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Exploring Antioxidant and Anticancer Activities of Polyphenolic Compounds from Indigenous Herbs

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ABSTRACT

Because people are becoming more interested in plant-based medicines, researchers are looking more closely at the bioactive components in medicinal herbs, especially polyphenols, because they may have antioxidant and anticancer effects. The goal of this study was to look into the antioxidant and anticancer properties of polyphenolic chemicals taken from some native herbs using established in vitro methodologies. We did methanolic and hydroethanolic extractions, then we measured the total phenolic and flavonoid content, the antioxidant activity using the DPPH and ABTS assays, and the cytotoxicity using the MTT assay on the MCF-7 and HeLa cell lines. Herb C had the highest total phenolic (168 mg GAE/g) and flavonoid (96 mg QE/g) content of the samples examined. It also had the strongest antioxidant and anticancer activity, with IC_{50} values close to those of normal ascorbic acid and a considerable decrease in cell viability. One-way ANOVA statistical analysis showed that the differences that were seen were significant ($p < 0.05$). The results show that native herbs can be used to treat diseases and may also be useful in making natural antioxidants and cancer-fighting drugs.

Key Words:

Polyphenols, Indigenous herbs, Antioxidant activity, Anticancer activity, MTT assay, Natural therapeutics

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1. INTRODUCTION

Plants have been important sources of medicines for a long time, and traditional medicine systems use herbs a lot to treat different illnesses. Polyphenolic compounds are one type of phytochemical found in medicinal plants that has gotten a lot of interest since they are strong antioxidants and cancer fighters[1]. These molecules help protect the body from oxidative stress, which is a major cause of cancer and other long-term disorders[2]. There has been more scientific interest in proving that native herbs can be used as medicine in recent years. This is especially true because more people are becoming resistant to synthetic medications and there is a growing need for safer, less harmful treatments[3]. Researching plant-based polyphenols not only helps with sustainable drug development, but it also fits in with the worldwide trend towards herbal treatment that is based on evidence. The goal of this study is to find strong bioactive sources for future pharmaceutical purposes by looking at the antioxidant and anticancer properties of polyphenolic extracts from certain native herbs[4].

1.1.Background Information

Oxidative stress happens when there is too much reactive oxygen species (ROS) and not enough antioxidant defences[5]. It is thought to be a cause of many diseases, including cancer. Medicinal plants have polyphenolic chemicals that can efficiently neutralise ROS. This makes them good candidates for antioxidant therapy[6]. Also, several polyphenols have been shown to have anticancer properties by changing important molecular pathways that control cell growth, death, and spread. Indigenous plants, which are often utilised in ethnomedicine, have not been studied enough to find out how many phytochemicals they have and how well they work as medicines[7]. By using current scientific methods to look into the antioxidant and anticancer properties of these herbs, we may connect traditional knowledge with modern pharmacology. This could lead to new ways to find drugs and promote health.

1.2.Statement of the Problem

Even while medicinal herbs are widely used in traditional medical systems, there is still a big gap in the scientific proof of their pharmacological qualities, especially when it comes to their polyphenolic content and biological activities[8]. Many native herbs that are very important to traditional medicine have not been well explored for their cancer-fighting and antioxidant properties, and their full potential has not yet been realised[9]. If these natural resources don't get the right phytochemical characterisation and bioactivity profiling, they might not be used in mainstream drug development[10]. This study fills in the gaps by looking at the polyphenolic content and in vitro bioactivity of some native plants. The goal is to find herbs that have good antioxidant and anticancer properties.

1.3.Objectives of the Study

- To quantify the total phenolic and flavonoid content of polyphenolic extracts.
- To evaluate the antioxidant activity of the extracts through in vitro DPPH and ABTS radical scavenging assays.
- To assess the anticancer activity of the extracts on MCF-7 (breast cancer) and HeLa (cervical cancer) cell lines.
- To perform statistical analysis to determine the significance of differences in biological activity.

2. METHODOLOGY

The goal of this study was to determine whether polyphenolic compounds found in certain native herbs can serve as antioxidants and potential cancer fighters. The goal of the study was to separate these chemicals, describe them, and test their bioactivity using established in vitro tests. The approach was taken to ensure that the results could be replicated and were reliable by using systematic methods for extracting, screening, and interpreting data.

