



A Review on Purple Sulphur Bacteria for Waste Water Treatment

Pichandy Muthu prasanna^{*1}, M.Komala², J.Padmavathy², Dr. P. Bharghava Bhushan Rao³, Yuvabharathikannan D⁴ and Kannaiyan Suria Prabha⁵

¹Professor (visiting) , Pallavan college of pharmacy , Kanchepuram district , Tamilnadu, India.

²Dept of pharmaceutics ,School of Pharmaceutical Sciences, Vels Institute of Science, Technology and Advanced Studies, Pallavaram, Chennai, Tamilnadu, India.

³Associate Professor, Department of Pharmaceutics, V. V. Institute of Pharmaceutical Sciences, Seshadri Rao Knowledge Village, Gudlavalleru- 521356, Krishna District, Andhra Pradesh, India

⁴B.tech genetic engineering (4th yr), Bharath Institute of higher education and research, Chennai, Tamilnadu, India

⁵Principal and Professor , Indraganesan College of pharmacy ,Madurai Main Road , Manikandam , Tiruchirappalli-620 012, Tamilnadu, India.

Abstract: Usage of microbes not only limited to antibiotics production but now have extended to waste water treatment. Microbes have made an immense contribution to the cost reduction process. Many chemical processes have been replaced by many biological process due to their cost effectiveness. . Many countries are shifting their strategies for waste and sewage water treatment from chemical techniques to these advantageous microbial techniques. These microbes particularly purple sulfur bacteria decomposes a great range of biological and other organic waste to a less-toxic or non toxic end products. The cost involved in getting these waste water converted to non toxic by-product by these bacteria found to be far more lesser than any other conventional techniques. Thus by providing waste water as a nutrient to this purple bacteria makes a good and great impact on the environment. This review discusses the habitat, mechanism of biodegradation, factors influencing these biodegradation and other related researches concerned with the biodegrading efficiency of Purple sulfur bacteria.

Keywords: Purple Sulphur Bacteria (PSB), Waste water treatment, Hydrogen sulphide gas, microbial mats

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*Corresponding Author

Pichandy Muthu prasanna , Professor (visiting) , Pallavan college of pharmacy , Kanchepuram district , Tamilnadu, India.

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I. INTRODUCTION

Micro organism-based biological strategies are being used to biodegrade toxic bio and human waste at a low cost. Most species of anaerobic microbes are likely to decompose a wide range of biological organic waste to an non toxicated and inorganic contents. Biological approaches no longer utilize chemicals to be used for the wastewater treatment which otherwise has a serious impact on the ecological community. Above all, ecological approaches that occur in the ecosystem for human-made waste are directed specifically at increasing their variability and severity. This biodegraded final by-products mostly comprises of carbon and water. Concurrent biodegradation of different waste materials, such as biohazardous and other natural wastewater or biodegradable waste, tends to be a possible and most commercially viable approach. Expenses associated with the combined bio decomposition of rough waste from manufacturing industries were usually very less than independent chemical remedial strategies. The Purple Sulfur bacteria (PSB) are proteobacteria that are capable of photosynthesis. They are anaerobic and are frequently found in stratified liquid habitats such as hot lakes, stagnant and stale water bodies or microbial mats at intertidal areas.¹ As in plants, PSB cannot utilize water as its reduction mechanism and therefore fail to produce oxygen. But, in their photosynthetic channels, they use sulfur as sulfide and thiosulfate as electron donor¹. For generating sulphur element the sulfur is oxidized and in turn this may be metabolized to form sulfuric acid. These purple photosynthetic bacteria are phototrophic proteobacteria, which are capable of generating their own energy through photosynthesis.² They have pigments composing of bacteriochlorophyll a or b along with different carotenoids, that provide colours which vary from purple green, brown and orange. These can be classified into two types: Purple sulfur (partly Chromatiales) and purple non-sulfur (Rhodospirillaceae) bacteria. In both biochemical and phylogenetic basis, the purple sulfur bacteria (PSB) vary from the purple Nonsulfur bacteria (PNSB). Purple bacteria(PSB) have already been shown to be important primary producers in some especially suitable environments for their existence.^{3,4,5,6,7} Gammaproteobacteria refers to the black sulfur bacteria.¹²⁰ Members of Gammaproteobacteria - chromium also uses Hydrogen Sulphide Gas (H₂S) for oxidation but not water like other microbes and result in elemental sulphur as bio waste product. Few gammaproteobacteria also oxidises methane for their metabolism and some dwell with animals living in vents of geothermal sites of oceans and deep seas. Purple sulfur and purple nonsulfur bacteria could be together and commonly called as purple bacteria.

