

Evaluation Of Phytoconstituents And Antioxidant Properties Of Polyherbal Formulations As Potential Antidiabetic Agents.

Yashwant Singh¹, Shilpi Sonker²

¹Research Scholar, Lords university Alwar Rajasthan

²Associate Professor, Lords university Alwar Rajasthan

***Corresponding Author:**

Yashwant Singh

Email:ID: yashwantobcms@gmail.com

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ABSTRACT

The aim of the study to investigate the phytochemical and antioxidant activity of polyherbal formulation in antidiabetic potential. Ethanolic extract of the polyherbal formulation was prepared by the Soxhlation process, Phytochemical screening, Total Phenolic Content and Total Flavonoid Content of the extracts were estimated using standard methods. Extracts were analysed for its antioxidant potential using DPPH, and Reducing power methods to assay their free scavenging activity. Results shows that plant extract (Tinospora Cordifolia and Epipremnum aureum) contains Flavonoids 0.65 ± 0.03 and 55.2 ± 0.05 mg/g, Total phenol 5.2 ± 0.29 mg/g and 8.3 ± 0.19 mg/g, preliminary phytochemical screening revealed the presence of tannins, alkaloids, flavonoids, glycosides. Antioxidant study for DPPH and reducing power of plant extract were 37.1 ± 0.31 , 47 ± 0.28 and 88.99 ± 0.22 , 43.8 ± 0.18 , at $100 \mu\text{g/ml}$ respectively. these findings may provide efficient, supportive data on the use of medicinal herbs for treatment of numerous health ailments associated with the accumulation of harmful free radicals and reactive oxygen species. It was concluded that flavonoids component were present in all solvent extracts of T. cordifolia and Epipremnum aureum. TFC was high in methanolic leaves extract of T. cordifolia and Epipremnum aureum. Furthermore, these results of plant sources were found to be highly significant. study has revealed that the ethanol extract of T cordifolia and Epipremnum aureum contains substantial amount of phenolics and thus, can be inferred that these phenolics are responsible for its marked antioxidant activity

Keywords: Tinospora Cordifolia and Epipremnum aureum, phytochemical analysis and antioxidant.

INTRODUCTION

Plants have been used as sources of essential medicine since time immemorial as isolated active components or classical combinations. Nowadays, 75 80 percent of the population in most places in the world and in developing countries especially depends on the herbal remedies as the main form of healthcare.^{1,2} As the popularity of natural healing methods increases, more and more plant-based products are becoming the means of control and treatment of different health issues. It is quite remarkable that plants contain natural antioxidants which can assist in abolishing the dangerous free radicals and reactive oxygen species (ROS). Such antioxidants defend the cells against oxidative stress hence playing a crucial role in preventing or limiting the cases of chronic diseases and facilitating healthy body in general.^{3,4}

Free radicals are at least molecules or atoms with one or more unpaired electrons, which occur naturally in the body, in the process of metabolic activity such as food breakdown. Although they are influential in the activation of the genes, receptors and transmission of signals, too many free radicals may be detrimental to the living cells. It is an imbalance called oxidative stress, which leads to the occurrence and growth of diverse chronic diseases, such as cancer, diabetes, and cardiovascular diseases. In healthy circumstances, there is a balance put in place between the production and the removal of free radicals within the body. When such a balance is impaired however, there is cellular damage and thus the necessity of antioxidants to counter excess free radicals to preserve health.^{5,6} When the pro-oxidants and antioxidants are not in equilibrium, oxidative stress takes place. The excessive amounts of reactive oxygen species (ROS) may cause the violation of the normal cell functioning, which results in a low activity of the cell and aging of cells and the development of different illnesses. This cellular damage highlights the need to have oxidative balance. Various bioactivities are seen to exist in plant extracts which have given rise to the significance of the extracts as a source of therapeutic compounds. They have antioxidant effects which aid in the reduction of these oxidative stresses and similar useful properties by having anti-allergic, anti-inflammatory,

antimicrobial, antifungal, antiviral, antidiabetic, and anticancer activities thus ensuring that people enjoy good health and are able to control various chronic illnesses.^{7,8} Antioxidants are essential as they break the oxidative chain reaction in the body thus offering protection against oxidative damages. They do this through donation or acceptance of their hydrogen atoms which form more stable free radicals hence producing less reactive intermediates. Majority of the antioxidants are phenolic compounds

comprising various substances like antioxidant vitamins and minerals as well as phytochemicals. Important ones are flavonoids, catechins, carotenoids, beta-carotene, lycopene, diterpenes and their derivatives. Its compounds act in many ways which include; Scavenging reactive oxygen species (ROS), chelating metal ions, neutralizing free radicals, inhibiting oxidative enzymes, converting hydroperoxides into non-radical, absorption of UV radiation and singlet oxygen quenching.^{9,10}

