



Research Article

PHYSICOCHEMICAL, PHYTOCHEMICAL AND HPTLC EVALUATION OF *EKANGAVEERARAS*, *KSHEERABALATAILA*, *BALAMULAKWATHA* AND *PANASAPATRA*

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KEYWORDS: Physicochemical, Phytochemicals, *Ekanagaveeraras*, *Ksheerabalataila*, *Panasapatra*, *Balamulakwath*.

ABSTRACT

Arditavata is a disease where *Mukhasankocha*, *Vakrata of Nasa*, *Bhru*, *Akshi*, *Lalata*, *Hanu* etc, will be seen. Mouth deviation, Deafness etc are the symptoms of Bell's palsy. The treatments like *Panasapatra*, *Ksheeradhuma* and *Ekanagaveeraras* has been tried on *Ardita*.

To highlight its mode of action accurately the Pharmacological analysis and HPTLC Study on the same helps in proper understanding and interpretation of drug action. This article enlightens about the Pharmacological analysis and HPTLC Study of *Ekanagaveeraras*, *Balamulakwatha churna*, *Ksheerabalataila* and *Panasapatra*.

As per the Analysis reports the presence of Carbohydrates, Steroids, Alkaloids, Flavonoids and Glycosides etc is positive. The pharmacologically active elements like carbohydrate, alkaloid and flavanoids are both Alcohol and water soluble whereas the others like steroids and glycosides are alcohol soluble extracts. Phytochemical Analysis of *Panasapatra Choorna* are both Water and alcohol soluble. Proteins, Tannins, triterpenoids and flavanoids are water soluble. Steroids alone are only alcohol soluble. The presence of these elements in the final product is directly proportional to the biological activity expressed by the product. With this in mind the Qualitative analysis of the final product holds good. By understanding of different active principles present in different formulation helps in explaining their mode of action scientifically.

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INTRODUCTION

Arditavata is one among *Vataja Nanatmaja vyadhi* explained by *Acharya Charaka*. *Acharya Sharangadhara* also said *Ardita* is one among *Vatavyadhi* and explained it's *Nirukti*. *Arditavata* is a disease where *Mukhasankocha*, *Vakrata of Nasa*, *Bhru*, *Akshi*, *Lalata*, *Hanu* etc, will be seen. Mouth deviation, Deafness etc. are the symptoms of Bell's palsy.

The patients are increasing and are approaching Ayurvedic doctor with lot of positive hopes. The lesion is within facial canal and may be due to reactivation herpes simplex virus/ infection. The treatments like *Panasapatra*, *Ksheeradhuma* and *Ekanagaveeraras* has been tried on *Ardita*. To highlight its mode of action accurately the Pharmacological analysis and HPTLC Study on the same helps in proper understanding and

interpretation the drug action. This article enlightens about the Pharmacological analysis and HPTLC Study of *Ekanagaveeraras*, *Balamulakwatha churna*, *Ksheerabalataila* and *Panasapatra*.

Ekanagaveeraras [1]

Ksheerabalataila [2]

Balamulakwathachurna [3, 4]

Panasapatra [5]

Determination of proximate values

a) Total Ash

Weigh accurately 2 gm of the air dried crude drug in a tared platinum or silica dish and incinerate at a temperature not exceeding 450°C until free from carbon, cool and weigh. If a carbon free ash cannot be obtained in this way, exhaust the charred mass with hot water, collect the residue on an ashless filter paper, incinerate the residue and

the filter paper until the ash is white or nearly so, add the filtrate, evaporate to dryness and ignite at a temperature not exceeding 450°C. Calculate the percentage of total ash with reference to the air dried drug.

b) Acid Insoluble ash

Boil the total ash obtained with 25 ml of 2M hydrochloric acid for 5 min, collect the insoluble matter in a Gooch crucible or on an ashless filter paper, wash with hot water, and ignite, cool, in a desiccator and weigh. The percentage of acid-insoluble ash with reference to the air dried drug was noted.

c) Sulphated ash

Weigh accurately 2 gm of the air dried crude drug in a tared platinum or silica dish and treat the powdered drug with dil. H₂SO₄ before incineration. Here all the oxides and carbonates are converted to sulphates. Then ignition is carried out at 600°C. The percentage of sulphated ash with reference to the air dried drug was calculated.

d) Water soluble ash

Boil the total ash obtained with 25 ml of distilled water for 5 min, filter and evaporate the filtrate cool, in a desiccator and weigh. The percentage of water soluble ash with reference to the air dried drug was noted.

e) Ethanol soluble extractive

Macerate 5 gm of air dried drug, coarsely powdered, with 100 ml of ethanol of the specified strength in a closed flask for 24 hrs, shaking frequently during the first 6 hrs and allowed to stand for 18 hrs. Thereafter filter rapidly taking precautions against loss of ethanol, evaporate 25 ml of the filtrate to dryness in tared flat-bottomed shallow dish, dry at 105°C and weigh. Calculate the percentage of ethanol soluble extractive with reference to the air dried drug.

f) Water soluble extractive

Add 5 gm of air dried crude drug to 50 ml of water at 80°C in a stoppered flask. Shake well and allow to stand for 10 mins, cool, add 2 gm of kieselghur and filter. Transfer 5 ml of the filtrate to a tared evaporating dish, 7.5 cm in a diameter, evaporate the solvent on a water bath, continue drying for 30 min, finally dry in a steam oven for 2 hrs and weigh the residue. Calculate the percentage of water-soluble extractive with reference to the air dried drug.

g) Loss on drying (Moisture content)

Weigh a glass stoppered, shallow weighing bottle that has been dried under the same conditions to be employed in the determination.

Transfer to the bottle 5 gm of crude drug, cover it and accurately weigh the bottle and the contents. Distribute the drug as evenly as practicable by gentle sidewise shaking to a depth not exceeding 10 mm. Place the loaded bottle in the drying chamber (oven or desiccator). Dry the drug to constant weight or for the specified time and at constant temperature. After drying is completed, open the drying chamber, close the bottle promptly and allow it to cool to room temperature in a desiccator before weighing. Weigh the bottle and the contents.

Preliminary Phytochemical investigations

The fractions were subjected to qualitative phytochemical investigation using following standard tests to identify the type(s) of phyto-constituents.

