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Pharmaceutical design & development of a Unani Emulgel dosage form: A novel approach

Shamim and Mohammad Idris

Abstract
The combined dosage form of gel and emulsion are referred as emulgel. Both oil-in-water and water-in-oil emulsions are extensively used as vehicles to deliver various hydrophilic as well as hydrophobic drugs to the skin in emulgel formulation. They also have a high ability to dissolve drug and penetrate the skin. The emulgel has more advantages, such as more absorption, better skin penetration, greaseless, thixotropic, easily spreadable, easily removable, emollient, non-staining, water-soluble, longer shelf life, bio-friendly, better stability, high loading efficiency, more economic with low-cost production. A new formulation was conceptualized in the form of Unani emulgel based on the commonly used anti-inflammatory and analgesic single drugs (*mufradat*), such as Aak(*Calotropis procera*), Bakayan (*Melia azedarach*), Baid injeer (*Ricinus communis*), Dhatura (*Datura stramonium*), Sanbhaloo (*Vitex negundo*), Sahajna (*Moringa oleifera*), Thuhar(*Euphorbia neriifolia*), Babuna (*Matricaria chamomilla*) and Kunjad (*Sesamum indicum*). The novel formulation based on the concept of rapid penetration through the skin i.e., emulgel has been prepared in the laboratory with standard operating procedure (SOP). The formulation is based on the above mentioned Unani single drugs along with excipients, namely Gum acacia, Olic acid, menthol oil, Carbopol- 940 etc. The study indicates that the test Unani anti-inflammatory and analgesic emulgel (UAAE) was found to be physicochemically active, and pharmaceutically possessed all characteristics of a standard emulgel. Hence, this study generates the pharmaceutical evidence for scientific validation of this new dosage form, i.e., emulgel, and moreover showed path for modernization of Unani pharmaceutics. Further, a clinical study is warranted to assess the therapeutic efficacy and safety of UAAE.

Keywords: Unani dosage form, unani Emulgel, unani pharmaceutics, Uaae, anti-inflammatory

Introduction
In Unani system of medicine, analgesic and anti-inflammatory drugs are used in the form of single (*mufrad*) drugs and compound (*murakkab*) dosage forms. There are various topical drugs available for the effective and safe management and treatment of inflammatory indications. The topically used Unani dosage forms are mainly based on *Roghan* (medicated oil). Thus, they are poorly absorbed and delayed in action. In a field / observational study, it was observed that the Unani dosage forms, especially employed topically are desired to be modified in terms of efficacy, application, safety and packaging. The major problem of these dosage forms confronts with their slow duration of action and not user-friendly approach. After a thorough survey of Unani classical literature, it was decided to envisage a study based on designing and development of a new pharmaceutical dosage form as emulgel.

Topical drug administration is a localized drug delivery system anywhere in the body mainly through skin, partly from ophthalmic, rectal, and vaginal as topical routes. The dosage forms used topically range in physicochemical nature from solid through semisolid to liquid form. Drugs are administered topically for their action at the site of application or for systemic effects. The enhancement of drug absorption through skin is based on two major factors, namely if the drug substance is in liquid form, it has a favorable lipid / water partition coefficient; secondly, if it is a non electrolyte, i.e., non polar. For the most part, pharmaceutical preparations applied to the skin are intended to serve some local action, and as such are formulated to provide prolonged local contact with minimal systemic drug absorption, It is interesting to note that in spite of the popular Unani drug dosage forms (UDDFs), namely *safoof* (powder), *hub* (pill) *qurs* (tablet), *majoon* (electuary) etc. devised and developed exclusively for the oral route.

A separate class of UDDFs had also been designed and developed for joint disorders/diseases, such as *marham* (ointment), *Roghan* (medicated oil), *tila* (liniment) etc. administered topically. These UDDFs have been more effective and safe in clinical practice since centuries.

However, there is no denying the fact that these dosage forms have some disadvantages in terms of delayed onset of action and cumbersome to use. Thus, it needs to be understood in the present perspective. The need of the hour is to revisit the Unani dosage forms in terms of prevailing situation, i.e., need of fast relief and user-friendly approach. It necessitates meeting the challenge by developing Unani fast reliving drug delivery system (UFRDDS), especially employed as analgesic and anti-inflammatory medicament. The joint disorder/disease, muscular inflammatory conditions are the most suitable case for UFRDDS.

Material and Methods

The present study is based on the pharmaceutical design and development of a Unani emulgel dosage form along with its standard operating procedure (SOP). An innovative approach has been made to design and develop a new anti-inflammatory and analgesic emulgel.

Materials

Test Formulation

Table 1: List of ingredients included in the test formulation

S.No.	Name	Botanical name	Part used
1.	Aak	*Calotropis procera*	Barg
2.	Bakayan	*Melia azedarach*	Barg
3.	Baidinjeer	*Ricinus communis*	Barg
4.	Dhatura	*Datura stramonium*	Barg
5.	Sanbhaloo	*Vitex negundo*	Barg
6.	Sahajna	*Moringa oleifera*	Barg
7.	Thuhar	*Euphorbia neriifolia*	Barg
8.	Babuna	*Matricaria chamomilla*	Flower
9.	Kunjad	*Sesamum indicum*	Seeds'oil

Table 2: List of chemicals used in the test formulation

S.No.	Material	Grade	Manufacturer
1.	Tween 20	Pharma grade	Pioneer in –Organics, Delhi
2.	Tween 80	Pharma grade	Pioneer in –Organics, Delhi
3.	Gum acacia	Pharma grade	Pioneer in –Organics, Delhi
4.	Carbopol 940	Pharma grade	Pioneer in –Organics, Delhi
5.	Hpmc	Pharma grade	Pioneer in –Organics, Delhi
6.	Oleic acid	Pharma grade	Pioneer in –Organics, Delhi
7.	Propylparaben sodium	Pharma grade	Pioneer in –Organics, Delhi
8.	Methylparaben sodium	Pharma grade	Standard chemicals, New Delhi
9.	Liquid paraffin	Pharma grade	Sisla laboratories, New Delhi
10.	Buffer solution	Pharma grade	General drug house, New Delhi

