Development of HPTLC Fingerprints of 'Itrifal Kishneezi' : A Unani Classical Formulation

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Introduction

trifal is a semi-solid preparation where one or more single drugs of plant origin are mixed, where triphala (Halela, Balela, Amla) used as main ingredients. This Poly-herbal Unani formulation is therapeutically considered as stomachic, laxative and carminative. It is used for the treatment of bleeding piles, conjuctivites, chronic catarrah, gastric headache, etc. The ingredients used in this formulation are: Amla (*Emblica officinalis*), Post- e-Halela Zard (*Terminalia chebula*), Post- e- Halela Kabli (*Terminalia chebula*), Halela Siyah (*Terminalia chebula*), Kishneez Khushk (*Coriendrum sativum*), Balela (*Terminalia bellerica*). Other formulations of similar catergories viz Itrifal Zamani (Siddiqui *et al.*, 1991), Itrifal- e- mulayin (Veeresh *et al.*, 2004) and Itrifal muqil (Arfin *et al.*, 2007) have been worked out. Botanical identification of the ingredients of *Itrifal Kishneezi* was carried out by Negi *et al.* (2009). Pharmacopoeial monograph on Itrifal Mulayyan ,Itrifal Muqawwi Dimagh ,Itrifal-e-MuqilMumsik,Itrifal-e-Mus-hil,Itrifal-e-Sana,Itrifal-e-Zabeeb,Itrifal Zamani,Itrifal-e-Muqil Mulayin were published by (Annonymous, 2009; 2010).

Material and method

Itrifal Kishneezi was prepared as per National Formulary of Unani Medicine (Anonymous, 2006). All the ingredients of the formulation were procured from the Khari Baoli market of Delhi and authenticated with the help of pharmacopoeial standards (Anonymous, 2007) and finally compared with the museum samples of PLIM, Ghaziabad. The compound formulation was prepared in the laboratory as per the procedure laid in NFUM (2006).

The physico-chemical study of the drug was carried out and for HPTLC profile CAMAG HPTLC system equipped with a sample applicator Linomat V, automatic multiple Developer-2 chamber, TLC scanner 3, Reprostar-3 and Win- cats an integrated Software 4.02 (Switzerland) was used.

Formulation/Composition: *Itrifal kishneezi* is a semi- solid preparation consists of the following herbal ingredients in the composition:

Table-1

S.No.	Unani Name	Botanical Name	Part used	Quantity
1	Amla	Emblica officinalis Gaertn.	Fruit	100g.
2	Post-e-Halela Zard Terminalia chebula Retz		Fruit pulp	100g.
3	Post-e-Halela Kabli	Terminalia chebula Retz	Fruit pulp	100g.



S.No.	Unani Name	Botanical Name	Part used	Quantity
4	Halela siyah	Terminalia chebula Retz	Fruit	100g.
5	Balela	<i>Terminalia bellerica</i> Roxb.	Fruit pulp	100g.
6	Kishneez khushk	Coriendrum sativum Linn.	Fruit	100g.

Procedure: The formulation was prepared as per methodology given in NFUM (Anonymous, 2006).

