

Quality Standards of Polyherbal Powder Formulation *Safoof-e-Binai*

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ABSTRACT

Objective: To standardize *Safoof-e-Binai* (SB), a polyherbal Unani Powder preparation on the basis of organoleptic characters and physico-phytochemical analysis. **Methods:** The drugs were cleaned, dried in shade and powdered by passing through sieve # no. 80 as per the method described in UPI/ National Formulary of Unani Medicine. SB was evaluated using physicochemical tests: powder characterization, extractive value, alcohol and water-soluble matter, Ash value, LOD at 105°C, pH and HPTLC fingerprinting. Statistical analysis used: Mean \pm SEM. **Results:** Organoleptic characters of the formulation are Brownish green colour, characteristic odour, sweet and pungent taste and fine texture. Physicochemical parameters displayed water soluble extractive (11.23 \pm 0.04), alcohol soluble extractive (9.45 \pm 0.33), total ash (11.33 \pm 0.12), acid insoluble ash (0.29 \pm 0.02), water soluble ash (5.51 \pm 0.08), LOD at 105°C (11.40 \pm 0.04), pH of 1% and 10% solution were 7.33 \pm 0.33 and 7.66 \pm 0.33 respectively. Phytochemical qualitative analysis displayed presence of alkaloids, tannins, flavanoids, ter-

penoids, carbohydrates, volatile oil. HPTLC finger-printing data was also set in. **Conclusion:** The standardization of this formulation was done and the data obtained would be used as a standard for future reference.

Key words: Physicochemical, Poly-herbal, Formulation, Standardization, *Safoof-e-Binai*, Unani Medicine.

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INTRODUCTION

Standardization of herbal formulations is vitally important for evaluating the quality of medicines and its therapeutic value. Herbal medicines are prepared by using various plant materials like leaves, roots, barks, fruits and seeds, they carry various types of biologically active ingredients in it which helps for curing ailment.¹ In the preceding years, there has been more and more rapid increment in the ally of herbal and traditional medicines, as these drugs have obtained great demand in developed and progressing countries because of their natural origin. Traditional medicines are extracted or prepared from the natural resources like minerals, medicinal plants and organic matter. Many plant origin drugs are mixed together to prepare a formulation, there is a need to develop a monograph for standardization of these traditional formulations. *Safoof-e-Binai* (SB) is a polyherbal formulation used in the Unani System of Medicine for treatment of *Zofe ishteha* (Anorexia), *Zofemeda* (Stomach insufficiency) and also indicated in *Nazla* (Cold / Rhinitis) due to its antisecretory activity, its pharmacological action mentioned are *Habis* (Styptic), *Qabiz* (Astringent) and *Muhallil-e-Warm-e-Meda* (Stomach resolvent). The name of *Safoof-e-Binai* has been credited after the name of Sheikh Bina as mentioned in '*Biyaz Khas*'.^{2,3} The formulation is also mentioned in National Formulary of Unani Medicine,³ its ingredients are Ood saleeb (*Paeonia officinalis* Linn., Root), Qaranful (*Syzygium aromaticum* Linn., Flower), Nana (*Mentha viridis* Linn., Leaf) and Badiyan (*Foeniculum vulgare* Mill., Seeds). [Figure 1] Physicochemical evaluation of *Safoof-e-Binai* was attempted in this work to develop a quality standard. Review of literature does not suggest previous work related to physicochemical standards of SB.

MATERIALS AND METHODS

All the ingredients were procured from the local market and identification done by experts of National Institute of Unani Medicine, Bangalore. The drug *Safoof-e-Binai* was prepared as per the formulation composition given in NFUM, Part-IV.³ Analysis was performed as per pharmacopoeial prescribed parameters. Parameters for quality standards of the formulations includes colour, odour, taste identification, Loos on drying at 105°C, Ash value, acid insoluble ash value, water soluble ash value, pH value etc.⁴ Authentic finger print of SB was developed, with TLC /HPTLC with appropriate solvent system and extract by trial and error.

Preparation of formulation

All the medicines are initially cleaned and dried in shade and then made into powder form by passing through sieve #80. Preparations methodology was followed as per National Formulary of Unani Medicine.³ Ingredient of the formulation is depicted in [Table 1], [Figure 2].

Physico-chemical standardization

Organoleptic evaluation

Safoof-e-Binai (SB) formulation was evaluated for colour, odour, taste, appearance and texture.⁵

Powder characterization

Angle of repose

The angle of repose gives an indication of the flow ability of the substance. Funnel was adjusted such that the stem of the funnel lies 2 cm above the horizontal surface. The drug powder was allowed to flow from the fun-

nel under the gravitational force till the apex of the pile just touched the stem of the funnel, so the height of the pile was taken as 2 cm. Drawing boundary along the circumference of the pile and taking the average of six diameters determined the diameter of the pile. These values of height and diameter were then substituted in the following equation: Angle of Repose (θ) = $\tan^{-1}[2h/d]$. Where, h -Height of the pile and d -Diameter of the pile.