Antioxidant compounds are only effective when a number of factors are considered such as their chemical structure, their concentration, temperature, solubility, their interaction with oxidizable substrates and existence of synergistic or antagonizing compounds. The activity of the antioxidant is also affected by the physical state and stability of the antioxidant. Antioxidants work in counteracting the oxidant-antioxidant balance in the body by destroying pro-oxidant molecules and decreasing the concentration of ROS. This balance has the vital role in cellular homeostasis and in stopping the oxidative damage related to choice of cell aging and a variety of chronic diseases other than cancer, diabetes, and cardiovascular diseases.^{11,12} This makes antioxidant use effective in mitigating the level of reactive oxygen species (ROS) and free radicals resulting in the minimization of the chances of lipids peroxidation, oxidative stress, protein oxidation, and mutation of DNA. The majority of such protective antioxidants are found in plants foods. Although synthetic antioxidants are capable of stopping bad redox reactions, they are usually more expensive whereas less accessible and can have an adverse effect. However, in comparison, natural antioxidants have been highly attributed because they are safe, cheap and always available in vast varieties of plants. The natural compounds also have a potential of replacing the synthetic antioxidants, in protecting the body against oxidative damages and the consequent chronic illnesses. They have extensive potential in treatment, and their low side effects help in enhancing health and preventing degenerative diseases that are associated with oxidative stress.^{13,14} Production of reactive oxygen species (ROS) in the body is a normal by-product of normal metabolic processes in the body. Superoxide radicals, hydrogen peroxide, hydroxyl radicals and singlet oxygen are some of the common ROS. Although they are critical components in cellular signaling response and cellular defense, excess accumulation of ROS involves oxidative stress. When ROS production becomes excessive in relation to the body antioxidant protection, proteins, lipids and DNA may be damaged and this can contribute to the age-related process and other chronic illnesses. The regulation of the ROS levels is of great importance as regards to cellular health and avoiding harmful effects of oxidation.^{15,16} Oxidative stress contributes significantly in causing and worsening many diseases. It happens when the formation of reactive oxygen species (ROS) exceeds the capacity of the body to mediate their presence through the use of antioxidants. This disorder is also related to a diversity of health issues such as cancer, coronary heart diseases, Alzheimer disease, neurological disorders, atherosclerosis, cataracts, inflammation, as well as diabetes mellitus. Oxidative stress produces cell damage by affecting DNA, proteins and lipids in a number of ways that lead to the development and exacerbation of these chronic illnesses.^{17,18} Autopsy and brain biopsies have shown that there was high oxidative stress loads in the brain of people suffering with neurodegenerative diseases. These are protein carbonyls, trans-4-hydroxy-2-nonenal (4-HNE) and malondialdehyde (MDA) that is indicative of both protein and lipid oxidative damage. These biomarkers depict the pernicious consequences of reactive oxygen species (ROS) and are linked with the development of diseases such as Alzheimer and Parkinson. This gives a nod to their presence of close association between oxidative stress and the pathogenesis of neurodegenerative diseases, which necessitates antioxidants-based treatment approaches.^{19,20} Uncontrolled free radicals production is closely associated with the onset and consequences of diabetes because oxidative stress hinders the capability of insulin to perform and destroys pancreatic cells. The antihyperglycemic effects of many medicinal plants are mostly characterized by their antioxidant activity due to neutralization of free radicals and suppression of oxidative stress. Such herbs are compounds that sustain blood glucose levels by defending the insulin-releasing cells, and enhancing insulin sensitivity. This makes the natural plant-derived antioxidants a potential lead drug in the control of diabetes and a reduction of its long-term effects on health.^{21,22} Most medicinal plants have the healing qualities which scientists attribute to high contents of phytochemicals which display antioxidant properties. Polyphenols, flavonoids and phenolic acids are some of the compounds that are important in protecting the cells under oxidative stress because they counter the toxic free radicals. Such antioxidants are used to lessen inflammation, and to provide support to the body in general and to prevent cell destruction. Their occurrence in the medicinal plants resulted in a range of therapeutic properties such as antidiabetic, anti-inflammatory properties as well as anticancer and thus they are useful in both traditional and the modern medicines in the prevention and treatment of diseases.^{23,24} Consequently, the phytochemicals have a wide range of health benefits through the counter of oxidative stress and its sequel. They aid in the prevention of oxidation of low-density lipoproteins (LDL), decrease inflammation, prevent the occurrence of cancer, and decrease the level of blood sugar and also protect the cell against the oxidative damage. These compounds help the body in defence and prevention of diseases through neutralisation of reactive oxygen species (ROS). Owing to their antioxidant and therapeutic potential, phytochemicals play an important role in the treatment and the control of chronic diseases and the general wellbeing of individuals using natural or plant-based strategies.^{25,26} The evaluation of the ability of a plant to resist free radicals is currently performed with the help of biochemical assays which are quite the most effective

and reliable. These tend to assist in quantifying the antioxidant activity via examining the interaction of plant extracts with any reactive species. A single antioxidant may tend to take a different behaviour in different environments of testing and hence necessitating not one, but several assays to be carried out in order to get the correct results. These are simple assays and most normal are DPPH assay, ABTS, FRAP assay and nitric oxide scavenge. With the extensive usage of a range of assays, a more detailed picture of the antioxidant capacity is obtained, and the mechanism of activity of the certain bioactive compounds included in the researched plant extracts can be revealed.^{27,28}

