

Fabrication of Nitrogen-containing Nanocarbons for the Detection of Hydrogen Peroxide Utilizing Layered Silicate Magadiite as a Template

Zhaoming Liu^{1*}, Shengying Wang¹, Mingliang Ge²

¹Key Laboratory of Polymer Processing Engineering of Ministry of Education, National Engineering Research Center of Novel Equipment for Polymer Processing, School of Mechanical & Automotive Engineering, South China University of Technology, Guangzhou 510641, China

²Key Laboratory of Polymer Processing Engineering Ministry of Education, Key Laboratory of Technique and Equipment for Macromolecular Advanced Manufacturing, South China University of Technology, Guangzhou 510640, China

*Corresponding author: melzming@mail.scut.edu.cn

Received March 04, 2023; Revised April 08, 2023; Accepted April 17, 2023

Abstract In search of novel artificial mimic enzymes with catalytic properties, new nitrogen-containing carbon nanomaterials were designed and fabricated using polypyrrole as a precursor and magadiite, a naturally occurring layered clay, as a template. X-ray diffraction spectroscopy (XRD), Fourier infrared transform spectroscopy (FTIR), X-ray photoelectron spectroscopy (XPS), and field emission scanning electron microscopy (FESEM) were used to describe the structure, morphology, and functional components of carbon nanomaterials. The reducing substrate used was 3,3',5,5'-tetramethylbenzidine (TMB), which could be oxidized and discolored by the nanozyme in the presence of H₂O₂. Initial simulated enzymatic activity probes demonstrated that the nanocarbon material containing nitrogen had a strong affinity for the chromogenic substrates TMB and hydrogen peroxide. The Michaelis constant for the substrate TMB is 0.452 mM, which is comparable to that of the natural peroxidase HRP, whereas the Michaelis constant for the substrate hydrogen peroxide is 1.89 mM, which is approximately 50% lower than that of HRP, indicating that its affinity for the substrate H₂O₂ is greater than that of HRP. The nitrogen-containing carbon nanomaterials' enzyme-like activity maintained high catalytic activity in extreme conditions such as strong acid and high temperature, giving them the potential for widespread use and stable storage.

Keywords: nitrogen-containing nanocarbon, magadiite, peroxidase, H₂O₂

Cite This Article: Zhaoming Liu, Shengying Wang, and Mingliang Ge, "Fabrication of Nitrogen-containing Nanocarbons for the Detection of Hydrogen Peroxide Utilizing Layered Silicate Magadiite as a Template." *Journal of Materials Physics and Chemistry*, vol. 11, no. 1 (2023): 22-28. doi: 10.12691/jmpc-11-1-3.

1. Introduction

In 2004, Scrimin and Pasquato [1] identified the exceptional catalytic characteristics of gold-zinc nanoclusters, and based on these findings, the notion of "nanozyme", a material with both nanomaterial and mimicking enzyme properties [2], was developed for the first time. Due to the rapid growth of nanotechnology, scientists have created a vast array of nanozymes with a variety of natural enzyme capabilities. Traditional biological enzymes lack the benefits of high stability, simple storage, controlled activity, and low cost. Nevertheless, synthetic enzymes provide these qualities. Utilizing nanozymes, numerous chemical sensors [3] may be fabricated. These sensors have been utilized in the detection of food [4], monitoring of the environment [5], and treatment of disease [6]. Currently, the creation and application of nanozymes are expanding their boundaries, and research into their underlying mechanisms is advancing.

Being an essential subfield within the study of nanozyme, carbon-based nanozyme have garnered the interest of numerous scientists in recent years. From the zero-dimensional fullerene [7] and carbon quantum dot nanozyme [8] to the two-dimensional graphene nanozyme [9] and the three-dimensional mesh-like structure of carbon-based nanozyme [10], carbon-based nanozyme with a variety of morphological structures exhibit outstanding characteristics including a variety of simulated enzymatic activities, simple preparation, simple functionalization, low cost, and high enzymatic activity. Doping carbon materials [11] improves their electrocatalytic activity in the field of electrochemistry, and the addition of heterogeneous elements disrupts the normal arrangement of carbon elements and offers more active sites on the material surface. In the field of nanozyme, nitrogen-doped carbon materials have comparable improved simulated enzymatic activity. Calculations based on the density flooding theory (DFT) indicate that the addition of nitrogen increases the number of catalytic reactive sites surrounding peroxides [12], hence increasing the

generation of free radicals that attack peroxides. However, the drawback of graphene and carbon nanotubes, which are frequently reported in research [13,14], is that nanoparticles tend to aggregate, reducing the contact between the active site and the substrate to be tested, which is not conducive to the catalytic reaction. This issue must be resolved. As an ideal substitute for natural enzymes, nitrogen-containing carbon compounds offer superior catalytic activity, stability, and absence of metal contamination [15]. In prior research [16], we discovered that carbon nanomaterials containing nitrogen had a significant specific surface area and may create superoxide radicals and breakdown tetracycline in the presence of water. This shows that carbon nanostructures containing nitrogen have the potential to facilitate the catalytic reduction of substrates.

