2 Literature on the treatment methodologies for removal of dyes from the wastewater

The textile industry is one of the leading industries in releasing polluted effluents, including dyes, salts, and other organic complexes, in the soil and water. It hence poses severe risks to the aquatic lives and environment. As per the world dyes consumption report 2017 (Source: IHS Markit), the Asian countries are using ~75% of the total produced synthetic dyes, with China and India as the top two consumers. It is estimated that more than 10,000 kinds of dyes and pigments are being devoured in the environment with their yearly production of more than 7×10^5 tons (da Silva Leite et al. 2016). The existence of dyes, even at low concentrations, diminishes the level of water transparency and, therefore, decreases the process of photosynthesis and triggers anaerobic conditions eventually in the water bodies (Tan et al. 2016; Robinson et al. 2001; Pirkarami and Olya 2017; El Bouraie and El Din 2016). The presence of lethal and non-biodegradable segments, such as complex aromatic and polymeric structures in the textile dyes, can be exceedingly poisonous with mutagenic and carcinogenic hazards to the aquatic and terrestrial biota (Ito et al. 2016). It necessitates the expulsion of dyes from the textile effluents for aesthetic reasons and the conservation of lives and ecological parameters (van der Zee and Villaverde 2005b).

Based on their fundamental working principles, the three steps of treatment processes are physical, chemical, and biological methods. However, the wide variety of robust and stable industrial dyes and pigments are the primary concern source since they are the principal polluting agent in textile effluent. These organic contaminants mainly comprise heavy metal ions, salts, stabilizing agents, and surfactants, which enable them to cling to textile materials for extended periods, despite natural processes, water wash, and laundry operations. Most modern-day dyes are too powerful to be destroyed by aerobic and anaerobic treatment.

2.1 Physical Methods

The conventional physical processes such as adsorption, membrane separation, coagulation/ flocculation, filtration, etcetera have been successfully used to treat the wastewater. Sedimentation/floatation is a typical sub-physical process separating dye-containing components from a mixture. Due to gravity, the consistent segregation of these suspended solids/wastes occurs in the water treatment process. Strong turbulence of solvent initially amuses these dye-related particles, then permitted to stand still for some time until all of the sediments are appropriately settled against a barrier. This may be explained as their movement in the fluid in reaction to external forces like centripetal force or gravitational strength (J. Classen et al. 2013). Segregation of dissolved dyes and their metallic ions does not occur in this system, which is a drawback. Dehghani et al. explain that four different coagulants, including alum, iron sulfate, polyaluminum chloride, and granular ferric hydroxide, can decolorize the dangerous colors methyl orange and methylene blue into minute sediments that can increase flotation (Dehghani, Karimi, and Rajaei 2016).

Coagulation occurs when the zeta potential on the surface area of/among the particles fluctuates, resulting in flocculated agglomerates or tiny molds. When wastewater is discharged into rivers, it includes a high level of contaminants and pollutants, which must be removed using coagulants (Dotto et al. 2019). To keep the system pure, iron coagulants such as ferrous sulfate, ferric chloride sulfate, and ferric chloride are introduced. Other chemical coagulants, such as hydrated lime and magnesium carbonate, aid in the adsorption of dyes and their by-products (Islam and Mostafa 2020). When a coagulant is introduced to a solution and forcefully agitated, the coagulant precipitates, trapping the organic contaminants and impurities. Polyaluminium ferric chloride (PAFCl), Polyaluminium chloride (PACl), Polyferric chloride (PFCl), and Polyferrous sulfate (PFS) are a few chemicals based coagulants, some plant-based coagulants like starch, gum, moringa seeds, etc., a few animals based coagulants like chitosan (a single polymeric chain of D-glucosamine and N-acetyl-D-

glucosamine built from chitin) have been used by the community of researchers. For the adsorption process, some physio-chemical variables such as the nature of the adsorbate (dye) and the adsorbent, surface area, particle size, porosity, pH, temperature and pressure, contact duration, etc. must be taken into consideration (Nigam et al. 2000; Sham and Notley 2018). Even though this approach achieves a more significant percentage of color removal and COD degradation than sedimentation, it has its limitations. Firstly, the problem comes in the recovery of the synthetic chemicals used in the preparation of coagulants and reusing them, which is not recommended in any case. Second, the safe disposal of colored coagulants, which are finally thrown away, is quite an issue. Therefore, these physical methods do not completely degrade the dyes, and hence the environmental concerns persist due to the generation of secondary pollutants in the form of sludge (Bilińska, Gmurek, and Ledakowicz 2016; Ahmad et al. 2015).

