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FABRICATION OF AG@CO$_3$(PO$_4$)$_2$ – C$_3$N$_4$ FOR BIOSENSING OF BOVINE SERUM ALBUMIN SENSOR

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Abstract
INTRODUCTION: A critical area of research in areas including healthcare, environmental monitoring, and food safety is the creation of sensitive and selective biosensors. Numerous advantages, in particular high sensitivity, rapid response, and simplicity of miniaturization, are associated with electrochemical biosensors. Recently, nanomaterial-based composites have received a lot of interest for enhancing the performance of biosensors. One such substance is Ag@Co$_3$(PO$_4$)$_2$ – C$_3$N$_4$, which combines the unique properties of graphitic carbon nitride, cobalt phosphate, and silver nanoparticles (AgNPs).
AIM: The aim of this study is to prepare the Ag@Co$_3$(PO$_4$)$_2$ – C$_3$N$_4$ nanocomposite and immobilize bovine serum albumin (BSA) on its surface for the development of a biosensor. The synthesized composite will be assessed for its potential use in the sensitive and specific detection of particular infections, contaminants, or biomarkers.
MATERIALS AND METHODS: Synthesis of Ag@Co$_3$(PO$_4$)$_2$ – C$_3$N$_4$ Nanocomposite: The Ag@Co$_3$(PO$_4$)$_2$ – C$_3$N$_4$ nanocomposite is synthesized through a suitable method, such as a one-pot hydrothermal or co-precipitation process. The Ag nanoparticles are incorporated into the Co$_3$(PO$_4$)$_2$ matrix, which is further combined with C$_3$N$_4$ to form the final nanocomposite.
RESULTS: Ag compound is highly sensitive, highly compatible, high in strength, and also highly selective. The composite material demonstrates positive results even at low concentrations of BSA, indicating its effectiveness for detecting and quantifying BSA in various samples.
CONCLUSION: The research on Ag@Co$_3$(PO$_4$)$_2$ – C$_3$N$_4$ biosensors for BSA detection provides a foundation for potential forensic applications in various areas of analysis and investigation

KEY WORDS : Ag@Co$_3$(PO$_4$)$_2$ – C$_3$N$_4$ Biosensor , bovine serum albumin , nano composite

INTRODUCTION:
Few of the sectors that give the creation of sensitive and specific biosensors a high priority include healthcare, environmental monitoring, and food safety. Biosensors provide an appealing technique to examine the content of a biological sample since they directly convert a biological event to an electrical signal. (1) For the detection and measurement of biomolecules in a variety of domains, such as healthcare, environmental monitoring, and food safety, biosensors have become highly effective tools. Accurate and dependable detection depends on the creation of biosensors with high sensitivity, selectivity, and stability.

Bovine serum albumin, a widely used model protein, serves as an important target analyte in biosensing studies. Its detection and quantification are of great significance in various applications, including clinical diagnostics, pharmaceutical research, and bioprocessing. Therefore, the development of a sensitive and reliable biosensor for BSA detection is highly desirable(2)

$g\text{-}C_3N_4$ can be used as a support material for enzymes in biosensors. Its high surface area and good biocompatibility make it an ideal platform for enzyme immobilization.(3,4) The negatively charged surface of $g\text{-}C_3N_4$ can interact with positively charged DNA, facilitating its immobilization and providing a stable and biocompatible matrix.The bandgap of semiconductor g-C3N4 is typically between 2.7 and 2.9 electron volts (eV). This qualifies it for a range of optoelectronic and electrical applications.As a result of its outstanding thermal and chemical stability, it may be used in high-temperature applications and is corrosion-resistant.(5)

$Co_3(PO_4)_2$ nanoparticles have been investigated for drug delivery applications. The porous structure and high surface area of $Co_3(PO_4)_2$allow for efficient drug loading and controlled release.$Co_3(PO_4)_2$based drug delivery systems can enhance the stability, bioavailability, and targeted delivery of therapeutic agents.(6,7)

Ag nanoparticles can serve as excellent electrode materials in electrochemical biosensors. They provide a large surface area for biomolecule immobilization and facilitate efficient electron transfer, leading to enhanced sensitivity and lower detection limits(8).BSA biosensors find applications in various biomedical research and diagnostic fields. They can be used to detect BSA as a biomarker for specific diseases or conditions(9). Additionally, BSA biosensors can aid in studying protein-protein interactions, assessing drug-protein binding, and monitoring protein adsorption on biomaterial surfaces.