In this study, polypyrrole was utilized as a precursor to incorporate layered silicate magadiite into the structure in order to discover efficient nanozyme mimics, which were inspired by the theories presented above. Utilizing XRD, FTIR, XPS, and FESEM, novel nitrogen-containing carbon nanomaterials with a high specific surface area were developed, fabricated, and characterized. By observing the color shift of the chromogenic substrate TMB, its imitated enzymatic activity was demonstrated. The preliminary simulated enzyme activity probes revealed that the nitrogen-containing carbon nanomaterials had a higher affinity for the chromogenic substrates TMB and hydrogen peroxide than the natural enzyme HRP, and its maximum reaction rates of TMB and H_2O_2 were 2.52 and 3.05 times that of HRP, respectively, indicating that the nitrogen-containing carbon nanomaterials prepared in this study possessed a greater peroxidase-like activity. In addition, it has excellent sensitivity to the detection limit of hydrogen peroxide. This work gives a specific foundation and direction for future research.

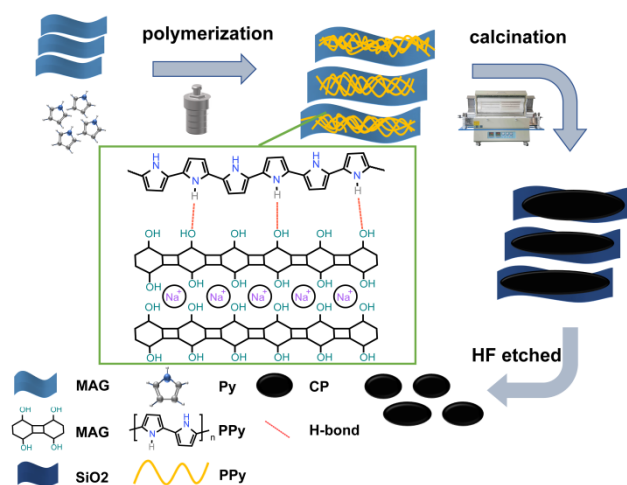


Figure 1. Synthesis route of nitrogen-containing carbon nanomaterials

2. Materials and Methods

2.1. Chemicals and Instrumentation

All solvents and reagents were purchased from commercial suppliers. The generated nitrogen-containing nanocarbon material was designated as CP-X ($X=0,1,2,3$),

CP-0 is the nitrogen-containing nanocarbon material without a magadiite template, and MP was designated as the precursor. For the measurement of the simulated enzyme activity, a 0.2 M buffer solution of acetic acid and sodium acetate (HOAc-NaOAc) at a pH of 4.0 was utilized. Nicolet 2000 UV-Vis spectrophotometer (Nicolet, USA) was used to collect the UV spectral data of the solutions.

Field emission scanning electron microscopy (FESEM) images were recorded and analyzed by Nova Nano SEM 430 (FEI, USA). Energy dispersive X-ray spectroscopy (EDS) was measured with an X-MaxN20 dual detector system (Oxford, UK). Fourier transform infrared (FT-IR) spectra were tested by a NEXUS 670 (Nicolet, USA). X-ray photoelectron spectra (XPS) were collected by a Kratos Axis Ultra DLD X-ray photoelectron spectrometer with an Al $K\alpha$ achromatic X-ray source (Kratos, UK).

2.2. Evaluation of Peroxidase-Like Activity of Nitrogen-Containing Nanocarbons (CPs)

As the solution environment for the assessment of the simulated enzyme activity, a 0.2 M HAc-NaAc buffer solution (pH=4.0) was used. Several forms of nanocarbon materials containing nitrogen (CP-0, CP-1, CP-2, and CP-3) and ultrasonically dispersed, homogenous 1 mg/mL water solutions of the four CPs were used. The peroxidase-like activity was evaluated based on the findings of H_2O_2 's catalytic oxidation of TMB. In a typical technique, the catalytic reaction was conducted in 20 mM TMB and 50 mM H_2O_2 in HAc-NaAc buffer (0.2 M, pH 4.0). The total volume of the reaction was 1.5 mL. The absorption spectra were measured using an ultraviolet spectrophotometer between 500 and 800 nm. Following a time of reaction, the solution was withdrawn to evaluate the color change, and photographs were taken to identify the nitrogen-containing porous nanocarbon material with the greatest simulated enzyme activity under the same circumstances.

2.3. CP-based Colorimetric Detection of Hydrogen Peroxide

By fixing the CP and TMB concentrations, a specific ratio exists between the catalytic reaction rate and H_2O_2 concentration, from which the H_2O_2 concentration in the system can be calculated based on the absorbance of oxidized TMB (TMB_{ox}) at 652 nm, and this ratio can be used to detect the H_2O_2 content in the system, which is essential for disease detection and environmental testing. This proportional relationship was determined by preparing 25 mM, 100 mM, and 500 mM H_2O_2 solutions and sequentially adding buffer solution, TMB-ethanol solution, and CP solution to 14 centrifuge tubes based on the optimal enzyme activity conditions determined by screening. A specified amount of H_2O_2 solution was introduced separately to create a gradient of low to high final H_2O_2 concentration in the system (0-10mM). After 5 minutes of reaction, the absorbance at 652 nm was measured using a UV-Vis spectrophotometer. The experiment was repeated three times for each hydrogen peroxide concentration, and the mean absorbance was recorded. Using absorbance data, the absorbance- H_2O_2 concentration relationship curve was constructed, and fit

analysis was done to determine the H_2O_2 concentration detection range.

