RESEARCH PAPER

Colorimetric Detection of Ammonia in Water by Using Biosynthesized Gold Nanoparticles

Shyamal Mandal[1], Nitin Sahai[1,2*]
[1]Department of Biomedical Engineering, North Eastern Hill University, Shillong, Meghalaya, India – 793022.
Email: shyamalmandaliit@gmail.com
[2]Department of Electronics and Communication Engineering, North Eastern Hill University, Shillong, Meghalaya, India – 793022
***Corresponding Author:** nitinbiomedical@gmail.com

Hydrogen and oxygen combine to form the chemical ammonia (NH_3). It can have a major impact on aquatic plants and animals and is commonly found in wastewater. It's crucial for maintaining the balance of ecosystems and human health. Less than 1.5 mg/L is the threshold level of ammonia in drinking water that should be present. Main sources of NH_3 in river water is biomass and various types of fertilizers. Ammonia can be detected in water using a variety of techniques, such as pH and ion-selective electrode approaches, but gold nanoparticle-based colorimetric methods are the most distinctive and straightforward because they allow ammonia to be easily recognized, even though the NH_3 solution colorless. The capacity of gold nanoparticles (AuNPs) to detect NH_3 at room temperature has been created and tested. The presence of ammonia in water is indicated by the redshift of a resonant peak in the absorption spectra, which is mostly explained by the chemical bonding that forms between AuNPs and NH_3 at the defect sites. Additionally, it mostly reacts with oxygen absorbents on AuNPs' surface, which causes it to exhibit sensing behavior. In this study we successfully achieved the green synthesis of AuNPs using pineapple extract which perform as reducing and capping agent. In present work we successfully measured the ammonia level at 200 µl to a maximum of 4900 µl. Results also conclude that if material surfaces were modified with AuNPs can be applied for drug loading using electrostatic interaction.

Keywords: Colorimetric, Ammonia (NH_3), Gold nanoparticles (AuNPs), Sensing, Spectra, Colour change, Drug loading
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1. Introduction:

Ammonia and its salts can be dangerous and corrosive, and they dissolve easily in water. Ammonia concentrations over 0.5 micromoles per litre have the potential to have a major negative effect on the environment. Ammonium predominates in natural settings, whereas ammonia makes up less than 25% of ammonium. Ammonia is exceedingly dangerous and should be thoroughly investigated, even if its concentration in the environment is just nanomoles per litre, much lower than that of ammonium. This is because dissolved ammonia is the primary cause of ammonia nitrogen's toxicity in water. High concentrations of ammonia and ammonium increase plankton populations' primary production, which causes fast algal development and eutrophication of water bodies, which can harm the aquatic ecosystem as a whole. The sensitivity of marine fish and invertebrates to ammonia toxicity is increased in anoxic conditions. Furthermore, fish, crabs, and humans are all poisonous to ammonia and its salts, with aquatic creatures being disproportionately more affected. Ammonia can cause the loss of entire populations of fish and invertebrate larvae, endangering numerous crucial ecosystems and fisheries. It is particularly detrimental to fish and invertebrate larvae during their growth and infancy [1-3]. Furthermore, under specific circumstances, ammonia can undergo nitrification and be transformed into nitrite and nitrate, both of which are harmful [4-9]. Nitrogen from ammonia is essential for life, productivity, and environmental ecology. Therefore, it's critical to identify, sense, and keep an eye on ammonia nitrogen levels in lakes, rivers, saltwater, and other various waterways to achieve sustainable water resource use and safeguard aquatic creatures from the acute and long-term consequences of ammonia. Several detection methods have been developed to control and monitor ammonia nitrogen levels, including the Nessler and indophenol blue (IPB) protocols [10] looked at how these methods for ammonium detection were developed [11,12] and evaluated these methods' accuracy and precision in freshwater and saltwater aquaculture, respectively. Due to restrictions on lower concentrations and environmental protection, these methods aren't always preferable. To fond the solution for this challenge, several analytical and sensory approaches have been developed. However, these techniques are not always applicable because of requirements for low concentrations and environmental protection. Many analytical and sensing techniques have been developed to address this problem [13]. compiled the benefits and drawbacks of gas phase molecular absorption spectrometry, chromatography, and distillation-titration for the detection of ammonia nitrogen in aqueous environments. Gold nanoparticles have been applied extensively in biology and medicine, and bio-nanotechnologists are quite interested in synthesizing them. Gold nanoparticles that have been functionalized with the right biomolecules are ready for specific uses. Researchers have interest on the various forms of gold nanostructures. Numerous processes for creating gold nanoparticles are detailed in the literature. Most of the techniques are based on inorganic chemical pathways. However, given the trend of using gold nanoparticles in biological sciences, more environmentally friendly ways to create gold nanoparticles are required. Because of excellent exceptional surface biocompatibility, gold nanoparticles (AuNPs) are among the most often used engineered materials in bio-imaging and medicinal treatments [14, 15]. AuNPs' powerful affinity for thiols makes it easier for them to combine with biomolecules like DNA and antibodies. It's also helpful for the development of different types of biosensors [16,17]. Optical sensor also used for finding of ammonia[18].

