

# Pharmacological Activities of Essential Oils from Medicinal Plants

## A systemic Review

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### Abstract:

Through an analysis of the oil's phytochemical makeup, the antifungal, insecticidal, and antioxidant capabilities of Haplophyllum tuberculatum essential oil not entirely settled and analyzed. The usage of gas chromatography-mass spectrometry analysis resulted in the discovery of fifty chemicals, with  $\beta$ -Caryophyllene being the major fixing identified among these compounds. The results of the study revealed that HTEO demonstrated a significant level of effectiveness against *Aspergillus flavus*, as confirmed by a hindrance rate of  $87.61\% \pm 1.71\%$ . The antifungal effects of this substance, then again, were not nearly as intense as the antibacterial effects. Furthermore, the results of the contact and inhalation tests indicated that it had a vital insecticidal action against *C. maculatus*. This was affirmed by the LC50 values of 30.66 and 40.28  $\mu\text{L}/100\text{g}$ , respectively. It was resolved that the essential oil possessed an antioxidant capacity of 869.45 mg AAE/g. This was mostly because of the presence of  $\gamma$ -Terpinyl isobutyrate, which is an antioxidant, as well as  $\gamma$ -Cadinene and p-Cymen-7-ol, which are principally responsible for their antifungal action. With regards to the treatment of illness, the utilization of HTEO as a sustainable and environmentally friendly alternative to synthetic medications is turning into an increasingly prevalent practice.

**Keywords:** Haplophyllum Tuberculatum, Pharmacological Activities, Medicinal Plants, Essential Oil

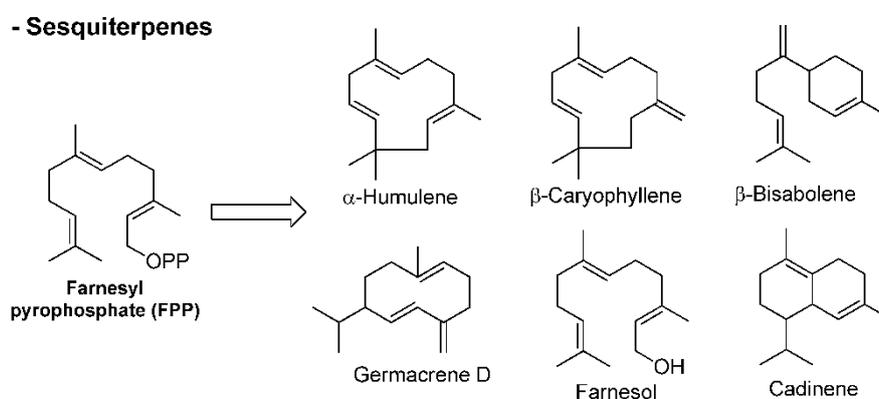
### I. INTRODUCTION:

Medicinal plants have become increasingly popular for use in the treatment and prevention of diseases due to the therapeutic benefits they offer and the fact

that they are relatively inexpensive. The insecticidal, antibacterial, and antioxidant characteristics that they possess are just some of the varieties of bioactive compounds that they possess. In spite of this, research is being conducted to

determine whether the essential oil (EO) of aromatic plants, such as *Haplophyllum tuberculatum* (*H. tuberculatum*), can be utilised for the treatment of fungal diseases such as *Aspergillus flavus* and *Fusarium oxysporum* [1]. These diseases have a negative impact on agricultural yields and can be harmful to individuals who have immune systems that are compromised. The use of synthetic pesticides to treat respiratory ailments, metabolic issues, neurological disorders, oxidative stress, and

DNA damage is commonplace, despite the fact that these pesticides can cause these illnesses. Because of their relatively low toxicity to animals and their ability to exert a variety of effects, essential oils have become viable alternatives. Pesticides that are generated from essential oils or their constituents have been shown to be effective against a wide variety of pests that are involved in the transmission of diseases to humans and agricultural crops.



**Figure 1: The Chemistry and Pharmacological Activities of Essential Oils**

An extensive number of research have been conducted to investigate the antioxidant potential of essential oils that have been isolated from medicinal plants in order to produce external antioxidants that are appropriate for human usage [2].