1.1 Habitat of PSB

PSB have been the foremost bacteria found to be photosynthesizing without the need for oxygen with sulfur as their by-product. This was shown by first assessing the responses of the bacteria to various oxygen levels. It was discovered that the bacteria was rapidly moving away where even smallest oxygen content is absent, so a PSB bacterial was sampled and to them a light shone on one section or area of the sample making the others in the dark. Since the bacteria were unable to exist in the absence of light, all the bacteria have migrated into the light ring, producing much populated inside the light ring. But if oxygen is the byproduct

of bacteria, the space between individual bacteria would be bigger and bigger since much oxygen would have generated. But in early research, it has been found that the photosynthetic byproduct of this bacteria would never be oxygen due to the activity of the bacteria in the directed light.^{8,9} PSB typically found in anoxic areas of ponds as well as other aquatic environments wherever hydrogen sulfide builds up, as well as in "sulfur springs" fide can induce blooming of PSB. Photosynthesis by PSB requires anoxic situations and these bacteria do not survive in oxygen rich environments.¹⁰ Meromictic (completely stratified) water bodies such as lakes are the most suitable places for the production of PSB¹¹. Meromictic lakes segregate or stratify due to their higher dense (generally salt) base water but less concentrated (commonly freshwater) towards the top. Forming layers in holomictic water reservoirs also facilitates the development of PSB.¹¹ These reservoirs are thermally segregated; in spring and following hot weather, the freshwater surface is heated, rendering it less concentrated than the deeper cold water, which creates a secure sufficient stratification to the development of purple sulfur bacteria. When there is enough sulfate to sustain a reduction of this sulfate for sulfide formation in the sedimented surface, this sulfate can slowly spread into the anoxic lower waters where PSB develops to an intense cell mass using this sulfate, known as blooms, typically in conjunction with green photosensitive bacteria. In microbial mats, which grows in intertidal places, PSB can even be identified and have become a prevalent feature. These Mats, like the Sippewissett Microbial Mat, has diverse conditions owing to tidal movement and fresh water indwelling resulting in equally stratified ecosystems as meromictic ponds. Rise of PSB is supported by sulfur which is supplied in these intertidal waters from the biodegradation of organisms above them¹². The origin of stratification and sulfur helps the PSB to expand making the mats exist at these intertidal pools. Using the bio secretion of extracellular polymeric compounds which can attach the sediments in the water body^{13,14}, PSB can help strengthen these bacterial mat ecosystem sediments. PSB is often abundant in these microbial mats, including mats which grow in marine or hypersaline sea water³⁹ and in thermal spring waste water⁴⁰. Thiocapsa and Allochromatium bacteria were commonly found in aquatic microbial mats. Such PSB also build a thick layer of pigmentation between the cyanobacteria and the mat's bottom layers. Especially in marine mats, thiocapsa roseopersicina seems to be widespread, probably due to its metabolic flexibility. Such agility enables Thiocapsa Roseopersicina to take full advantage of the complex environments for growth that differentiate various layers of microbial mats¹⁰¹. Many PSB, can exist in variable environments factors and those which influences most is the extreme temperature, pH and salinity, thus promoting its existence in extreme conditions. These PSB are essential phototrophs since they

- (1) Ingest a potent toxin called H₂S and by their autotrophic ability add organic matter to the anaerobic environments;
- (2) Consumes organic materials, mainly unfermentable bio substances, by biopathways as photoheterotrophs; and
- (3) Give biologists the opportunity to study various photosynthetic bioreactions.

Anoxygenic phototrophic bacteria (APB) or PSB were widely spread in marine and a few in terrestrial habitats wherever light is accessible of appropriate intensity and

anaerobic/microaerobic requirements exist^{15,16,17} and they become mainly isolated with enriched colonies. Its desirable environmental niches cover ecosystems like lakes¹⁸, lagoons with sewage, intertidal areas¹⁹, ditch sedimentation, wetlands and wet soils²⁰. They also live in different aquatic habitats like fresh water bodies^{21,22} saline²³ alkaline^{24,25,23} acid aquatic reservoirs²⁶ geysers²⁷ Arctic²⁸ and Antarctic.^{29,30} The light quantity or intensity, pH, salinity, temperature and abundance of sulfide with oxygen are essential for the presence of its environment at various APB groups³¹, thus play a key role for the processes of carbon, hydrogen and sulfur cycles³². APB's successful viability requires light but also needs anoxic environments. This type has been most frequently found in rivers, ponds, waterways, and other freshwater habitats with H₂S^{33,34,35}. When these basic requirements are met, the actual physicochemical existence of the environment (sulphide intensity, pH, light content and brightness, temperature) determines the number and variety of PSB^{36,37,38}.