Bulk density and tapped density

The weighed quantity (20 gm) of SB was carefully put into a measuring cylinder without any losses. The initial volume was noted and the sample was then tapped until no further reduction in volume was noted. The initial volume gave the bulk density value and after tapping the volume gives the value of tapped density.

Hausner's ratio

Hausner ratio has been also used as indirect method of quantifying powder flowability from bulk density. Hausner ratio = D_t/D_b .

Where D_b = Bulk density and D_t = Tapped density. The experiment was done in triplicate.^{6,7}

Loss on drying at 105°C

An accurately weighed 5g of SB was taken in a petri dish. The crude drug was heated at 105°C in an oven till a constant weight and percentage moisture content of the sample was calculated with reference to the weighed SB sample.⁸

Total ash Water soluble ash and Acid insoluble ash (% w/w) was done as per method mentioned in Unani Pharmacopeia (UPI).⁸

Determination of pH

pH Strip method

We used pH indicator strips measuring a pH value in the range of 2.0 to 10.5 (Merck Specialities Pvt. Ltd, Mumbai). The original strips were cut into eight pieces (2 mm in width), which were easy to insert into the in-

terproximal site. After insertion for 10s, the pH value (One decimal) was assessed by comparing the color of the strip with the color index scheme supplied by the manufacturer.⁹

Extractive value (cold maceration)

Determination of alcohol and water-soluble extractive were done as per protocol for testing of Ayurvedic, Siddha and Unani Medicines.¹⁰

Successive extractive value

The coarse powder of SB was extracted successively using Soxhlet apparatus with different solvent, in increasing order of polarity, petroleum ether → benzene → chloroform → ethanol. 10 g powdered drug was taken and subjected to successive extraction with each solvent for 6 h. After that the extracts were filtered first by using filter paper (Whatman No. 1) and dried on water bath. The extractive values were determined with reference to the weight of the drug taken (w/w). The procedure was repeated 3 times to calculate mean extractive values.

Non-successive extractive value

The coarse powder of SB was extracted separately in different solvent (Water, ethyl alcohol and petroleum ether) using Soxhlet apparatus. 10 g powdered drug was taken and subjected to separate extraction with each solvent. The extracts were filtered first by using filter paper (Whatman No. 1) and evaporate on water bath. Extractive values were determined with reference to drug taken (w/w).¹¹

Qualitative estimation of constituents

Organoleptic features of the powdered medicines were scrutinized by using the standard chemical reagents and by this various phytochemical constituents were tested such as alkaloids, tannins, glycosides, resins, terpenes, flavonoids, carbohydrate and saponins.^{12,13}

HPTLC

The weighed quantity (10g) of *Safoof-e-Binai* (SB) was extracted in a Soxhlet apparatus for 6h using 100ml of solvent (Benzene, Chloroform and Ethanol) at a controlled temperature. HPTLC was performed on 20cm × 10cm aluminium backed plates coated with silica gel 60F254 (Merck, Mumbai, India). Standard solution of sample solution were applied to the plates as bands by use of a Camag (Muttens, Switzerland) Linomat V sample applicator equipped with a 100 µl Hamilton (USA) syringe. Ascending development to a distance of 80 mm was performed at room temperature (28±2°C) in respective mobile phases separately viz. Toluene: Ethyl acetate:Formic acid (5:4:1), Toluene: Ethyl acetate (12:3) and Toluene: Ethyl acetate (12:3)(v/v), in a Camag glass twin-trough chamber previously saturated with mobile phase vapour for 20 min. Af-

Table 1: Ingredient of Safoof-e-Binai.3

S. No	Unani name	Botanical name	Part used	Quantity
1.	Ood Saleeb	<i>Paeonia officinalis</i> Linn.	Root	2 gm
2.	Qaranful	<i>Syzygium aromaticum</i> Linn	Flower Bud	2 gm
3.	Nana	<i>Mentha viridis</i> Linn	Leaf	20 gm
4.	Badiyan	<i>Foeniculum vulgare</i> Mill	Seeds	20 gm

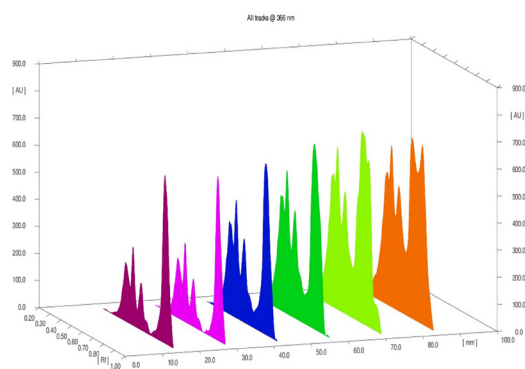


Figure 1: Ingridients of Safoof-e-Binai.



Figure 2: Safoof-e-Binai.

Table 2: Powder Characterization of *Safoof-e-Binai*.

