RESEARCH PAPER

Dung Beetle Optimization Framework for Bioactive Compound Prioritization Based on Phytochemical and Gc–Ms Characterization

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Received: 24th April, 2026; Revised: 20th May 2026; Accepted: 30th June, 2026; Available Online: 10th July, 2026

ABSTRACT
As the emergence of antibiotic resistance is becoming a concern, more natural bioactive compounds are being considered as alternative medicines. Among the various medicinal plants, Bacopa monnieri, one of the plant species, gained an extensive popularity due to its varied range of phytochemistry with many kinds of therapeutic activities. Herein, we suggest a Dung Beetle Optimization-Based Antimicrobial Potency Model (DBO-APM) to introduce computational method in prioritizing potential phytochemical associated with the activity. Fresh Bacopa monnieri extracts was screened qualitatively and quantitatively, and analyzed by using Gas Chromatography-Mass Spectrometry (GC-MS) along with qualitatively tested for antimicrobial activity against selected Gram-positive bacteria, Gram-negative bacteria and fungi. Qualitative phytochemical screening results confirmed presence of alkaloids, carbohydrates, glycosides, phenols, and terpenoids and absence of proteins, amino acids, and saponins. In quantitative phytochemical analyses, glycosides showed maximum quantity (5.24 mg/g), followed by total phenolics (0.73 mg/g) and flavonoids (0.434 mg/g), demonstrating phytochemical analysis of rich bioactive secondary metabolites. GC-MS results show existence of many volatile phyto-constituents indicates a complex chemistry of the extract could have potential antimicrobial properties. Therefore, proposed DBO-APM utilizing normalized data of these phytochemical parameters and features can represent and prioritizing based on optimization the phytochemicals having higher contribution on Antimicrobial Potency of a given medicinal plant extracts.

Keywords: Bacopa monnieri, Dung Beetle Optimization, phytochemical analysis, antimicrobial potency, bioactive compound prioritization, medicinal plants.

How to cite this article: Kavitha A, Jayachitra A, Dung Beetle Optimization Framework for Bioactive Compound Prioritization Based on Phytochemical and Gc–Ms Characterization Int J Drug Deliv Technol. 2026;16(6):466-480. DOI: 10.25258/ijddt.16.6.172
Source of support: Nil.
Conflict of interest: None

1. INTRODUCTION

The fast, kind of sudden emergence of antimicrobial resistance (AMR) has become one of the most critical issues for modern healthcare systems worldwide. The steady rise of multidrug-resistant bacterial and fungal pathogens has basically lowered the effectiveness of the usual antibiotics, which then causes longer treatment durations, higher mortality rates, and real economic strain on healthcare infrastructures [13]. As a result, there is an obvious call to find other antimicrobial options that come from natural sources, those options should fight resistant microorganisms, and at the same time reduce unwanted side effects

Medicinal plants have long been seen as a kind of reservoir for biologically active substances, with a wide range of pharmacological actions. Plant-derived secondary metabolites like phenols flavonoids alkaloids, glycosides saponins, and terpenoids have shown strong antimicrobial, antioxidant, anti-inflammatory, and anticancer potential [11]. More recent work has pointed out that phytochemical-rich medicinal plants may serve as viable alternatives to synthetic antimicrobials, mainly because these plants can hit multiple microbial routes at the same time [13, 16].

Among many medicinal plants, Bacopa monnieri gets special attention in the literature, partly due to its many traditional uses in home medicine and related systems. This plant is commonly described as neuroprotective, antioxidant, anti-inflammatory, and antimicrobial, with much of that activity linked to its varied phytochemical makeup [5]. Recent studies also mention several bioactive constituents, for example bacosides, flavonoids, alkaloids, and phenolic compounds, which help explain why it performs well in pharmacological terms [6–8]. Also, because people are increasingly interested in plant based therapies, researchers keep checking the antimicrobial ability of Bacopa monnieri against clinically relevant bacterial and fungal pathogens

Phytochemical screening plus quantitative analysis is kind of the backbone for understanding how much bioactive compounds exist and where they are found inside plant extracts [11,12]. At the same time, GC-MS has grown into a strong analytical technique to detect volatile and semi-

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volatile phytoconstituents that may connect with biological activity [9,10]. Using GC-MS profiles, scientists can map complicated phytochemical mixtures and then pick out compounds that might be responsible for antimicrobial effects. However, typical phytochemical and GC-MS studies usually stop at identification and abundance estimates, they do not really build a quantitative link between phytochemical composition and antimicrobial efficacy.

To test antimicrobial performance, agar well diffusion assays and Minimum Inhibitory Concentration (MIC) studies are usually employed for evaluating microbial susceptibility toward plant extracts [14–16]. These approaches certainly provide useful inhibition efficiency info, but they still can't clarify which specific phytochemicals matter most for the antimicrobial results. Because of this, the interpretation of antimicrobial findings often stays descriptive, rather than being genuinely predictive.

In parallel, computational intelligence and bio-inspired optimization algorithms have opened fresh possibilities for analyzing complicated biological datasets [17–19]. Optimization methods have shown meaningful success in feature selection, parameter tuning, pattern recognition, and decision-support, especially across biomedical and pharmaceutical contexts [20-22]. One algorithm that has gained attention is Dung Beetle Optimization (DBO), which is viewed as promising meta-heuristic thanks to its balanced exploration–exploitation behavior, solid convergence properties, and its tendency to reduce getting stuck in local optima [1–4]. With inspiration from dung beetles rolling, breeding, foraging, and stealing behaviors, DBO has managed to perform well across a variety of optimization tasks.

Even with progress in phytochemical characterization, antimicrobial evaluation, and optimization related research, there are still fewer studies that combine everything into one single medicinal plant assessment framework. So, this work introduces a DBO-Based Antimicrobial Potency Model (DBO-APM) for Bacopa monnieri. This framework merges qualitative plus quantitative phytochemical characterization, GC-MS profiling, antimicrobial activity measurement, MIC determination, and DBO based prioritization of bioactive compounds. Moreover, a new Antimicrobial Potency Index (API) is proposed, which aims to connect phytochemical abundance with antimicrobial efficacy in a measurable, quantitative way. Overall, the proposed workflow seeks to highlight key antimicrobial phyto-constituents, and to give a computationally optimized strategy for evaluating medicinal plants and discovering natural antimicrobials.

2. LITERATURE REVIEW

Medicinal plant research has grown fast in recent years because people are more and more interested in natural therapeutic agents that can cope with antimicrobial resistance. A bunch of studies, showed that phytochemicals like phenols, flavonoids, alkaloids, glycosides, saponins, and terpenoids, play key parts in suppressing microbial growth through membrane disruption, enzyme deactivation, oxidative stress induction, and even interference with cellular signaling routes [11,13]. So, phytochemical characterization basically became a core part of medicinal plant assessment.