3. Results and Discussion

3.1. Characterization of Nitrogen-Containing Nanocarbons(CPs)

Copies of the spectra of the characterization data and the target products have been included in the supplementary materials.

3.2. Optimization of Synthesis Routine

3.2.1. The Influence of Temperature on the Structure of Precursors MP

The system temperature during the polymerization process is one of the major parameters influencing the shape of polypyrrole. The FTIR spectra of the nitrogen-containing carbon material precursors generated in an ice bath (0°C) and at room temperature (25°C) are shown in Figure 2. The MP precursors synthesized in the ice bath exhibit N-H stretching vibration peaks at approximately 3442 cm^{-1} , the absorption peak at 1540 cm^{-1} corresponds to the C=C bond stretching vibration on the pyrrole ring, and the absorption peak at 1460 cm^{-1} corresponds to the C-N stretching vibration on the pyrrole ring [17]. At 25°C , the broad peak at 1700 cm^{-1} - 1800 cm^{-1} was slightly shifted to the right and lost intensity, whereas the characteristic N-H absorption peak of PPy at 3442 cm^{-1} was enhanced, indicating that a portion of the PPy molecule chain did not completely intercalate into the template MAG interlayer, and more Py completed the oxidative polymerization without entering the template compared to the compounding process in the ice bath.

Figure 3 depicts the SEM images of the MP precursors synthesized at various temperatures, revealing that the MP precursor synthesized at 25°C has poor regularity, the polypyrrole particles have uneven diameters, there are fewer lamellar template agents visible in the photographs, and the polypyrrole particles are aggregated into clusters. In contrast, the lamellar MAG template is observable in the SEM picture of the MP precursor produced in a cold bath, and the polypyrrole particles have a more uniform shape and are mostly bound to the lamellae of the template agent.

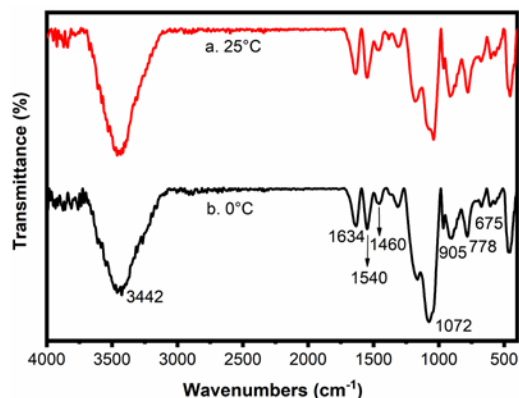


Figure 2. XRD images of MP precursor synthesized at different temperatures

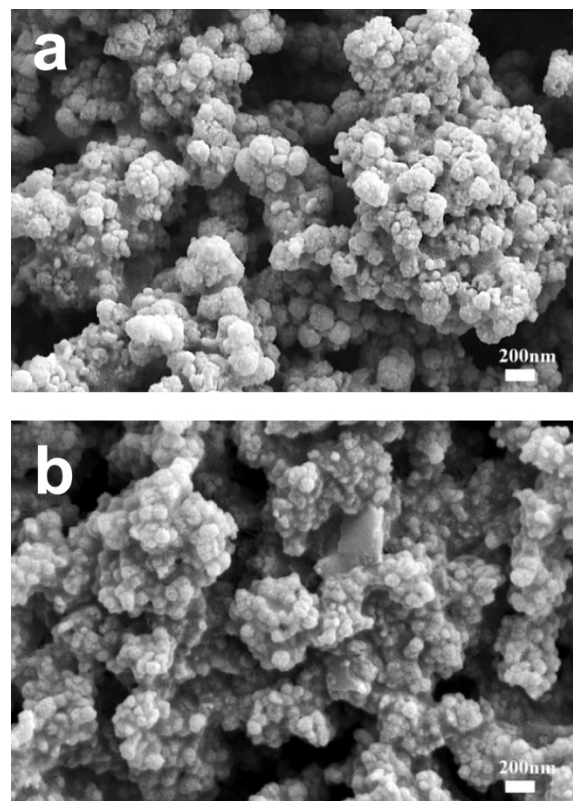


Figure 3. SEM images of MP precursor synthesized at different temperatures a) 25°C ; b) 0°C

3.2.2. Effects of Polymerized Gases on the Structure of Precursors MP

The precursors of MP produced with and without nitrogen protection are shown in Figure 4, respectively. In comparison to spectral line a, the relative intensities of the N-H stretching vibration absorption peak at 3459 cm^{-1} , the C=C stretching vibration absorption peak of the pyrrole ring, and the C-N stretching vibration absorption peak at 1169 cm^{-1} are all increased. These peaks in absorption are all classic absorption peaks for polypyrrole. Due to its high activity, it is possible that the pyrrole monomer is especially vulnerable to self-polymerization in the presence of oxygen. Thus, there are a considerable number of PPy molecular chains in the composite system that have not intercalated into the MAG template, which is represented in the IR spectrogram by the increased relative strength of the PPy absorption peaks.