S. No	Parameters	Percentage mean (n=3) ± SEM
1	Bulk Density (gm/ml)	0.285 ± 0.004
2	Tapped Density	0.377 ± 0.000
3	Hausner's ratio	1.320 ± 0.018
4	Carr's index	24.270 ± 1.082
5	Angle of repose θ	44° ± 0.577

Table 3: Physicochemical Characteristics of *Safoof-e-Binai*.

S. No	Parameters	Percentage mean (n=3) ± SEM
1	Loss on drying (%)	11.40 ± 0.04
2	Ash content	
(a)	Total ash (% w/w)	11.33 ± 0.12
(b)	Water soluble ash (% w/w)	5.51 ± 0.08
(c)	Acid insoluble ash (% w/w)	0.29 ± 0.02
3	pH	
(a)	pH 1%	7.33 ± 0.33
(b)	pH 10%	7.66 ± 0.33

Table 4: Successive Extraction and Non-Successive Extraction of SB.

Successive Extraction (% w/w)	
Petroleum ether	7.45 ± 0.05
Benzene	0.92 ± 0.03
Chloroform	0.99 ± 0.02
Ethanol	1.18 ± 0.04
Non-Successive Extraction (% w/w)	
A. Cold Extraction	
Water soluble	11.23 ± 0.04
Ethyl Alcohol	9.45 ± 0.33
B. Hot Extraction	
Petroleum ether	7.48 ± 0.05
Benzene	9.01 ± 0.04
Chloroform	12.33 ± 0.01
Ethanol	19.91 ± 0.03

ter development, the plates were dried and then scanned at 254 nm and 366 nm with a Camag TLC Scanner with WINCAT software, using the deuterium lamp.^{14,15}

RESULTS

The organoleptic properties of *Safoof-e-Binai* indicated the colour was Brownish green, odour was characteristic, taste was aromatic, pungent and sweet, texture was soft fine powder. The powder characterization of SB is depicted in [Table 2]. The physicochemical Evaluation of SB is mentioned in [Table 3] and water and alcohol soluble extractive values

Table 5: Phytochemical Screening of *Safoof-e-Binai*.

Parameters	Result
Alkaloids	+
Sugars	+
Phenols	+
Tannins	+
Proteins	+
Flavonoids	+
Essential oil and Volatile oil	+

were found to be 11.23±0.04 (%w/w) and 9.45±0.33 (%w/w) respectively. Successive and Non-Successive extraction, Phytochemical Screening and HPTLC of SB is depicted in [Table 4-11] respectively.

DISCUSSION

Standardization is an important measure for knowing the quality and purity of the formulation. It is essential for the identity of the materials. Finished product of *Safoof-e-Binai* (SB) was Greenish brown in colour as per colour chart (No. PMS 105 of Pantone color chart), taste was aromatic, pungent and sweet, texture characteristic in odor and without any clumping and aggregation.

The mean values of bulk density, tapped density, angle of repose, Hausner's ratio and compressibility index were 0.285±0.004, 0.377±0.000, 44°±0.577°, 320 ± 0.018 and 24.270 ± 1.082 respectively. Powder characterization methods are the simple and popular method to determine the flow characteristics of powder which depend on the size, shape, size distribution of particles and moisture content.¹⁶ The compressibility index of SB below 30, Hausner's ratio 1.25- 1.5 and Angle of repose displayed passable flow property.¹⁷ [Table 2]. The mean percentage of loss of weight on drying (Moisture content) of SB was 11.40 ± 0.04. [Table 3]. The presence of excessive amount of moisture in plant drugs causes hydrolysis of constituents, growth of bacteria/ fungi and biochemical reactions, excessive amounts of water causes deterioration of products.¹⁸ The ash value is an important parameter because a high ash value is indicative of contamination, substitution, adulteration or carelessness in preparing the drug or drug combinations. The total ash of SB was found to be 11.33±0.12 % w/w. is reasonably low indicating low contamination. Water-soluble ash is the part of the total ash content. It is a good indicator of either previous extraction of water-soluble salts in the drug or incorrect preparation. Thus, it is the difference in weight between the total ash and the residue obtained after treatment of total ash with water. The acid insoluble and water-soluble ash values of SB were 0.29 ± 0.02 % w/w and 5.51 ± 0.08 % w/w respectively [Table 3] pH of SB in 1% solution was 7.33 ± 0.33 while the pH of 10% solution was 7.66 ± 0.33. [Table 3] It is basic in nature.

The mean percentage of water-soluble and alcohol soluble extractive values of SB were 11.23±0.04 and 9.45±0.33 respectively. The mean percentage of successive extractive values in petroleum ether, benzene, chloroform and ethanol were 7.45±0.05, 0.92±0.03, 0.99±0.02 and 1.18±0.04 respectively. Non-successive extractive values in petroleum ether, benzene, chloroform and ethanol were 7.48±0.05, 9.01±0.04, 12.33±0.01 and 19.91 ± 0.03 respectively. [Table 4] Extractive value of a drug in definite solvent is an index for checking the purity of a drug. Organic constituent's viz. alkaloids, tannins, flavanoids, sugars, phenols, proteins were found present on qualitative estimation tests. [Table 5]

HPTLC plates of Benzene extract of *Safoof-e-Binai* (SB) in mobile phases viz. Toulene: Ethyl acetate: Formic acid (5:4:1) were examined. R_f value, numbers of peaks, peak area and peak height were analysed under 254nm and 366nm. [Figure 3, 4, 5, 7] Area percentage of peak no. 4 of SB analysed under 254 nm in Toulene: Ethyl acetate: Formic acid (5:4:1) was highest (57.67%). [Table 6] Area percentage of peak no. 4 of SB analysed under 366 nm in Toulene: Ethyl acetate: Formic acid (5:4:1) was highest (54.23%). [Table 7].