2. METHODOLOGY

2.1 Collection of material

Tinospora Cordifolia and *Epipremnum aureum* Leaves were collected. Plant materials were dried under shade and powdered coarsely before extraction.

2.2. Pre –Extraction Process

2.2.1 Collection and drying

Leaves of *Tinospora Cordifolia* and *Epipremnum aureum* were collected and washed and dried in shaded area.

2.2.2 Extraction of plant material

The powdered plant material was processed using Soxhlet instruments with various solvents. The crude extract was dried over a bath of water. Soxhlet extraction is only required if the target compound has minimal solubility in a solvent and impurity in that solvent is insoluble. If the compound you want is very soluble in a solvent to extract the compound from the insoluble substance with a simple filtration. The benefit of this approach, rather than transfer several sections of warm solvent to the sample, is that it removes only one batch of solvent. This process cannot be used with thermolabile compounds as intense heating allows the compound to degrade.

2.2.3 Preparation of Petroleum ether extract: A dry shade was packed well into a soxhlet apparatus (200 gm) and extracted with petroleum ether (60-80°C) before the extraction, which was confirmed by the colour of the syphoned oil, was finished. The extract was purified during warming and the extract was extracted in vacuum to fully isolate the solvent and dried in a dessicator subsequently. The extract was measured and the percentage yield for air-dried content determined.

2.2.4 Preparation of ethyl acetate extract: The marc was completely dry during the pet ether procedure. The ground powder plant material (200 gm) was well packaged in a Soxhlet appliance and extracted with ethyl acetate (100°C) before the extraction was finalized and checked by a syphoned fluid colour. The extract was purified during warming and the extract was extracted in vacuum to fully isolate the solvent and dried in a desiccator subsequently. The extract was measured and the percentage yield for air-dried content determined.

2.2.5 Preparation of 70% methanol extract: Each dried and weighted sample was collected and purified in 70% methanol for 24 hours. The whole procedure has been replicated three times to ensure full extraction. The extract was purified during warming and the extract was extracted in vacuum to fully isolate the solvent and dried in a desiccator subsequently. The extract was measured and the percentage yield for air-dried content determined. **2.2.6 Phytochemical characterization**

Extract phytochemical screening was carried out using normal methods. Extracts have been preliminarily phytochemically tested to classify different plant substances in them: alkaloids, terpenoids, glycosides, hormones, triterpenoids, flavonoids, carbohydrates, saponins and tannins.

2.2.6.1 Tests for carbohydrates

A. Molisch test: To 1ml of test + 2-3 drops of α - naphthol + Conc. sulphuric acid, appearance of purple ring.

B. Fehling's test: To 1 ml of test sample+ Fehling 's solution A and B + heat, brick red precipitate.

C. Benedict's test: To 1 ml of test sample, equal quantity of Benedict 's reagent was added and boiled. Red colour precipitate confirmed the presence of carbohydrates.

2.2.6.2 Test for alkaloids

All the extracts were first treated with dil. hydrochloric acid separately and then filtered. The filtrate of all the extracts was subjected to following tests:

A. Mayer's test: 1 ml of the filtrate + 1ml of Mayer's reagent, cream precipitate

B. Hager's test: 1 ml of the filtrate + 1mL of Hager's reagent, yellow precipitate

C. Wagner's test: 1ml of the filtrate + 1ml of Wagner 's reagent, reddish brown precipitate

2.2.6.3 Tests for terpenoids

A. Salkowski test: Chloroform solution of test sample was treated with equal amount of conc. sulphuric acid. The presence of steroid components in the test sample was observed by the appearance of red color in chloroform layer and green fluorescence in acid layer.

B. Libermann - burchard test: To 2ml of test sample, chloroform was added before the addition of 2-3 drops of acetic anhydride and conc. sulphuric acid. The test solution was observed for colour change from red to blue and then finally to bluish green which confirmed the presence of steroids in the test extracts.

2.2.6.4 Tests for flavonoids

A. Lead acetate test: Lead acetate solution was added to the extract. Flavonoid was confirmed on the basis of formation of yellow precipitate.