2.2   Biological Methods

Bacterial treatments (known as biodegradation) have decolorized the dyeing wastewater (Nawaz and Ahsan 2014). Although biodegradation is a very economical process for degrading dyes, it is an unrealistically slow process. On the other hand, the chemical processes (advanced oxidation processes, AOPs) for the remediation of the dyeing wastewater are fast and easy to use. Still, these methods are not cost-effective, particularly at large-scale processing. Further, the chemical processes may not be environmentally friendly due to the generations of hazardous byproducts. Lately, the integration of AOPs with biodegradation (sometimes referred to as AOP-biodegradation hybrid in the literature) (Tee et al. 2016; Kalyuzhnyi and Sklyar 2000) has been explored by different research groups for developing a methodology optimized in terms of dye removal efficiency, processing time, cost, toxicity, and environmental impact of the byproducts (Hai, Yamamoto, and Fukushi 2007).

Biodegradation utilizes microbial metabolism to break down the contaminants present in the wastewater. Biodegradation is one of the most widespread techniques used to decompose textile effluents. In this process, the microorganisms with diverse strains utilize enzymes to degrade the dye-waste either in off-site (ex-situ) or in onsite (in-situ) mode (Rai et al. 2005). The efficiency of biodegradation depends on the genotype and population density of the microbes, the accessibility of contaminants to the active sites of the enzymes, and the environmental conditions such as pH, temperature, nutrients, as shown in Figure 2.1.
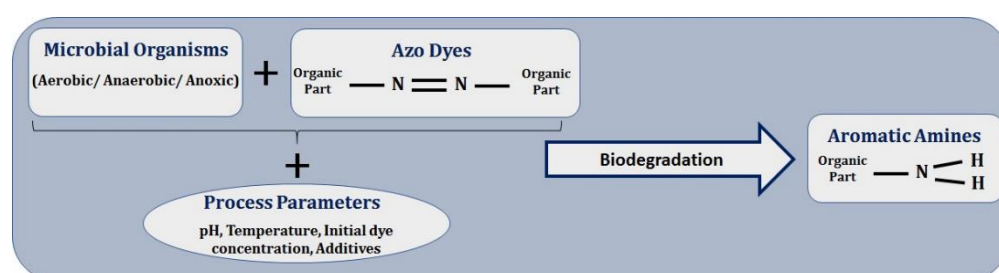

Figure 2.1 Schematic of biodegradation of an azo dye (Chaturvedi et al. 2021b)

Generally, a biological treatment involves bioaccumulation, biosorption, and biodegradation. Bioaccumulation is the accumulation of contaminants in the living cells of the microbes, while biosorption refers to the adsorption of the contaminants onto the live or dead microbial biomass. Although bioaccumulation and biosorption assist in waste treatment, they can impair the growth of microbes and pose an environmental concern during the disposal of the contaminated biomass. Biodegradation is a desired step where the organic matters (such as azo dyes) are decomposed into other by-products utilizing the enzymatic response of different micro-organisms (Wolicka et al. 2009; Sharma, Dangi, and Shukla 2018). The scale-up and process optimization of such biodegradation processes require detailed knowledge of the bacterial growth kinetics to achieve an economical treatment of the dyeing wastewater. Several research works on the modeling of bacterial growth kinetics in the biodegradation of dyeing wastewater have been published in the literature in the last few decades (Baranyi et al. 1993; Lineweaver and Burk 1934; Monod 1949; Sponza and Işık 2004). However, in most

studies, the bacterial growth rates in the log phase were estimated using the exponential growth kinetics applicable only in the accelerating log phase. Few studies barely mentioned the successive decelerating log phase occurring due to inhibition caused by toxic metabolites/by-products (Talaiekhozani et al. 2015; Waldrop 2009a). Understanding and incorporating kinetics of the decelerating log phase is essential to predict a precise overall growth rate of the bacterial mass in the entire log phase.

