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Introducing a High Throughput Nanozymatic Method for Green Nanozyme-Mediated Degradation of Organic Dyes in Real Water Media

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Abstract: In this work, a high throughput nanozymatic method was developed for green nanozyme-mediated biodegradation of organic dyes in real water media. The nanozymes were synthesized and characterized for their size and morphology by TEM and DLS analysis. The nanozymatic properties of the as-prepared nanomaterials were evaluated by standard enzyme activity assay, revealing high peroxidase-like performances for the as-prepared nanozymes. Therefore, for exploring more precise, the kinetics behavior of the as-prepared nanozymes was evaluated by the standard Lineweaver-Burk method, revealing a V_{max} as high as 0.263 μM sec^{-1} and a K_m as low as 0.03 mM, revealing high catalytic efficiency and affinity of the as-prepared nanozymes. Hence, the as-prepared nanozymes were utilized for organic dye degradation in real water media using methylene blue as a model organic dye. The effective factors on the dye degradation including pH, ionic strength, degradation time, nanozyme amount, etc. were optimized by the one-factor-at-a-time optimization method. At optimal experimental conditions, the as-prepared nanozymes can degrade about 99.0% of the organic dye within a degradation time as low as 7 min. The developed method was finally employed for the nanozyme-mediated degradation of methylene blue from real water media such as pool, mineral, river, and tap water samples. The results of this study revealed an excellent biodegradation yield of over 94.3%-99.0% for the different real samples, proving the suitability of the developed method for dye degradation in real water media.

Keywords: nanozyme-mediated dye degradation; organic dye; nanozyme; methylene blue; real water media

Introduction

Industrial wastewaters commonly contain several toxic wastes with high water solubility, for instance, toxic organic dyes, phenols, and pharmaceutically active compounds which are non-biodegradable. Hence, entering these non-biodegradable into the environment makes several serious environmental damages [1,2]. Considering these issues, the high throughput removal of wastes is necessary from an environmental safety view. There are several different methods for the removal of contaminates (e.g., dyes) from water media including simple adsorption-based systems or catalytic degradation of dyes to mineral and safe materials [2]. In the case of catalytic degradation, up to now, several methods are introduced for dye removal such as chemo-degradation [3], photo-degradation [4], biodegradation [5], photo-induced nanozymatic degradation [2], multienzyme-based degradation [6], and chemo-bio degradation [7]. Among different methods, nanozyme-based dye degradation recently attracted more attention due to their high catalytic efficiency, fast degradation, green properties, and high stability against environmental harsh conditions compared to natural enzymes. In fact, despite the high specificity and catalytic efficiency of the native enzymes, they suffer several drawbacks such as low stability against environmental changes [8]. Hence, replacing native enzymes with high stable nanozymes can be considered a new point in the biocatalysis field. The fast development of material science and nanotechnology led to introduce several novel nanoscale materials such as carbon dots [9] nanoporous MOFs [10,11] and ZSM-5@ Al-MCM nanocatalysts [12],

noble metal nanoparticles [13–18], and magnetic nanoparticles [19]. Among different nanoparticles, some of them show high enzyme-like activity, for instance, silver nanoparticles [14], Fe_2O_3/Au hybrid nanozyme [20], Fe/Cu single-atom nanozymes [21], manganese dioxide nanoparticles [22], BSA-Au nanoclusters [23,24], and BiOI-NFs [2]. Considering the high enzyme-like activity of these nanozymes, researchers aimed to utilize them as enzyme alternatives to overcome the difficulties of natural enzymes [2,8,23,24]. In this regard, nanozymes had been used for developing different analytical sensing and biosensing methods [8], and organic dye degradation [2,6].

However, the focus of the nanozyme-based research is on developing sensing and biosensing-based methods. In contrast, studies on their potential application toward the enhancement of environmental safety are limited. Hence, developing high throughput nanozyme-based methods for the biodegradation of organic dyes in real water media can be considered as high-impact and applicable systems for the enhancement of environmental safety. Considering the above-mentioned facts, in this contribution, a high throughput nanozymatic method was developed for green nanozyme-mediated degradation of organic dyes in real water media. The protein-protected gold nanozymes were synthesized and characterized for their size, morphology, and peroxidase-like activity. The nanozyme activity was evaluated by standard enzyme-activity assay. Besides, kinetics studies were carried out for the developed nanozymes. Thereafter, the as-prepared nanozymes were utilized for methylene blue degradation in real water media. The effective factors on the dye degradation including pH, ionic strength, degradation time, nanozyme amount, etc. were optimized by the one-factor-at-a-time optimization method. Finally, the method was employed for the nanozyme-mediated degradation of methylene blue from real water media such as pool, river, and tap water samples.

