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# Phytochemical Fingerprinting and Quality Profiling of *Gokshuradi* Dispersible Tablet- a Polyherbal Formulation

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**Abstract**

Ayurveda is an emerging field in healthcare. Ayurveda uses herbomineral pharmacological management along with its other non-pharmacological management. With growing demand for Ayurveda in various lifestyle disorders and other conditions, there is also a necessity for development of novel formulations. Fingerprinting of these formulations ensures identification and quantification of marker compounds. This article proposes analysis of one such novel formulation *Gokshuradi* Dispersible Tablet including phytochemical analysis of the drug. Various analytical methods and tools were used for organoleptic, physiochemical and HPTLC fingerprinting of this durg. This study concluded that *Gokshuradi* Dispersible tablet shows bioactive markers to support the theory of its therapeutic benefits. It also demonstrates a unique chromatographic marking which can be used in future to ensure same chemical composition in every batch of drug.

**Key words :** *Gokshuradi* Dispersible Tablet, Ayurveda, Fingerprinting.

1. **A**yurveda is science of holistic approach to health. Acceptance at international platform needs evidence-based research. The demand on global level for Ayurveda has increased in last few years. Along with this, demand in standardised and assurance in quality of herbal formulation has also increased. Ayurveda poly formulation contains many different phytoconstituents in contrast to single drugs. This diversity poses a challenge to consistency and compliance. To surmount this issue, profiling based on chromatographic profiling is recommended.

2.High-Performance Thin Layer Chromatography (HPTLC) is advanced version of Thin- layer chromatography (TLC) which is efficient in quantitative analysis of compounds. HPTLC is used for fingerprint profiling, identification of marker compounds

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and comparative analysis making it highly useful in the quality control of herbal medicines.

3. *Gokshuradi* Dispersible Tablet is a novel polyherbal Ayurvedic formulation composed of *Gokshura* (Tribulus terrestris), *Punarnava* (Boerhavia diffusa), *Varuna* (*Crataeva nurvala*), *Brahmi* (*Bacopa monnieri*), *Jatamansi* (*Nardostachys jatamansi*), and *Shankhapushpi* (*Convolvulus pluricaulis*). These herbs individually possess various pharmacological actions including *mutrala* (diuretic), *shothahara* (anti-inflammatory), *medhya* (neuroprotective), *anulomana, nidrajanan*(sleep-inducing). This study is done to analyse and create an analytical base for its proposed use in essential hypertension. For the preparation of the manuscript relevant literature[1-15] has been consulted.

***Aim:***

To conduct organoleptic, physiochemical analysis and phytochemical fingerprinting of *Gokshuradi* Dispersible Tablet.

1. Plant material and aunthentication –

| Name | Latin name | Parts used | Quantity |
|---|---|---|---|
| *Gokshura* | *Tribulus terrestris* L. | *Panchanga* | 1 part |
| *Punarnava* | *Boerhavia diffusa* L.non.cons | Root | 1 part |
| *Varuna* | *Crataeva nurvula* (Buch-Ham.) | Bark | 1 part |
| *Brahmi* | *Bacopa monnieri* (L.) Pennell. | *Panchanga* | 1 part |
| *Jatamansi* | *Nardostachys jatamansi* (D.Don.)DC. | Rhizome | 1 part |
| *Shankhapushpi* | *Convolvulus pluricaulis* (Forssk) | *Panchanga* | 1 part |

All ingredients of *Gokshuradi* Dispersible Tablet were collected from a local market. The raw material was authenticated and specimen was deposited for drug analysis.

2. Physiochemical evaluation- *Goksh uradi* Dispersible Tablet was evaluated for various parameters including description, colour, odour taste and consisteblity. Tablet weight, limit test for heavy metals was performed, loss of drying, ash value, acid insoluble ash were determined, hardness was measured using hardness tester, dissolution time was recorded.