The biodegradation can be classified based on the nature of microorganisms as aerobic, anaerobic, and anoxic (composite of aerobic and anaerobic) (Geed, Prasad, et al. 2018). Most bacteria cannot utilize dyes as a carbon source in an aerobic condition and require an additional carbon source (co-substrate). Some aerobic bacteria possess an oxidoreductase enzyme which can degrade the azo dyes (Sarayu and Sandhya 2010). The biodegradation of azo dyes occurs preferentially under an anaerobic condition wherein the azo linkages are reduced into colorless aromatic amines. The breaking-down of chromophore azo bonds is facilitated by azoreductase enzyme in which nicotinamide adenine dinucleotide (NADH) and flavin adenine dinucleotide (FADH) act as reducing agents (Jamee and Siddique 2019). The generated aromatic amines are carcinogenic and generally do not show degradation in an anaerobic environment, but they can be degraded into more specific products by aerobic treatments (García-Martínez et al. 2015). Anoxic conditions utilize less oxygen (less than 1 mg/L), and the operating conditions are similar to aerobic treatment (Jamee and Siddique 2019). Generally, a combination of anaerobic and aerobic process conditions is preferred to decolorize azo dye.

The advantages of biodegradation include high selectivity, cost-effectiveness, and good removal efficiency (Bhatia et al. 2017; Sharma, Dangi, and Shukla 2018). Biodegradation can be further enhanced either by adding nutrients to stimulate the growth of native microbes (bio-stimulation) or by adding exogenous /genetically-modified microbial strains to accelerate the process of microbial metabolism (bioaugmentation). The

biodegradation process requires identifying and growing the effective microbial strains and optimizing the process conditions to yield high degradation efficiency (Roy, Verma, et al. 2018). The combination of different microbes (such as bacteria and fungi) has also been utilized to enhance further the degradation of azo dyes (Moutaouakkil et al. 2003; Sen et al. 2016). Although biodegradation of the textile dyes is an economical and eco-friendly process, it is limited by the following (Chaturvedi et al. 2021b):
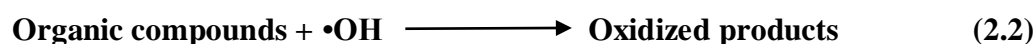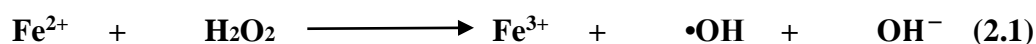
a. Biodegradation at a large scale is a prolonged process due to its dependence on the metabolic activity of microbes.

b. Contaminants must be biodegradable (BOD_5/ COD ≥ 0.2).

c. Biodegradation is highly specific, and therefore complete mineralization and discoloration of a mixture of various dyes and their recalcitrant metabolites would be challenging to achieve (Vikrant et al. 2018).

2.3   Chemical Methods

The AOPs are chemical treatments intended to degrade complex organic compounds that are difficult to remove by the conventional physical-biological routes. Highly oxidizing species drive the degradation of organic substances in radicals, molecules, or ions (Ghatak 2014; Oturan and Aaron 2014). Most AOPs use the oxidative power of hydroxyl radicals to transform the complex organic compounds into simple molecules. One of their fundamental advantages, contrasted with the conventional technologies, is that they quickly degrade the recalcitrant components without creating a secondary waste stream like other physical-biological processes (Kordkandi, Ashiri, and Policy 2015; Ribeiro et al. 2015; Moreira et al. 2017; Matafonova and Batoev 2018; Cheng et al. 2016). The AOPs can be classified into two types based on the oxidation pathways: free-radical oxidation and molecular oxidation. The AOPs based on Fenton, photo-catalysis, and ozonation are prevalent in literature.

2.3.1 Fenton process

The Fenton reaction is metal-catalyzed oxidation in the presence of hydrogen peroxide. The ferrous ion catalyzes the production of hydroxyl radicals (•OH) from hydrogen peroxide (H_2O_2) via electron transfer according to the following reactions

$$Fe^{2+} + H_2O_2 \longrightarrow Fe^{3+} + •OH + OH^- \quad (2.1)$$

$$\text{Organic compounds} + •OH \longrightarrow \text{Oxidized products} \quad (2.2)$$

As the standard redox potential of the hydroxyl radical ($E^0 = 2.80$ V) is significantly higher than that of the hydrogen peroxide ($E^0 = 1.77$ V) (Tarr 2003), the hydroxyl radical is a much stronger oxidizing agent than the hydrogen peroxide. The hydroxyl radicals are liable for the oxidative degradation of various organic compounds, including textile dye, phenols, formaldehyde, aromatic amines, surfactants, and pesticides. One of the significant drawbacks of the Fenton process is that the oxidation reaction, as shown in Eq 2.1, is impeded after the complete consumption of the ferrous ions, resulting in large deposits of the ferric hydroxide sludge. During the Fenton oxidation, the precipitation of ferric ions can be avoided by carrying out the reaction in an acidic environment (Kordkandi, Ashiri, and Policy 2015). However, the Fenton process requires the constant addition of ferrous ions to ensure continuous oxidation of the organic compounds.

