Plant Archives Vol. 21, Supplement 1, 2021 pp. 2179-2183 e-ISSN:2581-6063 (online), ISSN:0972-5210

Plant Archives

Journal homepage: http://www.plantarchives.org
doi link : https://doi.org/10.51470/PLANTARCHIVES.2021.v21.S1.358

PHYTOCHEMICAL SCREENING AND HPTLC OF SPECIES OF *SOLANUM* (SOLANACEAE)

Anju Kamboj[1] and Sanjana Piplani[2]
[1]Chandigarh College of Pharmacy, Landran, Mohali. India
[2]CT Institute of Pharmaceutical Sciences, Jalandhar and IKGPTU, Kapurthala. India

ABSTRACT

The members of genus *Solanum* possess high economic potential. The current research goals to hold out the phytochemical screening in addition to the HPTLC fingerprint profiling of three species of *Solanum.* Methanolic extract of each plant was subjected to qualitative phytochemical screening. Standard spectrophotometric methods did the quantification of total alkaloids, flavanoids and phenols. HPTLC method was developed to separate the chemical constituents within the extracts and HPTLC of the methanolic extract was carried out on silica gel pre-coated aluminium plates of Merck with the aid of automatic TLC applicator, using the solvent system toluene: ethylacetate: acetic acid: methanol (4.5:1:0.7:0.3). Phytoconstituents like alkaloids, flavanoids, steroids, carbohydrates and glycosides were found to be present after the preliminary phytochemical screening was done.. Amongst all the three extracts studied the alkaloid content was found to be highest in *Solanum indicum* and flavanoid content was found to be highest in *Solanum torvum.* Scanning of HPTLC fingerprint at 254 nm for methanol leaf extracts showed ten peaks for *Solanum indicum*, six peaks for *Solanum surattense,* and ten peaks for *Solanum torvum* having Rf values within range 0.18 to 0.79.

Keywords **:** *Solanum,* High-Performance Thin Layer Chromatography fingerprinting, Phytochemical screening.

Introduction

Aromatic plants and their medicinal constituents endow with raw material in pharmaceutical cosmetics and drug industries (Bhatacharjee, 2008). A wide range of chemical components are found in plants and frequently, biosynthetic plants liberating these compounds differ in different plant groups. Primary metabolites like amino acids, carbohydrates are vital, meant for protecting the living processes, on the other hand secondary metabolites such as alkaloids, phenolics, steroids, terpenoids possess toxicological, pharmacological and ecological significance (Sharma *et al.,* 1996).

Chromatographic methods are meant for the partitioning and purification of phytochemicals based on their charge, form or dimension (Helftmann, 1992). Thin-layer chromatography (TLC) is often regarded as a reliable and reproducible technique for the study of different drugs. This technique is broadly adopted for rapid investigation of drugs and drug preparations. The main function of fingerprinting is to establish the chromatogram of the pharmacologically active extracts. The chromatographic fingerprint approach can be used for the detection and authentication of natural medicines, even if several samples of the drug differ qualitatively and quantitatively. Therefore, it is vitally significant to attain dependable chromatograms that match to therapeutically active and chemical characteristic components of the herbal drug (Patil and Shettigar, 2010; Liang *et al.,* 2014; Ong, 2002; Xie., 2001).

HPTLC is probably the utmost refined type of Thin-Layer Chromatography, which consists of the use of chromatographic technique of extreme parting efficiency including the usage of state-of-the-art instrumentation at all steps within the procedure. These include accurate application of extract, development of uniform and reproducible chromatogram as well as software-controlled assessment. Chief advantage of HPTLC is its way to analyse quite a lot of samples concurrently using a small quantity of mobile phase (Modi *et al.,* 2008).

The biggest amid the angiosperms with grand potential for meal safety on the earth is the genus *Solanum* belonging to the family Solanaceae (Kochhar, 1981; Gnana Sundari *et al.,* 2013). It includes 1200 species and is very famous throughout the arena. It is discovered to be abundant in alkaloids, scattered in all components of the plant (Cronquist, 1981). The major components of pharmaceutical importance, isolated from roots and leaves are solinidine and some steroids (Milner *et al.,* 2011; Simmonds and Choudhury, 1976). Mostly *Solanum* species are used in folk medicines. The economical and medicinal importance of this genus all over the world is because of the existence of steroidal alkaloid solasodine, a vital precursor for the generation of steroid hormones (Silva *et al.,* 2005a; Barbosa-Filho *et al.,* 1991).

