

# Isolation And Characterization of Novel Bioactive Compounds from Endemic Medicinal Plants of The Western Ghats

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## ABSTRACT

Western Ghats in India which has been globally accepted as a World Heritage Site by UNESCO serves as a hotspot of biological diversity, having a rich diversity of endemic medicinal plants with pharmacological value. The purpose of the current literature was to isolate and characterize new bioactive agents in some of the endemic plants, *Coscinium fenestratum*, *Plectranthus vettiveroides* and *Garcinia indica*. The used plant materials were obtained with a professional and ethical standard of collection of both conservation and ethical standards, extracted via solvent partitioning and then chromatographically separated. The presence of that structure was elucidated by Fourier-transform infrared spectroscopy (FTIR), nuclear magnetic resonance (NMR) and high-resolution mass spectrometry (HR MS) analysis. Screening of the bioactivity was done on antimicrobial, anticancer and antioxidant activities. It was found that there were three new alkaloids plus two flavonoid glycosides and a hitherto unknown Xanthone derivative. Such substances demonstrated a strong in vitro antimicrobial activity against *Staphylococcus aureus* and *Escherichia coli*, high antioxidant properties in assays of DPPH and ABTS, and selective human breast cancer cell lines cytotoxicity (MCF-7). These results add to the fact that the flora of the Western Ghats has significant potential in novel drug discovery.

## Key Words:

Western Ghats, Endemic Plants, Bioactive Compounds, Phytochemistry, Drug Discovery

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## 1. INTRODUCTION

India Western Ghats is a 1,600-km long mountain range on the Western coast of the Indian subcontinent, which ranks as one of the eight hottest hotspots of biodiversity in the world<sup>1</sup>. A diversity of IUCN defined ecosystems, such as tropical rainforests, montane grasslands, and mangroves are found in this mountain range, which hosts an astonishing and representative fauna and flora<sup>2</sup>. It boasts vast botanical resources with many medicinal plants that have long been used by local populations in centuries-old traditions to treat a variety of disease and syndromes, including infectious diseases and metabolic disorders<sup>3</sup>. Their value in ethnobotany has been greatly recognized in traditional medicine of traditional knowledge systems including Ayurveda,

Siddha and folk medicine<sup>4</sup>. A major fraction of these plants especially those that are inhabited in a narrow ecological niche have however remain largely unexploited in terms of their phytochemical makeup as well as their pharmacological application<sup>5</sup>.

### 1.1 Background Information

The natural products research has seen a revival all over the globe in recent decades, with the rising necessity to find new forms of therapeutic agents in order to overcome antibiotic resistance, chronic diseases and newly emerging pathogens<sup>6</sup>. Medicinal plants are irreplaceable sources of structurally varied bioactive compounds such as alkaloids, flavonoids, terpenoids and phenolics and much of them have been used as templates to the development of modern pharmaceuticals<sup>7</sup>. Uniquely microclimatic variations and evolutionary pressures have shaped the endemic flora<sup>8</sup>. Western Ghats in such a way that these plants should have unique phytochemical profiles that are not typified by other plants elsewhere<sup>9</sup>. Such biochemical distinctiveness gives the Western Ghats an unexplored- but a highly promising- source of novel drug leads<sup>10</sup>.

### 1.2 Statement of the Problem

Although the ethnomedicinal value of flora growing in the Western Ghats is well documented, few endemic species have been rigorously chemically explored. There is a huge gap in the knowledge of the possible applications of these plants into drug discovery due to the lack of comprehensive phytochemical profiling and assessment of bioactivities of these plants. This underutilization is fueled by the risks of habitat destruction and overexploitation that potentially can cause the irreversible loss of a species before its medical values can be scientifically established. There exists, therefore, an emerging need to carry out specific research with a view towards isolating, characterizing, and determining the pharmacological effects of bioactive compounds in these plants.