2. Experimental Section

2.1. Materials

H_2O_2, acetic acid, NaCl, $HAuCl_4.4H_2O$, bovine serum albumin (BSA), NaOH, 3,3′,5,5′ tetramethylbenzidine (TMB), and methylene blue (MB) were from Merck Company. Deionized water was supplied by Zolal Teb Shimico (Iran) company.

2.2. Instrumentation

The UV-Visible measurements for the investigation of the peroxidase-like activity of the as-prepared nanozymes as well as for probing the dye degradation were performed by using an Ultrospec 4000 UV-Vis spectrophotometer manufactured by Pharmacia Biotech (Biochrom) Ltd equipped with SWIFT Software. Besides, a Metrohm 827 pH meter equipped with a combined glass electrode and a transmission electron microscope (Zeiss, model EL10C) were utilized for pH measurements and size and morphology evaluation, in order. For exploring more precise on size reporting, the DLS pattern was provided using a Shimadzu particle size analyzer (model: SALD-301V, Japan).

2.3. Green synthesis of gold nanozymes

Protein protected-gold nanozymes were synthesized via a simple, high throughput and green method at physiological temperature via incubation of 10 mL mixed solution (pH=10.5-11.0) of a 1:1 volume ratio of $HAuCl_4.4H_2O$ solution and 50 mg mL^{-1} at 37.0 ℃ for 12 hours. The as-prepared nanozymes were then collected and stored at 4 ℃.

2.4. Dye degradation protocol

The dye degradation activity of the as-prepared nanozymes toward organic dye, methylene blue, was evaluated by quantification of the dye concentration during the nanozyme-mediated reaction. In a typical experiment, 120.0 μL of the as-prepared nanozymes and 100.0 μL of 30.0% H_2O_2 were added to 10.0 mL of 20.0 mg L^{-1} methylene blue solution (pH 4.0-5.0). After 7.0 min, the dye bio-

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degradation process was probed by UV-Visible spectroscopy. The dye concentration was calculated by measuring its absorbance at 661.0 nm before and after incubation with the as-prepared nanozymes and the nanozyme-mediated degradation efficiency was estimated by the following formula;

$$Degradation\ efficiency\ (\%) = \frac{C_0 - C}{C_0} \times 100$$

It is notable that C_0 (mg L^{-1}) and C (mg L^{-1}) are the MB initial and final concentrations, respectively.

3. Results and discussion

3.1. Characterization of as-prepared nanozymes

The as-prepared nanozymes were synthesized via a simple, high throughput and green method at physiological temperature and then characterized for their morphological properties, and size by TEM imaging method, and DLS analysis, in order. The morphological properties and the mean size of the as-prepared nanozymes were investigated by the TEM imaging method. To do this, the TEM image of the as-prepared nanozymes was recorded. The results are shown in Figure 1A, as shown in this figure, the as-prepared nanozymes show a semi-spherical morphology with uniform particles. It is mentionable that, the uniformity of the particles of the as-prepared nanozymes is a significant advantage from an enzymatic point of view because the uniform particles showed higher enzyme-like activity than the particles with low uniformity. Besides, the results showed that the as-prepared nanozymes have a narrow size distribution of 7.7-18.3 nm with a mean size of about 13.2 nm which makes them suitable for enzyme-mimicking applications, considering the fact that the size of nanozymes can strongly affect their enzyme-like activity [24].

For exploring more precise size reporting of the as-prepared nanozymes, the DLS analysis was performed. The results are shown in Figure 1B, as shown in this Figure, the as-prepared nanozymes show a size distribution over 7.3-.31.3 nm with a mean diameter of 13.16 nm. However, the maximum of particles has a size in the range of 10.5-17.2 nm. Besides, the mode of the particles has a size of about 13.0 nm. It is notable that the results of the DLS analysis (mean size of 13.16 nm) are close to those of the TEM imaging method (mean size of 13.2 nm).

