

ORIGINAL RESEARCH PAPER

Investigating Feasibility of C. I. Direct Yellow 50 dye Degradation and Detoxification in Synthetic Aquatic Solution Effluent by UVA/TiO₂ Nanophotocatalytic process using Daphnia Magna

Feizollah Dinarvand^{1*}, Nematollah Jaafarzadeh², Mehdi Ahmadi Moghadam², Mohammad Bagher Miranzadeh¹, Nezam Mirzaei¹

¹Kashan University of Medical Science, Kashan, Iran

²Research center, Ahvaz Jundishapur University of Medical Sciences, Ahvaz, Iran

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ABSTRACT

Compounds containing dye are toxic, carcinogenic, and mutagenic for aquatic organisms and lead to mutagenicity, carcinogenicity, and dysfunction of human beings' kidneys, liver, brain, reproductive system, and central nervous system. Advanced oxidation processes can remove pollutants faster than other processes due to active hydroxyl radical production. This study was aimed at investigating the feasibility of dye removal using UVA/TiO₂ process. This study was done in a batch reactor and the effects of initial dye concentrations, TiO₂ nanoparticles dosage, time, pH, and interference compounds on the efficiency of dye degradation were investigated. The Daphnia Magna as bioassay test and biodegradability index (BOD₅/COD rate) were used for detoxification assessment. The D.Y50 dye effluent degradation at pH 2, 20 mg/l initial dye concentration and 1 g/l TiO₂ catalyst was (lnC₀/C= 1.4), (lnC₀/C= 3) and (lnC₀/C= 2.9) respectively. The dye removal rate by 50 mg/l COD concentration was (88%), the Daphnia Magna mortality rate after maximum contact time (96 h) decreased from 96.7% to 43.3% and the biodegradability index increased from 0.25 to 0.68. The current study indicated that UVA/TiO₂ process after optimizing operational factors, has high efficiency in the degradation and detoxification of D.Y 50 dye effluent.

Keywords: Direct Yellow 50 dye, UV/TiO₂, Photodegradation, bioassay, Daphnia Magna

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INTRODUCTION

The POPs (Persistent Organic Pollutants) are frequently detected in treated sewage, surface, groundwater, and even drinking water, and have been observed to be capable of long-range transport, Bioaccumulate in human and animal tissue, Biomagnify in food chains, and to have potentially significant impacts on human health and the environment [1]. The discharge of wastewater containing dye effluents is a significant source of pollution in the ecosystem. These

effluents are proven to be carcinogenic agents, in terms of esthetic and prevailing aquatic flora and fauna [4]. The wide discharge range of synthetic dyes produced and being increasingly used by different industries in ecosystems made this subject a major environmental problem [5]. Dyes are basic daily chemical compounds and are widely used in many fields to color their products such as textile, leather, paper, rubber, printing, plastics, etc [2]. About 200 L of water is needed to produce 1kg of textile and about 1.6 million L of water is consumed for an average-sized textile mill per day [3]. Dyes

* Corresponding Authors Email: dinarvand-f@kaums.ac.ir



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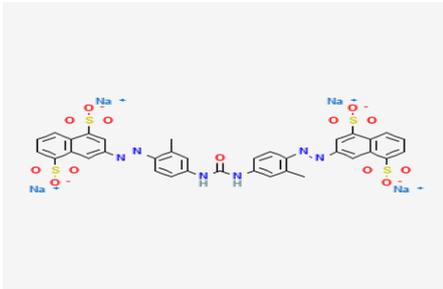
molecules in wastewater lead to mutagenicity, carcinogenicity, and dysfunction of human beings' kidneys, liver, brain, reproductive system, and central nervous system [13]. Azo dyes are not biodegradable and their

presence in water, in addition to adverse esthetic effects, reduces light penetration into water and interferes with aquatic life. Additionally, they are toxic, carcinogenic, and mutagenic to humans [6]. The coloring system of azo dyes includes a group (-N=N-) with one or more aromatic groups [11]. Azo dyes widely use in the textile, ink, paper, plastics, food, and cosmetics industries [14] and Compounds containing azo dye are toxic, carcinogenic, and mutagenic for aquatic organisms [4, 10, 12]. They are also highly stable in the environment and pose a threat to human, animal, and plant species if they enter the environment [14]. The development of advanced water treatment technologies with low-cost and high efficiency to treat the POPs contaminated wastewater is desirable [10]. Currently, available water treatment technologies such as adsorption or coagulation merely concentrate the pollutants present by transferring them to other phases, but they are not completely "eliminated" or "destroyed" [7]. Other conventional water treatment methods such as sedimentation, filtration, chemical oxidation, and biotechnology suffer from some disadvantages such as incomplete removal, high consumption of chemical reagents, high treatment cost, time-consuming, and generation of toxic secondary pollutants [9]. Biological and enzymatic treatment and detoxification, Membrane technologies, photolysis, and electrochemical coagulation for the degradation of hazardous dyes are the new dye removal processes. The photodegradation process of POPs has attracted increasing attention during the past decades due to its effectiveness in rapidly degrading and mineralizing recalcitrant organic compounds and its very high efficiency as well as the absence of secondary contaminants and is used to remove various types of contaminants such as dyes [8,17,18, 19]. The advantages of heterogeneous photocatalytic techniques that make them superior to traditional methods include the removal of pollutants in the range of ppb, production avoidance of polycyclic compounds, and the cost reduction of afferent procedures[76]. Photocatalysis has emerged as one of the most promising and sustainable technologies utilizing the renewable energy of either natural sunlight or artificial

indoor illumination sources which are plentifully available [77]. Various photocatalysts including CdS, SnO₂, WO₃, SiO₂, ZnO, Nb₂O₃, and Fe₂O₃ were being studied but the nano-TiO₂ photocatalyst is well-known among the metal oxides for its high efficiency, low cost, physical and chemical stability, widespread availability, and noncorrosive property [16]. TiO₂ nanomaterials mediated advanced wastewater treatment technique has considerable potential for the removal of various types of complex organic pollutants from wastewater under visible, solar, and UV light sources [20, 23, 24]. Titanium dioxide is technically an important compound that has been widely considered. This compound has useful features, such as low cost, commercial accessibility, chemical stability, biocompatibility, and nontoxicity [78]. In general, AOPs are processes in which by various methods, including a strong oxidizing agent such as hydrogen peroxide, ozone and a catalyst such as zinc oxide, titanium dioxide, and iron, in the presence or absence of ultraviolet radiation, that produces active hydroxyl radicals [29,30,33]. The rationales of AOPs are based on the in situ generation of highly reactive transitory species (i.e. H₂O₂, OH•, O₂⁻, O₃) for the mineralization of refractory organic compounds, water pathogens, and disinfection by-products [26, 27]. Due to their high oxidation capacity of 2.8v, hydroxyl radicals can convert organic chemical pollutants to minerals and have a high efficiency in the oxidation of resistant organic compounds [28]. Due to the possibility of converting raw materials into intermediate compounds with more toxicity in these processes, bioassay studies using different species of living organisms such as fish, algae, bacteria, animal cells, zooplankton, and freshwater organisms done like *Daphnia Magna* [53]. Due to short reproduction, high sensitivity, ease of use, low laboratory costs, and most importantly, the ability to reproduce and produce offspring of a genus with genetic similarity, to monitor the effluent and determine the efficiency of treatment plants to Reduction of toxicity has been high, *Daphnia Magna* regarded by researchers [55]. In a study using *Daphnia magna* and *Vibrio fisheri*, experiments indicated that among these organisms, *Daphnia magna* is more sensitive to environmental pollutants [21]. In this study, decolorization and detoxification of dye solution containing D.Y 50 dye and real textile effluent by nano photocatalytic process were investigated using *Daphnia Magna*. The effect of factors such as TiO₂ dosage and



Table 1. Characteristic of Direct Yellow 50 dye

Parameter	Information
Dye molecule structure	
Dye chemical formula	$C_{35}H_{24}N_6Na_4O_{13}S_4$
MW (g/mol)	956.8
Purity (%)	60
λ -max	402
Company	Alvan Sabet Hamedan

initial dye concentration, irradiation time, pH, and interfering factors in the environment on the efficiency of the decolorization process was investigated and after optimizing these factors, using the bioassay method by *Daphnia Magna* Toxicity reduction was studied.

MATERIALS AND METHODS

The textile azo dye, C.I. Direct Yellow50 (D.Y50), was procured from AlvanSabet, Hamedan Company. The molecule structure and structural information of the dye are shown in Table 1. The TiO_2 nanoparticles were procured from Nano Green Company, Tehran. The physical and chemical properties of TiO_2 nanoparticles are shown in Table

2. Other chemicals used in this study were obtained from Merck Germany with a purity of >99% and deionized laboratory water was used for making aqueous mixtures.

