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RESEARCH ARTICLE

# DEVELOPMENT OF HPTLC FINGERPRINTS OF FLAVANOIDS USING QUERCETIN AND BAICALEIN AS MARKERS IN *OROXYLUM INDICUM* (L.) VENT

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**ABSTRACT**

The study was aimed to develop the HPTLC fingerprints of Flavanoids using Quercetin and Baicalein as markers for leaf , stem and roots of the medicinally important plant *Oroxylum indicum* (L.) Vent collected from two different geographical regions of India viz., Western Ghats and North East. Preliminary phytochemical screening was done followed by extraction of flavanoids and then by HPTLC studies. Chloroform: methanol: formic acid (8.8:0.7:0.5) was used as mobile phase along with anisaldehyde $H_2SO_4$ as derivatizing agent for the separation of flavanoids. Quercetin and Baicalein were used as a marker compounds and were detected in all the plant parts collected from both the regions. A good seperation of flavanoids was observed at 540 nm after derivatisation. The study revealed diverse forms of flavanoids in large number in the root stem and leaf of *Oroxylum indicum* (L.) Vent. from both the regions. It can be concluded that HPTLC fingerprint analysis of root, stem and leaf extract of *Oroxylum indicum* (L.) Vent can be used as a diagnostic tool for the correct identification of the plant and it is useful as a phytochemical marker. These separated flavanoids in addition of being developed directly as drugs can also served as prototype drug molecules known as "Lead Compounds" and as pharmacological probes to help better understand biochemical and physiological mechanisms.

**Key words:** *Oroxylum indicum* (L.) Vent, flavanoids, HPTLC fingerprint

## INTRODUCTION:

Chemical fingerprints obtained by chromatographic techniques are strongly recommended for the purpose of quality control of herbal medicines since they might represent appropriately the "chemical integrities" of herbal medicines and therefore be used for authentication and identification of herbal products. The chemical fingerprint of a plant material tends to focus on identification of the stability of the chemical constituents which can used to ensure that the presence of the chemical marker in the newly collected plant material facilitates the manufacturer in confirming the identity of the plant material. A chromatographic fingerprint of a plant material is a chromatographic pattern of pharmacologically active and or chemically characteristic constituents present. HPTLC is the term for a method that meets all quality requirements of today's analytical labs even in a fully regulated environment.

Medicinal plants constitute the main source of new pharmaceuticals and healthcare products[1] . Phytochemical screening of plants has revealed the presence of numerous chemicals including alkaloids, tannins, flavonoids, steroids, glycosides, saponins etc. However, the complexity of composition of the herbal extracts in contrast to synthetic chemical drug compounds is a challenge for analytical methods used for quality assurance. The effect of natural product depends on the concentration of the active ingredients that is affected by a large number of variables like climate conditions, soil composition, time of harvesting and storage conditions. Therefore, a good quality control should confirm that the composition of a herb is exactly as expected. For example, herbal extracts can be manipulated by conscious adulterations (e.g. use of a less expensive plant) or use of similar looking plant inadvertently.

*Oroxylum indicum*(L.) Vent is an important herb in Ayurvedic medicine and indigenous medical system for over thousands of years. Every part of this tree possesses medicinal value. *Oroxylum indicum* (L.) Vent has been used as a single drug or as a component of well known Ayurvedic formulations like dashmularista, Shyonakapatpak and Bruhatpanchamulayadikwath, Dashmula, Narayan taila, Bhrama Rasayna, Dhanwatara and Chyawanprash.[2-4]



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Root bark of the plant is an astringent used in diarrhea, dysentery, rheumatism and as it contains ellagic acid[5] . Stem bark past contains oroxylin A, chrysin, tannic acid, scutellarein-7-rutinoside and is applied for the cure of scabies and to treat arthritis. Leaves contain baicalein-7-glucuronide. All plant parts are a good source of chrysin[6]. Leaf decoction is given in treating stomachache, rheumatism, enlarged spleen[7]. In Burma, Vietnam and Philippines, the bark is used to treat dysentery and rheumatism.[8]

Despite the extensive traditional use of Oroxylum species, it is remarkable that Oroxylum species has never been the subject of extensive HPTLC studies. Claims of the efficacy of *Oroxylum indicum* (L.) Vent in its traditional usage therefore requires validation and accurate documentation. For this purpose, such studies were initiated as a basis for scientific verification for identification of the plant *Oroxylum indicum* (L.) Vent .

## MATERIALS AND METHODS

### Selection and authentication of plant

Whole plant parts of *Oroxylum indicum* (L.) Vent*.,* were collected from two different geographical regions, Western Ghats (Village, Pophli, kumbharli Ghats, near Chiplun, district Raigarh, Maharashtra) and another from Dispur, Assam, North east part of India. The plants were identified and authenticated at Blatter's Herbarium, St. Xavier's College, Mumbai (Authentication no 54436). The plants collected from different regions were sorted out and individual plant parts were separated. The plant parts were then washed and the leaves and other parts were separated from other parts, cleaned and dried for further use.

### Test for flavonoids

(i) Water extract of the sample was reduced to dryness on the boiling water bath. The residue was treated with dil. NaOH, followed by addition of dilute HCl, solubility and colour were noted. A yellow solution with NaOH, which turns colourless with dil HCl confirms flavonoids.

(ii) To dry extract , add 5 ml of 95 % ethanol, few drops of hydrochloric acid and 0.5 g of magnesium turnings . A pink colour formation indicates the presence of flavonoids[9].

### Establishment of profile of flavanoids by HPTLC

One gram powdered material was extracted with 10 mL methanol on water bath (60 °C / 5 min). The filtrate was condensed by evaporation, a mixture of water and ethyl acetate (10:1 mL) was added and mixed thoroughly. The ethyl acetate phase thus retained was used for chromatography. The flavanoids spots were separated using Chloroform: methanol: formic acid (8.8:0.7:0.5) solvent mixture and anisaldeyde $H_2SO_4$ as derivatizing reagent. The colour and Rf values of these spots were recorded under ultraviolet (254 nm and 366 nm) and visible light (540 nm) [9].

### Preparation of standard solutions of Quercetin and Baicalein

Quercetin and Baicalein (purity 98%), was purchased from Sigma-Aldrich Chemie GmbH (Aldrich, Division, Steinbeim, Germany). Prepration of Standard stock solutions concentration (2.0µg/µl) was prepared in 10 mL standard volumetric flask, by dissolving 20.0 mg of accurately weighed Quercetin and Baicalein, in about 5.0 mL of methanol, followed by vortex and finally making up the volume of solution to 10.0 mL, with methanol. 5.0µg/µl was prepared in 10 mL standard volumetric flask, by dissolving 50.0 mg of accurately weighed Quercetin and Baicalein , in about 5.0 mL of methanol, followed by vortex and finally making up the volume of solution to 10.0 mL The two concentrations thus prepared were used for identification of Flavonoids[9-10].

### Chromatographic conditions:

The HPTLC analyses were performed on aluminum plates pre-coated with silica gel 60F254 (Merk, Germany). 10 µl of each extract were applied on the plate of 10 X 10 cm by HPTLC as bands of 10 mm width of each with the help of CAMAG linomat IV sample applicator. The plates were developed in a CAMAG twin- trough chamber previously equilibrated with a mobile phase for 20 minutes. Different solvent systems were used to develop HPTLC fingerprint profile for different secondary metabolite groups separately.[10-11] Each plate were developed up to 8 cm, air dried and scanned at wavelength of 254 & 366 nm using CAMAG TLC Scanner 3. The chromatograms were recorded. Then the plates were derivatized with respective chemical reagents and heated at 105 $^0$C on hot plate till the development of colour of bands and observed under white light. The colour of recorded bands and Rf values were recorded.

## RESULT AND DISCUSSION

Phytochemical examination of extracts of leaf, stem and root of the plant *Oroxylum indicum* (L.) Vent collected from two different regions revealed the presence of Flavanoids. Different compositions of the mobile phase for HPTLC analysis were tested in order to obtain high resolution and reproducible peaks. The desired aim was achieved using chloroform: methanol: formic acid (8.8:0.7:0.5) as mobile phase. Derivatizing agent - Anisaldehyde $H_2SO_4$ was found to be the best. The HPTLC fingerprint of flavanoids which was developed for different plant parts collected from two different geographical regions along with standard Querecetin and Baicalein as markers is seen in Plate no. 1, 2 and 3. The densitogram and corresponding Rf values at 254nm, 366nm and 540nm have been exhibited in figure 1-12.



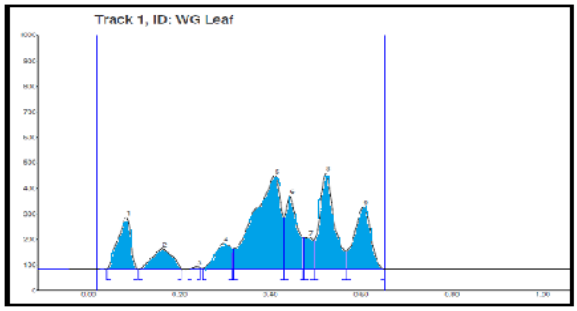
The 3D display of HPTLC chromatogram of flavanoids is seen in Figure 13-15. The variation in flavanoids composition from different plant parts is seen in Table no. 1-3.

The pigments that colour most flowers, fruits and seeds are flavanoids. More than 6000 different flavanoids have been identified and surely this number will increase [12]. The different flavanoids have diverse biological functions, including protection against UV radiation and phytopathogens, signaling during nodulation, make fertility, auxin transport as well as the coloration of flowers as a visual) signal that attracts pollinators[13--15].

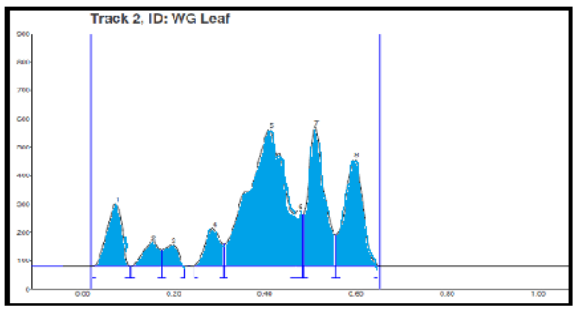
A good separation of Flavanoids has been observed. The anti oxidant activity of phenolics and polyphenolics is often regarded to be the basis of health promoting activity. Plants containing flavonoids and anthraquinones (such as quercetin and emodin) are good antibacterial agents against many human pathogenic bacteria such as Escherichia coli, Streptococcus sp, Staplylococcus aureus and Pseudomonas aeruginosa. Also flavanoids are reported to have anti oxidant anticancer anti allergic, antiinflamatory, anti carcinogenic and gastroprotective properties[16-19]. Flavanoids possess extraordinary antioxidant activity and estrogenic, antiviral, antibacterial and anticancer activities. The putative health protecting functions of flavanoids have stimulated significant research towards flavanoids.

Recent interest by the scientific community in the types and levels of flavanoids in food centers on the antiodicative[18], antimicrobial[20], and anti-inflammatory[21-22] properties which may have possible cardio protective and/or anticarcinogenic effects associated with certain of them. It was observed reduced risk of coronary heart disease (CHD) with high intakes of flavanoids[23]. Flavonoids also exert the effects of anti-oxidants, free radical scavengers and as chelators of divalent cations[24].

