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# EVALUATION OF ANTIOXIDANT AND ANTI-CHOLESTEROL ACTIVITY OF ETHANOLIC LEAF EXTRACT OF GYMNEMA SYLVESTRE – AN IN VITRO STUDY

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***ABSTRACT--****Gymnema sylvestre, which is extensively used for serving as traditional remedy for various ailments like diabetes and malaria, belongs to the family Asclepiadaceae and genus gymnema. Recent research has proved that Gymnema can be used as a supplement in the treatment of both type 1 and type 2 diabetes mellitus. Since the herb has potential anti diabetic property research can be done to evaluate its anti-cholesterol and antioxidant potential.Antioxidant and anticholesterol potential of ethanolic leaf extract of the Gymnema sylvestre plant was investigated.The Gymnema sylvestre leaf extract showed a good free radical scavenging activity. The antioxidant potential is comparable to that of VIT C. High cholesterol levels in blood is one of the causes of deadly cardiovascular disease. The result exhibited is the significant anti-cholesterol property of plant extract.From this study, it can be concluded that the Gymnema sylvestre leaf extract possess antioxidant and anti-cholesterol activity. Further in vivo studies can be done so that the extract can be made into a drug.*

***Keywords—****antioxidant,anti-cholesterol activity ethanolic leaf extract Gymnema sylvestre – An in vitro study*

## I. INTRODUCTION

Gymnema sylvestre, which is extensively used for serving as traditional remedy for various ailments like diabetes and malaria[1,2], belongs to the family Asclepiadaceae and genus gymnema. It also means, “destroyer of sugar” [3]. This plant species is universally known as “periploca of the woods”. It has the ability to serve as a laxative, diuretic, and a cough suppressant. The chief function of Gymnema sylvestre is that, it improves cholesterol and triglycerides levels, which reduces the risk of heart diseases. It also plays a prominent role in fat absorption and lipid levels. Also, Gymnema is a supplement that is being used as a complementary treatment for both type 1 and type 2 diabetes.

Antioxidants are substances that inhibit oxidation [4]. Vitamin C & E, selenium and carotenoids such as beta-carotene, lycopene, lutein and zea xanthin are all examples of antioxidants. Antioxidants can be obtained from

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natural plant-based sources or artificial sources. Endogenous antioxidants are a variety of antioxidants that are products of our body's metabolism, which may be enzymatic or non-enzymatic. Dietary antioxidants are another type of antioxidants that include ascorbic acid, β- carotene, selenium, which contributes to the total antioxidant capacity of cells and plasma. Hence, antioxidant activity is defined as the ability of certain compounds to delay and prevent oxidation of substrates such as DNA and lipid materials. On the contrary, anti- cholesterol substances are those that tend to reduce the level of cholesterol in the blood. Most anti- cholesterol drugs, otherwise known as statins, suppress the production of cholesterol in liver, our richest source of cholesterol. The most common anti-cholesterol drugs are lovastatin, atorvastatin, pravastatin, etc.

Gymnema sylvestre plant is not very well known for its anti-cholesterol and antioxidant activity. Hyperlipidaemia, commonly known as high cholesterol is a familiar inherited disorder, occurring in about one out of 200 persons in a population. One specific type of hyperlipidaemia is hypercholesterolemia, which is mainly caused due to high content of LDL cholesterol in the patient's blood [5]. Hyperlipidaemia can also be one of the main root effects of cardiovascular diseases [6]. Hydroalcoholic extract of Gymnema sylvestre leaves results in significant decrease of total serum cholesterol, triglycerides, low density lipoproteins (LDL), very low-density lipoproteins and which hence gives an outcome of increase in high density lipoproteins (HDL). [7,8]The significant antihyperlipidemic activity of hydroalcoholic extract of Gymnema sylvestre leaves may be due to the presence of acidic compounds, flavonoids, phenols, saponins and tannis (Phenolic compounds). [9,10]

The principle aim of this in-vitro study is to evaluate the anti-cholesterol and antioxidant activity of the extract of Gymnema sylvestre plant. Since the herb has potential anti-diabetic property, research can be done to evaluate its anti-cholesterol & antioxidant potential.