Design and construction of a reactor

The photocatalytic degradation was carried out in a 2 L cylindrical Plexiglass reactor with 7 UVA (PL, HHZM. Co. 9W, low-pressure Hg lamp close to bottom) The UV light sources were surrounded by a Quartz glass tube to prevent direct contact with the solution and the reactor schematic is shown in Fig. 1. The intensity of the lamps was measured by a radiometer EC1 UVA by Silicon photodiode detector, and the measuring range was between 1

Table2. Characteristic of TiO_2 Nanocatalyst

Parameter	TiO_2 Nanocatalyst
MW (g/mol)	89.866
Appearance	Solid white
Density (g/m ³)	4.23
Solubility in water (mg/100ml at 30°)	Insoluble
Particle size (nm)	21
Company	Nano Green Technology

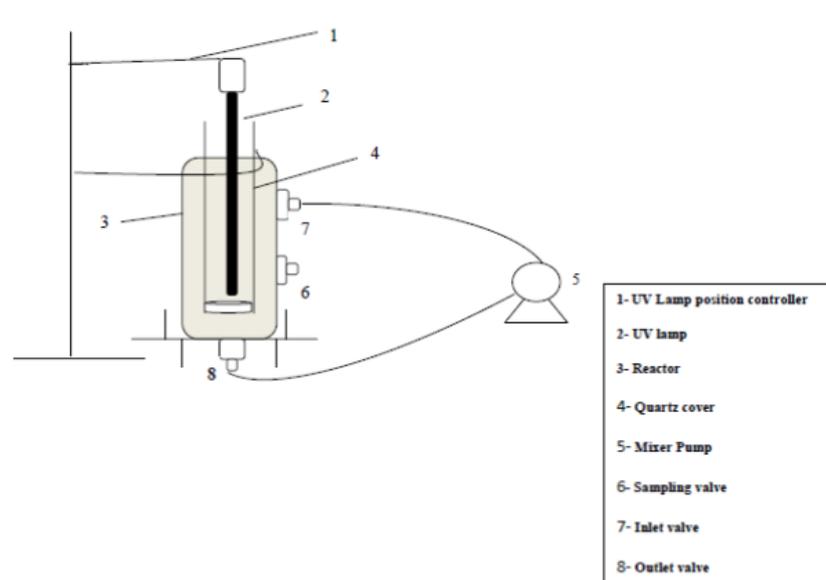


Fig. 1: Photocatalytic Process reactor [73]

to 6 mw/cm^2 . It was $\text{UVA} = 5.4 \text{ (mw / cm}^2\text{)}$. The wavelength range was UVA (315-400).

Preparation of D.Y50 dye calibration curve

To determine the λ -max of the D.Y 50 dye sample, the dye sample was scanned by Shimadzu/UV Probe2700i. It is a double monochromator system capable of 8-Abs measurements. This probe uses the integrating sphere, for the analysis of samples in the wavelength range of 300nm to 1400nm. The D.Y 50 dye λ -max obtained 402 nm. The dye stock solution (1000 mg/l) was prepared and then standard concentrations of 0.25 to 500 mg/l stock solution were prepared to draw the absorption calibration curve of the dye.

Photocatalytic experiments

The photocatalytic experiments were conducted to investigate the degradation of the D.Y 50 dye in aqueous suspension. The dye removal by TiO_2 in a reactor containing a UVA lamp was performed by following steps: 4 ranges 2, 5, 7, and 9 of pH were used to evaluate the effect of pH on the reactions. Initially, pH was obtained by using constant concentrations of 80 mg/l dye and 2 g/l TiO_2 . The next step, was applying the optimal pH, various initial dye concentrations 20, 40, 60, and 80 mg/l, and various TiO_2 concentrations 0.1, 0.25, 0.5, 1, 1.5, and 2 g/l respectively. Sampling was conducted from the reactor at intervals of 5, 10, 15, 30, 60, 90, and 120 minutes. The effect of pH,

catalyst dosage, initial dye concentration, contact time, and the presence of interfering materials on the efficiency of the studied processes was investigated. The experiments were triplicated. Control samples were considered to investigate the effect of each catalyst and UVA light alone on the dye decomposition process. To investigate the effect of interfering agents, the experiments were repeated under optimal conditions with real dyeing wastewater, and the reaction rate constant was compared to synthetic conditions. To determine the characteristics of real wastewater, the COD test was performed by reverse distillation method. A sample is refluxed in a strongly acid solution with a known excess of potassium dichromate. After digestion, the remaining unreduced dichromate was titrated by ferrous ammonium sulfate to determine the amount of potassium dichromate consumed and the oxidizable matter calculated in terms of oxygen equivalent. The BOD method consists of filling with samples. To overflow, an airtight bottle of the specified size and incubate at the specified temperature for 5 days. Dissolved oxygen was measured initially and after incubation, and the BOD was computed from the difference between the initial and final DO (40). The pH was adjusted to the required value by 1N H_2SO_4 or 1N NaOH. The pH is measured by a German Cyber scan pH meter. An injection pump (AEG- MS633-4 - Germany, 60 Lit/h, 220v) was used to mix and prepare a homogeneous solution during the

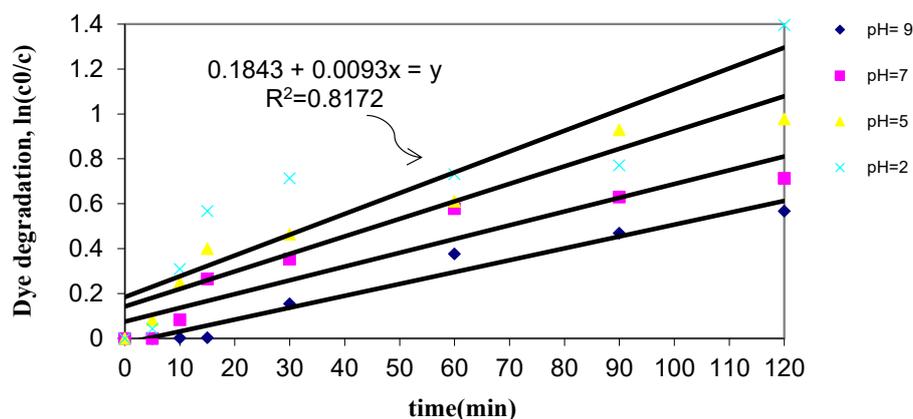


Fig. 2: Effect of pH on D.Y 50 dye degradation by UVA- TiO₂ process (D.Y 50 dye =80 mg/l, TiO₂ = 2 g/l)

experiment. After UV light irradiation for a certain time interval, each sample was centrifuged to separate the suspensions of the TiO₂ nanoparticles by the Hettich Universal x1000U/min at 4000 rpm at ambient temperature for 10 minutes and the residual dye concentration was determined by a Genesys 8 Type Spectrophotometer during the maximum wavelength was measured at 402 nm. The Genesys 8 Type Spectrophotometer include an automatic 7-position sample holder as standard and Wavelength Range is about 200 to 1100 nm. To determine the reaction kinetics, zero, one, and two models are used, and the model that shows the highest R² was determined as the reaction model, and its kinetic parameters were reported.

pH_{zpc} Determination

To determine this parameter, 0.1 M NaCl was used as an electrolyte, and 0.1 M caustic soda and hydrochloric acid solutions were used as pH control. Poured 35 ml of the electrolyte solution into six Erlenmeyer 50 ml and the pH of solutions adjusted in the range of 2 to 12 using acid and caustic soda. 0.2 g of catalyst nanoparticles were added to each Erlenmeyer. To control the results, an Erlenmeyer containing electrolyte without nanoparticles was considered as a control. The contents of each Erlenmeyer were centrifuged (3000 rpm for 15 minutes) and after the above time elapsed, the final pH was read. The pH_{zpc} point was determined by plotting the initial pH values against the final values. [22, 41].

Bioassay

The first generation of the daphnia used has preyed from its natural habitat. The culture medium used for Daphnia culture was based on

the standard method, which includes ordinary municipal dechlorinated water with nutrients 0.192 g/l sodium bicarbonate, 12 g/l calcium sulfate, 0.008 g/l potassium chloride, and yeast. The culture medium was aerated using an aquarium pump for 6 hours a day. In the first step, one of the preyed D. magna was cultivated according to the above-mentioned instructions. D. magna were then nourished to reach their reproductively active life cycle. At 20°C D. magna reach sexual maturity in 6–8 d releasing their eggs into a brood chamber. The embryos complete their development inside the brood chamber and hatch as free-swimming neonates on days 8–10. In the following 2–4 d the mature females release a second brood of neonates with reproduction peaking around the third brood (day 12–14) or fourth brood (day 14–17). As the adult daphnids become older the time between broods will increase and the size of the brood will decrease [33,42]. This generation has the same mother that was used for herd cultivation. Therefore, all of the D. magna used in the present study were genetically quite the same. To test for toxicity in each series, 4 containers with concentrations of 0, 5, 10, and 20 mg/l of dye were prepared and in each concentration, 10 Daphnia with the same genetic traits were used. The colorless sample was used as a control sample. All experiments are triplicated. The tests were performed according to standard procedures and EPA-821-R-02-012 directives. Data analysis and calculation of LD₅₀ were performed using Probit statistical method with SPSS 16.0.