As a result of research, it has been discovered that extracts from *H. tuberculatum* have the ability to speed up the healing process of wounds, as well as possess anti-HIV, anti-inflammatory, and pain-relieving agents. Among the clinically identified Gram-positive and Gram-negative bacteria, silver nanoparticles generated from *H. tuberculatum* extracts

demonstrate substantial antibacterial activity. Furthermore, these nanoparticles are efficient against faecal and total coliform colonies.

The insecticidal effects of *H. tuberculatum* and the essential oil's potential properties as broad-spectrum antifungals have not been fully explored, despite the fact that *H. tuberculatum* has been the subject of substantial research. Note that the bioactive

chemicals found in these extracts differ from one source to another. This is something that should be taken into consideration [3]. Through the utilisation of both in vitro and in silico approaches, the

purpose of this work is to investigate the phytochemical composition of HTEO and assess its insecticidal, antioxidant, and antifungal capabilities against designated pathogens such as *C. maculatus*, *A. flavus*, *A. niger*, and *F. oxysporum*.

## II. Techniques For Obtaining Essential Oils:

According to the definition that is given by AFNOR, essential oils can be extracted using either cold expression (on account of Hesperides) or distillation, which can be accomplished through either steam entrainment or hydro-distillation. These three approaches can be utilized in a system that is either continuous or discontinuous, at atmospheric pressure, in overpressure, or in depression. Without first characterizing the processes that take place during the distillation and/or steam training, giving a full description of these approaches is impossible:

### i. The phenomena occurring during extraction:

- To start, there is the stage of real extraction, also known as hydro-diffusion, which involves the release of volatile chemicals into the aqueous medium [4]. A physical increase in the internal pressure of plant matter that has swollen as a result of passive or osmotic water absorption is responsible for this release. Additionally, a chemical peculiarity that is applied by water is also responsible for this release.
- Then comes the co-distillation of water and volatile elements.

- Finally, the separation of the essential oil from the condensates involves coalescence and settling.

### ➤ Hydro-diffusion:

This occurs not just as a result of the osmotic exchanges that take place between the water phase and the substrate plant, yet additionally as a result of the physical forces that are at play.

**Physical forces:** At the point when a plant mass is immersed in a medium that is saturated with water, it is inevitable that various different hydraulic pressures will accumulate. An individual plant particle that is submerged in water experiences total hydraulic pressure, which may be separated into the following components:

- Osmotic pressure
- Matrix pressure: the water that is adsorbing on the surface of the particles and the particles themselves.
- Static pressure: At atmospheric pressure, the pressure that is exerted on the plant membranes is equal to zero because of the conditions that are in place [5].
- Gravity.

In addition to these many pressures, there are also a variety of migrations that can occur. The movement of volatile chemicals throughout the plant substrate; migrations:

- The porosity of the plant mass contributes to the capillary diffusivity that occurs.
- Molecular diffusivity: the components of essential oils are

able to move across plant tissues through a process known as simple molecular diffusion.

➤ **Co-distillation:**

Right now, the petroleum is transferred from the surface of the plant molecule to the aqueous medium, where it gradually disperses all through the surroundings. Two distinct physical rules are responsible for the entrainment of natural molecules that happen during the distillation process [6].

- **Dalton's law:** There is a relationship between the pressure of the vapour mixture and the total amount of tension vapours produced by the individual ingredients:

$$PT = TH + TE. \quad (1.1)$$

- **Raoul's law:** Contingent upon the voltage and fume density at the distillation temperature that is selected, the proportion of the quantities of products that are distilled simultaneously is a component of the voltage.

➤ **Coalescence and settling:**

**MoleH/MoleE=THMoleH/MoleE=THE1**  
:

To a considerable degree, the process of isolating unpredictable molecules is subject to the solubility of those compounds in water. Because the distillate can be pretty much plentiful in polar constituents, we then separate between the two:

- A piece of the distilled oil is dissolved in water, this sum is on the request for 1%, seldom over 2% and

for some phenolic compounds polar over 5%.