B. Alkaline reagent test: 1mL of test sample was dissolved in dilute sodium hydroxide solution that resulted in formation of yellow colour precipitate.

2.2.6.5 Tests for tannins and phenolic compounds

A. 5% FeCl₃ solution: Few drops of 5% FeCl₃ solution was added to the small amount of extract that leads to the formation of deep blue-black colour complex.

B. 10% lead acetate solution: To 2 ml of extract, few drops of 10% lead acetate solution were added, white precipitate was formed.

C. Gelatin test: After dissolving some quantity of extract in distilled water, 2 ml of 1% gelatin solution containing 10% NaCl was added which lead to the formation of white precipitate indicating the presence of phenolic compounds.

2.2.6.6 Tests for saponins

A. Froth test: 1 ml of test sample + 20 ml of distilled water + shaken for 15min, Formation of persistent foam.

2.2.6.7 Test for proteins and amino acids

A. Ninhydrin test: 3 ml of the test solution + 3 drops of 5% ninhydrin + heat for 10min + observe colour change

B. Biuret test: Test sample was treated with same volume of 1% copper sulphate and 4% sodium hydroxide solution and appearance of violet or pink colour was observed.

C. Million's test: 3 ml of extract was mixed with 5 ml of Million 's reagent. White precipitate formed which on heating turned to brick red, indicated the presence of proteins.

2.2.6.8 Tests for glycosides:

A. Borntrager's test: Dil. H₂SO₄ was added to 3 ml of extract solution and boiled for 5 min. The solution was filtered and cooled. Same amount of chloroform was added while shaking the mixture. Ammonia was added to the organic solvent layer. Change in color of ammonical layer to pinkish red colour confirmed the presence of anthraquinone type glycosides.

B. Legal test: 1 ml of sodium nitroprusside was added in 1 ml of pyridine solution containing test sample and colour change was observed.

C. Keller killani test: To 2 ml of extract, 3 ml of glacial acetic acid and 1 drop of 5% ferric chloride were added. This solution was carefully transferred to the surface of 2 ml concentrated H₂SO₄ and the observation was noted down.

2.2.6.9 Tests for fats and oils

A. Spot test: One drop of extract was placed on filter paper and solvent was allowed to evaporate. An oily stain on filter paper indicated the presence of fixed oil.

2.3 Antioxidant activity of *Tinospora Cordifolia* and *Epipremnum aureum*

2.3.1. Total phenolic content

Total soluble phenolic content was estimated by Folin-Ciocalteu reagent method using gallic acid as a standard phenolic compound. 1ml of stock solutions of extracts was prepared (1g/ml) from which different aliquots were pipetted out into test tubes. The volume was made up to 3 ml with distilled water to which freshly prepared Folin-Ciocalteu reagent was added. After 3 min, 2 ml of 20% sodium carbonate solution was added to each tube and mixed thoroughly. The tubes were placed in boiling water for one minute, cooled and the absorbance was measured at 650 nm and 765nm in a spectrophotometer against a reagent blank. The concentrations of the total phenolic compounds in the extracts were obtained by extrapolating the absorbance of gallic acid on standard gallic acid graph. The experiment was repeated thrice and concentration of total phenols was expressed as mg /g of dry extract.

2.3.2 Estimation of total flavonoid content: -

The total soluble flavonoid content was estimated by aluminum chloride colorimetric method for both aqueous and solvent extracts. 0.5ml of stock solution (1g/ml) of the extract, 1.5 ml methanol, 0.1ml potassium acetate (1M) was added to reaction test tubes and volume was made up to 5 ml with distilled water. After incubation at room temperature for 30 min, the absorbance of the reaction mixture was measured at 415 nm and 420nm. Total flavonoid content was calculated by extrapolating the absorbance of reaction mixture on standard curve of rutin. The experiment was repeated thrice and the total flavonoid content was expressed as equivalent to rutin in mg/ g of the extracts.

2.3.3 DPPH radical scavenging activity

The free radical scavenging activity of the leaf extracts was assayed using a stable free radical, 1, 1- diphenyl-2-picryl hydrazyl (DPPH). The DPPH scavenging assay employed in the present study was a modification of the procedure of Moon & Terao (1998). 0.1 ml of test sample at different concentration (0.1 - 0.9 mg/ml) was mixed with 0.9 ml of Tris-HCl buffer (pH 7.4); then 1 ml of DPPH (500 µM in ethanol) was added. The mixture was shaken vigorously and left to stand for 30 min. The absorbance of the resulting solution was measured at 517 nm in a spectrophotometer and compared with that of BHA. The experiment was repeated thrice. The capability to scavenging the DPPH radical was calculated by using the following formula.

$$\text{DPPH scavenging effect (\% inhibition)} = \{(A_0 - A_1)/A_0\} * 100\}$$

Where, A₀ is the absorbance of the control reaction,

and A₁ is the absorbance in presence of all of the extract samples and reference.