Test for carbohydrates

Molisch's test: Test solution + few drops of Molisch's reagent + 2ml of concentrated sulphuric acid along the sides of the test tube. A purple ring formed at the junction of two liquids indicates the presence of carbohydrates.

Fehling's test: Test solution + HCl heat + neutralize with NaOH + Fehling's solution A and B in equal proportions, Heat on water bath. Reddish brown precipitate indicates the presence of carbohydrates.

Barfoed's test: Test solution + Barfoed's reagent, boiled on water bath. Brick red precipitate indicates the presence of carbohydrates.

Benedict's test: Test solution + Benedict's reagent, boiled on water bath. Reddish brown precipitate indicates the presence of carbohydrates.

Test for sterols

Salkowaski test: Test solution + concentrated sulphuric acid, shaken and allowed to stand. The lower layer turns red indicating the presence of sterols.

Liebermann-Burchard test: Test solution + few drops of acetic anhydride. + Concentrated sulphuric acid along the sides of the test tube. Brown ring forms at the junction of the two liquids and the upper layer turns green.

Sulphur test: Sulphur when added to the test solution, it sinks to the bottom indicating the presence of sterols.

Test for alkaloids (general)

Mayer's test: Test solution + Mayer's reagent (Potassium mercuric iodide) gives cream coloured precipitate.

Wagner's test: The acidic test solution with Wagner's reagent (Iodine in potassium iodide) gives brown precipitate.

Hager's test: The acidic test solution with Hager's reagent (Saturated picric acid solution) gives yellow precipitate.

Dragendorff's test: The acidic test solution with Dragendorff's reagent (Potassium bismuth iodide) shows reddish brown precipitate.

Test for proteins and amino acids

Millon's test: Test solution + Millon's reagent, heated on a water bath. Yellow colouration indicates the presence of protein.

Xanthoproteic test: Test solution + concentrated nitric acid, on boiling gives yellow precipitate.

Biuret test: Test solution + 40% sodium hydroxide + dilute copper sulphate solution. Blue colour indicates the presence of protein.

Ninhydrin test: Test solution + Ninhydrin reagent gives blue colour.

Test for tannins

Ferric chloride test: Test solution + few drops of ferric chloride solution gives dark red colour.

Gelatin test: Test solution + gelatin solution gives white precipitate.

Test for Saponin glycosides

Foam test: Saponins when mixed with water and shaken shows the formation of froth, which is stable at least for 15 min.

Haemolysis test: 2 ml each of 18% sodium chloride solution was taken in two test tubes. To one test tube 2 ml of distilled water and to another test tube 2 ml of test sample was added. A few drops of blood

was added to both the test tubes, mixed and observed for haemolysis under microscope. Haemolysis of blood cells indicates the presence of saponin glycosides.

Test for triterpenoids

Salkowski test: Test solution + few drops of concentrated sulphuric acid, shaken and allowed to stand, lower layer turns yellow indicating the presence of triterpenoids.

Liebermann-Burchard test: Test solution + few drops of acetic anhydride. + Concentrated sulphuric acid along the sides of the test tube. Development of deep red colour indicates the presence of triterpenoids.

Test for flavanoids

Ferric chloride test: Test solution + few drops of ferric chloride solution give intense green colour.

Shinoda test: Test solution + few fragments of magnesium ribbon + concentrated hydrochloric acid, shows pink to magenta red colour.

Zinc-Hydrochloric acid reduction test: Test solution + zinc dust + few drops of hydrochloric acid shows magenta red colour.

Alkaline reagent test: Test solution + sodium hydroxide solution shows increase in the intensity of yellow colour which becomes colourless on addition of few drops of dilute acid.

Lead acetate solution test: Test solution + few drops of lead acetate (10%) solution gives yellow precipitate.

RESULTS

Proximate values (Values are average of two replicates) for Balamulakwathachurna

S.No.	Evaluation parameter	Value (%w/w)
01	Total ash value	4.3
02	Acid insoluble ash value	0.12
03	Water soluble ash value	0.05
04	Sulphated ash value	10
05	Alcohol soluble extractive value	8.6
06	Water soluble extractive value	7.5
07	Moisture content	12

Proximate values (Values are average of two replicates) for Panasaptra (Jack fruit leaf) powder

S.No.	Evaluation parameter	Value (%w/w)
01	Total ash value	12
02	Acid insoluble ash value	2.2
03	Water soluble ash value	1.1
04	Sulphated ash value	05
05	Alcohol soluble extractive value	9.4
06	Water soluble extractive value	12.5
07	Moisture content	10

Qualitative Phytochemical analysis of *Balamulakwathachurna*

S.No.	Name of the Phytoconstituent	Ethanol extract	Aq. extract
01	Carbohydrates	+	+
02	Steroids	+	-
03	Alkaloids	+	+
04	Proteins and Amino acids	-	-
05	Tannins	-	-
06	Saponins	-	+
07	Triterpenoids	-	+
08	Flavonoids	+	+
09	Glycosides	+	-

'+' = Present '-' = Absent

Qualitative Phytochemical analysis of *Panasapatra* (Jack fruit leaf) powder

S.No.	Name of the phytoconstituent	Ethanol extract	Aq. extract
01	Carbohydrates	+	+
02	Steroids	+	-
03	Alkaloids	+	+
04	Proteins and Amino acids	-	+
05	Tannins	-	+
06	Saponins	+	+
07	Triterpenoids	-	+
08	Flavanoids	-	+

'+' = Present '-' = Absent

Qualitative phytochemical analysis of *KsheerabalaTaila*

S.No.	Name of the phytoconstituent	Observations
01	Carbohydrates	+
02	Steroids	+
03	Alkaloids	+
04	Proteins and amino acids	-
05	Tannins	-
06	Saponins	+
07	Triterpenoids	-
08	Flavanoids	-
09	Cardiac glycosides	+
10	Lipids	+

'+' = Present '-' = Absent

Qualitative phytochemical analysis of *Ekangaveeraras*

S.No.	Name of the phytoconstituent	Observations
01	Carbohydrates	+
02	Steroids	+
03	Alkaloids	+
04	Proteins and Amino acids	-
05	Tannins	-
06	Saponins	+
07	Triterpenoids	+
08	Flavanoids	+
09	Coumarin glycosides	-

'+' = Present '-' = Absent

HPTLC

Ksheerabalataila

10.0 ml of *Ksheerabalataila* sample was partitioned with 20.0ml of methanol in a separating funnel and methanol soluble portion was made upto 10.0ml. 3, 6, 9 μ l of the sample were applied on a pre-coated silica gel F₂₅₄ on aluminum plates using Linomat 5 TLC applicator. The plate was developed in Toluene: Ethyl acetate (9:1) in developing chamber. The developed plate was visualized and scanned at UV 254 nm and 366 nm. Then it was dipped in Vanillin sulphuric acid reagent followed by heating at 105 °C till the spots appeared. The R_f, colour of the spots and densitometric scan were recorded.