- Another part is emulsified in water at the degree of 10%.
- The last piece is emulsified with water and natural molecules thirds playing the job of surfactants, it can surpass 10%.

**ii. Methods and equipment for obtaining essential oils:**

▪ **Extraction by entrainment with water vapor:**

After the vegetable mass has been exposed to a stream of steam (with next to no earlier maceration), the fume that is saturated with unpredictable components is condensed and afterward emptied for further processing.

▪ **Extraction by hydro-distillation:**

After the plant material has been submerged in water, the whole combination is heated to the point of boiling under tension, which is regularly atmospheric pressures.

▪ **Hydro-distillation under pressure:**

It is strongly suggested for essential oils that are hard to distil or potentially incorporate components that are more susceptible to temperature changes [7]. The truth is that unstable chemicals with a huge sub-atomic mass, such as those tracked down in sandalwood, ginger, and vetiver, can't be pressure-distilled at a surrounding temperature that is satisfactory to forestall their crumbling.

▪ **The heat pump system:**

With the single or twofold impact still, the intensity from the condenser is used to add to the making of the fume that will pass through the natural substance. This is finished to accomplish the desired impact. This is mostly to conserve energy (60%) and warming water (90%).

▪ **Turbo-distillation:**

The main contrast among this and typical distillation is the presence of a turbine that pulverizes the plant material and stirs it about. This sort of distillation is known as sped up bunch hydro-distillation, and it is done under constant atmospheric pressure [8]. Because to this last, the surface contact between the steam and the substrate is increased, which thus leads to a rise in the yields of energy and creation. Vapors from an improvement system can be installed in this apparatus; in any case, most of the time, it is a reflux system.

▪ **Microwave assisted distillation:**

Despite the way that this is a lab procedure, it has always been unable to track down a position in the industry because of specialized problems. This strategy is exceptionally charming because of the way that it allows for the acquisition of additional time and because of its presentation.

▪ **Cold expression:**

Cold expression is a strategy that is used exclusively for the extraction of unstable chemicals from the pericarps of Hesperides. Tormenting the pericarps, which are bountiful in secretory cells, is the mechanical treatment that is being presented here [9]. Because the petroleum

that has been discharged is gathered by a stream of water and acquires each of the common products of entrainment with water fume, the expression "essential oil" (AFNOR) was begat to describe this peculiarity.

**III. Materials and methods:**

**i. Plant material:**

The month of January 2023 saw the collection of aerial sections of *Haplophyllum tuberculatum* from a particular location. The taxonomic identification was conducted out by a university, and the reference sample was stored at the herbarium of the Faculty of Sciences.

**ii. HTEO extraction:**

Using the hydrodistillation technique, the essential oil of *Haplophyllum tuberculatum* was extracted and stored at a temperature of -4 degrees Celsius until it was required.

**iii. Chemical analysis of HTEO:**

In this experiment, a gas chromatograph was utilised, and it was equipped with a fire ionisation locator as well as a CP-Sil-5CB Varian narrow segment. The segment was heated from 40 degrees Celsius to 280 degrees Celsius, while the injector and locator were maintained at temperatures of 250 degrees Celsius and 280 degrees Celsius, respectively [10]. An investigation into the components of HTEO compounds was carried out with the assistance of a PolarisQ spectrometer and a Follow GC-ULTRA gas chromatograph.

At a rate of three degrees Celsius per second, the temperature of the section increased from forty degrees Celsius to two

hundred and eighty degrees Celsius. Following the process of reducing the concentration of HTEO in hexane, it was introduced into the gas chromatograph with minimal fractionation, employing a volume of 1  $\mu$ L within the apparatus. Measurements of the retention index (RI) of essential oil compounds were utilised in order to determine the components of essential oil that are unstable. This was accomplished by using a homologous sequence of n-alkanes.