HPTLC plates of Chloroform extract of SB in mobile phases viz. Toulene: Ethyl acetate (12:3) were examined. R_f value, numbers of peaks, peak area and peak height of SB were analysed under 254nm and 366nm. [Figure 8-12] Area percentage of peak no. 2 of SB analysed under 254nm in Toulene: Ethyl acetate (12:3) was highest (28.09%). [Table 8] Area percentage of peak no. 1 of SB analysed under 366 nm in Toulene: Ethyl acetate (12:3) was highest (38.64 %). [Table 9].

HPTLC plates of Ethanol extract of SB in mobile phases viz. Toulene: Ethyl acetate (12:3) were examined. R_f value, numbers of peaks, peak

area and peak height of SB were analysed under 254nm, 366 nm. [Figure 13-17] Area percentage of peak no. 14 of SB analysed under 254nm in Toulene: Ethyl acetate (12:3) was highest (28.20%). [Table 10] Area percentage of peak no. 9 of SB analysed under 366nm in Toulene: Ethyl acetate (12:3) was highest (41.32%). [Table 11].

Further studies can be done by the help of standards for quantitative estimation and identification of the ingredients peak. Present HPTLC fingerprinting data can help in authentication and identification of SB in the tested solvent system and extract.

Paeoniflorin present in Ood Saleeb shows a smooth muscle relaxant, vasodilatory, anti-inflammatory, immune-stimulating and CNS depressant activity in animal studies. Eugenol, present in Qaraful is antibacterial. Essential oil of *Mentha viridis* are menthol possesses both antibacterial and antifungal properties.¹⁹ Review of literature on ingredient of SB revealed promising constituent and pharmacological activity. *Paeonia officinalis* contains many important constituents like glucoside, essential oil, fixed oil, tannin and terpene;²⁰ *Mentha viridis* contains essential oil (0.2

Table 6: R_f value, No. of Peaks, Peak area and Height of Benzene Extract of *Safoof-e-Binai* (SB) in Toulene: Ethyl Acetate: Formic Acid at 254nm.

Peak	Start position	Start height	Max position	Max Height	Max %	End position	End Height	Area	Area %
1	0.42 R_f	19.3 AU	0.48 R_f	150.2 AU	18.22	0.53 R_f	59.1 AU	6426.6 AU	19.36
2	0.53 R_f	59.4 AU	0.55 R_f	120.5 AU	14.62	0.59 R_f	0.4 AU	2767.1 AU	8.34
3	0.59 R_f	0.1 AU	0.63 R_f	161.8 AU	19.63	0.69 R_f	6.1 AU	4855.5 AU	14.63
4	0.73 R_f	0.0 AU	0.85 R_f	391.7 AU	47.53	0.90 R_f	10.5 AU	19143.0 AU	57.67

Table 7: R_f value, No. of Peaks, Peak Area and Height of Benzene Extract of *Safoof-e-Binai* (SB) in Toulene: Ethyl Acetate: Formic Acid at 366nm.

Peak	Start position	Start height	Max position	Max Height	Max %	End position	End Height	Area	Area %
1	0.40 R_f	13.0 AU	0.48 R_f	217.6 AU	16.73	0.53 R_f	135.1 AU	11008.7 AU	19.52
2	0.53 R_f	136.2 AU	0.55 R_f	289.7 AU	22.28	0.59 R_f	66.5 AU	8053.6 AU	14.28
3	0.59 R_f	66.6 AU	0.63 R_f	173.1 AU	13.31	0.71 R_f	5.9 AU	6750.5 AU	11.97
4	0.72 R_f	6.0 AU	0.85 R_f	620.3 AU	47.69	0.92 R_f	21.9 AU	30586.4 AU	54.23

Table 8: R_f Value, No. of Peaks, Peak Area and Height of Chloroform Extract of *Safoof-e-Binai* (SB) in Toulene: Ethyl Acetate: at 254nm.

