

HPTLC FINGER PRINTING OF *Tinospora Cordifolia* AND *Trigonella Foenum Graecum* EXTRACTS

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ABSTRACT

The plates are developed using solvent systems with toluene-ethyl acetate-formic acid (7:3:0.05) for ethanolic extract at different concentration say 10 μ L and 15 μ L in each plate respectively. Then the plates are scanned using camag's scanner at 254 nm & 366 nm. The third plate is derivitized with vanillin sulphuric acid and scanned at 520 nm. The Rf values and peak areas are shown along with finger print chromatogram in Fig.18 (a), (b), (c). Similarly HPTLC plates of *Trigonella foenum graecum* using solvent system with chloroform, methanol and formic acid (8:2:0.5) are used. Its Rf values in different nm are shown in the Fig.19 (a), (b), (c). The contamination or any impurity during plant collection for the preparation of drugs can be identified if there is any change in Rf values, peak areas and no of peaks. These HPTLC chromatograms finger prints are very much needful for authentication of the study plants.

Key Words-: Chromatogram, HPTLC, Ethanolic extract, *Trigonella foenum graecum*, *Tinospora cordifolia*

INTRODUCTION

The plant parts including leaves, roots, rhizomes, stems, barks, flowers, fruits, grains or seeds, employed in the control or treatment of a disease condition and therefore contains chemical components that are medically active. These non-nutrient plant chemical compounds or bioactive components are often referred to as phytochemicals („phyto-„from Greek - phyto meaning „plant“) or phytoconstituents and are responsible for protecting the plant against microbial infections or infestations by pests (Rioset *al.*, 2005, Adekunle and Adekunle , 2009). The phytochemistry is the study of natural products. Phytochemicals have been isolated and characterized from fruits such as grapes and apples, vegetables such as broccoli and onion, spices such as turmeric, beverages such as green tea and red wine, as well as many other

sources. Natural products have been an integral part of the ancient traditional medicine systems, e.g. Chinese, Ayurvedic and Egyptian (Sarker *et al.*, 2007). Over the years they have assumed a very central stage in modern civilization as natural source of chemotherapy as well as amongst scientist in search for alternative sources of drugs. In the developing world about 3.4 billion people depend on plant-based traditional medicines. About 88 per cent of the world's population depends on traditional medicine for their primary health care. According to the World Health Organization, a medicinal plant is any plant which, in one or more of its organs, contains substances that can be used for therapeutic purposes, or which are precursors for chemo-pharmaceutical semi synthesis.

MATERIALS AND METHODS

PLANT COLLECTION AND AUTHENTICATION

Tinospora cordifolia leaves are collected from near Glocal University campus Saharanpur and *Trigonella foenum graecum* seeds are collected from local market, Saharanpur, Uttar Pradesh India.

HPTLC finger printing of *Tinospora cordifolia* and *Trigonella foenum graecum* extracts

HPTLC finger print analysis

The extract of the sample is dissolved with respective solvent ethanol used for extraction. The sample extract (25 μ L) is applied for the thin layer chromatography and High performance thin layer chromatography study with suitable solvent system for *Tinospora cordifolia* Toluene: Ethyl acetate: Formic acid (7:3:0.5) for *Trigonella foenum graecum* chloroform: Methanol: formic acid (8:2: 0.5). After development the plates are allowed to dry in air and examined under UV- 254 nm, 366 nm and visible light after derivatized using vanillin sulphuric acid at 520 nm. R_f value of the spots are recorded. (WHO, 1998; stahl, 1969; Saraswathi 2003; Arunadevi *et al.*, 2015).

