Detection of Dopamine via Green Synthesis of Gold Nanoparticles Dipped Carbon-Fiber Microelectrodes
American University, Washington DC

Introduction

With applications in biomedicine and material sciences, the field of nanotechnology is a current focus of many researchers. Nanoparticle research targets gold nanoparticle production due to their optoelectric properties and their wide range of applications. For example, gold nanoparticles have been used in sensory probes, therapeutic agents, drug delivery systems, etc. because of their biocompatibility, inertness, non-toxicity, and ability to readily attach to biological molecules.

Specifically, the interdisciplinary bio-directed synthesis (or green synthesis) of gold nanoparticles intrigues many from biology to chemistry to material science engineering. Bio-materials are often cheaper and more sustainable than inorganic material synthesis. Thus, replacing synthetically-produced with green nanoparticles will help prevent toxic laboratory waste and lead to a cleaner environment. Literature has shown that using natural honey as the reducing and capping agent for the synthesis of gold nanoparticles at room temperature is greener than all other bio-directed synthesis methods. Fructose — the primary ingredient in honey—is most likely the ingredient responsible for the reduction of gold. The literature also points to glucose and sucrose as potential contributors to the reduction. Last semester, reducing agent stock solutions of fructose, galactose, sucrose, sodium citrate (Na$_3$Ct), ascorbic acid (H$_2$Asc) were prepared and used for the synthesis of gold nanoparticles. Fructose was shown to produce gold colloids along with chitosan and Na$_3$Ct. This study will produce gold nanoparticles from natural honey as the experimental group and gold nanoparticles from synthetic fructose, sucrose, and glucose as controls.

Although green nanoparticles are well researched, few studies have then used the nanoparticles in an application. This study furthers this area of research by first producing the green nanoparticles and then depositing them onto carbon-fiber microelectrodes (CFMEs) to determine how the green nanoparticles work compared to synthetically-produced nanoparticles. Electrochemical detection is rapidly becoming significant for the detection of biological species. CFMEs are the standard electrodes used in the detection of neurotransmitters and are best used in biomedicine due to several factors. Firstly, carbon fibers are biocompatible and have little to no toxicity to cells. Also, they are small in size - usually less than ten micrometer in diameter - making them optimal for implantation because there would be less tissue damage than the larger standard electrodes. The electrochemistry of CFMEs has been studied extensively, and CFMEs are known to possess good electrochemical characteristics. However, building electrochemical sensors from noble metals, such as gold instead of bare carbon fibers could potentially be advantageous in producing cost-effective multiplexed electrochemical detection schemes.
While not as ideal as carbon fibers, gold electrodes render a simple platform for electrode surface modifications and microfabricated electrode arrays, allowing them to detect and distinguish several neurotransmitters. Furthermore, gold is able to covalently bond to functional groups such as thiol groups, forming self-assembled monolayers ideal for electrochemical sensors. The adsorption of catecholamines, such as dopamine onto gold has been researched using slow scan cyclic voltammetry; however, this method lacks in selectivity and sensitivity for real-time in vivo detection. Due to the differences in electrochemical characteristics of gold and carbon fibers, the electroactivity of dopamine on gold-modified CFMEs should be characterized using fast scan cyclic voltammetry (FSCV), and then compared to CFMEs as a reference.

FSCV is a technique that employs rapid scanning, providing an output of current versus voltage (cyclic voltammogram) in the identification of specific analytes. The large charging current produced by fast scanning is relatively stable at carbon fibers. This current can also be subtracted from the analyte signal to yield a background-subtracted cyclic voltammogram. The neurotransmitter is oxidized and the concentration is proportional to the oxidation peak; thus, yielding a biosensor capable of detecting dopamine. CFMEs continue to be significant in comprehending the basic mechanism of neurotransmission in in vivo studies.