Balamoolakwathachurna

1gm of *Balamulakwathachurna* sample was kept in a stoppered conical flask with methanol for cold percolation for 24hrs, followed by filtration and volume was made upto 10.0ml with methanol. 3, 6, 9 μ l of *Balamoolakwathachurna* were applied on a pre-coated silica gel F₂₅₄ on aluminum plates using Linomat 5 TLC applicator. The plate was developed in Toluene: Ethyl acetate (9:1) in developing chamber. The developed plate was visualized and scanned at UV 254 nm and 366 nm. Then it was dipped in Vanillin sulphuric acid reagent followed by heating at 105 °C till the spots appeared. The R_f, colour of the spots and densitometry scan were recorded.

Ekangaveerarasa tablet

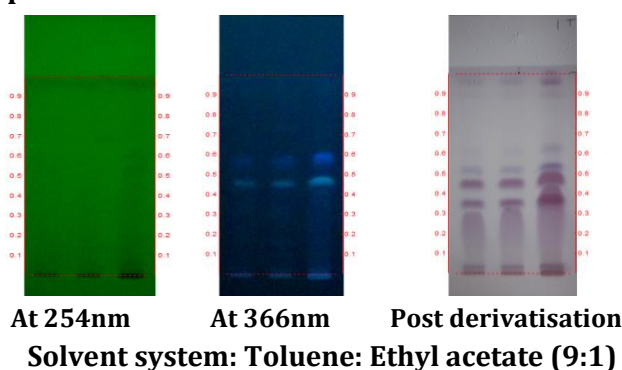
1gm of *Ekangaveera rasa* tablet powder was kept in a stoppered conical flask with methanol for cold percolation for 24hrs, followed by filtration and volume was made upto 10.0ml with methanol. 4, 8, 12 μ l of of the sample were applied on a pre-coated silica gel F₂₅₄ on aluminum plates using Linomat 5 TLC applicator. The plate was developed in Ethyl acetate: Toluene (9: 1) in developing chamber. The developed plate was visualized and scanned at UV 254 nm and 366 nm. Then it was dipped in Vanillin sulphuric acid reagent followed by heating at 105 °C till the spots appeared. The R_f, colour of the spots and densitometric scan were recorded.

Panasapatra

1gm of *Panasapatra* leaf powder was kept in a stoppered conical flask with methanol for cold percolation for 24hrs, followed by filtration and volume was made upto 10.0ml with methanol. 4, 8, 12 μ l of the sample were applied on a pre-coated silica gel F₂₅₄ on aluminum plates using Linomat 5 TLC applicator. The plate was developed in Ethyl acetate: Toluene (9: 1) in developing chamber. The developed plate was visualized and scanned at UV 254 nm and 366 nm. Then it was dipped in Vanillin sulphuric acid reagent followed by heating at 105 °C till the spots appeared. The R_f, colour of the spots and densitometric scan were recorded.

Part C: Results

Figure 1: HPTLC photo documentation of methanolic extract of *Ksheerabalataila*



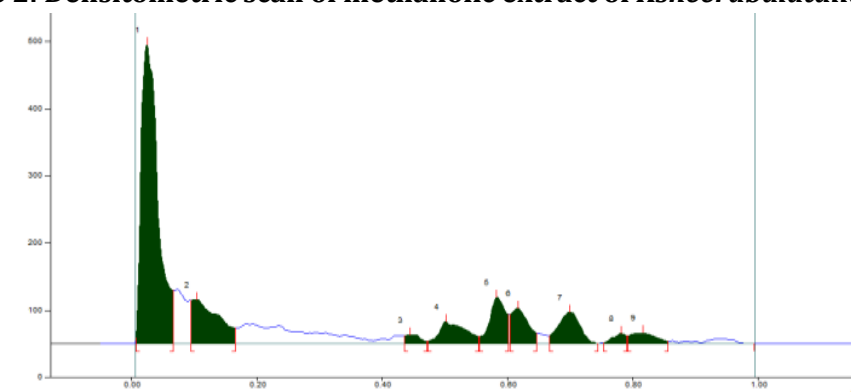
Track 1: *Ksheerabalataila* (3 μ l); Track 2: *Ksheerabalataila* (6 μ l); Track 3: *Ksheerabalataila* (9 μ l)

Table 1: R_f values of *Ksheerabalataila*

254nm	366nm	Post derivatisation
-	-	0.04 (L. pink)
-	-	0.36 (D. pink)
-	0.46 (FL. blue)	0.46 (D. pink)
-	-	0.52 (D. blue)
0.54 (L. green)	-	-
-	0.57 (FD. blue)	-

0.61 (L. green)	-	-
-	-	0.63 (L. pink)
-	-	0.91 (L. pink)
-	-	0.96 (D. pink)

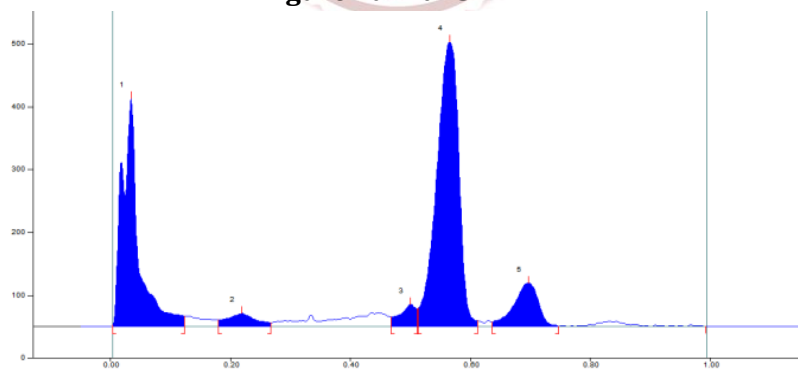
Figure 2: Densitometric scan of methanolic extract of *Ksheerabalataila* (9µl)