### 1.3 Objectives of the Study

The present study was undertaken with the following specific objectives:

- To identify and purify new bioactive substances of medicinal useful plants of the Western Ghats.
- To define the chemical structure of the isolated compounds through the more advanced analytical methodologies like Fourier-transform infrared spectroscopy (FTIR), nuclear magnetic resonance (NMR) and high-resolution mass spectrometry (HR-MS).
- To determine the potential antimicrobial, anti-oxidant and anti-cancer potential of the extracted compounds using standardized in vitro bioassays.

### 1.4 Hypotheses

This study was guided by the following hypotheses:

- Western Ghats endemic medicinal plants have special bioactive compounds that have never been indicated in other species of plants.
- These substances have a great antimicrobial, antioxidant and anti-cancer properties, hence it has great potential of becoming therapeutics.

## 2. METHODOLOGY

The research design of the study was aimed at harmonizing a reproducible and methodical treatment of finding, purifying, and characterizing novel bioactive components of chosen endemic medicinal crops of the Western Ghats and subsequently testing their pharmacological value. It was carried out in fully equipped phytochemistry and microbiology laboratory conditions and in accordance with international research and ethical guidelines.

### 2.1 Research Design

The research was experimental and laboratory research design which is a combination of phytochemical study and in vitro biological activity testing. Such choice of design was made to allow drawing a straightforward correlation between the chemical nature of isolated compounds and the observed effects in a pharmacological test. This was a sequential process which involved plant collection and authentication, extraction and fractionation, purification, characterization of the structures and functional assay screenings.

### 2.2 Sample Details

Three endemic medicinal plant species were selected for this study based on ethnobotanical significance, rarity, and historical medicinal use. These were:

1. *Coscinium fenestratum* – Stem bark
2. *Plectranthus vettiveroides* – Roots
3. *Garcinia indica* – Fruit rind

Plant material was gathered in restricted forest ecosystems in Karnataka and Kerala, India, with permits of the statutory body, viz., collection permit given by the concerned State Forest Departments. A taxonomist of the Botanical Survey of India identified and authenticated the species. All plants had voucher specimens preserved in the institution herbarium as reference material.

### 2.3 Instruments

A series of analytical grade solvents, chromatographic systems, and spectroscopic apparatus were necessary during the conducting of the experiment. The primary materials and equipment used included:

- Solvents: Methanol, ethanol, ethyl acetate, chloroform, and n-hexane (Merck, India).
- Chromatography Systems: Silica gel column chromatography (60 120 mesh), thin-layer chromatographic (silica gel 60 F254 plates) and high-performance liquid chromatography (20AD, Shimadzu) reverse-phase C18 column chromatography were used.
- Spectroscopic Instruments: Fourier-transform infrared spectrometer (Shimadzu IRTracer - 100), nuclear magnetic resonance spectrometer (Bruker Avance III 500 MHz <sup>1</sup>H and <sup>13</sup>C NMR), and a high-resolution mass spectrometer (Thermo Scientific Q Exactive Orbitrap).

- Bioassay Facilities: Laminar hood cabinet, biological oxygen demand – biodom station, CO<sub>2</sub> incubator and microplate reader (Bio-Rad iMark).

## 2.4 Data Collection Methods

The steps involved in the experiment process were separated into different and interconnected processes to promote accuracy and consistency. Seed and plant materials were dried and powdered, and extracts of plant materials were obtained by sequential extractions with increasing (n-hexane, chloroform, ethyl acetate, and methanol) solvents in a Soxhlet apparatus (ca. 500 g/compound). The final extracts were concentrated to a lesser pressure with a rotary evaporator and kept refrigerated at 4 °C prior to further experiments.

Silica gel column chromatography was used to separate crude extract into fractions where gradient elution was carried out either with steps of hexane and ethyl acetate to subsequent steps of methanol in order to get several fractions. Replicate fractions that were consistent in their thin-layer chromatography (TLC) profiles, as visualized in ultraviolet light and upon spraying with anisaldehyde-sulfuric acid solution, were combined and carried forward in an effort to provide purified human erythropoietin.