The photo-Fenton process regenerates ferrous ions via a photochemical route, to reduce the generation of secondary pollutants, one of the significant disadvantages of the conventional Fenton process (Ameta et al. 2018). The photo-Fenton oxidation utilizes UV irradiation in addition to the Fenton's reagent (hydrogen peroxide and ferrous ion). UV irradiation facilitates the photo-reduction of Fe^{3+} to Fe^{2+} ions and enhances the production of hydroxyl radicals as per equations 2.3 and 2.4.

$$Fe^{3+} + H_2O + h\nu \longrightarrow Fe^{2+} + •OH + H^+ \quad (2.3)$$

$$H_2O_2 + h\nu \longrightarrow •OH + •OH \quad (2.4)$$
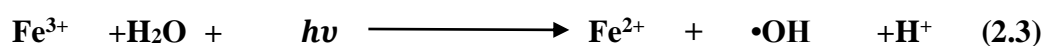
Numerous studies are available in literature focusing on optimizing various Fenton's process parameters such as temperature, pH, and concentration of H_2O_2, Fe^{2+}, and dye to degrade azo dyes (Ghanbari and Moradi 2015). Ertugay and coworkers (2017) studied the degradation of Direct Blue 71 (DB 71) dye in the textile wastewater using the Fenton process and reported the optimum values of the process parameters as Fe^{2+}=3 mg/L, H_2O_2=125 mg/L, and pH=3.0 (Ertugay and Acar 2017).

Although the homogeneous Fenton process is a promising and economical solution for the degradation of dyes, it has a lot of disadvantages. The homogeneous catalysts, typically iron salts, used in the Fenton reaction are difficult to be retained in the reactor during the degradation process (Pliego et al. 2015). Since the Fe^{++} concentration of 50-80 ppm or even higher is used in the Fenton process, releasing iron ions with the treated dyeing wastewater into the environment can generate secondary pollutants (Hartmann, Kullmann, and Keller 2010). Further, the release of iron ions with the effluent causes a reduction in the iron concentration in the reactor, demanding a regular addition of iron salt in the reactor to ensure a continuous reaction. Various research groups have utilized iron minerals (ferrite, magnetite, hematite, goethite, pyrite) and iron-containing industrial wastes (such as fly ash and pyrite ash) to overcome this problem. Iron/ iron oxide loaded support materials (such as activated carbon, silica, clay, zeolite, alumina, biosorbents, hydrogels) as catalysts in the 'heterogeneous' Fenton reactions (Nidheesh 2015). The leaching of iron ions to the treated effluent is contained, and the catalyst is recovered and reused after the reaction (Rossi et al. 2012). A similar study reported by Sreeja et al. (Sreeja and Sosamony 2016) found that the heterogeneous photo-Fenton process is more efficient than the homogeneous one for the COD and color removal. The removal efficiencies of the COD and color were found to be 47% and 82%, respectively, in the case of the homogeneous Fenton process; however, the same was obtained as 62% and 85% respectively for the heterogeneous Fenton process. Further, it was also observed that the sludge formation was significantly lower in the heterogeneous photo-

Fenton process than the homogeneous Fenton method because of the recovery of most of the heterogeneous catalysts from the effluent.

Several modifications of the Fenton process are available in the literature by associating the Fenton reagent with another external energy source, such as ultrasonication, to improve its dye degradation efficiency (Babaei et al. 2017). The utilization of sonication or UV + sonication with the Fenton process is called *Sono-Fenton*, SF, or *Sono-Photo-Fenton*, SPF, respectively. In ultra-sonication, the existence of ultrasound (US) irradiation enhances the generation of hydroxyl radicals in the solution through *acoustic cavitation*. In the process of *acoustic cavitation,* the ultrasound energy is concentrated in the form of micro-bubbles to generate local hot spots with very high temperatures (>5000K) and pressure (>500 atm) (Dükkancı, Vinatoru, and Mason 2014). These hot spots facilitate thermal dissociation of water molecules to generate hydroxyl (•OH), oxygen (O•), and hydroperoxyl (HOO•) radicals to oxidize organic pollutants (Ma, Xu, and Liu 2006; Wang et al. 2008).