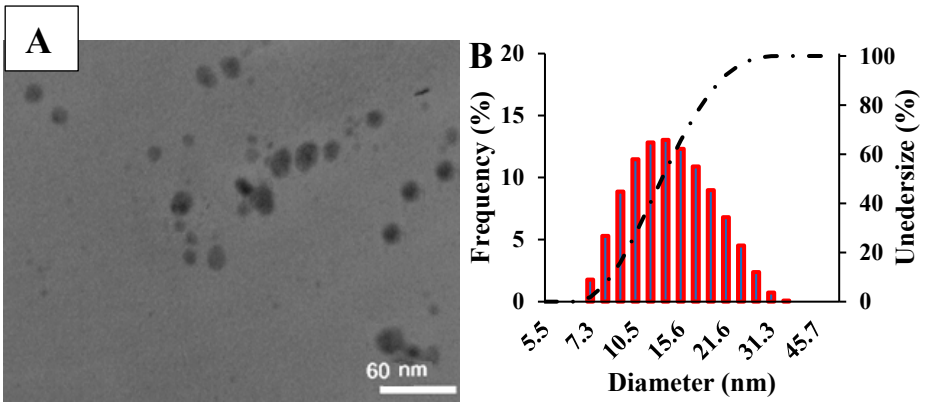

Figure 1. (A) TEM image and (B) DLS pattern of the as-prepared nanozymes.

3.2. Evaluation of nanozymatic properties

The nanozymatic properties of the as-prepared nanozymes including their peroxidase-like activity and their kinetics parameters toward enzyme-mediated oxidation of 3,3′,5,5′-tetramethylbenzidine (TMB) were evaluated to prove the enzyme-like activity and power as well as to estimate the catalytic efficiency of the as-prepared nanazymes.

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3.2.1. Peroxidase-like activity the as-prepared nanozymes

The peroxidase-like (nanozymatic) activity of the as-synthesized nanozymes was investigated toward 3,3′,5,5′-tetramethylbenzidine (TMB) oxidation as the standard peroxidase substrate [8,14]. The activity was evaluated by probing the absorbance of the blue-colored oxidation product (i.e., TMB-ox) at 655 nm based on the standard enzyme assay. It is notable that to prove the oxidation catalysis by the as-prepared nanozymes, the oxidation process was performed in the presence and the absence of the as-prepared nanozymes. The results are shown in Figure 2A, as seen in this figure, the oxidation of TMB to its corresponding blue-colored product cannot efficiently proceed in the absence of the as-prepared nanozymes, revealing very slow kinetics of the oxidation process of TMB by hydrogen peroxide. In contrast by introducing the as-prepared nanozymes into the mixture of TMB and hydrogen peroxide, the oxidation quickly proceeded and a significant absorbance was observed at 658 nm which is related to the oxidation product of TMB. Hence, it can be concluded that the as-prepared nanozymes show characteristic peroxidase-like activity. In fact, the as-synthesized nanozymes can produce active hydroxyl radicals by acting on hydrogen peroxide, as reported [8], then, the produced hydroxyl radical reacts with the TMB molecules and oxidize them to their corresponding cation radicals through a 2-electron reversible oxidation mechanism [8].

