WORLD JOURNAL OF PHARMACEUTICAL RESEARCH
SJIF Impact Factor 8.084
Volume 12, Issue 17, 1242-1251. Review Article ISSN 2277–7105

PHYSICO-CHEMICAL STANDARDIZATION STUDIES OF AN IMPORTANT UNANI DRUG: ARQ-E-MUSSAFFI-E-KHOON

Asma Sattar Khan*, Shoeb Ahmed Ansari, Usha Devi, Suryansh Kashyap, Reesha Ahmed and Firoz Ahmad Ansari

Drug Standardization Research Institute, CCRUM, Kamla Nehru Nagar, Ghaziabad-201002, UP, India.

Article Received on
22 August 2023,

Revised on 12 Sept. 2023,
Accepted on 02 Oct. 2023

DOI: 10.20959/wjpr202317-29864

***Corresponding Author**
Dr. Asma Sattar Khan
Drug Standardization Research Institute, CCRUM, Kamla Nehru Nagar, Ghaziabad-201002, UP, India.

ABSTRACT

Unani remedies are a significant healthcare alternative for sustaining good health, therefore standardizing their herbal formulas is crucial in light of the growing interest in ancient systems of medicine. To take advantage of worldwide potential, the validation of Unani formulation critically needs a systemic scientific process. Herbal formulations are typically created using conventional techniques in accordance with the steps outlined in classical literature, which occasionally lacks scientific support. To assure their legitimacy and medicinal effects, it is crucial to standardize the production process and phytochemical characteristics. Arq-e-Mussaffi-e-Khoon, a poly-herbal Unani formulation that is frequently used as "blood purifiers," has been standardized using a number of quality control parameters. This medication is regarded as safe for long-term therapy and is a highly effective blood purifier (Mussaffi-e-Dam) and antiseptic (Dafa-e-Ta'affun). Arq-e-Mussaffi-e-Khoon was made using the ingredients and procedures outlined in the National Formulacy of Unani Medicine (NFUM). The formulation was evaluated using standard analytical procedures, including organoleptic evaluation (color, taste, aroma and consistency) and physico-chemical evaluation (pH as such, weight per ml, Refractive index, optical rotation, volatile oil, and alcohol flavor assessment. In order to determine the quality of the medicine, evaluations of WHO parameters such heavy metals, aflatoxins, pesticide residues, and microbiological contaminations were also made in the formulation. The information gathered in this study will be used to create pharmacopeial standards that will ensure Arq-e-Mussffi-e-Khoon's quality and batch to batch consistency.

KEYWORDS: Arq; heavy metal; Physico-chemical analysis; Pharmacopeial standard; WHO parameter.

INTRODUCTION

The use of medicinal plants as a rich source of therapeutic substances for the treatment of diseases and afflictions has been recognized for millennia and is highly regarded throughout.[1] The usage of traditional medicine, which is mostly based on plant material, is a necessity for more than 60% of the world's population.[2] The World Health Organization (WHO) emphasizes and promotes the study of traditional medicines due to its accessibility, affordability, and ease of availability. India has been designated as the hub for integrated medicine by WHO. According to the National Policy of Indian System of Medicine and Homeopathy from 2002, the Indian government has official structures in place to control the quality, safety, effectiveness, and documentation of herbal medicine.[3]

The bio-efficacy and repeatable therapeutic impact of herbal medicines are affected by a number of parameters, including temperature, geographic location, period and time of collection, age and section of the plant gathered, collecting method, and a number of other characteristics. This makes it difficult to standardize herbal medicines.[4]

Adulteration, contamination, and the partial or complete omission of or replacement of expensive constituents are risky production processes that ultimately undermine the formulations' quality. The importance of rigorous quality control and standardization of herbal medicines through appropriate scientific testing and evaluation procedures has been highlighted by this.[5]

The national formulary of unani medicine, part 1, classifies Arq-e-Mussaffi-e-Khoon, a liquid poly-herbal formulation, under the 'Saiyyalat' category.[6] Araqiyat is a sort of liquid preparation that is created employing medications with a plant source through the distillation process, which includes the phases of evaporation and condensation. Most of the time, arqiyat are transparent as crystal.

There are numerous conventional formulas employed as blood purifiers (Mussaffi-e-Dam) in the Unani system of medicine. A common medication used as an antibacterial and blood purifier is Arq-e-Mussaffi-e-Khoon. Even if ingested for a longer period of time, it is safe and

does not build habits. Additionally helpful in pustules, boils, scabies, itchiness, pityriasis alba, and pityriasis migra.