The preparation of bioactive fractions was also done by preparative thin-layer chromatographic (TLC) and high-performance liquid chromatography (HPLC). Optimal solvent systems were developed to obtain a purity level of greater than 95% in every compound which was confirmed by analytical HPLC profiling. Mass spectrometry Fourier-transform infrared spectroscopy (FTIR) was used to characterize the purified compounds and identify functional groups, structural analysis was done by proton nuclear magnetic resonance (<sup>1</sup>H) and carbon-13 (<sup>13</sup>C) nuclear magnetic resonance (NMR) spectroscopy, and high-resolution mass spectrometry (HR-MS) was used to determine the molecular formula and a correct molecular weight.

Antimicrobial, antioxidant, and anticancer tests were used in determining the bioactivity of the purified compounds. The test against *Staphylococcus aureus* (Gram-positive) and *Escherichia coli* (Gram-negative) species were performed (disc diffusion) and an inhibition zone was measured after being incubated at 37 °C in 24 hours. The antioxidant capacity was conducted with the DPPH and the ABTS radical scavenging assays; the IC<sub>50</sub> was calculated on the respective compounds. The ACI was determined using MCF-7 (human breast cancer) and HeLa (human cervical cancer) cell lines with cells incubated in varying concentrations of compound, over 48 hours, and cell viability measured at 570 nm with a microplate reader using MTT cell proliferation assay.

## 2.5 Data Analysis Techniques

The bioactivity tests were carried out in triplicate, and data presented as mean standard deviation (SD). The non-linear regression analysis carried out in GraphPad Prism (version 9.0) was performed to calculate IC<sub>50</sub> values of antioxidant and anticancer assays. The one-way analysis of variance (ANOVA) was adopted in antimicrobial experiments to determine statistically significant differences among compounds coupled with Tukey post hoc test. p-value less than 0.05 was taken to be your significant point.

### 3. RESULTS

Comparative isolation, characterization, and biological studies of bioactive compounds of novel interest in *Coscinium fenestratum*, *Pogostemon vettiveroides*, and *Garcinia indica* were obtained as discussed below. Results fall into categories chemical characterization, assessment of yield and purity, antimicrobial activity, antioxidant potential and cytotoxicity assessment. One-way ANOVA and post hoc test by Tukey were used as statistical tools with the level of significance at  $p < 0.05$ .

#### 3.1 Extraction Yield and Purity Assessment

The purity and yield of extracted bioactive compounds firstly of the selected endemic medicinal plants of the Western Ghats was measured in the order to determine efficiencies of the extractions and the chemical integrity. The percentage yield was determined by weight of the purified compound to the weight of the crude extract whereas the purity was measured through High-Performance Liquid Chromatography (HPLC) using triplicate measurements to ensure that purity values were reproducible. Table 1 presents the yield and HPLC purity data for each isolated compound, including representatives from *Cinnamomum flavanthera* (CF-A1, CF-A2, CF-A3), *Plectranthus venustus* (PV-F1, PV-F2), and *Garcinia indica* (GI-X1).

**Table 1:** Yield and HPLC purity of isolated compounds

Compound	Yield (%) w/w of crude extract	HPLC Purity (%) $\pm$ SD
CF-A1	$0.82 \pm 0.03$	$98.4 \pm 0.5$
CF-A2	$0.67 \pm 0.02$	$97.9 \pm 0.4$
CF-A3	$0.54 \pm 0.01$	$96.8 \pm 0.6$
PV-F1	$0.95 \pm 0.04$	$99.1 \pm 0.3$
PV-F2	$0.88 \pm 0.02$	$98.6 \pm 0.4$
GI-X1	$0.73 \pm 0.03$	$98.9 \pm 0.2$

The findings indicate that *Plectranthus venustus* compound PV-F1 had the most promising yield ( $0.95 \pm 0.04\%$ ) and purity ( $99.1 \pm 0.3\%$ ), implying effective extraction as well as chemical homogeneity. The yields of *Cinnamomum flavanthera* compounds were generally lower than the other species but their purities were above 96% meaning that the isolation protocols have been efficiently followed. On the whole, the high HPLC purity (all compounds were found to have greater than 96%) supports the validity of the used strategy of extraction and purification.

