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Evaluation Of Preliminary, Chromatographic And Antioxidant Activity Of Commiphora Mukul

Shweta Hingwasiya[1], Kamini Dubey[2*]

[1]Assistant Professor, M.L.B. Govt Girls P.G.(Autonomous)College,Bhopal(M.P.)
[2]Assistant Professor, Govt.P.G. College Narsinghgarh (M.P.)*

***Corresponding Author:**Kamini Dubey*
Email: kamini.neeraj@gmail.com

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Abstract:

The qualitative analysis, chromatography, and antioxidant studies of the*Commiphoramukul* extract are the topics of this research. Physiochemical parameters such as Ph (3.6), total ash value (2.16%), acid insoluble matter (0.25%), water soluble value (41.29%), and moisture content (7.2%) were measured for this study.The important information gathered throughout the study indicates that saponins are absent from a certain plant, it has a variety of phytochemicals including alkaloids, tannin, carbohydrates, protein, and steroids.These plant-based bioactive substances have a distinct, typical biological effect on human anatomy. Rf=0.89 for Googlesterone was obtained with a good resolution in TLC tests when the mobile phase (toluene: ethyl acetate) was in the ratio (93:07)v/v under UV light at 272 nm. According to the investigations, *Commiphoramukul* is a rich source of a variety of secondary metabolites. This can be further explored by economically isolating and identifying biologically active molecules with medical use. The alcoholic percentage was spectroscopically analyzed using UV spectra, which reveal the highest observance of the alcoholic fraction of Plant.The current study's findings demonstrate that alcoholic plant extract has strong in vitro antioxidant activity at increasing concentrations in both DPPH free radical scavenging activity and $FeCl_3$ reduction power.

Keywords: *Commiphoramukul*, Physicochemical properties, Phytochemical analysis, TLC studies

INTRODUCTION

The usage of medicinal herbs is one of the oldest and most varied cultural traditions in India. Medicinal herbs are essential to human life and the lives of all living things. These plants possess every quality that is necessary for living things to utilize for both sustenance and wellness [1]. Since there was no modern medical research back then, our forefathers employed these plants as medicine. They gather information about plants via study, research, and collection, then impart it to the following generation.

The next generation carries on this legacy by researching botanicals to discover potential treatments for illnesses. Over 75% of people in underdeveloped nations could not afford sophisticated allopathic medicine in

2003, according to a World Health Organization (WHO) research [2]. As a result, they favoured using plant-based remedies to address their basic medical needs. In its 16 Agro-favourable zones, which span over 64 million hectares of forest area, India's medical science has officially claimed that more than 45,000 plant species and more than 7000 types of medicinal plants flourish there [3].

The broad study of how humans obtain medication from bushes, plants, water, fungi, animals, or other remaining natural resources is known as ethnopharmacology [4]. As things stand, the field's primary focus has been on developing novel medications based on the medicinal plants that indigenous peoples use. One of the most often used struggles to preserve civilization and a decent, organic variety has been the discovery that traditional knowledge about medicinal plants may include hints to curing ailments [5]. Numerous bioactive substances that have been discovered by scientists and researchers have improved the efficacy of herbal medicine. These substances are referred to as plant secondary metabolites. One important method for creating and refining new medications is pharmacological research, source testing, and source modification [6]. Furthermore, the chemical and medicinal qualities that secondary metabolites of plants have are the most beneficial to human health [7]. Alkaloids, cardiac glycosides, saponins, flavonoids, phenol, and steroids are examples of secondary metabolites or bioactive chemicals that are used to treat a variety of disorders [8,9]. A number of these metabolites have also been shown to be effective in preventing and delaying the progression of different forms of cancer. While people all around the world have different opinions about the benefits and medical qualities of plants, they are all completely dependent on them for their environment. These plants can be used to cure most illnesses. The bark, stems, roots, and leaves of the plant are all useful for treating various ailments [10].
Ayurveda values *Commiphoramukul*, a Burseraceae family medicinal plant, for its profoundly penetrating effect and special capacity to remove poisons that naturally accumulate in the body's pathways. Oleo-gum resin, which is derived by tapping the stem and branches of*Commiphoramukul* (Hook. Ex Stocks) Engl; Balsamodendronmukul, is what makes up guggul.[11] The plant, often referred to as guggul tree, grows in desert regions of Bangladesh, Pakistan, and India. India's Rajasthan, Gujarat, Assam, Madhya Pradesh, and Karnataka are among the states that contain it.[12] The tree is tiny and bushy, with prickly branches and little ducts scattered throughout its bark that release guggul, a yellowish gum resin. The bark of the trees is cut in order to tap them. The flowing resin is given time to solidify before being collected.[13] .November through January is when the tree is tapped, and May through June is when the resin is collected[14]. Every collecting season, a guggul tree produces 250–500 g of dry resin [15,16].