Major purpose of present research is to conduct the phytochemical analysis and HPTLC chromatogram development of methanolic leaf extract of three species of *Solanum, viz. Solanum indicum, Solanum surattense and Solanum torvum* that can be utilized for detection, confirmation and characterization.

Experimental

Plant Material

The herbal samples for the planned study have been amassed from Tirumala Hills, Tirupathi District, Andhara Pradesh, India and certified by Dr K Madhava Chetty, Asst. Professor, Dept. of Botany, Sri Venkateswara University, Tirupati, Andhara Pradesh, India.

Plant material Preparation and Extraction

Washed the collected leaves using distilled water, dried in the shade, powdered and stored at 4˚C till further use. Prepared the crude extracts using methanol as solvent. The extraction was done at room temperature. Filtered the extracts through Whatmann No. 1 filter paper and dried by means of rotary vacuum evaporator to give a concentrated extract. The extracts were dried with the help of drier and subjected to analysis.

Phytochemical Screening

Carried out the chemical tests with the crude extracts of each plant.

Qualitative analysis

Qualitative analysis was done to discover the existence of following constituents – alkaloids, flavanoids, phenols, steroids, carbohydrates and glycosides using standard procedures. 5, 6

Quantitative Detection

The coarse powder was then used for quantitative analysis of alkaloids, flavanoids and phenols, after confirming the presence of various phytochemicals.

Determination of total alkaloids

Weighed 5 g of the powder in a 250 ml beaker and added 200 ml, 10% ethanolic acetic acid, covered and set the mixture to macerate for 4 hr. Concentrated the filtrate to one-fourth of the original volume and added concentrated ammonia, dropwise to the extract till complete precipitation. The solution was allowed to settle, precipitates collected, washed using dilute ammonia solution and filtered. The residue was dried and weighed.

Determination of total flavanoids

Extracted 2 g of the powder repetitively using 100 ml of 80% aqueous methanol at room temperature. Filtered the solution by means of Whatmann filter paper no. 42 (125mm). Transferred the filtrate in a crucible and dried till steady weight.

Determination of total phenols

Extracted 100 mg of powder by means of 5 ml of 80% ethanol and centrifuged at 2000 rpm. The supernatant was used for assay. Added 1 ml of folin-ciocalteau reagent to 0.5 ml of supernatant followed by adding 2 ml of dilute sodium carbonate and gently warmed for 1 minute. Made up the volume to 10 ml with distilled water. Prepared the blank solution by mixing all the reagents apart from sample. The absorbance was read at 650 nm in UV spectrophotometer.

HPTLC

The evaluation of herbal drug is an essential part for analysing its identity. The observations of this work thus, serve as a base for proper analysis of the plant. Chromatograms developed were studied by HPTLC.

Preparation and Application of sample

Prepared 5 mg/ml concentration of dried extracts in solvents of particular grade and were filtered using Whatmann filter paper no. 1. The spots were marked on precoated TLC aluminium sheets silica gel 60 F 254 (Merck) 07 µl, with a band length of 5 mm using Linomat 5 sample applicator at speed of 150 nl / sec.

Solvent system

After using the variety of solvent systems, the acceptable resolution was obtained from the solvent toluene : ethyl acetate : acetic acid : methanol (4.5:1:0.7:0.3)

Chromatogram development

Chromatogram development was brought about in twin trough glass chamber using the solvent toluene : ethyl acetate : acetic acid : methanol (4.5:1:0.7:0.3) for 20 minutes at a distance of 80 mm.

Scanning and determination of spots

The air dried TLC plates were observed in UV radiation. Scanning was performed by CAMAG HPTLC Densitometer (Scanner) at 254 and 366 nm using Deuterium lamp and CAMAG winCATS software, at 60% relative humidity.

Results and Discussion

Prelimnary phytochemical screening of the methanolic extracts of leaves of *Solanum indicum, Solanum surattense* and *Solanum torvum* showed the presence of various components. [Table 1] All the three species showed the existence of constituents like alkaloids, flavanoids, steroids, carbohydrates and glycosides.

Quantitative determination of alkaloids, flavanoids and phenolics is carried out to find out the percentage composition of these components in the extracts. The results are tabulated in [Table 2]. Among all the three extracts studied, *Solanum indicum* had the highest concentration of alkaloids and *Solanum torvum* had maximum concentration of flavanoids. The concentration of total phenolics is significantly lesser compared to total alkaloids and flavanoids. Flavanoids can modulate the various enzymatic activities due to their interaction with various biomolecules since they have potent antioxidant properties (19).