RESULTS AND DISCUSSION

One of the important techniques used in phytochemical study is HPTLC finger print analysis for authentication and standardization of the study plants. The plates are developed using solvent systems with toluene-ethyl acetate-formic acid (7:3:0.05) for ethanolic extract at different concentration say 10 μ L and 15 μ L in each plate respectively. Then the plates are scanned using camag's scanner at 254 nm & 366 nm. The third plate is derivitized with vanillin sulphuric acid and scanned at 520 nm. The R_f values and peak areas are shown along with finger print chromatogram in Fig.18 (a), (b), (c). Similarly HPTLC plates of *Trigonella foenum graecum* using solvent system with chloroform, methanol and formic acid (8:2:0.5) are used. Its R_f values in different nm are shown in the Fig.19 (a), (b), (c). The contamination or any impurity during plant collection for the preparation of drugs can be identified if there is any change in R_f values, peak areas and no of peaks. These HPTLC chromatograms finger prints are very much needful for

authentication of the study plants. (Arunadevi et al., 2015)

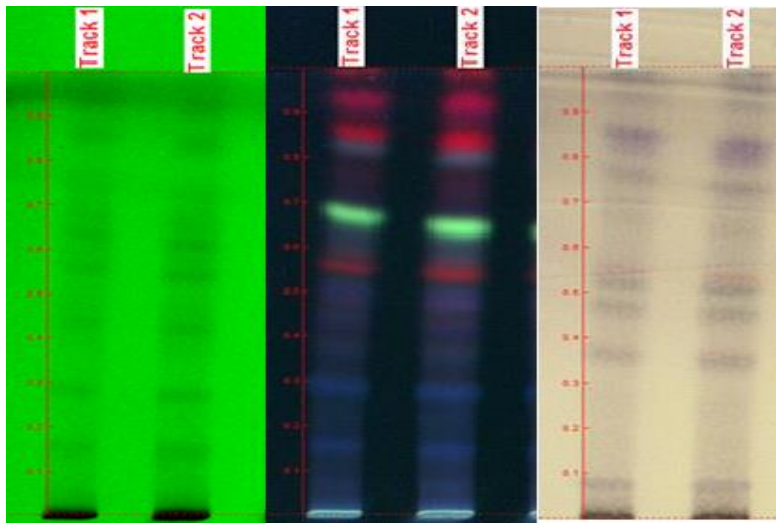


Fig.18 TLC Photo documentation of ethanol extract of *Tinospora cordifolia*

Solvent system: Toluene: Ethyl acetate: Formic acid (7:3:0.5)

Track 1- Sample A ethanol extract- 10µl, Track 2- Sample A ethanol extract- 15µl.