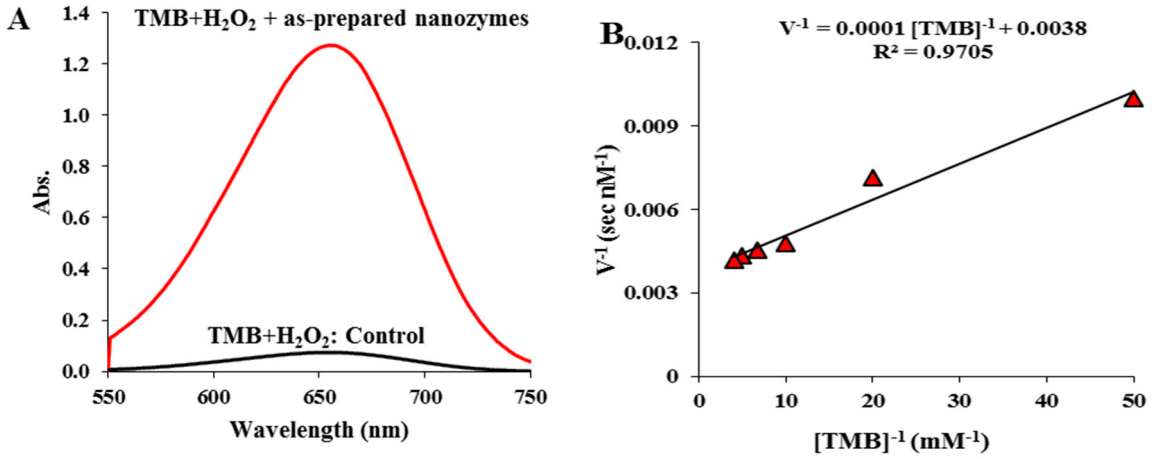

Figure 2. (A) The peroxidase-like activity of the as-prepared nanozymes vs. control sample (TMB+H_2O_2) and (B) Lineweaver–Burk plot for calculating the kinetics parameters of the as-prepared nanozymes.

3.2.2. Kinetics behavior of the as-prepared nanozymes

To explore more precise on the peroxidase-like activity and enzymatic power of the as-prepared nanozymes, the kinetics studies were carried out by estimating the nanozyme activity as a function of TMB concentration and then estimating the nanozymatic kinetics parameters, the standard Lineweaver–Burk plot was provided by plotting the inverse of the velocity of the nanozymatic reaction (V^{-1}) as a function of $[TMB]^{-1}$. It is notable that the velocity of the reaction was calculated based on the estimation of the concertation of the produced TMB-ox per one second of the reaction ($\mu M\ sec^{-1}$), considering this fact that the molar absorption efficient of TMB-ox at 655 nm was 39000 $\mu M^{-1}\ cm^{-1}$. The results are shown in Figure 2B, revealing a V_{max} and a K_m of 0.263 $\mu M\ sec^{-1}$ and 0.03 mM, in order. It is notable that the V_{max} showed the catalytic efficiency of the nanozyme and the K_m showed the affinity of an enzyme to its substrate, as reported. The higher V_{max} and lower K_m are assigned to higher efficiency and affinity, respectively [25–27]. Hence, based on the above considerations, it can be concluded that the as-prepared nanozymes reveal high peroxidase-like activity which makes them suitable for application in biocatalysts.

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3.3. Nanozyme-mediated organic dye degradation

For developing a nanozyme-mediated dye degradation method utilizing the as-prepared nanozymes, methylene blue (20.0 mg L^{-1}) was used as a model organic dye. The nanozyme-mediated dye degradation efficiency was calculated by probing its UV-Visible absorbance at 661 nm and calculating its concentration before and after the reaction. The factors affecting the nanozyme-mediated dye degradation including, pH, degradation time, nanozyme amounts, hydrogen peroxide volume, and ionic strength were optimized to reach the best dye degradation performances. It should be mentioned that the nanozyme-catalyzed degradation efficiency for MB was calculated as high as 99.0% at optimal experimental conditions.

3.3.2. Effect of degradation time

Since the nanozyme-mediated catalytic systems are time-dependent, hence, the effect of the degradation time on the nanozyme-mediated degradation of MB was evaluated over 0-10 min. The UV-Visible spectra of 20.0 mg L^{-1} MB as a function of degradation time are shown in Figure 3A, revealing that the concentration of MB was decreased by increasing the degradation time and reached its minimum concentration after 7 min and after this time, the concertation of MB was not affected by the degradation time. Besides, the plot of degradation yield as a function of time was shown in Figure 3B, as seen, the degradation yield reached its maximum percentage at a degradation time as low as 7 min and then leveled off.