3. HPTLC analysis – Analysis was performed at Centre of Research for Development, Parul University.

- Test sample solution preparation- 5gm of *Gokshuradi* Dispersible Tablet was accurately weighed and transferred into a glass beaker. 100ml methanol as extraction solvent was added to it. 15 min sonication was performed to make sure that the phytochemicals were

extracted completely. The solution was cooled to room temperature and filteration was done using a simple filter paper. The filtrate was used as test solution for HPTLC analysis.

• HPTLC instruments and conditions- Analysis was performed on 10 × 10 cm thin layer chromatography (TLC) plates coated with 0.2 mm layers of silica gel 60 F254 (Merck). The samples were applied to the plate as 6 mm wide bands by means of a Linomat 5 sample applicator (CAMAG, Switzerland). The analysis was carried out at concentrations of 5.0 µL , 10.0 µL, 15.0 µL and 20.0 µL. It was done under 254 nm and 366 nm visualisations which depict UV-absorbing compounds and fluorescent compounds respectively. The plate was developed at a distance of 8.0 cm with Toluene: Ethyl Acetate :Formic acid (5:4:1 v/v/v) as mobile phase in a CAMAG twin-trough chamber saturated with mobile phase vapor.

• Drying and scanning- The plate was then dried at room temperature for five minutes and was scanned at 254 nm and 366 nm by the use of a CAMAG TLC scanner 3 using win CATS 4 software (CAMAG, Switzerland).

1. Physio-Chemical Parameters of *Gokshuradi* Dispersible Tablet:

| Parameters | Result |
|---|---|
| Description | Tablet |
| Colour | Greyish |
| Odour | Pleasant |
| Taste | Bitter |
| Consistebility | Punched tablet |
| Tablet Weight | 500 mg |
| Limit test for heavy metals | Non -reactive |
| Loss on drying at 110 c | 0.4% |
| Total Ash Value | 6.5% |
| Acid-insoluble ash | 1.4% |
| Hardness | 3 |
| Dissolution time | Less than 1 minute |
| Test for Phytochemicals | Alkaloids, Glycosides and Phenols present |

2. *Phytochemical analysis :*

The results of HPTLC analysis at various concentrations and visualisation are depicted as follows-

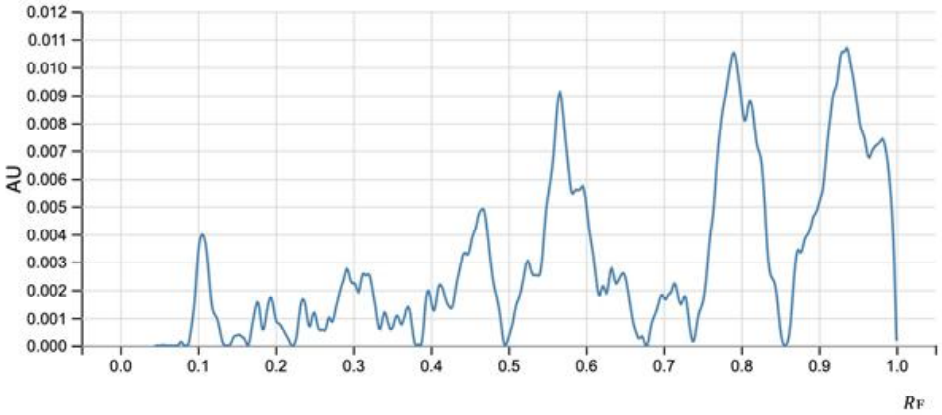


Fig. 1. HPTLC Chromatograph of *Gokshuradi* Dispersible Tablet @254 nm and 5 µL volume (A= Peak height, B= Rf Value and Area percentage) 1(A)