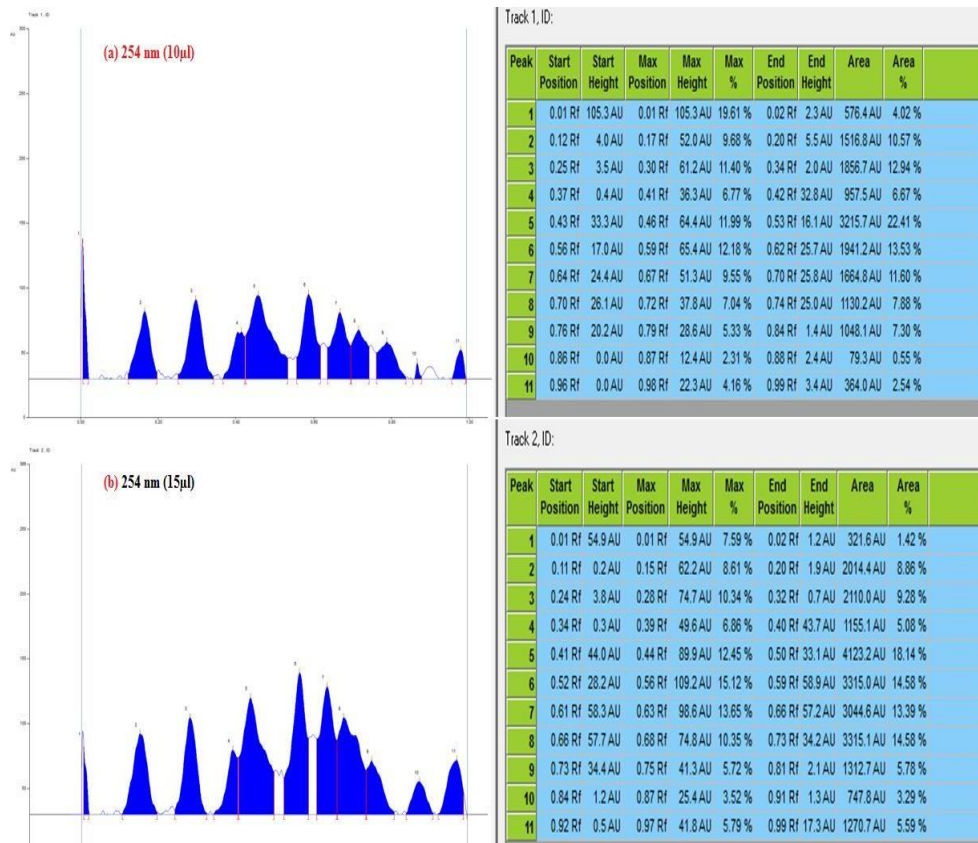


Fig.18(a) HPTLC fingerprint profile of *Tinospora cordifolia* extract scanning at 254 nm (a) Track 1 (10 µl) (b) Track 2 (15 µl)

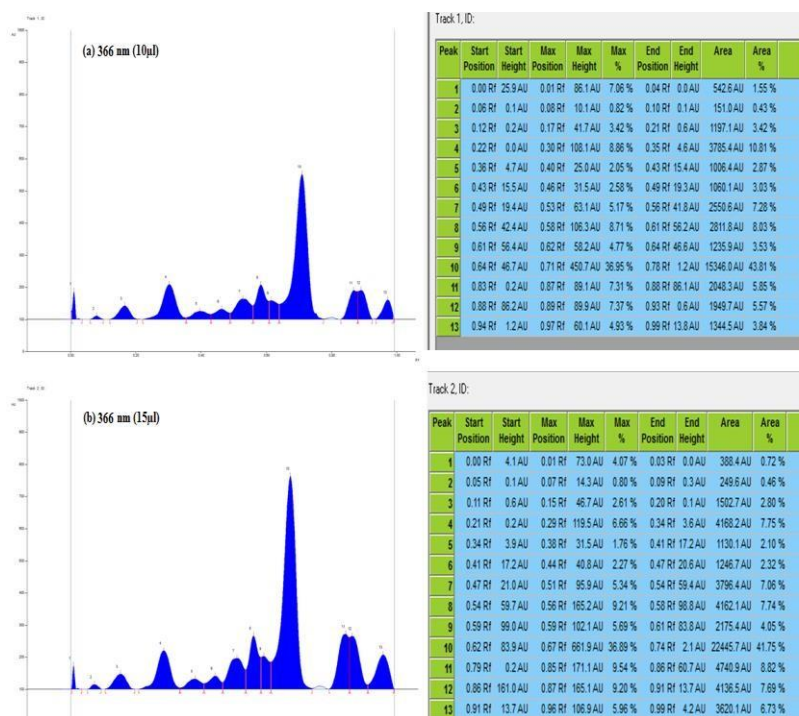


Fig.18(b) HPTLC fingerprint profile of *Tinospora cordifolia* extract scanning at 366 nm (a) Track 1 (10 µl) (b) Track 2 (15 µl)