The aim of the study is to analyze the biosensing activity of the fabricated compound using bovine serum albumin assay.

MATERIALS AND METHOD

The prevailing approach to synthesizing g-C3N4 involves a straightforward method known as direct thermal polymerization. In this process, a specific precursor material is placed inside either a ceramic crucible or a quartz boat. Subsequently, the setup is subjected to high temperatures, typically ranging around 500 °C. To ensure the success of the reaction, this heating step is carried out under an inert atmosphere (such as nitrogen or argon) or in a vacuum furnace. As a result of this controlled heating, the precursor material undergoes polymerization and condensation, leading to the formation of g-C3N4.

In an appropriate reaction vessel, combine the silver precursor (for example, silver nitrate, AgNO3), NABH4 as the reducing agent, and a stabilizing agent (such as a surfactant or polymer) to prevent the nanoparticles from clustering together. Thoroughly blend these constituents in the reaction vessel to achieve a consistent and even distribution.

Prepare a cobalt precursor solution by dissolving a cobalt salt, like cobalt nitrate, in water or a suitable solvent. Simultaneously, prepare a phosphate solution by dissolving a phosphate salt, such as sodium phosphate, in water or a compatible solvent. Combine the cobalt precursor solution and the phosphate solution in a sealed reaction vessel. Then, raise the temperature of the reaction vessel to a specific degree (180 °C) and maintain it at this level for 16 hours to facilitate the reaction and the subsequent formation of cobalt phosphate. Once the hydrothermal reaction is complete, cool down the reaction vessel and collect the resulting cobalt phosphate precipitate. To eliminate any impurities, cleanse the precipitate using an ethanol solvent, and then employ filtration methods to dry it. Finally, subject the dried cobalt phosphate to a temperature of 150 °C for 12 hours to complete the process.

Then we add the reduced Ag nanoparticles to the above-mentioned materials using ultrasonication for 3 hours, followed by filtration and drying for 3 hours at 90 °C

RESULT:

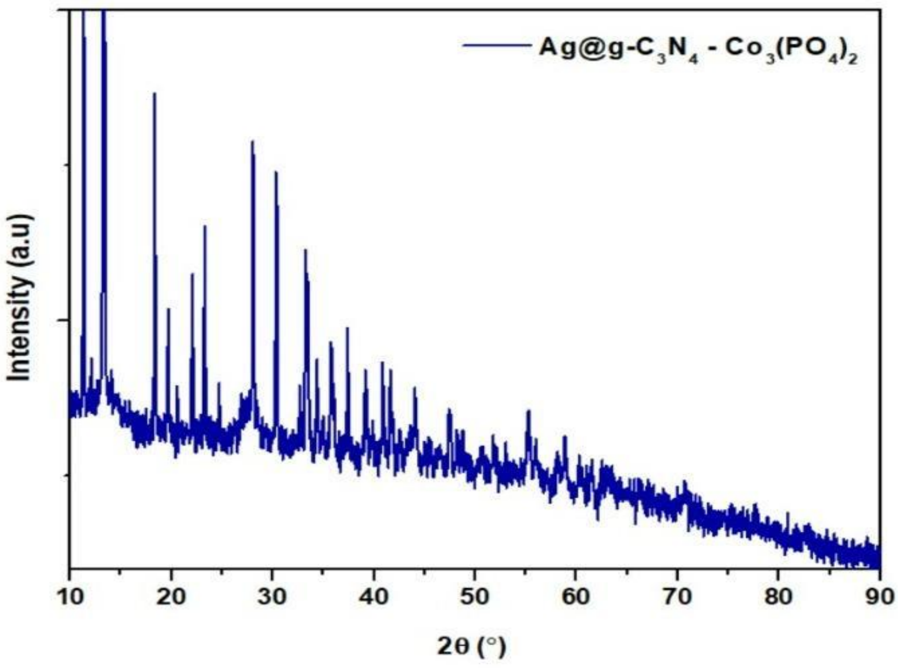

Fig1: XRD OF $Ag@Co_3(PO_4)_2$

The XRD shows highest peak at 15°

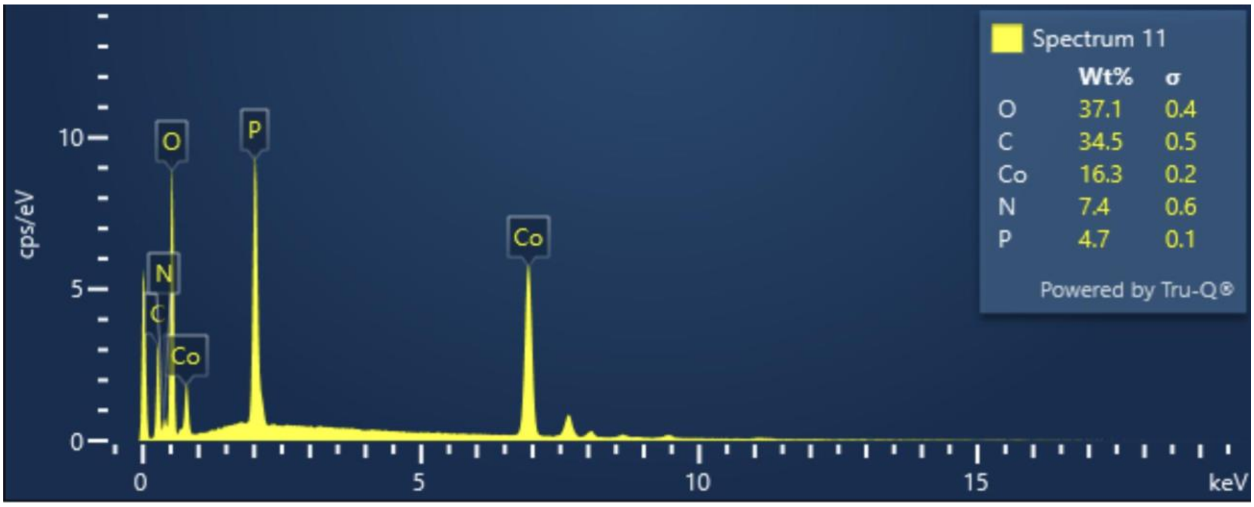

Fig2 : EDS analysis

The EDS shows the presence elements that has been formulated

Element	Line Type	Apparent Concentration	k Ratio	Wt%	Wt% Sigma	Standard Label	Factory Standard	Standard Calibration Date
C	K series	1.53	0.01531	34.51	0.50	C Vit	Yes	
N	K series	0.94	0.00168	7.43	0.64	BN	Yes	
O	K series	4.41	0.01482	37.05	0.42	SiO2	Yes	
P	K series	1.93	0.01079	4.66	0.07	GaP	Yes	
Co	K series	4.42	0.04423	16.35	0.20	Co	Yes	
Total:				100.00				