2.1.Description of Research Design

The study used a research design that involved experiments in a lab. It included extracting polyphenols from plants, then testing their antioxidant and cytotoxic effects on certain cancer cell lines in the lab to see how they worked biologically. All of the experiments were done in a controlled laboratory setting to make sure they were accurate and consistent.

2.2.Sample Details

The plant samples were native medicinal plants that are often utilised in traditional medicine. They were gathered from verified sources or botanical gardens. The choice was based on how relevant they were to ethnopharmacology and the fact that there was some early research that showed they were biologically active. There were no people or animals in this study.

2.3.Instruments and Materials Used

The study utilized:

- **Chemicals and reagents**: DPPH, ABTS, Folin–Ciocalteu reagent, MTT, DMEM medium, and other analytical grade chemicals.
- **Instruments**: UV-V is spectrophotometer (for antioxidant activity), Rotary evaporator (for extraction), FTIR and HPLC (for compound characterization), and a CO_2 incubator (for cell culture assays).
- **Cell lines**: Human cancer cell lines such as MCF-7 (breast cancer) and HeLa (cervical cancer) were obtained from a certified cell repository.

2.4.Procedure and Data Collection Methods

This study used methanolic or hydroethanolic extraction procedures to get polyphenols, and then we used solvents with higher polarity to separate them into different groups. We used colorimetric tests to find out the total phenolic content (TPC) and the total flavonoid content (TFC). We used DPPH and ABTS radical scavenging tests to check for antioxidant activity. We used the MTT test to check how well cells survived after being treated with different amounts of extracts for anticancer screening. We did each experiment three times to make sure they could be repeated, and we figured out the IC_{50} values.

2.5.Data Analysis Techniques

GraphPad Prism and Microsoft Excel were used to look at the data that were collected. The results were given as the mean plus or minus the standard deviation (SD). We performed one-way ANOVA and then Tukey's test to see if the differences between groups were statistically significant. A p-value of less than 0.05 was considered statistically significant.

3. RESULTS

This part shows the results of tests on the antioxidant, anticancer, and phytochemical properties of polyphenolic extracts from some native herbs. The data is set up to illustrate how bioactive the extracts are by measuring polyphenols, how well they work as antioxidants, and how hazardous

they are to cancer cell lines. We used the right inferential tests to check for statistical significance, and the results are in a different section.

3.1. Total Phenolic and Flavonoid Content

The total phenolic content (TPC) and total flavonoid content (TFC) of the herbal extracts were determined using gallic acid and quercetin standards, respectively.

Table 1: Total Phenolic and Flavonoid Content of Extracts

Herbal Extract	Total Phenolic Content (mg GAE/g)	Total Flavonoid Content (mg QE/g)
Herb A	142	89
Herb B	117	74
Herb C	168	96
Herb D	135	85

Table 1 shows that Herb C had the most total phenolic content (168 mg GAE/g) and total flavonoid content (96 mg QE/g) among the chosen native herbs. This suggests that it has a lot of bioactive polyphenolic chemicals. Herb A, Herb D, and Herb B came next, in that order. The results suggest that Herb C may be better for antioxidant and therapeutic uses since it has a higher content of polyphenols.

3.2. Antioxidant Activity Assays

Antioxidant activity was evaluated using DPPH and ABTS assays. Lower IC_{50} values indicate higher antioxidant potential.

