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DPPH FREE RADICAL SCAVENGING ACTIVITY OF THUJA OCCIDENTALIS AND ARAUCARIA HETEROPHYLLA PLANTS OF METHANOLIC EXTRACT

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ABSTRACT

In terms of antioxidant activity, the hydroalcohol extract of the leaves and stem came in second place after the methanol extract. In the DPPH scavenging procedure, the extract obtained from methanol provided the highest percentage of scavenging activity, followed by the extract obtained from hydroalcoholic. Methanol extract performed significantly better than hydroalcohol extract did in the hydrogen peroxide assay.

Kew Words-: *Anioxidant, Thuja occidentalis, , Araucaria heterophylla*

INTRODUCTION

Medical plants serve as a key source of organic chemicals, which are used for physiological effects as well as medical purposes. Some species used primarily for decoration have tremendous value in the field of medicine, such as the separated components of Catharanthus roseus that are employed in cancer treatment. *Araucaria* is a member of the Araucariaceae family of plant families. *Araucaria* and *Agathis* are the only two genera within this small family, which has a total of 38 different species of tree. Christmas trees are typically made out of Araucaria heterophylla, which is a type of generic columnar tree. Investigations on the chemical and biological properties of the resin of this genus, which was rumored to exhibit gastroprotective activities , were carried out. The plant known as *Thuja occidentalis*, which is a member of the *Cupressaceae* family, is yet another option. Rheumatism, migraines, and uterine carcinomas are some of the conditions that can be helped by using this. Anticancer, antiviral,

anti-inflammatory, insecticidal, and diabetes-fighting properties have all been uncovered by it . The use of plants in the production of medicine is a practice that has been passed down through generations. Plants have served as a fertile breeding ground for medicinal compounds, which are now an essential component of the healthcare system . Because India is the largest producer of medicinal herbs, the country is sometimes referred to as the botanical garden of the globe, which is an apt description. Plants are capable of producing secondary metabolites, which have a wide variety of biological and pharmacological properties, such as anti-allergic, antibacterial, hypoglycemic, and anti-carcinogenic. Plants that are utilized as remedies include a wide array of elements that have the potential to treat a number of ailments, including those that are chronic and infectious. Over the course of the past few years, gas chromatography–mass spectrometry has established itself as the primary technological program for characterizing secondary metabolites across all plant groups. Natural materials have, in addition to their use in the treatment of human ailments, also been put to use in the treatment of a variety of diseases. Unani and Ayurveda are two of the most well-known medical practices in the world that make use of ingredients derived from natural sources. These methods, along with other forms of folklore from a variety of countries, continue to rescue a significant section of the world's population by making use of items derived from the natural world. The majority of well-known inhibitors are organic inhibitors, but in addition to having strong corrosion inhibition efficacy, these inhibitors are expensive, non-biodegradable, and hazardous for both life and the environment. As a result of the drawbacks of existing inhibitors, the researchers were motivated to look for an eco-friendly green inhibitor. The use of extracts derived from plants as corrosion inhibitors is becoming increasingly common in modern times.

MATERIALS AND METHODS

Collection of plants

Plants of ***Thuja occidentalis*** and ***Araucaria heterophylla*** of procured from Saharanpur, Uttar Pradesh.

- **Thuja occidentalis**

This species is a member of the ***Cupressaceae*** plant family. Rheumatism, migraines, and uterine carcinomas are some of the conditions that can be helped by using ***Thuja occidentalis***. Anticancer, antiviral, anti-inflammatory, insecticidal, and diabetes-fighting properties have all been discovered in it .

- ***Araucaria heterophylla***

Araucaria is a member of the Araucariaceae family of plants. There are just 38 different species of trees that belong to this tiny family, which include the genera agathis and araucaria. It is a common columnar tree that is decorated to seem like a Christmas tree . Investigations on the chemical and biological properties of the resin of

this genus, which was rumored to exhibit gastroprotective activities , were carried out.