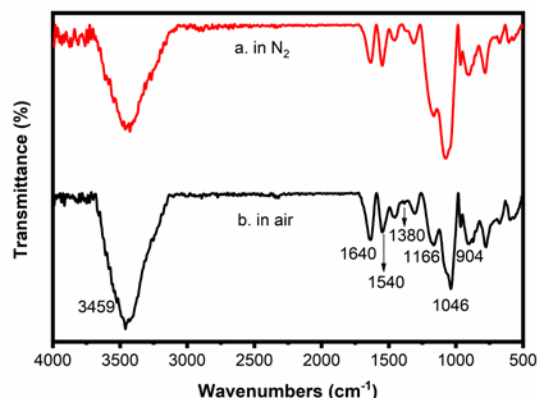


Figure 4. FTIR diagram of the precursor a) synthesized in a nitrogen atmosphere and the precursor b) synthesized in air

3.2.3. Effect of Polymerization Time on the Structure of Precursors MP

The preparation of MP precursors was conducted at a constant reaction temperature of 0°C and under nitrogen protection, and the FTIR characterization of the final products was performed at various reaction times; the results are depicted in Figure 5. During the reaction, it was observed with the naked eye that approximately 30 minutes after the addition of the oxidant, the formerly milky-white mixed solution changed to dark green, then to black, and continued to darken as the reaction time was extended. With the prolongation of the reaction time, the relative intensity of the C-H stretching vibration absorption peak at 1640 cm^{-1} gradually increases, whereas the C=C stretching vibration peak at 1552 cm^{-1} gradually decreases, and the broad absorption peak of MAG near 1072 cm^{-1} is gradually flattened by the C-N stretching on the polypyrrole ring vibration peak at 1170 cm^{-1} [18]. This suggests that the rate of polymerization of polypyrrole rises with reaction time and that a high number of molecular chains penetrate between the lamellae of the MAG template. The FTIR images of the MP precursors after 12 h and 18 h of reaction have virtually identical peak shapes, indicating that the polymerization reaction is essentially complete after 12 h.

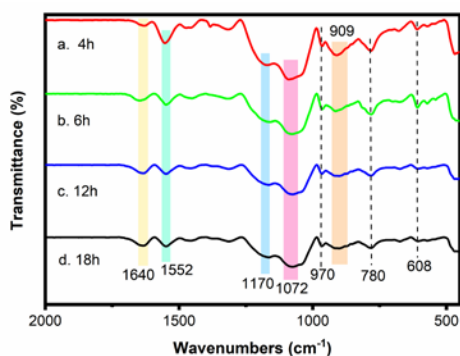


Figure 5. The effect of reaction time on the structure of the precursor: FTIR diagram of the precursor after reacting for 4h, 6h, 12h, 18h

3.2.4. Effect of Calcination Temperature on the Structure of Nitrogen-Containing Nanocarbon Materials

Before calcination, the precursors of nitrogen-doped nanocarbon materials undergo a thermogravimetric (TG) test analysis in nitrogen to aid the selection of an appropriate carbonization temperature (Figure 6a). The precursor experiences around three phases of weight loss at temperatures between 0 and 800 °C. After the TG curve was produced, three heat absorption temperatures were obtained. At 68.9°C, desorption of physically adsorbed water molecules occurred in all three samples, resulting in a mass loss of 3.86, 4.84, and 5.74 percent, respectively. Consequently, a greater proportion of weight was lost between 200°C and 400°C and beyond 400°C. When the temperature rises, the rate of molecular chain breakdown and the degree of carbonation both increase progressively. After the commencement of PPy carbonization, the precursors were selected to be calcined at 400°C, 600°C, and 800°C.

After calcination and etching, the nitrogen-doped nanocarbon materials were analyzed using energy spectroscopy (EDS) to determine the elemental distribution of the products, and the findings are shown in Figure 6b. Comparing the three nitrogen-doped carbon nanomaterials at various calcination temperatures, we discovered that their elemental compositions follow the pattern C>N>O>Si, and the Si concentration is below 0.2%, which is insignificant. The carbon-to-nitrogen ratio of nitrogen-doped carbon nanomaterials calcined at 400°C is close to 5:1, which is compatible with polypyrrole's structural makeup. Polypyrrole's N loss rapidly increased as the calcination temperature rose. The study of the rise in the ratio of carbon to nitrogen may be attributable to the increase in calcination temperature and the conversion of N atoms in the five-membered ring of polypyrrole into other small molecule compounds lost during the chain-breaking breakdown of molecular chains. It was discovered that as the temperature in the tube rose, white powder condensed at the lower ends of the tube furnace temperature.