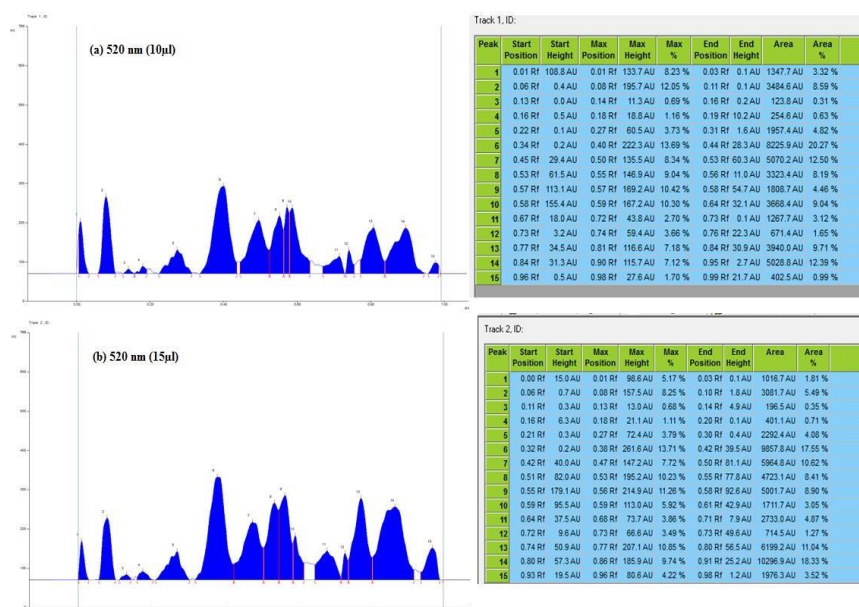


Fig.18.(c) : HPTLC fingerprint profile of *Tinospora cordifolia* extract scanning at 520 nm (a)
Track 1 (10 μ l) (b) Track 2 (15 μ l)

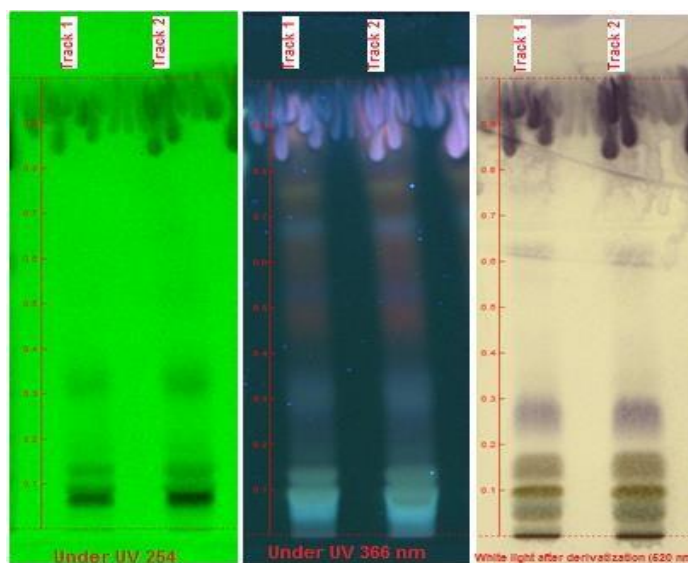
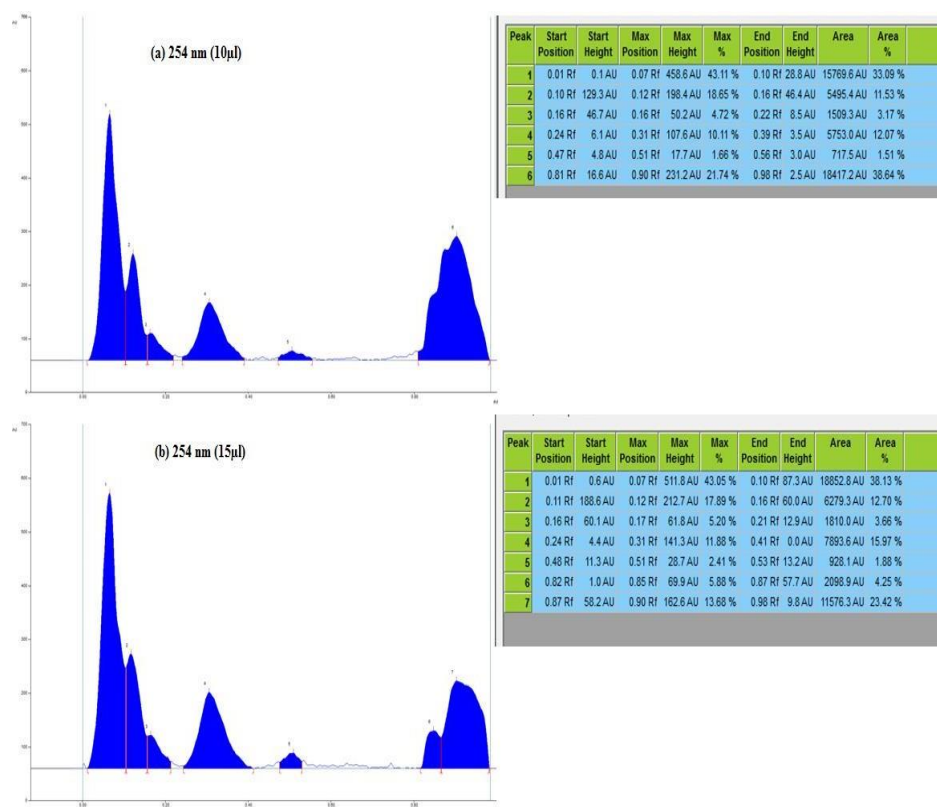