Table 3: List of instruments/equipments

S. No.	Instrument	Manufacturer
1.	Artificial membrane	Sigma Aldrich chemicals Pvt. Ltd. Missouri, USA
2.	Electronic balance	Citizen India
3.	Franz diffusion cell	Alpha Scientific Equipments, Delhi
4.	Hot air oven	Uts Pvt. Ltd. India
5.	Hot plate with Magnetic stirrer	Altis Instruments, India
6.	Mixer and Grinder	Lords Gold, India
7.	Ph meter	Decibel Instruments, India
8.	Pharmaceutical sieves	Parasava Natha Wire Netting Store, Delhi
9.	Soxhlet apparatus	Alpha Scientific Equipments, Delhi
10.	UV Spectrometer	Shimadzu –UV-1601, Kyoto, Japan
11.	Water Bath	Altis Instruments, India
12.	Electrical Microscope	Magnus, India

Methods

Pre-formulation studies

Collection of drugs: Leaves of *Aak, Bakayan, Baid injeer, Dhatura, Sanbhaloo, Sahajna, Thuhar* and flowers of *Babuna* were collected from the herbal garden at A&U Tibbia College & Hospital, Karol Bagh, New Delhi. *Roghan -e-Kunjad* (Sesame oil) was procured from Ballimaran *bail kolhu* (traditional oil expeller), Delhi. All foreign matters were inspected with naked eyes and cleaned properly.

Identification of drug

Sample of ingredients were separately packed, and sent for identification. The identity of all drugs was established by the experts of Raw Material Herbarium and Museum, CSIR-National Institute of Science Communication and Information Resources (NISCAIR), New Delhi. The specimen vouchers of all ingredients were deposited in the Drug Museum, Department of Ilm-ul-Advia, A&U Tibbia College & Hospital, Karol Bagh, New Delhi.

Preformulation Method

Process of extraction

Method of preparation of aqueous extract

Leaves of *Aak, Bakayan, Baidinjeer, Dhatura, Sanbhalu, Sahajna, Thuhar* and flowers of *Babuna* were taken in equal quantity, i.e. 10 grams each, and crushed by using iron mortar and pestle. 800 ml distilled water was added in crushed ingredients, and run over 6 hours at 70 ^{0}C by Soxhlet apparatus. The quantity of obtained extract was 550 ml.

Formulation of different versions of emulsion

Optimization and Preparation of emulsion

In the preparation of emulsion (oil in water), different surfactants, namely tween 20, tween 80, gum acacia were used. The aqueous extract was used as a water phase, and oil phase was made by *Roghan -e-Kunjad.* The emulsion was made in the following percentage ratio of oil phase: surfactant: aqueous phase. The surfactant was used in 5 to 20%.

Table 4: Ratio of different surfactant

S.No.	Version	Ratio (oil: surfactant: aqueous extract)	S.No.	Version	Ratio (oil: surfactant: aqueous extract)
1.	Version-1.0	45:05:50	21.	Version-2.7	50:15:35
2.	Version-1.1	50:05:45	22.	Version-2.8	40:20:40
3.	Version-1.2	40:10:50	23.	Version-2.9	35:20:45
4.	Version-1.3	50:10:40	24.	Version-2.10	45:20:35
5.	Version-1.4	40:15:45	25.	Version-2.11	30:20:50
6.	Version-1.5	45:15:40	26.	Version-2.12	50:20:30
7.	Version-1.6	35:15:50	27.	Version-3.0	45:05:50
8.	Version-1.7	50:15:35	28.	Version-3.1	50:05:45
9.	Version-1.8	40:20:40	29.	Version-3.2	40:10:50
10.	Version-1.9	35:20:45	30.	Version-3.3	50:10:40
11.	Version-1.10	45:20:35	31.	Version-3.4	40:15:45
12.	Version-1.11	30:20:50	32.	Version-3.5	45:15:40
13.	Version-1.12	50:20:30	33.	Version-3.6	35:15:50
14.	Version-2.0	45:05:50	34.	Version-3.7	50:15:35
15.	Version-2.1	50:05:45	35.	Version-3.8	40:20:40
16.	Version-2.2	40:10:50	36.	Version-3.9	35:20:45
17.	Version-2.3	50:10:40	37.	Version-3.10	45:20:35
18.	Version-2.4	40:15:45	38.	Version-3.11	30:20:50
19.	Version-2.5	45:15:40	39.	Version-3.12	50:20:30
20	Version-2.6	35:15:50			

Preparation of emulsion

In this study, 50 ml extract was taken in a beaker and placed on magnetic stirrer, 35 ml *Roghan-e-Kunjad* was taken in another beaker, in this beaker 15 grams gum acacia was added and mixed well. Then the beaker containing medicinal extract was placed on magnetic stirrer at the room temperature and *Roghan-e-Kunjad* was added along with surfactant and mixed for 6 hours.

Preparation of 2.5%gel

2.5 grams Carbopol-940 was dispersed in 100 ml of distilled water for 24 hours for hydration, then it was stirred thoroughly using magnetic stirrer and consistency was checked.

Preparation of Emulgel using Carbopol940 as a gelling agent in 2.5%: Emulsion was prepared and gel in 2.5% Carbopol 940 was added. It was mixed, stirred thoroughly by using magnetic stirrer, and checked its consistency.

In the final version of emulgel, methyl paraben 0.1% was added and placed at room temperature. Its stability was checked for a period of 6 months.