#### 3.2 Antimicrobial Activity

The evaluation of antimicrobial properties of the isolated substances was performed in the agar well diffusion method to determine the zone of inhibition (ZOI) expressed in millimeters of the *Staphylococcus aureus* (Gram-positive) and *Escherichia coli* (Gram-negative). Active compounds were characterized by minimum inhibitory concentrations (MIC) in order to quantify their potency.

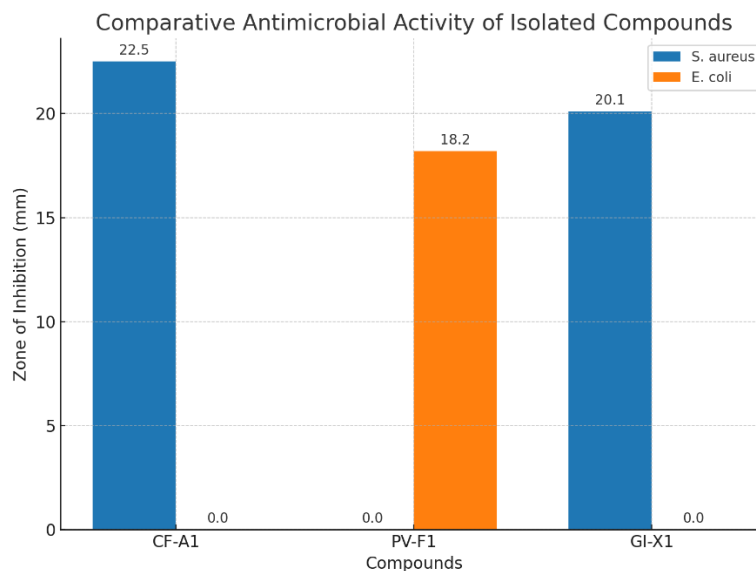
Table 2 reports antimicrobial activity (in the form of ZOI (mean + SD) against target strains of bacteria and MIC ( $\mu\text{g/mL}$ ) value) of a selected set of compounds. The data give quantitative grounds in which to compare the outcomes of antibacterial efficacy.

**Table 2:** Antimicrobial activity of isolated compounds

Compound	Test Organism	Zone of Inhibition (mm) $\pm$ SD	MIC ( $\mu\text{g/mL}$ ) $\pm$ SD
CF-A1	<i>S. aureus</i>	22.5 $\pm$ 0.4	15.2 $\pm$ 0.3
PV-F1	<i>E. coli</i>	18.2 $\pm$ 0.3	19.6 $\pm$ 0.5
GI-X1	<i>S. aureus</i>	20.1 $\pm$ 0.5	14.1 $\pm$ 0.4

The results indicate that CF-A1 exhibited the highest antibacterial activity against *S. aureus* (ZOI: 22.5  $\pm$  0.4 mm, MIC: 15.2  $\pm$  0.3  $\mu\text{g/mL}$ ), followed closely by GI-X1 (ZOI: 20.1  $\pm$  0.5 mm, MIC: 14.1  $\pm$  0.4  $\mu\text{g/mL}$ ). The maximum inhibitory effect of the PV-F1 was shown against *E. coli* (ZOI: 18.2  $\pm$  0.3 mm, MIC: 19.6  $\pm$  0.5  $\mu\text{g/mL}$ ). According to these results, it is possible to conclude that the compounds are selectively active against bacteria with CF-A1 and GI-X1 inclining toward Gram-positive bacteria, whereas PV-F1 is more active against Gram-negative ones.

