in metal removal as- precipitation, adsorption, coagulation etc (Gaikwad 2022). Technology approaches for heavy metal ion containing water and industrial waste treatment through ion exchange, carbon-based adsorption, membrane technology is way more expensive to execute. So, it's not easy to apply on large scale (Kavitha et al. 2014). The recent concern of impactful method to remove contamination is new research area as result a new method of bio electro catalysis came into existence, that can deploy or mobilize pollutant for reduction in sub- surface (Khanal et al. 2017). It is emergent of bio- electrochemical system (BES) and microbial fuel cells (MFCs).

Metal contamination of water can be purified /dealt with MFC 's anode and cathode. Positively charged anode are used to carry electron to cathode (negative charged) and result in precipitation through terminal electron acceptor. Bio-electro system treat waste-water through oxidation and reduction.

Conclusion

Day-to-day, huge amounts of waste water is generated from various sources including domestic sewage, industries, agricultural operations and many more. Waste water contain suspended solids, colloidal matter, dissolved solids toxic substances, pollutants and other contaminants that can be lethal to organism also affect the environment, induce water-borne diseases in humans and harm nearby vegetation and animals (Malisa et al. (2019). So as solution various treatment approaches are being considered for pollution remediation. Biological method is one of most appropriate method, economically realistic and easily applicable. Biological system is governed by the ability and action of microorganism to decompose and degrades the target contaminants. Anaerobic, aerobic processed are used in water treatment (Cornwell et al. 2019). Bacteria, protozoa and some useful, microorganism are responsible for degrading the organic matter and removing toxins in wastewater. Thus, in biological method microorganisms play an important role in water treatment. It is necessary to control the

activity of microorganism, so their efficiency can be utilized for the purpose of bioremediation. Moreover, their enzyme is effective catalysis for each substrate to degrade. It has low cost and chemical free, microbial wastewater treatment is environment friendly but also have drawback to work on as slow process, it takes long duration to be executed and also pathogen rise during bioremediation, that can obstruct the process.

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