Track 3, ID: Ksheerabala taila

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.01 Rf	8.2 AU	0.03 Rf	445.2 AU	58.82 %	0.07 Rf	78.8 AU	8650.3 AU	53.75 %
2	0.10 Rf	64.4 AU	0.10 Rf	65.8 AU	8.70 %	0.17 Rf	23.5 AU	2061.3 AU	12.81 %
3	0.44 Rf	11.5 AU	0.45 Rf	13.3 AU	1.75 %	0.47 Rf	3.0 AU	231.7 AU	1.44 %
4	0.47 Rf	3.2 AU	0.50 Rf	33.2 AU	4.38 %	0.55 Rf	10.2 AU	981.5 AU	6.10 %
5	0.56 Rf	10.3 AU	0.58 Rf	68.9 AU	9.10 %	0.60 Rf	43.6 AU	1257.9 AU	7.82 %
6	0.60 Rf	43.6 AU	0.62 Rf	52.5 AU	6.94 %	0.65 Rf	15.6 AU	1050.4 AU	6.53 %
7	0.67 Rf	11.5 AU	0.70 Rf	46.8 AU	6.19 %	0.75 Rf	0.0 AU	1167.5 AU	7.25 %
8	0.75 Rf	1.4 AU	0.78 Rf	15.3 AU	2.02 %	0.79 Rf	11.3 AU	227.1 AU	1.41 %
9	0.79 Rf	11.6 AU	0.82 Rf	15.9 AU	2.10 %	0.86 Rf	3.6 AU	467.3 AU	2.90 %

Figure 2a: At 254nm

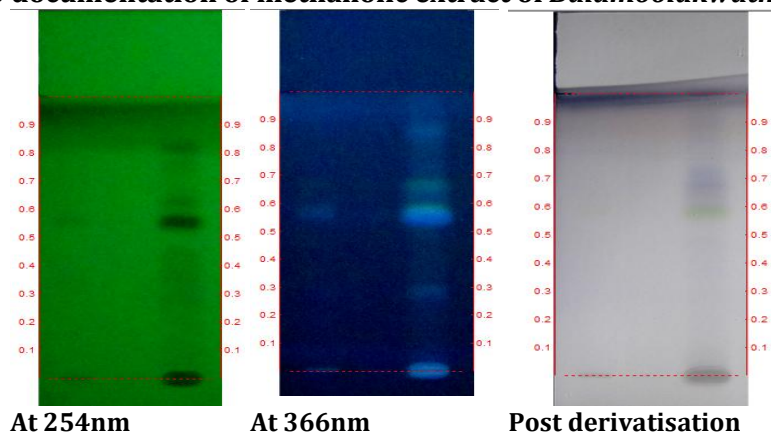


Track 3, ID: Ksheerabala taila

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.00 Rf	2.5 AU	0.04 Rf	363.0 AU	38.57 %	0.12 Rf	16.2 AU	7190.8 AU	31.84 %
2	0.18 Rf	10.0 AU	0.22 Rf	20.7 AU	2.20 %	0.27 Rf	6.1 AU	712.8 AU	3.16 %
3	0.47 Rf	15.4 AU	0.50 Rf	35.4 AU	3.76 %	0.51 Rf	28.3 AU	690.2 AU	3.06 %
4	0.51 Rf	29.0 AU	0.57 Rf	452.8 AU	48.11 %	0.61 Rf	8.8 AU	11946.9 AU	52.91 %
5	0.64 Rf	6.4 AU	0.70 Rf	69.2 AU	7.35 %	0.75 Rf	0.4 AU	2040.5 AU	9.04 %

Figure 2b: At 366nm

Figure 3: HPTLC photo documentation of methanolic extract of *Balamoolakwathachurna*



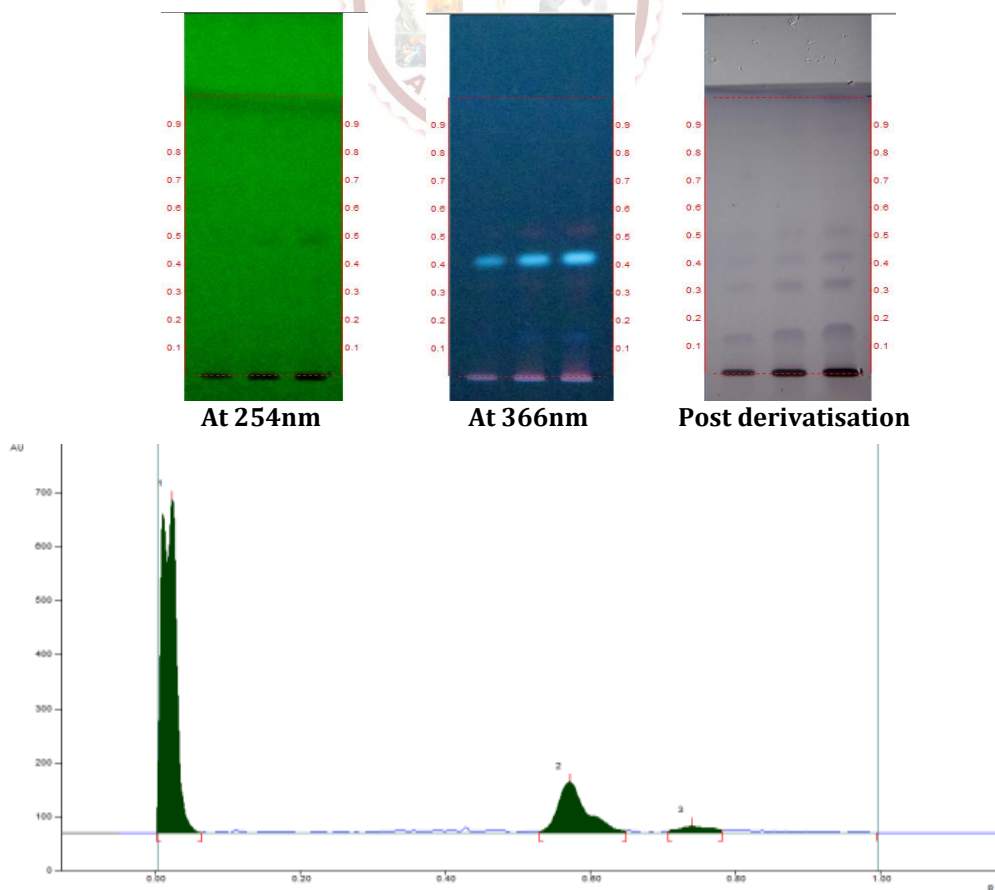
Solvent system: Toluene: Ethyl acetate (9:1)