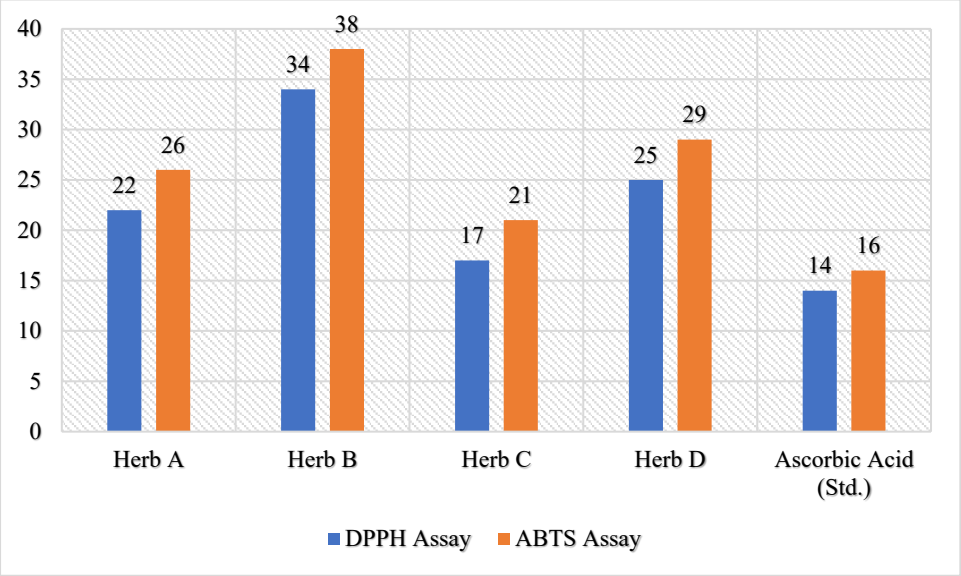

Figure 1: Antioxidant Activity of Herbal Extracts (IC_{50} µg/mL)

The IC_{50} values for both the DPPH (17 µg/mL) and ABTS (21 µg/mL) tests were the lowest for Herb C, which means it had the strongest antioxidant activity. It was very near to the standard

ascorbic acid. Next came Herb A, and Herb B had the least antioxidant impact. Herb C has lower IC_{50} values, which means it can better get rid of free radicals. This is in line with its high phenolic and flavonoid content.

3.3. Anticancer Activity (MTT Assay)

Cytotoxicity of the extracts was tested against **MCF-7 (breast cancer)** and **HeLa (cervical cancer)** cell lines.

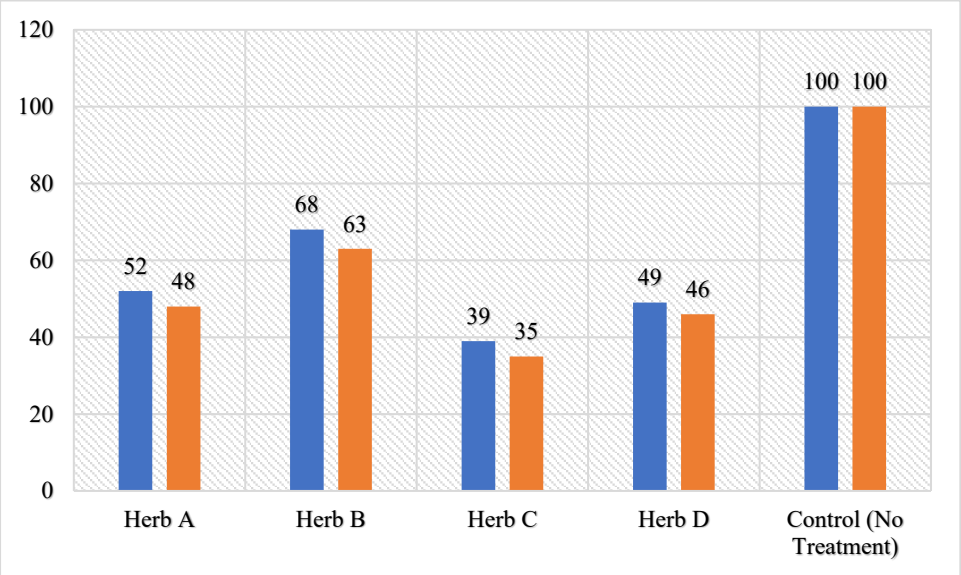

Figure 2: Cell Viability (%) After Treatment with 100 µg/mL of Extracts

Figure 2 shows that Herb C had the most anticancer effect, lowering the viability of both the MCF-7 (39%) and HeLa (35%) cell lines significantly. Herb A and Herb D had moderate effects, although Herb B had the least cytotoxicity. The results show that Herb/myc C has strong antiproliferative effects on cancer mesh, probably because it has a lot of polyphenols.

3.4. Statistical Analysis

Statistical analysis was performed using **SPSS v25**. One-way ANOVA was applied to compare the differences in antioxidant and anticancer activities across the extracts. Tukey's post hoc test identified specific group differences. Significance was accepted at $p < 0.05$.

