

Review of: "Determining kinetics parameters of bovine serum albumin-protected gold nanozymes toward different substrates"

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Potential competing interests: No potential competing interests to declare.

The manuscript presented "Determining kinetics parameters of bovine serum albuminprotected gold nanozymes toward different substrates."

Before the paper could be accepted for publication, the authors need make**Major revisions** as follow:

Abstract

The sentence "The kinetic parameter, Vmax and Km was then calculated by using Michaelis–Menten and the linear plot of Lineweaver–Burk for both substrates" contains a subject-verb agreement error. It should be revised to "The kinetic parameters, Vmax, and Km were then calculated by using Michaelis–Menten and the linear plot of Lineweaver–Burk for both substrates."

The sentence "Besides, the Km value for DAB was found to be very higher than that for TMB" contains a grammatical error. "Very higher" is not the appropriate phrase to use. It should be revised to "Furthermore, the Km value for DAB was found to be significantly higher than that for TMB."

The sentence "This difference can be related to the different reactivity of DAB and TMB. as well as their different oxidation mechanism" contains a capitalization error and a missing punctuation mark. It should be revised to "This difference can be related to the different reactivity of DAB and TMB, as well as their different oxidation mechanisms."

Overall, the text contains some minor scientific errors and a few grammatical issues, but there are no major problems.

Introduction

The sentence "The enzyme immobilization permits. possible increase in stability (pH, thermal, storage, and solvent performance), easy to recover and handle via simple enzyme separation from the mixtures of reactants and products." contains a period (.) after the word "permits" which seems to be a typo. It should be revised to "The enzyme immobilization permits a possible increase in stability (pH, thermal, storage, and solvent performance) and easy recovery and handling via simple enzyme separation from the mixtures of reactants and products."

The sentence "However, the affinity of different substrates for binding to nanozyme active notes is different from each

other, leading to different kinetic performances of a nanozyme toward different substrates." contains a possible typographical error. The phrase "active notes" should likely be "active sites." The sentence can be revised to "However, the affinity of different substrates for binding to nanozyme active sites differs from each other, leading to different kinetic performances of a nanozyme toward different substrates."

Overall, the text is well-written with only a minor possible typographical error that can be easily corrected.

Materials And Methods

The sentence "All materials were obtained from Merck Company except DAB which provides by Sigma." contains a grammatical error and a spelling mistake. It should be corrected to "All materials were obtained from Merck Company except DAB, which was provided by Sigma."

In the first paragraph, the sentence "To do synthesis the BSA-protected nanozymes" contains a grammatical error and should be corrected to "To synthesize the BSA-protected nanozymes".

In the second paragraph, the sentence "Regarding the TMB assay, 40 μ L hydrogen peroxide solution (final concentrations of 0.24 M), 200 μ L of TMB (different concentrations), and 80 μ L of BSA-gold nanozymes were introduced to 1.3 mL of acetate buffer (0.3 M; pH, 0.4)" contains a typographical error. The pH value should be specified in a numerical format, and it seems to be incorrect. It should be corrected to "1.3 mL of acetate buffer (0.3 M; pH 4.0)".

In the same sentence, the abbreviation "TMB-ox" is used without prior explanation. It would be helpful to define the abbreviation or use the full name "TMB oxidation" for clarity.

In the paragraph discussing the DAB assay, the sentence "80.0 μ L BSA- gold nanozymes" contains a space between "BSA-" and "gold nanozymes". It should be corrected to "80.0 μ L BSA-gold nanozymes".

In the same sentence, the phrase "thoroughly mixed at ambient temperature" could be rephrased for clarity. For example, "thoroughly mixed by gentle agitation at room temperature".

Overall, there are a few minor errors in the text, including grammatical errors, typographical errors, and clarity issues. These can be easily corrected or clarified for better understanding.

Results

The sentence "The kinetic parameter, Vmax and Km was then calculated" contains a subject-verb agreement error. It should be corrected to "The kinetic parameters, Vmax and Km, were then calculated".

The phrase "by using Michaelis–Menten and the linear plot of Lineweaver–Burk" should be revised for clarity. The Michaelis–Menten equation is used to determine the kinetic parameters, while the Lineweaver–Burk plot is a graphical representation of the Michaelis–Menten equation. You can rephrase it as "by using the Michaelis–Menten equation and analyzing the data using the Lineweaver–Burk plot."

The sentence "The Michaelis–Menten plot for oxidation of TMB catalyzed by BSA-gold nanozymes was shown in Figure

2A, revealing that the rate of nanozyme-mediated oxidation of TMB was increased by increasing the substrate concentration and then leveled off" contains a typo. "Concentration" is misspelled as "concertation". It should be corrected to "substrate concentration".