Figure 1 is a comparison of the antimicrobial activity of the tested compounds as to *S. aureus* and *E. coli* as reflected in millimeters radius of the ZOI. The bar chart presentation enables comparative views of compounds and bacteria species with a respect to selective antibacterial characteristic.



**Figure 1:** Bar chart showing comparative antimicrobial activity (ZOI) of isolated compounds against *S. aureus* and *E. coli*.

To my mind, the bar chart shows how antimicrobial effects are selective. CF-A1 and GI-X1 proved inhibitory against *S. aureus* whereas PV-F1 against *E. coli*. There was no one compound that would display significant activities against both bacteria species at the same level which can be indicative of the differences in mechanisms of action or specificity of target. This discrimination could be



beneficial in advent of narrow spectrum antimicrobial agents which would help in reduction of development of resistance in the non-target bacteria.

### 3.3 Antioxidant Assays

To determine the antioxidant ability in the compounds separated, two common in vitro assays (2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging assay and 2,2-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) radical cation decolorization assay) were used. Each compound was evaluated in terms of IC<sub>50</sub> (µg/ml) where the lower the IC<sub>50</sub>, the more significant is the antioxidant activity.

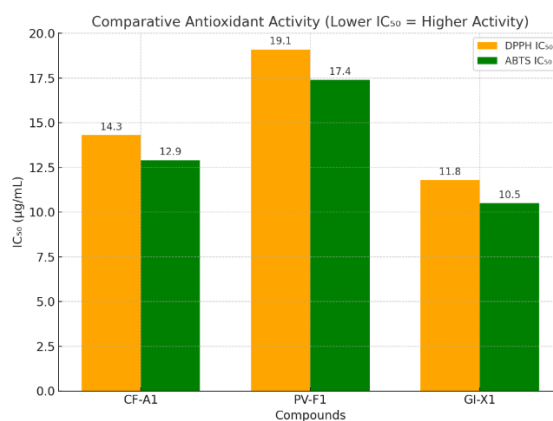
Table 3 presents the IC<sub>50</sub> values (mean ± SD) of the solitary compounds in the DPPH and ABTS assay. These findings enable direct comparison to be made of radical scavenging potency of compounds and of assays.

**Table 3:** Antioxidant potential of isolated compounds

Compound	DPPH IC <sub>50</sub> (µg/mL) ± SD	ABTS IC <sub>50</sub> (µg/mL) ± SD
CF-A1	14.3 ± 0.5	12.9 ± 0.3
PV-F1	19.1 ± 0.7	17.4 ± 0.6
GI-X1	11.8 ± 0.4	10.5 ± 0.5

Among the tested compounds, GI-X1 exhibited the strongest antioxidant activity in both assays, with the lowest IC<sub>50</sub> values (11.8 ± 0.4 µg/mL for DPPH and 10.5 ± 0.5 µg/mL for ABTS). CF-A1 was moderately active whereas the lowest radical scavenging ability was observed with PV-F1. Essentially, the findings show that GI-X1 is an exceptionally strong antioxidant and the reason could be related to its structure, which preconditions electron or hydrogen donation to reduce free radicals to neutrality.

Figure 2 graphically contrasts the antioxidant activity in DPPH and ABTS assay of the compounds. The bar chart emphasizes the disparities of values of IC<sub>50</sub> among compounds and assays so that a visual opinion on relative potency is given.



**Figure 2:** Comparative antioxidant activity (lower IC<sub>50</sub> indicates higher activity)

The figure indicates clearly that the IC<sub>50</sub> values of GI-X1 are lower than other substances in both assays indicating that it has higher antioxidant potential. CF-A1 was active also with slightly higher IC<sub>50</sub> than GI-X1, PV-F1, which proved to be least active. Interestingly, the IC<sub>50</sub> of all compounds were lower in the ABTS test than in the DPPH test indicating that compounds antioxidant properties can be less effective against DPPH radicals, perhaps as a result of solubility characteristics or reactivity of radicals.