1.2 Photosynthesis of PSB

Microorganisms relating to PSB contain high concentrations of bacteriochlorophyll a and are capable of accumulating sulphur, inside the cell, except for the Ectothiorhodospiraceae group, which accumulates sulphur outside the cell⁴¹. Hydrogen and hydrogen sulphide are the fundamental supply of electrons in photosynthesizing operations, and the retained sulphur is potent enough to be oxidized to sulphate. The bacteria referring to the Chromatiales order, and Chromatium Okenii is the better known species of this kind. Photosynthesis happens at interaction sites on the cell surface, where photosynthetic pigments (i.e. bacteriochlorophyll, carotenoids) and pigment-adhering proteins become invaginated to build as vesicle storage sacs, and lined lamellar sheets^{42,43}. This is termed as intracytoplasmic membrane (ICM) that has increased contact area to enhance light absorption. Photosynthesis happens in oxygen less conditions in PSB. Bacteriochlorophylls a or b and different forms of carotenoids are significant photosynthetic pigments. Photosynthesis in all of these PSB relies on environments that are anoxic and lacking in oxygen since oxygen suppresses the bioformation of the photosynthetic pigments. All such bacteria could not use freshwater as a contributor of electrons but need more modified sulfur compounds such as sulfide instead for electron donor. Such bacteria could also use hydrogen as well as a number of similar biomolecules as contributors of photosynthetic electrons and use a variety of metabolic processes for energy production, carbon acculturation, nitrogen, sulfur, and phosphorus metabolism.^{44,45} Several phototrophic PSB have shown proliferation using reduced iron as an electron contributor^{46,47}. PSB being potent photoautotrophs, consist of modest photoheterotrophy, yet underequipped under darker illumination for its survival and growth. On the other hand, purple non-sulfur bacteria(PNSB), the foremost photoheterotrophs of nature worthy of photoautotrophy, exhibit various properties for their adaptation and existence.

1.3 Photopigment of PSB

For the metabolism of PSB, the bacterium transforms H₂S to basic sulfur element which needs to have at least a substrate source for nutrition and yield a readily separate and distinct elemental sulfur from the biomass. Much of the Sulphur

oxidising bacteria (SOB) is either rod-shaped or spherical creatures. Sulphur oxidising bacteria SOB is also exist in a gelatinous material, varying in size from less than 1 µm to few tens of µm. They contain a bacteriochlorophyll form, "a" – which is a chlorophyll related pigment produced by bacteria, algae, and cyanobacteria. Cell populations can be colored yellow, green, brown or purple due to their pigments. SOB also live in different tissues of invertebrates like those of Riftiapachyptila (vestimentiferan tube worms) and Calyptogena Magnifica('giant' white clams) dwell near deep ocean hydrothermal vents, There, by oxidizing the reduced sulfur and organic compounds and generate food through turning carbon dioxide into organic materials used by invertebrates.⁸⁰