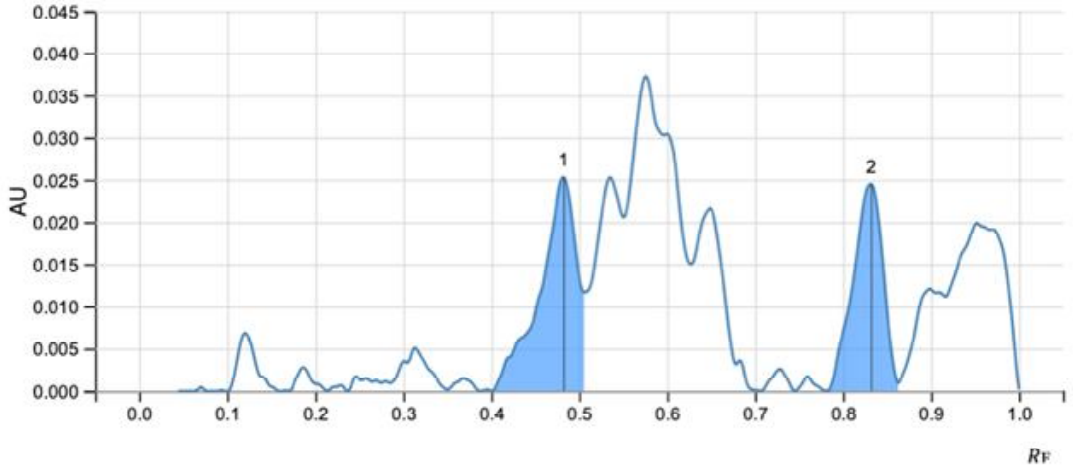


Fig.2. HPTLC Chromatograph of *Gokshuradi* Dispersible Tablet @254 nm and 10 µL volume (A = Peak height, B= Rf Value and Area percentage) 2(A)

2(B)

| Peak # | Start | | Max | | | End | | Area | |
|---|---|---|---|---|---|---|---|---|---|
| | $R_F$ | H | $R_F$ | H | % | $R_F$ | H | A | % |
| 1 | 0.401 | 0.0000 | 0.482 | 0.0253 | 50.83 | 0.507 | 0.0117 | 0.00123 | 55.98 |
| 2 | 0.782 | 0.0000 | 0.832 | 0.0245 | 49.17 | 0.863 | 0.0010 | 0.00097 | 44.02 |

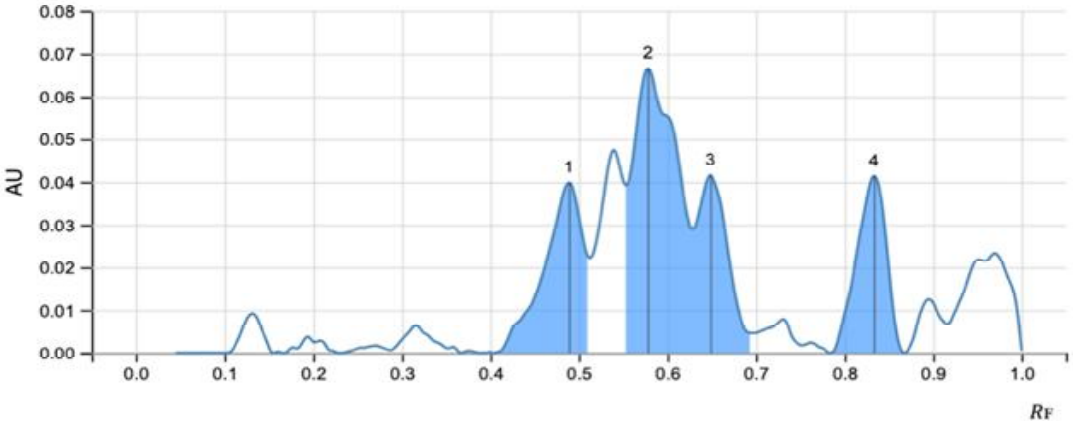


Fig. 3. HPTLC Chromatograph of *Gokshuradi* Dispersible Tablet @254 nm and 15 µL volume (A = Peak height, B= Rf Value and Area percentage) 3(A)