Table1: Elemental composition of C,N,O,P,Co

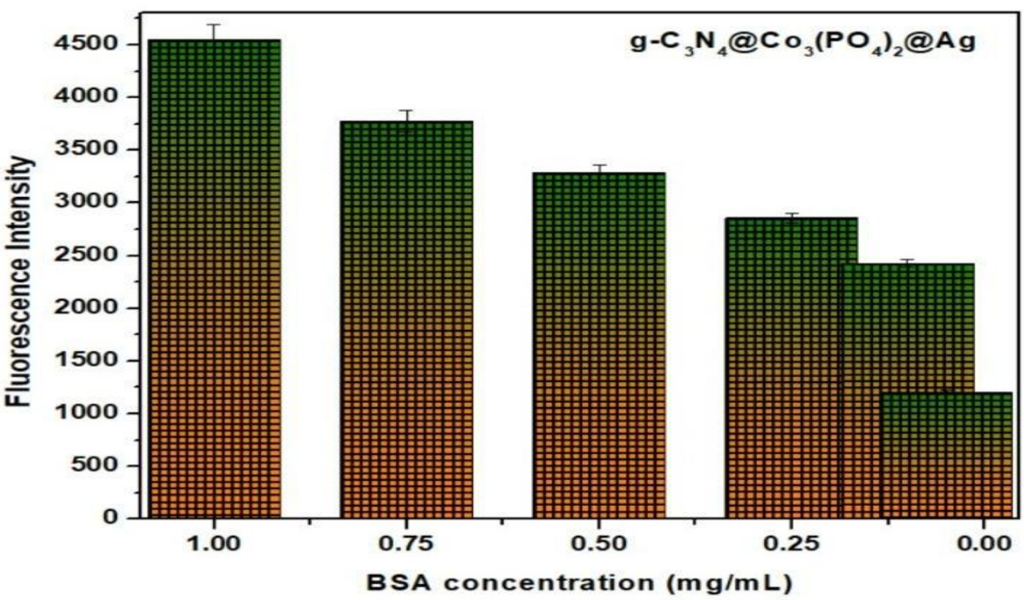

Fig3: BSA concentration

In the lowest the compound shows biosensing activity

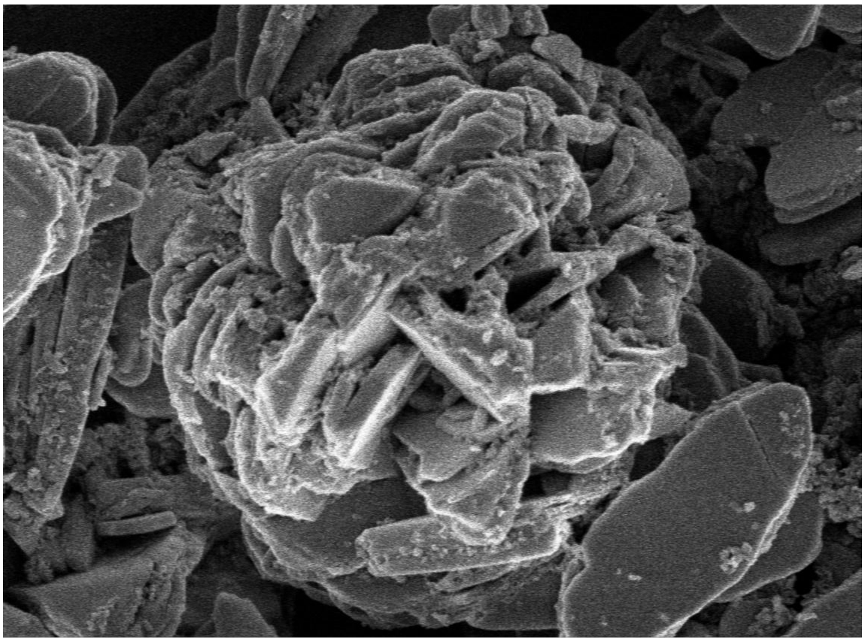

Fig4 : SEM of Cobalt Phosphate

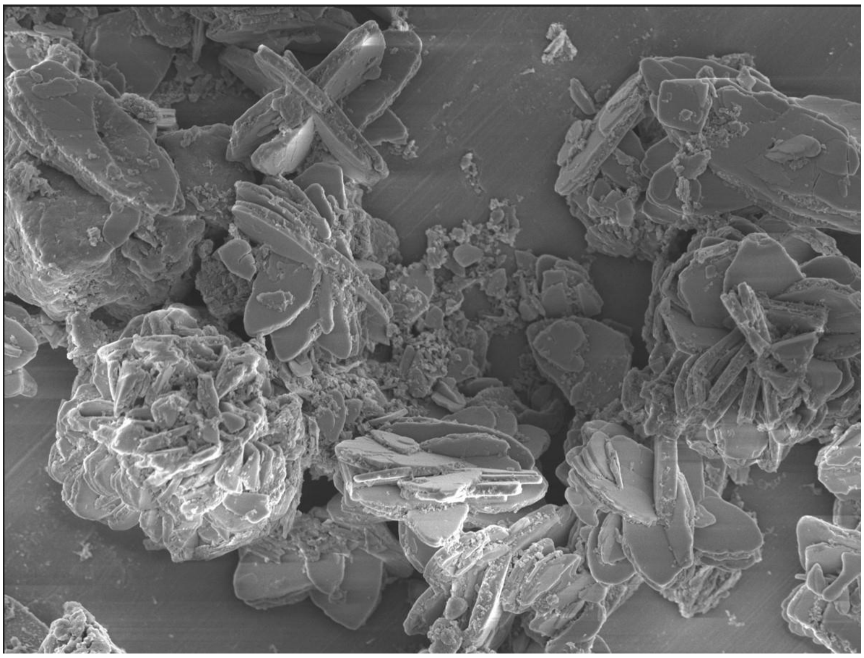

Fig5 : SEM of $Ag@Co_3(PO_4)_2$ – C_3N_4

DISCUSSION

$Ag@Co_3(PO_4)_2$–g-$C_3N_4$was synthesis by simple cost effective and environment friendly method.It was characterized by X ray diffraction study, SEM, EDS,elemental composition.

Form the XRD the material composed by amorphous and crystal in nature and the corresponding diffraction planes confirms the Ag, graphic carbon and Cobalt phosphate

The surface morphology was explained by SEM images . Cobalt phosphate looks like flower like structure and high crystallinity the flower composed by small sheets attached together,self assembled to the structure

Further it is confirmed by EDX Analysis, the material Composed by Co,P,O,C,N the absence of Ag due to low concentration.

Optimized material used for BSA molecule The result show even in the lower concentration on the material is capable to sense the BSA molecule

The use of cobalt phosphate nanostructures as a peroxidase mimic for colorimetric detection of glucose. The cobalt phosphate nanostructures exhibit high catalytic activity and can be used for sensitive glucose detection.(10)

Cobalt phosphate nanosheet-based electrochemical biosensor for the ultrasensitive detection of carcinoembryonic antigen (CEA). The cobalt phosphate nanosheets provide a large surface

area for CEA capture and exhibit excellent electrochemical properties, enabling sensitive and selective CEA detection.(11)