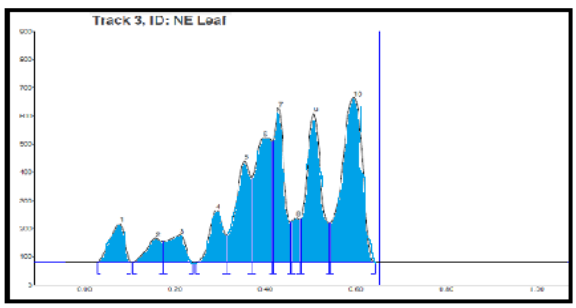
**Figure 1: Densitogram and corresponding Rf values of Flavanoids of leaves of *Oroxylum indicum* (L.) Vent from two regions (Western Ghats and North East) at wavelength 254 nm**




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.04 | 1.1 | 0.08 | 207.0 | 11.35 | 0.11 | 1.3 | 4936.8 | 8.74 |
| 2 | 0.11 | 0.5 | 0.16 | 83.2 | 4.56 | 0.21 | 1.1 | 3115.9 | 5.52 |
| 3 | 0.22 | 0.3 | 0.24 | 12.7 | 0.69 | 0.25 | 8.2 | 172.6 | 0.31 |
| 4 | 0.25 | 6.6 | 0.30 | 104.5 | 5.73 | 0.32 | 80.5 | 3090.5 | 5.47 |
| 5 | 0.32 | 80.8 | 0.41 | 368.6 | 20.21 | 0.43 | 204.2 | 18500.6 | 32.77 |
| 6 | 0.43 | 206.3 | 0.44 | 291.7 | 15.99 | 0.47 | 123.6 | 6607.7 | 11.70 |
| 7 | 0.48 | 123.9 | 0.49 | 127.7 | 7.00 | 0.50 | 115.6 | 1975.5 | 3.50 |
| 8 | 0.50 | 116.6 | 0.52 | 381.0 | 20.89 | 0.57 | 72.8 | 10450.2 | 18.51 |
| 9 | 0.57 | 73.4 | 0.61 | 247.5 | 13.57 | 0.65 | 1.0 | 7612.3 | 13.48 |



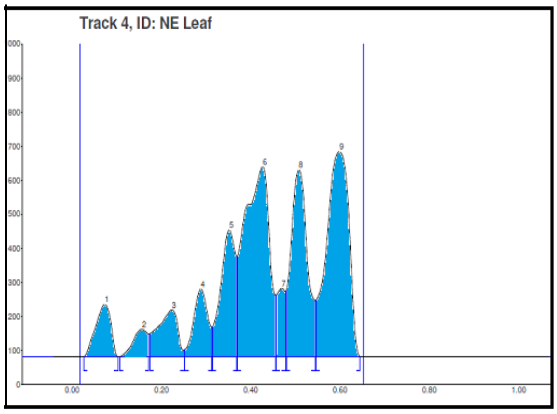

| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.02 | 0.5 | 0.07 | 222.0 | 10.72 | 0.10 | 0.8 | 6159.6 | 8.00 |
| 2 | 0.11 | 0.2 | 0.15 | 86.0 | 4.15 | 0.17 | 57.4 | 2523.4 | 3.28 |
| 3 | 0.17 | 57.6 | 0.20 | 75.9 | 3.67 | 0.22 | 0.2 | 1967.6 | 2.56 |
| 4 | 0.24 | 0.4 | 0.29 | 134.6 | 6.50 | 0.31 | 78.5 | 3781.5 | 4.91 |
| 5 | 0.31 | 80.0 | 0.41 | 483.8 | 23.36 | 0.47 | 165.1 | 32365.2 | 42.04 |
| 6 | 0.47 | 185.8 | 0.47 | 197.3 | 9.53 | 0.48 | 182.9 | 2473.7 | 3.21 |
| 7 | 0.49 | 184.7 | 0.51 | 493.0 | 23.81 | 0.56 | 113.1 | 14889.5 | 19.34 |
| 8 | 0.56 | 113.2 | 0.60 | 378.3 | 18.27 | 0.65 | 4.5 | 12830.4 | 16.66 |




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.03 | 0.8 | 0.08 | 138.4 | 4.42 | 0.10 | 0.1 | 3762.8 | 4.28 |
| 2 | 0.11 | 0.5 | 0.16 | 86.8 | 2.77 | 0.17 | 72.4 | 2419.8 | 2.75 |
| 3 | 0.17 | 73.1 | 0.21 | 97.9 | 3.12 | 0.24 | 0.6 | 3277.3 | 3.73 |
| 4 | 0.25 | 0.4 | 0.29 | 184.9 | 5.90 | 0.31 | 96.8 | 4649.0 | 5.29 |
| 5 | 0.32 | 96.9 | 0.35 | 361.5 | 11.54 | 0.37 | 299.6 | 9849.4 | 11.21 |
| 6 | 0.37 | 299.8 | 0.40 | 442.2 | 14.11 | 0.42 | 480.0 | 13729.3 | 15.63 |
| 7 | 0.42 | 431.9 | 0.43 | 547.4 | 17.47 | 0.46 | 143.6 | 10637.2 | 12.11 |
| 8 | 0.46 | 144.3 | 0.47 | 159.8 | 5.10 | 0.48 | 153.0 | 2307.1 | 2.63 |
| 9 | 0.48 | 154.6 | 0.51 | 529.5 | 16.90 | 0.54 | 140.5 | 14752.6 | 16.79 |
| 10 | 0.54 | 141.1 | 0.60 | 584.9 | 18.67 | 0.64 | 0.6 | 22478.7 | 25.58 |

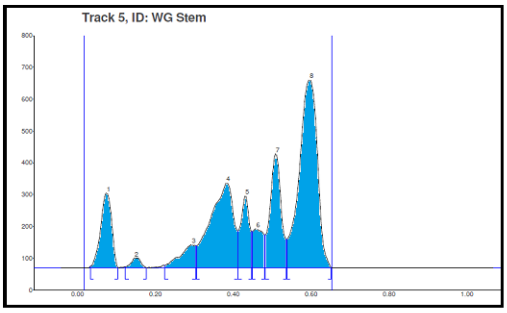


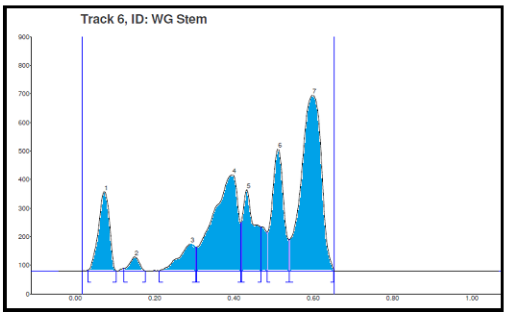



| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.03 | 0.4 | 0.07 | 154.6 | 5.40 | 0.10 | 0.5 | 4428.4 | 4.47 |
| 2 | 0.11 | 0.0 | 0.16 | 82.2 | 2.87 | 0.17 | 68.1 | 2238.6 | 2.26 |
| 3 | 0.17 | 69.1 | 0.22 | 138.6 | 4.84 | 0.25 | 20.4 | 5191.7 | 5.24 |
| 4 | 0.25 | 20.8 | 0.29 | 200.3 | 6.99 | 0.31 | 87.7 | 5306.0 | 5.36 |
| 5 | 0.32 | 88.9 | 0.35 | 373.1 | 13.03 | 0.37 | 294.7 | 10262.6 | 10.36 |
| 6 | 0.37 | 295.3 | 0.43 | 559.2 | 19.53 | 0.46 | 181.2 | 26142.8 | 26.38 |
| 7 | 0.46 | 182.0 | 0.47 | 203.2 | 7.10 | 0.48 | 192.6 | 3112.2 | 3.14 |
| 8 | 0.48 | 195.3 | 0.51 | 549.7 | 19.19 | 0.55 | 166.1 | 16940.2 | 17.10 |
| 9 | 0.55 | 167.1 | 0.60 | 603.0 | 21.05 | 0.65 | 1.0 | 25461.5 | 25.70 |

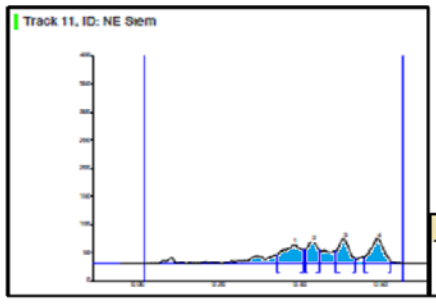
**Figure 2: Densitogram and corresponding Rf values of Flavanoids of stem of *Oroxylum indicum* (L.) Vent from two regions (Western Ghats and North East) at wavelength 254 nm**




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.03 | 4.8 | 0.08 | 235.9 | 12.35 | 0.10 | 0.8 | 5600.6 | 9.24 |
| 2 | 0.12 | 3.0 | 0.15 | 31.7 | 1.66 | 0.18 | 0.2 | 638.4 | 1.05 |
| 3 | 0.22 | 8.6 | 0.29 | 73.6 | 3.86 | 0.30 | 69.6 | 2410.1 | 3.98 |
| 4 | 0.31 | 69.3 | 0.38 | 267.9 | 14.03 | 0.41 | 116.2 | 13210.0 | 21.81 |
| 5 | 0.41 | 116.8 | 0.43 | 226.7 | 11.87 | 0.45 | 113.7 | 4270.4 | 7.05 |
| 6 | 0.45 | 114.0 | 0.46 | 122.6 | 6.42 | 0.48 | 103.7 | 2780.6 | 4.59 |
| 7 | 0.48 | 103.8 | 0.51 | 358.9 | 18.79 | 0.54 | 89.8 | 8519.8 | 14.06 |
| 8 | 0.54 | 89.9 | 0.60 | 592.4 | 31.02 | 0.65 | 1.2 | 23151.2 | 38.22 |




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.03 | 3.6 | 0.07 | 278.2 | 13.33 | 0.10 | 0.8 | 6494.9 | 8.93 |
| 2 | 0.12 | 8.6 | 0.15 | 49.6 | 2.38 | 0.18 | 0.3 | 992.3 | 1.36 |
| 3 | 0.21 | 1.2 | 0.29 | 94.9 | 4.54 | 0.30 | 82.1 | 3188.1 | 4.38 |
| 4 | 0.31 | 82.6 | 0.40 | 336.6 | 16.12 | 0.42 | 167.9 | 17671.6 | 24.30 |
| 5 | 0.42 | 171.0 | 0.43 | 285.0 | 13.65 | 0.47 | 155.2 | 7350.9 | 10.11 |
| 6 | 0.48 | 136.4 | 0.51 | 427.5 | 20.48 | 0.54 | 109.7 | 10735.9 | 14.76 |
| 7 | 0.54 | 110.6 | 0.60 | 615.8 | 29.50 | 0.65 | 4.1 | 26282.1 | 36.14 |



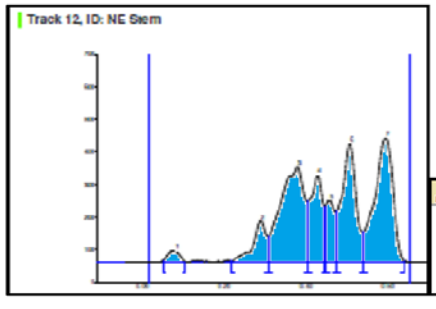
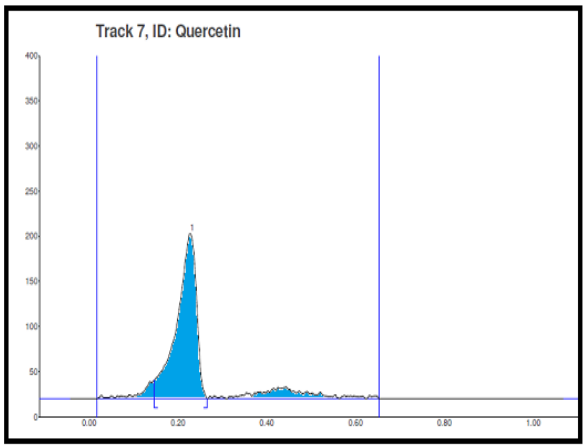



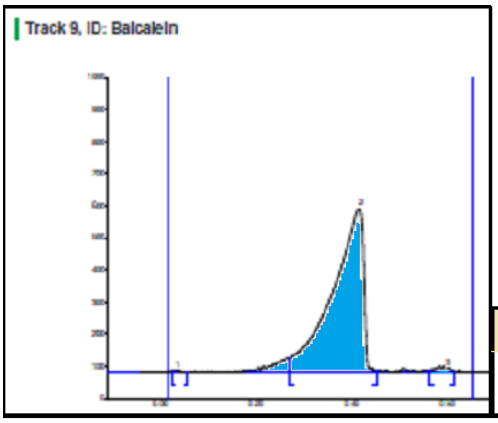


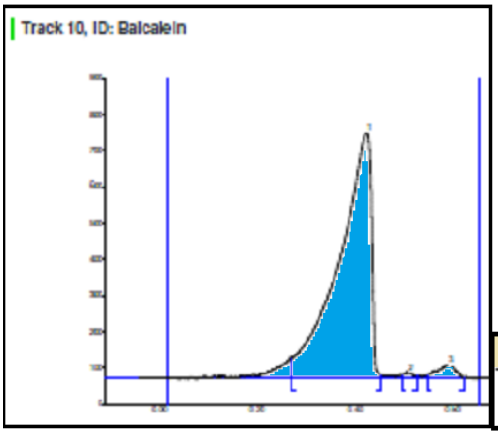
**Figure 3: Densitogram and corresponding Rf values of Querecetin and Baicalein at wavelength 254 nm**




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.15 | 20.9 | 0.23 | 183.8 | 100.00 | 0.27 | 0.0 | 6741.4 | 100.00 |