All the tests were performed in triplicates and the results were averaged.

2.3.4 Reducing power

The determination of reducing power was performed. Various extracts (0.1 - 0.9 mg/ml) were mixed with phosphate buffer (500 µl, 20 mM, pH 6.6) and 1% potassium ferricyanide (500 µl), and incubated at 50°C for 20min; 500µl of 10% trichloro acetic acid were added, and the mixture was centrifuged at 2500 rpm for 10 min. The supernatant was mixed with distilled water (1.5 ml) and 0.1% ferric chloride (300 µl) and the absorbance was read at 700nm. The experiment was repeated thrice. Increase in the absorbance of the reaction mixture indicated increase in the reducing power.

3. RESULT AND DISCUSSION

3.1 *Tinospora Cordifolia*

3.1.1 Percentage yield

The percentage yield in methanol is high.

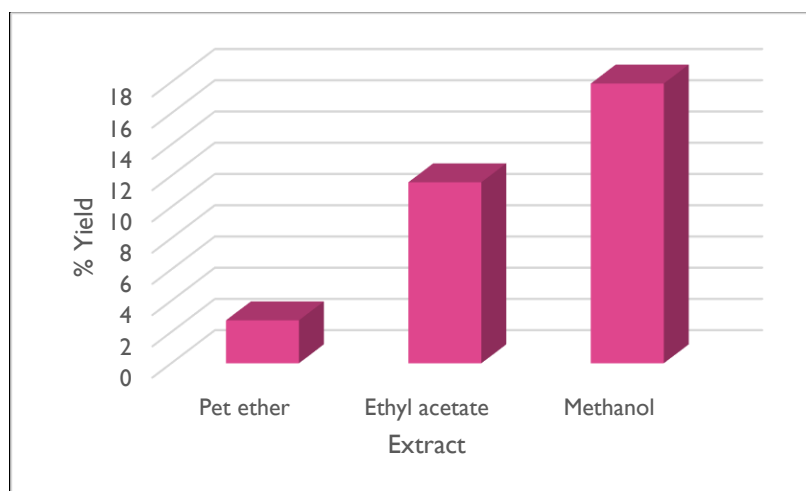


Figure 1. Yield of crude extracts of *Tinospora Cordifolia* leave extract

3.1.2 Preliminary observations

Three solvents were used to prepare the crude extracts. All the extracts were semi-solid in nature, and as solvent polarity rose, colour varied from light brown to dark brown. They smelled pungent and tasted salty.

Extract	Colour	Taste	Odor
Pet ether	Pale Yellow	Bitter	Unpleasant
Ethyl acetate	Light brown	Bitter	Unpleasant
Methanol	Light brown	bitter	Unpleasant

Table 1. Physical parameters of extracts of *Tinospora Cordifolia* leaves

S.no	Solvents	Pet ether extract	Ethyl acetate extract	Methanolic extract
1	Pet ether	Soluble	Insoluble	Insoluble
2	Ethyl acetate	Insoluble	Soluble	Soluble
3	Methanol	Insoluble	Soluble	Soluble
4	DDW	Insoluble	Insoluble	Soluble
5	DMSO	Insoluble	Insoluble	Soluble

Table 2. Solubility of extracts in different solvents

3.1.3 Phytochemical Screening

Test	Pet ether	Ethyl acetate	Methanol
Carbohydrate	-	-	+
Alkaloids	+	+	+
Terpenoids & Steroids	-	-	-
Flavonoids	+	+	+
Tannin & Phenolics	-	+	+
Saponins	-	+	+
Protein & amino acids	-	-	+
Glycosides	-	-	+
Fats	-	-	-

Table 3. Phytochemical evaluation of *Tinospora Cordifolia* leaves

3.1.4 Antioxidant activity of *Tinospora Cordifolia*

3.1.4.1 Total phenolic and flavonoid content

Results obtained in the present study revealed that the level of polyphenols in the methanol extract was 5.2 ± 0.29 mg/g which was higher when compared to methanol, chloroform, hexane and aqueous extracts of *T. cordifolia*. Ethanol extract of the leaves had a flavonoid content of 0.65 ± 0.03 mg/g. The flavonoid content of other extracts tested was lower than the ethanol extract. Aqueous extract had the least polyphenol and flavonoid content.