RESULTS

pH effect

pH is considered an important factor since it influences the surface charge properties of the

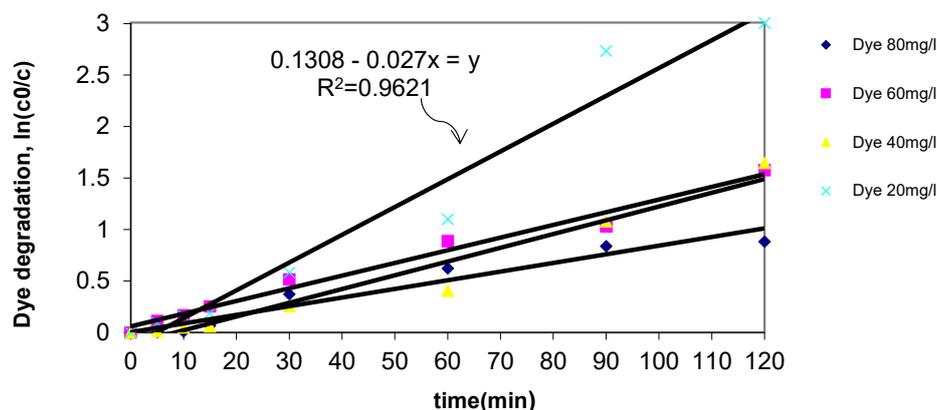


Fig. 3: Effect of Initial dye concentration on D.Y 50 dye degradation by UVA- TiO₂ process (TiO₂ = 1g/l)

catalyst. To study the effect of initial pH on the degradation of D.Y 50 dye, experiments were conducted by varying the pH in the range of 2–9. The amount of active hydroxyl radical produced by UV rays and the amount of effective dye removal depends to a large extent on the pH values of the reaction solution [35]. The pH_{zpc} of TiO₂ was 6. The experimental conditions were an initial dye concentration of 80 mg/l, a TiO₂ concentration of 2 g/L, and an irradiation time of 120 min. Fig. 2 shows the effect of pH on the D.Y 50 dye degradation. The degradation after 120 min irradiation was ($\ln C_0/C = 1.4$) at pH 2. In the UVA/TiO₂ process at pH 2, the dye concentration half-life and the reaction rate constant had the lowest and highest values was 214 min and 0.1542 min⁻¹ respectively.

Effect of Initial Dye Concentration

The effect of initial dye concentration on the removal rate was investigated at the optimum pH of 2 before being determined. The results showed that in UVA/TiO₂ process, by increasing the initial dye concentration and contact time, the dye removal decreased. The highest ($\ln C_0/C = 3$) and lowest ($\ln C_0/C = 0.6$) dye removal efficiency in UVA/TiO₂ photocatalytic process by 1g/l TiO₂ concentration, occurred in 20 mg/l and 80 mg/l initial dye concentration respectively. Fig. 3 shows the effect of initial dye concentration in the UVA/TiO₂ photocatalytic reactor. As shown in Fig. 3, the dye removal efficiency decreased by increasing the initial dye concentration, these results indicate when the dye concentration increased from 20 to 80 mg/l, the dye removal efficiency decreased respectively. The results revealed a gradual decrease in photodegradation activities with increasing the concentration of D.Y 50 dye. Low concentration of

D.Y 50 dye (20 mg/L) has high removal efficiency reaching $\ln C_0/C = 3$. This is due to the presence of a sufficient surface area of the provided catalyst and low intermediate production at a low D.Y 50 dye concentration [73]. However, the rich formation of intermediate products at high concentrations of D.Y 50 dye at constant photocatalyst dose would be competing with the D.Y 50 dye molecules themselves onto the photocatalytic surface, which might explain the decreased degradation of D.Y 50 dye [74]. To control the above steps under optimal pH conditions, the initial dye concentration of 20 mg/l and 1g/l TiO₂ catalyst without UVA radiation (dark condition) was taken as a control sample. The results showed the dye removal percentage in these conditions was very insignificant (0.54%). Also, as a control sample, the dye removal in 20 mg/l initial dye concentration and UVA radiation (without catalyst) was measured, and the dye removal percentage in these conditions was very low (0.06%). According to the first-order kinetic equation, the highest reaction rate constant of the UVA/TiO₂ process was $K = 0.4342 \text{ min}^{-1}$, which indicates the highest reaction rate at the mentioned concentration of titanium dioxide and initial dye concentration compared to other concentrations. Also, according to these equations, the D.Y 50 dye half-life ($t_{1/2}$) was the lowest in the initial concentration of 20 mg/l. As expected there is a reverse relationship between the photocatalytic degradation of D.Y 50 dye and its initial concentration. It is when the initial dye concentration was increased, more dye molecules adsorbed on the surface of TiO₂. The existence of large amounts of adsorbed dye results in the lack of any direct contact with the holes or hydroxyl radicals. This might have an inhibitive effect on dye degradation. Another possible reason for

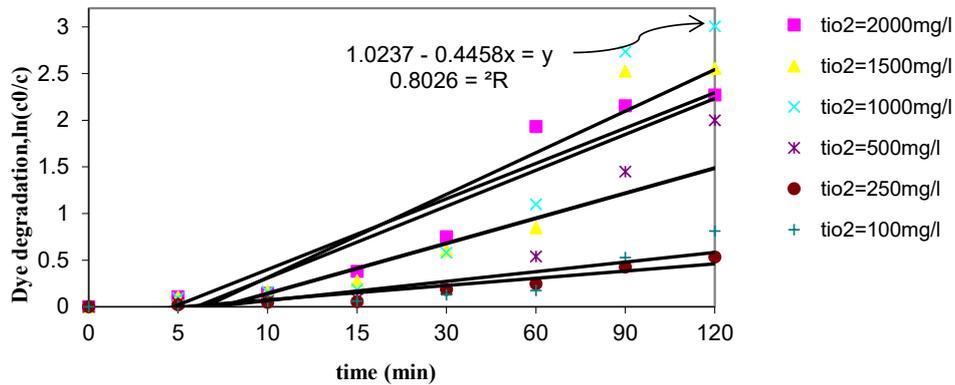


Fig. 4: Effect of TiO₂ concentration on D.Y 50 dye degradation by UVA- TiO₂ process (Initial dye concentration = 20 mg/l, pH= 2)

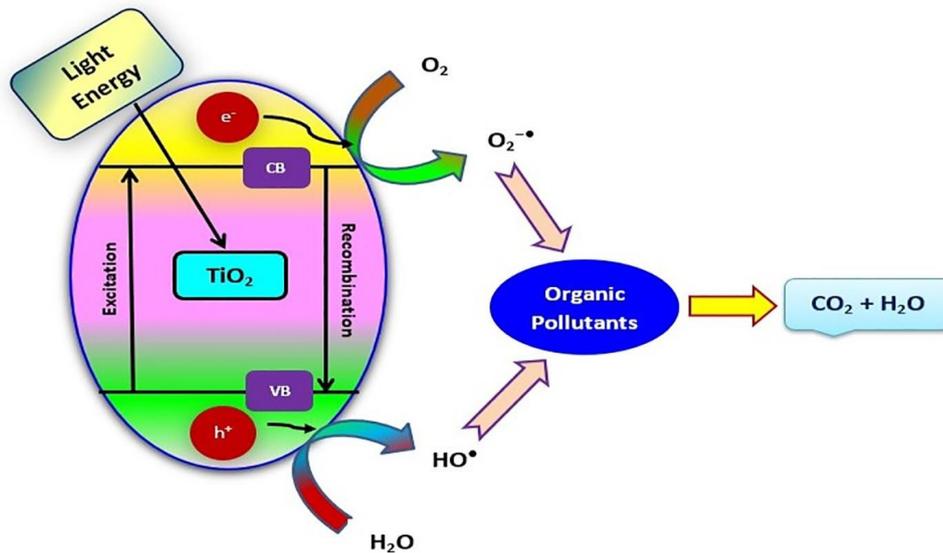


Fig. 5: Photocatalytic degradation of pollutant using TiO₂ Nanomaterial [84]

these results is the effect of UV screening of the dye. At high dye concentration, the solution with a higher concentration may absorb more fraction of the emitted UV radiation than a lower initial dye concentration, and as a result, the number of available photons may decrease, leading to a reduction in the formation of OH[•] radicals. On the other hand, the hydroxyl radical concentrations remain constant for all dye concentrations while the dye molecule increases as the initial concentration of dye increases, hence the degradation efficiency decreases [64]. Another possible reason is the formation of intermediates during the dye molecules' degradation. [59,61].

Effect of Catalyst Dosage

The effect of TiO₂ dosage on the D.Y 50 dye

removal was studied and the results of the UVA/ TiO₂ process are shown in Fig. 4. The experiments were carried out at 20 mg/l dye concentration for 120 min. The removal efficiency increases by increasing the catalyst dosage and then become constant in a specific catalyst dosage. The most effective decomposition of D.Y 50 dye was observed at 1 g/l of TiO₂ dosage. The enhancement of photocatalytic efficiency as a result of increasing photocatalyst dose could be explained based on increasing the available surface area of the photocatalyst and the formation of more active radicals and thought this observation is thought to be that all dye molecules are adsorbed on TiO₂[75]. In addition, the present results showed a relative decrease in photocatalytic efficiency that was observed at dose 2 g/L compared with 1.5 g/L due to the lack of light penetration and

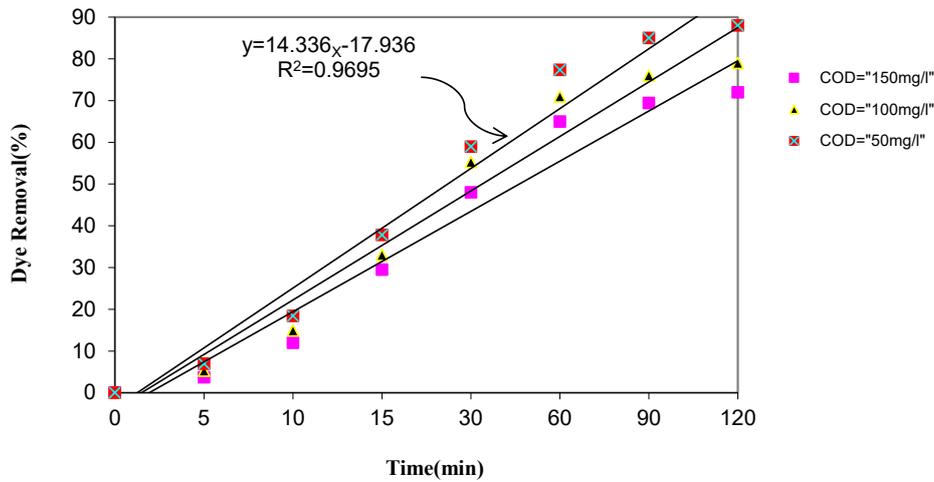


Fig. 6: Effect of Organic Substances on D.Y 50 dye removal by UVA- TiO₂ process (Initial dye concentration = 20 mg/l, TiO₂ = 1g/l)

the photocatalytic cumulative effect in the solution [76]. The addition of higher quantities of TiO₂ would not affect the degradation efficiency. The cause of this is supposedly an increased opacity of the suspension, brought about as a result of excess TiO₂ particles [50,57]. Fig. 4 shows the highest decolorization rate (lnC₀/C = 2.9) in the presence of 20 mg/l D.Y 50 dye initial concentration and 1g/l TiO₂ catalyst. When the TiO₂ dosage increases more than 1g/l, the catalyst particles may agglomerate and this results decreasing in removal percentage [58].