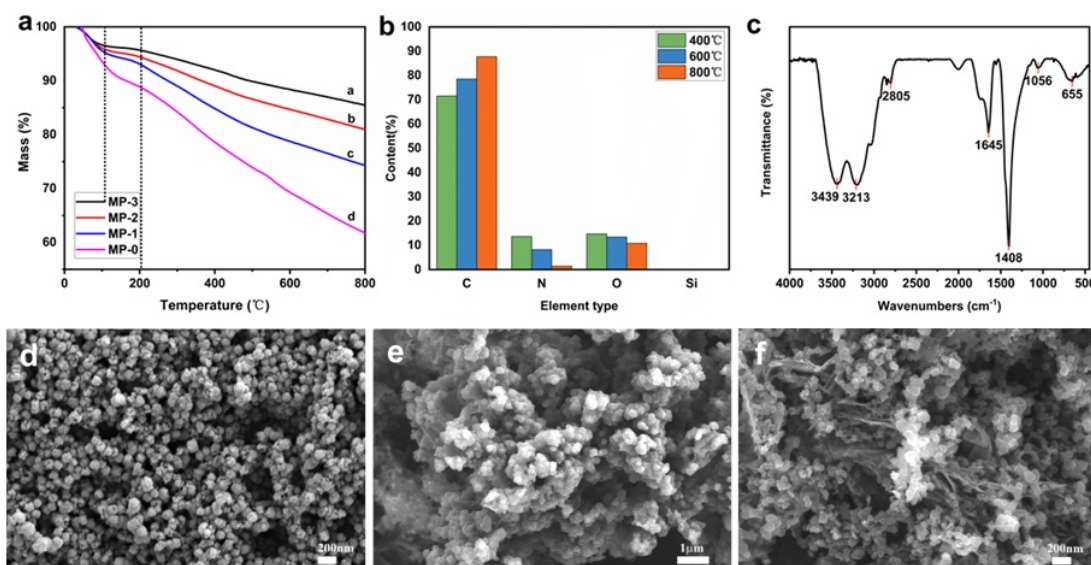


Figure 6. a) TG curve of the precursor of nitrogen-doped porous carbon material: a.MP-3, b.MP-2, c.MP-1, d.MP-0; b) Distribution image of C, N, O, Si elements of nitrogen-doped porous carbon material MP; c) FTIR diagram of decomposition products of calcined MP precursor; SEM images of CP nitrogen-doped carbon nanomaterials with calcination temperature of d) 400°C e) 600°C f) 800°C

As shown in Figure 6c, the white particles were analyzed by infrared spectroscopy (FTIR) as a mixture of benzamide and phenol, all of which were generated during the pyrolysis of the laboratory-prepared MP precursors.

The SEM in Figure 6 reveals that when the calcination temperature is 400°C, the nitrogen-doped carbon material is in the form of particles with a particle size between 20-40 nm and a relatively uniform particle size distribution; when the calcination temperature is 600°C, the morphology is no longer entirely composed of particles, and many flake-like structures appear; with further increase in temperature, when the calcination temperature is 800°C, CP nitrogen-doped nanocarbon emerges as an ultrathin sheet structure like graphene that joins the remaining particles to create a three-dimensional mesh. The findings indicate that the calcination temperature has a significant effect on the microstructure of the nitrogen-doped nanocarbon material. When the temperature rises, the degree of carbonization increases and the structure of the material transforms from granular to graphene-like. To produce a greater degree of nitrogen doping and N element protection, the calcination temperature was ultimately set at 400°C.

Table 1. Comparison of element content of various nitrogen-doped carbon materials

Sample name	C (at%)	O (at%)	N (at%)	Si (at%)
CP-400°C	71.53	14.67	13.61	0.19
CP-600°C	78.54	14.4	7.22	0.2
CP-800°C	87.75	10.84	1.35	0.06
NG@H-MMT [19]	88.6	7.2	4.2	—
CDs [20]	97.37	-	2.63	—
FN-CNF [21]	73.82	9.25	13.93	—

3.3. Evaluation of Enzyme Activity and Adaptive Conditions

In order to determine if the nitrogen-containing nanocarbon material CP has simulated enzymatic activity, the type of simulated enzymatic activity, and whether its activity originates from CP itself, four systems were set up for the catalytic oxidation of TMB, and their UV-visible absorption spectra were measured between 500 and 800 nm. As seen in Figure 7, under all other circumstances, only the [TMB+ H₂O₂+CP-1] system exhibited evident UV absorption at 652 nm, and the solution's color changed from colorless and clear to blue, suggesting that the substrate TMB was oxidized. In contrast, the [TMB+ H₂O₂], [TMB+CP-1], and [H₂O₂+CP-1] systems exhibited no visible absorption at 652 nm, and there was no visible change in the solution. This shows that CP-1 has excellent peroxidase-like activity and that this activity arises from CP-1 itself. CP-1 may possess some mimic peroxidase activity, but it is negligible in comparison to the mimic peroxidase activity.

Figure 8a depicts an examination of the simulated peroxidase activity of CP-1, CP-2, CP-3, and CP-0 with varying amounts of template agent. Under the same other conditions, CP-containing nitrogen carbon nanomaterials prepared with the highest simulated peroxidase-like activity when the precursor was the template agent to pyrrole monomer ratio of 1:2, and their absorbance after 5 min of reaction was approximately 7 times that of CP-0 obtained by calcination of pure polypyrrole. This may be because

the template effectively enhances the material's specific surface area, creating more active spots where TMB molecules may be adsorbed and so facilitating the positive reaction. In Figure 8b, the enzyme with the greatest enzymatic activity, CP-1, was chosen for additional investigation to determine the influence of its concentration on the system's catalytic oxidation process. When the concentration of CP-1 rises, the initial reaction rate of the system increases, the absorbance after 5 minutes of reaction increases in order, and the system's color transforms from colorless to dark blue. It was determined that the rate of catalytic oxidation of the system was positively connected with the concentration of CP-like peroxidase, and that the reaction rate of its catalytic oxidation increased as the concentration of CP-1 rose from 0 to 80 µg/mL. This is consistent with the fact that the natural HRP reaction rate [22] is dependent on its own concentration.