Observations and Results

1A. Macroscopical/Organolaptic features of Ingredients of Itrifal Kishneezi:-

- Amla (*Emblica officinalis* Gaertn.): Broken pieces of greyish black coloured fruit globuler with a wrinkled surface. The fruit breaks easily, exposing a section of dried pulp and nut which contains tri-gonous seeds of yellowish brown colour, odour- mild and characteristic, taste-acidic (Fig. 1A).
- 2. Post-e-Halela Zard (*Terminalia chebula* Retz.): Broken pieces of reddish brown fruit of various sizes, longitudinally wrinkled surface having five ribes. Odour-agrreable,Taste-astringent (Fig.1B).
- **3.** Post-e-Halela Kabli (*Terminalia chebula* Retz.): Broken pieces of yellowish brown fruit of various sizes, wrinkled surface, having five ribes. Odour- agrreable; taste- astringent (Fig.1C).
- 4. Halela siyah (*Terminalia chebula* Retz.): The small sized variety of young immature fruits which are upto 2.5 cm. long and 8mm. broad Ovoid with a longitudinally wrinkled surface and black colour, Odour- None, Taste-Astringent (Fig. 1D).
- **5.** Balela (*Terminalia bellerica* Roxb.): Broken pieces of yellowish brown fruit, surface velvetty, covered with close falvous tomentum having light amber colour (Fig.1E).
- 6. Kishneez khushk (*Coriendrum sativum* Linn.): Fruit ovoid or sub-globular, yellowish green in colour, external surface is covered with longitudinally running primary and secondary ridges. The former are wavy and inconspicuous while the latter are straight and prominent on whole, the fruit gives a ribbed appearance, Odour- spicy, taste-pungent and aromatic (Fig. 1F).





Amla (Emblica officinalis Gaertn.)



Post-e- Halela Zard(Terminalia chebula Retz.)



Post-e- Halela Kabli (Terminalia chebula Retz.)



Halela Siyah (Terminalia chebula Retz.)



Post- e- Balela (*Terminalia bellerica* Roxb.)



Kishneez khushk (Coriendrum sativum. L.)

Fig. 1: A, B, C, D, E, F - Photographs of the crude drugs



2. Physico-Chemical Analysis

Physico-chemical analysis of compound formulation: The data obtained in study is given in Table-2.

Parameter	Value
Alcohol soluble matter	27.90%
Water soluble matter	77.64%
Total ash	0.98%
Acid insoluble ash	0.065%

3. Thin Layer Chromatography: Five g powdered drug was extracted in 60 ml of absolute alcohol under reflux on water bath for 10 min. Filtered and concentrated the filtrate up to 4 ml. The extract obtained was applied on a precoated silica gel plate and developed in Ethyl acetate: Methanol: Water (100: 13.5: 10) system in developing chamber. The plate was dried and sprayed with Anisaldehyde- Sulphuric acid reagent and again the plate was dried and kept in an oven for heating at 105° c for 10 minutes, (Fig. 2A, B, C).



HPTLC Profile: Itrifal Kishizeezi

148





UV 366 nm



I-Itrifal Kisnizee, II-Amla, III-Post-e-Halela Zard, IV-Post-e-Halela Kabli, V- Halela Siyah, VI- Balela, VII- Kisneez Khushk Solvent System: Ethyl acetate: Methanol: Water (100: 13.5: 10) Spray Reagent: Anisaldehyde-Sulphuric Acid Reagent

Fig. 2: (A, B, C)

Table-3: TLC Fingerprint Data

Ingredient / Formulation	RF Value
Amla (ingredient)	0.13, 0.39, 0.81
Post-e- Halela Zard (ingredient)	0.13, 0.23, 0.39, 0.72
Post-e- Halela Kabli (ingredient)	0.13, 0.39, 0.72
Halela siyah (ingredient)	0.1, 0.29, 0.38, 0.72

149

Ingredient / Formulation	RF Value	
Balela (ingredient)	0.11, 0.72	
Kishneez khushk (ingredient)	0.13, 0.39, 0.72	
Itrifal kishneezi (formulation)	0.13, 0.23, 0.29, 0.39, 0.72, 0.81	

1B. Organoleptic features of formulation: Colour- raddish brown, semisolid, sweet but slightly bitter in taste, Smell aromatic.

Conclusion

Authentification of ingredients by Macroscopy (Fig. 1), along with physicochemical parameters (Table - 2) followed by HPTLC Profile (Fig-2, Table - 3) demonstrates the genuineness and purity of Itrifal kishneezi, that may help ensuring the quality of other indigenous medicine as well.

Acknowledgement

The authors are highly thankful to the Director General, CCRUM, New Delhi for providing necessary research facilities and Dr. G. P. Garg, Project Director D.S.R.I. (Unani), CCRUM, Ghaziabad, for continuous encouragement.

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