Track 1: *Balamoolakwatha churna* (3 μ l); Track 2: *Balamoolakwatha churna* (6 μ l); Track 3: *Balamoolakwatha churna* (9 μ l)

Table 2: Rf values of *Balamoolakwathachurna*

254nm	366nm	Post derivatisation
-	0.15 (FD. Blue)	0.15 (D. Purple)
-	-	0.32 (D. Purple)
-	0.34 (FD. Red)	-
-	0.41 (FL. blue)	0.41 (D. Purple)
0.49 (L. green)	-	-
-	0.51 (FD. Red)	0.51 (D. Purple)
-	-	0.89 (D. Purple)

Figure 4: Densitometric scan of methanolic extract of *Balamoolakwathachurna* (9 μ l)

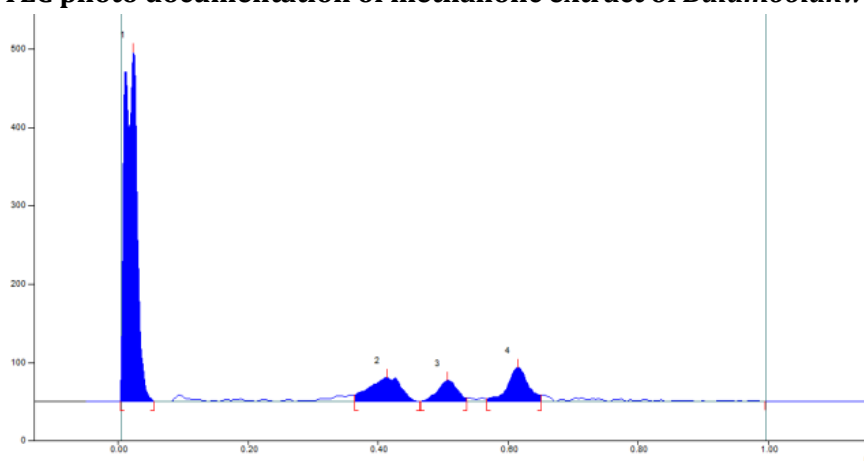


Track 7, ID: Balamoola kwatha churna

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.00 Rf	0.0 AU	0.02 Rf	617.4 AU	85.11 %	0.06 Rf	0.7 AU	9086.2 AU	74.61 %
2	0.53 Rf	3.1 AU	0.57 Rf	94.5 AU	13.03 %	0.65 Rf	3.8 AU	2676.0 AU	21.97 %
3	0.71 Rf	2.9 AU	0.74 Rf	13.5 AU	1.86 %	0.78 Rf	5.2 AU	416.6 AU	3.42 %

Figure 4a: 254nm

Figure 3: HPTLC photo documentation of methanolic extract of *Balamoolakwathachurna*



Track 7, ID: Balamoola kwatha churna

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.00 Rf	0.0 AU	0.02 Rf	445.9 AU	81.63 %	0.06 Rf	0.4 AU	6169.3 AU	70.22 %
2	0.36 Rf	8.6 AU	0.41 Rf	30.4 AU	5.57 %	0.46 Rf	0.1 AU	1058.6 AU	12.05 %
3	0.47 Rf	0.2 AU	0.51 Rf	26.9 AU	4.92 %	0.54 Rf	4.5 AU	574.2 AU	6.54 %
4	0.57 Rf	3.2 AU	0.62 Rf	43.0 AU	7.88 %	0.65 Rf	8.0 AU	983.8 AU	11.20 %

Figure 4b: 366nm

Figure 5: HPTLC photo documentation of methanolic extract of *Ekangaveerarasa tab*

Solvent system: Ethyl acetate : Toluene (9:1)

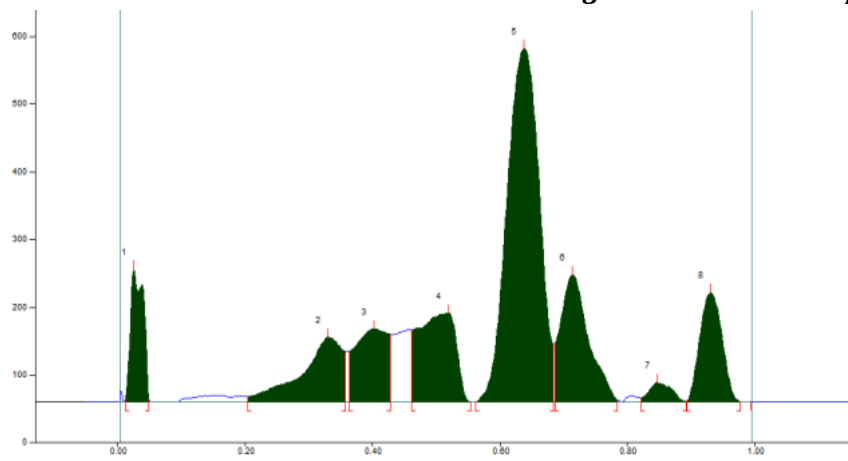
Track 1: *Ekangaveerarasa tab* (4µl); Track 2: *Ekangaveerarasa tab* (8µl); Track 3: *Ekangaveerarasa tab* (12µl)