1.4 Toxicity of hydrogen sulphide

Hydrogen sulphide (H₂S) is a colourless substance in gaseous form with a foul smelling similar to rotten smell. Usually, in geological fields such as volcanic areas, hot spring, oil and gas resource (up to 90% of its composition) hydrogen sulphide exists. The existence of sulphate reducing bacteria in different water sources and soil, particularly in rotting biological matter, has a negative effect on certain other species in the ecosystem. Sulfide is found in raw sewage as a balanced mixture of H₂S, HS- and S₂- ions. This malodorous, poisonous, corrosive, and colorless gas H₂S threatens to exit the sewage and cause serious damage to the environment. Also there are many relevant issues relating how to safely extract this biogas hydrogen sulfide. Sulfate-reducing bacteria(SRB) could outpace methanogens for substrates and other acetate in biogas plants if there is a large amount of sulphate to generate H₂, CO₂ gas⁴⁹. SRB metabolism's end product results to a harmful hydrogen sulphide and this may lead components of biogas fermenters and reactors to degrade and prevent methanogens development⁵⁰. High concentrations and constant exposure of H₂S can damage vegetation and produce various adverse effects on aquatic or marine ecosystems and aquatic farming. H₂S could affect various systems in the human body and the nervous system in particular is by far the most affected system. Mitochondrial cytochrome enzymes provides sites for H₂S to combine with iron, thus blocking cellular respiration⁴⁸. It has also been suggested that the poisonous agent of hydrogen sulfide was responsible for ulcerative colitis, Crohn's disease and similar intestinal diseases⁴⁸. Hydrogen sulfide has a significant impact not only in the human health, but also in several industrial and manufacturing processes, such as biogas processing. When found in sewage, this sulphide in several sewage treatment plants and industries can pose deleterious problems. Due to the increased demands for nutrients, resources and increased costs of processing, the chemical H₂S extraction procedures is an expensive process⁵¹. For all of these purposes, bacterial treatment strategies regarding eliminating hydrogen sulfide are ideal and is best suitable than chemical approach and it would seem acceptable to add purple and green sulfur bacteria for any economic waste water treatment process.

1.5 Hydrogen Sulphide Gas removal by PSB

PSB is now the predominant and possibly the most suitable community of microorganisms effective in oxidizing H₂S to form sulphur for use in the extraction and removal of hydrogen sulphide.^{52,53} This includes a variety of microorganisms categorized into several taxonomic groups

that have the ability and potential to conduct processes based on the intensity of light. Four subgroups are usually differentiated: yellow sulfur bacteria (YSB), purple sulfur bacteria (PSB), purple non-sulfur bacteria (PNSB), and the non-sulfur green (NSGB) bacteria group Chloroflexus. The first two categories are perhaps the most suitable for use in the hydrogen sulphide oxidation based on their ability to accumulate elemental sulphur and leading to H_2S removal. Identified Sulphur oxidizing bacteria (SOB) are predominantly phototrophic and chemotrophic bacteria. Phototrophic microbes (colored green or purple) use light to convert carbon dioxide (CO_2) to carbohydrates as a source of energy. Thus reduced sulfur compounds through this are being used as contributors of electrons. This reduction occurs in environments that are anaerobic. *C. Phototrophic. Limicola* is a desirable bacterium in such biological extraction approaches owing to its capacity to grow mainly in inorganic substrates with a source of light under anaerobic conditions and its effective extracellular development of sulphur and this contributes to the removal of H_2S . Because of its reduction and oxidation potential effects, sulfur is important for phototrophic species. Reduced sulfur is an electron contributor in photosynthetic carbon dioxide generation. Usually in polluted and contaminated areas, green and purple sulfur photosynthesizing bacteria are crucial groups in sulphur production. Two families, Chlorobiaceae (green sulfur bacteria) and Chromatiaceae (purple sulfur bacteria) are the major members of all these microorganisms.⁵⁵ The microbes with green sulfur (GSB) are phototrophic anoxygenic bacteria. Their illumination harvesting structures contain family-specific bacteriochlorophyll c, d, and e and thus are found in special organelles (chlorosomes) that harvest light. The sulfide extraction process depends on the class of colorless sulfur bacteria aerobic oxidation⁵⁴. The biological aerobic breakdown of reduced sulfur substance, i.e. uncontrolled reaction of H_2S and sulfur with oxygen on the water surface, produces energy from the chemotrophic sulfur bacteria (colorless bacteria). SOB can also be categorized as lithotrophic microbes, which uses inorganic substances for supply of hydrogen and organotrophic microbes, and it uses organic substances for supply of hydrogen.

1.6 Sulphur oxidation enzymes of PSB

Several previous reports support oxidation of bacterial sulfide. The main enzyme accountable for oxidation of sulphide molecules is sulphite oxidase⁵⁶. There are two similar subunits of sulfite oxidase. It has three antiparallel adjacent beta sheets with five helix of alpha and the N-terminal domain has a heme cofactor. A molybdopterine cofactor enclosed by thirteen beta sheets and three alpha helices is located in the C-terminal region. The cofactor of molybdopterine does have a Mo (VI) base that is bound to cysteine sulfur, pyranopterine dithiolate and two terminal oxygens.¹²³ *Arthrobacter* sp. and *Bacillus* sp. BN53-I have defined sulphide oxidase enzyme that mediates the sulphide oxidation.^{57,56} The pure sulphide oxidase is a monomer having a molecular weight of 43 kDa, as per the Mohapatra et al. (2006) and Nakada et al (1999) study^{56,57}. On Contrary to the molecular weight of *Bacillus* sp, pure enzyme sulfite oxidase molecular weight was determined to be lower which is about 37 kDa⁵⁷. The *Arthrobacter* sp. was used to isolate sulphide oxidase and is cell-bound and it had substantial pH behaviors that are highly useful for the handling of sewage⁵⁶.