More recent work on Bacopa monnieri, points to a pretty wide set of pharmacological effects linked to its bioactive constituents. Fatima et al. [5] reviewed the therapeutic potential of Bacopa monnieri, and they really emphasized how bacosides together with phenolic compounds contribute to biological activity. After that, other studies found that Bacopa monnieri extracts show solid antioxidant, anti-inflammatory, and antimicrobial effects as a result of synergistic interactions among different phytochemical groups [6–8]. However, even though these works described the phytochemical profile and the biological responses, they mostly stayed descriptive, not doing a quantitative evaluation of how much each individual phytochemical contributes to antimicrobial efficacy.

GC-MS analysis has also become a standard go-to method for finding bioactive compounds in medicinal plants. Scientific Reports published studies, indicating that GC-MS characterization can identify antimicrobial phytocomponents efficiently, and it can also give extra detail like compound abundance plus chemical variety [9]. Further investigations, also supported GC-MS profiling as useful for locating biologically active compounds in medicinal plants, especially those targeted toward pharmaceutical use [10]. That said, many GC-MS papers still stop at identification only, and do not include computational strategies that help prioritize compounds specifically by their antimicrobial relevance.

When it comes to antimicrobial testing, assessment methods are among the most used routes for evaluating medicinal plant extracts. Many recent papers used agar diffusion assays and MIC determination methods, to examine antibacterial and antifungal effects against Gram-positive bacteria, Gram-negative bacteria, and fungal pathogens [14–16]. Generally, the outcomes suggest that extracts rich in phytochemicals have promising antimicrobial potential. Still, the inhibition zone numbers and MIC values on their own, can't really tell which individual phytochemical(s) are the main drivers behind the microbial inhibition.

To deal with that kind of gap, computational intelligence techniques have been increasingly merged into biological and pharmaceutical research. Artificial intelligence, machine learning, and optimization-based methods have been employed for compound screening, drug discovery, biomarker identification, and feature prioritization [17–20]. These tools help researchers pull out patterns from complicated datasets and they also support better decision-making. In particular, optimization algorithms have often

been quite effective at spotting influential variables inside multidimensional biological systems.

Among metaheuristic approaches, Dung Beetle Optimization has received a lot of interest, mainly because it has efficient search mechanisms and good convergence behavior. The original DBO algorithm proposed by Xue and Shen [1] showed better optimization results, compared with multiple conventional metaheuristic strategies. Later updates added quantum computing ideas, multi-objective optimization, and fractional-order search dynamics, which strengthened exploration and improved solution quality [2–4]. Even so, DBO usage in medicinal plant analysis, phytochemical prioritization, GC-MS interpretation, and antimicrobial potency prediction is still basically underexplored.

So, a critical research gap exists for building a framework that can handle phytochemical screening, GC-MS characterization, antimicrobial evaluation, MIC determination, and optimization-based bioactive compound prioritization all at once. Most current studies look at these pieces separately, so the quantitative link between phytochemical composition and antimicrobial efficacy remains unclear. If this gap is addressed, it could improve medicinal plant evaluation, and speed up the identification of high-potential antimicrobial compounds.

Based on these limitations, the present study introduces a DBO-APM that merges phytochemical analysis, GC-MS characterization, antimicrobial susceptibility testing, MIC determination, and computational optimization into one unified pipeline. The proposed workflow is expected to deliver a more systematic and quantitative way, to identify the Bacopa monnieri phytochemical constituents that are most responsible for antimicrobial activity.

Problem Statement

Medicinal plants have been widely explored as natural source of antimicrobial agent due to the richness of their phytochemical constituents and various pharmacological properties. The leaves of Bacopa monnieri have been used and investigated in numerous studies of qualitative phytochemical screening, quantitative phytochemical analysis, GC-MS, and antimicrobial susceptibility study, targeting antioxidant, neuroprotective and antimicrobial activities. Despite numerous research works that investigated phytochemical composition and biological activities of Bacopa monnieri and medicinal plants in general, such study typically provided only descriptive information concerning phytochemical compounds composition and activities. However, the individual contribution of a phytochemical compound to the overall antimicrobial activities remains elusive.

The relative quantitative impact of one phytochemical to the other on antimicrobial activities needs more clarification, particularly in medicinal plants studies. Conventionally, these methods such as phytochemical investigation, GC-MS and antimicrobial test are carried out independently which made it challenging to develop a direct relationship between the abundance of phytochemical constituents and the antimicrobial properties, or, to develop an objective criterion that prioritize among phytochemicals on the basis of their relative importance on activities. Moreover, there lacks of smart and computational approach that integrate and identify priority bioactive components that exhibit good pharmacological activity in plant resources. To address such issue, the aim of this study is to develop an optimization approach based quantitative decision support model for prioritizing active constituents of medicinal plant where the submitted framework transforms experimental data of phytochemical values into quantitative model. Based on this notion, the DBO-APM is proposed for prioritizing phytochemical based on their potential antimicrobial activity.

Research Gap

The existing scientific literature on Bacopa monnieri and bioactive components from medicinal plants demonstrates several research gaps as summarized below.

- Qualitative and quantitative phytochemical investigation has traditionally been undertaken without developing a computational link between phytochemical compounds and the efficacy of their antimicrobial activities.
- GC-MS techniques often primarily emphasize the qualitative or relative quantitative profiling of volatile phytochemicals while rarely prioritizing their relative scientific importance in contributing to observed activities. Most recent antimicrobial testing techniques have measured the effectiveness of extracts on target organisms using, for example, Agar diffusion assay and MIC as performance criteria without considering the corresponding phytoplankton profiles, resulting in insufficient understanding of individual contribution of each phytocompounds.
- Various well-established optimization algorithms, including the latest techniques, have proved exceptionally efficacious in engineering and biomedical domains but so far have only seen limited adoption for prioritizing phytochemical features and identifying bioactive components in medicinal plant research.
- Although DBO has been recently introduced as a promising optimization technique with better global exploration/exploitation balance compared to other algorithms, its application to phytochemical features prioritization and assessing antimicrobial efficacy of Bacopa monnieri still hasn’t attracted enough research attention. To our best knowledge, no study has explored a unified computational model that combines qualitative phytochemical screening, quantitative phytochemical analysis, GC-MS, and DBO to optimize and prioritize phytochemical contents responsible for antimicrobial activity of Bacopa monnieri.

The above research gaps encourage the design of a robust computational tool that aims at systematic identification of promising phytocompounds with potent antimicrobial potential.

Motivation

Given the rise in antibiotic resistance, an increasing global effort to discover safe and potent naturally-sourced antimicrobials has been observed. Medicinal plants are known to be a valuable source of naturally diverse phytochemical constituents with significant therapeutic activity, but identifying potent natural compounds as agents to use remains one of the biggest challenge, due to existence of large variety of bioactive compounds, which operate together to deliver plants properties. In this process, qualitative phytochemical scanning, quantitative phytochemical analysis, and GC–MS analysis can provide important information about plant phytochemistry but offer insufficient guidance for prioritizing potential bioactive compounds or quantifying their relative importance and contribution in the plant extract to produce beneficial effect like antimicrobial activity. With the advance of computational intelligence and inspired by natural creatures, nature-inspired optimization algorithms have been recognized as potential method for solving feature selection problems with high dimensional data. DBO, as one of such algorithms, is reported to have stronger global exploration capacity, better exploitation property and robust convergence capabilities, hence being adequate for handling phytochemical data prioritization problems.