Table 5: Composition of Unani Emulgel

S. No.	Composition	Quantity
1.	Medicinal extract	50 ml
2.	*Roghan -e-Kunjad*	35 ml
3.	Gum acacia	15 grams
4.	Carbopol 940	2.5grams
5.	Oleic acid	5.0 ml
6.	Menthol oil	2.5 ml
7.	Methyl paraben	0.1 gram

***In vitro* Release / Permeation Study of Emulgel**

In vitro release study was carried out by using Franz diffusion cell method. Franz diffusion cell was used for the drug release study. The gelified emulsion was applied onto the surface of artificial membrane. The membrane was clamped between the donor and the receptor chamber of diffusion cell. The receptor chamber was filled with freshly prepared phosphate buffer solution (PBS) (pH 7.4) solution to solubilise the drug. The receptor chamber was stirred by magnetic stirrer. The samples (3.0 ml) were collected at time interval of 5 minutes for first 30 minutes, for next 90 minutes at interval of 15 minutes, further next two hours at interval of 30 minutes, next two hours at interval of 60 minutes, next at interval of 6 hours, and finally after 24 hours They were filtered for analysis and replaced with an equal volume of the buffer solution to maintain a constant volume. The absorbance of the collected samples was measured by the UV spectrophotometer at λ max of 635 nm, using the same buffer solution as a control medium. The *in vitro* skin permeation study was carried out in triplicate for each formulation. Thereafter, the mean values and standard deviations for the amount of drug permeated were calculated and used for further calculations, and also these samples were analyzed for qualitative test (Haneefa, 2013, Masmoudi *et al*, 2006) [27, 44].

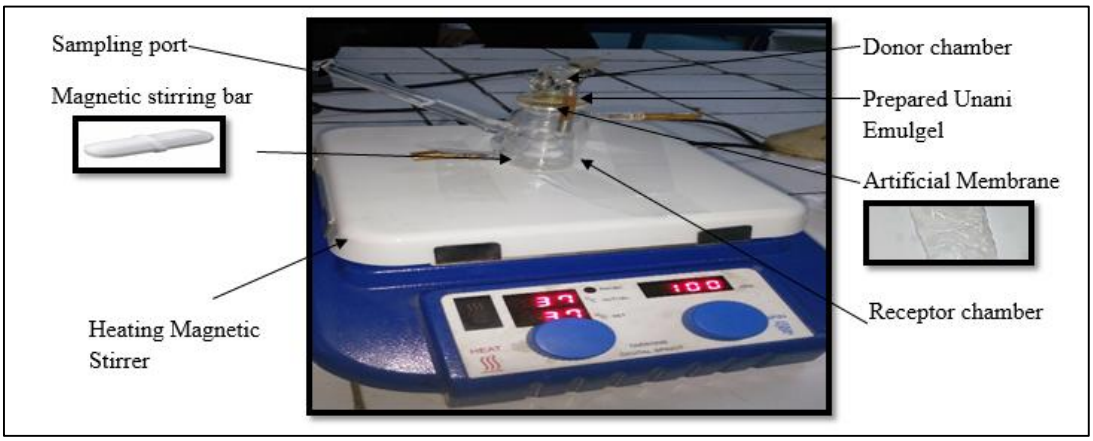

Fig 1: *In vitro* Release / Permeation Study of Emulgel by Franz Diffusion Cell Apparatus

Qualitative analysis of aqueous extract of formulation
The qualitative analysis of the different samples of aqueous extract of formulation was done by using various chemical tests.

(a) Tests for Alkaloids
Dragendorff's Test: 1.0 ml of the extract was taken in a test tube, added 1 ml Dragendorff's reagent were added and observed the change (s).

(b) Tests for Carbohydrates
Benedict's Test: 1.0 ml of extract was taken in a test tube and 5 ml of Benedict's reagent was added, boiled for 2 minutes, cooled, and observed the change (s).

(c) Tests for Proteins
Lead Acetate Test: 5.0 ml of extract was taken in a test tube, added 1.0 ml of lead acetate solution and observed the change (s).

Qualitative analysis of the formulation samples
The qualitative analysis of the different sample was done by using various chemical test.

(a) Tests for Alkaloids
Dragendorff's Test: 1 ml of the sample was taken in a test tube from the Franz diffusion cell, added 1.0 ml, Dragendorff's reagent was added and observed the change (s).

(b) Tests for Carbohydrates
Benedict's Test: 1.0 ml of sample was taken in a test tube and 5.0 ml of Benedict's reagent was added, boiled for 2 minutes, cooled and observed the change (s).

(c) Tests for Proteins
Lead Acetate Test: 5.0 ml of sample was taken in a test tube, added 1.0 ml of lead acetate solution, and observed the change (s).

Determination of water soluble matter
Ten (10) grams of formulation was macerated with 100 ml water in a conical flask for 24 hours, shaking frequently during 6 hours and was allowed to stand for 18 hours. Filtered rapidly, taking precautions against loss of the solvent, evaporated 25 ml of the filtrate to dryness in a tarred flat bottomed shallow dish on a water bath, and dried at 105 ^{0}C, to constant weight and weighed. The percentage of water soluble extractive with reference to the air dried drug was calculated. This procedure was repeated for three times the mean value and standard deviation was calculated (Anonymous, 2010; Anonymous, 2009) [15, 14].

Determination of alcohol soluble matter
Ten (10) grams of formulation was macerated with 100 ml alcohol in a conical flask for 24 hours shaking frequently during 6 hours and was allowed to stand for 18 hours. Filtered rapidly, taking precautions against loss of the solvent, evaporated 25 ml of the filtrate to dryness in a tarred flat bottomed shallow dish on a water bath, and dried at 105 ^{0}c, to constant weight and weighed. The percentage of alcohol soluble extractive with reference to the air dried drug was calculated. This procedure was repeated for three times. the mean value and standard deviation was calculated (Anonymous, 2010; Anonymous, 2009) [15, 14].

Loss in weight on drying at 105 ^{0}C temperature
Five (5) grams drug was spread uniformly in shallow Petri dish and was placed in oven temperature was maintained at 105 ^{0}C and then cooled in desiccators, weigh and calculated the percentage loss with respect to drug (Anonymous, 2010) [15].