Impact of interfering factors

The COD concentration in real wastewater was 610 mg/l. In the synthetic process 20 mg/l initial dye concentration and 1 g/l catalyst dosage, 50, 100, and 150 mg/l of COD were prepared. As shown in Fig. 6 the highest dye removal (85%) at 50 mg/l COD concentration occurred. Dye removal efficiencies at 100 and 150 mg/l COD concentrations were 75% and 70%, respectively. As shown in Table 3, the reaction rate constant in real wastewater under optimal conditions of the UVA/TiO₂ process was in the range of 0.28 and 0.12 While the reaction rate range in the synthetic sample was between 0.36 and 0.14.

Toxicity test

Comparing Figs. 7 and 8 which shows the Daphnia Magna death percent by contact with different D.Y 50 dye effluent concentration before and after UVA/TiO₂ process, indicate the high toxicity of D.Y 50 dye before degradation. In this

way, the D.Magna death percent when exposed to 20 mg/l initial dye concentration before detoxification at the maximum contact time (96 h) was % 97, while this rate after degradation by UVA/ TiO₂ process by the same initial dye concentration and contact time, decreased to % 41

DISCUSSION

pH effect

The pH influences the characteristics of the photocatalyst surface charge, hence pH of the solution is a significant parameter in performing the reaction on the surface of TiO₂ for the photocatalytic degradation of dyes [69]. According to the results, the dye removal rate increased by decreasing pH. The cause of this phenomenon is due to the anionic D.Y 50 dye structure and the TiO₂ catalyst pH_{zpc} which is 6. Accordingly, due to the positive charge of TiO₂ surface at pH below 6, as pH decreased, and the number of positive charges and the electrostatic attraction between catalyst and pollutants and dye removal efficiency increased respectively [44,48]. When pH increases, the number of negatively charged sites increases,

Table3. Parameter kinetic of first – order of D.Y 50 dye removal process

COD (mg/l)	TiO ₂ /UVA	
	K (min ⁻¹)	T _{1/2} (min)
150	0.1273	102
100	0.2401	85
50	0.2811	64



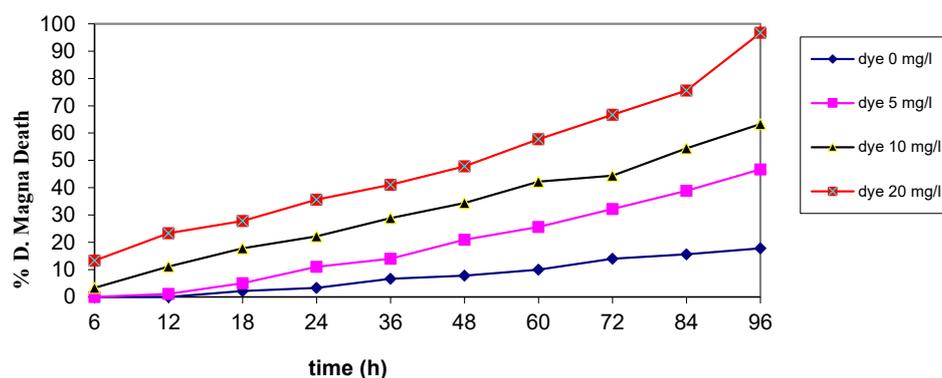


Fig. 7: Death percent of *D. magna* obtained from exposure to different untreated D.B 86 dye effluent (initial *D. magna* = 10)

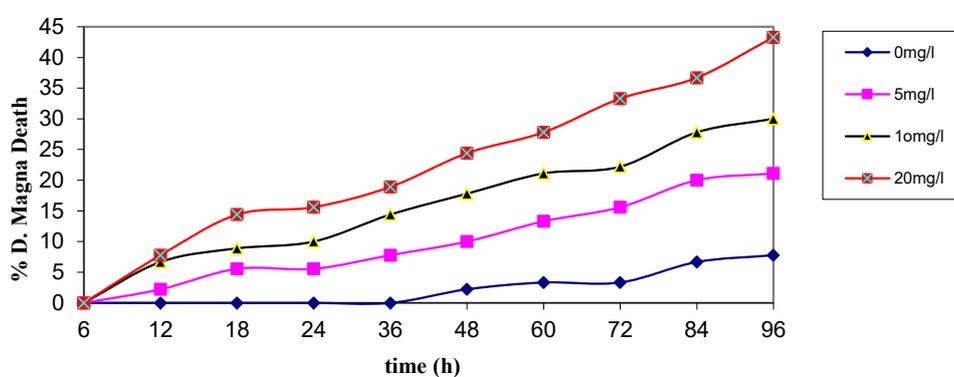


Fig. 8: Death percent of *D. magna* obtained from exposure to different treated D.Y 50 dye effluent after degradation by UVA/TiO₂ (initial *D. magna* = 10)

and since negative sites on the photocatalyst surface cannot be effective in adsorbing colored anions, increasing the pH will reduce the photocatalyst's ability to degrade and decolorize [55]. In addition, from the previous studies, the change in pH around the ZPC point, causes the stick together of catalyst particles, resulting in particle deposition, which will reduce the removal efficiency [31,38]. S. K. Patel et al. 2020, Investigated the photocatalytic degradation of reactive dye in aqueous solution by Fenton, Photo-Fenton and TiO₂. They observed that at a solution with pH of 3 and 4 and lower, dye removal efficiency was greater at UV-TiO₂ process. Mirzadeh et al showed that immobilized laccase of *P. variabile* on porous beads as an efficient biocatalyst for decolorization of synthetic dyes exhibited more than 80% of the maximum decolorization with a wider pH range between 4.5 and 6 [5]. At higher pH values, the dye removal efficiency decreased exponentially. The results of this study showed that most dye removal occurs in the acidic pH range, and the results are in agreement with the present study [15]. A study conducted by Kaur et al demonstrated

that as the pH decreased, dye-ionized molecules are adsorbed on the surface of the TiO₂ catalyst [32]. Mai et al and Shafaei et al, found that at pH less than 9, active levels of positively charged zinc oxide catalysts were coated by dye molecules. Therefore, the production of hydroxyl ions and consequently hydroxyl radicals is reduced. On the other hand, at pH above 9 due to the presence of hydroxyl ions, the surface of the catalyst has a negative charge and the dye adsorption decreases due to its structure at the catalyst level [34,38]. These studies confirm the results of the present study.

Effect of initial dye concentration

Initial dye concentration is one of the most important parameters at the photocatalytic dye decomposition rate which has a significant effect on decomposition efficiency. The photocatalysis depends on the adsorption of dyes on the surface of the photocatalyst. In the photocatalysis process, only the amount of dye adsorbed on the surface of the photocatalyst contributes and not the one in the bulk of the solution [72]. Dye removal decreased

with increasing initial dye concentration indicating that as the dye concentration increases, more dye substances are adsorbed on the surface of TiO_2 , whereas fewer photons are available to reach the catalyst surface and therefore less $\cdot\text{OH}$ radicals are formed, thus causing an inhibition in degradation percentage. Also according to Beer-Lambert law, as initial dye concentration increase, the amount of light or photons passing into the solution decrease, and when trapped and absorbed on the dye particle surface, it causes reducing the absorption of photons on the catalyst and production of radicals respectively. Eventually, it reduces the decomposition rates [37,44,71]. A study conducted by Ghanbarian et al, mentioned that a reason for the decrease in reactive orange dye removal efficiency was the UV-Screening of dye. At high dye concentrations, a large amount of UV radiation may be absorbed by dye molecules instead of being absorbed by TiO_2 particles, which reduces the efficiency of the catalytic reaction by reducing the concentration of hydroxyl and O_2 radicals [54]. Kumar et al. 2017, concluded that the initial methyl orange dye concentration reduction in the effluent caused turbidity reduction, and increasing its concentration caused adsorption on the catalyst surface, followed by a decrease in the active surface [46]. Previous studies have also reported the dependence of the UV- TiO_2 catalytic reaction rate on the initial concentration of contaminants. According to these studies, increasing the initial concentration of dye in this process causes saturation of the catalyst surface, thus reducing the UV absorbance on the catalyst surface and reducing the photocatalytic decomposition rate [17, 45]. The results of Chakrabarti and Mai's study confirm that at dye degradation by UV/ ZnO photocatalytic process, as initial dye concentration increased, the dye removal rate decreased. A reason for this reduction is the adsorption of contaminants on the catalyst surface and consequently declining of active sites on the catalyst surface [44, 51]. Mahvi et al. 2016 showed that by increasing the initial Direct Red 23 azo dye concentration from 25 to 200 mg/L, dye removal efficiency decreased from 99.2 to 20% [6]. According to obtained kinetic equations, the reaction rate constant for the UVA/ TiO_2 process at a concentration of 1 g/l titanium dioxide and initial dye concentration of 20 mg/l was $K = 0.4344$, which indicates a higher reaction rate than other concentrations of catalysts and dyes are present in

this reaction. Also, according to this equation, the D.Y 50 dye half-life ($t_{1/2}$) for the UVA/ TiO_2 process with the above conditions was 23 min.