Peak	Start position	Start height	Max position	Max Height	Max %	End position	End Height	Area	Area %
1	0.10 R_f	0.0 AU	0.13 R_f	42.0 AU	4.78	0.14 R_f	36.3 AU	717.5 AU	3.35
2	0.14 R_f	36.4 AU	0.17 R_f	264.4 AU	30.06	0.21 R_f	57.2 AU	6019.3 AU	28.09
3	0.21 R_f	57.5 AU	0.24 R_f	85.6 AU	9.74	0.27 R_f	0.1 AU	2030.7 AU	9.48
4	0.28 R_f	1.2 AU	0.31 R_f	120.4 AU	13.69	0.36 R_f	6.7 AU	2964.2 AU	13.84
5	0.36 R_f	7.0 AU	0.40 R_f	135.4 AU	15.40	0.46 R_f	2.2 AU	4411.9 AU	20.59
6	0.47 R_f	2.1 AU	0.52 R_f	97.7 AU	11.10	0.56 R_f	3.5 AU	1954.8 AU	9.12
7	0.57 R_f	0.6 AU	0.61 R_f	134.0 AU	15.23	0.65 R_f	0.2 AU	3326.8 AU	15.53

Table 9: R_f Value, No. of Peaks, Peak Area and Height of Chloroform Extract of *Safoof-e-Binai* (SB) in Toulene: Ethyl Acetate: at 366nm.

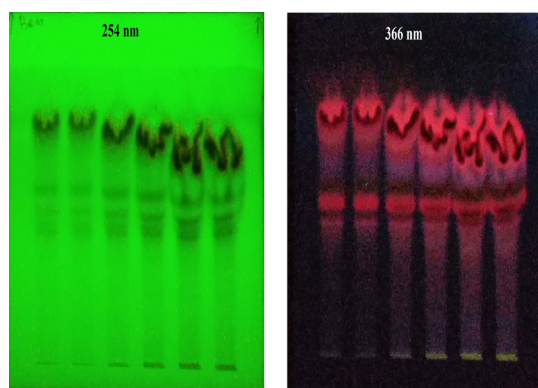
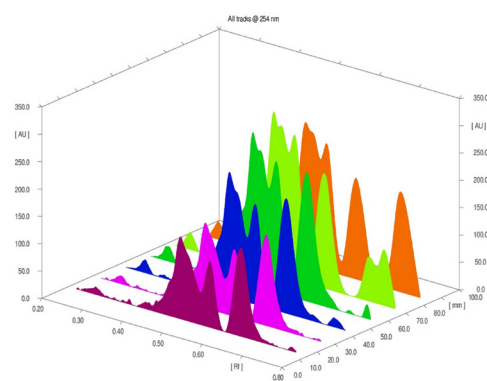
Peak	Start position	Start height	Max position	Max Height	Max %	End position	End Height	Area	Area %
1	0.10 R_f	0.5 AU	0.16 R_f	337.7 AU	38.30	0.20 R_f	35.6 AU	8278.4 AU	38.64
2	0.20 R_f	36.0 AU	0.22 R_f	77.4 AU	8.79	0.25 R_f	0.7 AU	1420.3 AU	6.63
3	0.26 R_f	0.3 AU	0.30 R_f	197.2 AU	22.37	0.32 R_f	51.4 AU	4201.5 AU	19.61
4	0.36 R_f	42.1 AU	0.41 R_f	88.5 AU	10.04	0.46 R_f	11.3 AU	3650.1 AU	17.03
5	0.46 R_f	11.5 AU	0.51 R_f	180.7 AU	20.49	0.58 R_f	1.5 AU	3876.8 AU	18.09

Table 10: R_f Value, No. of Peaks, Peak Area and Height of Ethanol Extract of *Safoof-e- Binai* (SB) in Toulene: Ethyl acetate: at 254nm.

Peak	Start position	Start height	Max position	Max Height	Max %	End position	End Height	Area	Area %
1	0.09 R_f	0.1 AU	0.11 R_f	56.4 AU	5.39	0.13 R_f	0.6 AU	663.6 AU	2.50
2	0.13 R_f	0.6 AU	0.14 R_f	33.5 AU	3.20	0.16 R_f	20.2 AU	408.1 AU	1.54
3	0.16 R_f	20.3 AU	0.19 R_f	200.2 AU	19.14	0.22 R_f	32.0 AU	4762.8 AU	17.97
4	0.24 R_f	32.7 AU	0.25 R_f	37.0 AU	3.54	0.28 R_f	0.1 AU	684.5 AU	2.58
5	0.29 R_f	0.7 AU	0.33 R_f	39.8 AU	3.81	0.35 R_f	16.9 AU	828.6 AU	3.13
6	0.35 R_f	17.1 AU	0.36 R_f	24.6 AU	2.35	0.39 R_f	8.6 AU	488.6 AU	1.84
7	0.39 R_f	8.8 AU	0.43 R_f	40.8 AU	3.90	0.47 R_f	4.9 AU	1231.1 AU	4.65
8	0.51 R_f	0.3 AU	0.53 R_f	22.1 AU	2.11	0.54 R_f	14.1 AU	300.6 AU	1.13
9	0.55 R_f	14.3 AU	0.56 R_f	20.2 AU	1.93	0.61 R_f	0.1 AU	517.7 AU	1.95
10	0.62 R_f	1.0 AU	0.67 R_f	33.4 AU	3.19	0.68 R_f	30.3 AU	750.0 AU	2.83
11	0.68 R_f	27.5 AU	0.72 R_f	68.7 AU	6.56	0.74 R_f	63.4 AU	1979.1 AU	7.47
12	0.74 R_f	63.4 AU	0.76 R_f	89.1 AU	8.52	0.78 R_f	62.2 AU	2279.8 AU	8.60
13	0.78 R_f	62.8 AU	0.81 R_f	153.6 AU	14.68	0.83 R_f	116.3 AU	4132.8 AU	15.60
14	0.83 R_f	117.0 AU	0.86 R_f	226.7 AU	21.67	0.93 R_f	2.4 AU	7471.6 AU	28.20

Table 11: R_f value, No. of Peaks, Peak Area and Height of Ethanol Extract of *Safoof-e- Binai* (SB) in Toulene: Ethyl acetate: at 366nm.