2.3.2 *Photo-catalysis process*

Photocatalytic oxidation is a heterogeneous AOP that utilizes metal oxide semiconductors (TiO_2, ZnO, CdS, ZnS, Fe_2O_3) to oxidize organic pollutants present in the dying wastewater (Fujishima, Rao, and Tryk 2000). Titanium dioxide (TiO_2) has become one of the most widely used photocatalytic materials because of its excessive availability, low cost, and biological and chemical stability over a wide range of pHs (Singaravadivel, Vanitha, and Balasubramanian 2012; Abdellah et al. 2018). TiO_2 semiconductor exists in three crystalline structures: anatase, rutile, and brookite. With the band-gap of 3.2eV, the anatase form absorbs photons in the UV range and is the most influential phase of TiO_2 for photocatalytic activity. The lifetime of the generated electron-hole pairs in the anatase phase is longer than the other phases. Therefore, the anatase TiO_2 becomes an effective photocatalyst for the oxidation of organic pollutants (Luttrell et al. 2014).

Photocatalytic oxidation involves the generation of electrons and holes in the conduction and valance bands, respectively, of TiO_2 particles after the absorption of photons with wavelengths below 390 nm. The produced electrons and holes cause reduction and oxidation reactions, respectively, on the surface of the TiO_2. Specifically, holes in the valance band react with the water molecules adsorbed on the surface of TiO_2 to generate hydroxyl radicals (•OH), and the oxygen molecules take up electrons in the conduction band to produce superoxide anion radicals ($•O_2^-$). The superoxide anion radicals get further protonated and form hydrogen peroxide, which finally dissociates into •OH (Ajmal et al. 2014). Because of their high oxidizing potential of 2.8V, the hydroxyl radicals oxidize the organic dye molecules and eventually convert them into CO_2 and H_2O.

Several studies reported in the literature on the photocatalytic degradation of azo dyes (Fujishima, Rao, and Tryk 2000; Velegraki et al. 2006). Pekakis et al. have investigated the oxidative discoloration of real dyehouse wastewater using photocatalytic degradation in the presence of TiO_2 (Pekakis, Xekoukoulotakis, and Mantzavinos 2006). They studied the effect of catalyst loading, pH of the medium, air-sparging, and addition of hydrogen peroxide on the degradation efficiency of different crystalline forms of TiO_2. The degradation of dyes was found to increase when the concentration of the TiO_2 catalyst was raised; however, this effect diminished beyond a certain level of TiO_2 concentration. The acidic pH conditions enhanced the degradation of dye molecules because the TiO_2 surface became positively charged at pH below 6.5 and, therefore, assisted in attracting the negatively charged dyes. Further, the air-sparging increased photocatalytic degradation because of dissolved oxygen.

One of the significant challenges in photocatalysis is separating TiO_2 (or other photocatalysts) slurry particles from the treated wastewater (Dhanya and Aparna 2016). The recovery cost of the photocatalysts can be very high at the largescale treatment of dyeing wastewater. Several researchers have immobilized TiO_2 particles onto various substrate materials to retain the photocatalysts (Mukherjee, Barghi, and Ray 2014). Mukherjee and co-

workers immobilized TiO_2 particles to a polymeric membrane made of PVP (Polyvinyl pyrrolidone) using physical and chemical cross-linking methods for the photocatalytic application (Mukherjee, Barghi, and Ray 2014). The polymeric materials used for immobilization must sustain the UV exposure during the process of photodegradation.

The photocatalyst materials such as TiO_2 and ZnO have also been supported on the surface of activated carbon to exploit their large surface area and porous structure (Suresh, Vijaya, and Kennedy 2014; Zeng et al. 2013). Suresh et al. (Suresh, Vijaya, and Kennedy 2014) synthesized ZnO and NiO nanoparticles supported on the activated carbon (Zn-AC, Ni-AC) to investigate the influence of activated carbon support on the photocatalytic degradation of the textile wastewater. They have reported significantly higher photocatalytic efficiencies in the case of Zn-AC and Ni-AC than the pure ZnO- and NiO-nanoparticles.

