FLUORESCENT CARBON NANODOTS AS AN NATURAL ALTERNATIVE FOR INDOCYANINE GREEN TO EFFECTIVELY VISUALIZE THE ARTERIES

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ABSTRACT

This project delves into the innovative realm of biomedical imaging by exploring the utilisation of fluorescent carbon nanodots derived from beetroot as a potential natural substitute for conventional contrast agents such as Indocyanine green (ICG) in arterial imaging. By leveraging computational docking techniques, the study aims to investigate the binding efficacy between bio-active compounds found in beetroot and cardiac markers, shedding light on potential applications in cardiovascular diagnostics. The methodology encompasses several crucial steps, including phytochemical analysis of beetroot extract, synthesis of carbon nanodots, and subsequent molecular docking studies with target proteins. Through meticulous experimentation and analysis, the findings underscore strong interactions between beetroot-derived compounds and cardiac markers, offering promising prospects for enhancing biomedical imaging techniques. This research not only showcases the ingenuity of nature-inspired technologies that are also particularly in the realm of cardiovascular diagnostics. This study opens up new avenues for the development of safer with potential implications for diagnosing conditions such as atherosclerosis. Overall, this project represents a significant contribution to the field of biomedical imaging, emphasizing the importance of exploring sustainable and nature-inspired solutions for improving healthcare outcomes.

Keywords: Beta Vulgaris, Carbon nanodots, Bioimaging, Atherosclerosis

Figure 1. Beta Vulgaris

INTRODUCTION

The advent of carbon nanodots (CDs) has revolutionized nanotechnology, challenging traditional quantum dots with their remarkable fluorescence and versatile properties. Initially discovered in the mid-2000s, CDs offer advantages such as low cost, easy synthesis, and biocompatibility. Synthesized through methods like laser ablation and thermal decomposition, CDs feature a core-shell structure and surface passivation, enabling diverse applications. Our project focuses on harnessing CDs derived from beetroot through hydrothermal carbonization for biomedical imaging, particularly targeting heart biomarkers like troponin. Our research explores CDs derived from beetroot Beta vulgaris as a sustainable replacement for indocyanine green (ICG) in non-invasive heart imaging. By employing computational docking techniques, we investigate the molecular interactions between troponin and CDs. This research aligns with the shift towards sustainable healthcare solutions, emphasizing the integration of nature-inspired technologies for enhanced diagnostics and ecological stewardship.

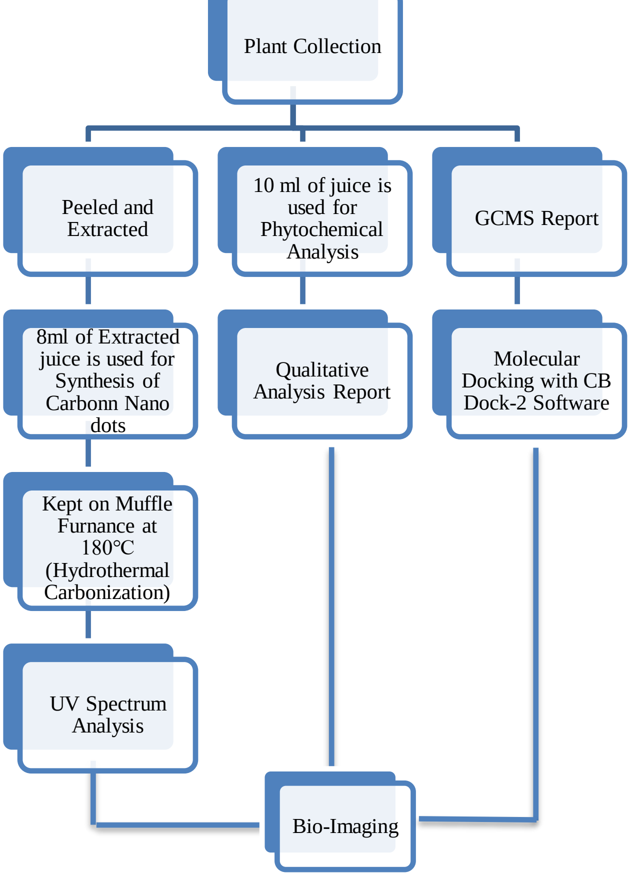
Significance of Indocyanine Green (ICG) in Bioimaging of Heart Block