Effect of Catalyst Dose

As mentioned and shown in Fig. 5, increasing the catalyst concentration increases the active levels of the catalyst and consequently, this process increases the hydroxyl production and other oxidizing radicals [43, 84]. Although this increase in catalyst concentration has a positive effect on the photochemical decomposition process but reduces the decomposition rate by increasing the catalyst concentration to higher than the optimum concentration due to the increase in turbidity, causing light scattering and decreasing the UV penetration [56]. Chakrabarti et al. 2004, showed that by increasing ZnO catalyst concentration, the photocatalytic decomposition rate increases due to an increase in active surfaces on the catalyst and consequently the increase in the production of hydroxyl radicals. Increasing the catalyst concentration higher than the optimal range reduces the decomposition rate. The main reason is the increase in turbidity and interference with the light passage into the solution [51]. Barakat et al. 2010, indicated that increasing TiO_2 dosage to the optimum dose increased the dye decomposition rate. The reason is the increase in dye adsorption on the catalyst surface due to an increase in its concentration [46]. According to Mahvi et al. 2015 as the dose of the nanoparticle increases from 0.2 to 1 g/L dye removal of RB 19 increases, when other parameters are kept constant. With increasing nanoparticles dose more adsorption sites are for the adsorption of RB 19 and consequently, dye removal raises [25].

Impact of interfering factors

The presence of interfering factors in the actual wastewater reduces the reaction rate due to the consumption of hydroxyl produced in the process. Sobczynski et al. 2001 found that a High concentration of pollutants in water saturates the TiO_2 surface and hence reduces the photonic efficiency and deactivation of the photocatalyst [70]. Daneshvar et al. 2007, show increasing organic compounds by increasing the turbidity in the solution, prevents the penetration of light and reduces the absorption of UV photons on the catalyst surface, and ultimately, reduces the dye

Table 4. LC50 and Toxicity Unit (TU) results of D.Y 50 dye before and after degradation by UVA/TiO₂ process

Toxicity Criteria	Time (h)			
	24	48	72	96
LC ₅₀ (mg/l), Untreated Effluent	66	48.3	27.9	16
Toxicity Unit (TU), Untreated Effluent	1.52	2.08	3.23	6.25
LC ₅₀ (mg/l), After Degradation	115	100	70.8	44.7
Toxicity Unit (TU), After Degradation	0.87	1	1.41	2.24

decomposition rate [52]. The above result confirms the results of the present study.

Catalytic process in the presence and absence of UV rays

The low decomposition percentage of D.Y 50 dye in the control sample (0.49%) is due to the major role of UV radiation in the production of highly active hydroxyl radicals [49]. By UV radiation alone, the dye removal efficiency was very low (0.04%). This indicates that hydroxyl radical production which requires the presence of the catalyst, has a major role in the dye decomposition initiation. Also, there was proved that the presence of the catalyst and UV radiation together is the vital factor for initiating and accelerating the photochemical reaction [46].

Toxicity

Most of the aromatic dyes and other aromatic pollutants could be toxic to various organisms, specifically aquatic organisms and plants [62,63]. One of the photocatalytic degradation processes aims is the decreasing or elimination of toxicity associated with the compounds being treated. The toxicity test results on *D. magna* showed that obtained lethal concentration of 50% (LC₅₀) from 24 to 96 hours contact time of D.Y 50 dye effluent before degradation varies from 66 to 16 mg/l. According to EPA guidelines, the D.Y 50 dye is in the relatively toxic group (LC₅₀ greater than 100 in the relatively non-toxic group, 10-100 in the relatively toxic group, 1-10 highly toxic, and less than 1 is ultimately toxic). According to the obtained results, the Toxicity Unit index (TU) of the dye varies from 1.52 to 6.25. As results show in Table 4, based on the EPA guideline, the effluent

after UVA/TiO₂ photocatalytic process is in a relatively non-toxic group (24-hour LC50 equal to 115 mg/l). Goyal et al. 2010, were used *D. magna*, *Vibrio fischer*, and microalgae to evaluate the degree of toxicity reduction in photocatalytic degradation processes. Results showed among these organisms, *D. magna* and *Vibrio fischer* were more sensitive to pollutants respectively [39]. Mahvi et al. 2007 and 2011 in studies conducted on the RO16 and AO7 dye toxicity reduction, concluded that UV/TiO₂ photocatalytic process and horseradish peroxidase (HRP) have a favorable effect on the removal and reduction of the dye toxicity [53, 79]. All results of the previous studies confirm the present study's findings.

Bioassay test and Biodegradability Index (BOD₅/COD rate) Comparison to confirm UVA/TiO₂ process detoxification efficacy

To confirm the photocatalytic degradation efficacy on detoxification of D.Y 50 dye effluent, bioassay test and biodegradability index (BOD₅/COD rate) were compared. The levels of biochemical oxygen demand (BOD₅) and chemical oxygen demand (COD) of wastewater could pose

Table 5. Comparison of ratios of various parameters to characterize wastewater [66]

Type of raw wastewater	BOD ₅ /COD
Untreated wastewater	0.29 – 0.79
Primary Sedimentation	0.41 – 0.59
Final Effluent wastewater	0.11 – 0.31

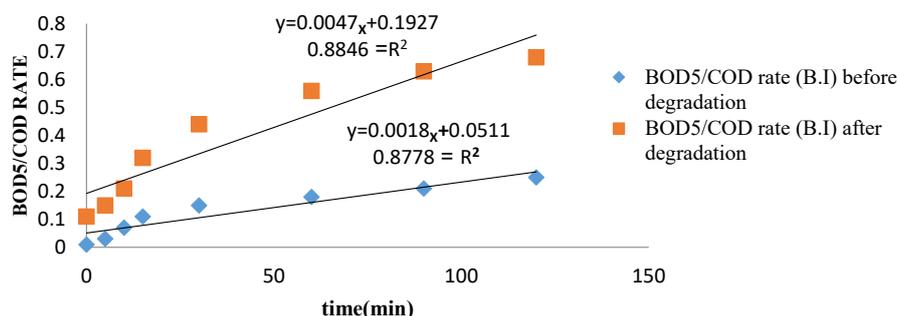


Fig. 9: Biodegradability index (BOD₅/COD rate) values before and after degradation of D.Y 50 dye effluent by UVA/TiO₂ process (dye concentration = 20 mg/l, TiO₂ = 1g/l, time = 120min)

potential pollution to water bodies in which they are discharged. Typical values for the ratio of BOD₅/COD for untreated municipal wastewater are in the approximate range of 0.3 to 0.8 as shown in Table 5 [65, 66, 67]. If BOD₅/COD is > 0.6 then the waste is fairly biodegradable and can be effectively treated biologically. If BOD₅/COD ratio is between 0.3 and 0.6, then seeding is required to treat it biologically, because the process will be relatively slow, as the acclimatization of the microorganisms that help in the degradation process takes time. If BOD₅/COD is < 0.3, biodegradation will not proceed, thus it cannot be treated biologically, because the wastewater generated from these activities inhibits the metabolic activity of bacterial seeds due to their toxicity or refractory properties [68]. It is called the Biodegradability Index (B.I.). It is generally considered the cut-off point between biodegradable and non-biodegradable waste [66]. According to Fig. 9 and comparing the B.I. values of effluent before and after degradation to the mean B.I. can assist in monitoring the presence of toxic and non-biodegradable substances in photocatalytic process effluent. As shown in Fig. 9, B.I. by the maximum exposure time of 120 min, in the control sample without the use of UV and catalyst was 0.25. As mentioned previously, B.I. < 0.3 means the presence of toxic or refractory substances in an effluent sample. Therefore, it shows that in the absence of UV and catalyst detoxification of D.Y 50 dye effluent does not happen. On the other hand, it showed that B.I. after D.Y 50 dye effluent degradation at the same condition was 0.68 and it was upper to 0.6 then the effluent was fairly biodegradable. So, the results showed that photocatalytic degradation of D.Y 50 dye effluent by the UVA/TiO₂ process enhanced the B.I. of effluent.

By comparing Figs. 9 and 10 and survey

changes in bioassay test and B.I. trends, could be interpreted that in photocatalytic degradation of D.Y 50 dye effluent by UVA/TiO₂ process over 120 min, B.I. increased from 0.25 to 0.68 and LC₅₀ over 96 h increased from 16 to 44.7 mg/l respectively and respect to Figs. 7 and 8 this means D. magna mortality rate over 96 h decreased from 97.7 to 43.3 respectively. Both of these experiments confirmed UVA/TiO₂ photocatalytic degradation process efficacy on detoxification of D.Y 50 dye effluent.