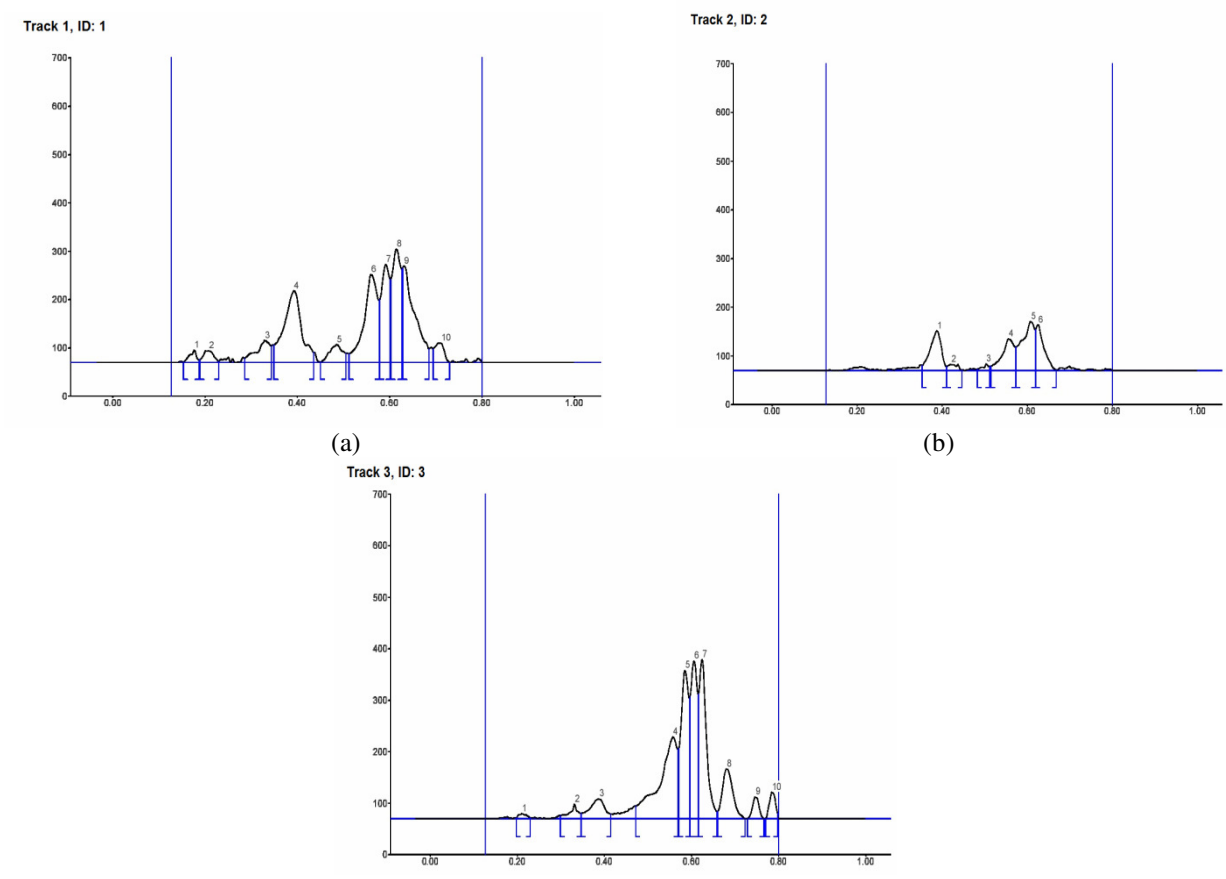
HPTLC Profile

The HPTLC fingerprint study revealed that all the three species of *Solanum* showed finest results in Toluene: Ethyl acetate: Acetic acid: Methanol 4.5: 1: 0.7: 0.3 solvent system for methanolic extracts. Further the HPTLC fingerprints, observed at wavelength 254 nm reported the existence of different polyvalent active constituents. The range for Rf values was found to be present between 0.18 to 0.79 for the three species. Further from the table and the chromatogram it has been found that out of 10 components in *Solanum indicum*, the component with Rf value 0.62 was more predominant as the percentage area is 20.53%. In the same way out of 6 polyvalent compounds in *Solanum surattense* and 10 polyvalent compounds in *Solanum torvum* the compounds with Rf value 0.61 and 0.63 were found to be present in higher content as the percentage area is 27.28%

and 23.25% respectively. TLC plates showed different colored phytoconstituents. HPTLC analysis in addition to giving the idea for the confirmation of the plant extracts and its constituents additionally helps in analysing the parameters for high quality of natural formulations. The members of the genus *Solanum* have valuable medicinal properties and hence possess high economic potential.