**iv. Antifungal activity of HTEO:**

A study was conducted at the University to determine whether or not the essential oil of *H. tuberculatum* possessed antifungal properties against *Aspergillus flavus*, *niger*, and *oxysporum*. With the help of agar diffusion, HTEO was examined to determine its antifungal activity. Following the inoculation of three different fungal strains into Petri plates, twenty microliters of *H. tuberculatum* essential oil was spread throughout the surface. On the seventh day, the percentage of inhibition was determined. The MIC of HTEO was determined by employing microdilution to test it against three different strains of fungus [11]. For the inoculation of microplates, HTEO was first dissolved in DMSO, then serially diluted with malt extract, and finally injected. The MIC endpoint was determined through the use of colorimetric tests as well as the direct observation of the growth of microorganisms. Using Tukey's test, comparisons were made between the treatment groups.

**v. In silico evaluation of antifungal, and antioxidant activities of EOHT:**

Using in-silico techniques, the research investigated the phytochemical activities of HTEO. The compounds were put through a series of tests with *A. flavus* and *A. niger* FAD glucose dehydrogenase, beta-1,4-endoglucanase, and NAD(P)H Oxidase to get data with respect to their cell reinforcement properties. In order to build compound structures, LigPrep and OPLS3 force field were utilised. These structures were taken from PubChem. The protein preparation wizard and the receptor grid construction tool were utilised in order to manufacture the structures of the proteins.

**vi. Insecticidal activity against *C. maculatus*:**

Utilising *Cicer arietinum* from Fez-Meknes, an evaluation was conducted to determine the insecticidal activity of *H. tuberculatum* essential oil against *C. maculatus*. Using a photoperiod of 10 hours and 14 hours of light, the insects were brought up in controlled conditions that were designed to mimic the natural light cycles that occur in the wild.

In order to examine the efficacy of HTEO as an insecticide against *C. maculatus*, the study utilised ten individuals to infest chickpea seeds for a period of two days. HTEO was applied to the seeds in a range of concentrations, and a control group was established throughout the experiment. The death rate of adult insects was determined after a day of confinement, and the quantity of eggs was counted after 12 and 28 days since the beginning of the experiment. Abbott's formula was utilised in order to make adjustments to the observed death rates.

$$Pc = 100 \times \frac{P0-Pt}{100-Pt} \quad (1.2)$$

In the study, a formula was used to determine the percentage of adults and eggs that were reduced when HTEO was present in varying concentrations in comparison to the control:

$$PR = \frac{NC-NT}{CN} \times 100 \quad (1.3)$$

The level of oviposition or decrease of arisen insects, which is shown by PR, as well as the number of eggs or insects that brought forth was thought about between the benchmark group and the treatment bunch.

The experiment consisted of using glass jars with cotton pieces suspended inside of them, and then depositing varying quantities of HTEO onto the cotton material. A total of ten *C. maculatus* insects were placed inside of each jar, and three separate experiments were carried out for each concentration. Through the application of the Abbott formula, the mortality rate was adjusted.

In order to determine whether or not HTEO possesses repellent qualities, the preferential surface area approach was administered to filter paper. Two halves of filter paper discs were cut in half, with one half containing different concentrations of HTEO and the other half containing any amount of acetone [12]. The number of insects that were present on the EO-treated area and the control area was counted after thirty minutes:

$$PR = \frac{NC-NT}{CN+NT} \times 100 \quad (1.4)$$

The percentage of repulsion is denoted by PR, while the quantities of insects in the control and treatment areas are denoted by NC and NT, respectively.

#### vii. Evaluation of antioxidant activity:

The cell reinforcement force of EOHT was assessed using three tests: DPPH assay, FRAP assay, and all out-cancer prevention agent limit measurement.

#### viii. Data analysis:

The death pace of *C. maculatus* insect was determined using the Abbott equation, and LC50 concentrations were resolved using the probit technique and IBM SPSS software.

### IV. Results and Discussion:

#### i. Essential oil yield and GC-MS analysis:

The amount of HTEO that was extracted using hydrodistillation was 0.27%, which was a fluctuation that was impacted by a number of factors including the stage of maturation, genetics, ambient circumstances, and geographic differences. The yields that were reported in earlier investigations ranged from 0.101% to 0.65% percent. The GC-MS investigation uncovered fifty different chemicals, with sesquiterpenes constituting fifty-seven-point seven percent of the total composition.