Moisture content
Ten (10) grams emulgel was weighed in a patri dish, and kept in desiccators for about 24 hours after that it was taken out and weighed again the percentage of moisture content was calculated on the basis of following formula

$$\text{Percentage af moisture content} = \frac{\text{Initial weight} - \text{Final weight}}{\text{Final weight}} \times 100$$

Determination of pH
pH of 1% solution: 1.0 gram drug was accurately weighed and dissolved in accurately measured 100 ml of water, then filtered and pH was checked with standardized glass electrode.

pH of 10% solution: 10 grams drug was accurately weighed and dissolved in accurately measured 100 ml of water, then filtered and pH was checked with standardized glass electrode (Anonymous, 2010; Haneefa, 2013) [15, 27].

Stability Study
Stability study was carried out at the room temperature a period of 6 months. The samples were analyzed for physical appearance, pH, viscosity and microbial contamination.

Spreadability
Two sets of glass slides of standard dimensions were taken. The test emulgel formulation was placed over one of the slides. The other slide was placed on the top of the emulgel, so that the emulgel was sandwiched between the two slides in an area occupied by a distance of 7.5 cm along the slide. 100 grams weight was placed upon the upper slide so that the emulgel between the two slides was pressed uniformly to form a thin layer. The weight was removed and the excess of emulgel adhering to the slides was scrapped off. The two slides in position were fixed to a stand without slightest disturbance and in such a way that only the upper slide to slip off freely by the force of weight tied to it. A 20 grams weight was tied to the upper slide carefully. The time taken for the upper slide to travel the distance of 7.5 cm and separated away from the lower slide under the influence of the weight was noted. The experiment was repeated by three (3) times and the mean time taken for calculation.
Spreadability was calculated by using the following formula:

$$S = m \times \frac{l}{t}$$

Where,
S - Spreadability
m - Weight tied to the upper slide (20grams)
l - Length of the glass (7.5 cm)
t - Time taken in seconds (Haneefa, 2013; Oswal and Naik, 2014) [27]

Extrudability
The Extrudability test was carried out using hardness tester. A 15 gm of gel was filled in aluminum tube. The plunger was adjusted to hold the tube properly. The presence of 1kg/cm2was applied for 30 sec. the quantity of gel extruded was weighed. The procedure was repeated at 3 equidistance places of the tube (Kumar *et al*, 2016: Haneefa, 2013) [27].

Viscosity
Viscosity of the test emulgel was determined by using Brookfield viscometer (Haneefa, 2013) [27].

Photomicrography
Morphology of emulsion was studied under light microscope. The emulgel was suitably diluted with lugols solution and sodium chloride (0.85 w/v), mounted on glass slide and viewed by light microscope under magnification of 40 X.

Determination of microbial load
For total bacterial count, total fungal count and presence of *E. coli*, *Salmonella*, *S. aureus* and *pseudomonas* was carried out by Shree Krishna Analytical Services, New Delhi. The test UMVT was evaluated as per Quality Control Manual for Ayurvedic, Siddha and Unani Medicine (2008), PLIM. Ghaziabad (UP).

Heavy metal analysis
The heavy metal analysis for Lead (Pb), Arsenic (As), Mercury (Hg) and Cadmium (Cd) was done as per Quality Control Manual for Ayurvedic, Siddha and Unani Medicine (2008), PLIM, Ghaziabad (UP) which was carried out by Shree Krishna Analytical Services, New Delhi.

Observations & Results
Present study was conducted for pharmaceutical design and development of a Unani emulgel dosage formulation along with its standard operating procedure (SOP). In this study, a number of procedures and tests were performed in order to design and develop a Unani emulgel.

Table 6: Organoleptic character of extract

S.No.	Organoleptic Character	Observation
1	Appearance	Liquid
2	Color	Brown
3	Smell	Agreeable

Table 7: pH of extract

S.No.	pH in 1%	pH in 10%
1	6.68	6.59
2	6.72	6.62
3	6.77	6.64
Mean ± S.D	6.72±0.045	6.61±0.025

Determination of λ max of extract
The peak wavelength λ max of the extract under UV spectrum was 635 in phosphate buffer at pH fixed at 7.4.

Table 8: Absorbance at 635 of extract

S.No.	Concentration (%)	Absorbance at 635
1	05	0.0319
2	10	0.0780
3	25	0.1852
4	50	0.4086
5	75	0.5951
6	100	0.8011