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % | Assigned substance |
|---|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.02 | 6.2 | 0.03 | 10.6 | 1.95 | 0.06 | 0.5 | 112.2 | 0.40 | unknown * |
| 2 | 0.27 | 46.5 | 0.42 | 514.6 | 94.60 | 0.45 | 4.5 | 27475.7 | 97.91 | unknown * |
| 3 | 0.56 | 6.4 | 0.60 | 18.7 | 3.44 | 0.62 | 2.8 | 473.2 | 1.69 | unknown * |

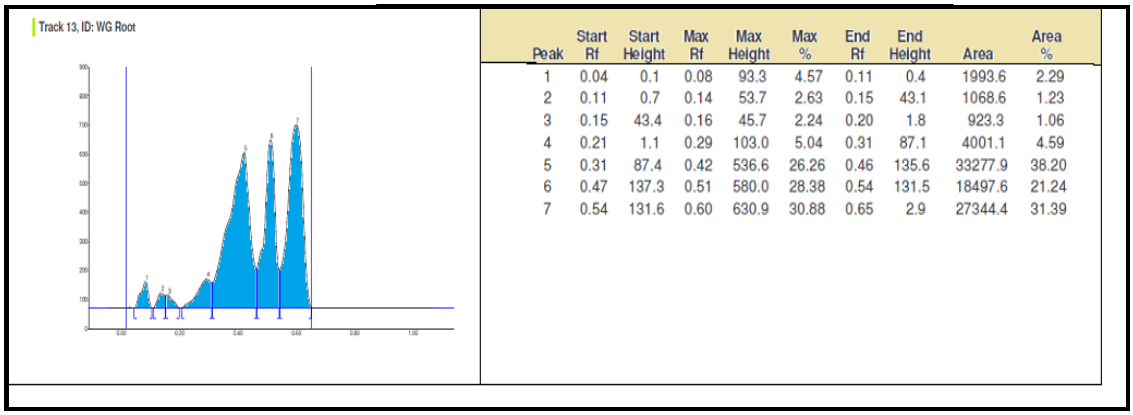



| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % | Assigned substance |
|---|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.27 | 57.0 | 0.43 | 680.3 | 92.24 | 0.45 | 10.8 | 37131.6 | 96.03 | unknown * |
| 2 | 0.50 | 9.2 | 0.51 | 18.9 | 2.56 | 0.53 | 5.6 | 299.0 | 0.77 | unknown * |
| 3 | 0.55 | 9.8 | 0.59 | 38.3 | 5.19 | 0.62 | 3.0 | 1237.8 | 3.20 | unknown * |

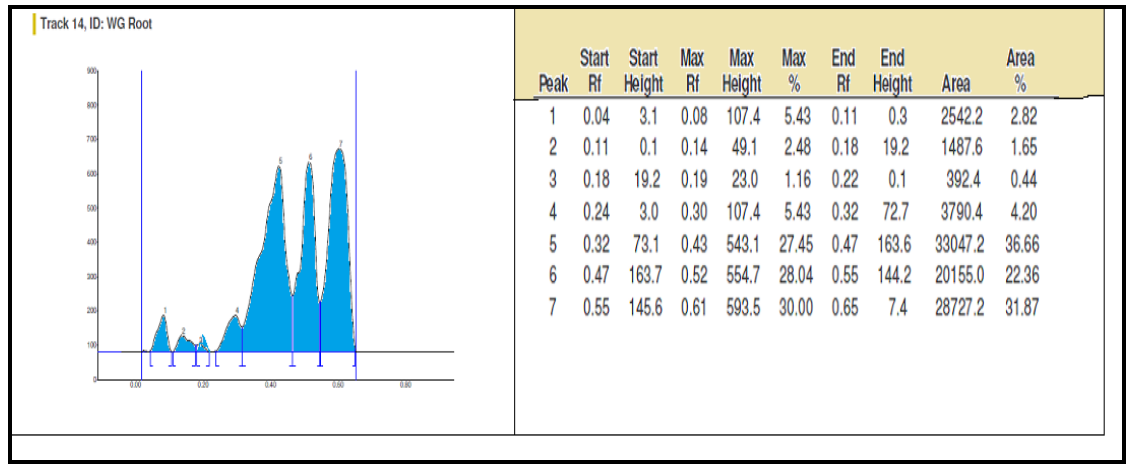


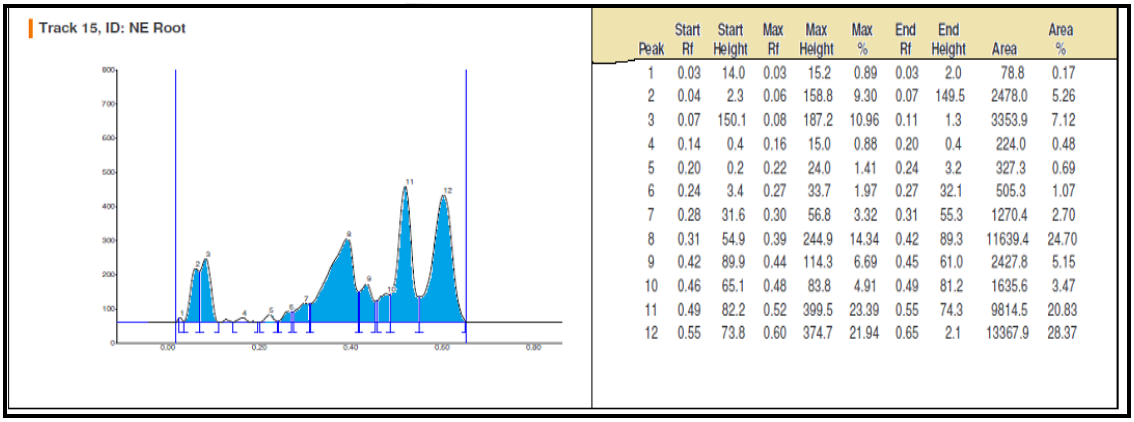
**Figure 4: Densitogram and corresponding Rf values of Flavanoids of root of *Oroxylum indicum* (L.) Vent from two regions (Western Ghats and North East) at wavelength 254 nm**



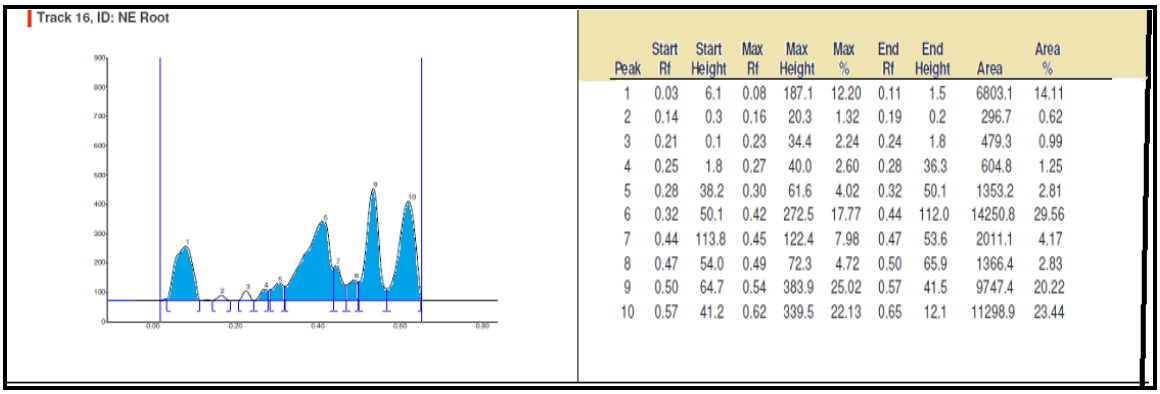

| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.04 | 0.1 | 0.08 | 93.3 | 4.57 | 0.11 | 0.4 | 1993.6 | 2.29 |
| 2 | 0.11 | 0.7 | 0.14 | 53.7 | 2.63 | 0.15 | 43.1 | 1068.6 | 1.23 |
| 3 | 0.15 | 43.4 | 0.16 | 45.7 | 2.24 | 0.20 | 1.8 | 923.3 | 1.06 |
| 4 | 0.21 | 1.1 | 0.29 | 103.0 | 5.04 | 0.31 | 87.1 | 4001.1 | 4.59 |
| 5 | 0.31 | 87.4 | 0.42 | 536.6 | 26.26 | 0.46 | 135.6 | 33277.9 | 38.20 |
| 6 | 0.47 | 137.3 | 0.51 | 580.0 | 28.38 | 0.54 | 131.5 | 18497.6 | 21.24 |
| 7 | 0.54 | 131.6 | 0.60 | 630.9 | 30.88 | 0.65 | 2.9 | 27344.4 | 31.39 |




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.04 | 3.1 | 0.08 | 107.4 | 5.43 | 0.11 | 0.3 | 2542.2 | 2.82 |
| 2 | 0.11 | 0.1 | 0.14 | 49.1 | 2.48 | 0.18 | 19.2 | 1487.6 | 1.65 |
| 3 | 0.18 | 19.2 | 0.19 | 23.0 | 1.16 | 0.22 | 0.1 | 392.4 | 0.44 |
| 4 | 0.24 | 3.0 | 0.30 | 107.4 | 5.43 | 0.32 | 72.7 | 3790.4 | 4.20 |
| 5 | 0.32 | 73.1 | 0.43 | 543.1 | 27.45 | 0.47 | 163.6 | 33047.2 | 36.66 |
| 6 | 0.47 | 163.7 | 0.52 | 554.7 | 28.04 | 0.55 | 144.2 | 20155.0 | 22.36 |
| 7 | 0.55 | 145.6 | 0.61 | 593.5 | 30.00 | 0.65 | 7.4 | 28727.2 | 31.87 |




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.03 | 14.0 | 0.03 | 15.2 | 0.89 | 0.03 | 2.0 | 78.8 | 0.17 |
| 2 | 0.04 | 2.3 | 0.06 | 158.8 | 9.30 | 0.07 | 149.5 | 2478.0 | 5.26 |
| 3 | 0.07 | 150.1 | 0.08 | 187.2 | 10.96 | 0.11 | 1.3 | 3353.9 | 7.12 |
| 4 | 0.14 | 0.4 | 0.16 | 15.0 | 0.88 | 0.20 | 0.4 | 224.0 | 0.48 |
| 5 | 0.20 | 0.2 | 0.22 | 24.0 | 1.41 | 0.24 | 3.2 | 327.3 | 0.69 |
| 6 | 0.24 | 3.4 | 0.27 | 33.7 | 1.97 | 0.27 | 32.1 | 505.3 | 1.07 |
| 7 | 0.28 | 31.6 | 0.30 | 56.8 | 3.32 | 0.31 | 55.3 | 1270.4 | 2.70 |
| 8 | 0.31 | 54.9 | 0.39 | 244.9 | 14.34 | 0.42 | 89.3 | 11639.4 | 24.70 |
| 9 | 0.42 | 89.9 | 0.44 | 114.3 | 6.69 | 0.45 | 61.0 | 2427.8 | 5.15 |
| 10 | 0.46 | 65.1 | 0.48 | 83.8 | 4.91 | 0.49 | 81.2 | 1635.6 | 3.47 |
| 11 | 0.49 | 82.2 | 0.52 | 399.5 | 23.39 | 0.55 | 74.3 | 9814.5 | 20.83 |
| 12 | 0.55 | 73.8 | 0.60 | 374.7 | 21.94 | 0.65 | 2.1 | 13367.9 | 28.37 |

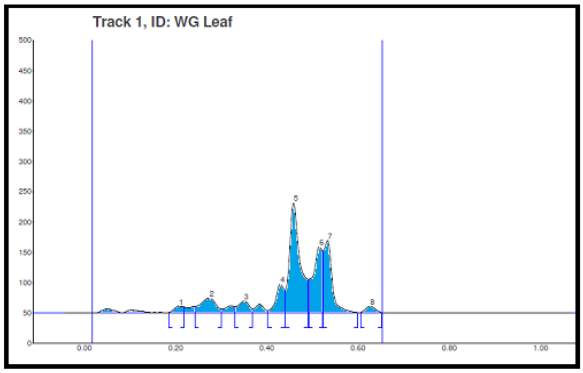



| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.03 | 6.1 | 0.08 | 187.1 | 12.20 | 0.11 | 1.5 | 6803.1 | 14.11 |
| 2 | 0.14 | 0.3 | 0.16 | 20.3 | 1.32 | 0.19 | 0.2 | 296.7 | 0.62 |
| 3 | 0.21 | 0.1 | 0.23 | 34.4 | 2.24 | 0.24 | 1.8 | 479.3 | 0.99 |
| 4 | 0.25 | 1.8 | 0.27 | 40.0 | 2.60 | 0.28 | 36.3 | 604.8 | 1.25 |
| 5 | 0.28 | 38.2 | 0.30 | 61.6 | 4.02 | 0.32 | 50.1 | 1353.2 | 2.81 |
| 6 | 0.32 | 50.1 | 0.42 | 272.5 | 17.77 | 0.44 | 112.0 | 14250.8 | 29.56 |
| 7 | 0.44 | 113.8 | 0.45 | 122.4 | 7.98 | 0.47 | 53.6 | 2011.1 | 4.17 |
| 8 | 0.47 | 54.0 | 0.49 | 72.3 | 4.72 | 0.50 | 65.9 | 1366.4 | 2.83 |
| 9 | 0.50 | 64.7 | 0.54 | 383.9 | 25.02 | 0.57 | 41.5 | 9747.4 | 20.22 |
| 10 | 0.57 | 41.2 | 0.62 | 339.5 | 22.13 | 0.65 | 12.1 | 11298.9 | 23.44 |