## II. MATERIALS & METHODS:

This research was carried out by preparing an ethanolic leaf extract of the Gymnema sylvestre plant. The leaves of this plant were cleaned and crushed to prepare the ethanolic leaf extract by carrying out the solvent extraction method.

1) Antioxidant Activity-

***i) DPPH free radical scavenging activity of G. sylvestre:***

DPPH solution (1.0 ml) was added to 1.0 ml of G. sylvestre extract at different concentrations (100, 200, 300,400 and 500µg/ml). The mixture was kept at room temperature for 50 minutes and the activity was measured at 517nm. Ascorbic acid at the same concentrations was used as standard. The capability to scavenge the DPPH radical was calculated and expressed in percentage (%) using following formula:

$$\text{DPPH radical scavenged \%} = \frac{\text{Control OD-Sample OD}}{\text{Control OD}} \times 100$$

***ii) Nitric oxide radical scavenging activity of G. sylvestre extract:***

In the total volume of 3ml reaction mixture, 2ml of sodium nitroprusside, 500µl of phosphate buffered saline (PBS) were mixed with 500µl of different concentrations (100, 200, 300,400 and 500µg/ml) of extracts of G.

sylvestre and incubated for 1 hour 30 minutes at 25°C. Then, 500µl of reaction mixture containing nitrite was mixed with 1 ml of sulfanilic acid and allowed to stand for 5 minutes for completing diazotization. Then, 1 ml of naphthyl ethylene diamine dihydrochloride was added, mixed and allowed to stand for 30 minutes at 25°C. A pink coloured chromophore is formed in diffused light. Ascorbic acid at the same concentrations was used as standard. The activity was measured at 550 nm and the results were expressed in percentage (%) using following formula:

$$\text{No radical scavenged \%} = \frac{\text{Control OD} - \text{Sample OD}}{\text{Control OD}} \times 100$$

***2) In-vitro Anti-cholesterol Activity of G. sylvestre Extract:***

Cholesterol was dissolved in chloroform at a concentration of 2.5 mg mL/ml. Ten microliter of the G. sylvestre extract was pipetted into micro titre plate followed by the addition of 2000 µL of R1 reagent and 10 µL of cholesterol as sample. Twenty microliter of distilled water and 2000 µL of R1 reagent were used as blank. Negative control comprised of 20 µL cholesterol and 2ml R1; standard comprised of 20 µL simvastatin and 2000 mL R1 reagent. The contents were incubated between 0-30 min at room temperature and the absorbance was read at 500 nm in a UV-Vis spectrophotometer against reagent blank. Anti-cholesterol assay of the extract was calculated using the following equation:

$$\text{Inhibition (\%)} = \frac{\text{Negative control} - \text{sample}}{\text{Negative control}} \times 100$$

***Statistical analysis:***

The data was subjected to statistical analysis using an one-way analysis of variance (ANOVA) & Duncan's multiple range test to assess the significance of individual variations between the groups. In Duncan's test, significance was considered at level of $p<0.05$.