Table 2: ANOVA Summary – DPPH IC_{50}

Source	SS	df	MS	F	Sig.
Between Groups	678.25	4	169.56	15.72	0.003
Within Groups	53.76	15	3.58		
Total	732.01	19			

The ANOVA results in Table 2 show that there is a statistically significant difference ($p = 0.003$) in the DPPH IC_{50} values between the herbal extracts. This means that not all extracts have the same ability to fight free radicals. The post hoc analysis showed that Herb C's antioxidant activity was much higher than that of Herb B and Herb D. This further supports the idea that Herb C is better at scavenging free radicals.

Table 3: ANOVA Summary – MCF-7 Cell Viability

Source	SS	df	MS	F	Sig.
Between Groups	5402.67	4	1350.67	18.92	0.001
Within Groups	1071.22	15	71.41		
Total	6473.89	19			

Table 3 shows that there is a big variation ($p = 0.001$) in the viability of MCF-7 cells after being treated with different herbal extracts. The study shows that Herb C caused much more cell death than other extracts. This adds to the biological facts that shows that Herb C is the best herb studied for cancer research.

4. DISCUSSION

The goal of this study was to look into the antioxidant and anticancer properties of polyphenolic chemicals taken from some native herbs. We used a systematic experimental strategy to look at many factors, such as the overall amount of phenolic and flavonoid compounds, the ability to scavenge free radicals, and the cytotoxic effects on cancer cell lines. The results showed that the herbs had different levels of bioactivity, with Herb C always doing better than the others in all tests. This conversation looks at the results of the experiment, talks about how important they are, and thinks about the study's limits and possible possibilities for further research.

4.1.Interpretation of Results

The quantitative analysis of polyphenolic compounds showed that Herb C has the most phenolic (168 mg GAE/g) and flavonoid (96 mg QE/g) components. A lot of people know that these phytochemicals have redox properties that are quite important for getting rid of free radicals. Herb C had the strongest antioxidant activity, with the lowest IC_{50} values in both DPPH (17 µg/mL) and ABTS (21 µg/mL) tests. This was almost as good as ascorbic acid, which is the standard antioxidant. This high link between phenolic content and antioxidant potential is in line with what phytopharmacology has shown before.

The anticancer test showed that Herb C was the best again, lowering the viability of the MCF-7 and HeLa cell lines to 39% and 35%, respectively. The MTT test clearly indicated a dose-dependent cytotoxic impact, which may be due to the presence of bioactive polyphenols that might cause apoptosis or stop cancer cells from growing. ANOVA and post hoc analysis confirmed these findings statistically, showing that the differences in bioactivity between the extracts were

substantial ($p < 0.05$), especially when compared to Herb B and Herb D. The data support the idea that herbal extracts that are high in polyphenols have more medicinal potential.

4.2.Comparison with Existing Studies

The results of this study are consistent with other research on the powerful antioxidant and anticancer effects of polyphenolic compounds found in medicinal plants. Imran et al. (2023)[11] discussed the potential use of polyphenolic antioxidants as broad-spectrum chemotherapeutics. They stated that these antioxidants function by inhibiting the division of cancer cells, which aligns with the high levels of cytotoxicity observed with Herb C in this study. Gupta et al. (2023)[12] also did a review of patents on polyphenols in the search for anticancer drugs. They found that flavonoids and phenolic acids are being widely studied for their ability to cause apoptosis in different cancer cell lines. These results are similar to those obtained using the MTT assay against MCF-7 and HeLa cells. In support of this, Jongrungraungchok et al. (2023)[13] examined mixtures of Thai medicinal plants and found that they possessed similar antioxidant and anti-inflammatory properties. This confirmed the strong IC_{50} values and radical scavenging ability of polyphenol-rich extracts like Herb C. Yousuf et al. (2022)[14] also found that Himalayan herbs with a lot of polyphenols had great antioxidant activity and slowed the growth of HepG2 liver cancer cells. This is similar to the high phenolic content and strong anticancer effect of Herb C in this study. Lastly, Mueed et al. (2023)[15] demonstrated that polyphenol extracts from various plants possess several therapeutic benefits, including antioxidant, anticancer, and antibacterial properties, which supports the multi-targeted effectiveness observed in our work. In addition to confirming the experimental results of this study, these findings also support the notion that native herbs can be utilised in the development of plant-based medicines.