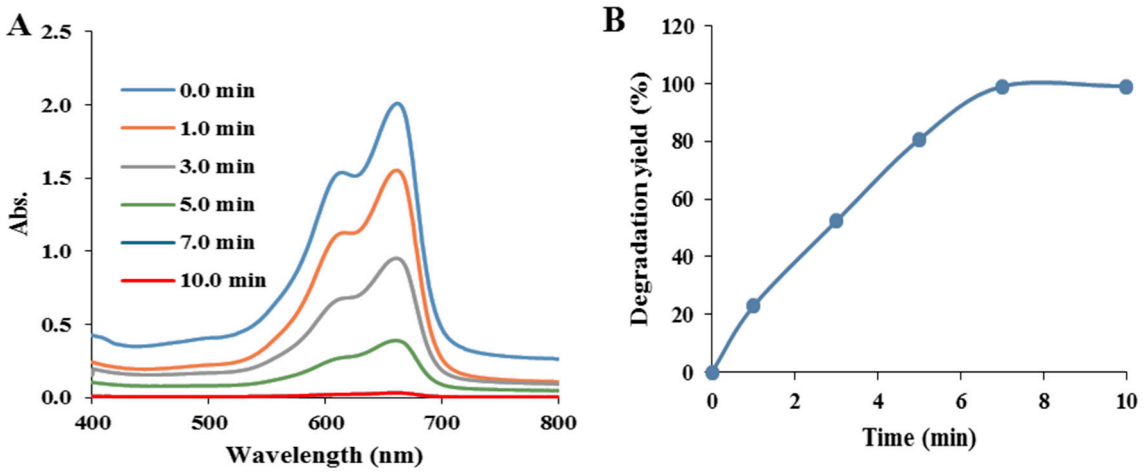

Figure 3. (A) UV-Visible spectra of 20.0 mg L^{-1} MB at different degradation times and (B) the effect of time on the nanozyme-mediated methylene blue degradation.

3.3.1. Effect of pH and ionic strength

The effect of pH on the yield of the nanozyme-mediated degradation of methylene blue as one of the most important and common parameters of the degradation process was investigated by testing a series of MB solutions with different pHs ranging from 2.0 to 8.0. The results shown in Figure 4A revealed that the nanozyme-mediated degradation efficiency reached its maximum value (99.0%) at pH=4.0-5.0 7.0 and then decreased by increasing the pH of the solution. It is maybe related to the degradation of hydrogen peroxide at higher pH values and decreasing the nanozymes activity at pHs higher than pH=4.0-5.0. Hence, pH=4.0-5.0 was selected as the optimal pH for nanozyme-mediated degradation of MB. Besides, the effect of ionic strength on the nanozyme-mediated dye degradation was also checked. To do this the ionic strength of the dye solutions was adjusted to the desired values using NaCl solutions with different concentrations over 1.0-5.0 mM. The results showed that the efficiency of the nanozyme-mediated dye degradation was not significantly changed by the variation of ionic strength (Figure 4B).

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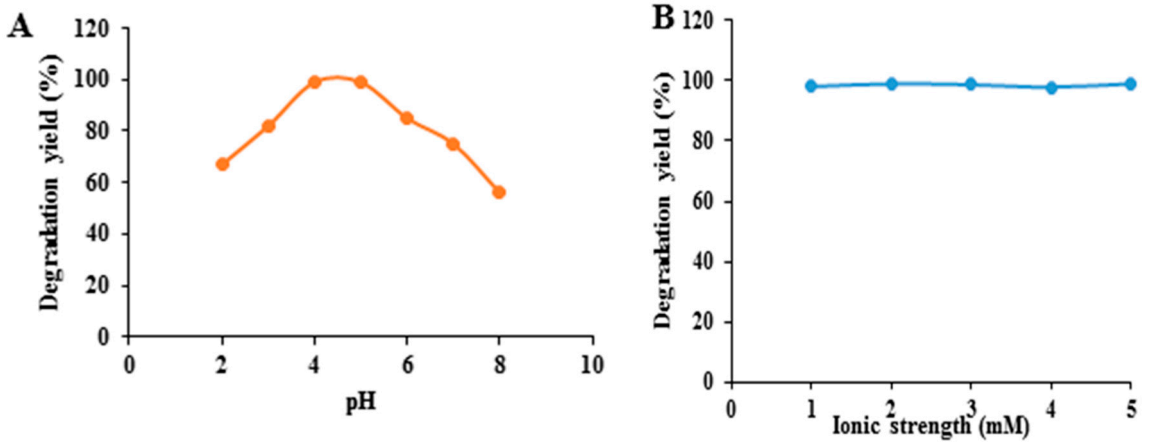

Figure 4. The effect of (A) pH and (B) ionic strength on nanozyme-mediated methylene blue degradation using the as-prepared nanozymes.