CONCLUSION

Decolorization and detoxification of dye solution containing C.I. direct Yellow 50 effluent by UVA/TiO₂ nano photocatalytic process were investigated using *Daphnia Magna*. The effect of factors such as TiO₂ dosage, initial dye concentration, irradiation time, pH, and interfering factors on the efficiency of the decolorization process was investigated and after optimizing these factors, toxicity reduction was studied by bioassay using *Daphnia Magna* and biodegradability index (BOD₅/COD rate). To remove dye by UVA/TiO₂ process, 4 ranges of pH 2, 5, 7, and 9, initial dye concentrations 20, 40, 60, and 80 mg/l, TiO₂ concentrations 0.1, 0.25, 0.5, 1, 1.5, and 2 g/l, at intervals of 5, 10, 15, 30, 60, 90 and 120 min and *daphnia magna* were used respectively. As a result, the D.Y 50 dye effluent degradation at pH 2, 20 mg/l initial dye concentration and 1 g/l TiO₂ catalyst was (lnC₀/C = 1.4), (lnC₀/C = 3) and (lnC₀/C = 2.9) respectively. dye removal rate by 50 mg/l COD concentration was (88%) and the *Daphnia Magna* mortality rate after maximum contact time (96 h) decreased from 96.7% to 43.3%. This study also found that by photocatalytic degradation of D.Y 50 dye effluent using UVA/TiO₂ process over 120 min, B.I. (BOD₅/COD rate) increased from 0.25 to 0.68. Both *Daphnia Magna* mortality rate and biodegradability index (B.I.) confirmed UVA/

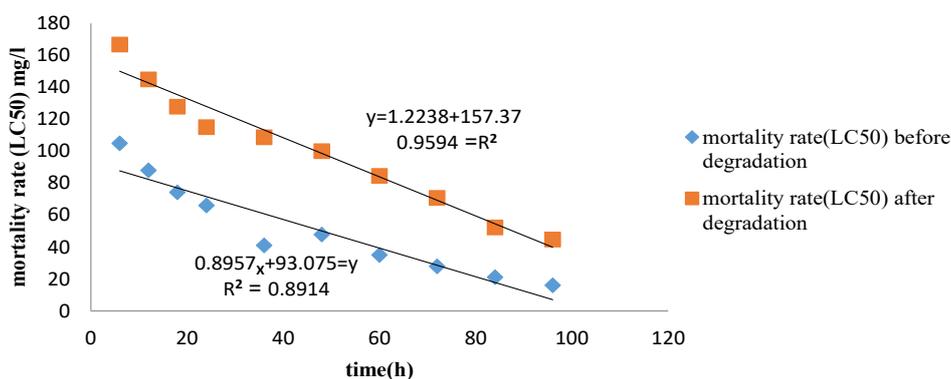


Fig. 10: D. magna mortality rate (LC₅₀) mg/l before and after degradation of D.Y 50 dye effluent by UVA/TiO₂ process (dye concentration = 20 mg/l, TiO₂ = 1g/l, time = 96h)

TiO₂ photocatalytic degradation process efficacy on degradation and detoxification of D.Y 50 dye effluent.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

REFERENCES

- Miranda-Garcia, N., Suarez, S., Sanchez, B., Coronado, J.M., Malato, S., Maldonado, M.I., 2011. Photocatalytic degradation of emerging contaminants in municipal wastewater treatment plant effluents using immobilized TiO₂ in a solar pilot plant. *Appl. Catal. B: Environ.* 103, 294-301. <https://doi.org/10.1016/j.apcatb.2011.01.030>
- Rafatullah, M., Sulaiman, O., Hashim, R., Ahmad, A., 2010. Adsorption of methylene blue on low-cost adsorbents: a review. *J. Hazard Mater.* 177, 70-80. <https://doi.org/10.1016/j.jhazmat.2009.12.047>
- Petcu, A.R., Lazar, C.A., Rogozea, E.A., Olteanu, N.L., Meghea, A., Mihaly, M., 2016. Nonionic microemulsion systems applied for removal of ionic dyes mixtures from textile industry wastewaters. *Separ. Purif. Technol.* 158, 155-159. <https://doi.org/10.1016/j.seppur.2015.12.002>
- V. Oskoei, M.H. Dehghani, S. Nazmara, B. Heibati, M. Asif, I. Tyagi, S. Agarwal, V.K. Gupta, Removal of humic acid from aqueous solution using UV/ZnO nano-photocatalysis and adsorption, *J. Mol. Liq.* 213 (2016) 374-380. <https://doi.org/10.1016/j.molliq.2015.07.052>
- Mirzadeh S.S, Khezri S.M, Rezaei S, Forootanfar H, Mahvi A.H, Faramarzi M.A. Decolorization of two synthetic dyes using the purified laccase of *Paraconiothyrium* variable immobilized on porous silica beads. *Journal of Environmental Health Science & Engineering.* 2014, 12:6. <https://doi.org/10.1186/2052-336X-12-6>
- Dalvand A, Gholibegloo E, Ganjali M.R, Golchinpoor N, Khazaei M, Kamani H, Hosseini S.S, Mahvi A.H. Comparison of Moringa stenopetala seed extract as a clean coagulant with Alum and Moringa stenopetala-Alum hybrid coagulant to remove direct dye from Textile Wastewater. *Environ Sci Pollut Res.* 2016, 23:16396-16405.7. Padmanabhan, P.V.A., Sreekumar, K.P., Thiyagarajan, T.K., Satpute, R.U., Bhanumurthy, K., Sengupta, P., Dey, G.K., Warriar, K.G.K., 2006. Nano-crystalline titanium dioxide formed by reactive plasma synthesis. *Vacuum* 80, 11-12. <https://doi.org/10.1007/s11356-016-6708-z>
- Chong, M.N., Jin, B., Chow, C.W.K., Saint, C., 2010. Recent developments in photocatalytic water treatment technology: a review. *Water Res.* 44, 2997e3027. <https://doi.org/10.1016/j.watres.2010.02.039>
- Zhou, Y.Y., Tang, L., Zeng, G.M., Chen, J., Cai, Y., Zhang, Y., Yang, G.D., Liu, Y.Y., Zhang, C., Tang, W.W., 2014. Mesoporous carbon nitride based biosensor for highly sensitive and selective analysis of phenol and catechol in compost bioremediation. *Biosens. Bioelectron.* 61, 519-525. <https://doi.org/10.1016/j.bios.2014.05.063>
- Soltani RDC, Jorfi S, Ramezani H, Purfadakari S. Ultrasonically induced ZnO-biosilica nanocomposite for degradation of a textile dye in aqueous phase. *Ultrason Sonochem* 2016; 28: 69-78. <https://doi.org/10.1016/j.ultrsonch.2015.07.002>
- Rizzo L. Bioassays as a tool for evaluating advanced oxidation processes in water and wastewater treatment. *Water Res* 2011;45(15): 4311-4340. <https://doi.org/10.1016/j.watres.2011.05.035>
- Yousefi Z, Bandpei AM, Tileki R Ad, Malaki A, Mohamadpur RA, Ghahramani E. Evaluation of the Combined GAC-SBR System Performance in the Removal of Yellow 3 and Disperse Yellow 3 Reactive Dyes from the Waste. *J Mazandaran Univ Med Sci* 2012; 21(1): 41-49 (Persian).
- Zhou, Y., Lu, J., Zhou, Y., Liu, Y., 2019. Recent advances for dyes removal using novel adsorbents: A review. *Environmental Pollution* 252, 352-365. <https://doi.org/10.1016/j.envpol.2019.05.072>
- Dutta M, Ghosh P, Basu JK. Application of artificial neural network for the decolorization of direct blue 86 by using microwave assisted activated carbon. *J Taiwan Inst Chem* 2012; 43(6): 879-888. <https://doi.org/10.1016/j.jtice.2012.06.007>