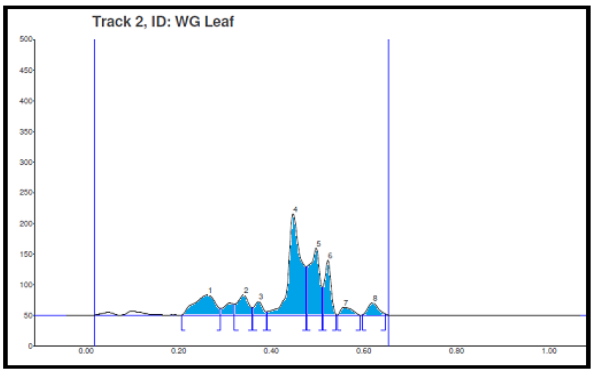


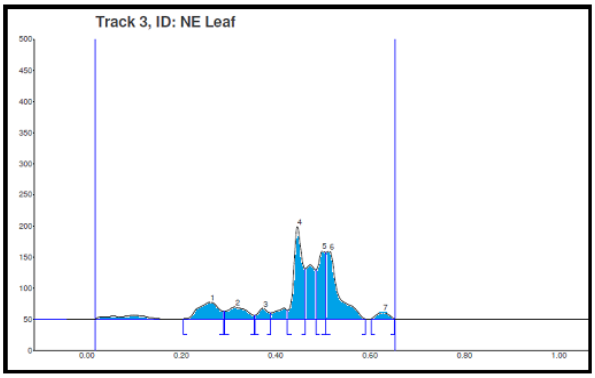
**Figure 5: Densitogram and corresponding Rf values of Flavanoids of leaf of *Oroxylum indicum* (L.) Vent from two regions (Western Ghats and North East) at wavelength 366 nm**



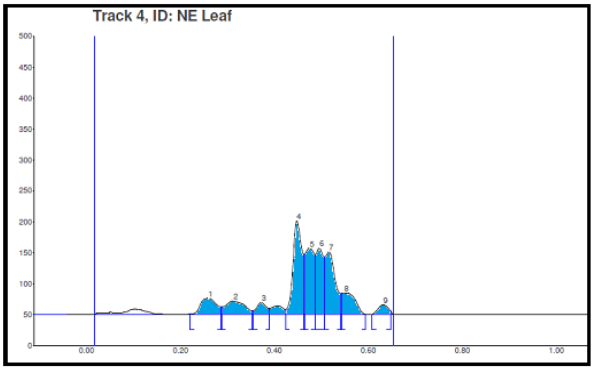

| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.18 | 0.3 | 0.21 | 10.6 | 2.04 | 0.22 | 9.4 | 178.4 | 1.89 |
| 2 | 0.24 | 10.3 | 0.28 | 24.0 | 4.59 | 0.30 | 5.6 | 681.5 | 7.21 |
| 3 | 0.33 | 10.0 | 0.35 | 19.6 | 3.76 | 0.37 | 6.2 | 392.2 | 4.15 |
| 4 | 0.40 | 3.6 | 0.43 | 47.1 | 9.03 | 0.44 | 37.2 | 704.1 | 7.45 |
| 5 | 0.44 | 37.4 | 0.46 | 181.5 | 34.77 | 0.49 | 55.0 | 3588.6 | 37.96 |
| 6 | 0.49 | 55.2 | 0.52 | 108.8 | 20.85 | 0.52 | 101.6 | 1880.5 | 19.89 |
| 7 | 0.52 | 101.7 | 0.53 | 119.5 | 22.91 | 0.60 | 0.2 | 1843.6 | 19.50 |
| 8 | 0.61 | 0.2 | 0.63 | 10.7 | 2.06 | 0.65 | 0.2 | 185.1 | 1.96 |




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.21 | 0.1 | 0.26 | 33.1 | 6.79 | 0.29 | 10.6 | 1243.3 | 12.66 |
| 2 | 0.32 | 17.8 | 0.34 | 32.9 | 6.77 | 0.36 | 11.9 | 670.7 | 6.83 |
| 3 | 0.36 | 12.1 | 0.37 | 22.6 | 4.64 | 0.39 | 5.2 | 327.6 | 3.34 |
| 4 | 0.39 | 5.2 | 0.45 | 165.7 | 34.04 | 0.48 | 78.5 | 3806.4 | 38.76 |
| 5 | 0.48 | 78.9 | 0.50 | 109.4 | 22.48 | 0.51 | 45.2 | 2083.1 | 21.21 |
| 6 | 0.51 | 45.5 | 0.52 | 89.7 | 18.43 | 0.54 | 0.5 | 1049.1 | 10.68 |
| 7 | 0.54 | 0.1 | 0.56 | 13.1 | 2.70 | 0.59 | 0.0 | 262.8 | 2.68 |
| 8 | 0.60 | 0.3 | 0.62 | 20.1 | 4.14 | 0.65 | 2.3 | 376.2 | 3.83 |




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.20 | 0.3 | 0.26 | 27.3 | 6.16 | 0.29 | 12.5 | 983.1 | 11.61 |
| 2 | 0.29 | 12.2 | 0.32 | 19.6 | 4.41 | 0.36 | 5.9 | 663.0 | 7.83 |
| 3 | 0.36 | 6.1 | 0.38 | 17.3 | 3.91 | 0.39 | 10.0 | 307.5 | 3.63 |
| 4 | 0.43 | 14.0 | 0.45 | 148.7 | 33.53 | 0.46 | 80.6 | 2331.3 | 27.52 |
| 5 | 0.49 | 78.9 | 0.50 | 110.0 | 24.81 | 0.51 | 106.7 | 1568.4 | 18.52 |
| 6 | 0.51 | 106.9 | 0.52 | 109.5 | 24.69 | 0.59 | 0.1 | 2382.4 | 28.12 |
| 7 | 0.60 | 0.1 | 0.63 | 11.1 | 2.50 | 0.65 | 0.6 | 235.4 | 2.78 |

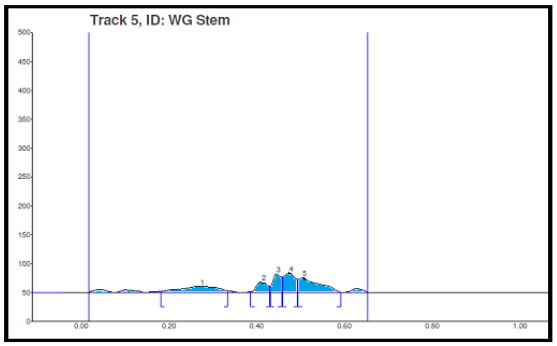



| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.22 | 0.3 | 0.26 | 26.0 | 4.46 | 0.29 | 11.9 | 734.4 | 7.09 |
| 2 | 0.29 | 11.9 | 0.31 | 21.8 | 3.74 | 0.35 | 5.5 | 739.4 | 7.14 |
| 3 | 0.36 | 5.6 | 0.37 | 19.6 | 3.36 | 0.39 | 9.9 | 336.2 | 3.25 |
| 4 | 0.43 | 7.7 | 0.45 | 151.6 | 25.96 | 0.46 | 97.0 | 2438.4 | 23.55 |
| 5 | 0.47 | 97.2 | 0.48 | 106.7 | 18.27 | 0.49 | 96.6 | 1738.8 | 16.79 |
| 6 | 0.49 | 96.7 | 0.50 | 107.4 | 18.38 | 0.51 | 92.7 | 1406.9 | 13.59 |
| 7 | 0.51 | 92.7 | 0.52 | 100.7 | 17.24 | 0.54 | 33.4 | 1857.2 | 17.94 |
| 8 | 0.54 | 33.6 | 0.55 | 34.7 | 5.94 | 0.60 | 0.1 | 829.4 | 8.01 |
| 9 | 0.61 | 0.2 | 0.63 | 15.5 | 2.65 | 0.65 | 5.6 | 273.0 | 2.64 |

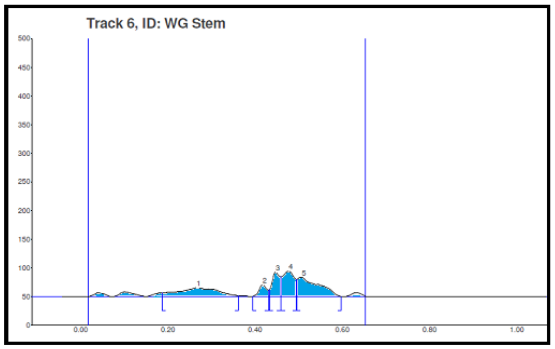


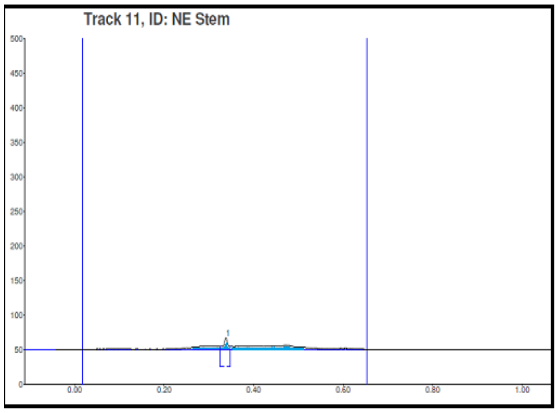
**Figure 6: Densitogram and corresponding Rf values of Flavanoids of stem of *Oroxylum indicum* (L.) Vent from two regions (Western Ghats and North East) at wavelength 366 nm**



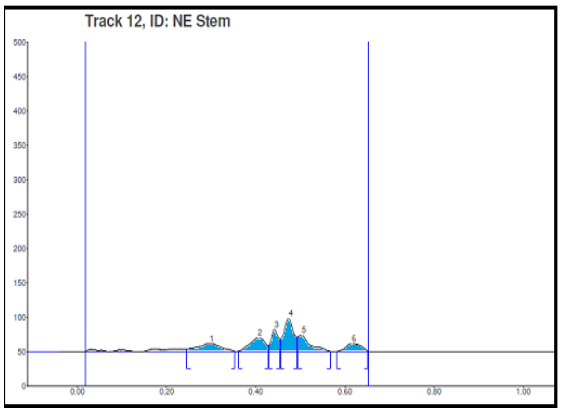

| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.18 | 1.4 | 0.27 | 10.5 | 8.83 | 0.33 | 2.5 | 739.3 | 22.16 |
| 2 | 0.39 | 0.3 | 0.41 | 17.9 | 15.03 | 0.43 | 10.2 | 366.3 | 10.98 |
| 3 | 0.43 | 10.6 | 0.45 | 32.1 | 27.02 | 0.46 | 26.8 | 495.5 | 14.85 |
| 4 | 0.46 | 26.9 | 0.47 | 33.1 | 27.83 | 0.49 | 22.0 | 740.1 | 22.18 |
| 5 | 0.50 | 22.1 | 0.51 | 25.3 | 21.28 | 0.59 | 0.0 | 995.6 | 29.84 |




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.19 | 5.7 | 0.27 | 14.8 | 9.56 | 0.36 | 0.8 | 1128.0 | 25.65 |
| 2 | 0.40 | 0.2 | 0.42 | 20.3 | 13.09 | 0.43 | 10.7 | 291.2 | 6.62 |
| 3 | 0.43 | 11.1 | 0.45 | 42.3 | 27.32 | 0.46 | 34.2 | 626.5 | 14.25 |
| 4 | 0.46 | 34.3 | 0.48 | 44.1 | 28.51 | 0.50 | 29.3 | 987.0 | 22.44 |
| 5 | 0.50 | 29.4 | 0.51 | 33.3 | 21.52 | 0.60 | 0.0 | 1365.1 | 31.04 |




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.33 | 4.5 | 0.34 | 17.2 | 100.00 | 0.35 | 4.3 | 133.4 | 100.00 |

