



Phytochemical Screening, Chromatographic analysis of Chloroform extract of *Conium maculatum*

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Abstract

Objective: To separate the bioactive compounds from crude chloroform extract of *Conium maculatum*, a herb from Jammu and Kashmir, India. **Methods:** Chromatographic separation was carried out by thin layer chromatography (TLC) of crude plant extract. **Results:** Preliminary and TLC technique has revealed the presence of Steroids and Terpinoids

Keywords: *Conium maculatum*, preliminary phytochemistry, TLC, Terpinoids.

Introduction

Some of the plants played important role in the treatment of few diseases even though they were toxic in nature. *Conium maculatum* dried leaf and juice was listed in Pharmacopoeias of London even though this is the most toxic members of the plant kingdom. Extracts of this species were used both as a sedative and an antispasmodic. Among these species the most poisonous is *Conium maculatum* is annual or biennial herb perennial herb in favourable conditions². Many secondary metabolites like alkaloids, phenolics, steroids characterized from *Conium maculatum*^{3,4}. *Conium maculatum* have been used as traditional medicines and its extract have proven activity against analgesic and anti inflammation⁵. Such activities could be attributed to the presence of biologically active anti inflammatory and analgesic compounds. Both Greek and Arabian physicians were in the practice of using it for the cure of indolent tumours, swellings and pains of the joints. The bitter juice of the plant was mixed with betony (*Stachys officinalis*) and fennel (*Foeniculum vulgare*) seeds for the bite of a mad dog. Later in history, this juice was administered as a last resort, as an antidote for strychnine and other strongly poisonous compounds⁶. Religious sects of the 15th and 16th centuries used the roasted roots for relieving the pains of gout. *Conium* remains a classic homeopathic agent with various uses⁷. Said to be a long-acting remedy, it is of special value in old age when the vital powers of the body are failing. It has been used against a serious type of malignant tumor.

So it is necessary to isolate characterize, determine the bioactivity of the lead compound for its pharmaceutical exploitation. Since Analytical methods play important roles in the discovery, development and manufacture of pharmaceuticals⁸. The traditional methodology of studying natural products includes the fractionation of a complex mixture, separation and isolation of the individual components using Column chromatography and structure elucidation using various spectroscopic methods^{9,10}. Isolation and separation of

bioactive compounds for further analysis are highly necessary. Analytical methods like thin layer chromatography (TLC) are the economical methods in the field of pharmacognosy. Chromatographic techniques are one of the simplest and cheapest methods for detecting plant constituents because these methods are easy to run, reproducible and requires little equipments^{11,12}.

Keep in view of the pharmacological importance of *Conium maculatum* the present study explores to isolate and characterization of bio active compounds.

In the present study, attempt has been made to identify the phytochemicals present in the crude extract and chromatographic separation of the compounds present in the extract.

Material and Methods

Collection of Plant Material: *Conium maculatum* plants were procured from JKMPIC (Jammu and Kashmir Medicinal Plants Introduction Centre) Nambalbal road, Pampore, J&K, India. The plant was authenticated By Dr. S. Gulzar, Head, JKMPIC.

Preparation of extracts: The plants were shade dried for one week and pulverized in pulverizer. This dried powder was packed in filter paper thimble and kept in Soxhlet apparatus, below this round bottom flask with chloroform (Ranbaxy Fine Chemicals, New Delhi, India) placed and set in heat mantle. The temperature was set to 80-110⁰C. Each filter paper thimble was changed until it gives colourless solution. Finally extract is obtained by concentrating to one fourth of its original volume under reduced pressure.

Preliminary Phytochemical screening: Phytochemical Screening of the reconstituted extracts thus obtained was done qualitatively for the presence of various phytochemical constituents like Alkaloids, flavonoids, tannins, sterols, and

phenols by using standard phytochemical methods of Swadhini et al, Pathak et al.

Chromatographic Characterization: TLC of Plant extract:

The TLC plates (Pre coated TLC Silica gel 60 F₂₅₄ Aluminium sheets 20X20) were trimmed to strips. The origin was marked as straight line with pencil to spot the Chloroform extract dissolved in chloroform and Petroleum ether, the solvent front also marked by a straight line. Chromatographic chamber contain the solvent mixture of n-Hexane: Ethyl acetate: Glacial acetic acid (8.5:1.5:0.1) at the level of 1cm beneath the origin and allowed to saturate. The chloroform extract was spotted on the origin this strip was put in chromatographic chamber and allowed to run. The solvent travelled up the plate by capillary action till it reached the solvent front. The strip was taken out of the chamber, sprayed with methanol and sulphuric acid spraying reagent after drying the strip. The spots were observed after the incubation of strip in hot air oven. The retention factor (Rf) values of all spots were determined by the following formula:

Retention factor = Distance travelled by the plant extract/ distance travelled by the solvent system.

Results and Discussion

Table-1 represents the presence of Sterols and Terpinoids in Chloroform extract of *Conium maculatum*. There were no Flavanoids, Alkaloids, Iridoits, Tannins and Phenols.