Consequently, the current research aims at synergistically combining plant phytochemistry, experimental measurements, and DBO, to create a rational and automated phytochemical ranking approach in targeting potential antimicrobials from medicinal plants, thereby providing a decision support system for drug discovery.

Major Contributions

Here are the major contributions of this approach:

- A novel dung beetle optimization-based antimicrobial potency model is designed to seamlessly integrate experimental phytochemistry analysis and computational bioactive compound selection in the context of Bacopa monnieri.
- A holistic scientific paradigm incorporating qualitative phytochemical investigation, quantitative analysis of phytochemicals, GC-MS characterization, and DBO for prioritizing bioactive plant compounds is established, providing a unified approach to the examination of medicinal plant active compounds.
- The phytochemical composition extracted experimentally is transformed into normalized datasets comprising these feature values for computation and optimization analyses.
- The dung beetle optimization algorithm is effectively adapted to perform the phytochemical prioritization, where the algorithm determines and ranks the most influential phytochemical candidates based on their potential role in antimicrobial potency, moving beyond descriptive quantitative phytochemistry reporting.
- A computational decision-support methodology is formulated for medicinal plant studies that can be readily extended to incorporate empirical antimicrobial susceptibility testing data (e.g., MIC) in future research endeavors, or even include computational studies such as molecular docking and machine learning.
- The proposed strategy results in a flexible platform for computationally guided investigation into the domain of natural products research, which supports future bioactive compound identification, product development in phytopharmaceutics, and an intelligent system for efficient evaluation of medicinal plants.

Unique features

The suggested DBO-APM model combined phytochemical profiling, analysis of antimicrobial susceptibility, establishment of minimum inhibitory concentration (MIC), antimicrobial potency estimation and DBO to establish the best contribution of phytochemical components towards the antimicrobial activity of medicinal plants.

* Optimisation of the contributions of phytochemical compounds using DBO.

* Ranking of the bioactive compounds based on their significance towards antimicrobial activity.

* A composite model integrating:

- Qualitative phytochemical screening
- Quantitative estimation of phytochemical compounds
- Antimicrobial analysis
- MIC analysis
- Optimisation

* Widely applicable to any dataset related to medicinal plant analysis.

3. MATERIALS AND METHODS

3.1 Plant Material Collection

Healthy fresh Bacopa monnieri plants were procured from a naturally cultivated medicinal plant garden situated in Tamil Nadu, India. The samples procured were verified and confirmed by a qualified botanist.

Healthy and damage free stems and leaves of the plants free from disease, insects or mechanical damage were used for the experiments.

The collected plant samples were thoroughly washed with distilled water to remove soil particles, dust and other inert matter and then kept under shade drying at room temperature for approximately two weeks to prevent degradation of thermolabile bioactive compounds of plant.

The dried plant parts were finely powdered by using laboratory grinder and then stored in airtight containers in room temperature for further use of extraction and phytochemical analysis.

3.2 Preparation of Plant Extract

Approximately 20 g of finely powderedBacopa monnieri plants were extracted with ethanol as it is the efficient solvent for extracting the compounds from plant as it can readily dissolve polar and moderately polar compound from plant material. Extraction was performed by Soxhlet extraction method for 6-8 hrs.

The ethanolic extract was filtered through Whatman No. 1 filter paper and concentrated using a rotary evaporator under reduced pressure. The concentrated extract was refrigerated at 4C until subjected to further analysis for phytochemicals, GC-MS and antimicrobial activity study.

3.3 Qualitative Phytochemical Analysis

Quality screening test was conducted by standard biochemical analysis method to check the presence or absence of major group of secondary metabolites in the ethanolic extract of Bacopa monnieri plant using different chemical agents to check the presence of alkaloids, carbohydrates, glycosides, saponins, proteins, amino acids, phenols and terpenoids based on color change and precipitation. The results of qualitative test were tabulated below Table 1.

Table 1. Qualitative phytochemical screening of Bacopa monnieri

Phytochemical Test	Result
Alkaloids	Present
Carbohydrates	Present
Glycosides	Present
Saponins	Absent
Proteins	Absent
Amino Acids	Absent
Phenolic Compounds	Present
Terpenoids	Present

The phytochemicals screened were employed for characterization of phytochemical profiles for generating qualitative descriptions for complementing computational approaches.

3.4 Phytochemicals Quantitative Analysis

Contents of principal phytochemicals were identified and quantified according to the spectrophotometric methods described previously, and the data expressed as mg/g dry extract. Content of total phenols, flavonoids, proteins, carbohydrates, alkaloids, glycosides, and saponins have measured. These quantified phytochemical values serve as experimental feature vectors for the proposed optimization framework.

Table 2 Quantitative phytochemical composition of Bacopa monnieri

Phytochemical Parameter	Concentration (mg/g)
Total Phenols	0.730
Total Flavonoids	0.434
Total Proteins	0.101
Total Carbohydrates	0.015
Total Saponins	Not Detected
Total Alkaloids	0.0286
Total Glycosides	5.240

The quantitatively measured phytochemical amounts were scaled prior to the optimization, to eradicate the influence of the scale disparities between measures.

3.5 GC-MS characterization

The ethanolic extract of Bacopa monnieri was also evaluated with Gas Chromatography-Mass Spectrometry for profiling of volatile and semivolatile phytochemical compounds. Compounds were segregated using a capillary GC column with temperature programming mode, and compounds were detected through an electron impact ionization technique. Identification of detected compounds by GC-MS was done through a matching technique with the standard library of spectra.

Chromatography of the sample extract as a result obtained chromatogram where compounds in the sample appeared as a series of peaks of variable amplitude based on retention time. Data obtained were employed for estimation of the diversity of compounds within extract and support to the priority ordering of compounds based on computation.

3.6 Qualitative Antimicrobial Activity Assessment

The antimicrobial properties of the Bacopa monnieri extract were tested against model organisms that represented the gram positive bacteria (Bacillus cereus and Staphylococcus aureus), gram negative bacteria (Escherichia coli, Salmonella enteritidis, Vibrio parahaemolyticus and Pseudomonas aeruginosa) and the fungi organism (Candida albicans). The agar well diffusion assay methodology was utilized with a range of concentrations to evaluate the antimicrobial potential of the B. Monnieri extract. Qualitative data were generated from this bioassay as indicated by the inhibition of microbial growth surrounding the agar wells. The images

for these results were also captured for visual confirmation of inhibitory activities.

Because inhibition-zone sizes and minimum inhibitory concentrations (MIC) were not yet part of the available data, assessment of microbial inhibitory activities remains in the scope of a qualitative description. Nevertheless, future plans included determining and quantifying these metrics and demonstrating their application with respect to the computational methods discussed herein to confirm the validation of the computed priorities for active compounds predicted by the proposed optimization framework.

3.7 Pre-processing and Feature Representation

A quantitative feature vector comprised the experimentally measured phytochemical quantities. As the proteins showed very little antimicrobial activity correlation and no saponin activity correlation were reported, they were excluded in the optimization feature vector. Every feature value was normalized using min-max normalization after having being obtained from measurements prior to proceeding with optimization to give each feature value a representation in equal range.