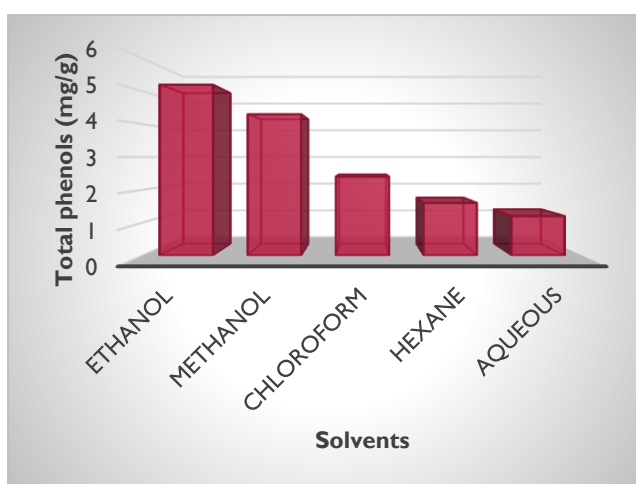


Figure 2. Phenol content

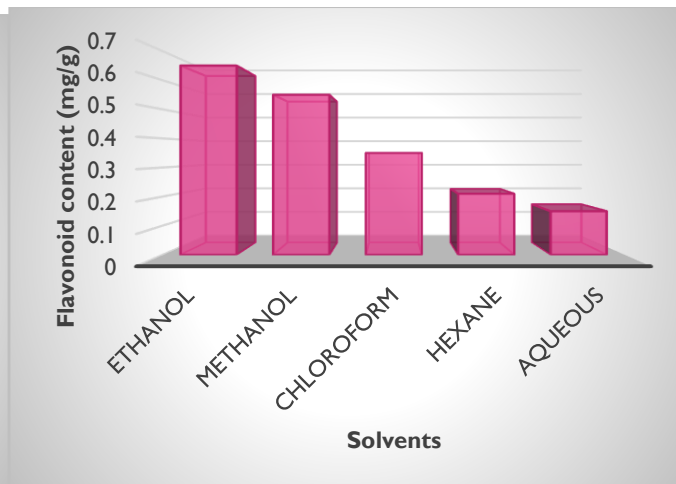


Figure 3. Flavonoid content

3.1.4.2 DPPH radical scavenging activity

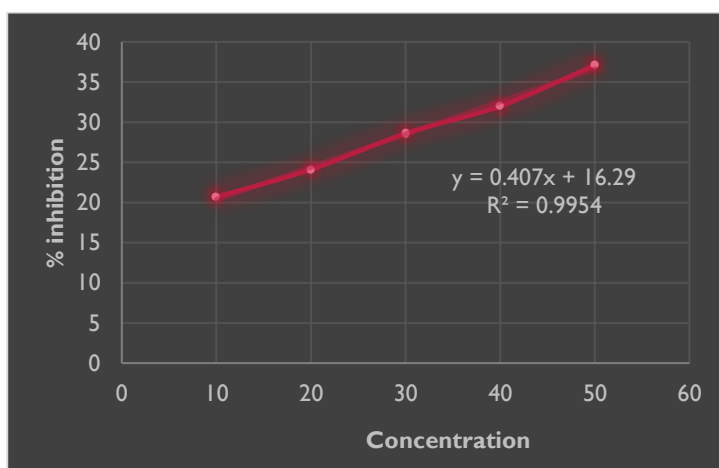


Figure 4. DPPH assay activity of methanol extracts of *Tinospora cordifolia*

3.1.4.3 Reducing power

The reducing power of different solvent extracts using the potassium ferricyanide method. The result indicates that the reducing ability of the extracts increased with the concentration. Among all the extracts tested for their reducing abilities ethanol extract of *T. cordifolia* showed better reducing power as shown by the increasing optical density at 700 nm.

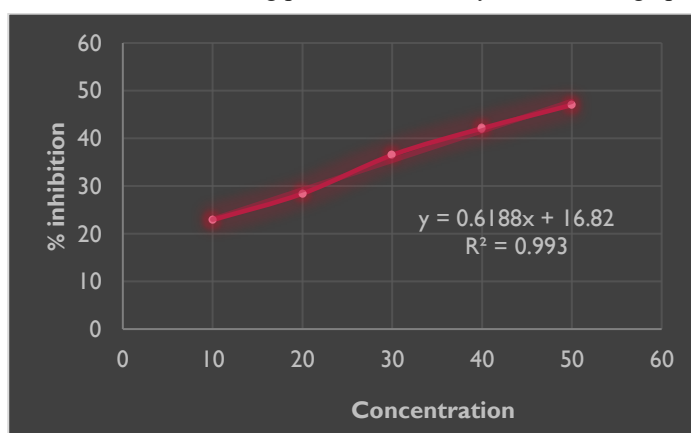


Figure 5. Reducing power of *Tinospora cordifolia* leaf extract

3.2 *Epipremnum aureum*

3.2.1 Percentage yield

The percentage yield in methanol is high.