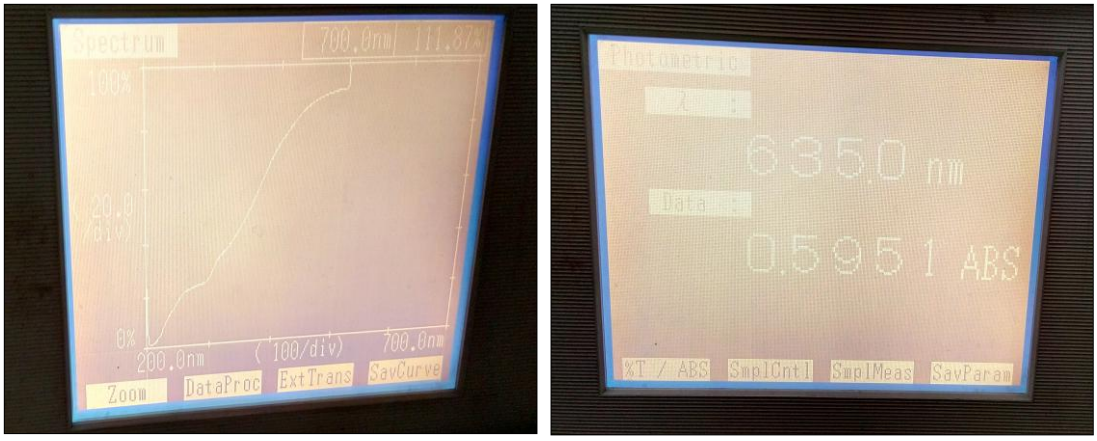

Fig 2: Absorbance at 635 of extract at 75%

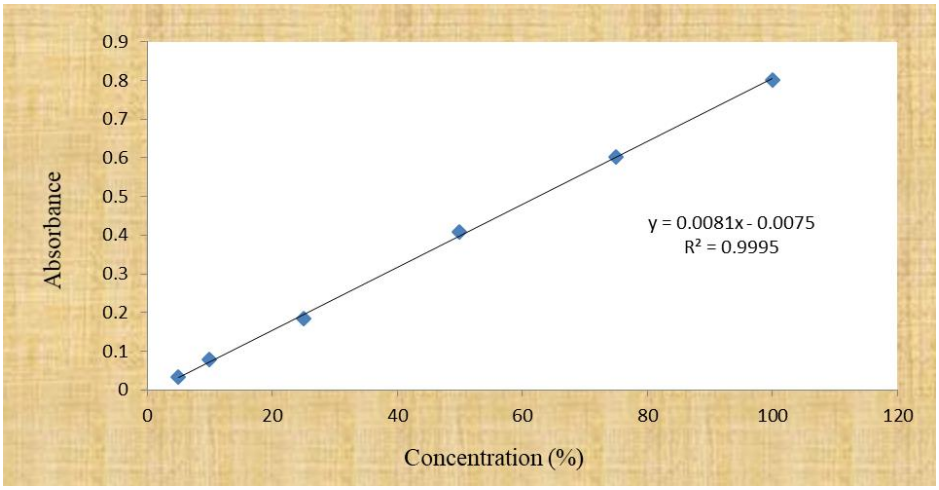

Fig 3: Absorbance at 635 of extract

Table 9: Version-1.0: Emulsion by using surfactant Tween 80

S.No.	Oil: Surfactant: Aqueous	Result
1.0	45:05:50	Separation after 10 minutes
1.1	50:05:45	Separation after 10 minutes
1.2	40:10:50	Separation after 2 hours
1.3	50:10:40	Separation after 2 hours
1.4	40:15:45	Separation after 18 hours
1.5	45:15:40	Separation after 18 hours
1.6	35:15:50	Separation after 18 hours
1.7	50:15:35	Separation after 18 hours
1.8	40:20:40	Thick, Separation after 36 hours
1.9	35:20:45	Thick, Separation after 36 hours
1.10	45:20:35	Thick, Separation after 36 hours
1.11	30:20:50	Thick, Separation after 36 hours
1.12	50:20:30	Thick, Separation after 36 hours

Table 10: Version-2.0: Emulsion by using surfactant Tween 20

S.No.	Oil: Surfactant: Aqueous	Result
2.0	45:05:50	Separation after 5 minutes
2.1	50:05:45	Separation after 5 minutes
2.2	40:10:50	Separation after 30 minutes
2.3	50:10:40	Separation after 30 minutes
2.4	40:15:45	Separation after 18 hours
2.5	45:15:40	Separation after 18 hours
2.6	35:15:50	Separation after 18 hours
2.7	50:15:35	Separation after 18 hours
2.8	40:20:40	Thick, Separation after 24 hours
2.9	35:20:45	Thick, Separation after 24 hours
2.10	45:20:35	Thick, Separation after 24 hours
2.11	30:20:50	Thick, Separation after 24 hours
2.12	50:20:30	Thick, Separation after 24 hours

Table 11: Version-3.0: Emulsion by using surfactant gum acacia

S.No.	Oil: Surfactant: Aqueous	Result
3.0	45:05:50	Separation after 18 hours
3.1	50:05:45	Separation after 18 hours
3.2	40:10:50	Separation after 72 hours
3.3	50:10:40	Separation after 72 hours
3.4	40:15:45	Stable emulsion no separation, no creaming nocracking & watched for 3 month.
3.5	45:15:40	Stable emulsion no separation, no creaming nocracking & watched for 3 month.
3.6	35:15:50	Stable emulsion no separation, no creaming no cracking &watched for 3 month.
3.7	50:15:35	Stable emulsion no separation, no creaming nocracking & watched for 3 month.
3.8	40:20:40	Emulsion, very thick.
3.9	35:20:45	Emulsion, very thick.
3.10	45:20:35	Emulsion, very thick.
3.11	30:20:50	Emulsion, very thick.
3.12	50:20:30	Emulsion, very thick.

Version-1.0: In the preparation of emulsion (oil in water), the aqueous extract was used as a water phase, and oil phase was made by *Roghan -e-Kunjad*, and the surfactant was used tween 80. It was observed that there was separation after 10 minutes in version-1.0 and version-1.1. In version-1.2 andversion-1.3, separation was noticed after 2 hours. In version-1.4, version-1.5, version-1.6 andversion-1.7, there was separation after 18 hours. In version-1.8, version-1.9, version-1.10, version-1.11, and version-1.12, emulsion was found thick, and separation noticed after 36 hours.

Version-2.0: In the preparation of emulsion (oil in water), the aqueous extract was used as a water phase, and oil phase was made by *Roghan -e-Kunjad*, and the surfactant was used tween 20. It was observed that there was separation after 05 minutes in version-2.0 and version-2.1. In version-2.2 andversion-2.3, separation was noticed after 30minutes. In version-2.4, version-2.5, version-2.6 andversion-2.7, there was separation after 18 hours. In version-2.8, version-2.9, version-2.10, version-2.11, and version-2.12, emulsion was found thick, and separation noticed after 24 hours.

Version-3.0: In the preparation of emulsion (oil in water), the aqueous extract was used as a water phase, and oil phase was made by *Roghan -e-Kunjad*, and the surfactant was gum acacia. It was observed that there was separation after 18 hours in version-3.0 and version-3.1. In version-3.2 andversion-3.3, separation was noticed after 72 hours. In

version-3.4, version-3.5, version-3.6 andversion-3.7, no separation, no creaming, no cracking was noticed during 3 months. Hence, these versions were found stable. In version-3.8, version-3.9, version-3.10, version-3.11, and version-3.12, emulsion was found very thick in consistency, and no separation, no creaming, no cracking was noticed.