3(B)

| Peak # | Start | | Max | | | End | | Area | |
|---|---|---|---|---|---|---|---|---|---|
| | $R_F$ | H | $R_F$ | H | % | $R_F$ | H | A | % |
| 1 | 0.404 | 0.0000 | 0.489 | 0.0398 | 21.04 | 0.511 | 0.0224 | 0.00201 | 21.93 |
| 2 | 0.553 | 0.0393 | 0.578 | 0.0666 | 35.17 | 0.628 | 0.0289 | 0.00384 | 41.87 |
| 3 | 0.628 | 0.0289 | 0.649 | 0.0415 | 21.94 | 0.694 | 0.0046 | 0.00171 | 18.65 |
| 4 | 0.783 | 0.0000 | 0.833 | 0.0414 | 21.85 | 0.867 | 0.0000 | 0.00161 | 17.55 |

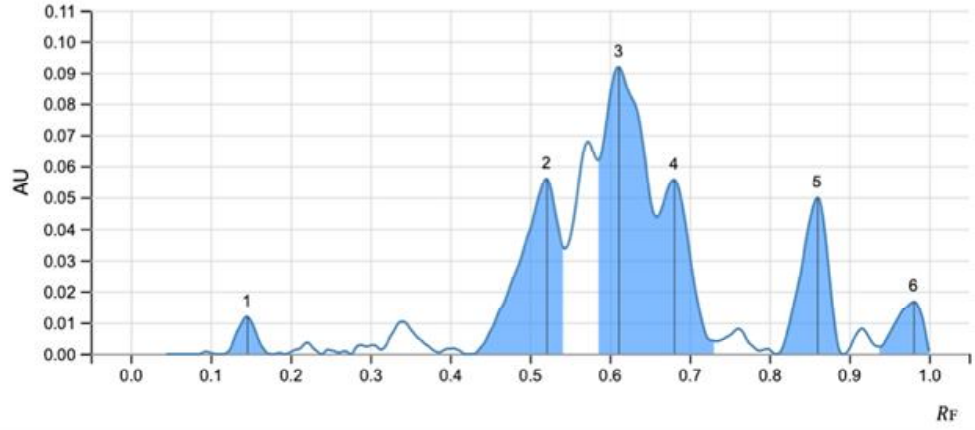


Fig.4. HPTLC Chromatograph of *Gokshuradi* Dispersible Tablet @254 nm and 20 µL volume (A = Peak height, B= Rf Value and Area percentage) 4(A)

4(B)

| Peak # | Start | | Max | | | End | | Area | |
|---|---|---|---|---|---|---|---|---|---|
| | $R_F$ | H | $R_F$ | H | % | $R_F$ | H | A | % |
| 1 | 0.117 | 0.0000 | 0.146 | 0.0118 | 4.20 | 0.175 | 0.0000 | 0.00032 | 2.29 |
| 2 | 0.429 | 0.0000 | 0.521 | 0.0560 | 19.87 | 0.543 | 0.0336 | 0.00318 | 23.01 |
| 3 | 0.586 | 0.0619 | 0.611 | 0.0918 | 32.59 | 0.658 | 0.0437 | 0.00534 | 38.61 |
| 4 | 0.658 | 0.0437 | 0.681 | 0.0557 | 19.76 | 0.733 | 0.0040 | 0.00244 | 17.63 |
| 5 | 0.810 | 0.0000 | 0.860 | 0.0498 | 17.68 | 0.892 | 0.0000 | 0.00191 | 13.79 |
| 6 | 0.938 | 0.0023 | 0.981 | 0.0167 | 5.91 | 1.000 | 0.0008 | 0.00065 | 4.67 |