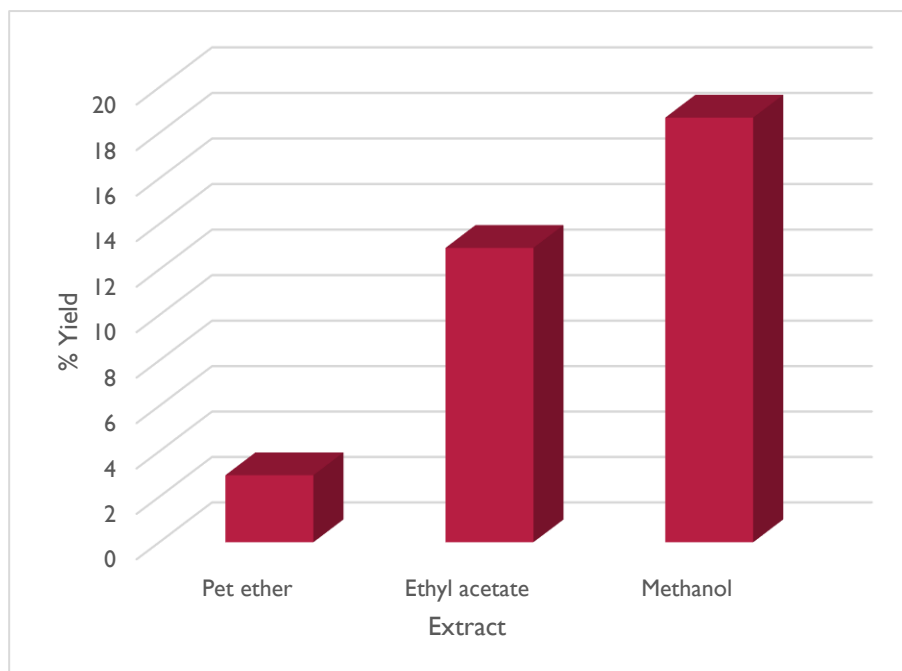


Figure 6. Yield of crude extracts of *Epipremnum aureum* leaf extract

3.2.2 Preliminary observations

Three solvents were used to prepare the crude extracts. All the extracts were semi-solid in nature, and as solvent polarity rose, colour varied from light brown to dark brown. They smelled pungent and tasted salty.

Extract	Colour	Taste	Odor
Pet ether	Pale Yellow	Bitter	Unpleasant
Ethyl acetate	Light brown	Bitter	Unpleasant
Methanol	Light brown	bitter	Unpleasant

Table 4. Physical parameters of extracts of *Epipremnum aureum* leaves

S.no	Solvents	Pet ether extract	Ethyl acetate extract	Methanolic extract
1	Pet ether	Soluble	Insoluble	Insoluble
2	Ethyl acetate	Insoluble	Soluble	Soluble
3	Methanol	Insoluble	Soluble	Soluble
4	DDW	Insoluble	Insoluble	Soluble
5	DMSO	Insoluble	Insoluble	Soluble

Table 5. Solubility of extracts in different solvents

3.2.3 Phytochemical Screening

Test	Pet ether	Ethyl acetate	Methanol
Carbohydrate	-	-	+
Alkaloids	+	+	+
Terpenoids & Steroids	-	-	-
Flavonoids	+	+	+
Tannin & Phenolics	-	-	+
Saponins	-	-	+
Protein & amino acids	+	+	+
Glycosides	+	+	+
Fats	-	-	-

Table 6. Phytochemical evaluation of *Epipremnum aureum* leaves

3.3 Antioxidant activity of *Epipremnum aureum*

3.3.1 Total phenolic and flavonoid content

Results obtained in the present study revealed that the level of polyphenols in the ethanol extract was 8.3 ± 0.19 mg/g which was higher when compared to methanol, chloroform, hexane and aqueous extracts of *Epipremnum aureum*. Ethanol extract of the leaves had a flavonoid content of 55.2 ± 0.05 mg/g. The flavonoid content of other extracts tested was lower than the ethanol extract. Aqueous extract had the least polyphenol and flavonoid content.