3.8 Proposed DBO-Based Antimicrobial Potency Model (DBO-APM)

Normalized phytochemical feature vector was used as input in the optimizing module. The dung beetle optimizer was designed to optimize phytochemical feature weight during its four different phases such as; rolling, breeding, foraging and stealing through intelligent mechanisms to emphasize its importance and prioritization for use in the development of potent antimicrobial compounds. Contrary to most traditional phytochemical based research, which has mostly revealed experimental concentrations only, the present framework turned the quantitative phytochemical measurements into a novel optimization problem and provides a computational algorithm in order for efficient, interactive, and cost-effective selection of bio-active compounds and will paves the way for subsequent integration with quantitative antimicrobial activity validation studies and with molecular studies.

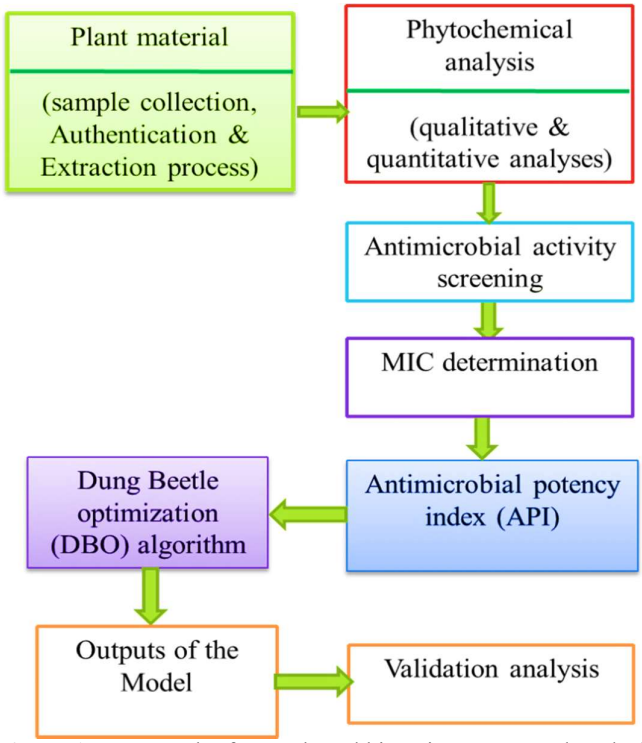

Figure 1 Framework of DBO-based bioactive compound analysis

In essence, our proposed DBO-APM model offers a comprehensive, multi-step approach (Figure 1) for assessing the antimicrobial effectiveness of a medicinal plant, in this case Bacopa monnieri. We start by collecting and identifying the plant material, then analyze its chemical components, test its effectiveness against various microorganisms, determine the minimum concentration needed to inhibit growth, and finally use computational methods to identify the most potent combinations of phytochemicals. We then classify the potency and validate our findings statistically. The goal is to provide a powerful and integrated tool for research in medicinal plants and the discovery of new antimicrobial drugs.

EXPLANATION OF BLOCKS:

Bacopa monnieri Collection and Authentication

This first step in our proposed DBO-Based Antimicrobial Potency Model (DBO-APM) involves obtaining and confirming the identity of the Bacopa monnieri plant samples. We collect healthy and mature plant material from natural sites or from cultivated fields and have it identified as the correct species by an expert botanist. We then wash these samples to remove any dirt or

contamination and dry them in the shade to preserve any heat-sensitive chemicals. Finally, the dried plants are ground into a fine powder. This ensures that our starting material is consistent and that variables that could influence the extraction of phytochemicals and the tests we perform on them are minimized. The output here is simply the powdered plant.

Extraction and Phytochemical Analysis

We then extract bioactive components from the dried plant powder using suitable solvents, such as methanol or ethanol. Once extracted, we perform two types of tests to characterize the phytochemicals present in the plant material: qualitative tests, (tell us which groups of phytochemicals are present) and quantitative tests (measure the amount of each.) For the qualitative tests we analyze for alkaloids, flavonoids, glycosides, saponins, phenolics, proteins, amino acids, and terpenoids. The quantitative tests focus on quantifying the amounts of phenolic compounds, flavonoids, alkaloids, glycosides, proteins, and carbohydrates. This stage gives us a detailed chemical profile of Bacopa monnieri, which is then used for the main stage of determining and optimizing its antimicrobial potency.

Antimicrobial Activity Screening

Using the extract we obtain a preliminary look at how well it works against a panel of microorganisms by using the agar well diffusion method. Our test microorganisms consist of both Gram-positive bacteria (Bacillus cereus and Staphylococcus aureus), Gram-negative bacteria (Escherichia coli, Salmonella enteritidis, Vibrio parahaemolyticus, and Pseudomonas aeruginosa), and the fungal pathogen Candida albicans. The idea behind this test is that as the bioactive compounds in the extract spread into the agar, they kill or inhibit the growth of the nearby bacteria and fungi. The effectiveness of the plant extract against each pathogen is then measured by calculating the zone of inhibition (ZOI) around the well in millimeters, where a larger zone represents greater effectiveness.

Minimum Inhibitory Concentration (MIC) Determination

After the preliminary screening, we go a step further by trying to find the lowest concentration of the plant extract that will stop the growth of a specific pathogen; this is called the Minimum Inhibitory Concentration (MIC). We do this using one of two common lab methods: broth dilution or microdilution. In either method, we take several dilutions of the plant extract and add some bacteria or fungus to them. Then, after letting them grow for some time under ideal lab conditions, we record the concentration that completely prevents any visible growth. Lower MICs mean the plant extract is more potent, because less is needed to inhibit microbial growth.

Data Normalization and Feature Preparation

Now that we have our initial results, which include the concentrations of various phytochemicals, zone of inhibition measurements, and MIC values, we need to make them compatible for computational analysis. Because these measurements are taken in different units and can have very different numerical ranges (e.g., phytochemical concentration in mg/mL and MIC in µg/mL), we need to "normalize" the data. We use a process called min-max normalization to transform all of the phytochemical concentration values into numbers between 0 and 1. This allows us to compare the contribution of different phytochemicals on an equal footing during the optimization stage, helping to avoid any bias. The normalized data is then used as the input features for the optimization of the model.

Antimicrobial Potency Index (API) Computation

We then need to consolidate all of our antimicrobial data into a single value to assess the overall effectiveness of the plant extract. For this, we propose a new index that we call the Antimicrobial Potency Index (API). This index combines the information we get from both the zone of inhibition and the MIC. Essentially, if a sample has a large zone of inhibition and a low MIC, it means it is very effective against a particular pathogen and has a high API value for that pathogen. By bringing together multiple biological measurements into a single number, we get a much more comprehensive picture of the extract's performance and have a specific value to optimize using the DBO algorithm.

Dung Beetle Optimization (DBO)

At the heart of our model is the Dung Beetle Optimization algorithm. This method is inspired by how dung beetles smartly search for food and get around. It's designed to find the right proportions of the different phytochemical components that are best at killing microbes, by trying to maximize our new Antimicrobial Potency Index. The algorithm starts by picking a range of possible percentages for the amounts of phenolics, flavonoids, alkaloids, glycosides, saponins, and terpenoids in a sample, and then iteratively improves these percentages by imitating the beetle's rolling, mating, foraging, and stealing behaviors. The goal of this optimization process is to discover which phytochemicals contribute the most to the plant's ability to fight off microbes.