There are 27 components that make up Arq-e-Mussaffi-e-Khoon (table 1), each of which has exceptional therapeutic qualities.[7]

The ingredient Barg-e-Neem is known for its Mudammil (Cicatrizant) and Mussaffi-e-Dam (blood purifier) action and used in Aatishak (syphilis), Juzam (leprosy) and Qurooh (ulcer); Post-e-Neem is Mohallil (anti-inflammatory) and Munaqqi-e-Qurooh (wound cleanser) in action and used in Istrakka (atony), Waja-ul-Mafasil (arthritis), Falij (paralysis) and Khujli (itching); Post-e-Bakayin due to its Muhallil-e-riya (carminative) and Mussaffi-e-Khoon (blood purified) action used in Qula (aphthous ulcer), Aatishak (syphilis) and Juzam (leprosy); Barq-e-Bakayin in Qatil-e-Deedan-e-Am'a in action and used in Bawaseer Amya (blind piles) and Bawaseer Damiya (bleeding piles). Post-e-Kachnal is well known for its Mujaffif (desiccant), Qabiz (astringent) and Mussaffi-e-Dam (blood purifier) action and used in Kanthamela (scrofula), Nafs-ud-Dam (haemoptysis) and Qula (aphthous ulcer). Barq-e-Hina is prescribed for Boil (phonsiya) and Jamda (facial erysipelas) due to its action Mohallil-e-Waram (anti-inflammatory) and Mujaffif (desiccant). Mundi (flower) is Mohallil (anti-inflammatory) and Muqammi-e-azae raisa (tonic for vital organ) in action and used in Hikka (pruritis), Yarqan (jaundice), Shizaak (gonorrhoea) and Warm (inflammation). Shahtra is well known for its action in Mussaffi-e-Dam (blood purifier) and used in Zarb (scabies), Daad (tinea), Kharish (pruritis) and Phoda Abscess. Sarphuka is Mudirr-e-Boul (diuretic) in action and used in Zeeq-un-Nafs (asthma), Jarb (scabies) and Khansi (cough). Gul-e-Nilofer is Muqammi-e-Dimag (brain tonic) in action is used in Dard-e-Sar (headache), Bars (vitiligo) and Surkh Bada (facial erysipelas). Burada-e-Sandal-Surkh is Mudammil (cicatrizant) in action is used in Hurqat-ul-Boul (burning micturition), Baul-ul-dam (haematuria) and Fasad-ud-dam (blood impurities). Burada-e-Sandal Safard is a well-known blood purifier (Mussaffi-e-Dam) and used in Is'hal Safrawi (bilious diarrhoea) and Khafqaan (palpitation). Burda-e-chob-e-Sheesham is Mujaffif (desiccant) in action and used in Bars (vitiligo), Juzam (leprosy) and Qula (aphthous ulcer).[8]

The current research intends to establish pharmacopeial standards for Arq-e-mussaffi-e-khoon by developing quality measures and evaluating the data. To determine the quality of the formulations, the traditional metrics, such as organoleptic parameters, such as heavy metal estimation, aflatoxins, microbial loads, and pesticide residue.

MATERIAL AND METHODS

Preparation of formulation

All of the substances were obtained from local raw drug dealers and were all botanically recognized by use of pharmacognostical techniques.[9,10,11] These constituents were further verified by comparison with the UPI Part 1 and API Part 1 Vol. 1 monographs.

All the ingredients were cleaned and dried under shade to remove the moisture if any. The ingredient nos. 1-26 were crushed separately by using iron mortar to obtained their coarse powders. The powders so obtained were mixed thoroughly and added to ingredient no.27. The whole content was kept for soaking for 24 hrs. and later transferred into a distillation assembly. The content was boiled at 100°C for 7-8 hrs in order to collect the distillate (Arq) in a round bottom flask. The Arq was allowed to cool at room temperature and stored in air tight glass container.[12,13]

Physicochemical analysis

The Arq-e-Mussaffi-e-Khoon's physicochemical properties such as pH, weight per mL, refractive index optical rotation, volatile oil, and test for alcohol, were examined using established techniques.[14,15]

Quality Control Analysis

Many people utilize unani medicines as effective and secure treatments all around the world. Regardless of the form of a unani preparation, its quality of check is crucial to maintaining the public's faith in unani medications. In order to assess the quality of Arq-e-Mussaffi-e-Khoon, various quality control measures, including microbiological load, heavy metals, aflatoxins, and pesticide residues, were used. The microbial load was calculated using the recommended methodology The examination of heavy metals and aflatoxins was done using, respectively, HPLC (Thermo Fisher) and Atomic Spectro photometer (LABINDIA). GC-MS system (Agilent) with mass selective detector was used for the analysis of pesticide residue in accordance with standard procedure.[16,17,18]

RESULTS AND DISCUSSION

Arq-e-Mussaffi-Khoon is a colourless liquid with characteristics smell and taste.