The catecholamine dopamine, is one of the neurotransmitters that has been extensively studied because of its all-round significance within neurobiological systems. Dopamine is an essential chemical messenger which allows information to be passed through neurons permitting proper neuropsychological behavior. Parkinson’s disease is commonly known for causing a deficiency of dopamine in certain areas of the brain, such as the basal ganglia; thus, causing involuntary movements. On the other hand, dopamine levels are increased when an individual ingests drugs causing addiction. This excess of dopamine in the brain causes the formation of new dopamine receptors; thus, inducing the dependency on the drug. Dopamine is also important for a variety of other functions in the human body such as processing pain, controlling one’s mood, pleasure, and the understanding of rewards.

Statement of Work

This project will be broken down into the following objectives which will be conducted in the following order:

1. Produce gold nanoparticles via natural honey mediated green synthesis
2. Produce gold nanoparticles from synthetic fructose/glucose/and sucrose
3. Develop three groups of carbon-fiber microelectrodes
   a. Green synthesized gold nanoparticles will be attached to the tapered ends of one electrode group
   b. Synthetic fructose gold nanoparticles will be attached to the tapered ends of one electrode group
   c. One electrode group will be prepared without any gold nanoparticles
4. Assess the detection of dopamine using these electrode groups

Since the nanoparticles will precipitate if not tested soon after they are produced, they will be added to the CFMEs and tested before moving to a different nanoparticle group. Thus, producing and testing the green synthesized gold nanoparticles and CFME Group (a) will be conducted first. Once testing of these group is finished, producing and testing the synthesized
gold nanoparticles and CFME Group (b) will be conducted. Finally, CFME Group (c) will be tested. Data will be analyzed throughout this process.

Hazards & Toxicity

The suggested project employs a WaveNeuro Fast Scan Cyclic Voltammetry (FSCV) Potentiostat System, which is an electrochemical instrument that generates alternating current. Proper control and managing skills are required to avoid electrical shock. Chloroauric acid (Hydrogen tetrachloroaurate (III) hydrate) and other chemicals will be used that might cause severe skin burns and/or eye damage if not handled properly. Due to their oxidizing properties, allergic skin reactions can also occur if not handled with care. The synthesis of carbon-fiber micro-electrodes requires the use of an oven to be set at 120˚C for 4 hours. In addition, the use of acetone will be necessary. Appropriate use of heat resistant gloves is recommended to prevent any burns. All other material usage should be fairly safe but care must be applied in every step of the way. Acetone is an extremely flammable solvent that can also be harmful if swallowed and inhaled. Proper PPE and goggles will be worn during its use.

Personnel & Facilities

Alexander Zestos— the PI for this project— is an Assistant Professor in the Department of Chemistry at American University. His work focuses on creating electrochemical sensors through nanomaterials. Specifically, he creates carbon-fiber microelectrodes to detect neurotransmitters and other relevant molecules via fast scan cyclic voltammetry.

The proposed work will be conducted at American University. Ultraviolet-visible Spectroscopy will be used to determine the presence of gold colloids— from both the natural honey and the synthetic fructose. Transmission electron microscope will be used to photograph the colloids produced. The crystalline nature of the gold nanoparticles will be confirmed via X-ray diffraction analysis. Fourier-transform infrared spectroscopy measurements will be conducted to identify the biomolecules responsible for capping and stabilizing the gold nanoparticles. An Inductively coupled plasma optical emission spectroscopy instrument will be used to properly characterize the concentration of solutions prepared in order to verify their concentration.

The synthesis of carbon-fiber microelectrodes requires a vacuum to delicately aspirate the carbon-fiber into the glass nanotube. Once aspirated, a PC-100 Puller is required to pull the carbon-fiber glass capillaries into two fine tapered sensors that can further be used to perform detection experiments. Afterwards, the carbon-fiber microelectrodes will be functionalized with Au nanoparticles and CNTs. In addition, the sensors will get covered with epoxy to close off the end of the sensor where the carbon fiber is protruding to enable a tight seal for its back filling with potassium chloride. Finally, the synthesized carbon-fiber microelectrodes with and without AuNPs, will be tested for their accuracy at identifying dopamine via FSCV to determine if their detection can be characterized at a different level.

References


