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**Gymnema sylvestre Leaf Hydroalcoholic Extract: Promising Source of Antioxidants with Remarkable Yield and Potent Scavenging Activity**

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**ABSTRACT:**

This research study aimed to compare the antioxidant activity and yield of different extracts of leaf of *Gymnema sylvestre*, including methanolic, hydroalcoholic, and aqueous extracts. In recent years, extensive research has been conducted to explore the antioxidant potential of *Gymnema sylvestre* extracts. However, a comprehensive comparison of these extracts in terms of their yield and antioxidant activity is lacking. Therefore, the objective of this study was to evaluate and compare the yield and antioxidant activity of different extracts of *Gymnema sylvestre*, specifically methanolic, hydroalcoholic, and aqueous extracts. The percentage yield of each extract was determined, providing valuable insights into the efficiency of the extraction methods. Furthermore, the antioxidant activity of the extracts was assessed by measuring their scavenging activity and determining the IC50 values. Methanolic, hydroalcoholic, and aqueous extracts of *Gymnema sylvestre* leaf were obtained using the Soxhlet extraction method after being finely pulverized. Within the limitations of this study, it was concluded that the hydroalcoholic extract of *Gymnema sylvestre* exhibited the highest percentage yield, with a recorded value of 10.27 percent. The IC50 values for the Gymnema extract and Ascorbic acid were determined to be 59.09 µg/ml and 44.90 µg/ml, respectively. These findings indicate the potential antioxidant activity of *Gymnema sylvestre* extracts, with the hydroalcoholic extract showing the most promising results in terms of both yield and scavenging activity. Further investigations are needed to explore the underlying mechanisms and assess the potential health benefits of these extracts in various applications.
Introduction:

*Gymnema sylvestre* is a medicinal plant that is indigenous to Asia’s tropical regions, and it has recently received a lot of attention because of its possible therapeutic benefits.¹ It has a lengthy history of usage in conventional medicine, especially in Ayurveda, where it has been used to treat a variety of conditions, such as diabetes, obesity, and inflammation.²

*Gymnema sylvestre* is well known for its bioactive chemicals, with gymnemic acid being one of the key components that have demonstrated remarkable pharmacological activities.³ By combating the negative effects of free radicals and lowering oxidative stress, antioxidants serve a critical role in preserving cellular health. Free radicals, such as reactive oxygen species (ROS), are produced in the body by normal metabolic processes as well as exposure to external factors such as pollution, radiation, and poisons.

Oxidative stress occurs when the body’s antioxidant defense mechanisms are overwhelmed by the generation of free radicals, resulting in cellular damage and the emergence of a number of chronic diseases. Natural antioxidants derived from plants have become promising therapeutic agents in treating oxidative stress-related disorders.⁴ *Gymnema sylvestre* has generated a lot of interest as a possible source of natural antioxidants due to its extensive phytochemical profile.

Numerous research has looked at the antioxidant activity of *Gymnema sylvestre* extracts and shown their capacity to scavenge free radicals, prevent lipid peroxidation, and guard against oxidative damage.⁵ However, there is still a need for a thorough comparison of diverse extracts prepared using various solvent systems, despite the expanding body of research on *Gymnema sylvestre* and its antioxidant potential. The antioxidant activity and composition of the extracted molecules can be greatly influenced by solvent choice.

Therefore, the primary objective of this study was to evaluate and compare the yield and antioxidant activity of different extracts of *Gymnema sylvestre* obtained through Soxhlet extraction using different solvents, including methanol, hydro alcohol and water. The Soxhlet extraction method was employed due to its efficiency in extracting bioactive compounds from plant material. The percentage yield of each extract was determined, providing valuable insights into the efficiency of the extraction process.

Additionally, the antioxidant activity of the extracts was assessed by measuring their scavenging activity and determining the IC50 values, which indicate the concentration at which 50% of the free radicals are inhibited. The findings from this study have significant implications for optimizing the extraction process and identifying the most potent extract with the highest yield and antioxidant activity. This knowledge is crucial for the development of *Gymnema sylvestre*-based products in the pharmaceutical, nutraceutical, and functional food industries. Moreover, understanding the antioxidant potential of *Gymnema sylvestre* extracts obtained through Soxhlet extraction using different solvents contributes to the broader field of natural antioxidants and may provide insights into their mechanisms of action and potential therapeutic applications.

In conclusion, this study aims to elucidate the antioxidant potential of different extracts of *Gymnema sylvestre* obtained through Soxhlet extraction using different solvents. By comparing the yield and antioxidant activity of the methanolic, hydroalcoholic, and aqueous extracts, this research contributes to the body of knowledge on *Gymnema sylvestre* and supports its potential as a natural source of antioxidants for maintaining and promoting human health.

**Materials and Methods**

**Collection of plant materials**
Gymnema sylvestre leaves were collected from the Delhi region in January 2023 while taking into account the seasonal conditions in order to obtain the greatest amount of phytoconstituents.

**Extraction of plant material**

The Gymnema (Gurmar) leaves are sorted, cleaned, and ground to the appropriate mesh (20–60). A soxhlet apparatus is then used to extract it using various solvents.

- **For methanolic extract**
  An extraction with methanol (500 ml at 40°C) was performed using a soxhlet apparatus on 10 g of dried and powdered plant material. After being filtered and completely dried off, the extract was cooled to room temperature.

- **For hydroalcoholic extract**
  Using a soxhlet apparatus, dried and powdered plant material (10 g) was extracted using hydroalcohol (70:30). The extract was cooled at room temperature, filtered and evaporated to complete dryness.

