

Department of Microbiology

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- **RT:** Biological Activities of *Cassia sieberiana* and *Detarium microcarpum* Extracts against Gastroenteritis-Associated Bacteria from Patients at Olabisi Onabanjo University Teaching Hospital
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AB: Gastroenteritis, a clinical condition involving frequent passage of watery or bloodstained stool, accounts for substantial morbidity and mortality among patients of all ages. Antibiotic treatment is almost ineffective due to inability of drugs to reach target sites. Studies have shown that silver nanoparticles (AgNPs) ensure effective drug delivery at target site. However, green synthesis of AgNPs using stem bark extracts of *Cassia sieberiana* and *Detarium microcarpum* have not been reported. Hence, biological activities of *C. sieberiana* and *D. microcarpum* crude and AgNPs extracts against gastroenteritis-associated bacteria from patients at Olabisi Onabanjo University Teaching Hospital (OOUTH) were investigated.

Dried stem bark of *C. sieberiana* and *D. microcarpum* were purchased and pulverized. Extracts were obtained by maceration using acetone, ethanol and methanol as solvents and their AgNPs synthesized respectively. Qualitative phytochemical screening was carried out. Chemical compounds were investigated using Gas Chromatography-Mass Spectrometry (GC-MS). The crude and AgNPs extracts were characterized using UV-Visible Spectroscopy (UV-VIS), Fourier Transformed Infrared Spectroscopy (FTIR), X-ray Diffraction (XRD), Scanning Electron Microscopy (SEM) and Transmission Electron Microscopy (TEM). Radical scavenging activities and phenolic contents were determined by Nitric Oxide (NO), Ferric Reducing Antioxidant Potential (FRAP), Lipid Peroxidation (LP), 2,2-Diphenyl-1-picrylhydrazyl (DPPH), Total Alkaloid Capacity (TAC), Total Flavonoid (TF) and Total Phenolic (TP) assays. Enteric bacteria from patients' stool samples at OOUTH were

identified using 16S rRNA gene sequencing. Antibacterial susceptibility and Minimum Inhibitory Concentrations (MICs) were determined. Data were analyzed using descriptive statistics.

Phytochemicals in the crude extracts were saponins, alkaloids, flavonoids, steroids, tannins, phlobatannins, terpernoids, cardiac glycosides, phenols and reducing sugar. The GC-MS analysis of C. sieberiana showed 65, 49, 51, 3, 3, and 4 chemical compounds in acetone, ethanol. methanol, acetone-AgNPs, ethanol-AgNPs and methanol-AgNPs extracts, respectively. The GC-MS analysis of D. microcarpum revealed 58, 37, 50, 3, 9 and 3 chemical compounds in acetone, ethanol, methanol, acetone-AgNPs, ethanol-AgNPs and methanol-AgNPs extracts, respectively. The FTIR identified biomolecules (proteins, carboxylic acid, amines and phenolic compounds) responsible for biogenic reduction of AgNPs at 3400-1645 cm-¹. The XRD indicated clear and distinct peaks confirming high purity and crystalline nature of phytosynthesized AgNPs. The SEM and TEM showed the AgNPs extracts were spherical in shape with average sizes \geq 50 nm. Some of nanocrystallite AgNPs were agglomerated and gave rise to lump due to the presence of extracting solvents and other attractive forces. The values (radical scavenging activities) of NO, FRAP, LP, DPPH, TAC, TFC and TPC in crude extracts were in the range of 20.09 to 70.77 µg AAE/mg, while NO, FRAP, LP, DPPH, TAC, TFC and TPC values in AgNPs extracts were 48.00-81.87 µg AAE/mg, respectively. The extracts showed MIC values of >50 µg/mL, while AgNPs showed MIC ranging between 0.35 and 1.40 µg/mL against Escherichia coli, Morganella morganii, Enterobacter hormaechei and Shigella flexneri.

The study concluded that *C. sieberiana* and *D. microcarpum* AgNPs extracts displayed higher biological activities against gastroenteritis-associated bacteria compared to crude extracts. The exploration and development of novel antimicrobials as well as toxicological study of the crude and AgNPs extracts are recommended for further studies.

Keywords: AgNPs, Chemical compounds, C. sieberiana, D. microcarpum, Gastroenteritis

Word Count: 500

Abbreviations: RFN: Researcher's Full Name, RD: Researcher's Department, RS: Researcher's School, RE: Researcher's Email, RAE: Researcher's Alternate Email, RP: Researcher's Phone Contact, RT: Registered Title, MS: Main Supervisor, ME: Main Supervisor's E-mail Address, SP: Main Supervisor's Phone Contact, CS: Co-Supervisor, CE: Co-Supervisor's E-mail Address, CP: Co-Supervisor's Phone Contact, AB: Abstract

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