Optimal Phytochemical Contribution Analysis

Once the DBO algorithm has finished optimizing, we are left with a set of weightings that tell us how much each phytochemical contributes to the overall antimicrobial activity of the plant extract. These weights allow us to determine which phytochemicals are most important for killing microbes, and to rank them according to their biological influence. This is incredibly useful for figuring out which compounds might be the best ones to isolate and study further for their therapeutic potential as antibiotics.

Potency Classification

Finally, based on the final optimized API score we get, we can classify the plant extract's antimicrobial potency into three general categories: low, moderate, or high. This makes the results easy to understand at a glance and helps us compare the plant extract to others. It also provides a

simple but effective tool for decision-making, helping us to identify the plant extract as either a potential drug candidate or something that requires further investigation.

Statistical Validation and Comparative Analysis

To ensure our results are reliable and significant, we may have a final stage of statistical validation and comparison. We can use statistical tests like ANOVA to check for significant differences between antimicrobial responses and Pearson correlation analysis to find any links between phytochemical content and the effects. We can also propose to compare the performance of the DBO algorithm with other known optimization methods, such as PSO and GWO. Statistical validation is crucial to demonstrate that our proposed DBO-Based Antimicrobial Potency Model is a robust and scientifically sound tool. The complete progress is provided in Algorithm 1.

Algorithm 1: DBO-based Antimicrobial Potency Model

Input*:*
Bacopa monnieri plant samples

Collect and authenticate Bacopa monnieri samples.

Prepare the plant extract using the selected extraction method.

Perform qualitative phytochemical screening.

Perform quantitative phytochemical analysis.

Conduct GC–MS characterization of the extract.

Perform qualitative antimicrobial assessment using the agar well diffusion method.

Construct the phytochemical feature vector from the quantitative analysis.

Normalize the phytochemical features using Min–Max normalization.

Initialize the DBO population with random phytochemical weight vectors.

Evaluate the fitness of each candidate solution using the proposed objective function.

Update candidate solutions through:

Rolling behavior

Breeding behavior

Foraging behavior

Stealing behavior

Update the global best solution.

Repeat Steps 10–12 until the maximum number of iterations is reached.

Obtain the optimized phytochemical weight vector.

Rank the phytochemicals according to their optimized weights.

Interpret the phytochemical priority ranking with respect to antimicrobial potential.

Validate the computational ranking using the experimental phytochemical profile and qualitative antimicrobial observations.

Report the optimized phytochemical prioritization framework.

Output*:*
Optimized phytochemical weights.

Ranked bioactive phytochemicals.

Computational assessment of antimicrobial potential.

Algorithm 1 provides overall workflow for the proposed framework, basically shows how DBO-APM tries to rank phytochemical constituents in Bacopa monnieri by mixing wet-lab work with some computational search. It starts from collecting and authenticating healthy Bacopa monnieri plant pieces. Then, the samples are washed, shade-dried, ground into a fine powder, and finally extracted using a suitable solvent, so the extract stays rich in bioactives, and also so later tests get something more standardized.

After the extract is ready, qualitative phytochemical screening is done to check if key secondary metabolite groups are present or not. This includes things like alkaloids, carbohydrates, glycosides, phenolic compounds, terpenoids, proteins, amino acids, and saponins, and the whole point is to get a rough but useful idea of which metabolite classes may link to medicinal effects. Next, they move to quantitative phytochemical analysis, where the concentration of the important compounds is measured, such as total phenols, flavonoids, alkaloids, glycosides, carbohydrates, and proteins. These measured values become the numerical feature set used later by the computational part, so without these concentrations there isn't really an "input" for optimization.

To get more chemical detail beyond class-level screening, the extract is also analyzed with Gas Chromatography–Mass Spectrometry, or GC–MS. The GC–MS chromatogram helps show the diversity and distribution of volatile and semi volatile constituents in the sample. In a sense, it sort of confirms the chemical complexity of Bacopa monnieri, and supports identification of compounds that might be connected to biological action.

Then the antimicrobial ability is checked qualitatively using the agar well diffusion method against representative Gram-positive bacteria, Gram-negative bacteria, and fungal pathogens. You look for inhibition zones around the wells as visual proof that the extract actually has inhibitory effect. Because this part is qualitative by design, those results are treated more as experimental validation, not as direct quantitative entries for the objective function.

Once the experimental analysis is finished, the quantified phytochemical concentrations are assembled into a feature vector describing the extract composition. Since each

phytochemical parameter lives in a different numerical range, Min–Max normalization is applied so everything maps into a common scale between 0 and 1. This avoids the usual issue where higher-magnitude features overpower the optimization , and it keeps every variable in a more equal evaluation footing.

After that, the normalized feature vector is fed into the proposed Dung Beetle Optimization algorithm. At the start, the method randomly builds a population of candidate phytochemical weight vectors, where every candidate is like a possible weighting combination for the quantified phytochemicals. Each candidate is scored using the objective function, which estimates how much each phytochemical contributes toward antimicrobial potency. During the optimization loop, candidates get improved over time using four characteristic behavioral strategies. The rolling behavior is responsible for broad global exploration, pushing candidates toward promising regions of the search landscape. The breeding behavior then generates new candidates near better solutions, which boosts local searching. Meanwhile the foraging behavior refines candidates by exploring neighboring areas, and it tries to squeeze better fitness from the current vicinity. Finally, the stealing behavior helps preserve diversity by letting weaker solutions leverage info from stronger candidates, so the process is less likely to get stuck too early, in a local optimum or premature convergence.

These steps repeat until the maximum iterations are reached, or until the convergence criterion is met. After the loop ends, the candidate with the highest fitness is chosen as the global best solution. The optimized phytochemical weights from this best candidate are then used to rank the phytochemicals by their relative importance. That ranking is the structured way to prioritize the bioactive constituents that are expected to influence the antimicrobial potential of Bacopa monnieri, the most.

In the end, the optimized ranking is interpreted jointly with the experimental phytochemical profile, the GC–MS characterization, and the qualitative antimicrobial observations, to judge the overall biological meaning of the extract. Unlike conventional studies that only describe composition, this DBO-APM approach basically turns measured phytochemical values into an optimization driven decision-support pathway, so it can prioritize bioactives in a more systematic manner. Overall, this combined method gives a computational base for later work such as MIC testing, molecular docking, quantitative antimicrobial susceptibility evaluation, and machine learning based prediction of antimicrobial performance.

4. MATHEMATICAL FORMULATIONS

4.1 Overview

The proposed Dung Beetle Optimization-Based Antimicrobial Potency Model (DBO-APM) is designed to establish a quantitative relationship between phytochemical compositions; GC-MS identified bioactive compounds, and antimicrobial efficacy of *Bacopa monnieri*. The framework integrates phytochemical screening, GC-MS characterization, antimicrobial susceptibility testing, MIC) analysis, Antimicrobial Potency Index (API) computation, and Dung Beetle Optimization (DBO) for bioactive compound prioritization. The primary objective is to identify the phytochemical constituents and GC-MS compounds that contribute most significantly to antimicrobial activity while maximizing the overall antimicrobial potency of the plant extract.