1. MATERIALS AND METHODS:

All the reagents were taken into consideration without more further purification. Deionized Milli Q water (DI) with a resistivity of 18.2 MΩ was used throughout the experiment. ($HAuCl4. 3H_2O$) and NaOH were bought from SRL Chemicals Mumbai, India. Pineapple brought from local market Shillong, India. Pineapple juice act as a reducing agent as well as capping agent.

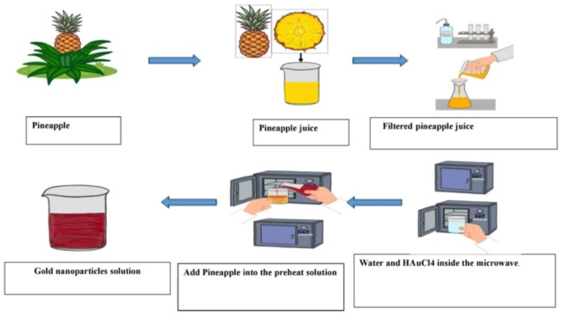


Figure 1: Schematic illustration of biosynthesis of gold nanoparticles.

2.1. Biosynthesis Mechanism:

The manufacturing mechanism for gold nanoparticles utilizing various biological agents has yet to be identified. Different chemicals in biogenic substances may act as reducing agents, interacting with metal ions to help them break down and, as a result, making metal nanoparticles. Researchers have proposed several hypotheses to understand the mechanism behind the production of gold nanoparticles. Recent studies indicate that biomolecules, including proteins, phenols, and flavonoids, found in fruit extracts, significantly contribute to the reduction of metal ions and the stabilization of nanoparticles [19].

$$HAuCl_4 + \text{Pineapple juice} = Au + Cl + H_2O$$

3. Results and discussion:

3.1. Transmission Electron Microscopy:

Figure 2 (a,b,c) shows the TEM micrographs (TECNAI G, FEI, Netherlands) of the as-synthesized gold nanoparticles. TEM photograph shows that Au nanoparticle's shape were uniform and size around (10 - 50 nm). Selective Area Electron Diffraction (SAED) pattern also confirmed that [111] zone axis with hexagonal symmetry. It also determined that nanoparticles are single crystalline in nature.

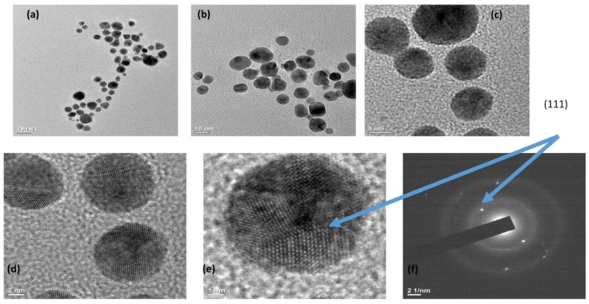


Figure 2(a-d), TEM micrograph of Au-NPs, (e) Different planes of AuNPs (f) SAED pattern of gold nanoparticles.