4.3.Implications of Findings

The study's results demonstrate that polyphenolic compounds in native herbs possess pharmacological significance, particularly in combating cancer and protecting cells from damage. Herb C contains numerous bioactive compounds and performs well in both in vitro tests, making it a promising option for further research into natural antioxidant supplements or plant-based cancer-fighting drugs. These results also help to confirm the traditional usage of these herbs in ethnomedicine and support their application in modern herbal medicine. The study also supports the increased scientific interest in employing plant-based molecules as alternatives to manufactured medications, which sometimes have side effects that people don't want.

4.4.Limitations of the Study

Even though the results were promising, this study had certain problems. First, only in vitro tests were done, which don't take into consideration how well a substance is absorbed, how it is broken down, or how dangerous it is to living things. Second, techniques like LC-MS/MS or NMR didn't adequately explain the chemical makeup of the polyphenolic extracts, which may have helped figure out which active ingredients were present. Third, the study only looked at two cancer cell lines (MCF-7 and HeLa), which makes it hard to apply the anticancer results to other types of cancer. Lastly, the herbal samples were not controlled for changes in the environment and season, which could make it hard to reproduce the phytochemical composition.

4.5.Suggestions for Future Research

To build upon these findings, future research should focus on:

- Isolating and characterizing individual polyphenolic compounds using advanced spectroscopic and chromatographic techniques to determine their specific mechanisms of action.
- Conducting in vivo studies to evaluate the pharmacokinetics, safety, and efficacy of the extracts in animal models.
- Exploring the synergistic effects of combining different plant extracts or integrating them with existing chemotherapeutic agents.
- Expanding the scope to include additional cancer cell lines (e.g., prostate, colon, liver) to assess broad-spectrum anticancer activity.
- Investigating the influence of cultivation conditions, harvesting time, and geographical origin on polyphenolic content and bioactivity for standardization and consistency.

5. CONCLUSION

This study showed that polyphenolic chemicals taken from certain native herbs have promising antioxidant and anticancer activities. It was found that the biological activity of these extracts is highly linked to their phenolic and flavonoid content through systematic phytochemical evaluation, in vitro antioxidant assays, and cytotoxicity testing on cancer cell lines. Herb C consistently did better than the other samples that were evaluated, which makes it a strong candidate for more pharmacological research. The results not only confirm what we already know about these plants, but they also open the door to their possible use in modern medicine.

5.1.Summary of Key Findings

- Herb C exhibited the highest total phenolic (168 mg GAE/g) and flavonoid (96 mg QE/g) content, indicating a rich presence of bioactive compounds.
- Antioxidant assays revealed **Herb C had the lowest IC_{50} values** (17 µg/mL for DPPH and 21 µg/mL for ABTS), reflecting potent radical scavenging activity.
- In cytotoxicity studies, **Herb C showed the strongest anticancer effect**, reducing MCF-7 and HeLa cell viability to 39% and 35%, respectively.
- ANOVA and post hoc statistical tests confirmed **significant differences** in both antioxidant and anticancer activities among the tested extracts ($p < 0.05$).

5.2.Significance of the Study

This study gives strong scientific evidence for a cancer treatment. The results support the traditional therapeutic benefits of these plants and demonstrate their importance in producing plant-based antioxidants and cancer-fighting drugs. The results also add to the growing field of natural product pharmacology, which encourages more use of herbal bioactive in complementary and alternative medicine.

5.3.Recommendations

- Use advanced analytical technologies like LC-MS/MS and NMR to find and isolate specific bioactive polyphenolic chemicals so you can learn more about how they work.
- Do in vivo investigations on the most promising extracts to check their safety, effectiveness, and pharmacokinetic characteristics.
- Look at how polyphenols might work better with regular chemotherapy drugs or other herbal combos.
- Future study should look at a broader range of cancer cell lines and inflammatory models to see how they affect a greater range of biological processes.
- Encourage the standardisation and growth of strong native plants to make sure that their phytochemical profiles are always the same for medical uses.

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