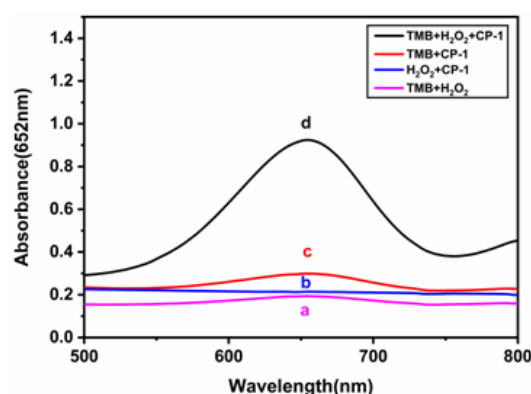


Figure 7. Using TMB as a chromogenic substrate to study the intrinsic peroxidase activity of CP-1: UV absorption Spectral of different reaction systems [(a) TMB+H₂O₂ (b) TMB+CP-1 (c) H₂O₂+CP-1(d) TMB+H₂O₂+CP-1]

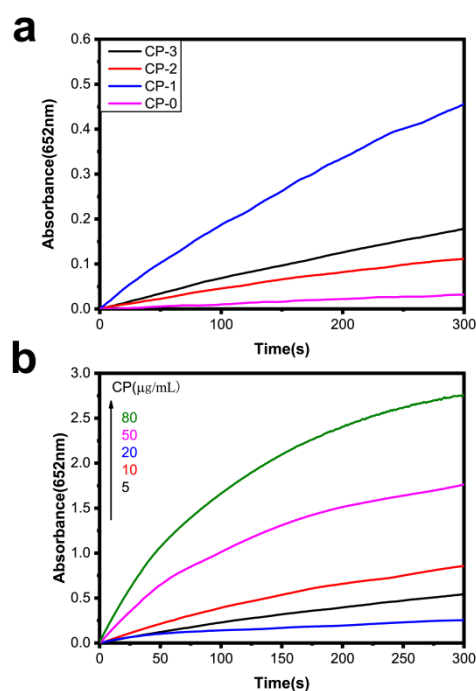


Figure 8. a) CP nitrogen-doped carbon nanometer mimic enzyme system with different template content a) The graph of absorbance change with time; b) Absorbance time curve of reaction system based on CP-1 nitrogen-doped carbon nanometer mimic enzyme under different material concentration conditions

The CP reaction with the substrates hydrogen peroxide and TMB followed the standard Michaelis-Menten model [23], as determined by kinetic calculations. Michaelis constant K_m for the substrate hydrogen peroxide is 1.89 mM, which is almost 50% lower than that of HRP, showing that its affinity for the substrate H_2O_2 is superior to that of HRP. In the meanwhile, the affinity of CP for TMB is greater than that of H_2O_2 , similar with the behavior of peroxidases such as HRP. Comparing the maximal reaction rate v_{max} revealed that the CP peroxidase v_{max} for TMB and H_2O_2 was about 2.52 and 3.05 times that of HRP, respectively.

3.4. CP-Based Colorimetric Detection of Hydrogen Peroxide

As seen in Figure 9a, at optimal reaction circumstances, the absorbance of the system rose sequentially with increasing hydrogen peroxide concentration after 5 minutes of reaction, and the wavelength scan curve exhibited a similar trend. Take the difference between the absorbance at each concentration and the absorbance when the concentration of hydrogen peroxide is zero, create a graph of the relationship between the difference and the concentration of hydrogen peroxide (Figure 9b), and make a linear fit; the results indicate that it has good linear characteristics between 20 and 6000 μM . The linear regression equation was $A_{652nm} = 7.7958 \times 10^{-5} [H_2O_2] + 0.0410$, while the correlation coefficient R^2 was 0.9795. Using the formula $LOD = 3\sigma/s$ (where σ is the relative standard deviation of the absorbance value of the blank sample and s indicates the slope of the standard curve), the limit of detection for H_2O_2 was determined to be 20 μM , indicating a high degree of sensitivity.

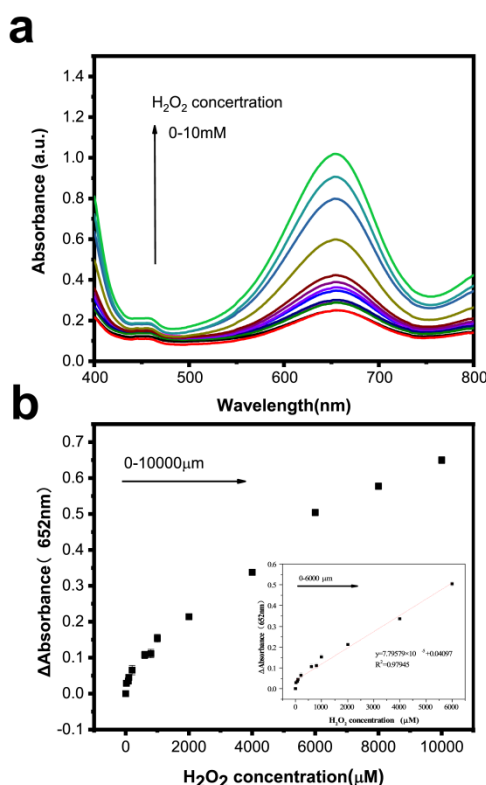


Figure 9. a) Wavelength scan of the system after the reaction; b) The detection range of hydrogen peroxide; inner inset: linear detection range

Table 2 presents the detection ranges, correlation coefficients, and detection limits of other mimetic enzymes reported in the literature for the colorimetric detection of hydrogen peroxide. The comparison reveals that the CP-like peroxidase experimentally prepared in this paper has a wide detection range and reasonable detection limits, making it suitable for the detection of hydrogen peroxide over a broad concentration range.