3.3.3. Effect of hydrogen peroxide and nanozyme volume

As reported in the literature, the semi-peroxidase nanozymes or peroxidase enzymes act on the hydrogen peroxide and produce active hydroxyl radicals. The produced radicals react with the organic dyes and degraded them to carbon dioxide and water [2,6]. Hence, the amount of hydrogen peroxide is a key factor that significantly affects the nanozyme-mediated degradation of methylene blue dye. Considering this fact, the amount of hydrogen peroxide was optimized by degrading 20.0 mg L^{-1} MB in the presence of different volumes of 30% hydrogen peroxides (Figure 5A). The results revealed that the degradation efficiency reached its maximal value when 100.0 µL H_2O_2 (30%) was used as the oxidant agent. Also, the amount of the as-prepared nanozymes was also optimized by probing the degradation of 20.0 mg L^{-1} MB in the presence of different volumes of the as-prepared nanozymes over 60-160 µL, the results showed that the degradation yield was increased by increasing the nanozyme amount, reached its maximum yield at 120 µL and then leveled off (Figure 5B).

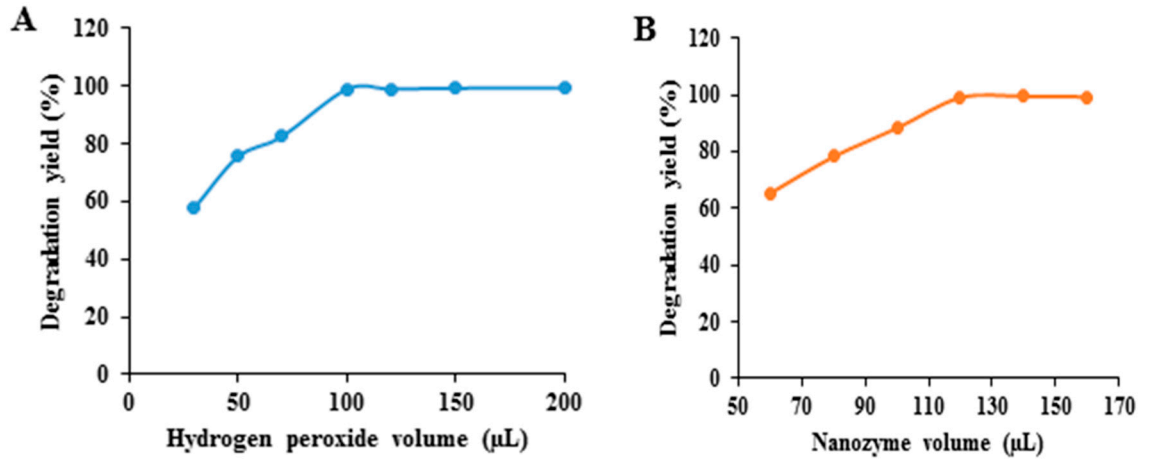

Figure 5. The effect of (A) hydrogen peroxide and (B) nanozyme amount on the nanozyme-mediated methylene blue degradation.

3.5. Practical application

The feasibility of the developed nanozyme-mediated methylene blue degradation for the degradation of this organic dye in the real samples was investigated by probing its degradation in different real water media including tap water, mineral water, pool water, and river water. To do the degradation experiments, 10.0 mg L^{-1} or 20.0 mg L^{-1} MB was initially spiked to each water sample, and the nanozyme-mediated dye degradation system was applied for the dye degradation. The results of this study were shown in Table 1, revealing an excellent biodegradation yield over a minimum value of 94.3% (river water) and a maximum value of 99.0% (tap water) for the different real samples, proving the suitability of the developed method for the dye degradation in real water media.

Table 1. Practical applications of the developed nanozyme-mediated dye degradation system.

Real sample	Spiked MB conc. (mg L^{-1})	Degradation yield (%)	RSD (%, n=3)
Tap water	10.0	99.0	2.7
	20.0	98.4	3.1
Pool water	10.0	97.5	3.5
	20.0	96.4	3.8
Mineral water	10.0	99.0	3.1
	20.0	98.5	3.5
River water	10.0	96.1	4.3
	20.0	94.3	4.6

