

**MAKERERE**



**UNIVERSITY**

**DEVELOPMENT OF A BIOACTIVE GAUZE DRESSING FUNCTIONALIZED WITH  
HERB-LOADED NANOPARTICLES TO FACILITATE WOUND HEALING**

**BY**

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## **DECLARATION**

I, **Namuga Catherine**, declare that this thesis is my original work and has not been submitted to any other university or institution for the award of any degree or academic qualification. Any work of others that has been used in this thesis has been duly acknowledged and appropriately referenced.

**Namuga Catherine**

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## LIST OF ABBREVIATIONS

**AFM:** Atomic Force Microscope

**AHL:** N-Acyl Homoserine Lactone

**ANOVA:** Analysis Of Variance

**ARRIVE:** Animal Research Reporting of In vivo Experiments

**ASTM:** American Society for Testing and Materials

**ATR:** Attenuated Total Reflectance

**ATCC:** American Association of Textile Chemists and Colourists

**CEDAT:** College of Engineering, Art, Design, and Technology

**COVAB:** College of Veterinary Medicine, Animal Resources and Biosecurity

**DLS:** Dynamic Light Scattering

**DMSO:** Dimethyl Sulphoxide

**DPPH:** 2, 2-diphenyl-1-picrylhydrazyl

**DGAL:** Directorate of Government Analytical Laboratory

**EPS:** Extracellular Polymeric Substance

**ECM:** Extracellular Matrix

**FTIR:** Fourier Transform Infrared

**FRAP:** Ferric Reducing Assay

**FWHM:** Full Width with the Half-Maximum

**GC-MS:** Gas Chromatography-Mass spectrometry

**HSD:** Honestly Significant Difference

**ICH:** International Council for Harmonisation

**IRB:** Institutional Review Board

**LC-MS:** Liquid Chromatography-Mass spectrometry

**MBEC:** Minimum Biofilm Eradication Concentration

**MBIC:** Minimum Biofilm Inhibition Concentration

**MDR:** Multi-Drug Resistant

**MHB:** Mueller Hinton Broth

**MIC:** Minimum Inhibitory Concentration

**MRM:** Multiple Reaction Monitoring

**MRSA:** Methicillin-Resistant Staphylococcus Aureus

**MTP:** Microtiter Plate

**MSCRAMM:** Microbial Surface Components Recognising Adhesive Matrix Molecules

**NCRI:** Natural Chemotherapeutic Research Institute

**NDP:** National Development Plan

**NIST:** National Institute of Standards and Technology

**OECD:** Organisation for Economic Co-operation and Development

**ORAC:** Oxygen Radical Absorbance Capacity

**PBS:** Phosphate Buffered Saline

**PDI:** Polydispersity Index

**PII:** Primary Irritation Index

**RSM:** Response Surface Methodology

**ROS:** Reactive Oxygen Species

**SDG:** Sustainable Development Goals

**SEM:** Scanning Electron Microscopy

**SVARREC:** School of Veterinary and Animal Resources Research Ethics Committee

**TEAC:** Trolox Equivalent Antioxidant Capacity

**TEM:** Transmission Electron Microscopy

**T-PACC:** Textile Protection and Comfort Centre

**TRAP:** Total Peroxyl Radical-Trapping Antioxidant Parameter

**TOSC:** Total Oxyradical Scavenging Capacity

**UIRI:** Uganda Industrial Research Institute.

**UNCST:** Uganda National Council of Science and Technology

**UV:** Ultraviolet

**UV-Vis:** Ultraviolet Visible

**VRE:** Vancomycin-Resistant Enterococci

**WHO:** World Health Organisation

**XRD:** X-ray Diffraction

**XPS:** X-ray Photonic Spectroscopy

## LIST OF PUBLICATIONS

1. Namuga, C., Muwonge, H., Nasifu, K., Sekandi, P., Sekulima, T., & Kirabira, J. B. (2024). *Hoslundia opposita* vahl; a potential source of bioactive compounds with antioxidant and antibiofilm activity for wound healing. *BMC Complementary Medicine and Therapies*, 24(1), 236. <https://doi.org/10.1186/s12906-024-04540-z>.
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3. Namuga, C., Muwonge, H., Lubwama, M., Janet, N., Sekulima, T., & Kirabira, J. B. (2022). Antibacterial activities of *Bidens pilosa* L, *Hoslundia opposita* Vahl, and *Ageratum conyzoides* L against some common wound pathogens. *African Journal of Pharmacy and Pharmacology*, 16(5), 64-78 <https://doi.org/10.5897/AJPP2022.5296>.