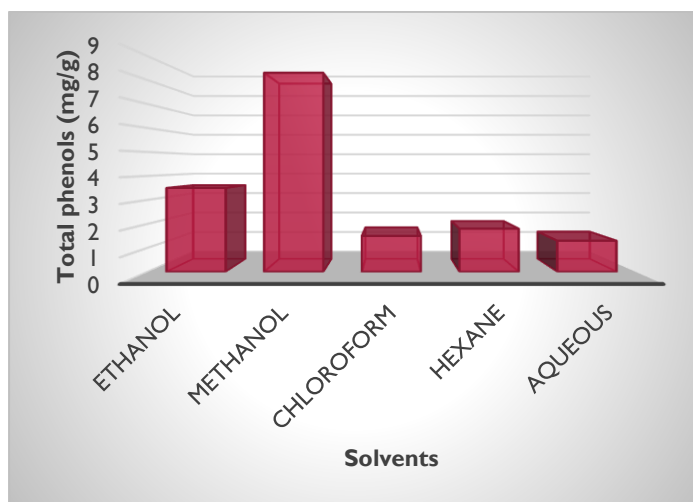


Figure 7. Phenol content

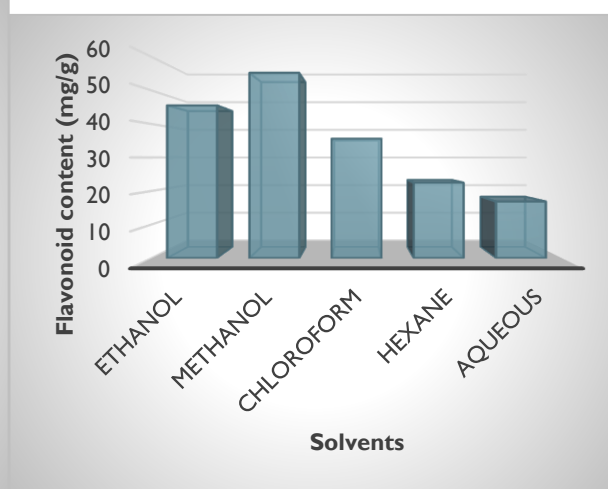


Figure 8. Flavonoid content

3.3.2 DPPH radical scavenging activity

The percentage of DPPH radical scavenging activity of methanolic extract of *Epipremnum aureum* Linn. Leaves show in figure. It showed that methanolic extract exhibited a maximum DPPH scavenging activity of 88.99 % at $100 \mu\text{g/ml}$.

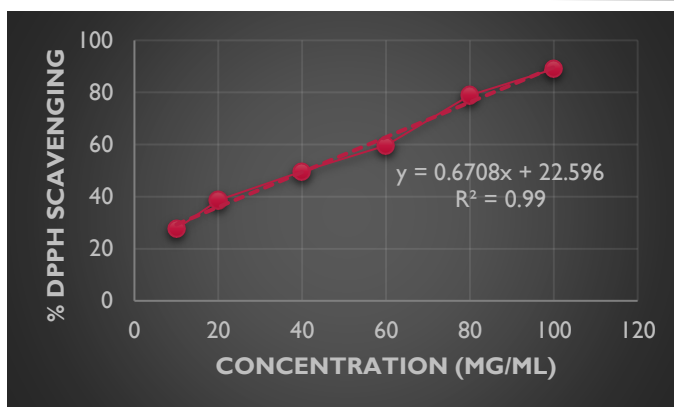


Figure 9. DPPH radical scavenging activity *Epipremnum aureum*

3.3.3 Reducing power

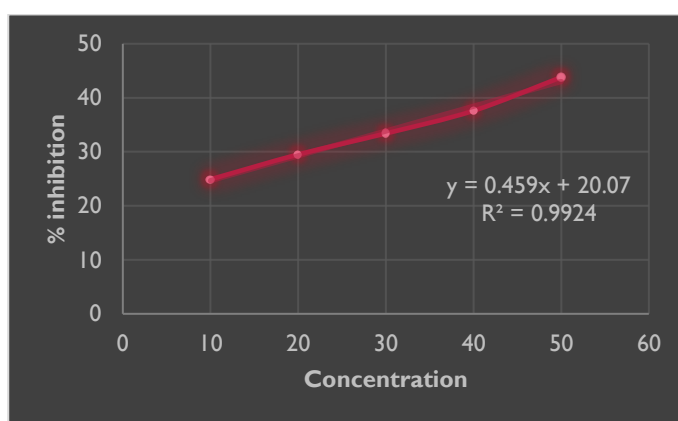


Figure 10. Reducing power ability of *Epipremnum aureum*

4. CONCLUSION

These results revealed that flavonoids component were present in all solvent extracts of *T. cordifolia* and *Epipremnum aureum*. TFC was high in methanolic leaves extract of *T. cordifolia* and *Epipremnum aureum*. Furthermore, these results of plant sources were found to be highly significant. Hence, there is more requirements to explore the applicability of these plant resources which are rich in phytochemicals/flavonoids and may have beneficial effect on health. This is observed that leaves and stem of the plant has antioxidant activity. Thus it can be use as natural antioxidants. The present study has revealed that the ethanol extract of *T cordifolia* and *Epipremnum aureum* contains substantial amount of phenolics and thus, can be inferred that these phenolics are responsible for its marked antioxidant activity. This is consistent with several reports that have shown close relationship between total phenolic contents and antioxidant activity of fruits, plants and vegetables. Therefore, *T cordifolia* and *Epipremnum aureum* leave extracts have considerable antioxidant properties and the consumption of this plant may play a role in preventing human diseases in which free radicals

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