Research Article

Design and Evaluation of Isolated Herbal Extracts of Aloin, Proanthocyanidin, Quercetin Loaded Microsponge as Sunscreen Gel

Venkatrajan Muruganantham[1,*], Bendi Sri Venkateswarlu[1], Vignesh Chinraj[2]

[1]Department of Pharmaceutics, Vinayaka Mission's College of Pharmacy, Vinayaka Mission's Research Foundation (Deemed to be University), Salem, Tamil Nadu, INDIA.
[2]Vinayaka Mission's College of Pharmacy, Vinayaka Mission's Research Foundation (Deemed to be University), Salem, Tamil Nadu, INDIA.

Submission Date: 22-05-2022; Revision Date: 30-06-2022; Accepted Date: 16-07-2022.

ABSTRACT

The goal of this research is to produce and create a technique for using microsponge gel as a polyherbal formulation, as well as methods for evaluating sunscreen qualities. Using the quasi-emulsion solvent diffusion method, polyherbal extracts as an active component for preparing microsponge formulations with changes in the ratio of a drug, i.e. Polyherbal extracts, ethylcellulose as a polymer, and Polyvinyl alcohol as an emulsifier present in the formulation, was successfully obtained. The significant characterisation was considered for the formulations developed. The physical description of a microsponge formulation with a Gel-3 covering shows that it has a higher loading efficiency and yield. The microsponge formulation was made with the assistance of a gel and carbopol, and then pH, viscosity, spreadability, drug content, and *in-vitro* release tests were done. The majority of microsponge preparations are made by putting drugs into them, and the results are easy to replicate. Using a Franz diffusion cell, the optimal formulation of microsponge-loaded gel of polyherbal extract. The microsponge-loaded sunscreen gel has an outstanding release rate, according to the release profile.

Keywords: Microsponge, Aloin, Proanthocyanidin, Quercetin, Quasi-emulsion solvent diffusion method, Sunscreen, Polyherbal microsponge gel.

Correspondence:
Prof. Dr. Venkatrajan Muruganantham,
Department of Pharmaceutics, Vinayaka Mission's College of Pharmacy, Vinayaka Mission's Research Foundation (Deemed to be University), Salem-636308, Tamil Nadu, INDIA.

Email: svmanand@gmail.com

INTRODUCTION

Herbal Medicine[1,2]

Plants and their components, which are used in medicine, might be a source of help in the early stages of human development. We have a varied variety of plant habitats and a huge number of plants that may be cultivated in different sections of the nation thanks to the environment. With 75-80 percent of the population adopting herbal treatment, the therapeutic use of plants in the form of herbal treatment is still a popular therapy for treating a range of disorders. The utilisation of plant extracts and active components is a key component of these traditional medicines. Secondary metabolites discovered in medicinal plants that have therapeutic value include alkaloids, steroids, tannins, phenol compounds, flavonoids, resins, fatty acids, and gums. Active components may be found in herbal remedies. Many herbal treatments' active constituents are yet unknown. A single active component produced from plants is found in a number of pharmaceutical medications. Herbal medicine practitioners feel that separating an active ingredient from the rest of the plant reduces its potency or makes it less safe.

SCAN QR CODE TO VIEW ONLINE

www.ajbls.com

DOI: 10.5530/ajbls.2022.11.49

Microsponge[3,4]

"Microsponges are porous microsphere-based polymeric delivery devices. There are tiny sponge-like particles with a large porous surface." Microsponge is a relatively

new technology for controlling medication release and delivering drugs to precise targets. They want to distribute API effectively at the lowest dose possible, as well as improve stability, eliminate side effects and adjust medication release. A patented, porous, highly cross-linked polymeric microsphere that can entrap a wide range of actives, release them into the skin over time, and then sponge to trigger is known as a microsponge delivery system. The technique was used to improve the performance of medications that were applied topically. Microporous beads, typically 10-25 microns in diameter and filled with an active agent, make up this one-of-a-kind technology for the controlled release of topical medicines. Drug release can be controlled by diffusion or a variety of other mechanisms when a microsponge delivery system is applied to the skin.