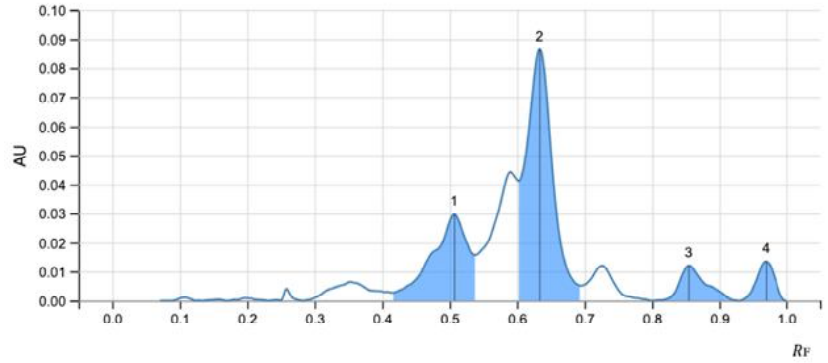


Fig. 5. HPTLC Chromatograph of *Gokshuradi* Dispersible Tablet @366 nm and 5 µL volume (A = Peak height, B= Rf Value and Area percentage) 5(A)

5(B)

| Peak # | Start | | Max | | | End | | Area | |
|---|---|---|---|---|---|---|---|---|---|
| | $R_F$ | H | $R_F$ | H | % | $R_F$ | H | A | % |
| 1 | 0.415 | 0.0027 | 0.507 | 0.0296 | 20.99 | 0.537 | 0.0156 | 0.00184 | 27.54 |
| 2 | 0.603 | 0.0413 | 0.633 | 0.0863 | 61.11 | 0.694 | 0.0051 | 0.00389 | 58.31 |
| 3 | 0.807 | 0.0002 | 0.854 | 0.0119 | 8.42 | 0.928 | 0.0000 | 0.00053 | 7.98 |
| 4 | 0.931 | 0.0000 | 0.969 | 0.0134 | 9.47 | 1.000 | 0.0000 | 0.00041 | 6.16 |

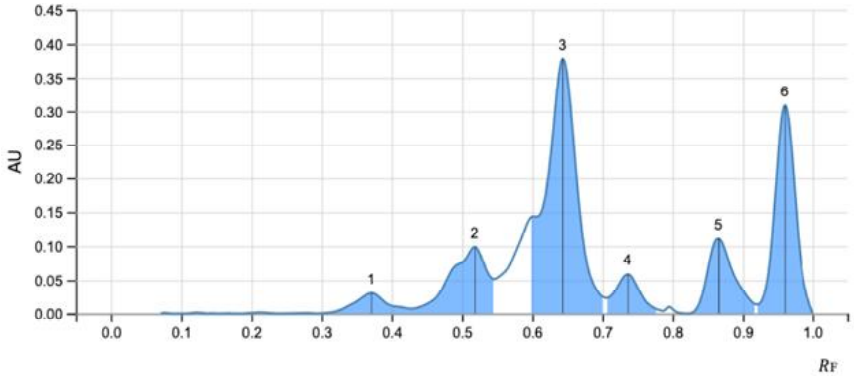


Fig.6. HPTLC Chromatograph of *Gokshuradi* Dispersible Tablet @366 nm and 10 µL volume (A = Peak height, B= Rf Value and Area percentage) 6(A)

6(B)

| Peak # | Start | | Max | | | End | | Area | |
|---|---|---|---|---|---|---|---|---|---|
| | $R_F$ | H | $R_F$ | H | % | $R_F$ | H | A | % |
| 1 | 0.294 | 0.0000 | 0.371 | 0.0305 | 3.09 | 0.426 | 0.0069 | 0.00169 | 3.77 |
| 2 | 0.428 | 0.0069 | 0.518 | 0.0988 | 10.02 | 0.546 | 0.0517 | 0.00609 | 13.63 |
| 3 | 0.599 | 0.1428 | 0.643 | 0.3780 | 38.33 | 0.704 | 0.0229 | 0.01907 | 42.69 |
| 4 | 0.706 | 0.0227 | 0.736 | 0.0590 | 5.99 | 0.785 | 0.0037 | 0.00246 | 5.51 |
| 5 | 0.822 | 0.0000 | 0.865 | 0.1113 | 11.29 | 0.919 | 0.0130 | 0.00490 | 10.97 |
| 6 | 0.921 | 0.0129 | 0.960 | 0.3086 | 31.29 | 1.000 | 0.0003 | 0.01047 | 23.42 |