Physicochemical analysis

Table 2 displays the physicochemical information about the medication Arq-e-Mussaffi-e-Khoon. The results of the quantitative analysis showed that the test for alcohol was negative. It indicates that the Arq is fresh and does not degrade or decay. The pH of Arq-e-Mussaffi-e-Khoon is in the 7.45–7.55 range, indicating neutrality and confirming the lack of any acidity. The presence of volatile oil in the range of 1.40-1.65 indicates that high-quality herbal material was used. Weight per milliliter (g) is between 0.9624 and 0.9786, indicating the Arq's thin consistency. The refractive index lies between 1.3327 and 1.3366.

Quality control parameters

Microbial Load

An extremely crucial factor in traditional remedies is the estimation of microbial growth. It indicates whether or not the medicine contains disease-causing and spoilage microorganisms, and if so, whether or not those organisms are present within WHO allowed limits. The evaluation is carried out to determine the total number of bacteria, total number of fungi, number of bacteria from the family *Enterobacteriaceae*, and number of pathogens such as *E. Coli*, *Staphylococcus aureus*, *Salmonella* spp., and *Pseudomonas aeruginosa*. Table 3's microbiological load data, which are within acceptable levels, revealed that the medicine is safe for internal use or consumption.

Aflatoxin

Table 4 lists the outcomes of the drug's analysis for aflatoxins. A number of molds, including *Aspergillus flavus*, *Aspergillus parasiticus* and *Aspergillus nomius* produce aflatoxins, which are hazardous by-products. The findings indicates that there were no aflatoxins (B1, B2, G1, or G2) present in Arq-e-Mussaffi-e-Khoon.

Pesticide residues

Table 5 provides the results of pesticide residues. Due to a number of circumstances, harvesting herbal material without the use of pesticides is quite challenging. However, in accordance with WHO rules, the main issue is whether or not the medicine has pesticide residue in an acceptable level. The medication was analyzed using GC-MS to assess the pesticide residue. The outcomes showed that the medication is safe to use and devoid of pesticide residue.

Heavy metal analysis

Table 6 presents the findings of the estimate of heavy metals. Injurious to human health, heavy metals have been linked to a number of fatal disorders. A heavy metal is dangerous or poisonous even at low concentrations because of its relatively high density and atomic weight. Arq-e-Mussaffi-e-Khoon's heavy metal content was discovered to be below the detection threshold, indicating that the medicine is free of heavy metal contamination and suitable for usage.

Table 1: Formulation composition of Arq-e-Mussaffi-e-Khoon.

S. No.	Unani Name	Botanical/ English name	Part used
1	Barg-e-Neem	*Azadirachta indica* A. Juss	Leaf
2	Post-e-Neem	*Azadirachta indica* A. Juss	Bark
3	Post-e-Bakayin	*Melia azedarach* L.	Bark
4	Barg-e-Bakayin	*Melia azedarach* L.	Leaf
5	Post-e-Kachnal	*Bauhinia racemose* Lam.	Bark
6	Post-e-Mulsari	*Mimusops elengi* L.	Bark
7	Dudhi Khurd	*Euphorbia hirta* L.	Whole
8	Barg-e-Bhangra Siyah	*Eclipta alba* Hassk.	Leaf
9	Shakh-e-Barg-e-Jawansa	*Ahagi pseudalhagi* (Bieb.) Desv.ex B.Keller and Sharp	Branches
10	Post-e-Gular	*Ficus racemose* Roxb.	Bark
11	Barg-e-Hina	*Lawsonia inermis* L.	Leaf
12	Mundi	*Sphaeranthus indicus* L.	Flower
13	Shahtra	*Fumaria parviflora* L.	Whole
14	Sarphuka	*Tephrosia purpurea* (L.) Pers.	Whole
15	Dhamaya/ Shukai	*Fagonia cretica* L.	Whole
16	Chob-e-Bijasar	*Pterocarpus marsupium* Roxb.	Wood
17	Gul-e-Nilofar	*Nymphae alba* L.	Flower
18	Burada-e-Sandal Surkh	*Pterocarpus santalinus* L. f.	Sawdust
19	Burada-e-Sandal Safaid	*Santalum album* L.	Sawdust
20	Gul Surkh	*Rosa damascena* Herm.	Flower
21	Kishneez Khushk	*Coriandrum sativum* L.	Fruit
22	Tukhm-e-Kasni	*Cichoriumintybus* L.	Seed
23	Beikh-e-Kasni	*Cichoriumintybus* L.	Root
24	Majeeth	*Rubia cordifolia* L.	Root
25	Barg-e-Bed Sada	*Salix alba* L.	Leaf
26	Burada-e-Chob-e-Sheesham	*Dalbergia sissoo* DC.	Sawdust
27	Aab	Water	As such