Table-1
Phytochemical Screening of *Conium maculatum*

S. No	Name of the Phytochemicals	Test name	Results
1	Sterols	Liebermann-Burchard test	+Ve
2	Terpinoids	Salkowski test	+Ve
3	Flavanoids	Ferric chloride Shinoda's Lead acetate	-Ve
4	Alkaloids	Mayer's wagner Dragan drop	-Ve
5	Iridoits	Wieffering test:	-Ve
6	Tannins	Ferric chloride test	-Ve
9	Phenol	Elegic acid test	-Ve

In order to obtain some information on the active components present in the crude extract of *Conium maculatum* a important medicinal herb from Jammu and Kashmir, India to characterize it, different types of Chromatographic techniques viz. TLC studies were undertaken.

Discussion: Over the centuries, people have been living in close association with the environment and relying on its flora and fauna as a source of food and medicine. As a result, many societies have their own rich plant pharmacopoeias. In

developing countries, due to economic factors, nearly 80% of the population still depends on the use of plant extracts as a source of medicine. Natural products also play an important role in the health care system in developed countries. Natural products can also be used as starting materials for semi synthetic drugs. The main examples are plant steroids, which led to the manufacture of oral contraceptives and other steroidal hormones. Today, almost every pharmacological class of drugs contains a natural product or natural product analog. The investigation of higher plants has led the discovery of many new drugs.

Table-2
TLC of Chloroform extract of *Conium maculatum*

S. No	Extract dissolved in Solvent	Mobile Phase	No. of Bands	Rf Values
1.	Chloroform	n-Hexane: Ethyl acetate: Glacial acetic Acid (8.5:1.5:0.1)	9	0.18, 0.22, 0.3, 0.34, 0.4, 0.5, 0.6, 0.67, 0.97.
2.	Petroleum ether	n-Hexane: Ethyl acetate: Glacial acetic Acid (8.5:1.5:0.1)	8	0.18, 0.24, 0.4, 0.42, 0.5, 0.62, 0.74, 0.97.

TLC is the simplest and cheapest method for detecting plant constituents because the method is easy to run, reproducible and requires little equipment. However, for efficient separation of metabolites, good selectivity and sensitivity of detection. They play an important role as an analytical supporting the work of phytochemists for the efficient localization and rapid characterization of natural products. Preliminary phytochemistry and TLC chromatogram of the plant extracts showed presence of aromatic compounds. Aromatic compounds such as sterols and Terpinoids posses diverse biological properties such as anti-inflammatory and antispasmodics agent.

Conclusion

Thus it is apparent from the present study that chloroform crude extract of *Conium maculatum* containing sterols and Terpinoids. Chloroform and petroleum ether dissolved extract has given 9 and 8 bands. The crude extract has to be purified and characterized for possible pharmaceutical utilization.

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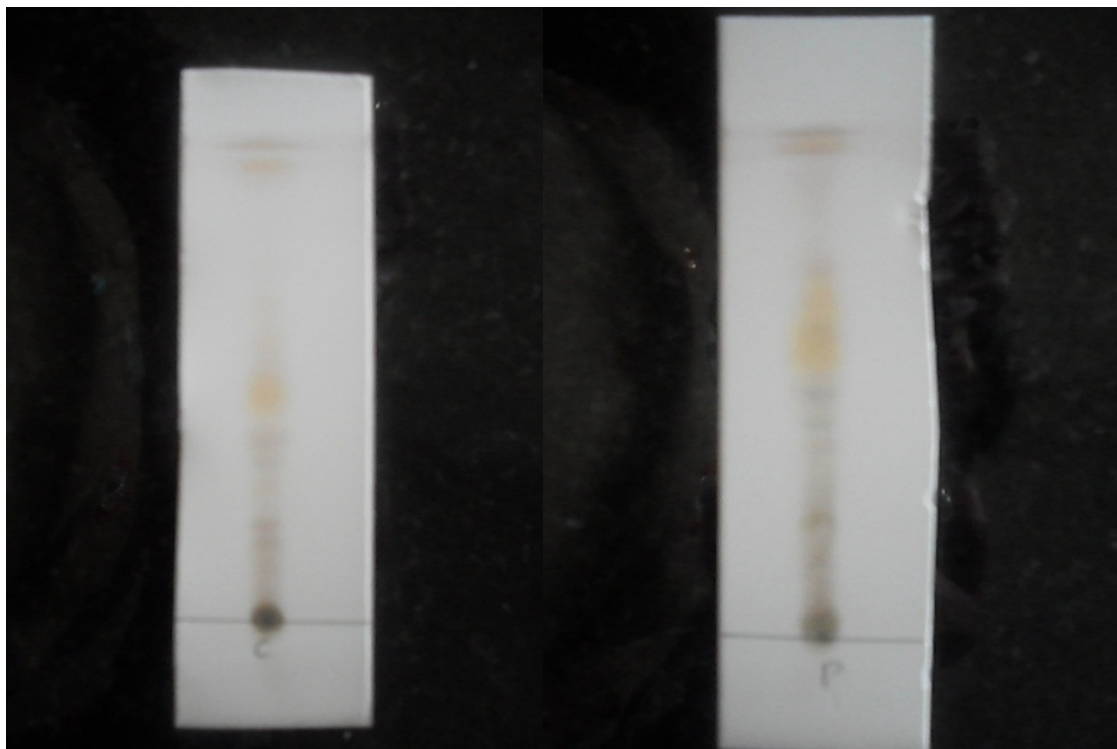


Figure-1
TLC of crude Conium extract dissolved in Chloroform and Petroleum ether

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