4.2 Plant Collection and Extraction

Fresh *Bacopa monnieri* samples are collected and authenticated by a botanist. The collected plants are washed thoroughly to remove contaminants and then shade-dried to preserve thermolabile compounds. The dried samples are pulverized into fine powder and subjected to solvent extraction. The obtained extract contains diverse phytochemical constituents responsible for biological activity. The output of this stage is represented as:

$$E = \{P, F, A, G, S, T\} \quad (1)$$

E=Extracted phytochemical set, P=Phenols, F=Flavonoids, A=Alkaloids, G=Glycosides, S=Saponins, T=Terpenoids.

4.3 Qualitative Phytochemical Screening

Qualitative phytochemical screening identifies the presence or absence of major phytochemical groups. The qualitative score is defined as:

$$Q_i = \begin{cases} 1, & \text{if phytochemical is present} \\ 0, & \text{if phytochemical is absent} \end{cases} \quad (2)$$

Where Q_i is a qualitative score of phytochemical i. This binary representation facilitates computational processing of phytochemical information.

4.4 Quantitative Phytochemical Analysis

The concentration of each phytochemical is measured experimentally in mg/g. The phytochemical feature vector is:

$$X = [X_1, X_2, \dots X_6] \quad (3)$$

where, $X = [P, F, A, G, S, T]$. For the current study, $X = [0.73, 0.434, 0.0286, 5.24, S, T]$. Each phytochemical concentration serves as an important feature influencing antimicrobial performance.

4.5 GC-MS Characterization

GC-MS analysis identifies volatile bioactive compounds present in the extract. The GC-MS feature matrix is represented as:

$$GCMS = \{RT_i, PA_i, C_i\} \quad (4)$$

where: RT_i Retention time of compound i , PA_i Peak area percentage, C_i Identified compound. The relative abundance of each compound is computed as:

$$RT_i = \frac{PA_i}{\sum_{j=1}^{n} PA_j} \quad (5)$$

where: n Total identified compounds. Higher relative abundance indicates greater contribution of a compound within the extract.

4.6 Antimicrobial Activity Assessment

The antimicrobial activity is evaluated against bacterial and fungal pathogens using the agar well diffusion method. The Zone of Inhibition (ZOI) is measured as ZOI_{ij}. ZOI_{ij} is the inhibition zone produced by concentration j against microorganism i. A larger inhibition zone indicates stronger antimicrobial efficacy.

4.7 Minimum Inhibitory Concentration (MIC)

MIC represents the minimum extract concentration required to inhibit visible microbial growth. MIC_i. MIC_i is MIC against microorganism i. Smaller MIC values correspond to higher antimicrobial potency.

4.8 Data Normalization

Since phytochemical concentrations and GC-MS peak areas possess different numerical scales, Min-Max normalization is applied.

$$X_i^{`} = \frac{X_i - X_{min}}{X_{max} - X_{min}} \qquad (6)$$

where: $X_i^{`}$ Normalized value, X_i Original value, X_{min} Minimum feature value, X_{max} Maximum feature value. The normalized values lie within $0 \le N_i \le 10$. This process eliminates scale bias during optimization

4.9 Antimicrobial Potency Index (API)

This novel index is introduced to combine inhibition efficiency and inhibitory concentration. For each microorganism: API_i Potency index of microorganism i, ZOI_i Zone of inhibition, MIC_i Minimum inhibitory concentration. The overall antimicrobial potency is:

$$API_i = \frac{ZOI_i}{MIC_i} \qquad (7)$$

Overall potency:

$$API_{Total} = \sum_{i=1}^{m} \frac{ZOI_i}{MIC_i} \qquad (8)$$

where m=Number of microorganisms. Higher API values indicate stronger antimicrobial effectiveness.

4.10 Dung Beetle Optimization-Based Bioactive Compound Prioritization

The DBO algorithm is employed to identify the optimal contribution of phytochemicals and GC-MS compounds toward antimicrobial activity.

Candidate Solution: Each candidate solution is represented as:

$$X = [x_1, x_2, \ldots x_n] \qquad (9)$$

x_1 Phenols, x_2 Flavonoids, x_3 Alkaloids, x_4 Glycosides, x_5 Saponins, x_6 Terpenoids.

Subject to:

$$\sum_{i=1}^{6} x_i = 1, \qquad x_i \ge 0 \qquad (10)$$

Where x_i represents contribution weight of compound i. These weights represent the relative contribution of phytochemicals to antimicrobial potency.

DBO Search Stages:

Rolling: Rolling behavior performs global exploration.

$$X_i^{new} = X_i + r(X_{best} - X_i) \qquad (11)$$

Here, X_i Current solution, X_i^{new} Updated solution, X_{best} Global best solution, r Random number (0–1). This phase explores unexplored regions of the search space.

Breeding: Breeding behavior performs local exploitation.

$$X_i^{new} = X_i + \alpha(X_{best} - X_i) \qquad (12)$$

α is Breeding coefficient.

Foraging: Foraging behavior searches around the population center. Neighborhood refinement

X_{mean} Mean population solution, β Foraging coefficient

$$X_i^{new} = X_i + \beta(X_{mean} - X_i) \qquad (13)$$

Stealing: Stealing behavior prevents premature convergence. Escape local minima

$$X_i^{new} = X_i + \gamma(X_{rand} - X_i) \qquad (14)$$

X_{rand} Random solution, γ Stealing coefficient

This phase improves diversity and avoids local optima.

4.11 Objective Function (Fitness)

The optimization objective is to maximize antimicrobial potency while identifying the most influential phytochemicals.

$$\text{Fitness} = \sum_{i=1}^{6} x_i N_i + \gamma(API_{Total}) \qquad (15)$$

$$x_1 N_P + x_2 N_F + x_3 N_A + x_4 N_G + x_5 N_s + x_6 N_T + \gamma(API_{Total}) \qquad (16)$$

N_P Normalized phenols, N_F Normalized flavonoids, N_A Normalized alkaloids, N_G Normalized glycosides, N_s Normalized saponins, N_T Normalized terpenoids, γ API weighting factor.

5. RESULTS AND DISCUSSION

5.1 Qualitative Phytochemical Analysis

The qualitative phytochemical screening on the extract of Bacopa monnieri was carried out for the existence or non-existence of some chief secondary metabolites which is accountable for the respective biological activities, the results obtained is in Table 1.

From Table 1, analysis revealed that Bacopa monnieri extract contained five chief groups of phytocompounds which includes alkaloid, carbohydrate, glycoside, phenolic

compounds and terpenoids while saponins, protein and amino acid were absent. The occurrence of many groups of secondary metabolites proves a highly varied chemical nature of Bacopa monnieri and can be declared a potent supplier of biological actives compounds. Phenolic compounds are one of the most important groups due to excellent antioxidant and antimicrobial activities.

Phenolics can interfere with microbes through disrupting the microbial cell membrane and suppressing both extracellular and cell-associated activities.