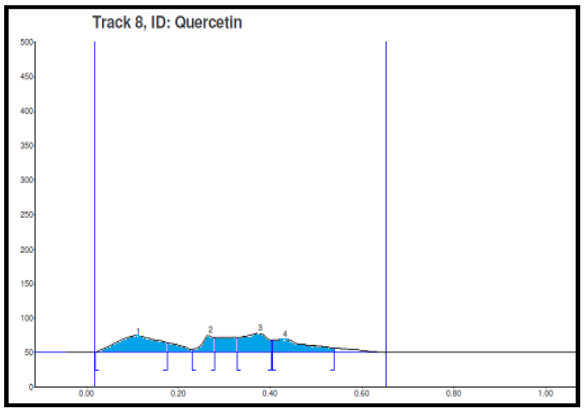



| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.25 | 3.7 | 0.30 | 11.9 | 7.98 | 0.35 | 0.4 | 536.1 | 16.10 |
| 2 | 0.36 | 0.6 | 0.41 | 20.2 | 13.56 | 0.43 | 8.9 | 583.5 | 17.52 |
| 3 | 0.43 | 9.3 | 0.44 | 32.4 | 21.73 | 0.46 | 18.2 | 423.8 | 12.72 |
| 4 | 0.46 | 18.5 | 0.47 | 48.3 | 32.37 | 0.49 | 21.0 | 904.7 | 27.16 |
| 5 | 0.50 | 21.3 | 0.50 | 24.2 | 16.24 | 0.57 | 0.4 | 539.6 | 16.20 |
| 6 | 0.58 | 0.3 | 0.62 | 12.1 | 8.12 | 0.65 | 0.6 | 342.9 | 10.30 |

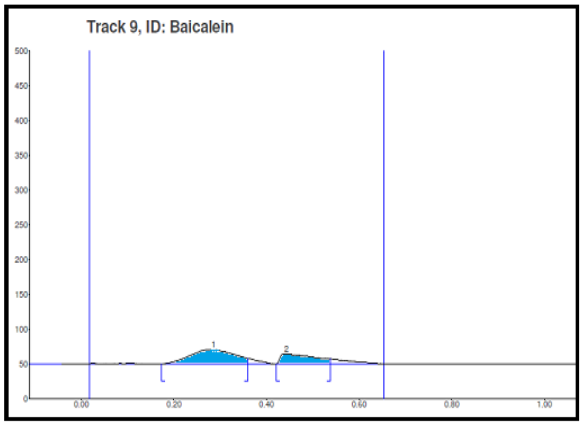


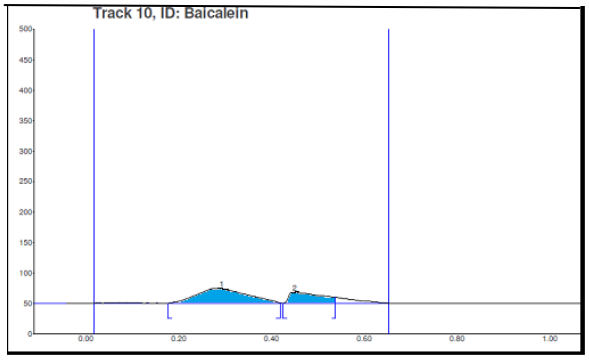
**Figure 7: Densitogram and corresponding Rf values of Querecetin and Baicalein at wavelength 366nm**




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.02 | 0.3 | 0.11 | 24.6 | 24.80 | 0.18 | 15.2 | 1897.8 | 36.96 |
| 2 | 0.23 | 5.0 | 0.27 | 26.1 | 26.24 | 0.28 | 21.7 | 572.1 | 11.14 |
| 3 | 0.33 | 22.2 | 0.38 | 28.6 | 28.82 | 0.40 | 18.5 | 1346.4 | 26.22 |
| 4 | 0.41 | 18.6 | 0.43 | 20.0 | 20.14 | 0.54 | 6.8 | 1317.7 | 25.67 |




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.17 | 0.2 | 0.28 | 20.8 | 58.59 | 0.36 | 8.0 | 1687.1 | 65.22 |
| 2 | 0.42 | 0.0 | 0.44 | 14.7 | 41.41 | 0.54 | 7.2 | 899.6 | 34.78 |

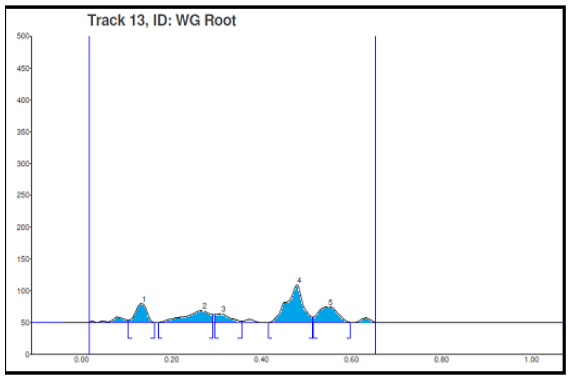



| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.18 | 0.3 | 0.29 | 24.3 | 57.61 | 0.42 | 1.0 | 2212.7 | 67.83 |
| 2 | 0.43 | 0.1 | 0.45 | 17.9 | 42.39 | 0.54 | 9.5 | 1049.2 | 32.17 |

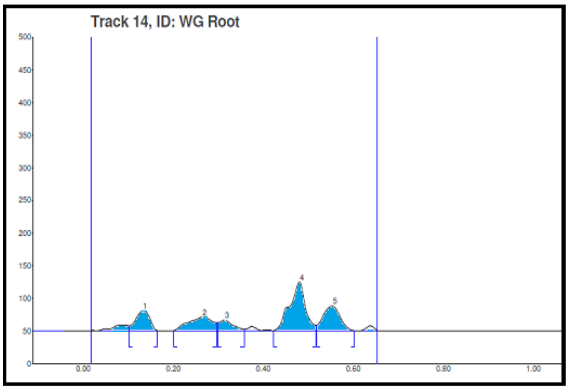


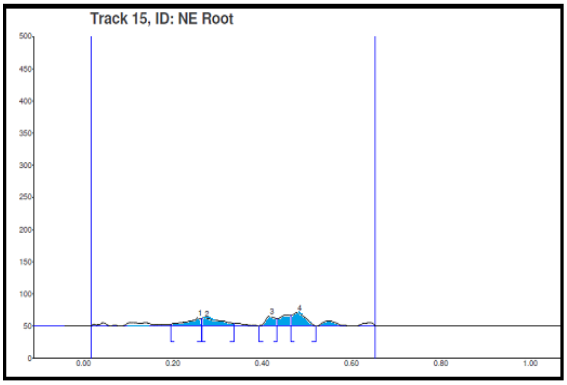
**Figure 8: Densitogram and corresponding Rf values of Flavanoids of root of *Oroxylum indicum* (L.) Vent from two regions (Western Ghats and North East) at wavelength 366 nm**



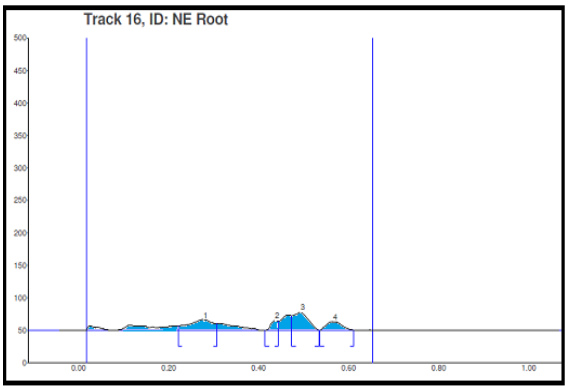

| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.10 | 3.2 | 0.13 | 29.9 | 20.43 | 0.16 | 0.0 | 625.6 | 13.65 |
| 2 | 0.17 | 0.1 | 0.27 | 18.6 | 12.71 | 0.29 | 12.1 | 883.1 | 19.27 |
| 3 | 0.30 | 12.0 | 0.31 | 13.5 | 9.25 | 0.36 | 1.9 | 365.9 | 7.98 |
| 4 | 0.42 | 0.1 | 0.48 | 59.5 | 40.66 | 0.52 | 8.6 | 1821.8 | 39.75 |
| 5 | 0.52 | 8.8 | 0.55 | 24.8 | 16.95 | 0.60 | 0.2 | 887.0 | 19.35 |




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.10 | 7.6 | 0.13 | 30.2 | 16.84 | 0.17 | 0.2 | 801.7 | 14.57 |
| 2 | 0.20 | 0.2 | 0.27 | 20.4 | 11.36 | 0.30 | 11.9 | 930.4 | 16.91 |
| 3 | 0.30 | 12.1 | 0.32 | 16.2 | 9.05 | 0.36 | 1.9 | 423.8 | 7.70 |
| 4 | 0.42 | 0.0 | 0.48 | 74.5 | 41.61 | 0.52 | 8.4 | 2161.5 | 39.28 |
| 5 | 0.52 | 8.4 | 0.56 | 37.9 | 21.14 | 0.60 | 0.1 | 1185.1 | 21.54 |




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.20 | 2.7 | 0.26 | 12.6 | 21.02 | 0.26 | 11.2 | 325.6 | 21.05 |
| 2 | 0.27 | 11.6 | 0.27 | 11.9 | 19.87 | 0.34 | 3.6 | 434.5 | 28.09 |
| 3 | 0.39 | 0.1 | 0.42 | 15.0 | 24.90 | 0.43 | 11.1 | 275.5 | 17.81 |
| 4 | 0.47 | 16.1 | 0.48 | 20.5 | 34.21 | 0.52 | 0.1 | 511.0 | 33.04 |

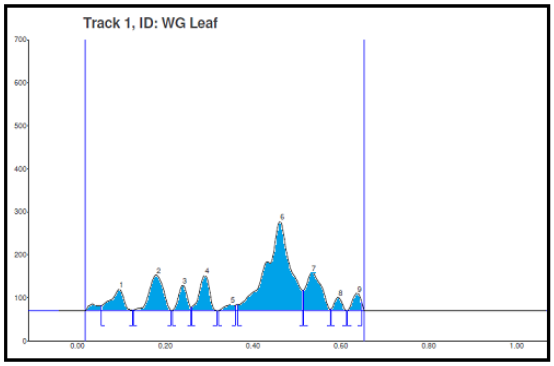



| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.22 | 6.2 | 0.28 | 15.7 | 22.01 | 0.31 | 9.9 | 702.1 | 35.09 |
| 2 | 0.42 | 0.0 | 0.44 | 14.9 | 20.93 | 0.44 | 13.8 | 185.1 | 9.25 |
| 3 | 0.47 | 21.9 | 0.49 | 27.7 | 38.73 | 0.53 | 0.1 | 750.2 | 37.50 |
| 4 | 0.54 | 0.0 | 0.57 | 13.1 | 18.33 | 0.61 | 0.0 | 363.4 | 18.16 |

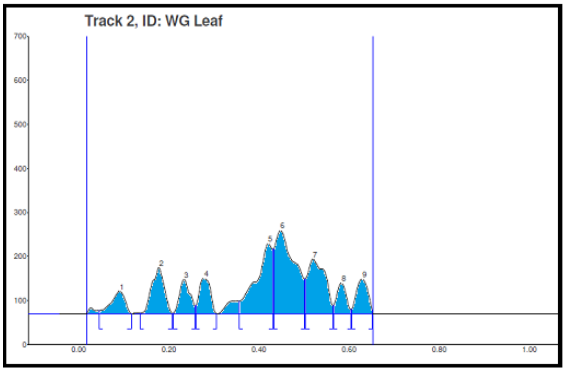


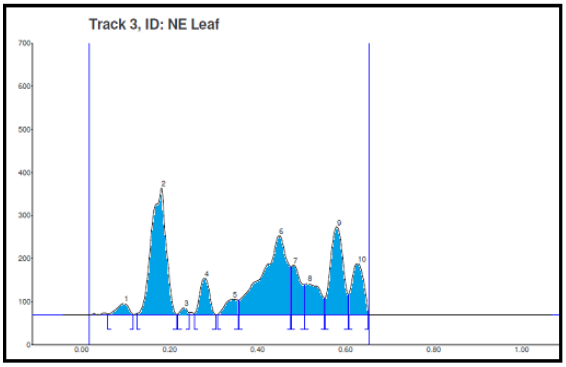
**Figure 9: Densitogram and corresponding Rf values of Flavanoids of leaves of *Oroxylum indicum* (L.) Vent from two regions (Western Ghats and North East) at wavelength 540 nm**