3.2. *XRD analysis of AuNPs*

The gold nanoparticle characteristics were determined by comparing the JCPDS data file (No: 04-0784). XRD spectrum for the synthesised AuNPs is present in Figure 3. It has been observed a dominant peaks of metallic Au. The characteristics peaks of AU corresponding to Miller indices (1 1 1) and (220) were observed which confirmed the FCC crystalline geometry of AuNPs based on the above XRD data.

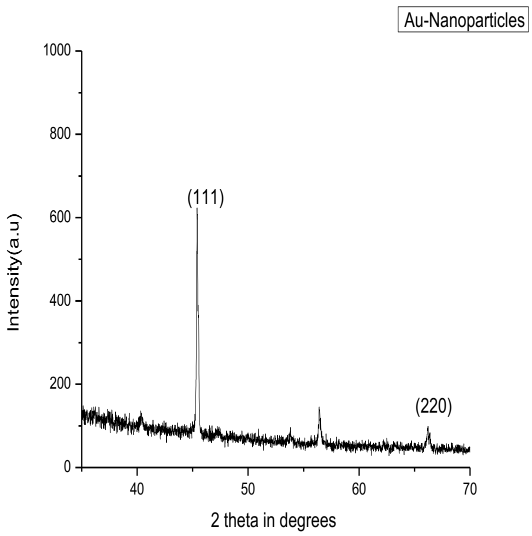


Figure 3: XRD pattern of gold nanoparticl

3.3 Zeta potential of different samples

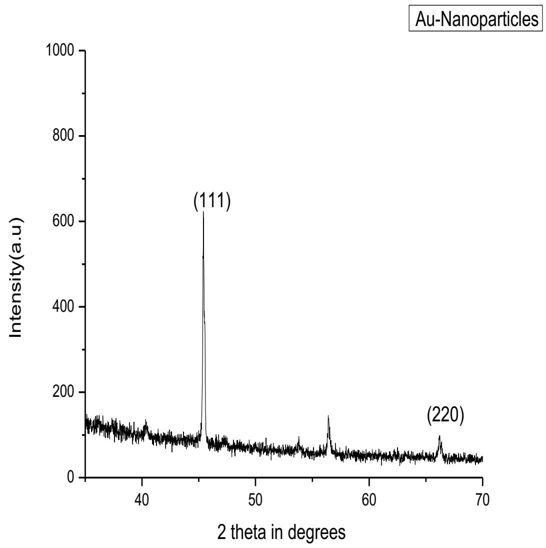
Zeta potential measurements reveal the nanoparticles are highly stable and have an average surface -25.6mV charge.it is mainly used to measure the diameter, zeta potential, electronic mobility, conductivity and Standard deviation of the solution.

Table 1: Describe the Zeta potential of AuNPs

Sample name	Zeta Potential
Gold nanoparticles	-25.6mV

4. Ammonia sensing:

The present study utilized a Perkin Elmer Lambda 750 spectrophotometer from the United States. Exactly 3 millilitres of AuNPs was added to the quartz cell, and the absorbance spectra were measured for a duration of 1 minute. The scanning wavelength range chosen was 300-800 nm. Figure 4 demonstrates that the pick's visual characteristics at a wavelength of 535 nm indicate SPR band of different concentration of ammonia. Colourimetric assays based on the unique SPR properties of gold nanoparticle have shown to be very much significant due to its simplicity, high sensitivity, low detection limit, low cost, quick response time and reproducibility. It may be noted that we have reported room temperature detection of aqueous ammonia at µl level by AuNPs based optical sensor.