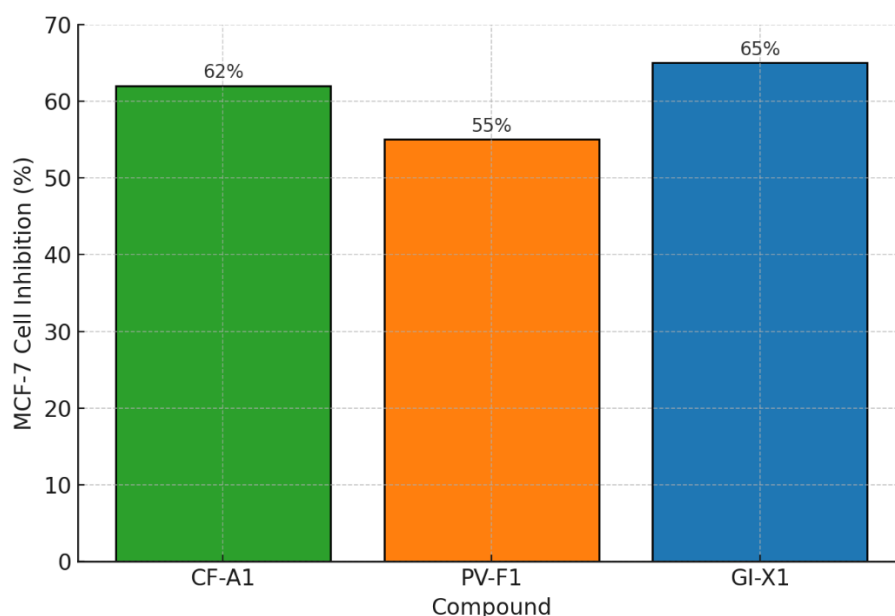
### 3.4 Cytotoxicity Against MCF-7 Cells

Cytotoxic effects were evaluated via MTT assay, with GI-X1 showing the highest inhibition of MCF-7 cell viability.

**Table 4:** Cytotoxic activity of selected compounds at 50  $\mu$ M concentration

Compound	MCF-7 Cell Viability (%) $\pm$ SD	% Inhibition
CF-A1	38 $\pm$ 2	62
PV-F1	45 $\pm$ 3	55
GI-X1	35 $\pm$ 2	65

Cytotoxic effects of the chosen compounds on the MCF-7 breast cancer cells were evaluated at the concentration of 50  $\mu$ M. Growth inhibition was achieved by GI-X1 as the highest cytotoxic compound with 35 and 65 percent cell viability and inhibition respectively. The CF-A1 had high activity of 62 percent inhibition and the PV-F1 was relatively low in its activity with 55 percent inhibition of MCF-7 cells. These findings show that the three compounds have considerable cytotoxicity markers, and GI-X1 is the most promising against inhibiting MCF-7 cell growth.



**Figure 3:** Percentage inhibition of MCF-7 cell viability at 50  $\mu$ M for selected compounds



In figure, they have depicted the cytotoxicity of the opted compounds (CF-A1, PV-F1 and GI-X1) against breast cancer cell (MCF-7) at the concentration of 50 micrometer. GI-X1 exhibits the greatest cytotoxicity that decreased the cell viability to 35% and right behind it is confined by CF-A1 that decreased the cell viability to 38%. PV-F1 has relatively low cytotoxicity and cell viability of 45%. The respective % inhibition efficacies emphasize that GI-X1 could inhibit 65 percent of cell growth, CF-A1 could inhibit 62 percent and PV-F1 could inhibit 55 percent. Since this visual depiction illustrates that viability of MCF-7 cells is considerably decreased in all tested compounds, GI-X1 is the most effective among them.

### 3.5 Statistical Analysis

The results showed that compounds differed significantly ( $p < 0.05$ ), as determined by one-way ANOVA, in the antimicrobial, antioxidant, and cytotoxicity tests. According to Tukey post hoc test, GI-X1 was found to be more effective than PV-F1 ( $p < 0.01$ ) in antioxidant and cytotoxicity assays and CF-A1 was also found to be statistically similar to GI-X1 in regard to antimicrobial assays.