15. Saurabh K, Patel. Sunil G, Patel. Geeta V. Patel. Degradation of Reactive Dye in Aqueous Solution by Fenton, Photo-Fenton Process and Combination Process with Activated Charcoal and TiO₂. Proc. Natl. Acad. Sci., India, Sect. A Phys. Sci. (October 2020) 90(4):579-591 <https://doi.org/10.1007/s40010-019-00618-3>
16. Herrmann, J., Duchamp, C., Karkmaz, M., Hoai, B., Lachheb, H., Puzenat, E.G., 2007. Environmental green chemistry as defined by photocatalysis. J. Hazard. Mater. 146, 624-629. <https://doi.org/10.1016/j.jhazmat.2007.04.095>
17. Chong, M.N., Jin, B., Chow, C.W.K., Saint, C., 2010. Recent developments in photocatalytic water treatment technology: a review. Water Res. 44, 2997e3027. <https://doi.org/10.1016/j.watres.2010.02.039>
18. Miranda-Garcia, N., Suarez, S., Sanchez, B., Coronado, J.M., Malato, S., Maldonado, M.I., 2011. Photocatalytic degradation of emerging contaminants in municipal wastewater treatment plant effluents using immobilized TiO₂ in a solar pilot plant. Appl. Catal. B: Environ. 103, 294e301. <https://doi.org/10.1016/j.apcatb.2011.01.030>
19. Alijani S, Vaez M, Moghadam AZ. Comparative Study on the Photodegradation of Acid Black 26 from Synthetic Wastewater using Slurry and Immobilized TiO₂ on the Sackcloth Fiber. IJHE 2013; 6(2): 243-56 (Persian).
20. Abdelhaleem, A., Chu, W., 2017. Photodegradation of 4-chlorophenoxyacetic acid under visible LED activated N-doped TiO₂ and the mechanism of stepwise rate increment of the reused catalyst. J. Hazard. Mater. 338, 491-501. <https://doi.org/10.1016/j.jhazmat.2017.05.056>
21. Czech B, Joško I, Oleszczuk P. Ecotoxicological evaluation of selected pharmaceuticals to *Vibrio fischeri* and *Daphnia magna* before and after photo oxidation process. Ecotoxicol Environ Saf 2014; 104:247-253. <https://doi.org/10.1016/j.ecoenv.2014.03.024>
22. Azizi S, Shookohi R, Fredmal J. Biosorption of Pentachlorophenol from Aqueous Solutions Using *Phanerochaete chrysosporium* Biomass. J Mazandaran Univ Med Sci 2014; 24(113):105-117 (Persian).
23. Hinojosa-Reyes, M., Camposeco-Solis, R., Ruiz, F., Rodríguez-González, V., Moctezuma, E., 2019. Promotional effect of metal doping on nanostructured TiO₂ during the photocatalytic degradation of 4-chlorophenol and naproxen sodium as pollutants. Mater. Sci. Semicond. Process 100, 130-139. <https://doi.org/10.1016/j.mssp.2019.04.050>
24. Yi, C., Liao, Q., Deng, W., Huang, Y., Mao, J., Zhang, B., Wu, G., 2019. The preparation of amorphous TiO₂ doped with cationic sand its application to the degradation of DCFs under visible light irradiation. Sci. Total Environ. 684, 527-536. <https://doi.org/10.1016/j.scitotenv.2019.05.338>
25. Dalvand A, Nabizadeh R, Ganjali M.R, Khoobi M, Nazmara S, Mahvi A.H. Modeling of Reactive Blue 19 azo dye removal from colored textile wastewater using larginine-functionalized Fe₃O₄ Nanoparticles: Optimization, reusability, kinetic and equilibrium studies, Journal of Magnetism and Magnetic Materials. 2015, 404: 179-189. <https://doi.org/10.1016/j.jmmm.2015.12.040>
26. Esplugas, S., Gimenez, J., Conteras, S., Pascual, E., Rodríguez, M., 2002. Comparison of different advanced oxidation processes for phenol degradation. Water Res. 36, 1034-1042. [https://doi.org/10.1016/S0043-1354\(01\)00301-3](https://doi.org/10.1016/S0043-1354(01)00301-3)
27. Pera-Titus, M., García-Molina, V., Bano, M.A., Gimenez, J., Esplugas, S., 2004. Degradation of chlorophenols by means of advanced oxidation processes: a general review. Appl. Catal. B: Environ. 47, 219-256. <https://doi.org/10.1016/j.apcatb.2003.09.010>
28. Banerjee, S., Pillai, S.C., Falaras, P., O'Shea, K.E., Byrne, J.A., Dionysiou, D.D., 2014. New insights into the mechanism of visible light photocatalysis. J. Phys. Chem. Lett. 5, 2543e2554. <https://doi.org/10.1021/jz501030x>
29. Farsani MH, Moghaddam MA, Shirmardi M, Mengelzadeh N-A, Farsani AH, Naeimabadi A. The effectiveness of the UV/H₂O₂/UV/H₂O₂ and ZrO₂/SiO₂ processes in removal of the nickel from the aqueous environments. Journal of North Khorasan University of Medical Sciences 2013; 5(4):741-750 (persian). <https://doi.org/10.29252/jnkums.5.4.741>
30. Moussavi G, Borghee M, Farzadkia M, Asl RA. Decolorization and mineralization of reactive red 198 in saline water: performance comparison of photolysis, UV/TiO₂, and UV/ZnO processes. Environ Eng Manag J 2015; 14(5): 1027-1036. <https://doi.org/10.30638/eemj.2015.113>
31. Venkatachalam N, Palanichamy M, Arabinoo B, Murugesan V. Alkaline earth metal doped nanoporous TiO₂ for enhanced photocatalytic mineralisation of bisphenol-A. Catalysis Communications. Catalysis Communications 2007; 8(7): 1088-1093. <https://doi.org/10.1016/j.catcom.2006.10.025>
32. Kaur S, Singh V. Visible light induced sonophotocatalytic degradation of Reactive Red dye 198 using dye sensitized TiO₂. Ultrason Sonochem 2007; 14(5): 531-537. <https://doi.org/10.1016/j.ultrsonch.2006.09.015>
33. Jorfi S, Rezaee A, Moheb-ali G-A, Jaafarzadeh NA. Pyrene removal from contaminated soils by modified Fenton oxidation using iron nano particles. Journal of Environmental Health Sciences & Engineering 2013; 11-17. <https://doi.org/10.1186/2052-336X-11-17>
34. Shafaei A, Nikazar M, Arami M. Photocatalytic degradation of terephthalic acid using titania and zinc oxide photocatalysts: Comparative study. Desalination 2010; 252(1-3): 8-16. <https://doi.org/10.1016/j.desal.2009.11.008>
35. American Public Health Association, American Water Works Association, and Water Environment Federation, Standard Methods for the Examination of Water and Wastewater, American Public Health Association/American Water Works Association/Water Environment Federation, Washington, DC, USA, 20th edition, 1998.
36. Peternel IT, Koprivanac N, Bozi'AM, Kusic HM. Comparative study of UV/TiO₂, UV/ZnO and photo-Fenton processes for the organic reactive dye degradation in aqueous solution. J Hazard Mater 2007; 148(1-2):477-484. <https://doi.org/10.1016/j.jhazmat.2007.02.072>
37. Kansal SK, Singh M, Sud D. Studies on photodegradation of two commercial dyes in aqueous phase using different photocatalysts. J Hazard Mater 2007; 141(3): 581-590. <https://doi.org/10.1016/j.jhazmat.2006.07.035>
38. Mai FD, Chen CC, Chen JL, Liu SC. Photodegradation of methyl green using visible irradiation. Determination of the reaction pathway and identification of intermediates by a high performance liquid chromatography-photodiode array-electrospray ionization-mass spectrometry method. J Chromatogr A 2008; 1189(1-2):35-365. <https://doi.org/10.1016/j.chroma.2008.01.027>
39. R.N. Goyal, V.K. Gupta, S. Chatterjee, Voltammetric biosensors for the determination of