Indocyanine green (ICG) is pivotal in bioimaging for heart block diagnosis and treatment. Through near-infrared fluorescence imaging (NIRF), ICG illuminates cardiac anatomy, function, and perfusion dynamics in real-time. This enables clinicians to track myocardial distribution, assess chamber dimensions, contractility, and identify perfusion deficits swiftly. Such dynamic insights aid in pinpointing structural and functional abnormalities associated with heart block, guiding interventions effectively for improved patient outcomes.

Significance of Carbon Nanodots (CNDs) in Non-Invasive Imaging of Heart Block:

Carbon nanodots (CNDs) offer unprecedented advantages in non-invasive heart block imaging. Their exceptional optical properties enable enhanced contrast imaging, providing precise visualization of affected cardiac tissues. CNDs allow real-time monitoring of cardiac function, detecting subtle dynamics changes crucial for timely intervention. Additionally, their potential in targeted drug delivery promises personalized therapeutic approaches, elevating the standard of care for heart block patients while minimizing invasiveness and maximizing efficacy.

FLOWCHART



MATERIALS AND METHODS

Plant Collection

The mature and healthy Beetroot (*Beta Vulgaris)* were collected from a local market in Trichy, Tamil Nādu, India. Upon collection, the beetroot was thoroughly washed to remove any surface impurities. Subsequently, the Beetroot were peeled, sliced, and finely grind to extract the juice. The extracted juice was then subjected to further processing for use in the project.

Extraction

A volume of 8 ml of extracted beetroot juice is filtered using filter paper to remove any impurities. The filtered juice is used for further analysis and synthesis of carbon nanodots.

Phytochemical Analysis of Beetroot - Qualitative Analysis

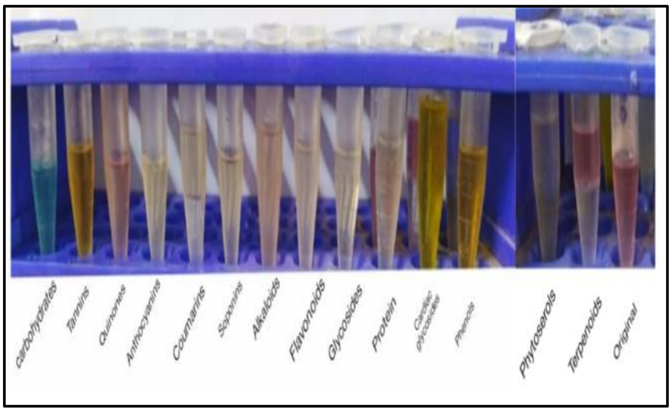


Figure 2. Phytochemical Test Results

Phytochemicals	Observations	Results
Alkaloids	No Creamy Precipitate is absorbed	-
Phenols	Dark Bluish Black is absorbed	+
Tannins	Dark Bluish Black is absorbed	+
Protein	No Blue/Violet Ring Formation	-
Carbohydrates	No Green Colour is obtained	-
Cardiac Glycosides	No Brown-Red Ring Formation	-
Phytosterols	Red Colour Formation	+
Saponins	No Persistent Foam is absorbed	-
Anthocyanins	Blue -Green Colour is absorbed	+
Coumarins	Yellow Colour is absorbed	+
Quinones	No Red Precipitate	-
Terpenoids	Reddish Brown	+

Synthesis of Carbon Nanodots

The extracted filtered juice is transferred to a crucible and subjected to hydrothermal carbonization by heating in a muffle furnace at 180°C for 90 minutes. This process transforms the beetroot juice into carbon nanodots, a crucial step in our project for synthesizing functional nanoparticles with unique properties for fluorescent and bioimaging applications.

GCMS Report

The GCMS report for the extract of *Beta Vulgaris* has been taken for identifying the bio-active components to interact with the cardiac markers (Myoglobin & Troponin-1).