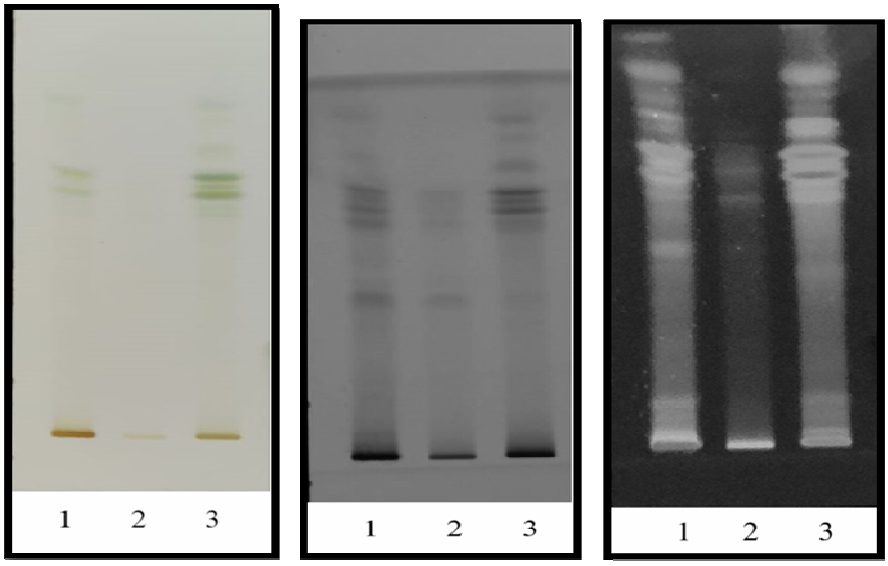
Discussion

HPTLC results explain that the methanolic leaf extract of three *Solanum* species contain a combination of compounds. The fingerprint images obtained can be referred as standard fingerprints for authentication, identification, purification, quality control evaluation as well as to separate the leaves of three species from their adulterants so as to ensure the medicinal efficacy. In future works have to be carried out for the characterization of more phytochemicals along with quantitative estimation with marker compounds. Even if this record can be used along with the previous information for fixing qualities to these plant species.



(a) (b) (c)

Fig. 1: Chromatogram of the methanol extract at 254 nm. (a) *Solanum indicum*; (b) *Solanum surattense*; (c) *Solanum torvum*



(a) **(b)** **(c)**

Fig. 2: High performance thin layer chromatography plate seen at (a) visible light (b) 254 nm (c) 366 nm. Track 1: *Solanum indicum*; Track 2: *Solanum surattense*; Track 3: *Solanum torvum.*

Table 1: Phytochemical composition of the leaves of *Solanum indicum, Solanum surattense and Solanum torvum.*

Name of the plant	Alkaloids	Flavanoids	Phenols	Steroids	Carbohydrates	Glycosides
Solanum indicum	+	+	-	+	+	+
Solanum surattense	+	+	-	+	+	+
Solanum torvum	-	+	-	+	+	+

Table 2: Quantitative analysis for total alkaloids, flavanoids and phenolics.

Plant name	Alkaloids	Flavanoids	Phenolics
Solanum indicum	9.9±0.21	9.63±0.82	1.13±0.57
Solanum surattense	7.22±2.52	9.43±0.82	0.84±0.24
Solanum torvum	2.36±0.22	10.86±1.50	0.824±0.21

Table 3: Data referring to HPTLC fingerprint of methanolic extracts of *Solanum indicum, Solanum surattense* and *Solanum torvum* at 254 nm.