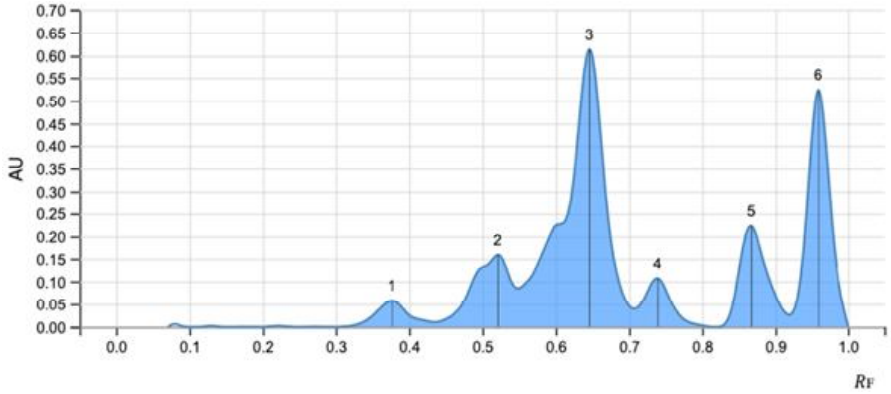


Fig. 7. HPTLC Chromatograph of *Gokshuradi* Dispersible Tablet @366 nm and 15 µL volume (A = Peak height, B= Rf Value and Area percentage) 7(A)

7(B)

| Peak # | Start | | Max | | | End | | Area | |
|---|---|---|---|---|---|---|---|---|---|
| | $R_F$ | H | $R_F$ | H | % | $R_F$ | H | A | % |
| 1 | 0.299 | 0.0000 | 0.376 | 0.0579 | 3.44 | 0.435 | 0.0105 | 0.00309 | 3.61 |
| 2 | 0.435 | 0.0105 | 0.521 | 0.1590 | 9.44 | 0.549 | 0.0847 | 0.00983 | 11.49 |
| 3 | 0.549 | 0.0847 | 0.646 | 0.6135 | 36.42 | 0.707 | 0.0398 | 0.03931 | 45.93 |
| 4 | 0.707 | 0.0398 | 0.739 | 0.1068 | 6.34 | 0.814 | 0.0000 | 0.00468 | 5.47 |
| 5 | 0.821 | 0.0000 | 0.867 | 0.2236 | 13.27 | 0.918 | 0.0263 | 0.00986 | 11.52 |
| 6 | 0.918 | 0.0263 | 0.958 | 0.5236 | 31.09 | 1.000 | 0.0006 | 0.01882 | 21.99 |