Table 3: Rf values of *Ekangaveerarasa tab*

254nm	366nm	Post derivatisation
-	0.30 (FL. Blue)	-
0.56 (D. Green)	0.56 (FD. Blue)	-
-	0.59 (FD. green)	0.59 (Green)
0.63 (L. Green)	-	-
-	-	0.65 (L. purple)
-	0.67 (FD. green)	-

-	-	0.69 (D. purple)
-	-	0.74 (D. purple)
0.77 (L. Green)	-	-
0.84 (D. Green)	-	0.84 (L. purple)
-	0.86	-
-	-	0.91 (L. purple)

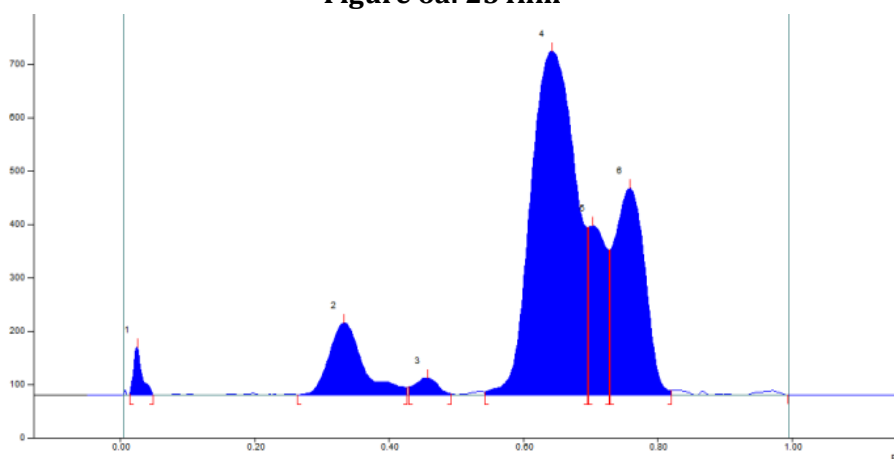
Figure 6: Densitometric scan of methanolic extract of *Ekangaveerarasa* tablet powder (12µl)



Track 3, ID: Ekangaveera rasa tablet

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.01 Rf	4.1 AU	0.03 Rf	195.9 AU	13.70 %	0.05 Rf	4.2 AU	2869.1 AU	6.17 %
2	0.20 Rf	8.0 AU	0.33 Rf	95.8 AU	6.70 %	0.36 Rf	74.8 AU	4346.6 AU	9.35 %
3	0.36 Rf	74.7 AU	0.40 Rf	107.9 AU	7.55 %	0.43 Rf	99.9 AU	3979.5 AU	8.56 %
4	0.46 Rf	106.4 AU	0.52 Rf	130.6 AU	9.13 %	0.55 Rf	0.1 AU	5584.9 AU	12.02 %
5	0.56 Rf	0.1 AU	0.64 Rf	522.2 AU	36.52 %	0.69 Rf	84.9 AU	18972.8 AU	40.82 %
6	0.69 Rf	86.2 AU	0.71 Rf	187.8 AU	13.13 %	0.79 Rf	3.3 AU	5960.2 AU	12.82 %
7	0.82 Rf	6.0 AU	0.85 Rf	28.2 AU	1.97 %	0.89 Rf	1.2 AU	728.8 AU	1.57 %
8	0.90 Rf	1.9 AU	0.93 Rf	161.5 AU	11.29 %	0.98 Rf	0.3 AU	4040.8 AU	8.69 %

Figure 6a: 254nm

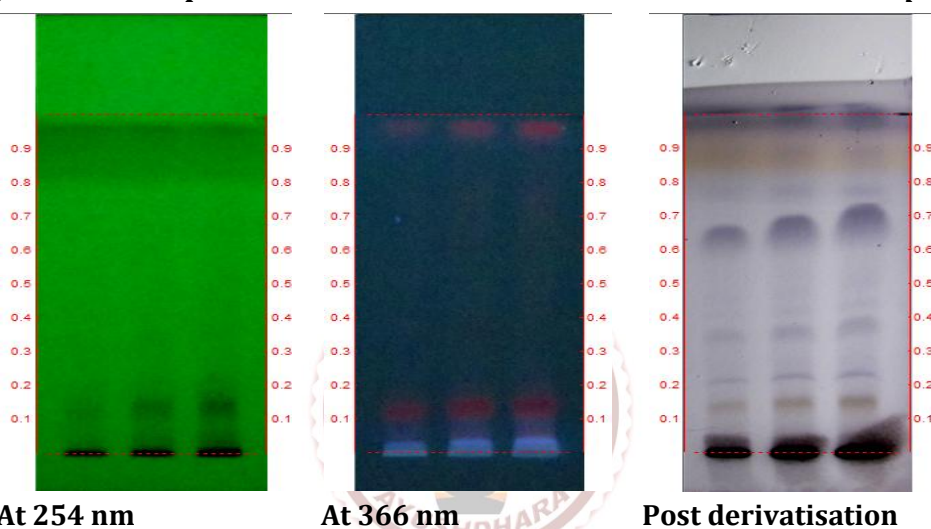


Track 3, ID: Ekangaveera rasa tablet

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.01 Rf	2.6 AU	0.03 Rf	89.7 AU	5.59 %	0.05 Rf	0.7 AU	805.9 AU	1.45 %
2	0.26 Rf	0.5 AU	0.33 Rf	134.6 AU	8.40 %	0.43 Rf	13.9 AU	4944.3 AU	8.88 %
3	0.43 Rf	15.0 AU	0.46 Rf	32.1 AU	2.00 %	0.49 Rf	2.6 AU	782.9 AU	1.41 %
4	0.54 Rf	5.9 AU	0.64 Rf	643.5 AU	40.14 %	0.70 Rf	13.7 AU	30664.0 AU	55.06 %
5	0.70 Rf	314.1 AU	0.70 Rf	316.2 AU	19.73 %	0.73 Rf	71.2 AU	5984.1 AU	10.75 %
6	0.73 Rf	271.8 AU	0.76 Rf	386.8 AU	24.13 %	0.82 Rf	8.6 AU	12507.5 AU	22.46 %