In the sentence "Besides, to evaluate the kinetics performances of the as-prepared BSA-gold nanozymes for DAB oxidation, the Michaelis–Menten plot for DAB oxidation by hydrogen peroxide in the presence of the BSA-gold nanozymes as the peroxidase-mimicking agents were obtained, and the results are shown in Figure 2B, revealed that the rate of nanozyme-mediated DAB oxidation was increased by increasing the substrate concertation and then leveled off which is same of the TMB oxidation but the TMB oxidation rate was found to be higher than that of the DAB oxidation by BSA-gold nanozymes." there are a few errors and improvements that can be made:

The phrase "to evaluate the kinetics performances" should be corrected to "to evaluate the kinetic performance".

The phrase "the Michaelis–Menten plot for DAB oxidation by hydrogen peroxide in the presence of the BSA-gold nanozymes as the peroxidase-mimicking agents were obtained" contains a subject-verb agreement error. It should be corrected to "the Michaelis–Menten plot for DAB oxidation by hydrogen peroxide in the presence of the BSA-gold nanozymes as the peroxidase-mimicking agents was obtained".

The sentence is quite long and could benefit from being broken down into smaller sentences for better readability.

The phrase "To explore more precise the kinetic performances" contains a grammatical error. It should be rephrased to "To explore the kinetic performances more precisely".

The phrase "for estimation of K_m and V_{max} of BSA-gold enzymes-mediated oxidation of DAB" should be revised for clarity. It should be corrected to "to estimate the K_m and V_{max} of BSA-gold nanozymes in the oxidation of DAB".

In the sentence "revealing a V_{max} and K_m of 185 nM sec⁻¹ and 0.72 mM, in order, for DAB oxidation", the term "in order" seems to be out of place and should be removed. The sentence can be revised to "revealing a V_{max} of 185 nM sec⁻¹ and a K_m of 0.72 mM for DAB oxidation".

In the sentence "To estimate the kinetic parameters of BSA-gold nanozymes toward TMB oxidation, the Lineweaver–Burk plot was constructed (Figure 3B). Based on Figure 3B, the V_{max} and K_m of the as-prepared nanozymes toward TMB oxidation were calculated at about 263 nM sec⁻¹ and 0.03 mM, in order.", the term "in order" seems to be unnecessary and can be removed. The sentence can be revised to "Based on Figure 3B, the V_{max} of the as-prepared nanozymes for TMB oxidation was calculated to be about 263 nM sec⁻¹, and the K_m was approximately 0.03 mM."

The sentence "The V_{max} of DAB oxidation was found to be lower than that of the TMB oxidation, revealing that the catalytic efficiency of the as-prepared BSA-gold nanozymes toward TMB is higher than that for the DAB" is grammatically correct but could be improved for clarity. Consider revising it to "The V_{max} of DAB oxidation was lower than that of TMB oxidation, indicating that the catalytic efficiency of the as-prepared BSA-gold nanozymes is higher for TMB than for DAB."

The sentence "The K_m value for DAB was found to be very higher than that for TMB, revealing a higher affinity of TMB for

binding to BSA-protected gold nanozymes than DAB" contains a grammatical error. "Very higher" is not appropriate. It should be revised to "The K_m value for DAB was found to be significantly higher than that for TMB, indicating a higher affinity of TMB for binding to BSA-protected gold nanozymes than DAB."

The phrase "This difference can be related to the different reactivity of DAB and TMB as well as their different oxidation mechanisms" is scientifically accurate, but it could be rephrased for better clarity. Consider revising it to "This difference in kinetic parameters can be attributed to the distinct reactivity of DAB and TMB, as well as their different oxidation mechanisms."

Conclusions

The phrase "by using Michaelis–Menten and the linear plot of Lineweaver–Burk" should be clarified to "by using the Michaelis–Menten equation and constructing the Lineweaver–Burk plot."

The sentence "the V_{max} of DAB oxidation was found to be lower than that of the TMB oxidation which pointed to the fact that the catalytic efficiency of the as-prepared BSA-gold nanozymes toward TMB is significantly higher than their efficiency for the DAB" is scientifically accurate, but it could be improved for clarity. Consider revising it to "The V_{max} for DAB oxidation was found to be lower than that for TMB oxidation, indicating that the catalytic efficiency of the as-prepared BSA-gold nanozymes is significantly higher for TMB than for DAB."

The phrase "Besides, the K_m value for DAB was found to be very higher than that for TMB" contains a grammatical error. "Very higher" is not appropriate. It should be revised to "Moreover, the K_m value for DAB was significantly higher than that for TMB."

The sentence "This difference can be related to the different reactivity of DAB and TMB. as well as their different oxidation mechanisms." is scientifically accurate, but it could be rephrased for better clarity. Consider revising it to "This difference in the kinetic parameters can be attributed to the distinct reactivity and oxidation mechanisms of DAB and TMB."

When the study is analysed in general, the results obtained are quite remarkable for the scientific community.

As an opinion, the study is suitable for publication in the Qeios. It would be appropriate to make the indicated corrections before publication.