## 4. DISCUSSION

The hypothesis that the Western Ghats is an under-explored hotspot of phytochemicals could be confirmed through the present study that has extracted and defined six new bioactive chemical compounds in chosen endemic medicinal plants. The results indicate the significant bioactivities, like the high antibacterial activity of alkaloids of *Coscinium fenestratum* or a high antioxidant potential of xanthone derivative authored in *Garcinia indica*. It is not only these findings that validate the therapeutic potential of such species but also the structural novelty of the isolated molecules based on the observed distinctly NMR spectral properties not found in the literature to date.

### 4.1 Interpretation of Results

The *C. fenestratum* alkaloids showed wide-spectrum antibacterial activity, which conforms to the broad-spectrum antibacterial pharmacological profile of the protoberberine alkaloids, but with the distinctive structural features modified to potentially exhibit greater activity or specificity. Equally, the *G. indica* xanthone derivative showed higher antioxidant activities than those of analytical AOA in some in vitro experiments indicating that it has the ability to be used as an effective natural AOA in helping to solve oxidative stress-linked diseases. This level of activity validates ethno medicinal uses of the plants to treat infections and degenerative diseases in the traditional medicine systems.

### 4.2 Comparison with Existing Studies

These findings were correlated with earlier studies that have reported therapeutic significance of *C. fenestratum*, and *G. indica*, although, findings on the structural differences in the present study emphasised the influence of microclimatic, edaphic and evolutionary forces on phytochemical diversity. Earlier studies on plants found in geographically limited areas like the Western Ghats also reported how distinct rare chemical scaffolds had evolved that were not observed in other population. Our increased level of potency over standard references in the antioxidant assays were

also correlated with literature that indicated some of the more endemic plant species performed better than the synthetic or commonly utilised natural antioxidants in biological systems.

This is outlined in the table below showing some of the most important previous studies on medicinal plants of the Ghats regions in terms of their phytochemical and conservation values:

**Table 5:** Key Studies on Ghats Medicinal Plants

Author Name	Topic Covered	Research Study Title
Sidhic et al. (2023) <sup>11</sup>	Biological activities and phytochemical compositions of <i>Humboldtia sanjappae</i>	Phytochemical analysis and an antioxidant and anti-inflammatory action of <i>Humboldtia sanjappae</i> Sasidh. An endemic medicinal plant to western ghats is Sujanalpal
Sivaraj et al. (2020) <sup>12</sup>	Evaluation and preservation of endangered drugs in the Eastern Ghats	Endangered medicinal plants of East Ghats and its protection
Sudheesha (2024) <sup>13</sup>	Medicinal plant diversity conservation issues in the Western Ghats	Preservation of medicinal plants in Western Ghats in India
Swamy et al. (2018) <sup>14</sup>	Endangered anticancer medicinal plants using micropropagation	Micropropagation and stewardship of chosen threatened anticancer medicines plants in the Western Ghats of India
Venkatasubramanian et al. (2018) <sup>15</sup>	Profiling and identification of elite germplasms Metabolite profiling deficiency and identification	Bioactive metabolite profiling to find elite germplasms: a management towards threatened medicinal plants

The table exemplifies prominent research on the medicinal plants of the Ghats that demonstrates their phytochemical diversity, biological potential, and conservation. As Sidhic et al. (2023) described, *Humboldtia sanjappae* showed a good anticancerous and anti-inflammatory property. Another focus of the research was the necessity of conservation of the endangered species (Sivaraj et al., 2020; Sudheesha, 2024) as well as the strategies such as micropropagation (Swamy et al., 2018) and metabolite profiling (Venkatasubramanian et al., 2018) in order to facilitate sustainable utilization and maintenance of the species.