Figure 6b: 366nm

Figure 7: HPTLC photo documentation of methanolic extract of *Panasapatra*



At 254 nm

At 366 nm

Post derivatisation

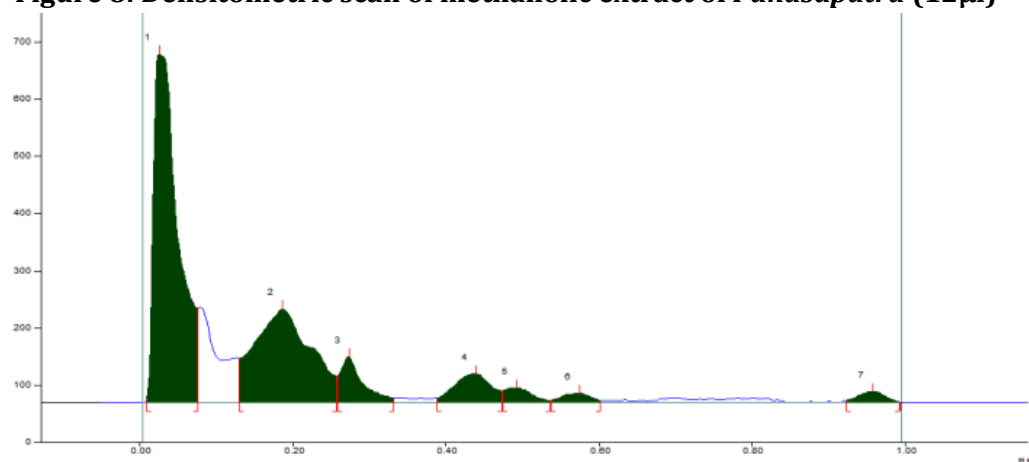
Solvent system: Ethyl acetate: Toluene (9:1)

Track 1: *Panasapatra* (4µl) Track 2: *Panasapatra* (8µl); Track 3: *Panasapatra* (12µl)

Table 4: Rf values of *Panasapatra*

254nm	366nm	Post derivatisation
0.14 (D. Green)	0.14 (FD. Red)	-
-	-	0.23 (D. Purple)
-	-	0.28 (L. Purple)
0.35 (L. Green)	-	0.36 (D. Purple)
-	-	0.43 (L. Purple)
-	-	0.50 (L. Purple)
-	-	0.65 (D. Purple)
-	0.78 (FL. pink)	0.78 (D. Purple)
-	-	0.86 (D. Purple)
-	-	0.91 (D. Purple)

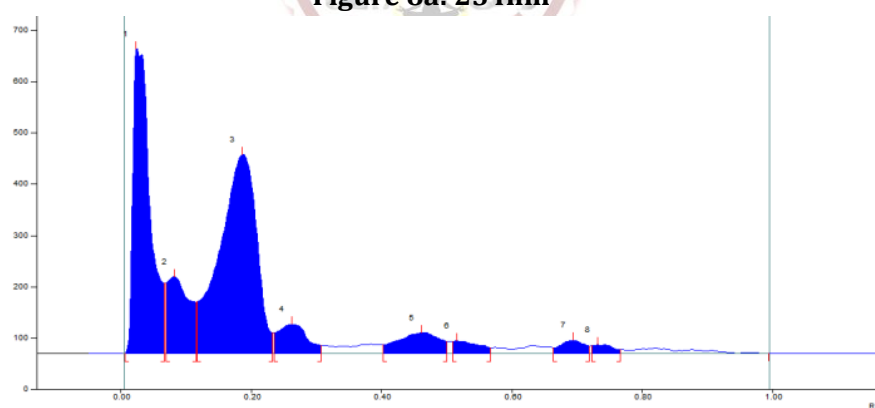
Figure 8: Densitometric scan of methanolic extract of *Panasapatra* (12µl)



Track 7, ID: Panasa patra

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.01 Rf	4.1 AU	0.03 Rf	608.6 AU	63.30 %	0.08 Rf	65.3 AU	14180.3 AU	51.30 %
2	0.13 Rf	76.8 AU	0.19 Rf	162.7 AU	16.92 %	0.26 Rf	45.9 AU	8560.8 AU	30.97 %
3	0.26 Rf	47.1 AU	0.27 Rf	79.8 AU	8.30 %	0.33 Rf	7.1 AU	1644.4 AU	5.95 %
4	0.39 Rf	7.6 AU	0.44 Rf	50.1 AU	5.21 %	0.47 Rf	20.7 AU	1716.1 AU	6.21 %
5	0.48 Rf	20.8 AU	0.49 Rf	25.2 AU	2.62 %	0.54 Rf	3.3 AU	636.1 AU	2.30 %
6	0.54 Rf	3.8 AU	0.58 Rf	16.3 AU	1.70 %	0.60 Rf	2.8 AU	428.9 AU	1.55 %
7	0.92 Rf	3.7 AU	0.96 Rf	18.8 AU	1.96 %	0.99 Rf	0.3 AU	476.7 AU	1.72 %

Figure 8a: 254nm



Track 7, ID: Panasa patra

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.01 Rf	0.1 AU	0.02 Rf	593.5 AU	45.83 %	0.07 Rf	36.6 AU	11413.3 AU	31.32 %
2	0.07 Rf	137.0 AU	0.08 Rf	149.5 AU	11.55 %	0.12 Rf	00.1 AU	3772.5 AU	10.35 %
3	0.12 Rf	100.5 AU	0.19 Rf	388.2 AU	29.98 %	0.23 Rf	40.0 AU	15759.8 AU	43.25 %
4	0.24 Rf	40.1 AU	0.26 Rf	56.9 AU	4.40 %	0.31 Rf	16.2 AU	1848.5 AU	5.07 %
5	0.40 Rf	16.9 AU	0.46 Rf	40.3 AU	3.11 %	0.50 Rf	23.3 AU	1875.0 AU	5.15 %
6	0.51 Rf	23.6 AU	0.52 Rf	24.2 AU	1.87 %	0.57 Rf	10.9 AU	711.2 AU	1.95 %
7	0.66 Rf	10.9 AU	0.70 Rf	25.7 AU	1.98 %	0.72 Rf	14.6 AU	676.8 AU	1.86 %
8	0.72 Rf	15.2 AU	0.73 Rf	16.7 AU	1.29 %	0.77 Rf	7.4 AU	381.2 AU	1.05 %

Figure 8b: 366nm

Part D: Remarks

HPTLC fingerprint *Ksheerabalataila*, *Balamoolakwathachurna*, *Ekangaveerarasa* and *Panasapatra* derived as per standard methodology* is documented in respective figures and tables. [6]

Interpretation of the Analysis***Balamoolakwatha choorna*****Total Ash Value**

The total Ash Value is calculated based on the % of ash with reference to the air dried drug.