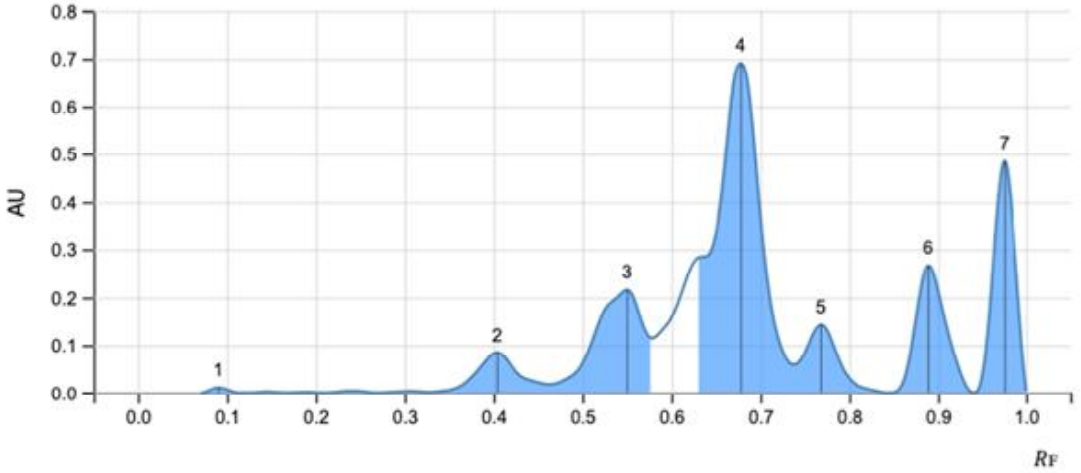


Fig.8. HPTLC Chromatograph of *Gokshuradi* Dispersible Tablet @366 nm and 20 µL volume (A = Peak height, B= Rf Value and Area percentage) 8(A)

**8(B)**

| Peak # | Start | | Max | | | End | | Area | |
|---|---|---|---|---|---|---|---|---|---|
| | $R_F$ | H | $R_F$ | H | % | $R_F$ | H | A | % |
| 1 | 0.072 | 0.0000 | 0.090 | 0.0110 | 0.58 | 0.114 | 0.0000 | 0.00023 | 0.26 |
| 2 | 0.326 | 0.0012 | 0.404 | 0.0846 | 4.46 | 0.463 | 0.0179 | 0.00470 | 5.32 |
| 3 | 0.463 | 0.0179 | 0.550 | 0.2156 | 11.37 | 0.578 | 0.1150 | 0.01366 | 15.47 |
| 4 | 0.631 | 0.2830 | 0.678 | 0.6907 | 36.42 | 0.738 | 0.0593 | 0.03921 | 44.43 |
| 5 | 0.738 | 0.0593 | 0.768 | 0.1433 | 7.56 | 0.842 | 0.0000 | 0.00631 | 7.15 |
| 6 | 0.849 | 0.0000 | 0.889 | 0.2659 | 14.02 | 0.939 | 0.0000 | 0.01037 | 11.75 |
| 7 | 0.942 | 0.0000 | 0.975 | 0.4852 | 25.59 | 1.000 | 0.0034 | 0.01378 | 15.61 |

*Interpretation :*

| Rf Range (from HPTLC) | Peak Area % (Interpretation) | Probable Phytochemical Class | Likely Plant Source(s) | Probable Pharmacological Role in Essential Hypertension | Citations |
|---|---|---|---|---|---|
| 0.11–0.20 | 0.17 (2.29%)→ Minor peak indicating trace hydrophilic compounds | Simple phenolics, sugars | *Gokshura* (fruit), *Punarnava* (root) | Mild diuresis ↓ plasma volume & preload → ↓BP | [4,13] |
| 0.42–0.55 | Major peak cluster: 0.50 (55.98%), 0.51 (21.93%), 0.54 (23.01%), 0.537 (27.54%), 0.546–0.549 (11–13%) → Dominant phytochemical zone | Saponins (protodioscin), flavonoids, phytosterols | *Gokshura, Punarnava, Varuna* | Strong diuretic activity, ACE-inhibition, smooth muscle relaxation, ↓ peripheral resistance | [1-3,10] |
| 0.62–0.73 | 0.62 (41.87%), 0.65–0.73 (17–38%) → Moderate–high intensity | Flavonoids (boeravinones), alkaloids | *Punarnava, Shankha-pushpi* | Renal vasodilation, ↓ edema, ↓ angiotensin signaling, antioxidant vascular protection | [7,8,14] |
| 0.69–0.71 | 0.69 (18.65%); 0.704–0.707 (42–45%) → Brahmi-dominant zone | Bacosides (saponins), phenolic glycosides | *Brahmi* (Bacopa) | ↓ sympathetic drive, anxiolytic, neurocalming → ↓ stress-induced hypertension | [9,12] |
| 0.78–0.85 | 0.785 (5.51%), 0.814 (5.47%), 0.842 (7.15%) → Low–moderate | Terpenoids, alkaloids, coumarins | *Jatamansi, Shankha-pushpi, Varuna* | CNS calming, ↓ heart rate, ↓ vascular tone | [5,6,15] |