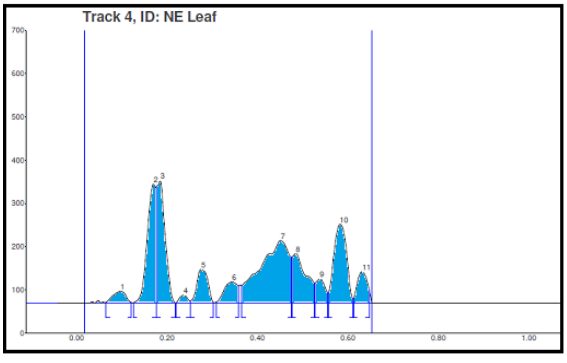

| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.05 | 11.5 | 0.10 | 49.5 | 7.57 | 0.13 | 0.8 | 1224.8 | 6.34 |
| 2 | 0.13 | 1.1 | 0.18 | 82.7 | 12.64 | 0.21 | 0.3 | 2178.4 | 11.28 |
| 3 | 0.22 | 0.3 | 0.24 | 59.4 | 9.08 | 0.26 | 7.2 | 950.0 | 4.92 |
| 4 | 0.26 | 8.1 | 0.29 | 81.9 | 12.52 | 0.32 | 0.0 | 1570.0 | 8.13 |
| 5 | 0.32 | 0.0 | 0.35 | 14.1 | 2.15 | 0.36 | 12.7 | 271.1 | 1.40 |
| 6 | 0.37 | 13.4 | 0.46 | 207.8 | 31.76 | 0.52 | 46.6 | 9589.8 | 49.65 |
| 7 | 0.52 | 47.3 | 0.54 | 88.0 | 13.45 | 0.58 | 3.1 | 2503.9 | 12.96 |
| 8 | 0.58 | 3.8 | 0.60 | 30.9 | 4.72 | 0.61 | 0.1 | 428.1 | 2.22 |
| 9 | 0.62 | 0.1 | 0.64 | 40.1 | 6.12 | 0.65 | 21.3 | 598.8 | 3.10 |




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.05 | 6.8 | 0.09 | 50.9 | 5.45 | 0.12 | 0.2 | 1289.5 | 5.04 |
| 2 | 0.14 | 0.9 | 0.18 | 104.3 | 11.16 | 0.21 | 0.0 | 2401.7 | 9.39 |
| 3 | 0.21 | 0.7 | 0.23 | 77.5 | 8.30 | 0.26 | 16.8 | 1504.8 | 5.88 |
| 4 | 0.26 | 18.6 | 0.28 | 80.3 | 8.60 | 0.31 | 0.2 | 1665.5 | 6.51 |
| 5 | 0.36 | 28.5 | 0.42 | 158.9 | 17.00 | 0.43 | 145.6 | 4788.9 | 18.72 |
| 6 | 0.43 | 146.4 | 0.45 | 189.7 | 20.31 | 0.50 | 78.7 | 6983.8 | 27.30 |
| 7 | 0.50 | 79.3 | 0.52 | 124.5 | 13.32 | 0.57 | 18.2 | 4044.7 | 15.81 |
| 8 | 0.57 | 18.5 | 0.58 | 69.3 | 7.42 | 0.61 | 9.0 | 1222.0 | 4.78 |
| 9 | 0.61 | 9.7 | 0.63 | 78.9 | 8.44 | 0.65 | 6.1 | 1683.7 | 6.58 |




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.06 | 1.1 | 0.10 | 26.3 | 2.28 | 0.12 | 0.3 | 554.8 | 1.74 |
| 2 | 0.13 | 2.1 | 0.18 | 295.1 | 25.62 | 0.22 | 0.0 | 8253.0 | 25.81 |
| 3 | 0.22 | 0.7 | 0.23 | 15.6 | 1.36 | 0.25 | 4.0 | 185.2 | 0.58 |
| 4 | 0.26 | 1.9 | 0.28 | 85.1 | 7.39 | 0.31 | 0.0 | 1520.1 | 4.75 |
| 5 | 0.31 | 0.2 | 0.34 | 36.1 | 3.14 | 0.36 | 33.0 | 844.7 | 2.64 |
| 6 | 0.36 | 33.1 | 0.45 | 183.4 | 15.92 | 0.48 | 110.7 | 8839.8 | 27.65 |
| 7 | 0.48 | 110.8 | 0.48 | 114.8 | 9.96 | 0.51 | 68.9 | 2061.4 | 6.45 |
| 8 | 0.51 | 69.2 | 0.52 | 72.8 | 6.32 | 0.55 | 37.8 | 2067.2 | 6.47 |
| 9 | 0.55 | 38.1 | 0.58 | 203.5 | 17.67 | 0.61 | 46.4 | 4940.8 | 15.45 |
| 10 | 0.61 | 46.5 | 0.63 | 119.1 | 10.34 | 0.65 | 8.1 | 2706.7 | 8.47 |

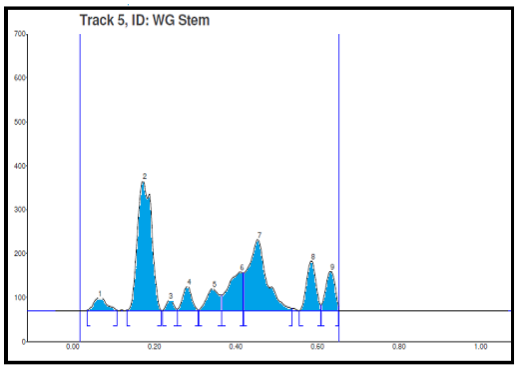



| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.07 | 0.4 | 0.10 | 27.3 | 2.11 | 0.12 | 0.5 | 617.7 | 2.10 |
| 2 | 0.13 | 0.3 | 0.17 | 274.5 | 21.23 | 0.18 | 267.5 | 4370.2 | 14.86 |
| 3 | 0.18 | 269.0 | 0.19 | 282.6 | 21.85 | 0.22 | 0.3 | 4326.9 | 14.71 |
| 4 | 0.22 | 0.4 | 0.24 | 17.3 | 1.34 | 0.25 | 4.9 | 242.1 | 0.82 |
| 5 | 0.25 | 5.1 | 0.28 | 76.6 | 5.92 | 0.30 | 1.0 | 1550.7 | 5.27 |
| 6 | 0.31 | 1.5 | 0.34 | 48.7 | 3.76 | 0.36 | 39.3 | 1190.1 | 4.05 |
| 7 | 0.37 | 40.1 | 0.45 | 144.1 | 11.14 | 0.48 | 104.9 | 7676.6 | 26.10 |
| 8 | 0.48 | 105.4 | 0.49 | 113.8 | 8.80 | 0.53 | 45.6 | 2911.0 | 9.90 |
| 9 | 0.53 | 46.1 | 0.54 | 54.9 | 4.25 | 0.56 | 23.4 | 930.1 | 3.16 |
| 10 | 0.56 | 24.2 | 0.58 | 181.7 | 14.05 | 0.61 | 8.9 | 4283.1 | 14.56 |
| 11 | 0.61 | 10.0 | 0.63 | 71.8 | 5.55 | 0.65 | 29.8 | 1309.2 | 4.45 |

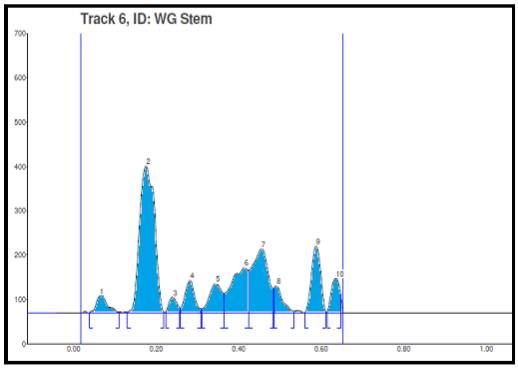


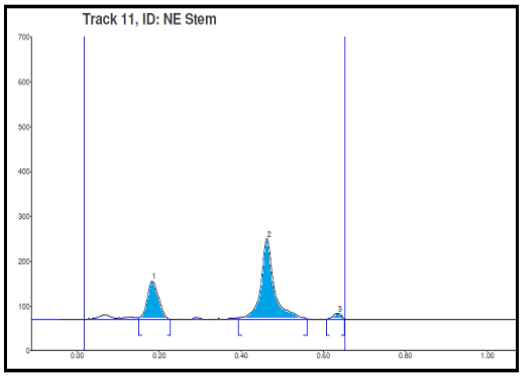
**Figure 10: Densitogram and corresponding Rf values of Flavanoids of stem of *Oroxylum indicum* (L.) Vent from two regions (Western Ghats and North East) at wavelength 540 nm**



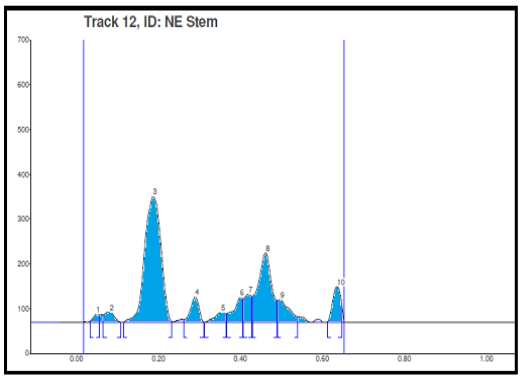

| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.03 | 0.5 | 0.06 | 28.2 | 3.12 | 0.11 | 1.5 | 777.3 | 3.16 |
| 2 | 0.13 | 1.2 | 0.17 | 295.9 | 32.66 | 0.22 | 0.8 | 8618.9 | 35.08 |
| 3 | 0.22 | 0.4 | 0.24 | 22.7 | 2.51 | 0.26 | 3.1 | 342.1 | 1.39 |
| 4 | 0.26 | 3.9 | 0.28 | 55.4 | 6.11 | 0.31 | 1.3 | 1002.3 | 4.08 |
| 5 | 0.31 | 1.4 | 0.34 | 49.8 | 5.49 | 0.37 | 34.6 | 1275.3 | 5.19 |
| 6 | 0.37 | 34.8 | 0.41 | 88.3 | 9.74 | 0.42 | 85.2 | 2540.2 | 10.34 |
| 7 | 0.42 | 85.3 | 0.45 | 162.2 | 17.90 | 0.54 | 4.8 | 6144.7 | 25.01 |
| 8 | 0.56 | 0.1 | 0.59 | 113.5 | 12.52 | 0.61 | 12.0 | 2163.7 | 8.81 |
| 9 | 0.61 | 12.6 | 0.63 | 90.0 | 9.94 | 0.65 | 7.5 | 1705.4 | 6.94 |




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.04 | 0.2 | 0.06 | 37.2 | 3.45 | 0.11 | 0.5 | 838.6 | 3.02 |
| 2 | 0.13 | 0.6 | 0.18 | 331.3 | 30.74 | 0.22 | 0.6 | 10103.5 | 36.33 |
| 3 | 0.22 | 1.1 | 0.24 | 34.8 | 3.23 | 0.26 | 9.7 | 556.4 | 2.00 |
| 4 | 0.26 | 9.8 | 0.28 | 73.5 | 6.82 | 0.31 | 7.4 | 1394.3 | 5.01 |
| 5 | 0.31 | 7.8 | 0.35 | 65.9 | 6.11 | 0.37 | 42.8 | 1683.7 | 6.05 |
| 6 | 0.37 | 43.3 | 0.42 | 102.4 | 9.50 | 0.43 | 95.0 | 3398.8 | 12.22 |
| 7 | 0.43 | 95.4 | 0.46 | 144.2 | 13.38 | 0.48 | 53.9 | 4695.1 | 16.88 |
| 8 | 0.49 | 54.5 | 0.49 | 60.7 | 5.63 | 0.54 | 3.5 | 1058.3 | 3.81 |
| 9 | 0.56 | 0.1 | 0.59 | 150.4 | 13.95 | 0.61 | 1.5 | 2795.5 | 10.05 |
| 10 | 0.62 | 0.7 | 0.64 | 77.5 | 7.19 | 0.65 | 43.1 | 1283.3 | 4.61 |




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.15 | 3.8 | 0.18 | 86.6 | 30.70 | 0.23 | 0.5 | 2061.4 | 27.69 |
| 2 | 0.39 | 3.5 | 0.46 | 181.2 | 64.24 | 0.56 | 1.6 | 5128.2 | 68.89 |
| 3 | 0.61 | 1.1 | 0.64 | 14.3 | 5.07 | 0.65 | 1.3 | 254.3 | 3.42 |