Table 2: Physico-chemical parameters of Arq-e-Mussaffi-e-Khoon.

S. No.	Parameter	Results (range)
1	pH as such	7.45-7.55
2	Weight per mL (g)	0.9624-0.9786
3	Refractive index (nD)	1.3327-1.3366
4	Volatile oil (% $^{v}/_{v}$)	1.40-1.65
5	Test of Alcohol	Negative

Table 3: Microbial load of Arq-e-Mussaffi-e-Khoon.

S. No.	Microbe analysed	Results
1	Total aerobic bacterial count (TABC)	3.6×10^3 CFU/ml
2	Total yeast and molds count (TYMC)	1.0×10^2 CFU/ml
	Enterobacteriaceae members	
3	*Escherichia coli*	ND
4	*Salmonella sp.*	ND
5	*Shigella sp.*	ND
6	*Klebsiella sp.*	ND
	Specific objectionable pathogens	
7	*Pseudomonas aeruginosa*	ND
8	*Staphylococcus aureus*	ND
9	*Candida albicans*	ND
	Aflatoxin producing fungi	
10	*Aspergillus flavus*	ND
11	*Aspergillus parasiticus*	ND

ND= Not detected

Table 4: Aflatoxins level of Arq-e-Mussaffi-e-Khoon.

S. No.	Parameter analysed	Results
1	B_1	ND
2	B_2	ND
3	G_1	ND
4	G_2	ND

ND= Not detected

Table 5: Heavy metals analysis of Arq-e-Mussaffi-e-Khoon.

S. No.	Metal analysed	Results
1	Arsenic	ND
2	Cadmium	ND
3	Lead	ND
4	Mercury	ND

ND= Not detected

Table 6: Pesticide Residue of Arq-e-Mussaffi-e-Khoon.

S. No.	Pesticide	Result (mg/kg)	Permissible limit (mg/kg)
1	Alachlor	BLQ	0.02
2	Aldrin (Aldrin and dieldrin combined expressed as dieldrin)	BLQ	0.05
3	Azinophos-methyl	BLQ	1.0
4	Bromopropylate	BLQ	3.0
5	Chlordane (cis, tans and oxychlordane)	BLQ	0.05
6	Chlorfenvinphos	BLQ	0.5
7	Chlorpyrifos	BLQ	0.2
8	Chlorpyrifos-methyl	BLQ	0.1
9	Cypermethrin (and isomers)	BLQ	1.0
10	DDT (all isomers, sum of p, p'-TDE (DDD) expressed as DDT)	BLQ	1.0
11	Deltamethrin	BLQ	0.5
12	Diazion	BLQ	0.5
13	Dichlorvos	BLQ	1.0
14	Dithiocarbamates (as CS2)	BLQ	2.0
15	Endosulphan (sum of isomers and Endosulphan sulphate)	BLQ	3.0
16	Endrin	BLQ	0.05
17	Ethion	BLQ	2.0
18	Fenitrothion	BLQ	0.5
19	Fenvalerate	BLQ	1.5
20	Fonofos	BLQ	0.05
21	Heptachlor (sum of Heptachlor and Heptachlor epoxide)	BLQ	0.05
22	Hexachlorobenzene	BLQ	0.1
23	Hexachlorocyclohexane isomer (other than ɤ)	BLQ	0.3
24	Lindane (ɤ- Hexachlorocyclohexane)	BLQ	0.6
25	Malathion	BLQ	1.0
26	Methidathion	BLQ	0.2
27	Parathion	BLQ	0.5
28	Parathion methyl	BLQ	0.2
29	Permethrin	BLQ	1.0
30	Phosalone	BLQ	0.1
31	Piperonyl butoxide	BLQ	3.0
32	Pirimphos methyl	BLQ	4.0
33	Pyrethrins (sum of isomers)	BLQ	3.0
34	Quintozene (sum of Quintozene, pentachloroanilene and methyl pentachlorophenyl sulphide)	BLQ	1.0

BLQ= Below the Limit of Quantitation.