A considerable portion of the chemicals were composed of  $\beta$ -Caryophyllene, followed by monoterpenes,  $\alpha$ -Phellandrene, Germacrene D, and Eugenol. Eugenol was the last of the chemicals. Oxygenated monoterpenes, monoterpene hydrocarbons,

and non-terpene hydrocarbons are comprised of the constituents that constitute the phytochemical profile of terpene-containing essential oil (HTEO). To add

insult to injury, this profile also contains oxygenated sesquiterpenes as well as sesquiterpene hydrocarbons.

**Table 1: Antifungal activity of HTEO**

| Microorganism | % Inhibition   | Inhibition MCI (mg/mL) |
|---------------|----------------|------------------------|
| A. niger      | 72.33 ± 3.22a  | 87.37 ± 6.19           |
| A. flavus     | 87.61 ± 1.71 b | 75.64 ± 6.09a          |
| F. oxysporum  | 39.62 ± 0.70c  | 61.51 ± 6.65           |

**ii. Antifungal activity:**

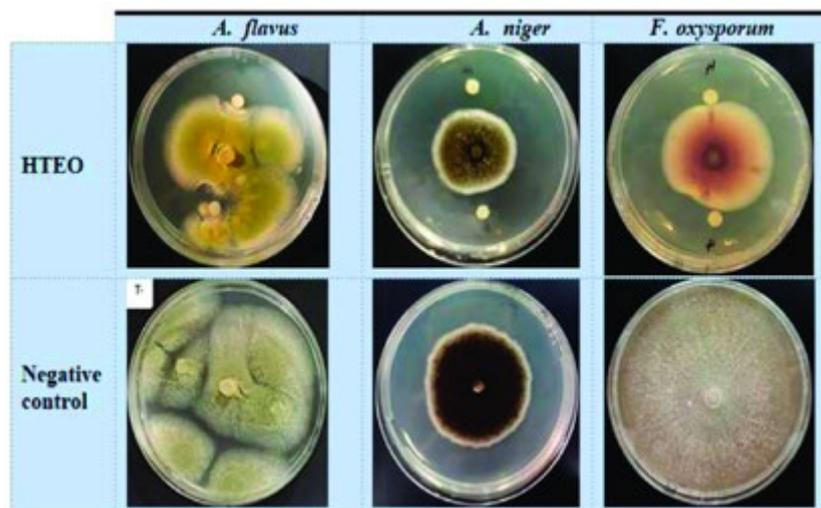
flavus was shown to have the highest sensitivity to the antifungal activity of HTEO, which was assessed in the study against three different fungal strains. While F. oxysporum exhibited a lower level of sensitivity, A. niger demonstrated a moderate level of sensitivity. Within the range of  $20.53 \pm 5.08$  to  $87.37 \pm 6.19$  mg/mL, the minimum inhibitory concentration (MIC) values were reported. When it comes to protecting against pathogenic moulds, HTEO's antifungal effectiveness is limited. Skimmianine and vulcanine have been shown to have antimicrobial activities against phytopathogenic bacteria and fungi, according to research that was conducted in the past on the antimicrobial characteristics of H. tuberculatum extracts.

Both the phytochemical content of the extract and the particular fungal strain both

play a role in determining the antifungal impact that HTEO has on Candida albicans. Nevertheless, HTEO is also capable of exhibiting antimycelial activity.

**iii. In-silico evaluation of the antifungal, antioxidant and activities of HTEO:**

During the course of the research, phytochemicals found in HTEO were subjected to targets, and the results uncovered their limiting affinities for proteins. The compounds  $\gamma$ Cadinene and p-Cymen-7-ol showed the highest liking for the FAD glucose dehydrogenase from A. flavus, whereas the compounds Carvacrol and Viridiflorol showed the best fondness for the  $\beta$ -1,4-endoglucanase from A. niger. The cancer prevention agent movement of  $\alpha$ -terpinyl isobutyrate was shown to be higher than that of NADPH oxidase. Through the development of hydrogen bonds, carvacrol took part in interactions with A. niger  $\beta$ -1,4-endoglucanase.