SOFTWARE REQUIREMENTS

Analyzing molecular docking interactions between protein-ligand becomes an emerging tool in drug design. The molecular Docking technique was used to prove a protein (Troponin-1 and Myoglobin) that are indicators of heart damage which interacts with small molecules (ligands) or phytochemicals of plant extract.

The molecular docking studies were performed into the Core domain of human cardiac troponin (PDB code: 4Y99) and Crystal Structure of Human Myoglobin Mutant K45R (PDB code: 3RGK) to observe the mode of interaction between the identified phytochemicals in the extract of beta vulgaris and (Troponin-1 and Myoglobin). Since the active site amino acids were unknown, the blind way of docking was preferred (whole protein was docked against the phytochemicals).

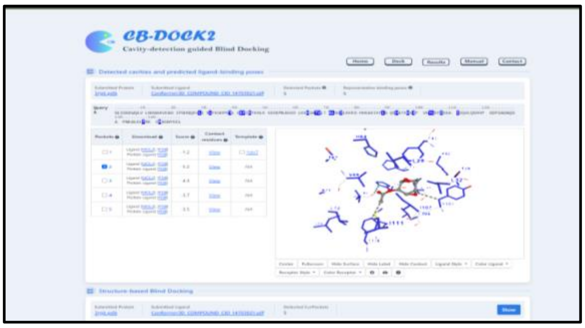


Figure 3. CB-DOCK 2 Software

GC-MS Analysis of *Beta Vulgaris* Extract

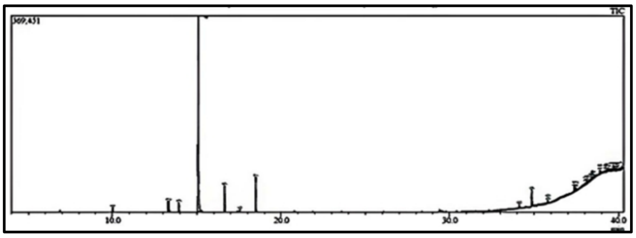


Figure 4: GC-MS report

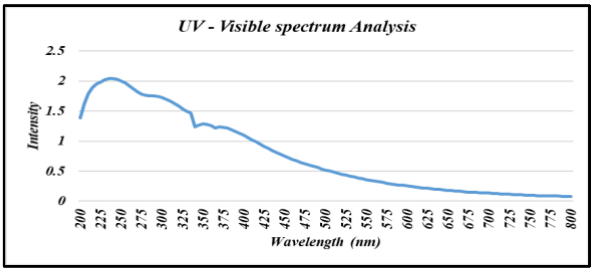
RESULT

Our Project "Fluorescent Carbon nanodots as a natural alternative for Indocyanine green (ICG) to effectively visualize the arteries" integrated with computational docking techniques offers a novel and versatile approach to arterial visualization and cardiovascular diagnostics. This innovative system harnesses the unique optical properties of carbon nanodots derived from beetroot *Beta vulgaris*, synthesized through hydrothermal carbonization. These carbon nanodots serve as a natural alternative to conventional contrast agents like Indocyanine green (ICG), overcoming the limitations associated with invasive administration and limited tissue penetration.

The Phytochemical qualitative analysis of beetroot extract revealed the presence of various bioactive compounds, including phenols, glycosides, flavonoids, tannins, coumarins, anthocyanins, terpenoids and phytosterols indicating their utility in fluorescence applications. Through computational docking studies using CB DOCK-2 docking tool, the binding efficacy between identified phytochemicals in beetroot extract and cardiac markers such as Troponin-1 and Myoglobin was investigated. The results revealed strong interactions between the bio-active compounds and the target proteins, suggesting their potential utility in cardiovascular diagnostics and therapeutics.

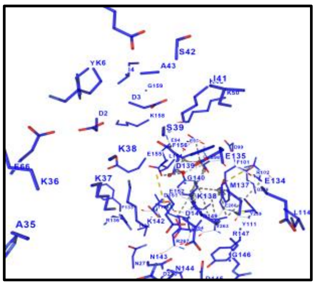
Our docking study revealed strong binding interactions between troponin 1 and myoglobin proteins with beetroot aquas ligands. Troponin 1 showed ionic and hydrogen bond interactions with acetic acid, 2-methyl pyrazine, N-nitroso-N-methyl urethane, N-formyl beta alanine, N2-pyrrolidin ylidene, and methyl linoleate. Myoglobin exhibited ionic interactions and hydrophobic contacts with acetic acid and palmitic acid.