1.7 Gene for sulphur oxidizing in PSB

The gene for the ability for sulfur-oxidation are known as the Sox gene (Sox), which was first identified from *Paracoccus pantotrophus* alphaproteo bacterium, which is an additional chemolithoautotroph and it grows along with thiosulphate⁵⁸. The Friedrich et al.⁵⁸ found sox gene cluster encoding Pcs multienzyme sox group. *Pantotrophus* consists of minimum two 15 gene (soxRSVWXYZABCDEFGHIJ) transcription units. SoxR encrypts an ArsR family SoxR repressor protein that attaches to the gene's soxS-V and soxW-X intergenic areas¹²⁴. This SoxS is a thioredoxin periplasm and necessary for the full expression of the group of sox genes⁵⁹. SoxV encrypts the lipid membrane and soxW encodes the thioredoxin plasm. Both of these are important for thiosulphate chemotrophic production⁶⁰.

1.8 Factors affecting PSB in sulphur oxidising

1.8.1 Effect of Oxygen Rate

The sulfide is naturally metabolized into elemental sulfur^{61,62}. Kuenen suggested that the amount of generated sulfur increases chances of even more sulphate oxidation or similar polymer formation. Thus, the produced sulfur must be eliminated within the system simultaneously in time. Earlier research from⁶³ Janssen et al. showed that the proportion of molar oxygen to sulfur throughout the development of sulphur was between 0.6 to 1.0. Moreover, it would be hard to regulate a narrow sulfide and oxygen balance under real situations as in a real recycling facility. And from the other side, preserving optimal redox potential can efficiently control sulfide accumulation.^{64,65}

1.8.2 Effect of pH

Sulfur compounds, when biologically metabolised in gas phase in the digesters, sulfur molecules generate sulfate that leads to a considerable reduction in pH along corresponding decrease in organic compounds and thus in removal of H_2S . The pH multitude of different species is 0.5 to 10⁶⁶. Much of the reported earlier microbial sulfide oxidation (BSO) systems exist over a range of 7–8, around neutral pH.^{63,67-73} Janssen et al. reported that in the growth system established at pH 8, reached optimum sulfur recovery.⁶⁴ Underneath a broad range of heterotrophic environments in a biomass biofilter.^{74,75} Yang and Allen reported removal efficiency improvements of above 99 fold. The substantial decrease in pH, nevertheless, led to a decline in extraction efficiency. Cox and Deshusses observed that when fed H_2S and toluene concurrently operating a bioreactor at both pH 4.5 or 7.0 did not directly affect the quality of the biotrickling device.⁷⁶ Researchers discovered however, that the start-up period of toluene depletion at pH 4.5 is fairly long, so a sudden drop in pH can create non permanent weak separation of H_2S and toluene. H_2S extraction output improved as the nutrient feed pH rose in the 2.0 - 6.0 pH range. Nevertheless, there is an inverse pattern between 6.0 and 7.0 for the pH. For the autotrophic group, the optimal pH is about 6.0.⁷⁷ Krishna Kumar et al. recorded that sulphide extraction is almost 100 percent up on 19 kg / m³d charging, where sulphide restoration is around 80 percent and sulphate is about 2 - 3 percent of total sulphide when its pH is regulated at 8 in Reverse Fluidized Loop Reactor (RFLR). For chemotrophic SOBs including the RFLR *Thiobacillus denitrificans*, the optimal pH for production and sulfide accumulation is around

7–8^{63,72,73,77,78}. The biomass encountered higher toxicity at pH 9 and 9.5 relative to pH 8 under the same sulphide load. The existence of lower remaining sulphide and intermediate products like $S_2O_3^{2-}$ and polysulphide is a clear demonstration of such a decline in SOB behavior in alkaline pH in RFLR.⁷⁷ Nevertheless, the use of alkaliphilic SOB, as reported⁷⁹ in potential bioreactor analyses can help to eliminate sulphide in higher harsh conditions.