After optimization of different versions of emulsion, it was observed that the separation occurred after some time when they were treated with tween 80 and tween 20. When aqueous phase and oil phase was treated with gum acacia it was noted that there was separation in version-3.0, version-3.1, vesion-3.2 and version-3.3, when quantity of gum acacia was up to 10%., but when quantity was increased up to 15% there was perfect emulsion, i.e., no creaming, no cracking, homogenous, globules was regular under electrical microscope. Hence, emulsion was stable in version-3.4, version-3.5, and version-3.6 and version-3.7. Out of these four versions, version-3.6(having 35%*Roghan -e-Kunjad*, 15% gum acacia and 50% aqueous extract) was selected for incorporation in gel. The emulsion was also found stable in version-3.8 to version-3.12, but these emulsions were found thick in consistency.

Optimization of Gel

Table 12: Result of gel optimization HPMC as a gelling agent

S. No.	% age of gelling agent	Result
a-1	0.5	Free flowing consistency
a-2	1.0	Free flowing consistency
a-3	1.5	Free flowing consistency
a-4	2.0	Gel consistency but less viscous
a-5	2.5	Gel consistency very good
a-6	3.0	Gel consistency but more viscous

Table 13: Result of gel optimization Carbopol -940 as a gelling agent

S. No.	% age of gelling agent	Result
b-1	0.5	Free flowing consistency
b-2	1.0	Free flowing consistency
b-3	1.5	Less viscous in consistency
b-4	2.0	Gel consistency but less viscous
b-5	2.5	Gel consistency very good
b-6	3.0	Gel consistency but more viscous

Optimization of Emulgel

Table 14: Result of prepared Emulgel HPMC as a gelling agent

S.No.	Version	Percentage	Result
1	Version-3.6 (a-1)	0.5	Free flowing consistency
2	Version-3.6 (a-2)	1.0	Free flowing consistency
3	Version-3.6 (a-3)	1.5	Less viscous in consistency
4	Version-3.6 (a-4)	2.0	Stable, good in consistency but less viscous
5	Version 3-6 (a-5)	2.5	Stable very good in consistency and sticky
6	Version-3.6 (a-6)	3.0	Stable, but more viscous consistency and sticky

Table 15: Result of prepared Emulgel Carbopol 940 as a gelling agent

S.No.	Version	Percentage	Result
1	Version-3.6 (b-1)	0.5	Free flowing consistency
2	Version-3.6 (b-2)	1.0	Free flowing consistency
3	Version-3.6 (b-3)	1.5	Less viscous in consistency
4	Version-3.6 (b-4)	2.0	Stable, good in consistency but less viscous
5	Version-3.6 (b-5)	2.5	Stable, very good in consistency
6	Version-3.6 (b-6)	3.0	Stable, but more viscous in consistency

Emulgel prepared with HPMC as a gelling agent

When version-3.6 was incorporated in different ratios of gel, it was observed that the emulgel was free flowing in consistency when quantity of HPMC was up to 1.5%. When concentration was increased up to 2%, the emulgel was found stable but less viscous. Again the concentration was increased up to 2.5%, emulgel was found stable and good in consistency, and sticky in nature. Further increasein concentration up to 3%, emulgel was found stable, but more viscous and sticky.

Emulgel prepared with Carbopol 940 as a gelling agent

When version-3.6 was incorporated in different ratios of gel, it was observed that the emulgel was found free flowing in consistency in concentration up to 1% of Carbopol 940.When concentration was increased up to 1.5%, less viscous consistency was observed. Again concentration was increased up to 2%, the emulgel was found stable, and less viscous consistency was observed. Further increase in concentration of Carbopol 940 up to 2.5%, the emulgel was found stable, very good in consistency whereas up to 3%, the emulgel was found stable and more viscous. Hence, the version-3.6 (b-5) having Carbopol 940 in the concentration of 2.5% was finalized for further study.

Table 16: Organoleptic characteristics of emulgel version-3.6 (b-5)

S. No.	Organoleptic Character	Observation
1	Appearance	Liquid
2	Color	Light brown
3	Grittiness	No
4	Homogeneity	Homogenous
5	Phase separation	None
6	Smell	Agreeable

Fig 4: Prepared emulgel version-3.6 (b-5)

In Vitro Permeation Study of Version-3.6 (b-5)

Table 17: Qualitative analysis

S. No.	Test	Extract	Emulgel	*Roghan*
1.	Alkaloids (Dragendorff's Test)	Strongly positive (orange red precipitate)	Positive (orange red precipitate)	Positive (orange red precipitate)
2.	Carbohydrates (Benedict's Test)	Strongly Positive (red precipitate)	Positive (red precipitate)	Positive (red precipitate)
3.	Proteins (Lead Acetate Test)	Strongly Positive (white precipitate)	Positive (white precipitate)	Positive (white precipitate)

Quantitative Analysis

Table 18: Quantitative analysis Emulgel version-3.6 (b-5)