Table 2. Comparison of the ability of CP and other peroxidases to detect hydrogen peroxide

Sample	Range/ μM	Detection limit/ μM	R^2
CP	20-6000	20	0.9795
CP ₆₀₀₋₆ [24]	20-200	15	0.9697
CWNS _s [25]	0.5-100	0.15	0.9947

4. Conclusion

In this study, polypyrrole macromolecules were introduced between layers of the layered silicate magadiite (MAG) using an in situ polymerization method, and CP nitrogen-containing carbon nanozymes with different template contents were prepared by post-calcination and etching processes. The results demonstrated that the nitrogen-containing porous carbon nanomaterials synthesized by MAG template intervention had a large specific surface area, good retention of N elements, and a high pore size distribution. Consistent with the substrate and catalyst concentration dependency of natural HRP activity, the activity of CP-like peroxidase increased when concentrations of the substrate hydrogen peroxide and the enzyme itself increased. Investigations of its catalytic mechanism show that the peroxidase-like activity of CP materials results from their large specific surface area, which catalyzes the oxidation of H_2O_2 by producing the intermediate product -OH in order to oxidize the substrate. The reaction kinetic data are consistent with the standard Michaelis-Menten model, and the K_m values for TMB and hydrogen peroxide are 0.45 and 1.89 mM, with v_{max} values of 25.22×10^{-8} and $26.6 \times 10^{-8} \text{ Ms}^{-1}$, respectively. The affinity for the substrate is greater and the detection limit for H_2O_2 is 20 μM . In conclusion, the current study not only presents a unique artificial peroxidase for effective sensors, but also encourages researchers to investigate further uses of MAG-based enzyme mimics.

Acknowledgements

This work was supported by the Guangdong Water Conservancy Science and Technology Innovation Project (Project No. 2017-24), Science and Technology Project of Guangzhou (Project No. 202102080477), and Guangdong Provincial Department of Education Featured Innovation Project (Project No. 2017KTSCX007).

References

- [1] F. Manea, F. B. Houillon, L. Pasquato and P. Scrimin, "Nanozymes: Gold-Nanoparticle-Based Transphosphorylation Catalysts", *Angewandte Chemie International Edition*, 43 (45). 6165-6169. Nov.2004.