Preparation of plant extract

The components made from the fresh plant parts were allowed to dry out and were then pulverized roughly. After defatting the plant parts powder, a soxhlet extractor was utilized in order to perform a sequence of consecutive methanolic extractions. A rotary vacuum evaporator was used to remove moisture from the methanolic extract as the pressure was lowered. Following extraction using a Soxhlet apparatus maintained at a temperature lower than 60 degrees Celsius, the extract was obtained. The Soxhlet extraction method was chosen for the process of plant extraction because it is simple to implement, requires a short amount of time, is economical due to the fact that only one sample is required for the entirety of the extraction, makes it simple to determine when the extraction process is complete, and presents a lower risk of contamination due to the fact that it is a closed system.

Materials

During the course of the experiment, many types of analysis, including weight loss, EIS, potentiodynamic polarization, and surface analysis, were performed using the metal sheet . To create a surface that was perfectly smooth, the metal sheet was first cut into squares measuring 2.5 cm2 by 2.5 cm2 before being cleaned using abrasive papers or Emery sheets of grades 320, 600, 1000, 1500, and 2000 respectively. They were cleaned with acetone to remove any oil, washed with distilled water, and then allowed to air dry before being submerged in the corrosive media .

Antioxidant activity

DPPH free radical scavenging activity

DPPH is an example of a stable organic radical that has the potential to cause damage to cellular membranes. The DPPH method is often utilized in the research that is conducted on the antioxidant capacity of both unprocessed extracts and pure compounds.When methanol is present, the presence of a stable free radical known as DPPH results in the production of a violet color. The transformation of the DPPH radical into DPPH-H could be observed when antioxidants were present because the color of the DPPH solution shifted from violet to yellow as a result of the transformation. Methanol was used to create a stock solution of DPPH with a concentration of 4 milligrams per one hundred milliliters. The antioxidant potential of the compounds was determined by measuring the absorbance of a DPPH solution at a wavelength of 517 nm.

Each individual solution was given an addition of approximately 2 milliliters of the DPPH solution in methanol, in addition to varying amounts of both standard ascorbic acid and plant extracts. After being violently stirred for

a quarter of an hour in the dark, the reaction mixtures were then observed. To create the control solution, 2 milliliters of methanol and 2 milliliters of DPPH solution were added. The absorbance of each reaction mixture as well as the control solution was determined by looking at it at 517 nm.

The percentage inhibition (%) was calculated by the following equation.

$$\% \text{ Inhibition} = \frac{(\text{AC } 517\text{nm} - \text{AS } 517\text{nm})*100}{\text{AC } 517\text{nm}}$$

Where,

AC is the absorbance of the control and AS is the absorbance of the sample

Higher antioxidant capacity is shown by a lower IC50 value. The average result and standard deviation of each test were computed after each test was carried out in triplicate.

RESULTS AND DISCUSSIONS

Antioxidant activity

DPPH radical- scavenging activity

DPPH scavenging assay potential of methanol extract of plants

The DPPH scavenging assay was used to assess the in-vitro DPPH scavenging capability of methanol extracts of leaf and stem derived from T. occidentalis and A. heterophylla (TOLMe, TOSMe, AHLMe, AHSMe). These extracts were given the names TOLMe, TOSMe, AHLMe, and AHSMe, respectively. Free radicals were inhibited at doses ranging from 20 to 100 micrograms per milliliter in methanol plants.

In the methanol extract of the plants, TOLMe exhibited the best percentage inhibition of DPPH radical at concentrations ranging from 20 to 100 micrograms per milliliter, with an IC50 value of 46.407 micrograms per milliliter. On the other hand, TOSMe showed the lowest percentage inhibition of DPPH radical at concentrations ranging from 20 to 100 micrograms per milliliter, with an IC50 value of 47.161 micrograms per milliliter.

AHLMe showed the highest percentage of inhibition of DPPH radical at concentrations ranging from 20 to 100 g/ml, with an IC50 value of 77.719 g/ml. In contrast, AHSMe showed the lowest percentage of inhibition of DPPH radical at concentrations ranging from 20 to 100 g/ml, with an IC50 value of 154.85 g/ml. Ascorbic acid showed a

percent suppression of the DPPH radical at concentrations ranging from 20 to 100 micrograms per milliliter, with an IC50 value of 35.03 micrograms per milliliter, as shown in Figure 12.