S. No.	Time (min)	Absorbance	% Drug Release	Mean% drug release ± S.D
1	0	0.0000	0.0000	0±00
2	3	0.0197	3.3375	3.6±0.357
3	5	0.0300	4.625	4.61±0.425
4	10	0.049	7.0	7.03±0.201
5	15	0.073	10.0	10.23±0.634
6	20	0.098	13.125	13.34±0.610
7	25	0.126	16.625	16.77±0.916
8	30	0.137	18	18.06±0.266
9	35	0.146	19.125	20.33±1.051
10	40	0.176	22.875	22.47±0.635
11	45	0.184	23.875	23.69±0.512
12	50	0.228	29.375	29.15±0.349
13	55	0.236	30.375	30.76±0.353
14	60	0.286	36.625	36.47±0.588
15	75	0.384	48.875	49.13±0.252
16	90	0.412	52.375	51.74±0.572
17	105	0.426	54.125	54.44±0.510
18	120	0.438	55.625	56.00±0.372
19	135	0.449	57.0	57.08±0.170
20	150	0.465	59.0	59.32±0.596
21	165	0.474	60.125	60.62±0.460
22	180	0.482	61.125	60.80±0.408
23	195	0.497	63.0	62.55±0.489
24	210	0.501	63.5	64.08±0.634
25	225	0.52	65.875	65.95±0.184
26	240	0.533	67.5	67.83±0.800
27	255	0.549	69.5	68.88±0.784
28	270	0.553	70.0	70.45±0.562
29	300	0.563	71.25	71.67±0.546
30	330	0.581	73.5	73.14±0.307
31	360	0.592	74.875	75.20±0.467
32	1440	0.628	79.375	78.82±0.557

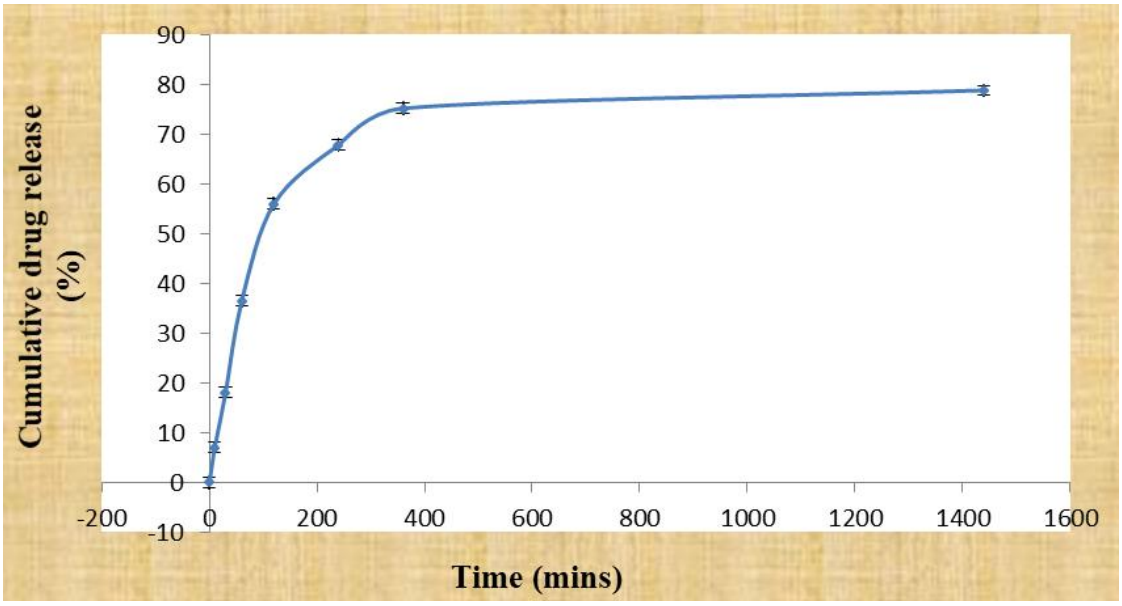

Fig 4: Cumulative Drug Release of emulgel version-3.6 (b-5)

The mean amount of cumulative drug release (CDR) of emulgel during first 60 minutes was rapid which slowed down as the experiment proceeded. The amount of drug release was 3.6±0.357% in first 3 minutes and increased to 10.23±0.634% within 15 minutes, and reached to 18.06±0.266% in 30 minutes, reached to 36.47±0.588% in 60 minutes, the drug release at the end of 120 minutes was 56.00±0.372%, and reached to 67.83±0.800% in 240 minutes. The CDR in 360 minutes was 75.20±0.467%. The mean CDR in 24 hours was 78.82±0.557%.

Table 19: Water soluble matter in emulgel

S. No.	Water soluble matter (%)
1.	72.2.
2.	73.14
3.	73.51
Mean ±SD	72.97 ± 0.631

Table 20: Alcohol soluble matter in emulgel version-3.6 (b-5)

S. No.	Alcohol soluble matter (%)
1.	48.62
2.	49.71
3.	47.98
Mean ± SD	48.77 ± 0.874

Table 21: Loss on drying of emulgel version-3.6 (b-5)

S. No.	Loss on drying%
1.	32.40
2.	33.16
3.	32.93
Mean ±SD	32.83 ±0.389

Table 22: Moisture content of emulgel version-3.6 (b-5)

S. No.	Moisture content
1.	78.82
2.	79.43
3.	78.19
Mean ±SD	78.81± 0.620

Table 23: Spreadability of emulgel version-3.6 (b-5)

S. No.	Spreadability(cm/sec)
1.	17.04
2.	17.13
3.	16.98
Mean ±SD	17.05 ±0.075

Table 24: Extrudability of emulgel version-3.6 (b-5)

S.No.	Extrudability
1.	0.98
2.	0.94
3.	0.95
Mean ± SD	0.95 ±0.020

Table 25: Viscosity of emulgel version-3.6 (b-5)

S.No.	Viscosity (cps)
1.	5.9×10^4
2.	6.2×10^4
3.	6.0×10^4
Mean ±SD	$6.03\pm0.152\times10^4$

Table 26: pH of emulgel version-3.6 (b-5)

S.NO.	pH in 1%	pH in 10%
1	7.21	7.11
2	7.18	7.15
3	7.20	7.18
Mean ± S.D	7.19±0.015	7.14±0.028

Table 27: Stability of emulgel version-3.6 (b-5)

S.No	Duration	Color	Phase separation (creaming / cracking)	Grittiness	Homogeneity	pH		Viscosity (cps) × 10^4
						1%	10%	
1.	0 day	Light brown	None	No	Homogenous	7.19	7.14	6.03
2.	60 days	Light brown	None	No	Homogenous	7.21	7.11	5.94
3.	120 days	Light brown	None	No	Homogenous	7.17	7.18	5.91
4.	180 days	Light brown	None	No	Homogenous	7.23	7.15	5.90

Contamination

No contamination was observed in the test version-3.6 (b-5) in 180 days of stability / shelflife.