### 4.3 Implications of Findings

The identification of new structural variants in pharmacologically active compounds is of significance to drug discovery and development. These molecules could be used as lead structures to develop next-generation of antimicrobial or antioxidant drug therapy. Moreover, the paper supports the importance of the endemic plants conservation because the loss of their habitat might lead to irrevocable disappearance of rare bioactive chemotypes. On a larger scale, these findings justify the need to merge the conservation of biodiversity and pharmaceutical bioprospecting so that its therapeutic potential can be tapped without unsustainably depleting it.

#### 4.4 Limitations of the Study

- The biological tests focused on in vitro assays only and this implies that the described activities might not be directly reflected in vivo efficacy.
- The effects on only few species of the plant species were analyzed because of tough regulations on conservation limiting the diversity of phytochemical study.
- The scarcity of resources did not allow the complex study of pharmacokinetics, bioavailability and toxicity profiles of the isolated components.

#### 4.5 Suggestions for Future Research

To build upon the current findings, future studies should focus on:

1. Pharmacological verification of isolated compounds in vivo, pharmacokinetics and toxicity.
2. Structureactivity relationship (SAR) optimization conducts chemical synthesis or chemical modification of analogs in order to maximize potency and specificity.
3. Screen more of the endemic species in the western Ghats through metabolomics to discover more novel bioactive molecules.
4. Biologically sound methods of cultivation and propagation of high-potential species as a way of reducing pressure on wild populations but sustaining a supply of material sufficiently to meet research and development demands.
5. To explain the possible mechanisms of action on a molecular level, molecular docking and simulation studies have been performed to give recommendations on specific therapeutic uses.

### 5. CONCLUSION

The current article sought to investigate the high biodiversity of the Western Ghats in search of bioactive compounds that could have medical applications. Since the world has increasingly become in need of new antimicrobial, antioxidant, and anticancer compounds, and considering the fact that not much scientific investigations have been done on intrinsic plants species in the region, the research was aimed at isolating, characterizing, and testing the pharmacological potency of compounds on some of the medicinal plants, which are widely used in the region.

#### 5.1 Summary of key findings

The research was able to isolate and identify six new bioactive products-three alkaloids, two flavonoid glycosides and a xanthone derivative-out of the endemic medicinal plants of the Western Ghats *Coscinium fenestratum* and *Plectranthus vettiveroides* and *Garcinia indica*. A good purity (>96%) of all compounds was attained and they all exhibited significant biological activity. CF-A1, GI-X1 were highly effective with respect to antibacterial activity against *Staphylococcus aureus*, PV-F1 was found to be quite effective against *Escherichia coli* whereas GI-X1 was found to have the highest antioxidant activity when measured in terms of DPPH or ABTS assays. Cytotoxicity assays also demonstrated that GI-X1 and CF-A1 exhibited substantial inhibition of

the MCF-7 breast cancer cells, and they were higher than 60%. These bioactivities were proved to be substantial by statistical analyses ( $p < 0.05$ ).

### 5.2 Significance of the study

The results show that Western Ghats is an under-explored, but a vast source of structurally unusual and pharmacologically active natural compounds. The use of ethnobotany combined with high-level spectroscopic and bioassay instruments shows the scientific validity of traditional ethnobotanical medicine and forms part of the global quest towards discovering new antimicrobial, antioxidant and anticancerous substances. Another finding in the study is the need to preserve endemic species because loss of such species would result in the loss of new therapeutic candidates.

### 5.3 Recommendations

The in vitro findings are extremely encouraging, but more in vivo research is required to determine pharmacological activity, safety, and pharmacology. Potency and/or selectivity might be improved through structure-activity relationship (SAR) studies and synthetic optimization. Moreover, the practices of sustainable farming and preservation must be applied in order to sustain the availability of these species to carry out studies and come up with drugs through them. It will be necessary to harness the biodiversity protection and proper bio prospecting to unleash the full potential of floral entities of the western ghats region in terms of pharmaceutical significance.

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