It is the indicator of the presence / absence of the organic matter in the final product.

The present sample exhibits the value 4.3 for the total ash which is indicative of the presence of minimal amount of organic matter in the *Balamoola Kwatha Choorna*.

Acid (insoluble) and water (soluble) ash value

The acid insoluble ash value and water soluble ash value are both calculated with reference to the air dried drug.

The Acid insoluble ash value being 0.12 % is indicative of the maximum solubility of the *Balamoola Kwatha Choorna* in the acid.

The Water soluble ash value being 0.05 indicates that *Balamoola Kwatha Choorna* in almost soluble in water.

Alcohol and water Soluble Extractive value

The % of Alcohol and Water Soluble Extractive value is derived with reference to the air dried drug.

The only differentiating factor being the liquid media in both i.e. ethanol in former and chloroform in the latter.

The Extractive Value 8.6 and 7.5 depicts the utmost solubility of *BMK Choorna* in both alcohol and water respectively.

Panasapatra choorna**Total Ash Value**

The total Ash Value is calculated based on the % of ash with reference to the air dried drug.

It is the indicator of the presence / absence of the organic matter in the final product.

The present sample exhibits the value 12 for the total ash which is indicative of the presence of certain amount of Organic matter in the *Panasapatra Choorna*.

Note: In comparison *Balamoola Kwatha Choorna* has less organic matter than *Panasapatra Choorna*.

Acid (insoluble) and water (soluble) ash value

The acid insoluble ash value and water soluble ash value are both calculated with reference to the air dried drug.

The Acid insoluble ash value being 2.2 % is indicative of the maximum solubility of the *Panasapatra Choorna* in the acid.

The Water soluble ash value being 1.1 indicates that *Panasapatra Choorna* in almost soluble in water.

Note: The acid insoluble ash value and water soluble ash value is comparatively less in *Balamoola Kwatha Choorna* than *Panasapatra Choorna*.

Indicating the more solubility of *Balamoola Kwatha Choorna* than *Panasapatra Choorna*.

The Alcohol and water Soluble Extractive value

The % of Alcohol and Water Soluble Extractive value is derived with reference to the air dried drug.

The only differentiating factor being the liquid media in both i.e. ethanol in former and chloroform in the latter.

The Extractive Value 9.4 and 12.5 depicts the solubility of *Panasapatra Choorna* in both alcohol and water respectively.

Note: The Alcohol and water Soluble Extractive value of PNP is higher In comparison with the *Balamoola Kwatha Choorna*.

Moisture Content

The moisture content in the *Panasapatra Choorna* and *Balamoola Kwatha Choorna* is 10 and 12 respectively.

This shows the % of reactivity in the external atmosphere.

The *Panasapatra Choorna* is less reactive to the external atmosphere than the *Balamoola Kwatha Choorna* in comparison.

The Qualitative Phytochemical Analysis of *Balamoola Kwatha Choorna*

The presence of the phytoconstituents plays an important role in the expression of biological activity by the plant material.

As per the Analysis reports the presence of Carbohydrates, Steroids, Alkaloids, Flavonoids and Glycosides etc is positive. (Later bifurcated based on the solubility and insolubility in alcohol and water extracts.)

The pharmacologically active elements like carbohydrate, alkaloid and flavanoids are both Alcohol and water soluble whereas the others like steroids and glycosides are alcohol soluble extracts. Saponins and triterpenoids are water soluble extracts.

The Qualitative Phytochemical Analysis of *Panasapatra Choorna*

The isolated secondary metabolite otherwise known as the phytoconstituents are therapeutically significant and hence studied here.

Among the lot Carbohydrates, Alkaloids and saponins are both Water and alcohol soluble. Proteins, AA, Tannins, triterpenoids and flavanoids are water soluble. Steroids alone are only alcohol soluble.

The Qualitative Phytochemical Analysis of *Ksheerabalataila*

The Therapeutically active components present in the final product are Carbohydrates, Steroids, Alkaloids, Saponins, Cardiac glycosides and lipids.

The Qualitative Phytochemical Analysis of *Ekangaveerarasa*

The Therapeutically active components present in *Ekangaveerarasa* are Carbohydrates, Steroids, Alkaloids, Saponins and triterpenoids.

Note: In general there are macro elements, Microelements and Micronutrients as well. They are both Organic and Inorganic type. Amongst them are the carbohydrate, proteins, fat and other mineral elements.

Others are the secondary metabolites which are complex chemical substances metabolized by the plants. Amongst them are the Alkaloids, Glycosides, Flavanoids and so on. These are of significance due to their therapeutic value in them.

They are initially separated from the unwanted components present and are later isolated and studied.

The presence of these elements in the final product is directly proportional to the biological activity expressed by the product. With this in mind the Qualitative analysis of the final product holds good.

CONCLUSION

The presence of these elements in the final product is directly proportional to the biological activity expressed by the product. With this in mind the Qualitative analysis of the final product holds

good. By understanding of different active principles present in different formulation helps in explaining their mode of action scientifically.

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Dr. B Ravishankar M.Sc., Ph.D. Director, Dr. KN Sunil Kumar M.Sc., Ph.D, Senior Research Officer, Suchitra N prabhu, M.Pharm Trainee Research Officer. S.D.M. Centre for Research In Ayurveda And Allied Sciences (AYUSH Centre for Excellence and Recognized SIROs by DSIR) Laxminarayana Nagar, P.O. Kuthpady -574 118 UDUPI [Karnataka] (HPTLC fingerprint *Ksheerabalataila*, *Balamoola kwatha churna*, *Ekangaveerarasa* and *Panasapatra* derived as per standard methodology* is documented in respective figures and tables.

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