Plant Name	Number of Peaks	Rf value	Area Percentage
Solanum indicum	10	0.18	2.27
		0.21	2.16
		0.33	4.02
		0.39	12.99
		0.49	3.26
		0.56	15.97
		0.59	17.77
		0.62	20.53
		0.63	17.50
		0.71	3.54
Solanum surattense	6	0.39	22.12
		0.42	3.58
		0.50	3.86
		0.56	17.73
		0.61	27.28
		0.63	25.42
Solanum torvum	10	0.21	0.75
		0.33	2.11
		0.39	2.90
		0.56	11.96
		0.59	21.65
		0.61	23.02
		0.63	23.25
		0.68	7.29
		0.75	3.21
		0.79	3.86

References

Barbosa-Filho J.M.; Agra, M.F.; Oliveira, R.A.G.; Paulo, M.Q.; Troling, G.; Cunha, E.V.L.; Ataide, J.R. and Bhattacharyya, J. (1991). Chemical and pharmacological investigation of *Solanum* species of Brazil- a search for solasodine and other potentially useful therapeutic agents. *Mem. I. Oswaldo Cruz.,* 86: 189-191.

Bhatacharjee, S.K. (2008). Handbook of Medicinal Plants. *Jaipur India: Pointer Publishers,* 5: 56-77.

Catapano, A.L. (1997). Antioxidant effect of flavanoids. *Angiology,* 48: 39-44.

Cronquist, A. (1981). An Integrated system of Classification of flowering plants. Columbia University Press, New York. 1262.

Gnana Sundari, S.; Rekha, S. and Parvathi, A. (2013). Phytochemical Evaluation of Three Species of Solanum. *Int. J. Res. Ayurveda Pharm.,* 4(3): 420-425.

Harborne, J.B. (1998). Phytochemical methods- A guide to modern techniques of plant analysis. *Springer Sci. Bus.,* 3: 5-30.

Helftmann, F. (1992). Chromatography: Fundamental and Applications of Chromatographic and Electrophoretic Techniques. *Orinda, USA: Elsevier,* 2: 520-522.

Kochhar, S.L. (1981). A Textbook of Economic Botany. Macmillan Publ. New Delhi.

Liang, Y.Z.; XIE, P. and Chan, K. (2014). Quality control of herbal medicines. *J. Chromatogr. B.,* 812: 53-70.

Milner, S.E.; Brunton, N.P.; Jones, P.W.; O'Brien, N.M.; Collins, S.G. and Maguire, A.R. (2011). Bioactivities of glycoalkaloids and their aglycones from *Solanum* species. *J. Agrie. Food Chem.,* 59: 3454-84.

Modi, K.P.; Patel, N.M. and Goyal, R.K. (2008). Estimation of L-dopa from *Mucuna prurien* LINN and formulations containing M. Pruriens by HPTLC method. *Chem. Pharm. Bull (Tokyo),* 56(3): 357-359.

Ong, E.S. (2002). Chemical assay of glycyrrhizin in medicinal plants by pressurized liquid extraction (PLE) with capillary zone electrophoresis (CZE). *J. Sep. Sci.,* 25: 825-831.

Patil, P.S. and Shettigar, R. (2010). An advancement of analytical techniques in herbal research. *J Adv Sci Res.,* 1: 8-14.

Sharma, R.K.; Bhagwan, D. and Samhita, C. (1996). Chowkhamba Sanskrit series. 2: 17-101.

Silva, T.M.S.; Agra, M.F. and Bhattacharyya, J. (2005b). Studies on the alkaloids of *Solanum* of northeastern Brazil. *Rev. Bras. Farmacogn.,* 15: 292-293.

Silva, T.M.S.; Costa, R.A.; Oliviera, E.J.; Barbosa-Filho, J.M.; Agra, M.F. and Camara, C.A. (2005a). Complete 1H and 13C NMR assignments of isojuripidine from *Solanum asterophorum. Mart J. Braz. Chem. Soc.,* 16(6B): 1467-1471.

Simmonds, N.W. and Choudhury, B. (1976). Egg Plant (*Solenum melongena* L.). Evolution of Crop Plants. Longman Inc. New York.

Tiwari, P.; Kumar, B.; Kaur, N.; Kaur, G. and Kaur, H. (2011). Phytochemical screening and extraction. *A review. Int. Pharm. Sci.,* 1: 98-106.

Wagner, H.; Bladt, S. and Zgainski, E.M. (1984). Plant drug analysis: A Thin Layer Chromatography Atlas. *Munich, Germany: Springer-Verlag,* 5-21.

Xie, P.S. (2001). A feasible strategy for applying chromatography fingerprint to assess quality of Chinese herbal medicine. *Trad. Chin. Drug Res. Clin. Pharm.,* 12: 141-169.