| | | | | | |
|---|---|---|---|---|---|
| 0.86–0.90 | 0.86 (17.55–44%), 0.89 (13.79%), 0.918–0.928 (8–11%) → Strong antio-xidant band | Flavonoids, lignans, phenolics | *Punarnava, Gokshura, Brahmi* | ↑ NO availability, ↓ vascular inflammation, improved endothelial function | [8,9] |
| 0.93–1.00 | 1.00 (4–23%), 0.939 (11.75%), 1.00 (15–22%) → High lipophilic band | Terpenoids (jatamansone), lipophilic phenolics | *Jatamansi, Brahmi, Shankha-pushpi* | Sedative, anxiolytic, improved sleep → ↓ sympathetic overdrive → ↓ BP | [6,11] |

• The HPTLC fingerprinting of *Gokshuradi* Dispersible Tablet revealed characteristic of multi-component profile which indicates its polyherbal composition. The chromatographic separation under the mobile phase Toluene: Ethyl acetate: Formic acid (5:4:1 v/v/v) produced bands across different Rf values under both 254 nm (UV absorbance) and 366 nm (fluorescence) wavelengths.

• Major peaks were seen at Rf 0.50-0.54, 0.62-0.69, and 0.78-0.86, representing dominant absorbance zones. The peak at Rf ~0.50 showed highest area contribution (55.98%), indictating possible presence of saponins, flavonoids, and phytosterols. Additional bands at Rf 0.62-0.73 represented flavonoids and alkaloids, while significant intensities at higher Rf values (0.86-1.00) indicated presence of lipophilic compounds like terpenoids and lipophilic phenolics .

• Strong fluorescent bands were noted at Rf 0.537, 0.694, 0.704-0.707, 0.918-0.928, and 1.00. The peak at Rf ~0.70 had highest area contribution (42-58%), indicating presence of bacosides and boeravinones, is an important bioactive markers of *Brahmi* and *Punarnava*.

• The HPTLC fingerprinting of *Gokshuradi* dispersible tablet provided a comprehensive representation of its phytochemical complexity. The chromatography displayed many well-defined peaks across the Rf range of 0.11 to 1.00, which indicates the presence of multiple phytoconstituents such as saponins, flavonoids, phenolics, terpenoids, and glycosides. These are biomarkers sourced from *Gokshura, Punarnava, Brahmi, Jatamansi, Varuna,* and *Shankha-pushpi*.

• The batch-to-batch uniformity was confirmed by the reproducibility of Rf values and peak intensity across different tracks, demonstrating formulation stability. Uniform peak appearance in repeated runs under same chromatographic conditions shows good manufacturing control, phytochemical preservation and suitability for clinical application.

• The HPTLC fingerprinting of *Gokshuradi* Dispersible Tablet showed its phytochemical

complexity, authenticity, and formulation quality. Multiple prominent peaks with consistent Rf values were observed across various concentrations, which highlighted the presence of characteristic phytoconstituents such as saponins, flavonoids, phenolics, bacosides, and terpenoids. Repeated peaks with uniform intensity highlights strong formulation consistency and confirms that the manufacturing process maintains chemical integrity and reproducibility.

• The results not only established a new chromatographic identity for the formulation but also the presence of bioactive markers responsible for its therapeutic activities, supporting its antihy pertensive potential. This provides a scientific basis for its application in evidence-based Ayurveda. This study also establishes a unique chromatographic signature which can be used in future to ensure same chemical composition in every batch of drug.

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