4. Namuga, C., Ocan, M., Kinengyere, A. A., Richard, S., Namisango, E., Muwonge, H., & Obuku, E. A. (2023). Efficacy of nano-encapsulated herbal extracts in the treatment of induced wounds in animal models: a systematic review protocol. *Systematic Reviews*, 12(1), 215. <https://doi.org/10.1186/s13643-023-02370-7>

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## ABSTRACT

Wound healing is crucial in healthcare due to its significant physiological and economic impacts on patients. Among wound dressings, gauze is the most widely used; however, it is associated with a high risk of bacterial wound infections that delay healing. Hence, enhancing its effectiveness remains critical. In Uganda, herbal medicines are still utilised in the treatment of wounds and other illnesses; however, they require high dosages and prolonged treatment, leading to poor patient compliance. Nanoencapsulation offers a promising solution by improving the drug's therapeutic effect through sustained release while minimising toxicity and dosage. The main objective of this study was to develop and evaluate a gauze dressing functionalized with herb-loaded chitosan nanoparticles for enhanced wound healing. This study explored the extraction of selected medicinal herbs (*Bidens pilosa* L., *Ageratum conyzoides* L., and *Hoslundia opposita* Vahl) using different methods and solvents with varying polarity. The extraction yield, in vitro antibacterial (on *Staphylococcus aureus*/Methicillin-Resistant *Staphylococcus Aureus* (MRSA), *Enterococcus faecalis*, *Escherichia coli*, *Klebsiella Pneumoniae*, and *Pseudomonas aeruginosa*), antibiofilm, and antioxidant activities, as well as phytochemicals present, were determined. The most efficacious extract was subjected to GC-MS and LC-MS analysis. It was nanoencapsulated in chitosan nanoparticles via the ionic gelation method, and optimisation was performed using Response Surface Methodology (RSM). The herb-loaded nanoparticles were evaluated for in vitro antibacterial activity and incorporated into the gauze dressing. Their in vitro antibacterial activity, in vivo wound healing in Wistar rats, water absorption, and retention capacity, as well as in vivo skin irritation in rabbits, were tested. High extraction yields were obtained for samples extracted with highly polar solvents. The methanol (100%) extract of *H. opposita* extracted by maceration, displayed better bioactivity (antibacterial, antibiofilm and antioxidant). The biological activity of the plants was attributed to the presence of various phytochemicals. The herb nanoparticles obtained were spherical with size 212 nm, zeta potential 40 mV, Polydispersity Index (PDI) 0.22, encapsulation efficiency 79.1% and loading capacity 9.82 %. Their in vitro drug-release profile showed sustained release of 50% over 24 hours, compared with 100% for the free extract. They exhibited enhanced antibacterial activity with minimum inhibitory concentrations (1.875 to 3.275 mg/mL) against *S. aureus*, MRSA, *E. faecalis*, *E. coli*, *K. pneumoniae*, and *P. aeruginosa*. Gauze incorporated with nanoparticles exhibited no antibacterial activity but significantly accelerated wound healing, achieving 93% wound closure by day 18 compared with 41% in untreated gauze. The dressing also exhibited improved water-holding capacity; no skin irritation was observed in rabbits, and its water absorption capacity remained unaltered. Extract fractionation identified rutin and 4<sup>111</sup>-acetylxitexin-2<sup>11</sup>-O-rhamnoside as the possible major bioactive compounds in the

methanol extract of *H. opposita*. In conclusion, integrating the *H. opposita* extract-loaded chitosan nanoparticles into gauze significantly enhanced wound healing, offering a promising advancement in wound care technology.

**Keywords:** *Wound healing, gauze dressing, herbal medicine, chitosan nanocarriers, nanoencapsulation, antibacterial activity, phytochemical analysis.*