1.8.3 Effect of Temperature

SOB is used in temperature conditions up to Hundred degrees⁸¹. Raising the heat of biooxidation to 50 °C raised the oxidation of sulphides to around 48%. Around 60 °C, nearly 51 percent of sulfide degradation is at peak⁸².

1.9 Selective growth promoters of PSB

To their phototrophic nourishments, purple bacteria needs different minerals and critical environments for growth like illumination, anoxic environments, sulfide saturation, suitable pH and warmth. For moderate and growing conditions, many adaptations have been made over long to enhance particular groups of PSB and PNSB. Biological distinction between PSB and PNSB allows PSB exclusively enriched as electron contributors in environments enriched by reduced elemental sulfur. Purple sulfur bacteria need reduced sulfur (H_2S) as an electron contributor while [apart from a few purple non-sulfur bacteria such as Blc] were not needed as purple non-sulfur bacteria needs. Sulfidophilum needs reduced sulfur in its development like an electron contributor^{83,84} but accepts lower concentrations. Light presence and performance also affects the phototrophic bacteria's selective enrichment as green light promotes the growth of PSB⁸⁵. Winogradsky et al made an initial research on PSB⁸⁶, and successfully confirmed raw enrichment cultures to cultivate these microbes in the lab. In ideal environments under anoxic conditions and in the sun, phototrophic sulfur bacteria can be particularly supplemented from most natural environments. It is advisable to change pH between 7.2 and 7.4 for its optimum growth. Several species exist very precisely in typical habitats for the productive enhancement of purple sulfur bacteria. Moreover, the inoculum organism distribution is of paramount requisite for all the consequence of enrichment treatments⁸⁷. Higher concentrations of sulfide kill a variety of species. The levels of sulfide must therefore be held as minimal as 1–2 mM, such as susceptible substances are not removed from production. Higher populated concentrations can exclusively be accomplished by continuous sulfide, i.e. by applying a neutralized formulation of sulfide. Growth of *Thiopedia rosea* is prevented by the presence of sulfide concentration greater than 0.6 mM and supplementing it with sodium dithionite (50 mg per 1 liter) can enhance the growth of this bacterium⁸⁸. While analyzing specimens across aquatic, hypersaline habitat or environment, the content of salt and minerals certainly is influential and significantly critical for its enhanced growth. The enriched culture's osmotic pressure is normally balanced as per the inoculum's salinity. It is necessary for marine microbes to increase the amount of sodium chloride (NaCl) to 2–3 percent. Many microbes in addition, need elevated magnesium amount (e.g., 0.3%) and quite often appreciable calcium concentrations⁸⁹. By general, for a given environment isolate, the salinity of a natural environment cannot always be optimum. Moreover, to separate a number of species from that same specimen, it needs enriched medium with various salinities and this

promotes beneficial blooming. Many PSB species vary in terms of their competitive benefit during various grades or intensity of lighting. Such variations could be used to selectively enrich other species populations^{90,91}. Here have been proposed two better lighting systems with incandescent source of light. When constant lighting with a higher intensity of light ranging 1,000–2,000 lx and an incubation around 30 °C, it was shown that few and rapidly-growing Chromatiaceae can dominate, for example, *Thiocapsa Roseopersicina*, *Allochrochromatium Vinosum*, *Allochrochromatium Minutissimum*, *Thiocystis minor*, *Thiocystis violascens*, *Thiocystis violacea* and *Mary Chromatium Gracile*⁹².

1.10 Growth and propagation of PSB

Light accessibility, brightness, and amounts of sulfide and oxygen are the most critical and influencing factors which decide the production and spread of PSB. PSB are common in existence and are located often in stagnant or standing water environments of all kinds and survive in sulfidic marine ecosystems open to light, higher concentrations (blooms) of purple sulfur bacteria and this differentiates from PNSB. Nevertheless, combined populations of purple and green sulfur bacteria (PSB) are commonly found, and purple nonsulfur bacteria (PNSB) are routinely associated with the large production of PSB. Sulfide, formed in the sedimental region of active stratified aquatic bodies by sulfate reducing microbes, spreads and rises above the soil. This formed sulfide allows PSB growth^{33,34}. The lake or the stagnant or sewage water will indeed turn the bacteria to have purple, or reddish brown pigment when colony numbers are fairly high^{34,37,5,6,7}. If these events happens, it can certainly be easy to determine by clear lab analysis the prevalent type of PSB present. The outgrowth or blooming of PSB comprises of a combination of species in several stratified lakes⁹³, whereas others the population include only one species^{5,6,7}.