Guggul has been used for thousands of years in the ancient Indian medical system to treat rheumatism, obesity, gout, inflammation, arthritis, and problems with lipid metabolism [17]. It goes under several names, including Indian bdellium, gugar, guggula, guggul, and guggal [18].

Guggul is found in tiny, vermicular bits that are pale yellow or brown in color, have a bitter, astringent taste, and are scented when fresh. [19]. Guggul is the main ingredient in a great deal of commercial polyherbal anti-inflammatory preparations [20].Investigating Commiphoramukul's physicochemical, phytochemical, and chromatographic investigations is crucial, nevertheless.gathering of vegetation.

MATERIAL AND METHODS

Collection of Botanical Specimens

In March 2023, the gum resin portion of *Commiphoramukul* was harvested from Bhopal's suburbs. The resins are employed in phytochemical screening and chromatography studies and research. Following a thorough washing with running water, the recovered plant components are quenched in distilled water. The cleaning material is then easily dried by cutting it into pieces that are 5-8 cm in size. After that, it is allowed to dry for eight to ten days at ambient temperature in a dark, shady area to prevent contamination. Afterwards, an electrical grinder is used to grind the fully dry materials into a coarse powder. This dried plant material powder was kept for use in chromatography and phytochemical analysis research in an airtight container.

Defatting of the substance

We soak the coarsely ground plant material in petroleum ether for a full day at room temperature in order to begin the defatting process. To separate the necessary material from the contaminants, the material is filtered after 24 hours using a funnel, spatula, and filter paper [21]. The filtered material should be dried and then kept in an airtight container.

Chemical ingredient

Fehling solutions A and B, Erdmann's reagent, acetic acid, diluted iodine, biuret reagent, hydrochloric acid, chloroform, sulfuric acid, and sodium hydroxide are important chemical reagents for phytochemical investigation.

Material extraction using the maceration method

The powdered plant material is submerged in a container of water, and alcohol is added intermittently. It is then left to stand at room temperature for 48 hours. Once the alcohol is still clear, filter it and let it sit at room temperature to evaporate the alcohol.

Preliminary analysis of the plant

To determine whether active chemical ingredients or phytoconstituents are present or absent, physicochemical investigations and phytochemical testing have been conducted.pH, Total Ash Value, Acid Insoluble Matter, Water Solubility Value, and Moisture Content are measured in physicochemical research. For the phytochemical test, flavonoids (alkaline reagent test), proteins (Biuret test), carbohydrates (Fehling test), proteins (Tannins test), phenolic compounds (reaction with diluted Iodine), steroids (Salkowski reagent), and saponins (Saponification test) are tested [22, 23, 24].

Thin layer chromatography

The method or technique used to separate non-volatile mixtures is called thin layer chromatography. Plant extract with alcohol was subjected to thin layer chromatography (TLC) using silica 60F254, 7*3cm (Merck) in accordance with the traditional one-dimensional ascending procedure. The extract was then cut with regular scissors. Pencil markings were made on the silica plate. Glass capillaries were employed in thin layer chromatography to spot the sample. A sample volume of one microliter was applied using capillaries spaced one centimeter apart at two tracks. Toluene and ethyl acetate are utilized in the solvent system in a ratio of 93:07 (v/v). They are utilized once the run plates have dried and have been pre-saturated with mobile phase for 20 minutes for development. Retention factor (Rf) values were computed to express the mobility of the active ingredient. Vanillium sulphuric acid, a derivatizing agent, was used to visualize the resolved component after the standard and sample were spotted on the TLC plate. The formula Rf=Distance travelled by solute / Distance travelled by solvent can be used to compute Rf. The Rf value of the spot was computed after the chromatogram was generated.