% Inhibition of DPPH radical by methanol extract of plants follows the order i.e., TOLMe>TOSMe>AHLMe>AHSMe

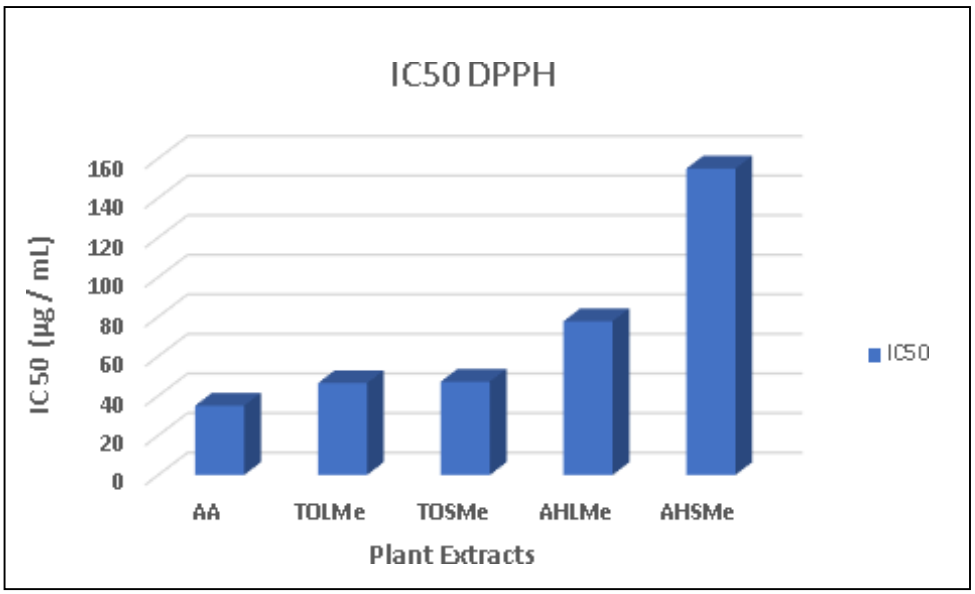

Figure 12: DPPH antioxidant potential of leaf and stem extracts in methanol of *Thujaoccidentalis* and *Araucaria heterophylla*

DPPH scavenging assay potential of hydro alcohol extract of plants

Hydroalcohol extracts of T. occidentalis and A. heterophylla were tested for their ability to scavenge DPPH radicals in vitro. The DPPH scavenging assay was utilized for the purpose of determining TOLHA, TOSHA, AHLHA, and AHSHA. The suppression of free radicals by hydro alcohol plants was dose dependent, occurring at concentrations ranging from 20 to 100 g/ml.[129]

Among the hydro alcohol extracts of the plants, TOLHA showed the highest percentage inhibition of DPPH radical at concentrations ranging from 20 to 100 g/ml and had an IC50 value of 92.993 g/ml. On the other hand, TOSHA showed the lowest percentage inhibition of DPPH radical at concentrations ranging from 20 to 100 g/ml and had an IC50 value of 115.657 g/ml. The IC50 value for the percentage of DPPH radical inhibition caused by AHLHA was found to be 87.629 g/ml, whereas the IC50 value for the percentage of DPPH radical inhibition caused by AHSHA was found to be 145.667 g/ml. Ascorbic acid showed a percent suppression of the DPPH radical at concentrations ranging from 20 to 100 micrograms per milliliter, with an IC50 value of 35.03 micrograms per milliliter, as shown in Figure 13.

% Inhibition of DPPH radical by methanol extract of plants follows the order i.e., AHLHA>TOLHA>TOSHA>AHSHA.