**Figure 2: Antifungal Activity of HTEO by Use of The Disc Method**

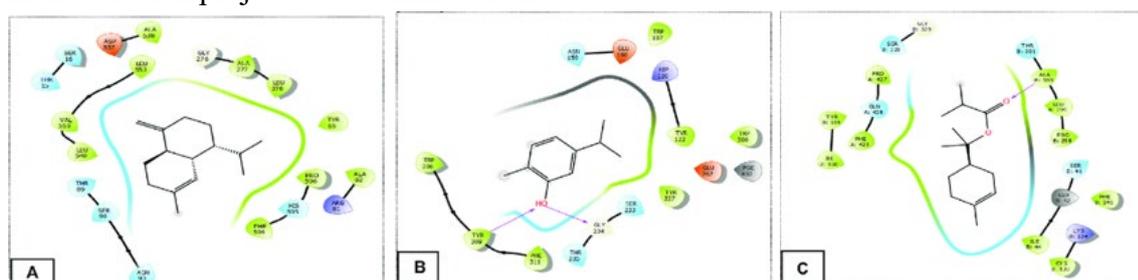
Within the scope of this study, the antioxidant properties of essential oils are examined, with particular emphasis placed on  $\alpha$ -Terpinyl isobutyrate and the hydrogen bonds that it forms within the NADPH oxidase active site. A number of chemicals found in essential oil, such as trans-caryophyllene and eugenol, have been identified as having persistent binding strengths in the acetylcholinesterase enzyme of *Tribolium castaneum*, according to the findings of the research. According to the findings of the study,  $\beta$ -caryophyllene, a significant molecule, demonstrates the ability to block enzymes such as  $\alpha$ glucosidase,  $\alpha$ -amylase, and xanthine oxidase.

#### iv. Insecticidal activity:

Both inhalation and contact tests were used in the research project to determine whether

or not HTEO possessed insecticidal properties against adult *C. maculatus*. According to the findings, the inhalation test demonstrated a low mortality rate at dosages of 10 and 20  $\mu$ L/100 g. On the other hand, the contact mortality tests demonstrated comparable outcomes. When administered at the highest dose (20  $\mu$ L/100 g), the death rate was 10%, and after 96 hours, it reached 63.33%.

In comparison to the inhalation test, the contact toxicity of HTEO was shown to be significantly higher, with lower LC50 and LC95 values. With the application of 20 microliters per one hundred grammes of HTEO, the emergence of bruchids was greatly reduced by 91.34 percent, and the number of eggs laid by female *C. maculatus* bruchids was significantly reduced as well.



**Figure 3:** The interaction between ligands and active sites is depicted 2D. (A) The active site of *A. flavus* FAD glucose dehydrogenase is bound to by  $\gamma$ -Cadinene. (B) The active sites of *A. niger* are occupied by carvacrol and  $\alpha$ -1,4-endoglucanase. (C) Interactions between  $\alpha$ -terpinyl isobutyrate and NADPH oxidase active sites

HTEO had a considerable impact on fecundity in *C. maculatus*, with doses of 5  $\mu$ L/100g producing a 48.85% reduction in oviposition and a 45.15% suppression of emergence. Additionally, dosages of 20  $\mu$ L/100g caused a reduction of 85.09% and 84.59%, respectively.

**v. Repulsive activity:**

The preferential surface approach was utilised in order to establish whether or not HTEO had a repellent impact on *C. maculatus*. The action was dose-dependent, with a repulsion rate of sixty percent after thirty minutes, which increased to eighty percent after one hundred twenty minutes.

This study delves into the investigation of HTEO's insecticidal action against *C. maculatus*, illuminating the compound's potent repellent properties. Both the inward breath test and the contact test were used to confirm the harmfulness, and both produced significant results with LC50 values of 14.59 and 14.68  $\mu$ L/100 g, respectively. The insecticidal potential of HTEO has been investigated in the past, both in relation to other plant extracts like *H. tuberculatum* and a wide range of other insect pests. According to multiple research projects, essential oils from different plants have different levels of activity against *C. maculatus*.