Column Chromatography

Utilizing column chromatography, phytoconstituents from a particular plant were separated. Permit a medium-sized borosilicate glass dry column, already equipped with a filter device, to stand.Create a slurry of silica gel G (grades 60–120) using the same solvent system, then use a vacuum pump to pack the column (as the stationary phase) for optimal packing.As a filter and parting agent in between the stationary phases, add dry inactivated sand, then extract the sample.Next, combine each plant's extract with Silica Gel G (grade 60–120) using the same solvent system, stirring until the solvent evaporates.Using a funnel, add the prepared extract sample on top of the Silica Gel G that has been packed into the column.Top-fill the produced extract sample with a solvent system (toluene: ethyl acetate, 93:07 v/v) and cover the column with silver foil from top to bottom for 12 hours.Following a 12-hour period, different fractions of the alcoholic extract of plant were obtained. Every eluted fraction is sent for additional TLC analysis, and if a solvent system detects a single spot, it suggests that there is a single component in that fraction, as confirmed qualitatively.

Shortly after the chemical is isolated, it undergoes thorough characterisation in order to confirm the likely structure of the drug using spectral analysis. [26]

Tests for antioxidant properties using biochemistry

Plant extract's antioxidant capacity was assessed using a variety of in vitro techniques.

1. *$FeCl_3$-induced power reduction*

The extract's reducing activity was ascertained using Oyaizu's technique (Oyaizu, 1986).[27]

2. *Radical Scavenging by DPPH*

Radical Braca et al. (2001) carried out the scavenging activity of 1,1 diphenyl-2-picryl hydrazyl (DPPH) free radical.[28]

RESULTS AND DISCUSSION

The physicochemical properties of the resin sample were examined and interpreted for the preliminary study, as shown in Table 1. The qualitative test conducted in the alcoholic extracts used in this investigation indicated the presence of metabolites.Different phytoconstituents were found in the alcoholic extract of *Commiphoramukul* at the time of Phytochemical investigation. According to study, the extract lacks saponin. There are proteins, sugars, steroids, tannins, and alkaloids.Based on the findings of this investigation, new medications that can treat more illnesses or disorders can be created by identifying these bioactive chemicals. Different phytochemicals' presence and absence are displayed in the Table 2.Medicinal plants contain a variety of bioactive chemicals with significant therapeutic benefits for both individual and community health. Fractionation and isolation of phytoconstituents from alcoholic extract, Thin layer chromatography was utilized to extract guggulsterone, with vanillin sulphuric acid serving as the derivation agent.Under UV light at 272 nm, guggulsterone exhibits yellowish brown luminous zones in alcoholic extract of resin at Rf 0.89 was shown in Table:-3. The TLC plate with the alcoholic extract of *Commiphoramukul* is displayed in Figure 1. TLC profiling of the plant's extract in a solvent system under the current conditions indicates that the plant has a variety of phytochemical kinds. The fraction that came out of column chromatography had a colour that was yellowish brown. It is insoluble in water but soluble in the majority of organic solvents.The resulting compounds display the UV absorption maximum (λ max) measured in nm using a Shimadzu spectrophotometer. The alcoholic fraction underwent spectroscopic investigation using UV light, and the resulting spectra in Figure 2 display the maximum absorbance (λ max) of the alcoholic fraction at 272 nm.The ability of the alcoholic extract of*Commiphoramukul* to scavenge free radicals produced by DPPH and convert ferric to ferrous ions was used to calculate the extract's antioxidant activity.Table 4 compares the antioxidant activity of the extract with ascorbic acid.The DPPH activity was significantly inhibited by the alcoholic extract of *Commiphoramukul*. Similarly, Table 5 displays the extracts' reductive powers in relation to ascorbic acid. The extract's reducing power is highly significant, and it increases with sample quantity.