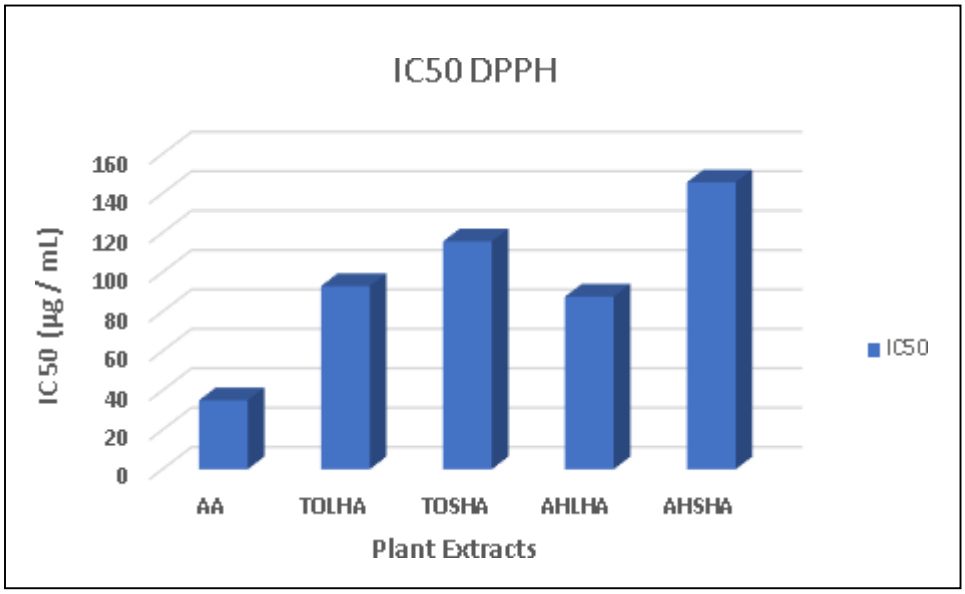

Figure 13: DPPH antioxidant potential of leaf and stem extracts in hydro alcohol of *Thujaoccidentalis* and *Araucaria heterophylla*

Table 4: Antioxidant capacity of leaf and stem extracts of *Thuja occidentalis* and *Araucariaheterophylla*.

EXTRACT	DPPH (IC50 µg/ml)
AA	35.03±0.0145
TOLMe	46.407±0.0003
TOLHA	92.993±0.0010
AHLMe	77.719±0.0012
AHLHA	87.629±0.0018
TOSMe	47.161±0.008
TOSHA	115.657±0.0012
AHSMe	154.85±0.0003
AHSHA	145.667±0.0006

The data represent mean± SEM of three determinants

*p< 0.05, significantly different from standard.

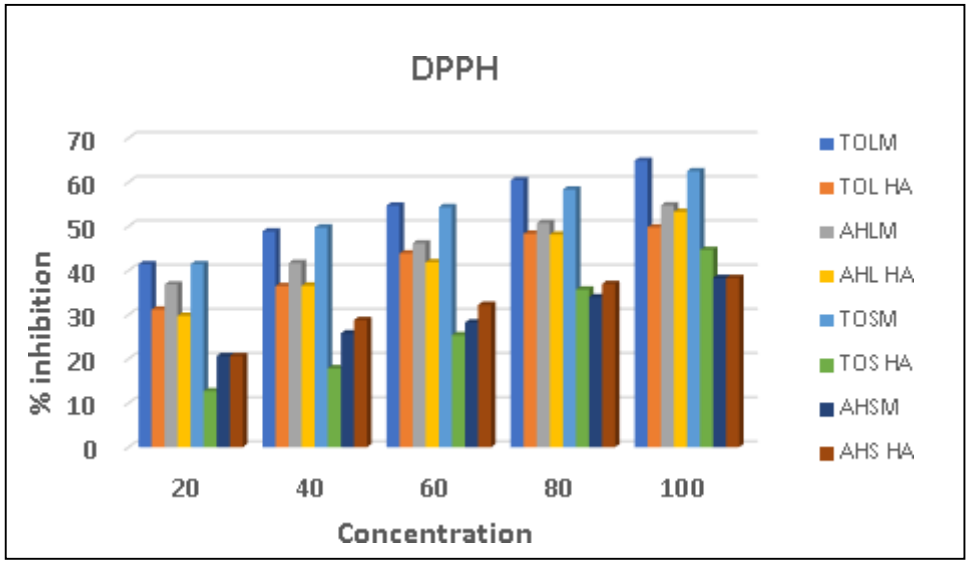

Figure 14: Comparison of % inhibition of *T. occidentalis* and *A. heterophylla* leaves andstem extracts against concentrations

CONCLUSION

The findings of a Antioxidant investigation of extract of Thuja occidentalis and Araucaria heterophylla leaves and stems have been given. The investigation was conducted on both of these plants. In terms of antioxidant activity, the hydroalcohol extract of the leaves and stem came in second place after the methanol extract. In the DPPH scavenging procedure, the extract obtained from methanol provided the highest percentage of scavenging activity, followed by the extract obtained from hydroalcoholic. Methanol extract performed significantly better than hydroalcohol extract did in the hydrogen peroxide assay.

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