**Table 2: Docking outcomes involve ligands in various receptors**

| Compound                       | 2CDU Glide GScore (Kcal/mol) | 4YNT Glide GScore (Kcal/mol) | 5I77 Glide GScore (Kcal/mol) |
|--------------------------------|------------------------------|------------------------------|------------------------------|
| 1,8-cineole                    | -4.488                       | -5.364                       | -4.159                       |
| 2-Carene                       | -4.64                        | -5.286                       | -3.869                       |
| $\alpha$ -Bisabolol            | -5.579                       | -5.715                       | -3.983                       |
| $\alpha$ -Cadinene             | -5.543                       | -5.385                       | -4.366                       |
| $\alpha$ -Guaiene              | -4.666                       | -5.233                       | -3.986                       |
| $\alpha$ -Gurjunene            | -5.278                       | -6.26                        | -4.385                       |
| $\alpha$ -Longifolene          | -4.815                       | -5.615                       | -4.129                       |
| $\alpha$ -Longipinene          | -5.286                       | -5.612                       | -4.579                       |
| $\alpha$ -Phellandrene         | -4.784                       | -5.448                       | -4.469                       |
| $\alpha$ -Selinene             | -4.92                        | -5.879                       | -3.742                       |
| $\alpha$ -Terpinyl isobutyrate | -6.299                       | -5.589                       | -3.544                       |
| Aristolochene                  | -4.675                       | -4.884                       | -3.88                        |

**vi. Antioxidant activity:**

In order to determine the level of antioxidant activity that HTEO, which is an

extract of a plant, possesses, the DPPH, FRAP, and TAC tests were utilised. With an antioxidant activity value of  $869.45 \pm 3.87$

mg AAE/g, the oil demonstrated a noteworthy level of antioxidant activity.

The values of EC50 and IC50 were slightly greater than those of typical compounds, which is an indication of the presence of powerful antioxidant activity. When compared to the essential oil, the aqueous extract demonstrated a lower level of free

radical scavenging activity, but it exhibited a slightly higher level of iron-reducing activity. The essential oil (EO) that was isolated from the leaves and stems of *H. tuberculatum* was also found to have a modest level of antioxidant activity. It was also noted that the essential oils included a significant concentration of monoterpenes.

**Table 3: According to the results of the antioxidant activity (DPPH, FRAP, and TAC),**

| Compound      | DPPH (IC50 mg/mL) | FRAP (EC50 mg/mL) | TAC (mg AAE/g ext) |
|---------------|-------------------|-------------------|--------------------|
| EOHT          | 0.364             | 0.385             | 869.45 ± 3.87      |
| Ascorbic acid | 0.1138            | 0.111888          | —                  |
| Quercetin     | 0.118386          | 0.118386          | —                  |

Highly concentrated essential oils (HTEO) are known for their potent antioxidant properties. One of the main contributing factors to this effect is the high concentration of monoterpene molecules in HTEO. This is because the total antioxidant capacity of a cell can be affected by the presence of many chemicals, giving rise to ideas like additivity, antagonism, and synergy. As a result, HTEO's strong antioxidant activity can be attributed to its makeup, particularly the high quantity of monoterpene chemicals it contains.

#### V. Conclusion:

In addition to providing definitive answers, the study sheds important light on the organic properties of high-temperature essential oil (HTEO), especially in relation to its potency against the infamous *C. maculatus* [13]. Additionally, the study investigates HTEO's potential as a biocide to manage infectious species like *F. oxysporum*, *A.*

*niger*, and *A. flavus*. In particular, HTEO had insecticidal efficacy that was greater than essential oils (EO) obtained from other plant species. Additionally, it demonstrated a high cell reinforcement limit, which can be linked to the presence of a mixture of different terpene components [14]. This information can be found in the previous sentence. In contrast to the synthetic pesticides that are typically employed in the fight against pathogenic fungus in agricultural products, the results of a sub-atomic docking analysis that was carried out to evaluate the antifungal action of HTEO revealed the presence of compounds that have the potential to serve as a viable alternative. EOHT, by and large, shows itself as a prospective choice for further research and use in cell reinforcement interventions and pest the board techniques, subsequently advancing sustainable and environmentally friendly solutions in these areas [15].

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