2.3.3   Ozonation process

Ozonation is one of the most effective AOPs and has been applied effectively to degrade recalcitrant azo dyes in various research studies (Bethi et al. 2016; Ghuge and Saroha 2018; He et al. 2019; Khamparia and Jaspal 2017). Being a powerful oxidant, Ozone reacts quickly with most organic compounds, including the aromatic rings and the double bonds in azo dyes (Tosik 2005). The oxidation of organic compounds in the presence of ozone takes place via two separate mechanisms (Özbelge and Erol 2008; Wijannarong et al. 2013): (A) Direct oxidation by molecular ozone (O_3), and (B) Indirect or advanced oxidation by free radicals (such as •OH, HO_2•) produced after the decomposition of ozone as shown in Figure 4. Direct oxidation has very slow reaction kinetics, and it is selective to primarily unsaturated hydrocarbons (such as alkenes, alkynes) and aromatic compounds having electron-donating groups (-OH, -CHO, etcetera). The direct oxidation reaction is favored in the acidic (low pH) environments or in the presence of radical scavengers (such as HCO_3^-, CO_3^{--}, acetic acid, tert-butyl alcohol, or chlorides), which inhibit the generation of hydroxyl radicals. The indirect oxidation reaction is non-selective; it oxidizes all organic compounds. Although indirect

oxidation has a very high reaction rate, this oxidation pathway is limited by the short lives of hydroxyl radicals. The indirect oxidation can be promoted in high pH environments or in a radical promoter (such as H_2O_2) which favors the generation of hydroxyl radicals.

Ozonation is one of the most popular oxidation processes to decolorize textile wastewater. Some of the recent works have been summarized in Table 1. The oxidation of an organic compound during ozonation can be maximized by optimizing various process parameters such as pH, temperature, time, and concentration of ozone and additives. Ozbelge et al. and others (Özbelge and Erol 2008; Wijannarong et al. 2013; Oller, Malato, and Sánchez-Pérez 2011), as listed in Table 1, have observed that ozone oxidizes the organic carbons gradually at acidic pH via direct oxidation. However, the oxidation reactions become very fast at basic pH and in the presence of radical initiators such as hydrogen peroxide, formic acid, and metal ions (Fe^{2+}, Ni^{2+}, Cd^{2+}). Fanchiang and coworkers (Fanchiang and Tseng 2009) examined the UV-visible spectrum of the intermediate compounds produced during the ozonation of the Reactive Blue-19 (RB-19) solution to study the oxidation mechanism. They reported complete disappearance of the color and anthraquinone rings of RB-19 after 10 min of the oxidation regardless of initial pH conditions; however, the TOCs were effectively removed only in the alkaline environment. The cause of the increased removal efficiency of TOCs at alkaline pH was attributed to the hydroxyl radical-based indirect oxidation of dyes and their by-products. Somensi et al. (Somensi et al. 2010) have studied the ozonation of dyeing wastewater at different pH conditions in a pilot-scale plant. They also observed a higher color and COD reduction of 67.5% and 25.5% respectively at alkaline pH of 9.1 than the same of 40.6% and 18.7% at acidic pH of 3.0 after 4h of ozone treatment. Different research groups (Venkatesh et al. 2015; Khadhraoui et al. 2009), listed in Table 1, have demonstrated that the pH values of various dyeing solutions decrease with increasing the ozonation time. This indicates the generation of organic acids as by-products upon the oxidation of different azo dyes during ozonation. Therefore, the oxidation pathway

during ozonation shifts from the *non-selective, indirect oxidation* to the *selective, direct oxidation* despite the initial pH level.

The actual dyeing wastewater contains various dyes and additives (such as inorganic and organic salts and polymers) to improve texturing, light resistance, and stability of dyeing performance. Although most chemical treatments of the artificial dyeing wastewaters available in the literature do not consider the effect of additives, the presence of additives can alter the efficiency of a degradation process. Pérez et al. investigated the impact of textile additives Na_2SO_4 and Na_2CO_3 present in the dyeing wastewater on the ozonation of Reactive Black 5 (RB5) and Direct Red 28 (DR28) dyes (Pérez, Poznyak, and Chairez 2013). They reported that additives could slow down the oxidation rate of the ozonation process due to either generation of the new oxidative species instead of hydroxyl radicals (as was the case with Na_2SO_4) *scavenging nature* of the additive compounds (such as Na_2CO_3) towards the free radicals. Further, additives may also enhance the concentrations of organic acids by-products (such as maleic, formic, oxalic, and fumaric acids) formed in the ozonation of dyes. This, in turn, would reduce the pH level of the reaction mixture, favor the direct oxidation pathway, and, therefore, reduce the oxidizing efficiency of the ozonation process.