## III. RESULTS AND DISCUSSION

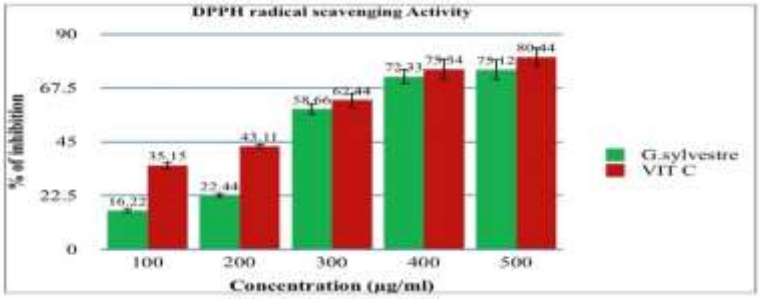


**Figure 1:** Dpph readial seavening activity

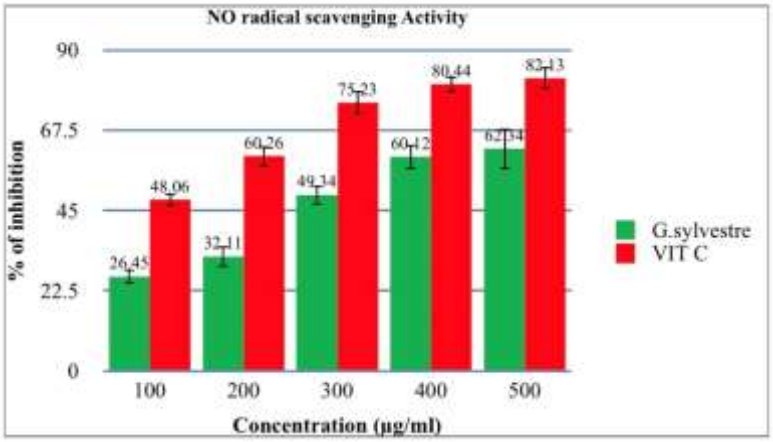


**Figure 2:** No radical seavening activity

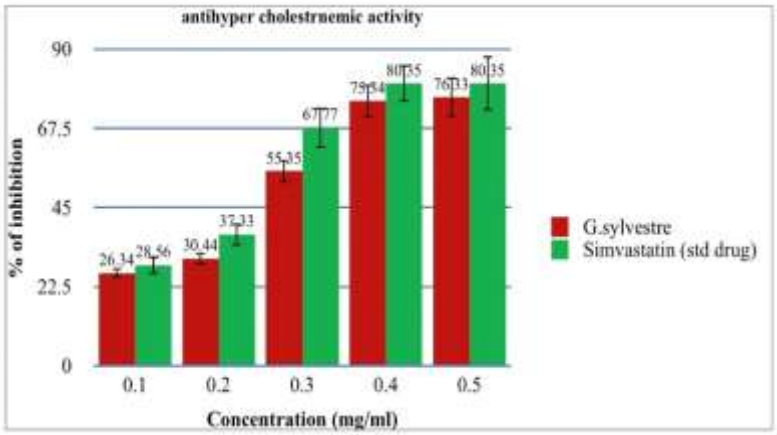


**Figure 3:** antihyper cholestrnemic activity

Each bar represents mean ± SEM of 3 independent observations. Significance at $p < 0.05$

In-vitro inhibition of DPPH & nitric oxide radical is a measure of antioxidant activity. The Gymnema sylvestre leaf extract showed a good free radical scavenging activity. The antioxidant potential is comparable to that of VIT C. Antioxidant effects of vitamin C have been demonstrated in many experiments in vitro. Antioxidants work by giving electrons to free radicals; the extra electron stabilizes them, preventing them from causing further damage to the cells in the body. In addition to neutralizing free radicals with its antioxidant properties, vitamin C also plays a role in controlling infections . The gymnema sylvestre plant exhibits antioxidant activity by inhibiting the DPPH, known as 2,2-diphenyl-1-picrylhydrazyl, which is a chemical compound used to test the antioxidant capacity of a substance and is also used to test the ability of compounds to act as free radical scavengers . It additionally has the reducing power ability due to the presence of Flavonoids, Phenols, tannis (Phenolic compounds) and Triterpenoids found in the preliminary phytochemical screening .

The result exhibited is the significant anti-cholesterol property of plant extract. Statin drugs are generally used as standard drugs to compare the anti-cholesterol property with other extracts. Some examples of statin drugs include atorvastatin, Fluvastatin, lovastatin, pravastatin, simvastatin, rosuvastatin, etc. Statins also are effective in

reducing triglyceride levels in patients with hypertriglyceridemia . Statins lower LDL levels by inhibiting HMG-CoA reductase activity leading to decreases in hepatic cholesterol content resulting in an up-regulation of hepatic LDL receptors, which increases the clearance of LDL. High cholesterol levels in blood is one of the causes of deadly cardiovascular disease .

## IV. CONCLUSION

In this study, it can be concluded that the Gymnema sylvestre leaf extract possess antioxidant and anti-cholesterol activity. Further in vivo studies can be done so that the extract can be made into a drug.

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