Table:-1 PHYSICOCHEMICAL PARAMETERS RESIN OF *COMMIPHORA MUKUL*

S.No	PARAMETERS	RESULTS
1	DESCRIPTION	BROWNISH
2	PH	3.6
3	TOTAL ASH VALUE	2.16%
4	ACID INSOLUBLE ASH	0.25%
5	WATER SOLUBLE ASH	41.29%
6	MOISTURE CONTENT	7.2%

Table 2 TEST FOR PHYTOCONSTITUENTS FOR DIFFERENT EXTRACT OF *COMMIPHORA MUKUL*

S.No	SECONDARY METABOLITES TEST	ALCOHOLIC EXTRACT
1	ALKALOID TEST	PRESENT
2	TANNIN	ABSENT
3	PHENOLIC COMPOUNDS	ABSENT
4	CARBOHYDRATES	PRESENT
5	PROTEIN	ABSENT
6	SAPONIN	ABSENT
7	STEROID TEST	PRESENT
8	FLAVANOIDS	PRESENT
9	GLYCOSIDES	PRESENT

Table 3 ESTIMATION OF GUGGULSTERONE FROM ALCOHOLIC *COMMIPHORA MUKUL* BY THIN LAYER CHROMATOGRAPHY

S,NO.	SAMPLE	SOLVENT SYSTEM	NO OF SPOTS	RF VALUES
01	MATERIAL	Toulene:Ethyl Acetate(93:7)(v/v)	01	0.89

Table 4 ANTIOXIDANT ACTIVITY OF ALCOHOLIC EXTRACT OF *COMMIPHORA MUKUL* BY DPPH METHOD

S.NO.	Concentration µg/ml	% Antiradical Activity	
		Alcoholic Extract	Ascorbic Acid
1	100	31.68	42.08
2	200	46.98	58.75
3	300	62.38	69.68
4	400	76.95	82.56
5	500	86.84	94.63

Table 5 ANTIOXIDANT ACTIVITY OF ALCOHOLIC EXTRACT OF *COMMIPHORA MUKUL* BY REDUCING POWER BY $FeCl_3$

S.NO.	Concentration µg/ml	% Antiradical Activity	
		Alcoholic Extract	Ascorbic Acid
1	100	21.76	38.59
2	200	37.63	47.32
3	300	52.69	63.67
4	400	67.49	76.45
5	500	76.96	87.13

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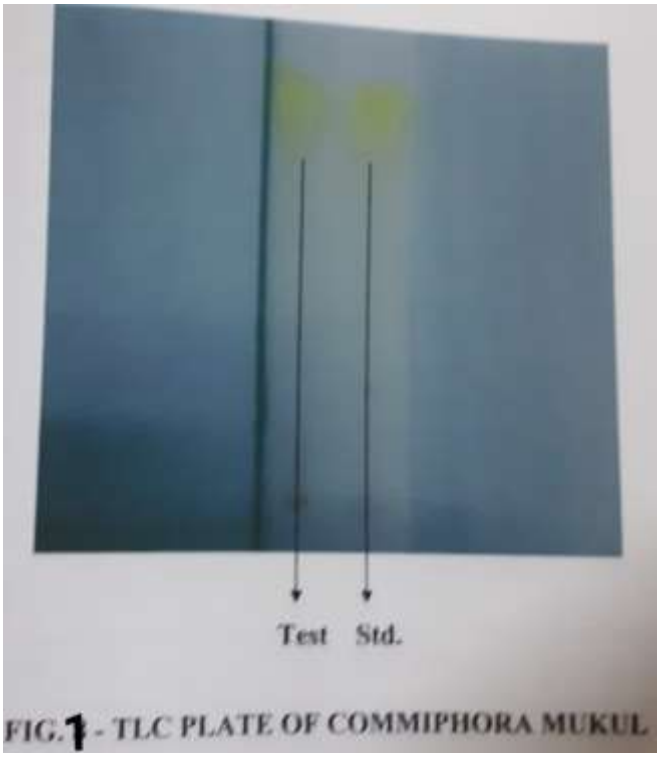

FIG.1 - TLC PLATE OF COMMIPHORA MUKUL