4. Conclusions

In this work, a high throughput nanozymatic method was developed for green nanozyme-mediated biodegradation of organic dyes in real water media. The nanozymes were synthesized and characterized for their size and morphology by TEM and DLS analysis. The nanozymatic properties of the as-prepared nanomaterials were evaluated by standard enzyme activity assay, revealing high peroxidase-like performances for the as-prepared nanozymes. Therefore, for exploring more precise, the kinetics behavior of the as-prepared nanozymes was evaluated by the standard Lineweaver-Burk method, revealing a V_{max} as high as 0.263 μM sec^{-1} and a K_m as low as 0.03 mM, revealing high catalytic efficiency and affinity of the as-prepared nanozymes. Hence, the as-prepared nanozymes were utilized for organic dye degradation in real water media using methylene blue as a model organic dye. The effective factors on the dye degradation including pH, ionic strength, degradation time, nanozyme amount, etc. were optimized by the one-factor-at-a-time optimization method. At optimal experimental conditions, the as-prepared nanozymes can degrade about 99.0% of the organic dye within a degradation time as low as 7 min. The developed method was finally employed for the nanozyme-mediated degradation of methylene blue from real water media such as pool, mineral, river, and tap water samples. The results of this study revealed an excellent biodegradation yield of over 94.3%-99.0% for the different real samples, proving the suitability of the developed method for dye degradation in real water media.

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Conflict of interest: None.

References

1. Heberer T. Occurrence, fate, and removal of pharmaceutical residues in the aquatic environment: a review of recent research data. Toxicology letters. 2002 May 10;131(1-2):5-17.
2. Ahmadi-Leilakouhi B, Hormozi Jangi SR, Khorshidi A. Introducing a novel photo-induced nanozymatic method for high throughput reusable biodegradation of organic dyes. Chemical Papers. 2023 Feb;77(2):1033-46.
3. Nitin KA, Kishor KS, Chandrakant LP. Integrated chemical and biological processes for the degradation of synthetic dyes. InCurrent Developments in Bioengineering and Biotechnology 2023 Jan 1 (pp. 567-600). Elsevier.
4. Chopra L. Photo-Degradation of dyes and drugs using aloe vera synthesized zinc oxide nanoparticles–A review. Materials Today: Proceedings. 2023 Jan 1;72:1613-7.
5. Yuan Y, Yin W, Huang Y, Feng A, Chen T, Qiao L, Cheng H, Liu W, Li Z, Ding C, Chen F. Intermittent electric field stimulated reduction-oxidation coupled process for enhanced azo dye biodegradation. Chemical Engineering Journal. 2023 Jan 1;451:138732.
6. Hormozi Jangi SR, Davoudli HK, Delshad Y, Hormozi Jangi MR, Hormozi Jangi AR. A novel and reusable multinanozyme system for sensitive and selective quantification of hydrogen peroxide and highly efficient degradation of organic dye. Surfaces and Interfaces. 2020 Dec 1;21:100771.
7. Nanjani DS, Keharia H, Paul D. Deciphering the Shift in Soil Functional and Microbial Diversity Upon Perturbation with Reactive Blue 28 Dye. Available at SSRN 4360527.
8. Hormozi Jangi AR, Hormozi Jangi MR, Hormozi Jangi SR. Detection mechanism and classification of design principles of peroxidase mimic based colorimetric sensors: A brief overview. Chinese Journal of Chemical Engineering. 2020 Jun 1;28(6):1492-503.
9. Jangi SR, Akhond M. Ultrasensitive label-free enantioselective quantification of d-/l-leucine enantiomers with a novel detection mechanism using an ultra-small high-quantum yield N-doped CDs prepared by a novel highly fast solvent-free method. Sensors and Actuators B: Chemical. 2021 Jul 15;339:129901.
10. Jangi SR, Akhond M. Synthesis and characterization of a novel metal-organic framework called nanosized electroactive quasi-coral-340 (NEQC-340) and its application for constructing a reusable nanozyme-based sensor for selective and sensitive glutathione quantification. Microchemical Journal. 2020 Nov 1;158:105328.
11. Hormozi Jangi SR. Synthesis and characterization of magnesium-based metal-organic frameworks and investigating the effect of coordination solvent on their biocompatibility. Chemical Research and Nanomaterials. 2023;1(4):1-9.
12. Hormozi Jangi SR. Low-temperature destructive hydrodechlorination of long-chain chlorinated paraffins to diesel and gasoline range hydrocarbons over a novel low-cost reusable ZSM-5@ Al-MCM nanocatalyst: a new approach toward reuse instead of common mineralization. Chemical Papers. 2023 Apr 21:1-5.
13. Hormozi Jangi SR, Akhond M. High throughput green reduction of tris (p-nitrophenyl) amine at ambient temperature over homogenous AgNPs as H-transfer catalyst. Journal of Chemical Sciences. 2020 Dec;132:1-8.
14. Hormozi Jangi, SR, Dehghani Z. Spectrophotometric quantification of hydrogen peroxide utilizing silver nanozyme. *Chemical Research and Nanomaterials.* https://crn.shiraz.iau.ir/article_701960.html?lang=en. *2023*
15. Hormozi Jangi SR. Effect of daylight and air oxygen on nanozymatic activity of unmodified silver nanoparticles: Shelf-stability. Qeios 2023, doi:10.32388/9RWVYI.3.
16. Hormozi Jangi SR. A comparative study on kinetics performances of BSA-gold nanozymes for nanozyme-mediated oxidation of 3,3′,5,5′-tetramethylbenzidine and 3,3′-diaminobenzidine. Preprints.org 2023, 2023060387. https://doi.org/10.20944/preprints202306.0387.v1
17. Hormozi Jangi SR. Evaluating the Effect of Shelf-Storage, Daylight, and Air Oxygen on the Peroxidase-like Activity of Unmodified Silver Nanoparticles. Preprints.org 2023, 2023052106. https://doi.org/10.20944/preprints202305.2106.v1
18. Hormozi Jangi SR. Determining kinetics parameters of bovine serum albumin-protected gold nanozymes toward different substrates. Qeios. 2023 doi:10.32388/07MWBC.
19. Chen H, Ma X, Zhang X, Hu G, Deng Y, Li S, Chen Z, He N, Wu Y, Jiang Z. Novel aerosol detection platform for SARS-CoV-2: Based on specific magnetic nanoparticles adsorption sampling and digital droplet PCR detection. Chinese Chemical Letters. 2023 Jan 1;34(1):107701.
20. Zeng X, Ruan Y, Chen Q, Yan S, Huang W. Biocatalytic cascade in tumor microenvironment with a Fe2O3/Au hybrid nanozyme for synergistic treatment of triple negative breast cancer. Chemical Engineering Journal. 2023 Jan 15;452:138422.
21. Chen X, Wang Y, Feng M, Deng D, Xie X, Deng C, Khattak KN, Yang X. Dual-active-site Fe/Cu single-atom nanozymes with multifunctional specific peroxidase-like properties for S2− detection and dye degradation. Chinese Chemical Letters. 2023 Jun 1;34(6):107969.
22. Hormozi Jangi SR, Akhond M, Absalan G. A field-applicable colorimetric assay for notorious explosive triacetone triperoxide through nanozyme-catalyzed irreversible oxidation of 3, 3′-diaminobenzidine. Microchimica Acta. 2020 Aug;187:1-0.