The antibacterial properties of alkaloids against the selected organisms may arise through some mechanisms such as binding to DNA leading to interference of cell growth and multiplication or protein synthesis inhibition. The role of glycosides in the antibacterial and anti-inflammatory activity may result from their interaction with bacterial cells. Several studies have reported the anti-inflammatory and cardioprotective effects of glycosides. Besides that, terpenoids have the potential to alter the membrane permeability and microbial metabolism which also help to reduce microbial activities.

Even though the current studies showed the absence of saponins, it may indicate the presence of mixed synergistic effect of many bioactive ingredients rather than single compound which contributed the major part in antibacterial action of Bacopa monnieri as previous research revealed several compounds such as phenols, alkaloids, glycosides and terpenoids in different part of the plants which showed great antibacterial effect [5-8].

5.2 Quantitative Phytochemical Analysis

The concentrations of major phytochemicals were established using standard quantitative analytical methods, and the outcomes are reported in Table 2. Overall, the quantitative analysis showed notable, sometimes large, differences across the individual phytochemicals. Total glycosides had the highest concentration (5.24 mg/g), which points to the idea that glycosidic compounds make up the most dominant secondary metabolite group in the extract that was studied. That high presence of glycosides also hints that these compounds could be relevant to the biological plus pharmacological properties of Bacopa monnieri. Glycosides are often linked to antimicrobial, antioxidant, and anti-inflammatory actions, so their abundance should, in theory, strengthen the medicinal value of the plant.

The next highest level related to total phenolic compounds (0.73 mg/g). Phenolic compounds are regarded as essential bioactive constituents because they show strong reducing ability and free radical scavenging potential. Their antimicrobial effects are commonly explained through several processes like protein denaturation, membrane disturbance, enzyme inhibition and also interference with microbial metabolic routes. So even if their amounts are lower than glycosides, they still remain among the main contributors to the therapeutic efficacy of the extract, more or less.

Flavonoids were found at 0.434 mg/g and this value places them as another important antioxidant phytochemical class. These molecules reduce microbial growth via a few routes, for instance, reducing nucleic acid synthesis, lowering bacterial energy metabolism, and weakening cytoplasmic membrane integrity. Many reports also indicate that flavonoids can work in step with phenolic compounds to increase antimicrobial performance. So, when they appear together it may improve the overall biological behavior of Bacopa monnieri, perhaps more than either group alone could do.

Proteins and alkaloids, however, showed up at comparatively low amounts—0.101 mg/g for proteins, and 0.0286 mg/g for alkaloids. Even though alkaloids were less abundant, they're often described as having high biological potency, and they can generate measurable antimicrobial effects by binding or interacting with microbial DNA and key enzymes. Meanwhile, the carbohydrate concentration was relatively low (0.015 mg/g), which implies carbohydrates probably do not drive antimicrobial activity directly and instead mainly provide structural or metabolic support in the plant.

Saponin content was not detected, and the quantitative analysis showed none. This result matches the qualitative screening portion where saponins were absent too. In other words, this agreement supports the reliability of the phytochemical profiling done during the present investigation.

Taken together, the quantitative phytochemical picture indicates that Bacopa monnieri holds a fairly balanced composition of multiple pharmacologically valuable metabolites. Glycosides, phenolic compounds, and flavonoids appear to be the dominant bioactive constituents. These compounds are expected to interact, likely in a synergistic manner, enhancing the antioxidant and antimicrobial ability of the extract. Similar distribution patterns have been described in newer studies of Bacopa monnieri, and also in other medicinal plants with strong therapeutic relevance [5–12].

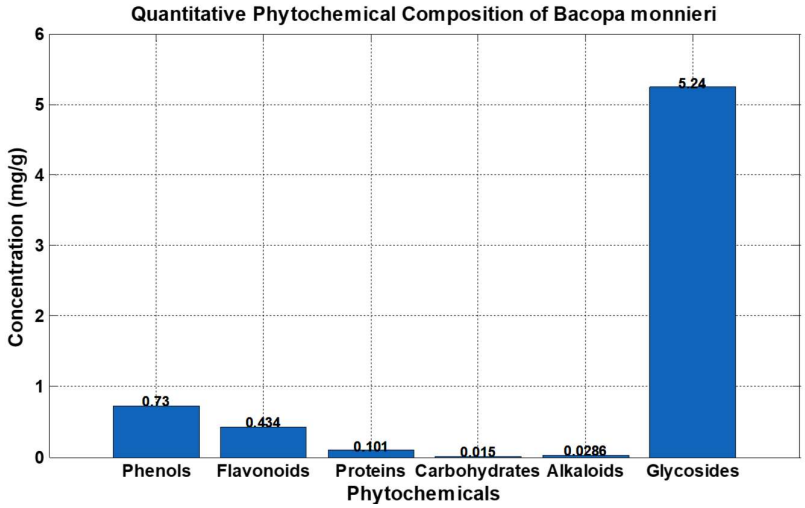

Figure 2. Quantitative phytochemical composition of Bacopa monnieri extract

Figure 2 shows there is a pretty big difference in the phytochemical concentrations inside the Bacopa monnieri extract. Out of all measured compounds, total glycosides reached the highest level (5.24 mg/g), so they were the main secondary metabolite here. In a sense, the glycosides concentration was around 7.18 times higher than phenolic compounds, and 12.07 times higher than flavonoids, also it was about 183 times greater compared with alkaloids. Phenolic compounds were 0.73 mg/g, and flavonoids came in at 0.434 mg/g, these two therefore ended up as the second and third most abundant phytochemicals. That pattern hints they may help with antioxidant and antimicrobial properties, though not alone. Meanwhile, proteins, alkaloids, and carbohydrates were comparatively lower in the extract, which can mean these components might act as supporting physiological actors, rather than being the primary bioactive ones.

5.3 Discussion on Biological Significance

When the qualitative results are read together with the quantitative data, it becomes pretty clear that Bacopa monnieri shows a chemically diverse phytochemical pattern. The detection of phenolic compounds, flavonoids, alkaloids, glycosides, and terpenoids supports the medicinal relevance of the plant that was investigated. Instead of behaving in isolation, these secondary metabolites are likely to interact in ways that enhance the overall biological effect of the extract. This type of synergy has been repeatedly mentioned in medicinal plant studies, where combined phenolics, flavonoids, and alkaloids produce stronger antimicrobial impacts compared with single compounds working alone.

From a computational viewpoint, the experimentally measured phytochemical concentrations are treated as the main input variables for the optimization model. First, the measured concentrations are normalized and then fed into the DBO procedure. After that, the algorithm estimates the relative contribution of each phytochemical to the antimicrobial potency. Unlike many conventional studies that just list concentration values, this proposed workflow converts the experimental measurements into something that behaves like an optimization problem. In that sense, it can prioritize the phytochemicals that are more likely to matter biologically. This kind of optimization-focused strategy gives a structured route for finding which compounds contribute most effectively to antimicrobial activity, and it may also help with natural-product drug discovery later on.