The total bacterial count was found to be 4855 micro-organisms per gram (NMT 100000 micro-organisms per gram).The total fungal count were found absent per gram. As regard to *E. coli, Salmonella, S. aureus* and *Pseudomonas* were absent.

The heavy metal analysis showed that Lead, Arsenic, Mercury and Cadmium were less than permissible limit as per WHO guidelines.

Discussion

The test formulation based on nine (9) single Unani drugs chosen has been a novel combination for anti-inflammatory and analgesic properties. It has also been mentioned that these nine (9) single Unani drugs possess potent *mohallil-e-auram* (anti-inflammatory) in almost all *qarabadeen* (pharmacopoeias). The uniqueness of this formulation lies in the fact that it is the one of the Unani classical formulations which has been used for over a long period of time.

The ingredients of test formulation were collected from the herbal garden at Ayurvedic & Unani Tibbia College & Hospital, Karol Bagh, New Delhi. *Roghan-e-Kunjad* (Sesame oil) was procured from traditional oil expeller (*bail kolhu*) at Ballimaran in old Delhi. Samples of ingredients were separately packed, and sent for identification. The identity of all drugs was established by the experts of Raw Material Herbarium and Museum, CSIR-National Institute of Science Communication and Information Resources (NISCAIR), New Delhi. The specimen vouchers of all ingredients were deposited in the Drug Museum, Department of Ilm-ul-Advia, Ayurvedic & Unani Tibbia College & Hospital, Karol Bagh, New Delhi. The authentication letter issued by NISCAIR.

In this study, a number of procedures and tests were employed to design and develop a Unani anti-inflammatory & analgesic emulgel (UAAE) with standard operating procedure (SOP).As many as thirty nine (39) versions of emulsion and twelve (12) versions of emulgel were prepared. In the preparation of emulsion (oil in water), different surfactants, namely tween 20, tween 80, gum acacia were used. The aqueous extract was used as a water phase, and oil phase was made by *roghan-e-Kunjad.* The emulsion was made in the various percentageratio of oil phase: surfactant: aqueous phase.

The surfactant was used in 5 to 20%. The separation was found in all twenty six (26) versions, i.e., thirteen each (13) versions having tween 20 and tween 80, respectively. As regard to thirteen (13) more versions prepared with gum acacia, it was observed that the four (4) versions prepared by using surfactant gum acacia up to 10% showed separation whereas the other four (4) versions in 15% were found stable. The remaining five (5) versions in 20% were found stable but consistency was found thick. Based on the stability. consistency, homogeneity and grittiness, the versions prepared in 15% were selected for incorporation in gel for making emulgel. Out of the versions prepared in 15%, the version-3.6 was finalized for emulgel making.

For preparation gel, twelve (12) different versions divided six (6) each versions having HPMC and Carbopol 940, respectively in the percentage of 0.5.to 3.0 were prepared. The version-3.6 was added in version a-1 to a-6 prepared with HPMC. In these version, it was observed that these versions were free flowing and sticky in nature. These version were found not up to mark. Thus, these versions were rejected.

In another set of six (6) versions based on Carbopol 940 in the percentage of 0.5.to 3.0 were prepared. The version-3.6 was added in version b-1 to b-6 prepared with Carbopol 940.

In these versions, it was observed that these versions were free flowing, viscous and stable. The version-3.6 (b-5) was found up to mark because it was found to be stable, homogenous, greaseless, thixotropic and emollient. Thus, this test version was finalized for further study. The test emulgel version-3.6 (b-5) was characterized as light brown in color, homogenous, no phase separation (i.e., no creaming, no cracking), no sign of grittiness, better stability, high loading efficiency, greaseless, thixotropic, easily spreadable, easily removable, emollient, non-staining, water-soluble, longer shelf life, bio-friendly, pleasing appearance and more production economical with low cost. The mean values pH of 1% & 10% solutions of prepared emulgel were 7.19 ± 0.015 & 7.14 ± 0.028, respectively. The mean Spreadability 17.05 ± 0.075 cm/sec, mean viscosity 6.03 ± 0.152×10^4cps, Extrudability 0.95 ± 0.020, water soluble matter 72.97 ± 0.631, alcohol soluble matter 48.77 ± 0.874, Moisture content 32.83 ± 0.389, Loss in weight on drying at 105^0C 78.81 ± 0.620.

The presence of alkaloids, carbohydrates and proteins in the test emulgel version-3.6 (b-5) was detected in the qualitative phytochemical tests.

The mean amount of cumulative drug release (CDR) of test emulgel during first 60 minutes was rapid which slowed down as the experiment proceeded. The amount of drug release was found to be 3.6 ± 0.357% in first 3 minutes, 10.23 ± 0.634% in 15 minutes, 18.06 ± 0.266% in 30 minutes, 36.47 ± 0.588% in 60 minutes, and in 120 minutes CDR was 56.00 ± 0.372%, 67.83 ± 0.800% in 240 minutes, and raised up to 75.20 ± 0.467% in 360 minutes. The mean CDR in 24 hours was 78.82 ± 0.557%. It showed a fare amount of drugs released in the body.

The test emulgel version-3.6 (b-5) was found stable during the period of 6 months. The total bacterial count was found to be 4855 micro-organisms per gram (NMT – 100000 micro-organisms per gram). The total fungal count was found absent per gram. *E. coli*, *Salmonella*, *S. aureus* and *Pseudomonas* were found absent.

The heavy metal analysis showed that Lead (Pb), Arsenic (As), Mercury (Hg) and Cadmium (Cd) were less than permissible limit as per WHO guidelines.

No contamination was observed in the test emulgel version-3.6 (b-5) in 180 days of stability / shelf life.

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