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Hypothesizing an Ecological and Non-Invasive Early Alzheimer’s Diagnosis: Intranasal Carbon Dot Biosensors for Ultrasensitive p-tau217 and pH Detection.

Abstract

Neurodegenerative diseases related to genetics are among the most dangerous in the world, and Alzheimer's disease is one of them. Currently, diagnostic methods for Alzheimer's are very expensive, invasive, and do not allow for early diagnosis. Considering this, this research hypothesizes if Carbon Dots can be used as a diagnostic tool that detects lysosomal pH fluctuations and the presence of specific biomarkers, by utilizing an intranasal delivery and fluorescence. Therefore, the results aim to democratize the diagnosis, change the global health system and propose a sustainable alternative.

Introduction

Alzheimer's Disease or AD is a neurodegenerative, irreversible and progressive condition that affects predominantly elderly people from all over the world, causing memory deterioration and cognitive disturbances. Pathologically, it is characterized by amyloid-beta plaques and neurofibrillary tangles that are caused by hyperphosphorylated tau protein **(Smith, M. D. A. C. (1999)**. However, recent research suggests these hallmarks are consequences of an earlier endolysosomal dysfunction: the failure to maintain an acidic pH within lysosomes impairs the degradation of toxic proteins, directly contributing to the accumulation of hyperphosphorylated tau and amyloid aggregates . The current methods used for AD detection are PET/MRI (cost-intensive) and CSF Analysis (highly invasive) but according to the existing literature: it is necessary to invest in the development of more precise biomarkers and imaging techniques to improve the diagnosis and treatment of the disease, which is hampered by ambiguous clinical presentations and a lack of specific biomarkers, leading to several delays in the treatment. Considering this, this research proposes the use of Nitrogen and Sulfur-doped Carbon Dots (biomass-derived nanoparticles) as a pH-sensitive biosensor, aiming for a more accessible and early-stage diagnostic tool.

Literature Review

Carbon Dots (CDs) are carbon particles with 1-10 nanometers in size and important properties: strong absorption, excellent light stability, low toxicity, eco-friendly, water soluble, great biocompatibility, and easy preparation **(Xia, Chunlei, et al 2019)**. The synthesis of CDs can be done in two ways: top-down and bottom-up, but the second method is cheaper. Bottom-up methods primarily use the hydrothermal or pyrolysis of small organic molecules – such as glucose and citric acid – that can be aromatic or non-aromatic and after this reaction, it can be mixed with Nitrogen or Sulfur to dope the CDs **(Ozyurt, Derya, et al. 2023)**.

According to contemporary literature (e.g., Dong et al., 2012), evidence suggests that doping these CDs improves their optical and electronic performance, because it creates new energy states, establishing their electron cloud. This chemical modification allows the nanoparticle to undergo reversible protonation/deprotonation, making its fluorescence intensity highly sensitive to local pH variations, a critical and useful feature for detecting lysosomal dysfunction in AD.

Even though studies on CDs are still recent, some researchers, such as **Jiang, Wei, et al. (2021)**, have delved into their use as biosensors - devices that convert a biological response into an electrical, optical, or thermal signal. Nevertheless, in the case of their application for the diagnosis of AD, they will be functional biosensors, since they signal through fluorescence and act in vivo, so fluorescence imaging can be used to ensure the diagnosis.

Methods

This study will be conducted according to **Khairol Anuar, Nurul Kamilah, et al.** Hydrothermal Method. Firstly, the CDs will be synthesized using fruit peels biomass (e.g., banana or mango peels) or sugarcane bagasse and for the doping Urea and L-cysteine (primarily fonts of N and S). After this, the method Hydrothermal Treatment (bottom-up) will be applied so the precursor substances will be dissolved in deionized water and placed in a Teflon-lined stainless steel autoclave reactor. To ensure complete carbonization of the biomass precursors while maintaining the stability of the functional groups, the hydrothermal treatment will be conducted at 160°C-200°C for 4-6 hours. After this, the solution will be cooled to room temperature and subjected to centrifugation for 20 minutes to remove large polymers chains. After cooling it, the solution will be filtered and the CDs will be functionalized with DNA aptamers designed to bind specifically to the p-tau217 protein. This bioconjugation creates a targeted sensor capable of identifying the disease's earliest biomarkers - such as p-tau217. Looking for a real non-invasive diagnosis, the biosensor will be delivered via an intranasal spray. This route utilizes the olfactory pathways to bypass the Blood-Brain Barrier (BBB**)**, allowing the particles to reach the brain's deeper regions directly. Combined with a mucoadhesive matrix to improve absorption, the sensors will provide a real-time diagnostic signal through ratiometric fluorescence, identifying lysosomal dysfunction, triggered by the local pH dysregulation (lysosomal acidification failure), and protein accumulation long before traditional symptoms appear. This signal will be monitored via Near-Infrared (NIR) imaging, providing a real-time diagnostic tool for early-stage AD.

Conclusion:

This study presents a suggestion of low-cost, and non-invasive alternative to traditional Alzheimer’s diagnostics by detecting p-tau217. By mixing green nanotechnology with intranasal delivery, this method not only bypasses the blood-brain barrier but also provides a scalable solution for global healthcare systems. Ultimately, these Carbon Dot biosensors represent a vital step toward transforming Alzheimer’s from a late-stage discovery into a manageable condition through early detection, saving families from all over the world.

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