- [2] J. Li, "Artificial enzymes: learning from nature and beyond", *Science China Life Sciences*, 66, 421-422. Nov.2022.
- [3] B. Liu, Y. Wang, Y. Chen, L. Guo and G. Wei, "Biomimetic two-dimensional nanozymes: synthesis, hybridization, functional tailoring, and biosensor applications", *Journal of Materials Chemistry B*, 8 (44). 10065-10086. Sep.2020.
- [4] Y. Huang, J. Ren and X. Qu, "Nanozymes: Classification, Catalytic Mechanisms, Activity Regulation, and Applications", *Chemical Reviews*, 119 (6). 4357-4412. Mar.2019.
- [5] N. Jiang, N. D. Tansukawat, L. Gonzalez-Macia, H. C. Ates, C. Dincer, F. Güder, S. Tasoglu and A. K. Yetisen, "Low-Cost Optical Assays for Point-of-Care Diagnosis in Resource-Limited Settings", *ACS Sensors*, 6 (6). 2108-2124. Jun.2021.
- [6] Y. Ai, Z.-N. Hu, X. Liang, H.-b. Sun, H. Xin and Q. Liang, "Recent Advances in Nanozymes: From Matters to Bioapplications", *Advanced Functional Materials*, 32 (14). 2110432. Dec.2021.
- [7] R. Li, M. Zhen, M. Guan, D. Chen, G. Zhang, J. Ge, P. Gong, C. Wang and C. Shu, "A novel glucose colorimetric sensor based on intrinsic peroxidase-like activity of C60-carboxyfullerenes", *Biosensors and Bioelectronics*, 47. 502-507. Sep.2013.
- [8] J. Qin, X. Yang, C. Shen, Y. Chang, Y. Deng, Z. Zhang, H. Liu, C. Lv, Y. Li, C. Zhang, L. Dong and C. Shan, "Carbon nanodot-based humidity sensor for self-powered respiratory monitoring", *Nano Energy*, 101. 107549. Oct.2022.
- [9] P. K. Boruah, G. Darabdhara and M. R. Das, "Polydopamine functionalized graphene sheets decorated with magnetic metal oxide nanoparticles as efficient nanozyme for the detection and degradation of harmful triazine pesticides", *Chemosphere*, 268. 129328. Apr.2021.
- [10] X. Liu, J. Wang, D. Wu, Z. Wang, Y. Li, X. Fan, F. Zhang, G. Zhang and W. Peng, "N-doped carbon dots decorated 3D g-C₃N₄ for visible-light driven peroxydisulfate activation: Insights of non-radical route induced by Na⁺ doping", *Applied Catalysis B: Environmental*, 310. 121304. Aug.2022.
- [11] L. Cao, T. Zhu, M. Zan, Y. Liu, X. Xing, Q. Qian, Q. Mei, W.-F. Dong and L. Li, "Red emissive N, Cl-doped carbon dots for detection of tea polyphenols and lysosomal imaging at the cellular level", *Sensors and Actuators B: Chemical*, 370. 132424. Nov.2022.
- [12] X. Zhang, C. Li, X. Wang, S. Yang, Y. Tan, F. Yuan, S. Zheng, D. D. Dionysiou and Z. Sun, "Defect Engineering Modulated Iron Single Atoms with Assist of Layered Clay for Enhanced Advanced Oxidation Processes", *Small*, 18 (52). 2204793. Nov.2022.
- [13] J. Shi, T. Yin and W. Shen, "Effect of surface modification on the peroxidase-like behaviors of carbon dots", *Colloids and Surfaces B: Biointerfaces*, 178. 163-169. Jun.2019.
- [14] A. Ahmed, P. John, M. H. Nawaz, A. Hayat and M. Nasir, "Zinc-Doped Mesoporous Graphitic Carbon Nitride for Colorimetric Detection of Hydrogen Peroxide", *ACS Applied Nano Materials*, 2 (8). 5156-5168. Aug.2019.
- [15] Q. Wang, H. Zhang, D. Yu, W. Qin and X. Wu, "Ultra-sensitive and stable N-doped carbon dots for selective detection of uranium through electron transfer induced UO₂(V) sensing mechanism", *Carbon*, 198. 162-170. Oct.2022.
- [16] M. Ge, X. Li, M. Zhang and Z. Liu, "Enhanced Photocatalytic Degradation Performance of Antibiotics Using Magadiite-Supported Carbon Nitride Under Visible Light Irradiation", *Journal of Inorganic and Organometallic Polymers and Materials*, 32 (2). 678-686. Feb.2022.
- [17] Z. Wang, X. Xu, J. Kim, V. Malgras, R. Mo, C. Li, Y. Lin, H. Tan, J. Tang, L. Pan, Y. Bando, T. Yang and Y. Yamauchi, "Nanoarchitected metal-organic framework/polypyrrole hybrids for brackish water desalination using capacitive deionization", *Materials Horizons*, 6 (7). 1433-1437. Apr.2019.
- [18] M. Ma, Z. Liao, X. Su, Q. Zheng, Y. Liu, Y. Wang, Y. Ma and F. Wan, "Magnetic CoNi alloy particles embedded N-doped carbon fibers with polypyrrole for excellent electromagnetic wave absorption", *Journal of Colloid and Interface Science*, 608. 2203-2212. Feb.2022.
- [19] W. Ding, Z. Wei, S. Chen, X. Qi, T. Yang, J. Hu, D. Wang, L.-J. Wan, S. F. Alvi and L. Li, "Space-Confinement-Induced Synthesis of Pyridinic- and Pyrrolic-Nitrogen-Doped Graphene for the Catalysis of Oxygen Reduction", *Angewandte Chemie International Edition*, 52 (45). 11755-11759. Sep.2013.
- [20] Y.-Q. Zhang, D.-K. Ma, Y. Zhuang, X. Zhang, W. Chen, L.-L. Hong, Q.-X. Yan, K. Yu and S.-M. Huang, "One-pot synthesis of N-doped carbon dots with tunable luminescence properties", *Journal of Materials Chemistry*, 22 (33). 16714-16718. Jun.2012.
- [21] Z. Wang, L. Qie, L. Yuan, W. Zhang, X. Hu and Y. Huang, "Functionalized N-doped interconnected carbon nanofibers as an anode material for sodium-ion storage with excellent performance", *Carbon*, 55. 328-334. Apr.2013.
- [22] L. Gao, J. Zhuang, L. Nie, J. Zhang, Y. Zhang, N. Gu, T. Wang, J. Feng, D. Yang, S. Perrett and X. Yan, "Intrinsic peroxidase-like activity of ferromagnetic nanoparticles", *Nature Nanotechnology*, 2 (9). 577-583. Sep.2007.
- [23] H. Liu, Y. Ding, B. Yang, Z. Liu, Q. Liu and X. Zhang, "Colorimetric and ultrasensitive detection of H₂O₂ based on Au/Co₃O₄-CeO_x nanocomposites with enhanced peroxidase-like performance", *Sensors and Actuators B: Chemical*, 271. 336-345. Oct.2018.
- [24] Z. Lou, S. Zhao, Q. Wang and H. Wei, "N-Doped Carbon As Peroxidase-Like Nanozymes for Total Antioxidant Capacity Assay", *Analytical Chemistry*, 91 (23). 15267-15274. Dec.2019.
- [25] W. Cao, P. Ju, Z. Wang, Y. Zhang, X. Zhai, F. Jiang and C. Sun, "Colorimetric detection of H₂O₂ based on the enhanced peroxidase mimetic activity of nanoparticles decorated Ce₂(WO₄)₃ nanosheets", *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy*, 239. 118499. Oct.2020.

