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Conveying the Accuracy and Reliability of Synthetases in Catfish Freshness Assessment

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Abstract

Seabed products, especially fish, like other muscle products, are highly perishable due to their remarkable natural composition. It can be detected colorimetrically by engineered nanozymes without the aid of xanthine oxidase). His trading of Hx in polyvinylpyrrolidone-modified platinum box nanomaterials (PVP-PtNC), where Hx blocks catalytically active sites on the surface, serves as the basis for the principle. The ability of PVP-PtNC to induce the oxidation of 3', 5'-tetramethylbenzidine (TMB) by H_2O_2 is hampered by this downregulation of its catalytic ability. This results in a 1:1 ratio between Hx attention and UV-Vis immersion drop-off versus colorimetric responsive color drop-off. Rapidly detectable, affordable, accurate and easy-to-use targeted colorimetric systems have been developed to monitor novelty in fish. In particular, the fact that Hx has been established in real fish suggests that the system has a bright future in the real world. Hx correlation response is set to the range of 0.5M

Keywords: Fish Freshness; Platinum Nanocube; Hypoxanthine

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Introduction

One of the most important components of the modern food industry is seafood, which provides the body with the calories it needs and various nutrients such as protein and docosahexaenoic acid. People are especially attracted to these products [1]. According to the official report on fisheries and aquaculture products (FAP), the daily consumption of aquaculture products is steadily increasing. China is largely responsible for the increase in global fish consumption, as its domestic market consumes the majority of its seafood and aquatic products. China is currently leading the world. However, seafood, especially fish, is highly perishable and undergoes rapid protein degradation, lipid oxidation, and changes in odor, taste and texture, resulting in a short shelf life after slaughter [2]. As a result, accurate assessment of fish freshness is favored by seafood spoilage. To assess the sensory properties of fish, consumers have traditionally relied primarily on sensory approaches based on perception and experience. This method is not effective in judging freshness due to its subjective nature and lack of reliability [3]. Long-term spoilage of food can even pose a serious threat to food safety. Therefore, the development of accurate analytical methods to accurately monitor the freshness of seafood is essential [4].

Thanks to the rapid development of seafood science and modern analytical chemistry, biochemical and microbiological levels are used to determine the freshness of seafood such as fish. A reaction that produces hypoxanthine (Hx) displaces adenosine triphosphate (ATP) when microorganisms grow in fish tissue. This transformation suggests that Hx accumulation is inversely proportional to storage time and thus can be used as an important indicator of fish freshness. Conventional methods, including mass spectrometry techniques, are currently used to meet freshness requirements [5]. Near-infrared spectroscopy, electronic nose, electronic tongue, fluorescence techniques, computer vision techniques, and gas chromatography-mass spectrometry (GC-MS) techniques have all been used to measure Hx. Despite their high reproducibility, precision, and reliability, these methods require expensive equipment, skilled workers, and specialized laboratories, and thus can only be used in limited situations. Efficient, accurate and easy-to-use detection methods are always in demand [6].

A new class of engineered enzymes known as nanozymes has recently emerged and made great strides. Nanozymes are highly efficient due to their enzyme-like properties. His seminal 2007

report on this work has been published by Yan's group. The first result of their investigation was the intrinsic peroxidase-like activity of ferromagnetic nanoparticles [7]. Various nanozymes such as graphene, carbon dots, WS2 nanoplates and Au nanoclusters have been used to build biosensors sensitive to proteins, nucleic acids, metal ions, inorganic and organic compounds, and other target analytes rice field. In particular, noble metal nanomaterials have become one of the most popular topics due to their unique chemical properties and excellent catalytic activity. In contrast to natural enzymes, engineered nanozymes are prone to inactivation, expensive to use and difficult to manufacture. It is also easy to manufacture, has high catalytic capacity, high stability, and low cost. These advantages are making nanozyme-based bioanalytical sensors more popular. These sensors may also be used to develop assay-responsive online detection strategies [8].

Xanthine oxidase (XOD) is normally required for conventional His Hx sensor platforms to convert His Hx to Her H_2O_2 and uric acid in the presence of oxygen. The generated H_2O_2 is further catalyzed to produce hydroxyl radicals (OH) and similar nanozymes to oxidize the chromogenic substrate from colorless to colored.