This review article provides an overview of cobalt phosphate nanomaterials for electrochemical biosensing applications. It discusses the synthesis methods, properties, and various biosensing strategies utilizing cobalt phosphate nanomaterials.(12)
These references highlight the potential of cobalt phosphate in biosensing applications, particularly in enzymatic and electrochemical sensing. They showcase its use for the detection of glucose, carcinoembryonic antigen (CEA), and other biomarkers. Further research and development are needed to explore the full range of applications and optimize the performance of cobalt phosphate-based biosensors.
Further research can focus on investigating the effect of different ratios of $Ag@Co_3(PO_4)_2$ nanoparticles and $g\text{-}C_3N_4$ nanosheets, as well as exploring alternative synthesis methods to enhance the performance of the nanocomposites , this research focused on BSA as the target protein, the biosensing platform can be expanded to detect other proteins of interest.

CONCLUSION

The fabrication of $Ag@Co_3(PO_4)_2\text{–}g\text{-}C_3N_4$ composite for the biosensing of bovine serum albumin (BSA) holds promise as a sensitive detection platform. The composite material demonstrates positive results even at low concentrations of BSA, indicating its effectiveness for detecting and quantifying BSA in various samples. Nevertheless, the research on $Ag@Co_3(PO_4)_2\text{–}g\text{-}C_3N_4$ biosensor for BSA detection provides a foundation for potential forensic applications in various areas of analysis and investigation.

ACKNOWLEDGEMENTS

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CONFLICT OF INTEREST

There are no conflicts of interests in the present study.

SOURCE OF FUNDING

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ETHICAL CLEARANCE

Since it is an in vitro study, an ethical clearance number is not required.