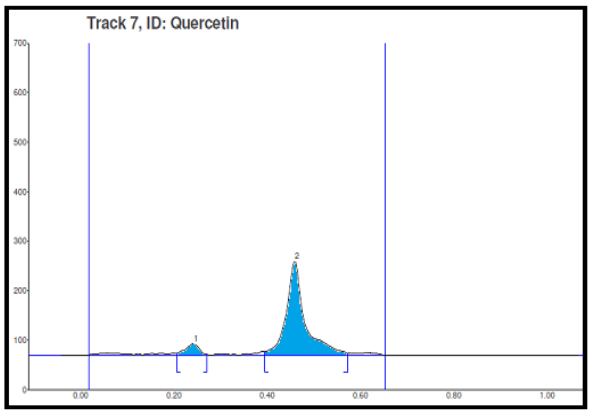



| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.03 | 2.0 | 0.05 | 18.0 | 2.25 | 0.06 | 15.9 | 211.2 | 1.05 |
| 2 | 0.07 | 16.1 | 0.08 | 21.0 | 2.64 | 0.11 | 0.1 | 435.0 | 2.17 |
| 3 | 0.12 | 0.1 | 0.19 | 281.1 | 35.20 | 0.23 | 0.6 | 9353.0 | 46.65 |
| 4 | 0.26 | 5.4 | 0.29 | 57.1 | 7.15 | 0.31 | 0.3 | 956.8 | 4.77 |
| 5 | 0.31 | 0.0 | 0.35 | 20.8 | 2.60 | 0.37 | 18.0 | 471.0 | 2.35 |
| 6 | 0.37 | 18.5 | 0.40 | 55.0 | 6.89 | 0.41 | 51.1 | 996.0 | 4.97 |
| 7 | 0.41 | 51.5 | 0.42 | 62.1 | 7.77 | 0.43 | 56.9 | 932.6 | 4.65 |
| 8 | 0.43 | 57.4 | 0.46 | 154.4 | 19.34 | 0.49 | 47.4 | 4246.1 | 21.18 |
| 9 | 0.49 | 47.7 | 0.50 | 50.0 | 6.26 | 0.54 | 12.0 | 1176.4 | 5.87 |
| 10 | 0.61 | 0.6 | 0.64 | 79.0 | 9.90 | 0.65 | 42.4 | 1269.5 | 6.33 |

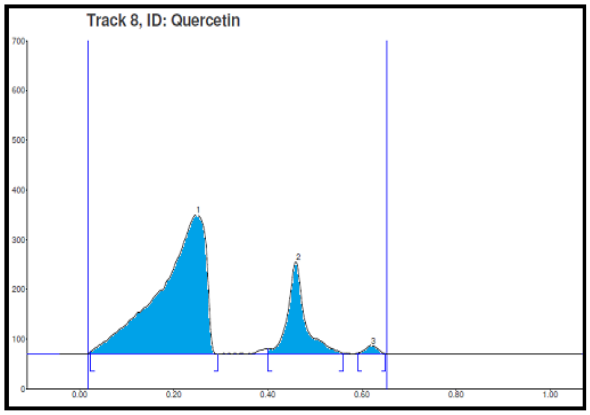


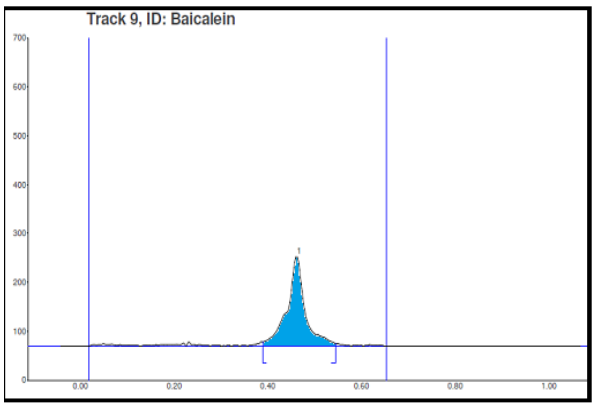
**Figure 11: Densitogram and corresponding Rf values of Standards Quercetin and Baicalein at wavelength 540nm**



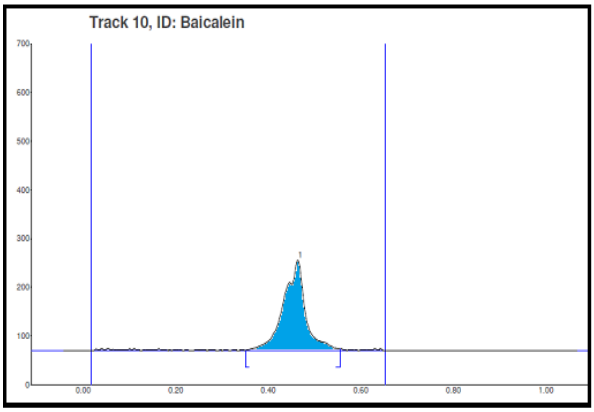

| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.21 | 3.6 | 0.24 | 23.2 | 10.92 | 0.27 | 0.9 | 542.5 | 7.89 |
| 2 | 0.40 | 6.7 | 0.46 | 189.6 | 89.08 | 0.57 | 3.6 | 6329.6 | 92.11 |




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.02 | 4.5 | 0.25 | 279.3 | 58.00 | 0.29 | 0.0 | 22548.3 | 78.65 |
| 2 | 0.40 | 9.6 | 0.46 | 185.2 | 38.44 | 0.56 | 1.8 | 5736.2 | 20.01 |
| 3 | 0.59 | 2.1 | 0.62 | 17.1 | 3.56 | 0.65 | 0.6 | 383.1 | 1.34 |




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.39 | 8.7 | 0.46 | 184.7 | 100.00 | 0.55 | 5.4 | 5852.1 | 100.00 |

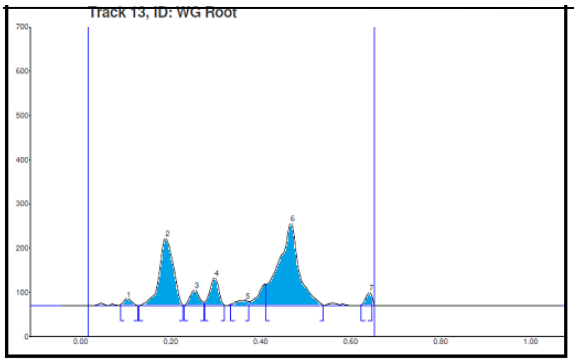



| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.35 | 0.3 | 0.47 | 185.9 | 100.00 | 0.56 | 2.7 | 7331.0 | 100.00 |

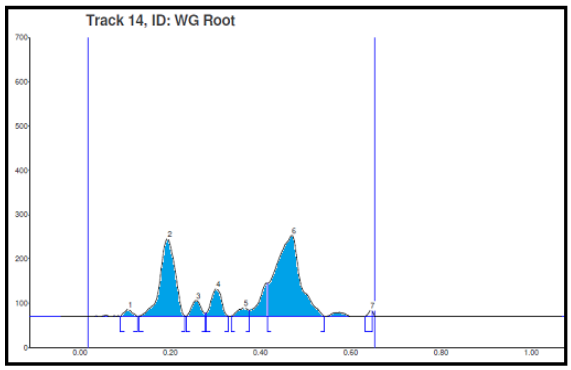


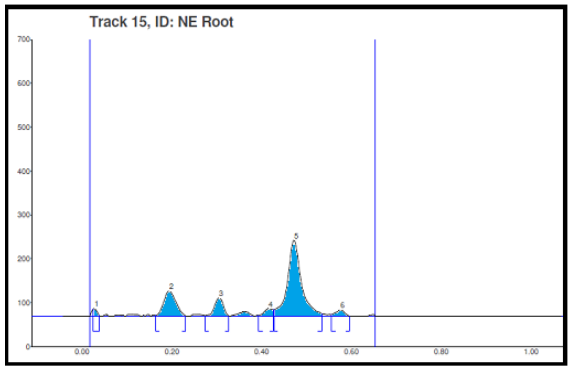
**Figure 12: Densitogram and corresponding Rf values of Flavanoids of root of *Oroxylum indicum* (L.) Vent from two regions (Western Ghats and North East) at wavelength 540 nm**



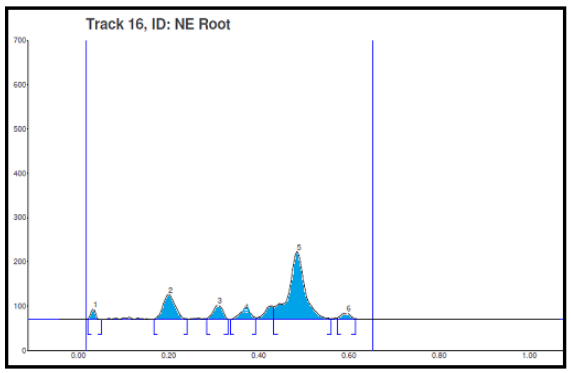

| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.09 | 1.8 | 0.10 | 15.1 | 3.06 | 0.13 | 0.0 | 229.1 | 1.70 |
| 2 | 0.13 | 0.1 | 0.19 | 152.2 | 30.84 | 0.23 | 0.7 | 3992.5 | 29.69 |
| 3 | 0.23 | 0.0 | 0.25 | 35.8 | 7.25 | 0.28 | 7.2 | 601.8 | 4.48 |
| 4 | 0.28 | 7.4 | 0.30 | 62.8 | 12.73 | 0.32 | 0.3 | 1030.6 | 7.66 |
| 5 | 0.33 | 2.2 | 0.37 | 11.4 | 2.30 | 0.38 | 9.2 | 255.6 | 1.90 |
| 6 | 0.41 | 47.6 | 0.47 | 186.5 | 37.79 | 0.54 | 0.2 | 7021.9 | 52.22 |
| 7 | 0.63 | 0.7 | 0.64 | 29.8 | 6.03 | 0.65 | 22.5 | 315.9 | 2.35 |




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.09 | 0.3 | 0.11 | 15.4 | 3.04 | 0.13 | 0.1 | 224.6 | 1.44 |
| 2 | 0.13 | 0.6 | 0.19 | 175.6 | 34.70 | 0.23 | 0.6 | 4864.5 | 31.27 |
| 3 | 0.24 | 0.2 | 0.26 | 35.8 | 7.07 | 0.28 | 7.3 | 616.5 | 3.96 |
| 4 | 0.28 | 7.5 | 0.30 | 62.2 | 12.30 | 0.33 | 1.0 | 1172.2 | 7.53 |
| 5 | 0.34 | 0.7 | 0.36 | 19.7 | 3.89 | 0.38 | 13.9 | 376.5 | 2.42 |
| 6 | 0.42 | 74.6 | 0.47 | 183.4 | 36.26 | 0.54 | 0.1 | 8214.3 | 52.80 |
| 7 | 0.63 | 0.2 | 0.65 | 13.8 | 2.74 | 0.65 | 12.8 | 90.2 | 0.58 |




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.03 | 15.7 | 0.03 | 16.9 | 5.31 | 0.04 | 0.6 | 121.1 | 1.72 |
| 2 | 0.16 | 0.6 | 0.19 | 57.1 | 17.95 | 0.23 | 0.2 | 1276.9 | 18.16 |
| 3 | 0.28 | 0.4 | 0.31 | 40.9 | 12.86 | 0.33 | 0.4 | 662.8 | 9.43 |
| 4 | 0.39 | 1.1 | 0.42 | 16.8 | 5.29 | 0.43 | 14.3 | 267.1 | 3.80 |
| 5 | 0.43 | 14.4 | 0.47 | 172.8 | 54.30 | 0.53 | 5.7 | 4480.4 | 63.73 |
| 6 | 0.56 | 3.8 | 0.58 | 13.7 | 4.30 | 0.60 | 0.0 | 222.4 | 3.16 |




| Peak | Start Rf | Start Height | Max Rf | Max Height | Max % | End Rf | End Height | Area | Area % |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 0.02 | 0.5 | 0.03 | 23.4 | 7.95 | 0.05 | 0.0 | 202.3 | 2.77 |
| 2 | 0.17 | 0.2 | 0.20 | 56.3 | 19.11 | 0.24 | 0.6 | 1256.5 | 17.18 |
| 3 | 0.28 | 1.4 | 0.31 | 31.4 | 10.66 | 0.33 | 0.2 | 569.6 | 7.79 |
| 4 | 0.34 | 0.2 | 0.37 | 17.7 | 5.99 | 0.39 | 3.5 | 426.1 | 5.83 |
| 5 | 0.43 | 28.9 | 0.49 | 152.6 | 51.80 | 0.56 | 0.6 | 4609.7 | 63.03 |
| 6 | 0.57 | 3.7 | 0.60 | 13.2 | 4.49 | 0.62 | 1.0 | 249.0 | 3.40 |