Against this background, the research goal is to build a conventional colorimetric sensor platform for judging fish freshness. We established a colorimetric method for Hx determination based on the downregulation of peroxidase activity in polyvinylpyrrolidone-modified cubic platinum nanomaterials (PVP PtNC) [9]. We found that Hx can directly modulate the extremely high catalytic capacity of PVP-PtNCs by blocking surface-active catalytic sites. Exposure to H_2O_2, which is used to catalyze the oxidation of TMB, a typical peroxidase substrate, significantly reduced peroxidase activity and effectively stopped the production of the oxidized blue product. As a result, this sensor platform can quantify Hx without the help of native enzymes (XOD). According to our findings, transmission electron microscopy (TEM) successfully demonstrated the morphology of PVP PtNCs [10]. The lower the UV-Vis absorption and the weaker the colorimetric reaction color, the lower the Hx concentration. This colorimetric nanosensor method was also adopted due to its reproducibility, low detection limit, high specificity, large linear response range, and the successful analysis of real fish samples in 10 minutes. Therefore, a nanozyme-based sensor platform with the advantage of detection at the smallest scale will open a new avenue for online monitoring of seafood freshness [11].

Materials and Method

PVP synthesis

PVP-PtNC nanomaterials were synthesized as described in the literature. 20 mg of KBr and 40 mg of PVP were first injected into 3.5 ml of glycol (EG) solution. After dissolution, the mixture was heated to reflux (about 180° C.) in an oil bath for 15 minutes. The mixture is then given for an additional 20 minutes to react with 0.5 ml of a 40 mg/ml K2PtCl6.6H2O solution in which EG is dissolved. The resulting solution was then immediately cooled in an ice bath [12]. Excess PVP was removed by centrifugation, and the final reaction product was washed with acetone and deionized water three times before being stored in 4 ml of ultrapure water at 4°C until use of the purified PVP-PtNC. The platinum concentration in PVP-PtNC was 0.473 g/L as determined by ICP-OES [13].

Exposure through PVP

Since Hx is sparingly soluble under neutral conditions, a standard Hx solution prior to detection was prepared by weighing 0.0136 g of Hx and dissolving it in 0.4 mL of 1 M HCl. To prepare a stock solution of Hx (10 mM), 9.6 mL of 0.05 M sodium acetate-acetic acid (NaAC-HAC) buffer, pH 4 was added to the mixture. Additional concentrations were obtained by diluting stock solutions of Hx with pH 4 0.05 M NaAC-HAC buffer. PVP-PtNC (100 µL, 5 M) and various concentrations of Hx solutions were mixed in 0.05 M NaAC-HAC buffer (pH = 4) for 30 min at room temperature for Hx colorimetric detection. For colorimetric catalysis, 100 mL of the resulting mixture was added to 25 mL each of TMB (25 mL, 15 mM) and H_2O_2 (25 mL, 1 M) for 10 min at room temperature. A UV-Vis spectrophotometer was used to measure the visible absorption spectrum from 400 to 800 nm. Sensor performance was evaluated by recording peak absorbance values at 651 nm [14].

Efficient Analysis

We used the PVP based sensor method to detect Hx in real fish samples prepared by injecting different concentrations of standard Hx into two batches of spoiled fish meat. This was after showing that Hx can be detected both qualitatively and quantitatively. The above is the pretreatment of the fish meat. Regardless of whether the fish samples were spoiled for 12 or 24 hours, Hx spiked at the indicated concentrations of 10, 100 and 1000 M all gave good recoveries of 102.6–106.0% and 100.9–101, respectively. On the other hand, the relative standard derivative (RSD) values were within acceptable limits. These results indicated that this method can be used to monitor fish freshness by measuring Hx levels in fish meat [15].

Conclusion

We have developed a colorimetric method for ultrafast Hx signaling based on the targeted downregulation of peroxidase activity in PVP-PtNC without the help of native enzymes. This study demonstrates the following advantages of colorimetric methods for Hx detection.

Hx was originally used to directly modulate the catalytic activity of nanozymes, reducing the detection cost and allowing Hx detection in one step at 10 min. This method has been successfully used to monitor the freshness of live fish. Second, the colorimetric approach does not require expensive and cumbersome equipment. Third, this colorimetric approach has a wide detection range and high sensitivity and specificity for detecting Hx. This method is expected to help determine the freshness of fish samples to ensure food safety.

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