Peak	Start position	Start height	Max position	Max Height	Max %	End position	End Height	Area	Area %
1	0.02 R_f	3.9 AU	0.04 R_f	137.5 AU	8.01	0.07 R_f	0.0 AU	1153.8 AU	2.41
2	0.10 R_f	0.1 AU	0.16 R_f	328.5 AU	19.14	0.21 R_f	38.1 AU	9243.9 AU	19.27
3	0.27 R_f	1.6 AU	0.30 R_f	79.8 AU	4.65	0.33 R_f	38.8 AU	1888.4 AU	3.94
4	0.49 R_f	0.1 AU	0.52 R_f	66.1 AU	3.85	0.54 R_f	49.4 AU	982.1 AU	2.05
5	0.54 R_f	49.9 AU	0.54 R_f	54.5 AU	3.18	0.60 R_f	1.7 AU	1266.7 AU	2.64
6	0.62 R_f	1.7 AU	0.66 R_f	57.4 AU	3.34	0.68 R_f	23.2 AU	1193.6 AU	2.49
7	0.68 R_f	23.4 AU	0.76 R_f	174.3 AU	10.16	0.78 R_f	104.8 AU	5807.4 AU	12.11
8	0.78 R_f	107.0 AU	0.81 R_f	276.6 AU	16.12	0.82 R_f	237.9 AU	6610.6 AU	13.78
9	0.82 R_f	239.6 AU	0.86 R_f	541.3 AU	31.54	0.94 R_f	1.4 AU	19815.6 AU	41.32

**Figure 3:** HPTLC Photos of *Safoof-e-Binai* (SB) Benzene Extract in Toulene: Ethyl Acetate: Formic Acid at 254 nm and 366 nm.**Figure 4:** HPTLC 3-D Densitometric Scan of Benzene extract of *Safoof-e-Binai* (SB) in Toulene: Ethyl Acetate: Formic Acid at 254 nm.

to 0.8 percent), terpene such as carvone (60%) and limonene (10%) are major constituents.²¹ *Mesfin et al.* studied the anxiolytic activity of fennel on adult mice.²² *Al-Mofleh et al.* demonstrated protective effect of fennel on gastric ulcer with reduced mucosal lining of the stomach. These functions were attributed to its antioxidant capacity.²³ The antiviral activity of eugenin, a compound isolated from *S. aromaticum* and from *Geum*

japonicum, was tested against herpes virus strains being effective at 5 μg / mL and one of the major targets of eugenin is the viral DNA synthesis by the inhibition of the viral DNA polymerase²⁴ The most important lacunae in traditional / alternative system of medicine for its globalization is difficulty to ensure uniformity and quality of drugs. Powder dosage form SB doesn't have any pharmacopoeial standards since yet. Owing to

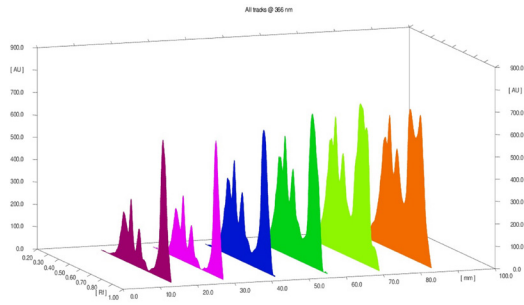


Figure 5: HPTLC 3-D Densitometric Scan of Benzene Extract of *Safoof-e-Binai* (SB) in Toulene: Ethyl Acetate: Formic Acid at 366 nm.

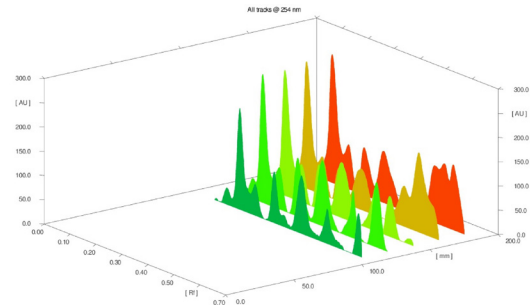


Figure 9: HPTLC 3-D Densitometric Scan of Chloroform Extract of *Safoof-e-Binai* (SB) in Toulene: Ethyl acetate at 254 nm.