CONCLUSION

It is clear that standardization is crucial for preserving the quality of any herbal composition. Proper identification of both single ingredients and complex formulations should be done. Arq-e-Mussaffi-e-Khoon was assessed using pharmacopeial standards, which unquestionably offer a guarantee of the drug's purity. The microbiological load, aflatoxins, pesticide residue,

and heavy metal values of the quality control parameters were determined to be under the WHO permitted limit, indicating that the medicine is free of harmful ingredients and can be used safely. Therefore, the current investigation validates the legitimacy, excellence, and effectiveness of the Unani remedy Arq-e-Mussaffi-e-Khoon.

ACKNOWLEDGEMENT

The authors are extremely grateful to the Director-General, CCRUM, New Delhi for his cordial encouragement and providing necessary research facilities.

REFERENCES

1. Sharma A., Shanker C., Tyagi L. K., Singh M., Rao CV (2008) Herbal medicine for market potential in India: An overview. Acad J Plant Sci, 1: 26-36.
2. World Health Organization (2019) WHO global report on traditional and complementary medicine. World Health Organization, Geneva, Switzerland.
3. Singh B., Kumar M., Singh A. (2013) Evaluation of implementation status of national policy on Indian system of medicine ad homeopathy 2002: stakeholders' perspective. Ancient Science of Life, 33: 103-108.
4. Kumari R., Kotecha M. (2016) A review on the standardization of herbal medicines. International Journal of Pharma Sciences and Research, 1: 97-106.
5. World Health Organization (1998) Quality control methods for medicinal plant materials. World Health Organization, Geneva, Switzerland.
6. Anonymous. National formulary of Unani medicine, Part 1, CCRUM, Ministry of Health and family welfare (Department of Ayush), Government of India, New Delhi, 2006; 218.
7. Kabeeruddin h. (YNM): Makhzanul Advia Al maroof Khawaas ul Advia, Ajaz Publication house, Darya Ganj, New Delhi. P: 131, 337, 379, 391, 396, 552 and 580.
8. Ghani h. (YNM): Khazainul Advia, Idare Kitabus Shifa, Darya Ganj, New Delhi. P: 379, 797, 927, 932, 933, 1263, 1286, 1330, 1329.
9. Johnson D. A., Plant micro-techniques, Mc-Graw Hill book Company Inc. New York and London, 1940; 13: 65-105.
10. Wallis TE. Text book of Pharmacognosy, 5th ed. CBS Publishers and Distributors Pvt. Ltd., New Delhi, 2005; 578: 493-494.
11. Trease GE, Evans W. C. Pharmacognosy Bailliere Tindall, London, 1989; 13: 5-9.

12. Anonymous. Unani Pharmacopoeia of India, Ministry of Health and family welfare, Govt. of India, Part 1; Vol. I, P- 68, 56; Vol II, P- 81, 87, 51, 75; Vol III, P- 10, 8, 207, 31; Vol IV, P- 100, 21, 104, 89; Vol V, P- 25, 80, 92; Vol VI, P- 35, 76, 95.
13. Anonymous, Ayurvedic Pharmacopoeia of India, Ministry of health and family welfare, Govt. of India, Part 1, Vol III, P: 207.
14. Anonymous. Physicochemical standard of Unani formulation, Part II, CCRUM, Govt. of India, New Delhi, 1987; 2: 268-281.
15. Anonymous. Quality control methods for medicinal plant materials. World Health Organisation, Geneva, 1998; 28-33.
16. Anonymous. WHO guidelines for assessing the quality of herbal medicines with references to containments and residues, World Health Organisation, Geneva, 2007; 27-28, 55-68.
17. Anonymous. Official methods of analysis, Horwitz W., Latiner G W (ed.) 18th ed. AVAC International; Merry land, 2005; 3: 10-11, 10:18-23 and 26:17.
18. Anonymous. Official analytical methods of the American spice trade association (ASTA), 4th ed., New Jersey, 1997; 149-152.