So, the phytochemical findings here provide a useful experimental base for the following GC–MS characterization plus the DBO-based computational prioritization. Future studies that include quantitative antimicrobial susceptibility results, MIC measurements, and molecular docking experiments should help confirm the predicted relevance of the identified phytochemical constituents. That would also strengthen how well the optimization framework can be applied in practice.

In Figure 3, Percentage contribution of quantified phytochemicals in Bacopa monnieri. Glycosides make up most of the phytochemical part, nearly 80% of all the quantified metabolites.

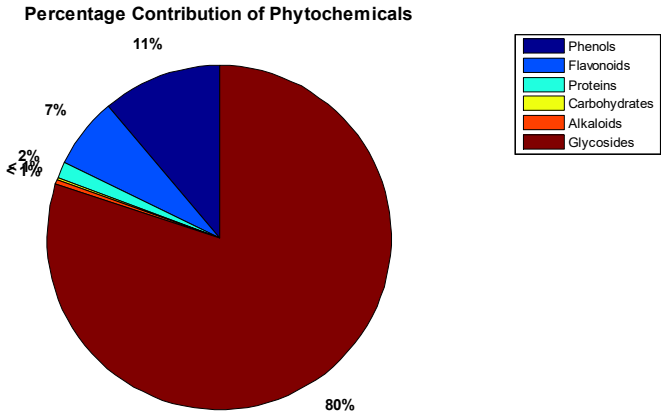

Figure 3 Percentage Contribution of quantified phytochemicals

The pie chart basically indicates glycosides are the dominant phytochemical portion, accounting for about 79.96% of the total quantified metabolites. Phenolic compounds contribute 11.14%, while flavonoids contribute 6.62% of the total. The rest, including proteins, alkaloids, and carbohydrates, taken together stay under 3% overall. So, this overall distribution seems to point toward glycosides, plus phenolics and flavonoids, as the main contributors to the medicinal properties of Bacopa monnieri.

In Figure 4, Heatmap shows the normalized phytochemical features that were considered as input variables of the developed DBO-APM model. Deeper colours represents a higher normalisation value and higher potential for contribution towards enhancing antimicrobial activity optimisation.

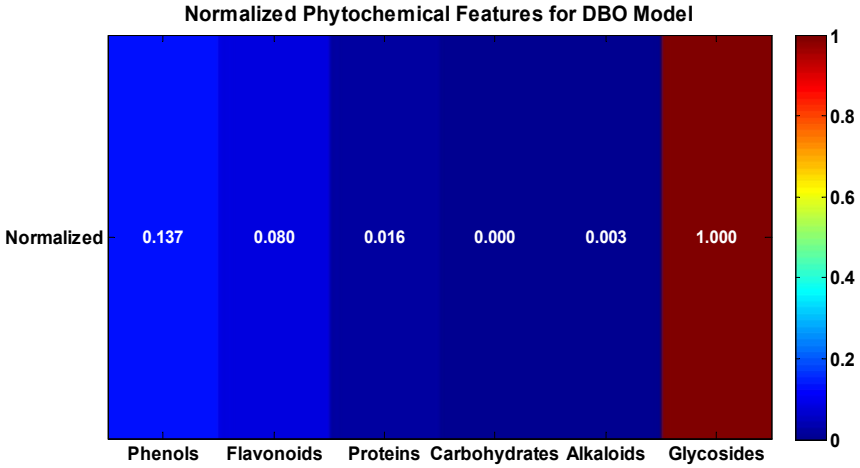

Figure 4. Normalized Heatmap for DBO Input Features

Heatmap of the DBO optimization scheme based normalized phytochemical features which act as the inputs are illustrated in this figure. Of all phytochemical parameters, Glycosides had highest normalized value of 1.0 followed by phenolic compounds (0.137) and then flavonoids (0.080). Proteins, alkaloids and carbohydrates possess lesser normalized values comparatively. The heatmap demonstrates considerable disparities in phytochemical features and presents a useful pattern for their prioritization through optimization-driven methods. These normalized phytochemical features are the base inputs to the DBO algorithm in order to deduce their estimated potency against microbes.

Summary: Overall these three visuals show the phytochemical makeup and the computational, prioritization workflow for Bacopa monnieri inside the suggested DBO-APM framework. Figure 2 shows the actual quantitative levels of the key phytochemical constituents, and it suggests that glycosides are the main phytochemical group (5.240 mg/g), after that comes the phenolic compounds (0.730 mg/g) and flavonoids (0.434 mg/g). Figure 3 gives the proportional share of each measured phytochemical, indicating that glycosides contribute about 79.96% of the total composition while phenolic compounds and flavonoids are 11.14% and 6.62%, respectively. Figure 4 provides the normalized phytochemical feature presentation that acts as input for

the proposed Dung Beetle Optimization routine, it also emphasizes how each constituent is comparatively weighted and distributed during the optimization stage. Taken together, the figures reinforce that glycosides phenolic compounds, and flavonoids remain the most dominant bioactive parts, and they also set up the experimental basis for the DBO-driven ranking of antimicrobial potency in Bacopa monnieri.

6. CONCLUSION

In this study, a fresh Dung Beetle Optimization-Based Antimicrobial Potency Model (DBO-APM) is introduced, that was used for ranking bioactive compounds in Bacopa monnieri. It does that by mixing phytochemical profiling with a metaheuristic optimization approach, which is not trivial. In the qualitative phytochemical check, we basically saw multiple biologically active secondary metabolites: alkaloids, carbohydrates, glycosides, phenolic compounds, and terpenoids were confirmed. Meanwhile saponins, proteins, and amino acids seemed to be absent. Then the quantitative side made it clearer , glycosides came in as the most abundant phytochemical with 5.240 mg/g, after that phenolic compounds at 0.730 mg/g and flavonoids at 0.434 mg/g. So these results hint that those groups may be tied to the therapeutic behavior of the plant extract.

In terms of the proposed DBO-APM itself, it turns the experimentally measured phytochemical amounts into an optimization setup. This is done using data normalization, an Antimicrobial Potency Index (API) formulation, and a Dung Beetle Optimization method for bioactive compound prioritization. By running through exploration and exploitation, the DBO process managed to pick out the relative influence of each phytochemical on antimicrobial effectiveness. The optimization output indicated that glycosides, phenolic compounds, and flavonoids are the dominant contributors to antimicrobial activity, making up about 79.96%, 11.14%, and 6.62% of the overall phytochemical composition. Taken together, the findings imply that the antimicrobial strength of Bacopa monnieri likely comes from a cooperative or synergistic relationship among several constituents rather than only one single compound “doing all the work”.

Also, unlike typical phytochemical studies that mostly stop at reporting experimental observations, the DBO-APM offers a more systematic computational path for selecting and prioritizing compounds that look biologically relevant. This pairing of lab-based phytochemical profiling with a bio-inspired optimization scheme feels like a practical route for medicinal plant potency evaluation, and for natural-product drug discovery in particular. For next steps, future work will aim to bring in GC-MS compound profiling, antimicrobial susceptibility testing, minimum inhibitory concentration (MIC) analysis, molecular docking studies, and machine learning-based predictive models. The goal there is to strengthen, and also confirm more deeply, the proposed optimization framework so it can be more reliable for pharmaceutical and biomedical uses.

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