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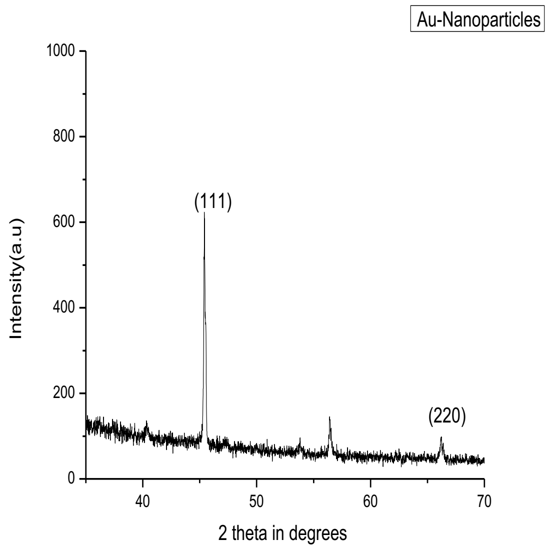

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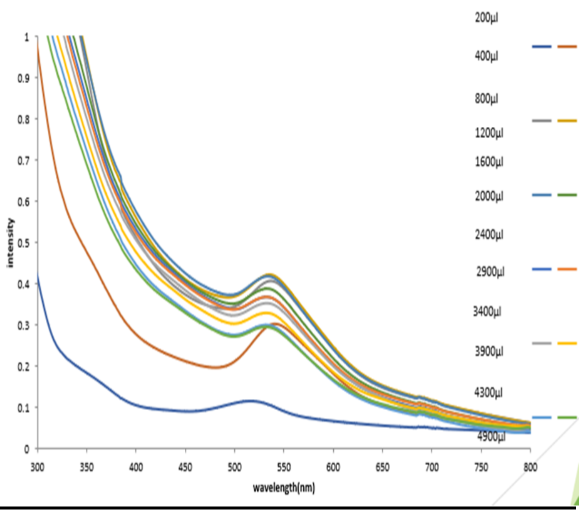

Figure 6: Graph of ammonia sensing

5.Ammonia sensing mechanism

The analysis of ammonia solution was conducted by colorimetric measurements. Understanding the optical properties of Au-NPs necessitates consideration of their composition, morphology, size, and immediate surroundings. The local dielectric environment encompasses both solvent molecules and additional nanoparticles. The interaction of electromagnetic fields from two distinct nanoparticles generates a complicated surface plasmon resonance (SPR). The aggregation of nanoparticles results in a shift of the surface plasmon resonance to lower energy compared to solitary nanoparticles. To prevent uncontrolled aggregation, the surfaces of nanoparticles have been modified with capping molecules that provide an electrostatically generated steric barrier between them. Alternatively, to facilitate regulated nanoparticle aggregation under specified environmental circumstances, capping molecules can be affixed to the surface of the nanoparticles. Consequently, pineapple juice serves as a capping agent for the stability of Au-NPs. The sensing capability of Au-NPs solution in response to escalating ammonia concentrations from 200 to 5000 µl is assessed by observing variations in the SPR location and amplitude using UV-Visible spectrophotometry. To investigate the ammonia sensing behavior, the Au-NPs solution was combined with an aqueous ammonia solution at varying concentrations of 200 to 5000 µl, diluted with 25% ammonia solution in deionized water just prior to the tests. The absorbance spectra of the Au-NPs colloidal solution at varying ammonia concentrations are depicted in Figure 4. The absorbance peak intensity at approximately 537 nm consistently diminishes with an increase in ammonia content. This process occurred due to the interaction of ammonia with Au^{3+} or Au^{+} ions, resulting in the production of the coordination complexes $[Au(NH_3)_2]^{3+}$ and $[Au(NH_3)_2]^{+}$.

6. Conclusion:

This research article describes the green synthesis of gold nanoparticles (AuNPs) using pineapple extract, which acts as both a reducing and capping agent. The synthesized AuNPs exhibited perfect stability in aqueous solvents, consisting to their high zeta potential. This approach offers a low cost and environmentally friendly method for producing gold nanoparticles. Colorimetric assays leveraging the unique surface plasmon resonance (SPR) properties of AuNPs are particularly valuable due to their simplicity, high sensitivity, low detection limits, cost-effectiveness, rapid response, and excellent reproducibility. We have demonstrated the detection of aqueous ammonia at the microliter level using an AuNPs-based optical sensor at room temperature. Given these promising characteristics, we aim to adapt our room-temperature optical ammonia sensor for clinical and medical diagnostics, enabling the detection of ultra-low ammonia levels in humans. Results also conclude that if material surfaces were modified with AuNPs can be applied for drug loading using electrostatic interaction.

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