CHARACTERISATION OF *Beta Vulgaris*:



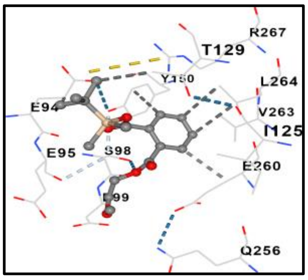
The UV-Visible spectrum for the synthesised nanodots were performed in the wavelength range from 200 to 800 nm and the results showed the indication of peaks at 240 nm and 370 nm which confirms the synthesis of carbon nanodots with fluorescent property.

Figure 5. Carbon Nano Dots



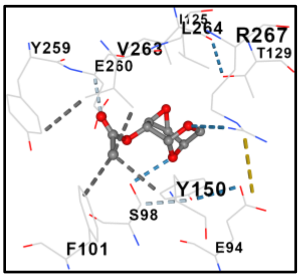
Butyl 2-methoxyethyl phthalate 91694261

Binding Energy: 5.6



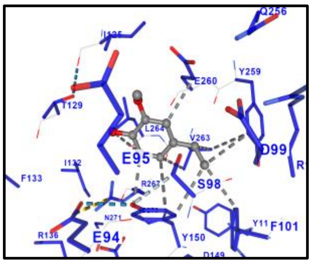
9,12-Octadecadienoic acid 3931

Binding Energy: 5.6



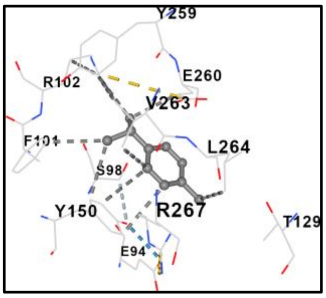
1,6;3,4-Dihydro-2-O-Acetyl-Beta-D Galactopyranose 14703021

Binding Energy: 5.7



2-Methoxy-4-vinylphenol 332

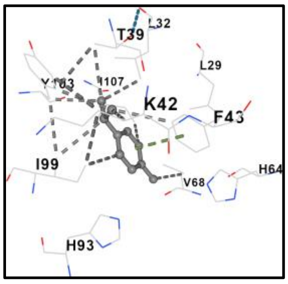
Binding Energy: 5.6



p-Cymene 7463

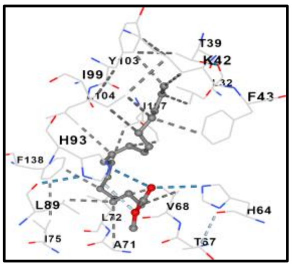
Binding Energy:5.9

Figure 6. Interaction Between Troponin-1 and Ligand



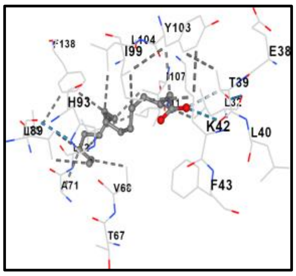
p-Cymene 7463

Binding Energy:6.2



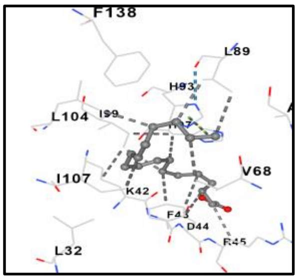
Methyl linoleate 528441

Binding Energy:6.6



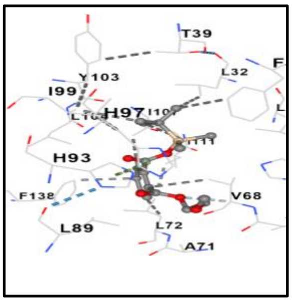
9,12-Octadecadienoic acid 3931

Binding Energy:6.1



Palmitic acid 985

Binding Energy: 6.3



Butyl 2-methoxyethyl phthalate 91694261

Binding Energy: 6.5

Figure 7 Interaction Between Myoglobin and Ligand

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