# Quality Control studies and Ethnopharmacological evaluation of *Natsiatum herpeticum* Buch.-Ham. ex Arn.

**Thesis submitted in partial fulfilment for the**

**Award of Degree**

## *Doctor of Philosophy*


By

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It is further certified that the student has fulfilled all the requirements of Comprehensive Examination, Candidacy and SOTA for the award of Ph.D. Degree.

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I, Ms. Sangeeta Hazarika, certify that the work embodied in this thesis is my own bonafide work and carried out by me under the supervision of Prof. Siva Hemalatha from July 2017-July 2023, at the Department of Pharmaceutical Engineering & Technology, Indian Institute of Technology (BHU), Varanasi. The matter embodied in this thesis has not been submitted for the award of any other degree/diploma. I declare that I have faithfully acknowledged and given credits to the research workers wherever their works have been cited in my work in this thesis. I further declare that I have not wilfully copied any other's work, paragraphs, text, data, results, etc. reported in journals, books, magazines, reports dissertations, theses, etc., or available at websites and have not included them in this thesis and have not cited as my own work.

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It is certified that the above statement made by the student is correct to the best of my knowledge.

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## ABBREVIATIONS

AAS : Atomic Absorption Spectroscopy

ALP : Alkaline phosphatase

ALT : Alanine transaminase

AMR : Antimicrobial resistance

ANOVA : Analysis of Variance

AST : Aspartate aminotransferase

BLAST : Basic Local Alignment Search Tool

CBOL : Consortium for the Barcode of Life

CPCSEA : Committee for the Purpose of Control and Supervision of Experiments on Animals

DHFR : Dihydrofolate reductase

DMNC : Density of Maximum Neighbourhood Component

DNA : Deoxyribonucleic acid

dNTPs : Deoxynucleoside triphosphates

EPC : Edge Percolated Component

FDR : False Discovery Rate

GHS : Globally Harmonized System

GO : Gene Ontology

$IC_{50}$ : Inhibitory Concentration 50

IRAK4 : Interleukin-1 receptor-associated kinase 4

KEGG : Kyoto Encyclopaedia of Genes and Genomes

LC-MS : Liquid Chromatography-Mass Spectrometry

LC-MS-MS : Liquid Chromatography Tandem Mass Spectrometry

$LD_{50}$ : Lethal Dose 50

MBC : Minimum Bactericidal Concentration

MCC : Maximal Clique Centrality

MD : Molecular Dynamics

MHA : Mueller-Hinton agar

MHB : Mueller-Hinton broth

MIC : Minimum Inhibitory Concentration

MM-GBSA : Molecular Mechanics Generalized Born Model and Solvent Accessibility

MNC : Multinetwork Clustering

NCBI : National Center for Biotechnology Information

NIH : National Institutes of Health

OECD : Organisation for Economic Co-operation and Development

PBP : Penicillin binding protein

PCR : Polymerase Chain Reaction

PPI : Protein-protein interaction

QToF : Quadrupole Time-of-Flight

QToF-MS : Quadrupole Time-of-Flight Mass Spectrometer

rGy : Radius of Gyration

RMSD : Root Mean Square Deviation

RMSF : Root Mean Square Fluctuation

SDG : Sustainable Development Goals

TNF : Tumour necrosis factor

TRAF6 : Tumour necrosis factor receptor–associated factor 6

UV : Ultraviolet

WHO : World Health Organization

## PREFACE

Natural resources, particularly plants, have long been used as a source of therapeutics, and over the past centuries, many valuable drugs have been created from these resources. They have been the source of privileged scaffolds for half of all medicinal compounds that are produced synthetically due to their complex structural spaces and physicochemical properties. Emerging risks to human health necessitate a concerted effort to find both preventive and therapeutic approaches, with natural products at the centre of initiatives to develop novel treatments and lower disease transmission and associated mortality. *Natsiatum herpticum* Buch.-Ham. ex Arn., a least-explored plant, is being considered a wild edible plant by some ethnic groups. In spite of several previous publications suggesting its ethnopharmacological use by different tribes, no documentation or scientific approaches have been made hitherto to validate its ethnopharmacological claims. The research work presented in the thesis addresses four objectives. The first objective of the study is to validate the pharmacognostical quality control assessment of *N. herpeticum* in accordance with standard guidelines. The second objective is to determine the toxicity profile of the plant. The third objective includes the qualitative screening of the aqueous plant extract and network pharmacology-based prediction of the antibacterial and anti-inflammatory potential of the detected compounds. The fourth objective deals with the experimental validation of the pharmacological activity of the aqueous extract of *N. herpeticum* using in silico-in vitro-in vivo approach.

The work embodied in this thesis has been organised into the following chapters:

**Chapter 1.** This chapter introduces the significance of natural products in drug discovery and development and discusses the significance of modern tools in natural product research. Further, the available ethnopharmacological claims on *N. herpeticum* have been described.

**Chapter 2.** This chapter deals with the literature available on *N. herpeticum*. It further summarises the application of different methodologies in natural product research.

**Chapter 3.** This chapter includes the rationale, objectives, and plan of work.

**Chapter 4.** In this chapter, a description of the methodologies employed in the research work has been provided.

**Chapter 5.** This chapter includes the findings of the quality control assessment, toxicity studies, network pharmacology-based prediction, and pharmacological evaluations.

**Chapter 6.** This chapter discusses the major findings of the study and their implications. Further, it describes the current limitations of the study.

**Chapter 7.** This chapter summarises the key points of the findings from the study and presents future prospective.

Apart from this, the references and appendix, including the additional supplementary data supporting the main findings, have been included.