Fig.19 : TLC Photo documentation of ethanol extract of *Trigonella foenum graecum*

Solvent system: Chloroform: Methanol: Formic acid (8:2:0.5)

Track 1- Sample B ethanol extract- 10 μ l, **Track 2-** Sample B ethanol extract- 15 μ l.

Fig.19.a : HPTLC fingerprint profile of *Trigonella foenum graecum* extractscanning at 254 nm (a) Track



1 (10 µl) (b) Track 2 (15 µl)

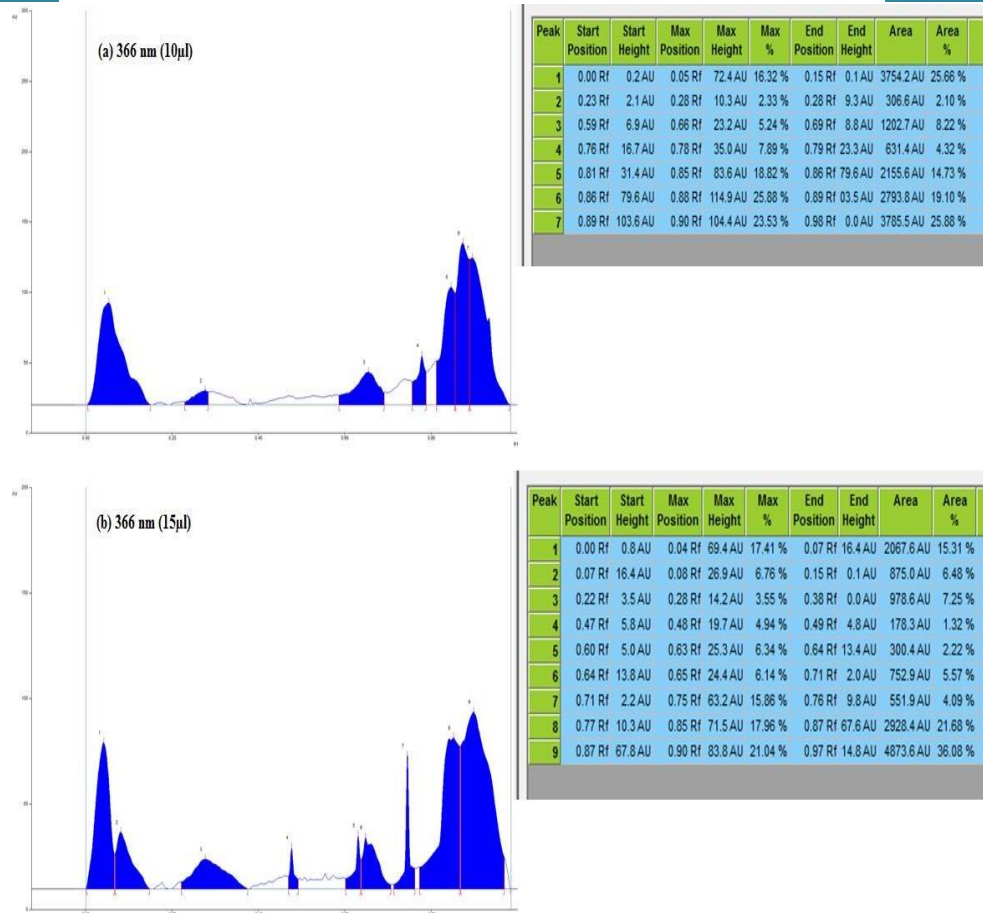
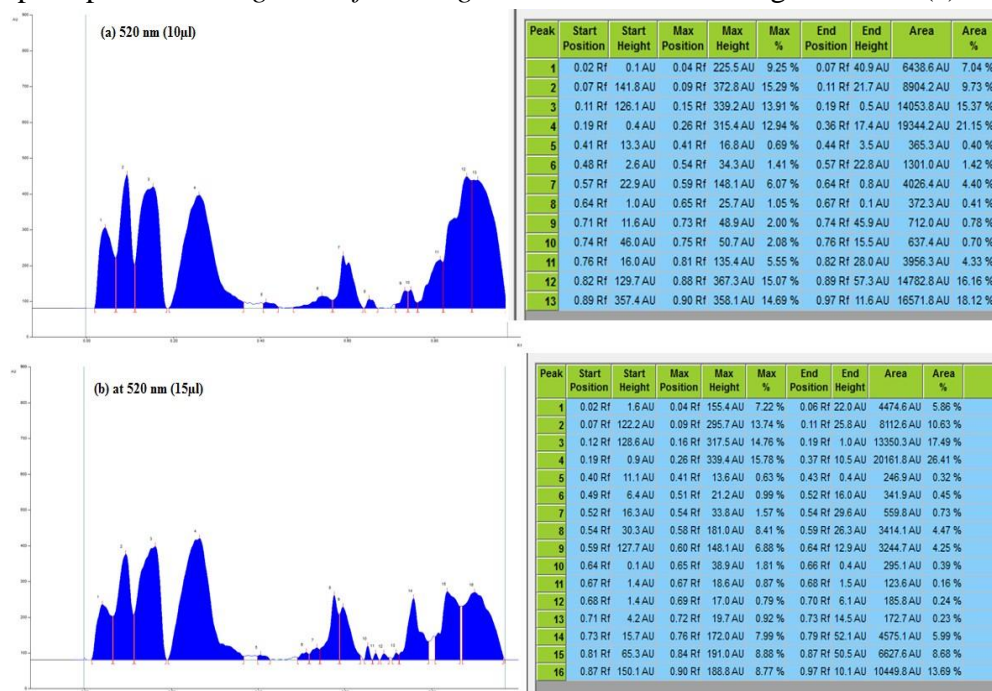


Fig.19.b: HPTLC fingerprint profile of *Trigonella foenum graecum* extract scanning at 366 nm (a) Track 1 (10 µl) (b) Track 2 (15 µl)

Fig.19.c : HPTLC fingerprint profile of *Trigonella foenum graecum* extract scanning at 520 nm (a) Track 1



(10 µl) (b) Track 2 (15 µl)

CONCLUSION

The present study for the two study plants *Tinospora cordifolia* and *Trigonella foenum graecum* which includes and in depth analysis of various physico-phytochemical, qualitative phyto constituents, elements present qualitatively and quantitatively are seen. By using the two study plants extracts individually, the synthesis of silver nano particles and its anti-microbial study are studied. Authentication and standardization of study plants by HPTLC finger print analysis. *In vitro* and *in vivo* screening methods are followed for studying antioxidant potential. By using spectral techniques one or more phytoconstituents are isolated and the structures are elucidated.

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