2.4 Integration of the AOPs with biological methods

The scientific communities are trying to overcome some of the challenges mentioned above of biodegradation by identifying and engineering the efficient microbial strains and utilizing the integration of AOPs with biodegradation to improve the overall degradation efficiency and reduce the toxicity level of the treated dyeing wastewater (Hai, Yamamoto, and Fukushi 2007). The AOPs are coupled with the biodegradation either as a pre-treatment or post-treatment step. Since the aromatic amines, which are generally formed after the biological treatments of azo bonds, are recalcitrant to biodegradation, AOP as a post-treatment step after the biological process may be preferred. However, some AOPs may also generate toxic by-products, which may be biodegraded if the biological treatment is done after

the AOPs. Therefore, the sequence of AOPs and biodegradation in the integrated process needs to be optimized per the chemical structure of dyes and their degradation pathways during chemical and biological treatments. Further, the extent of the AOPs can be reduced in the AOPs-biodegradation integration to bring down the overall cost of processing the dyeing wastewater. Various AOPs and their integrations with biodegradation for the treatment of azo dyes are discussed in detail in the following sub-section.

2.4.1 Integration of Fenton with biodegradation

Several studies have utilized photo-Fenton as a post-treatment step following the conventional biodegradation (Zhang et al. 2014; Yu et al. 2015; Azizi et al. 2015). In a study reported by Punzi and coworkers (Punzi, Anbalagan, et al. 2015), the effluent collected from an anaerobic biodegradation process of the synthetic textile wastewater (containing commercial Remazol Red, starch, and salts) was further treated in a photo-Fenton process at different initial concentrations of Fe^{2+} (1, 3 and 5 mM) and hydrogen peroxide (10, 15 and 20 mM). They observed an increased toxicity level and no further reduction beyond 55% in the COD after four days of anaerobic treatment of the simulated textile wastewater. The Fenton oxidation process has also been reported as a pre-treatment step before the conventional biodegradation for reducing the overall processing time and cost of the decolorization of azo dyes (Rodrigues, Madeira, and Boaventura 2009). The advantages of the photo-Fenton process include simple operation at room temperature, short reaction time, easy-to-handle reactants with regenerated catalyst, and no mass transfer barrier in case of a homogeneous catalytic reaction.

Nonetheless, this process has a few disadvantages, such as the loss of catalyst (ferrous ions) to the effluent in case of a homogeneous Fenton process and the generation of chemical sludge (secondary pollutants) because of leaching of the catalysts in case of heterogeneous Fenton process. The Fenton process integrated with aerobic/ anaerobic biodegradation either as a pre-step or as post-step has the potential to offer improved performance in terms of

decolorization efficiency, cost, and toxicity of the final products. However, the sequence of a combination of the Fenton and biodegradation needs to be optimized as per the nature of the microbes, dyes, and process conditions. For most of the integrated studies, biodegradation plays a significant role in the removal of the organics than the AOPs.

2.4.2 *Integration of photocatalysis with biodegradation*

A combination of photocatalysis and biodegradation can decolorize and detoxify the dyeing wastewater. However, there are relatively few studies reported in literature utilizing the integration of photocatalysis and biodegradation compared to other combined AOP-biodegradation methods. Brosillon et al. utilized photocatalysis followed by a biological treatment by *P. fluorescens* to degrade azo dyes Reactive Yellow 145 (Brosillon et al. 2008). They observed a reduction in the total organic carbon (TOC) level of the by-products produced after photocatalysis when processed through the subsequent biological treatment. This confirmed the feasibility of the integration of photocatalysis with biodegradation.

Recently, Waghmode et al. (Waghmode et al. 2019) examined an integrated photocatalytic and biological treatment to degrade the Methyl Red (MR) dye with an initial dye concentration of 500 mg/L. They obtained 70% decolorization and only 16% COD reduction when the photocatalytic degradation was employed *standalone* for 6hrs.
The advantages of photocatalysis include a simple and efficient process based on cheap and chemically inert metal oxides such as TiO_2. The research efforts performed in the last decade have focused on enhancing the photocatalytic efficiency of the metal oxide catalysts by optimizing particle size, shape, loading, and process conditions. One of the significant challenges associated with the dye effluents' photocatalysis is recovering the photocatalyst from the treated wastewater. Although few efforts are made to immobilize the photocatalyst on a polymeric matrix, the stability of the polymeric matrix during the duration of photo-treatment remains a concern. TiO_2 has been a popular choice as a photocatalyst due to its robust metal support and photocatalytic activity. The application of photocatalysis to process

textile wastewater at high volume faces additional challenges on an economic scale. These challenges include the requirement for a large amount of the photocatalyst and an efficient reactor design, allowing adequate flux of sun-/UV-light to be present everywhere inside the photocatalytic reactor. More research efforts are required in the photocatalysis-biodegradation integration process to treat the textile wastewater at large.