DURATION OF STUDY

The study was do for a period of 3 months

REFERENCES

1. *Garehbaghi S, Ashrafi AM, Adam V, Richtera L. Surface modification strategies and the functional mechanisms of gold nanozyme in biosensing and bioassay. Mater Today Bio. 2023 Jun;20:100656.*
2. *Jin M, Zhu S, Hou Y. Insight on Serum Albumin: From Structure and Biological Properties to Functional Biomaterials for Bone Repair. ACS Biomater Sci Eng. 2023 May 8;9(5):2235–50.*
3. *Kuan J, Zhang H, Gu H, Zhang Y, Wu H, Mao N. Adsorption-enhanced photocatalytic property of Ag-doped biochar/g-C3N4/TiO2 composite by incorporating cotton-based biochar. Nanotechnology [Internet]. 2022 May 17; Available from: http://dx.doi.org/10.1088/1361-6528/ac705e*
4. *Feng J, Chen T, Liu S, Zhou Q, Ren Y, Lv Y, et al. Improvement of g-C3N4 photocatalytic properties using the Hummers method. J Colloid Interface Sci. 2016 Oct 1;479:1–6.*
5. *Wang YX, Chen MN, Tao H. Preparation of Nested g-C_3N_4 Fiber Surrounding Graphene Oxide and Its Photocatalytic Degradation of Rhodamine B. J Nanosci Nanotechnol. 2020 Sep 1;20(9):5445–51.*
6. *López-Gallego F, Yate L. Selective biomineralization of Co3(PO4)2-sponges triggered by His-tagged proteins: efficient heterogeneous biocatalysts for redox processes. Chem Commun . 2015 May 25;51(42):8753–6.*
7. *Zheng X, Hu J, Cheng G, Xu Y, Zhao Y, Liu M. [Study on the interaction of copper-zinc superoxide dismutase with cobalt (II)-histidine by spectral analysis--II. Effect of amount of external added Co(His)n, phosphate]. Guang Pu Xue Yu Guang Pu Fen Xi. 1999 Dec;19(6):798–802.*
8. *Pan Y, Wei X, Guo X, Wang H, Song H, Pan C, et al. Immunoassay based on Au-Ag bimetallic nanoclusters for colorimetric/fluorescent double biosensing of dicofol. Biosens Bioelectron. 2021 Dec 15;194:113611.*
9. *Ambrósio JAR, Pinto BCDS, da Silva BGM, Passos JC da S, Beltrame Junior M, Costa MS, et al. BSA nanoparticles loaded-methylene blue for photodynamic antimicrobial chemotherapy (PACT): effect on both growth and biofilm formation by. J Biomater Sci Polym Ed. 2020 Dec;31(17):2182–98.*
10. *He Y, Li X, Xu X, Pan J, Niu X. A cobalt-based polyoxometalate nanozyme with high peroxidase-mimicking activity at neutral pH for one-pot colorimetric analysis of glucose. J Mater Chem B Mater Biol Med. 2018 Sep 28;6(36):5750–5.*
11. *Wang J, Hua X, Jin B. Ultrasensitive Detection of Carcinoembryonic Antigen by Chitosan/Polythiophene/CdTe Electrochemical Biosensor. ACS Omega. 2022 Dec 13;7(49):45361–70.*
12. *Holzinger M, Le Goff A, Cosnier S. Nanomaterials for biosensing applications: a review. Front Chem. 2014 Aug 27;2:63.*
13. *Sneka S, Preetha Santhakumar. Antibacterial Activity of Selenium Nanoparticles extracted from Capparis decidua against Escherichia coli and Lactobacillus Species. Research Journal of Pharmacy and Technology. 2021; 14(8):4452-4. doi: 10.52711/0974-360X.2021.00773*
14. *Vishaka S, Sridevi G, Selvaraj J. An in vitro analysis on the antioxidant and anti-diabetic properties of Kaempferia galanga rhizome using different solvent systems. J Adv Pharm Technol Res. 2022 Dec;13(Suppl 2):S505-S509. doi: 10.4103/japtr.japtr_189_22.*
15. *Sankar S. In silico design of a multi-epitope Chimera from Aedes aegypti salivary proteins OBP 22 and OBP 10: A promising candidate vaccine. J Vector Borne Dis. 2022 Oct-Dec;59(4):327-336. doi: 10.4103/0972-9062.353271.*
16. *Devi SK, Paramasivam A, Girija ASS, Priyadharsini JV. Decoding The Genetic Alterations In Cytochrome P450 Family 3 Genes And Its Association With HNSCC. Gulf J Oncolog. 2021 Sep;1(37):36-41.*