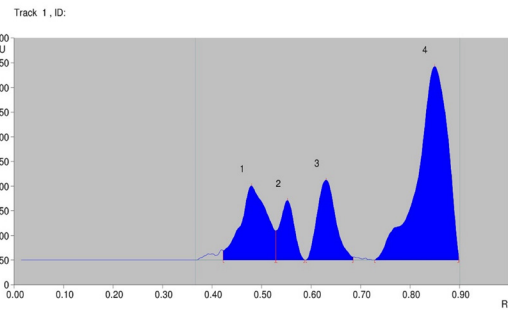


Figure 6: HPTLC fingerprint profile of Benzene Extract of *Safoof-e-Binai* (SB) in Toulene: Ethyl Acetate: Formic Acid at 254 nm

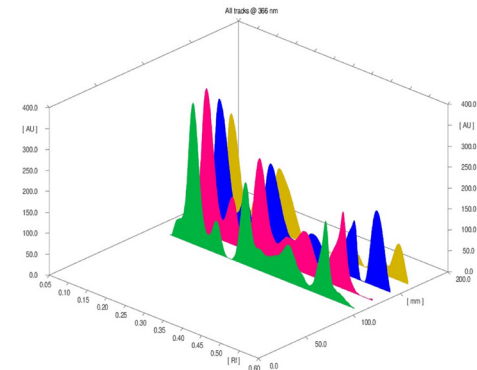


Figure 10: HPTLC 3-D Densitometric Scan of Chloroform Extract of *Safoof-e-Binai* (SB) in Toulene: Ethyl acetate at 366 nm.

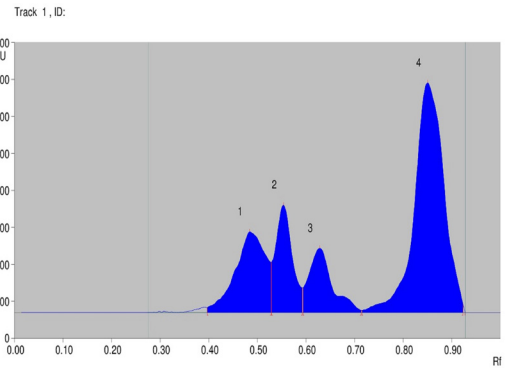


Figure 7: HPTLC fingerprint profile of Benzene Extract of *Safoof-e-Binai*(SB) in Toulene: Ethyl acetate: Formic acid at 366 nm.

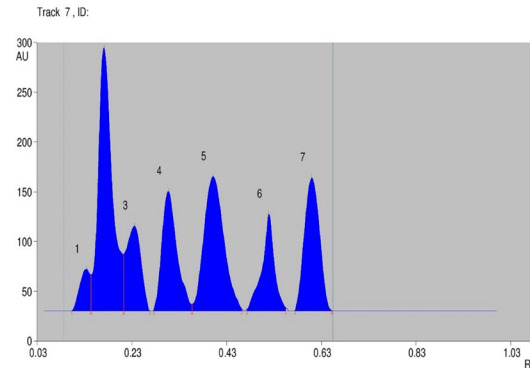


Figure 11: HPTLC Fingerprint Profile of Chloroform Extract of *Safoof-e-Binai* (SB) in Toulene: Ethyl acetate at 254 nm.

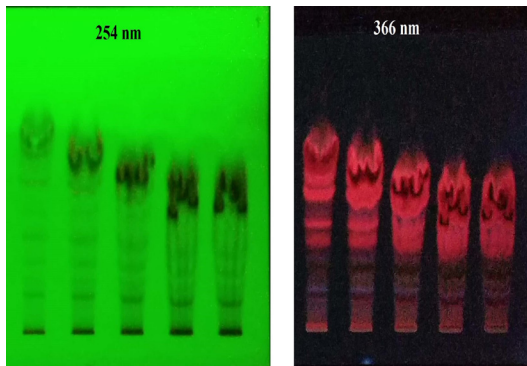


Figure 8: HPTLC Photos of *Safoof-e-Binai* (SB) Chloroform Extract in Toulene: Ethyl Acetate at 254 nm and 366 nm.

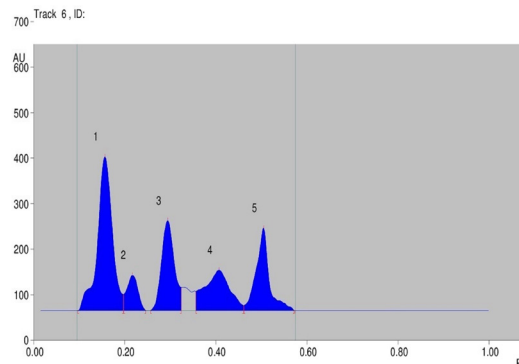


Figure 12: HPTLC Fingerprint Profile of Chloroform Extract of *Safoof-e-Binai* (SB) in Toulene: Ethyl acetate at 366 nm.