2.4.3 Integration of ozonation with biodegradation

The ozonation process generates toxic by-products and involves significant power consumption. As biological processes are naturally benevolent and cost-effective, the integration of ozonation and biodegradation can offer an effective and economical wastewater treatment, as presented in Table 1 (de Souza, Bonilla, and de Souza 2010; Cresson et al. 2006; Venkatesh, Venkatesh, and Quaff 2017; Paździor et al. 2017). Punzi and coworkers (Punzi, Nilsson, et al. 2015a), as listed in Table 1, developed a process having an anaerobic biofilm reactor followed by ozonation for the treatment of synthetic and real textile wastewaters containing Remazol Red dye. The anaerobic biofilm was used to decompose the biodegradable components of the dyeing wastewater. A 'short' ozone treatment was carried out afterward to break down the recalcitrant molecules to the biological degradation. The toxicity of the effluent after each step was estimated using the Microtox® toxicity meter and the shrimp *Artemia salina*. The Microtox® toxicity-meter (Modern Water Inc., New Castle, United States) utilizes luminescent bacteria (*Vibrio fischeri*) and determines the level of toxicity in a test sample by measuring the reduction in light generated by the bacteria when they are exposed to a toxic substance. Punzi and coworkers found the ozonation–biodegradation integration successful in removing color (by more than 99%), COD (by 85–90%), and toxicity from the 'synthetic' textile wastewater containing 100 – 1000 mg/L of Remazol Red dye. For a 'real' textile effluent, they reported an average reduction of 70% in COD and 95% in toxicity when the textile effluent underwent three days of biological treatment followed by six minutes of ozonation.

Ozonation has also been employed as a pre-treatment step for biodegradation. Qi and coworkers (Qi, Wang, and Xu 2011) treated textile wastewater using the ozonation followed by an aerated biological filter (BAF) for biodegradation. The BAF contained granular media, which provides a large specific surface area for the microbial community to reduce BOD and COD from the wastewater. It was found that the ozonation–BAF integration offered a higher color and COD reduction than that of the ozonation or BAF alone.

The ozonation process is a simple, effective, cheap, and sludge-free oxidation process for the degradation of textile wastewater. The research works available in the literature have conclusively demonstrated a very fast discoloration of the dyeing wastewater in ozonation. The process conditions such as pH, ozone concentration, time, and presence of additives have been shown to alter the reaction kinetics of the ozonation process. Therefore, these process parameters must be optimized for the given dye solution. One of the significant challenges associated with the ozonation process is the incomplete mineralization (poor TOC removal) of the dyeing wastewater (Fanchiang and Tseng 2009; Castro, Bassin, and Dezotti 2017). The addition of radical promoters, such as hydrogen peroxide, has been tried to enhance the mineralization to some extent successfully. Ozonation has also been used with a catalyst such as zerovalent iron (ZVI) to increase the COD and color removal because the ZVI increases the radical formation capabilities of the process (Malik et al. 2018). The generation of toxic by-products during the ozonation process remains a concern.

The integration of ozonation with biodegradation has been shown to have the potential to offer a complete decolorization and significant detoxification of the dyeing wastewater. A purely ozonation-based technology is generally not cost-effective at the large-scale processing of textile wastewater because of the high electricity consumption associated with the process. However, combining ozonation with biodegradation can be a cost-effective method even at the large-scale treatment of wastewater with high dye concentrations. In such integration, the ozonation can be used as a first step to reduce the dye concentration such that cheap

biological processes could subsequently treat it. Multiple cycles of ozonation and biodegradation in different orders (depending on the nature of the dyes and by-products/ metabolites in ozonation/ biodegradation) may also be considered at optimized process conditions. Future research efforts should be aimed in this direction to develop an integrated process that could yield complete mineralization of the high-volume textile azo-dyes at a minimal cost.