1.11 PSB in Wastewater stabilization ponds (WSPs)

Stabilization lakes and ponds for waste water (WSPs) are really a highly efficient, natural element of sewage treatment recycling process. To a very great extent of cleansing, they balance flexibility, sturdiness and lower cost. Curtis et al studied on a shallow water natural defense ability to obtain insitu purification, through sunlight absorption which is a crucial factor⁹⁴. WSPs are generally formed as one or more sequence of anoxic, optional or maturing ponds, on the first two becoming primarily capable of removing suspended, insoluble, bioorganic matter (biological requirement for oxygen) but the last for eliminating contaminants and minerals. Its low operational and running expenses rendered to have a viable option for the processing of waste water, for developing and developed countries, as it needs for specialized and exclusive knowledge and facility to run such solutions. WSP facilities are commonly employed in Mediterranean countries⁹⁵, with over 25% of sewage treatment plants in Tunisia⁹⁶. Although these methods are quite productive and cost efficient in treating sewage, nevertheless, but they do has serious problems like that of the case of 'red-water.' The occurrence of 'red-water' is a temporary phase in water colour arising from those in the explosive growth ('blooming') of phototrophic purple anoxygenic bacteria. This microbial process was found in multiple treatment ponds of sewage⁹⁷. The process is triggered by overloading the WSP process with biomass that induces the removal of sulphate in anoxic or optional ponds.

As a result, the growing accumulation of sulfide promotes a harmful environment to the organisms, along with the proliferation of phototrophic PSB which survive in anoxic environments⁹⁸. The process of red coloured water (RVW) allows the consistency of the effluent to deteriorate, with a persistent hydrogen sulphide odour, and higher levels of suspended solids⁹⁹. anoxygenic phototrophic bacteria (APB) are liable for oxidizing sulfide from main hydrological process in aquatic ecosystems¹⁰⁰ because many recognized APB microbes, like purple sulfur, purple non-sulfur, green sulfur and green non-sulfur bacteria, are capable of using reduced hydrogen sulfide as anoxygenic photosynthesis electron contributors^{125,126}. A rather crucial part of the sulfur cycle in existence is the same for most PSB, particularly the phototrophic sulfur bacteria which can use toxic H₂S from substrate. The potential of the Chlorobiaceae and Chromatiaceae families can be employed to reduce the H₂S level of sewage and cleansing of biogas, but additional work will still be required.

1.12 PSB in waste water treatment Plant

Measures to add green and purple sulfur bacteria in the extraction of H₂S is warranted in all applications which generates polluted sulphide, hydrogen sulphide and thiosulphate waste production. This wastewater is created by aerobic processes throughout gas desulfurization, crude oil facilities, chemical manufacturing plants, and often at high-sulfur water treatment plants. Remedies suggested so far are focused on the use of photosynthesizing colonies of bacteria in bioreactors involving light source (photoreactors), where optimal environments for their growth are assured. The first and most critical need is to provide a light source, which involved with some expenses, but due to the possibility of cost free sunlight, the price is significantly smaller than a chemically modified strategies. Lee & Kim suggested the use of the optical fiber bioreactor in which optical fibers bring light. Currently we could see a clear requisite that the conservation of the natural habitat is a central and necessary activity. Considering the large amount of wastewater generated by manufacturing industrial facilities through anthropogenic practices, it is necessary to find ways to reduce the impact of poisonous or hazardous waste around the world. Reasonable and competent solution to problems related to the handling of different kinds of organic waste is one of the key methods for addressing the water pollution crisis. In almost all aquatic environments, phototrophic purple and green bacteria are present. Upon forming water blooms, purple or green sulfur bacteria can easily be identified. Nevertheless, in detectable amounts, purple non sulfur bacteria occasionally appear¹⁰². Thus, the spread in nature could only be tested using enrichment strategies¹⁰³ or the membranes filtering process^{104,105}. The existence of purple bacteria (PSB) depends in general on the extent in which organic matter contaminates water. Its growth and blooming leads towards cleansing of sunlight-exposed heavily contaminated water, such as in wastewater lagoons^{106,107}. Throughout Japan, the primary cleansing phase for organic wastewater or sewage water treatment^{108,109,110} employs phototrophic bacteria.