- paracetamol at carbonnanotube modified pyrolytic graphite electrode, *Sens. Actuators B: Chem.* 149 (2010) 252-258. <https://doi.org/10.1016/j.snb.2010.05.019>
40. Nishio J, Tokumura M, Znad HT, Kawase Y. Photocatalytic decolorization of azo-dye with zinc oxide powder in an external UV light irradiation slurry photoreactor. *J Hazard Mater* 2006; 138(1): 106-115. <https://doi.org/10.1016/j.jhazmat.2006.05.039>
 41. Hoseinzadeh E, Samargandi MR, Alikhani MY, Roshanaei G, Asgari G. Antimicrobial Efficacy of Zinc Oxide Nanoparticles Suspension Against Gram Negative and Gram Positive Bacteria. *IJHE* 2012; 5(3):331-342 (Persian)
 42. U.S. Environmental Protection Agency Office of Water. Methods for measuring the acute toxicity of effluents and receiving waters to fresh water and marine organisms. Washington, DC. EPA .5th ed. 2002.
 43. Konstantinou IK, Albanis TA. TiO₂-assisted photocatalytic degradation of azo dyes in aqueous solution: kinetic and mechanistic investigations. *Appl Catal B* 2004;49(1): 1-14. <https://doi.org/10.1016/j.apcatb.2003.11.010>
 44. Naik L, Devi G, Kottam, Nagaraju. Photocatalytic Degradation of Azo Dyes over Mn²⁺ Doped TiO₂ Catalyst under UV/Solar Light: An Insight to the Route of Electron Transfer in the Mixed Phase of Anatase and Rutile. *Chin J Chem* 2010; 28(11): 2151-2161. <https://doi.org/10.1002/cjoc.201090356>
 45. Ochuma IJ, Fishwick RP, Wood J, Winterbottom JM. Optimisation of degradation conditions of 1,8-diazabicyclo[5.4.0]undec-7-ene in water and reaction kinetics analysis using a co-current down flow contactor photocatalytic reactor. *Appl Catal B* 2007;73(3-4): 259-268. <https://doi.org/10.1016/j.apcatb.2006.12.008>
 46. Kumar A, Pandey G. Photodegradation of Methyl Orange in Aqueous Solution by the Visible Light Active Co:La:TiO₂ Nanocomposite. *Chem Sci J.* 2017;8:164.
 47. Mahvi AH, Ghanbarian M, Naddafi K, Mahmoodi N. Investigation of the Toxicity Reduction in Reactive Dye Solution and Real Textile Wastewater by Nanophotocatalysis Process Using *Daphnia Magna*. *Journal of Color Science and Technology* 2008; 1(2):91-96 (Persian).
 48. Ghaneian MT, Ehrampoush MH, Dehvary M, Jamshidi B, Amrollahi M. A Survey of the Efficacy of Cuttle Fish Bone Powder in the Removal of Reactive Red 198 dye from Aqueous Solution. *Toloo Behdasht* 2012;10(3-4): 127-138 (Persian).
 49. Mai FD, Lu CS, Wu CW, Huang CH, Chen JY, Chen CC. Mechanisms of photocatalytic degradation of Victoria Blue R using nano-TiO₂. *Sep Purif Technol* 2008; 62(2): 423-436. <https://doi.org/10.1016/j.seppur.2008.02.006>
 50. D.E. Kritikos, N.P. Xekoukoulotakis, E. Psillakis, D. Mant-zavinos, Photocatalytic degradation of reactive black 5 in aqueous solutions: Effect of operating conditions and coupling with ultrasound irradiation, *J. water research.* 41, 2236 - 2246, (2007). <https://doi.org/10.1016/j.watres.2007.01.048>
 51. Chakrabarti S, Dutta BK. Photocatalytic degradation of model textile dyes in wastewater using ZnO as semiconductor catalyst. *J Hazard Mater* 2004; 112(30): 269-278 <https://doi.org/10.1016/j.jhazmat.2004.05.013>
 52. Daneshvar N, Rasoulifard MH, Khataee AR, F. Hosseinzadeh. Removal of C.I. Acid Orange 7 from aqueous solution by UV irradiation in the presence of ZnO nanopowder. *J Hazard Mater* 2007; 143(1-2):95-101. <https://doi.org/10.1016/j.jhazmat.2006.08.072>
 53. Mahvi AH, Ghanbarian M, Naddafi K, Mahmoodi NM. Investigation of the Toxicity Reduction in Reactive Dye Solution and Real Textile Wastewater by Nanophotocatalysis Process Using *Daphnia Magna*. *Journal of Color Science and Technology* 2007;1(2):91-96.
 54. Ghanbarian M, Mahvi AH, Nabizadeh R, Saeednia S. A Pilot Study of RO16 Discoloration and Mineralization in Textile Effluents Using the Nanophotocatalytic Process. *Journal of water and wastewater* 2008; 1: 45-51 (Persian).
 55. KHodadadi A, Ganjidoust H, Panah H. Purification and kinetics of synthetic wastewater containing naphthalene using titanium dioxide nanoparticles stabilized on activated carbon. *IJHE* 2012; 4(4): 401-10 (Persian).
 56. Behnajady MA, Modirshahla N, Hamzavi R. Kinetic study on photocatalytic degradation of C.I. Acid Yellow 23 by ZnO photocatalyst. *J Hazard Mater* 2006; 133(1-3): 226-232. <https://doi.org/10.1016/j.jhazmat.2005.10.022>
 57. Ch.H. Wu, H.W. Chang, J. Chern, Basic dye decomposition kinetics in a photocatalytic slurry reactor, *J. Hazardous Materials*. B137, 336-343, (2006). <https://doi.org/10.1016/j.jhazmat.2006.02.002>
 58. C.G. Joseph, G.L. Puma, A. Bono, D. Krishnaiah, Sonophotocatalysis in advanced oxidation process: A short review, *J. Ultrasonics Sonochemistry.* 16, 583-589, (2009). <https://doi.org/10.1016/j.ultsonch.2009.02.002>
 59. F. banat, S. Al-Asheh, M. Al-Rawashteh, M. Nusair, photo-degradation of methylene blue dye by the UV/H₂O₂ and UV/acetone oxidation processes, *J. Desalination.* 18, 225-232, (2005). <https://doi.org/10.1016/j.desal.2005.04.005>
 60. N. Shimizu, C. Ogino, M.F. Dadjour, T. Murata, Sonocatalytic degradation of methylene blue with TiO₂ pellets in water, *J. Ultrasonics Sonochemistry.* 14, 184-190, (2007). <https://doi.org/10.1016/j.ultsonch.2006.04.002>
 61. M. Mrowetz, C. Pirola, E. Selli, Degradation of organic water pollutants through sonophotocatalysis in the presence of TiO₂, *J. Ultrasonics Sonochemistry.* 10, 247-254, (2003). [https://doi.org/10.1016/S1350-4177\(03\)00090-7](https://doi.org/10.1016/S1350-4177(03)00090-7)
 62. Martins, M.; Santos, J.M.; Diniz, M.S.; Ferreira, A.M.; Costa, M.H.; Costa, P.M. Effects of carcinogenic versus non-carcinogenic AHR-active PAHs and their mixtures: Lessons from ecotoxicological relevance. *Environ. Res.* 2015, 138, 101-111. [CrossRef] [PubMed] <https://doi.org/10.1016/j.envres.2015.02.010>
 63. Alam, M.Z.; Ahmad, S.; Ahmad, M. Mutagenicity and genotoxicity of tannery effluents used for irrigation at Kanpur, India. *Ecotoxicol. Environm. Saf.* 2010, 73, 1620-1628. [CrossRef] [PubMed]. <https://doi.org/10.1016/j.ecoenv.2010.07.009>
 64. Feng J, Hu X, Yue PL, Zhu HY, Lu GQ (2003) Discoloration and mineralization of reactive red HE-3B by heterogeneous photo-Fenton reaction. *Water Res* 37(15):3776-3784. [https://doi.org/10.1016/S0043-1354\(03\)00268-9](https://doi.org/10.1016/S0043-1354(03)00268-9)
 65. Alsaqqar, Awatif Soaded.; Khudair, Basim Hussein.; and Makki, Ahmed, Performance Evaluation of the Organic Matter Removal Efficiency in Wastewater Treatment Plants; Case study Al- Diwaniyah WWTP in Iraq. *International*

- Journal of Science and Research 6(2): 334-338 (2017).
66. Metcalf and Eddy, Wastewater Engineering, Treatment and Reuse' 4th edition, McGraw Hill Com. Inc. New York (2003).
 67. A.M. Al-Sulaiman, B.H. Khudair, Correlation Between BOD5 AND COD for Aldiwaniyah Wastewater Treatment Plants to obtain the Biodegradability Indices. Pak. J. Biotechnol. Vol. 15 (2) 423-427 (2018).
 68. Khaled, Zaher Abdallaa.; and Gina, Hammam, Correlation between Biochemical Oxygen Demand and Chemical Oxygen Demand for Various Wastewater Treatment Plants in Egypt to Obtain the Biodegradability Indices. International Journal of Sciences: Basic and Applied Research (IJSBAR) (2014) Volume 13, No 1, pp 42-48.
 69. C.-S. Lu, C.-C. Chen, F.-D.Mai, and H.-K. Li, "Identification of the degradation pathways of alkanolamines with TiO₂ photocatalysis," Journal of Hazardous Materials, vol. 165, no.1-3, pp.306-316, 2009. <https://doi.org/10.1016/j.jhazmat.2008.09.127>
 70. Sobczynski A, Dobosz A. Water purification by photocatalysis on semi-conductors. Polish Journal of Environmental Studies. 2001;10(4):195- 205.
 71. L. C.Macedo, D. A. M. Zaia, G. J.Moore, and H. de Santana, "Degradation of leather dye on TiO₂: a study of applied experimental parameters on photoelectrocatalysis," Journal of Photochemistry and Photobiology A: Chemistry, vol. 185, no. 1, pp. 86-93, 2007. <https://doi.org/10.1016/j.jphotochem.2006.05.016>
 72. Reza KM, Kurny ASW, Gulshan F. Parameters affecting the photocatalytic degradation of dyes using TiO₂: a review. Applied Water Science. 2017;7(4):1569-1578. <https://doi.org/10.1007/s13201-015-0367-y>
 73. Ahmed, S., Rasul, M., Martens, W.N., Brown, R., Hashib, M., 2010. Heterogeneous photocatalytic degradation of phenols in wastewater: a review on current status and developments. Desalination 261, 3-18. <https://doi.org/10.1016/j.desal.2010.04.062>
 74. Kashif, N., Ouyang, F., 2009. Parameters effect on heterogeneous photocatalysed degradation of phenol in aqueous dispersion of TiO₂. J. Environ. Sci. 21, 527-533. [https://doi.org/10.1016/S1001-0742\(08\)62303-7](https://doi.org/10.1016/S1001-0742(08)62303-7)
 75. MirzaHedayat, B., Noorisepehr, M., Dehghanifard, E., Esrafil, A., Norozi, R., 2018. Evaluation of photocatalytic degradation of Dinitrophenol from synthetic wastewater using Fe₃O₄@SiO₂/TiO₂/rGO magnetic nanoparticles. J. Mol. Liq. 264, 571-578. <https://doi.org/10.1016/j.molliq.2018.05.102>
 76. Tayeb R, Mohammadzadeh Kakhki R, Audebert P, et al. A robust UV-visible light-driven SBA-15-PS/phthalhydrazide nanohybrid material with enhanced photocatalytic activity in the photodegradation of methyl orange. Appl Organometal Chem. 2018;e4391. <https://doi.org/10.1002/aoc.4391>
 77. Mohammadzadeh Kakhki R, Tayeb R, Hedayat S. Phthalhydrazide nanoparticles as new highly reusable organic photocatalyst in the photodegradation of organic and inorganic contaminants. Appl Organometal Chem. 2017;e4033. <https://doi.org/10.1002/aoc.4033>
 78. Javadi F, Tayeb R, Bahramian B. TiO₂/nanoclinoptilolite as an efficient nanocatalyst in the synthesis of substituted 2-aminothiophenes. Appl Organometal Chem. 2017; e3779. <https://doi.org/10.1002/aoc.3779>
 79. Gholami-Borujeni F, Mahvi A. H, Nasser S, Nabizadeh R, Alimohammadi M. Enzymatic Treatment and Detoxification of Acid Orange 7 from Textile Wastewater. Appl Biochem Biotechnol 2011, 165:1274-1284. <https://doi.org/10.1007/s12010-011-9345-5>