23. Akhond M, Hormozi Jangi SR, Barzegar S, Absalan G. Introducing a nanozyme-based sensor for selective and sensitive detection of mercury (II) using its inhibiting effect on production of an indamine polymer through a stable n-electron irreversible system. Chemical Papers. 2020 Apr;74:1321-30.
24. Hormozi Jangi SR, Akhond M, Absalan G. A novel selective and sensitive multinanozyme colorimetric method for glutathione detection by using an indamine polymer. Analytica Chimica Acta. 2020 Aug 29;1127:1-8.
25. Hormozi Jangi SR, Akhond M. Introducing a covalent thiol-based protected immobilized acetylcholinesterase with enhanced enzymatic performances for biosynthesis of esters. Process Biochemistry. 2022 Sep 1;120:138-55.
26. Hormozi Jangi SR, Akhond M, Dehghani Z. High throughput covalent immobilization process for improvement of shelf-life, operational cycles, relative activity in organic media and enzymatic kinetics of urease and its application for urea removal from water samples. Process Biochemistry. 2020 Mar 1;90:102-12.
27. Hormozi Jangi SR, Akhond M. High throughput urease immobilization onto a new metal-organic framework called nanosized electroactive quasi-coral-340 (NEQC-340) for water treatment and safe blood cleaning. Process Biochemistry. 2021 Jun 1;105:79-90.