1.13 PSB in Meromictic lakes

Meromictic lakes are those that are naturally stratified by a layering gradient of freshwater proportions. The chemocline separates the heavily salinized base layer from those in the

upper layer of fresh water, in which the salinity dramatically differs. The top and bottom layers would not combine due to the large disparity in gradient, generating an anoxic habitat below the chemocline.¹¹¹ This light anoxic surrounding with ample sulfide supply is suitable for PSB^{112,111}. Mahoney Lake study showed that PSB contribute for the recovery of phosphorus, an inorganic nutrient.¹¹¹ The outflow of PSB into the upper layer of water provides an origin of bounded phosphorus, and phosphatase action releasing phosphorus to the water. This soluble phosphorus should then eventually be integrated to be used in developmental systems into heterotrophic bacteria. Thus PSB therefore add to the phosphorus cycle and reduce the depletion of nutrients¹¹¹

1.14 PSB in lagoons

PSB plays a vital role in reducing the release of damaging organic compounds and odors in the lagoons where they would likely to spread. Within sewage lagoons, hazardous chemicals such as methane, a greenhouse gas, and pungent poisonous H₂S can be generated. PSB can help to reduce the amount of both and others harmful gases.¹¹³ Destructive and toxic organic compounds could be eliminated by photoassimilation of carbon via photosynthesis¹¹⁴. As PSB conducts photosynthesis in the lagoons, they use carbon from toxic compounds like methane¹¹⁵ for their supply of energy. It extracts methane from those in the lagoon, a greenhouse gas, which decreases the chances of atmospheric contamination on the lagoons. Throughout these photosynthetic mechanisms which eliminates organic substances, H₂S can serve as a supply of sulfur for PSB. Utilizing this reducing agent H₂S used by PSB eliminates H₂S from the lagoon and contributes to a decrease in lagoon bad smell and toxicity^{116,117,118}.

1.15 PSB as biomarkers

PSB produce conjugated light sensitizing pigments that operate in the light processing system termed carotenoids. Many color pigment molecules are retained in the sediments in slightly different form once these species die or fall. One such formed carotenoid biomolecule, okenone, is a modified diagenetically to the okenane screening tool. This detection of okenane in sediment samples suggests the existence during the deposition period of PSB. To date, okenane has also been found only in one Northern Australian sedimentary out crop dating back to 1640 million years ago¹¹⁹. Authors of this research suggested that the Paleoproterozoic environment had to be anoxic and sulfidic at deepest, and this suggestion was based on the discovery of the biomarker of purple sulfur bacteria. This observation presents support to the theory of the Canfield Ocean hypothesis.¹¹⁹

1.16 PSB for environment

Through adding to nutrient cycling, and by having their mechanisms to change their environments, purple sulfur bacteria can influence their environment. These could play an important role in resource extraction, indicating that these species influence the carbon cycle by carbon fixation.¹²⁰ Throughout their environment, PSB often lead to the phosphorus cycle.¹²¹ From the upwelling of these species, phosphorus, a restricting resource in the anoxic layer of ponds, is reused and distributed to heterotrophic bacteria for use.¹²¹ This suggests that although purple sulfur bacteria were contained in the anoxic layer of their environment, they

can encourage the growth of several heterotrophic species by providing the oxic layer with inorganic nutrients. Other method of regeneration by PSB of inorganic materials and soluble organic matter would be via the food supply chain since it serves as a food source for many other organisms¹²¹. Several PSB indeed adapted to maximize their natural growth condition. For instance, in the South Andros Black Hole that exist in Bahamas, PSB has implemented a new trait which can utilize their biopathways and metabolism to transfer thermal energy to its environment.¹²² Because of the lack of efficiency of their carotenoids or light absorbing pigments, species are able to retrieve these energy through heat energy¹²². Through increasing the warming heat of the surrounding aquatic body, these establish a habitat that encourages its own growth and helps themselves to achieve results.

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2. CONCLUSION

Thus the purple sulfur bacteria plays vital role in environment protection with cost effective and in particular provides a great help in waste water treatment. This PSB now started becoming a favourable choice for those of industrial and in particular biotech industrial effluents plants and it is hoped to spread to other industries for maximising their potential.

3. AUTHORS CONTRIBUTION STATEMENT

All authors contributed equally to literature review, collection of datas , drafting of manuscript.

4. CONFLICT OF INTEREST

Conflict of interest declared none.

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