**Table1: Total number of Flavanoids observed in different parts of *Oroxylum indicum* (L.) Vent from two regions (Western Ghats and North East) at 254 nm**

| Sample | Leaf | | Stem | | Root | |
|---|---|---|---|---|---|---|
| | 5 µl | 10 µl | 10 µl | 20 µl | 10 µl | 20 µl |
| Western Ghat | 9 | 8 | 8 | 7 | 7 | 7 |
| North East | 10 | 9 | 3 | 7 | 12 | 10 |

**Table 2: Total number of Flavanoids observed in different parts of *Oroxylum indicum* (L.) Vent from two regions (Western Ghats and North East) at 366 nm**

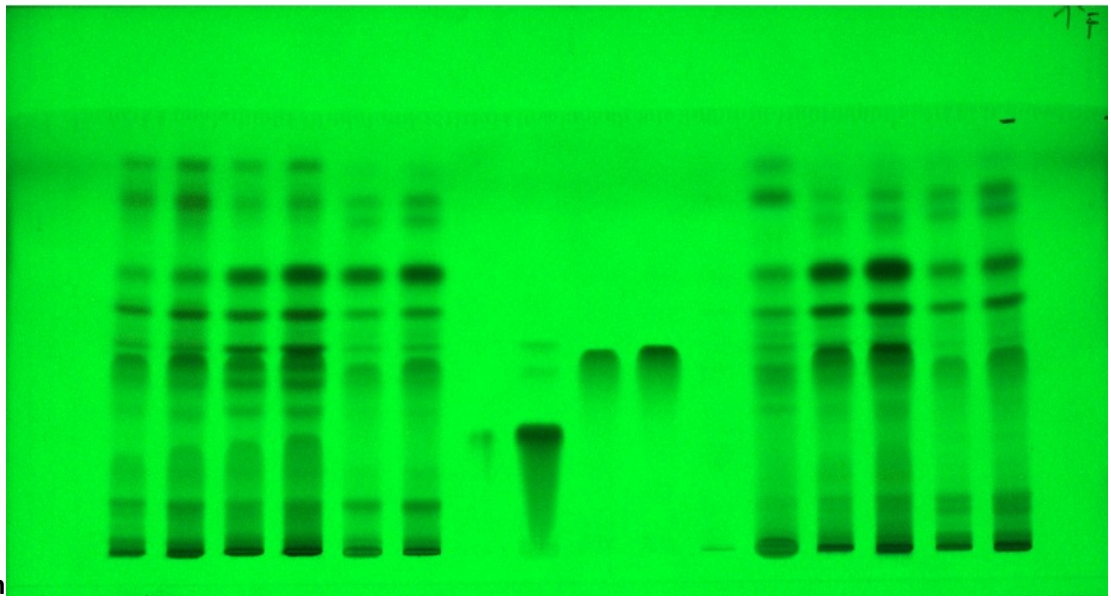
| Sample | Leaf | | Stem | | Root | |
|---|---|---|---|---|---|---|
| | 5 µl | 10 µl | 10 µl | 20 µl | 10 µl | 20 µl |
| Western Ghat | 8 | 8 | 5 | 5 | 5 | 5 |
| North East | 7 | 9 | 1 | 6 | 4 | 4 |

**Table 3: Total number of Flavanoids observed in different parts of *Oroxylum indicum* (L.) Vent from two regions (Western Ghats and North East) at 540 nm**

| Sample | Leaf | | Stem | | Root | |
|---|---|---|---|---|---|---|
| | 5 µl | 10 µl | 10 µl | 20 µl | 10 µl | 20 µl |
| Western Ghat | 9 | 9 | 9 | 10 | 7 | 7 |
| North East | 10 | 10 | 3 | 10 | 6 | 6 |

**Plate 1, 2 and 3: HPTLC studies on the Flavanoids observed in different parts of *Oroxylum indicum* (L.) Vent from two regions (Western Ghats and North East) at 254nm, 366nm and 540 nm respectively**

Plate 1: **Flavonoids at**



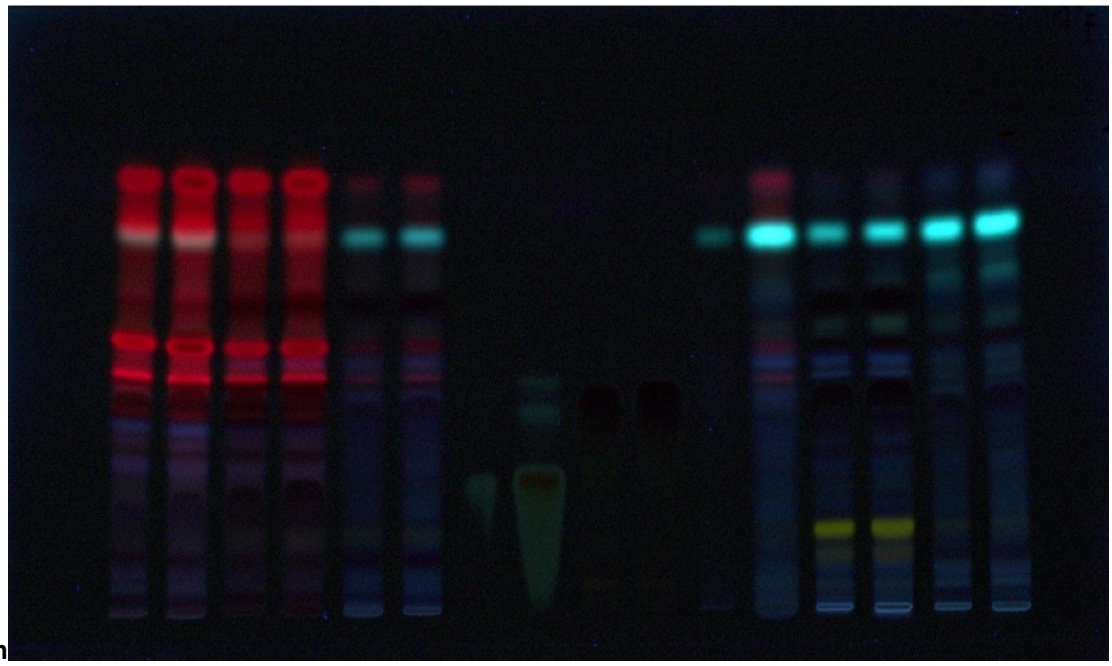
**254nm**

1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16



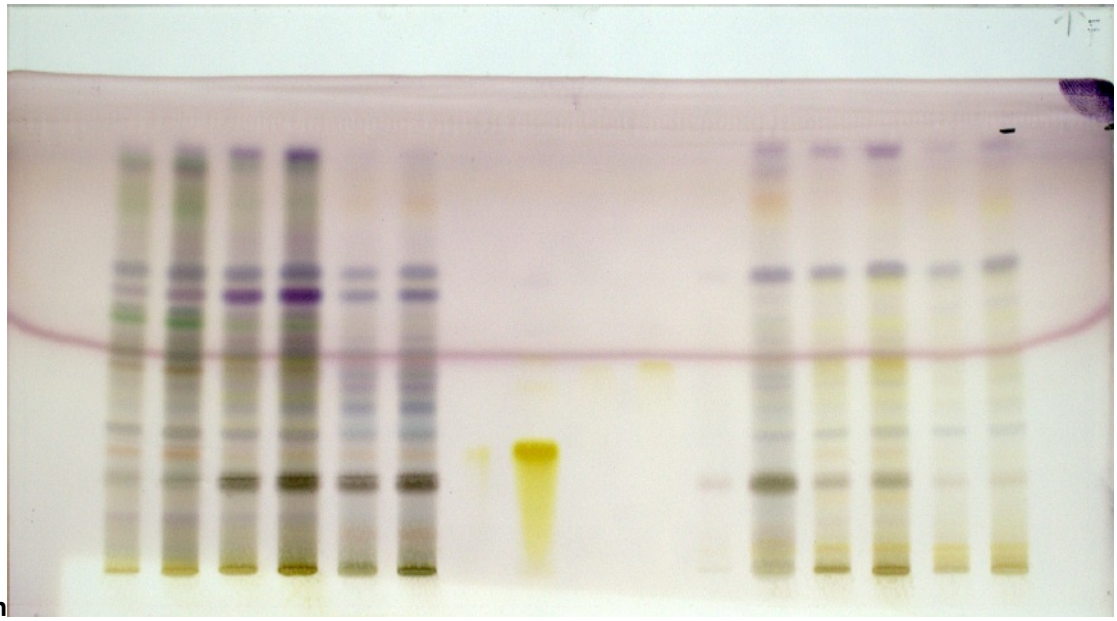
Plate 2: **Flavonoids at**



**366nm**

1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16

Plate 3: **Flavonoids at**



**540nm**

1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16

Track No 1 – WG Leaf (5 µl), 2 – WG Leaf (10 µl) , 3 – NE Leaf (5 µl), 4 - NE Leaf (10 µl), 5 -WG Stem ( 10 µl), 6- WG Stem ( 20 µl),7-Quercetin( 2 µl), 8 –Quercetin( 5 µl), 9 – Baicalein(2 µl) , 10 – Baicalein ( 5µl), 11- NE Stem(10 µl), 12- NE Stem(20 µl), 13- WG Root (10 µl), 14- WG Root (20 µl), 15- NE Root (10 µl), 16- NE Root (20 µl).



**Figure 13, 14 and 15: 3D display of HPTLC chromatogram of Flavanoids of *Oroxylum indicum* (L.) Vent (Leaf, stem and roots) collected from different geographical regions at wavelenth 254nm, 366nm and 540nm respectively.**

**Figure 13: (254nm)**

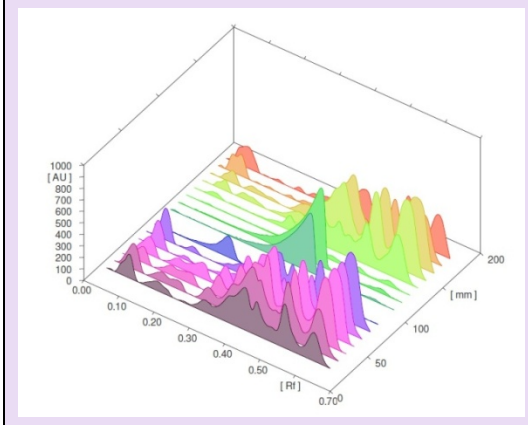



**Figure 14: (366nm)**

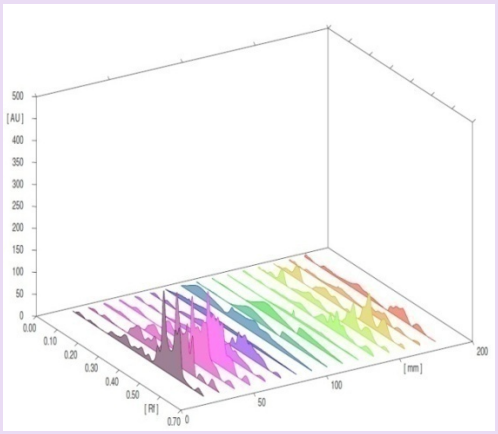
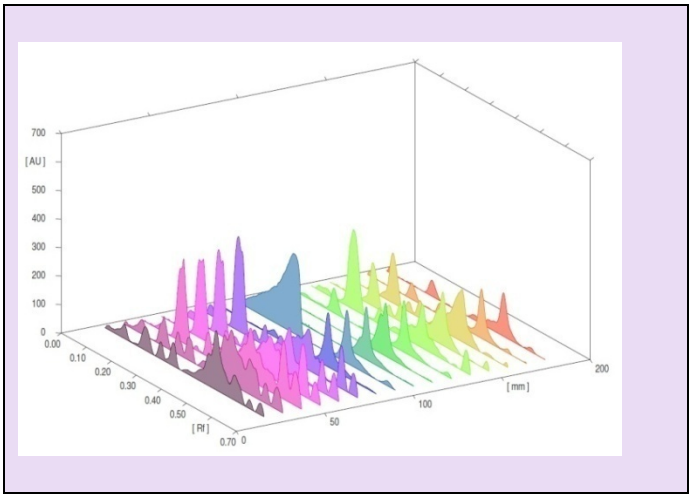



**Figure 15: (540nm)**




## CONCLUSION

The work carried out for the development of chromatographic fingerprint of *Oroxylum indicum* (L.) Vent demonstrate that HPTLC technique was successfully used for evaluating regional variations and quantitate chemical markers in the herbal raw material. The data also reveals that every plant part shows a presence of wide array of Flavanoids including Quercetin and Baicalein. Standardisation of plant materials is the need of the day. An HPTLC fingerprint is suitable for rapid and simple authentication. The HPTLC fingerprint developed may serve as a supplement chromatographic data and the information thus generated may be explored further as a tool for standardization.

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