- **For aqueous extract**
  Dried and powdered plant material (10 g) was taken and then extracted with water (aqueous) using soxhlet apparatus. The extract was cooled at room temperature, filtered and then evaporated until completely dry. The percentage yield of all the extracts was calculated.

**DPPH radical scavenging activity**

- The DPPH radical scavenging assay was used to evaluate the extracts' capacity to scavenge free radicals. The ability of the plant extractives to donate hydrogen atoms was assessed using the decolorization of a methanol solution of 2,2-diphenyl-1-picrylhydrazyl (DPPH). In a methanol solution, DPPH produces a violet or purple colour that, in the presence of antioxidants, fades to varying hues of yellow.

- The following equation can be used to determine the percentage of DPPH radical scavenging activity:

  \[
  \text{% DPPH radical scavenging activity} = \left( \frac{A_0 - A_1}{A_0} \right) \times 100
  \]

  Where,
  
  \( A_0 \) is the absorbance of the control
  
  \( A_1 \) is the absorbance of the extractives/standard.

  Then % of inhibition is plotted against concentration, and from the graph IC50 was calculated.

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Figure 2: 96 ELISA well plate

**Procedure for scavenging activity calculation of gymnema sylvestre:**

0.1 ml of standard ascorbic acid solution at concentrations of 10, 20, 40, 60, 80, and 100 g/ml, along with a plant extract, were added to 3 ml of a 0.004% methanol solution of DPPH. The control consisted of a mixture of methanol and DPPH. Absorbance at 517 nm was measured after 30 minutes of incubation in the dark. The percentage inhibitory activity was calculated using 

$$\frac{(A0 - A1)}{A0} \times 100$$

where A0 is the absorbance of the control and A1 is the absorbance of the extract/standard.

**Results and Discussion**

The extraction process using the Soxhlet apparatus was employed to obtain different extracts of Gymnema sylvestre, including methanolic, hydroalcoholic, and aqueous extracts. The percentage yield of each extract was calculated.

For the methanolic extract, the percentage yield was determined to be 8.77%.

Similarly, for the hydroalcoholic extract, the percentage yield was found to be 10.27%.

In the case of the aqueous extract, the percentage yield was determined to be 9.34%.

<table>
<thead>
<tr>
<th>Solvent</th>
<th>Dry weight</th>
<th>Extract weight</th>
<th>% yield</th>
</tr>
</thead>
<tbody>
<tr>
<td>Methanol</td>
<td>10g</td>
<td>0.877g</td>
<td>8.77%</td>
</tr>
<tr>
<td>Hydroalcoholic</td>
<td>10g</td>
<td>1.027g</td>
<td>10.27%</td>
</tr>
<tr>
<td>Aqueous</td>
<td>10g</td>
<td>0.934g</td>
<td>9.34%</td>
</tr>
</tbody>
</table>

According to the findings, the hydroalcoholic extract had the highest percentage yield (10.27%), followed by the aqueous extract (9.34%) and the methanolic extract (8.77%). These results imply that the solvent of choice has a substantial impact on the yield of Gymnema sylvestre leaf extracts.

**Antioxidant activity**

The extract’s antioxidant activity was quantified as an IC50 value. The concentration of extracts (in g/ml) that suppresses the production of DPPH radicals by 50% is known as the IC50 value. The graph was drawn using the average of three observations, and each test was run in triplicate.

<table>
<thead>
<tr>
<th>Solvent</th>
<th>IC50 of extracts(microgram per ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alcoholic extract</td>
<td>67.67</td>
</tr>
<tr>
<td>Hydroalcoholic extract</td>
<td>59.09</td>
</tr>
<tr>
<td>Aqueous extract</td>
<td>63.98</td>
</tr>
<tr>
<td>Ascorbic acid standard</td>
<td>44.90</td>
</tr>
</tbody>
</table>

The antioxidant activity of the extracts was evaluated by measuring their scavenging activity and determining the IC50 values. The IC50 value of Gymnema sylvestre extract and Ascorbic acid were found to be 59.09 µg/ml and 44.90 µg/ml respectively.

**Conclusion**

In conclusion, this research study compared the antioxidant activity and yield of different extracts of Gymnema sylvestre, including methanolic, hydroalcoholic, and aqueous extracts. The findings of this study provide important insights into the potential of Gymnema sylvestre as a
natural source of antioxidants. The results revealed that the hydroalcoholic extract of Gymnema sylvestre exhibited the highest percentage yield (10.27 percent) among the tested extracts.

This suggests that the hydroalcoholic extraction method was particularly effective in extracting bioactive compounds from Gymnema sylvestre. The IC50 values, representing the concentration required to scavenge 50% of free radicals, were found to be 59.09 µg/ml for the gymnema extract and 44.90 µg/ml for ascorbic acid. These findings suggest that Gymnema sylvestre extracts possess significant antioxidant potential, although the antioxidant activity of ascorbic acid was found to be more potent. The Soxhlet extraction method employed in this study proved to be a reliable and efficient technique for obtaining Gymnema sylvestre extracts with desirable antioxidant activity. The comparative analysis of the different extracts provides valuable information for researchers and industries seeking to harness the potential of Gymnema sylvestre as a natural antioxidant source. It is important to note that this study has certain limitations, including the focus on antioxidant activity and yield, and the specific extraction method employed. Further research is warranted to explore other bioactive components, investigate additional extraction methods, and evaluate the potential health benefits of Gymnema sylvestre extracts.

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Conflict of interest

The authors declare that there is no conflict of interest regarding the publication of this research paper.

References