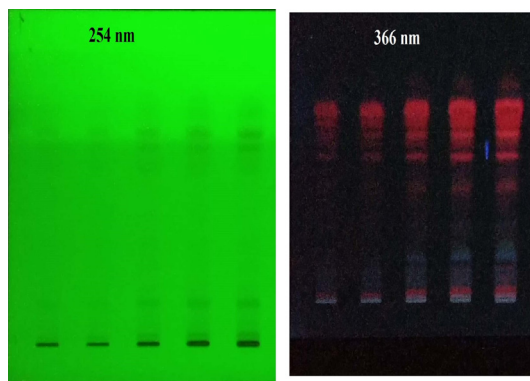


Figure 13: HPTLC Photos of *Safoof-e-Binai* (SB) Ethanol Extract in Toulene: Ethyl Acetate at 254 nm and 366 nm.

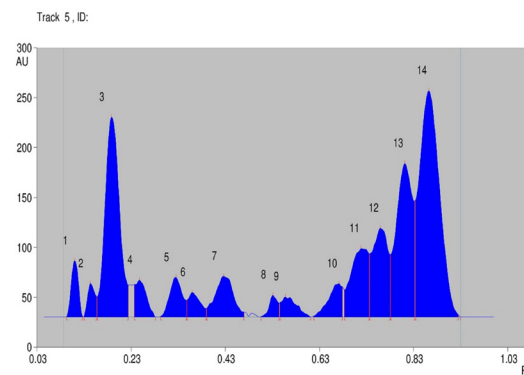


Figure 16: HPTLC Fingerprint Profile of Ethanol Extract of *Safoof-e-Binai* (SB) in Toulene: Ethyl acetate at 254 nm.

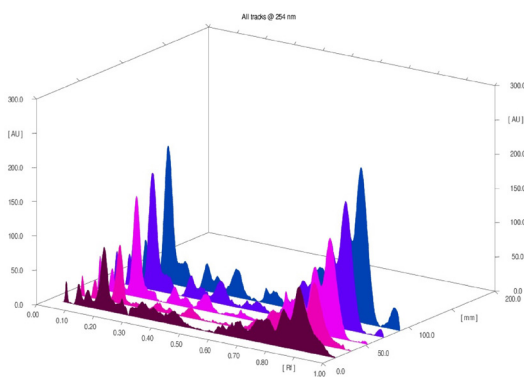


Figure 14: HPTLC 3-D Densitometric Scan of Ethanol Extract of *Safoof-e-Binai* (SB) in Toulene: Ethyl acetate at 254 nm.

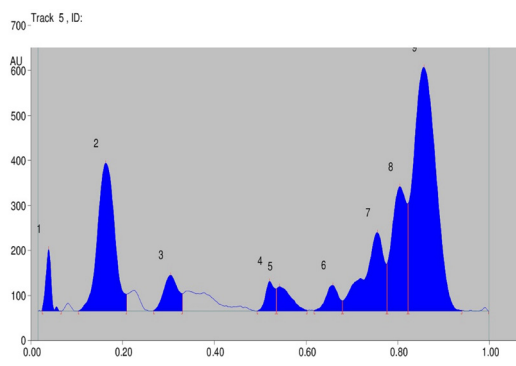


Figure 17: HPTLC Fingerprint Profile of Ethanol Extract of *Safoof-e-Binai* (SB) in Toulene: Ethyl Acetate at 366 nm.

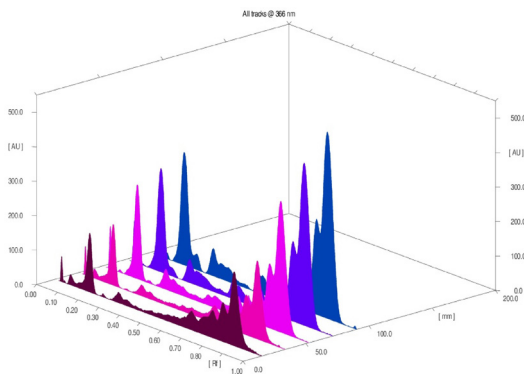


Figure 15: HPTLC 3-D Densitometric Scan of Ethanol extract of *Safoof-e-Binai* (SB) in Toulene: Acetate at 366 nm.

its indications and data derived for its reported activity and constituent of the formulation (SB) ingredients, present study will be very useful for quality control including identification and standardization of SB.

CONCLUSION

In present study *Safoof-e-Binai* (SB) was evaluated physico-chemically to set its physicochemical standards (loss of weight on drying, pH, total ash, water soluble, acid insoluble ash, extractive values, qualitative data for presence of organic constituents and HPTLC finger printing), which

may be used as standard monograph for identification, quality control and also for further evaluation or future research work on standardization of this formulation.

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CONFLICT OF INTEREST

There are no conflicts of interest.

ABBREVIATIONS

WHO: World Health Organisation; **USM:** Unani System of Medicine; **SB:** *Safoof-e-Binai*, **HPTLC:** High Performance Thin Layer Chromatography; **SEM:** Standard Error of Mean; **LOD:** Loss on Drying; **UPI:** Unani Pharmacopeia of India; **TLC:** Thin Layer Chromatography; **NFUM:** National Formulary of Unani Medicine.

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