Sunscreen Protedtion[5-7]

UVB radiation causes sunburn, however, ultraviolet A radiation may be more harmful to the skin. In an ideal world, sunscreens would block both wavebands. A sunscreen's sun protection factor is primarily determined by its ability to filter ultraviolet B rays. It doesn't assess how efficient it is at blocking ultraviolet A. Organic or inorganic compounds can be used as sunscreens The cosmetic acceptability of polyherbal unscreens may be improved if they are formulated as microsponges. This sunscreen formulation has antioxidant, anti-inflammatory, and antimutagenic properties. UVA (Longwave Radiation) The wavelength range of 320-400 nm. Although erythrogenic activity is minimal, it penetrates the dermis. There is a responsible for the gradual development of a natural tan and the majority of drug-induced photosensitivity occurs, UVA could amplify the effects of UVB. UVB (Middlewave Radiation) The wavelength range of 290-320 nm. The maximum level of erythrogenic activity is found and produces new pigments, sunburn, and vitamin D production. Skin cancer is caused by this substance and UVC (Shortwave or Germicidal Radiation) The wavelength range of 100-290 nm. Released from artificial ultraviolet sources and it does not reach the earth's surface.

MATERIALS AND METHODS

Herbal Materials

Collection of plant materials of the fresh leaf of *Aloe vera* and fresh bulb of *Allium cepa* collected from yercaud foothills garden and dried seeds of *Vitis labrusca* collected from a vegetable market in yercaud. Further authentication of samples was done in the ABS HERBAL GARDENS in Salem by botanist DR.A.BALASUBRAMANIAN. The plant materials obtained were shade dried and not contaminated for up to 3-4 weeks. Plant materials were ground using an electronic grinder, and the powered mixture was examined for physiological properties such as colour, odour, taste, and texture. Then the powdered plant materials were stored in air-tight containers for further analysis like the phytochemical and biological analysis.

Aloe Vera[7]

Aloe vera gel is extensively utilized in cosmetics and toiletries for its moisturizing and revitalizing action. It blocks each UVA and UVB rays and holds the skin's herbal moisture balance. When UV radiation damages molecules in the skin, the immune system attempts to restore the harm by releasing inflammatory proteins. Aloe vera additionally stimulates collagen manufacturing to enhance elasticity and dryness in the skin. Aloe vera gel or juice is antimicrobial and aids in the prevention of skin peeling after prolonged sun exposure. Aloe vera additionally has anti-inflammatory activity and excessive water content material that allows hydrating of the skin. Aloe vera is mainly used in the formulation based on Moisturises, Acne, Eczema, cuts, skin whitening, Psoriasis and Sunburn.

Grape seed[8]

Grape seed active constituent of Proanthocyanidin used as an Antioxidants, Anti-microbial, Anti-inflammatory and cell turnover, collagen synthesis. The antioxidant proanthocyanidin (OPC) acts as a DNA mutation inhibitor. Furthermore, OPC inhibits elastase, preserving the integrity of elastin in the skin, and works synergistically with vitamins C and E to protect and replenish the skin. Grape seed extract boosted the SPF/PPD value of sunscreen products and antioxidant activity.

Onion[9]

All onions include quercetin, but since it is a pigment, red and yellow onions include the most. Onions include many other vitamins and minerals, and they're mainly rich in vitamin biotin. Quercetin has anti-inflammatory and antioxidant consequences and acts as an immunomodulator. Quercetin and rutin have been examined as ability topical sunscreen elements in people and discovered to protect the UVA and UVB range. Quercetin is in a position to reduce redness, itching, and irritation of broken skin; it can additionally assist repair skin barrier function, growing hydration, and decreasing water loss. Onions may also aid in the formation and

maintenance of collagen due to their high vitamin C content.

Extraction process[9-11]

Extraction Aloin from Aloe vera

The aloe plant's juice is extracted and dried, and the quantity of extract contained is determined by using methanol or higher alcohols. Crude aloin has been separated after the removal of sugars, fats, and colours. During aloin purification, aloin is removed from its natural condition and reintroduced.

Extraction of Proanthocyanidin from Grape Seed

This maceration of one part of the powdered grape seed powder in three parts % ethanol was carried out for one week. As soon as the filtrate was collected, it was concentrated in a 40°C rotating vacuum evaporator. It took 2-3 days to dry the remaining filtrate to a consistent weight in a hot air oven set at 40°C. The crude extract of proanthocyanidins was isolated and purified.

Extraction of Quercetin from Onion

Quercetin may be extracted from powdered onion using this approach. The effects of time, temperature, and solvents employed in the extraction process were examined in the course of the study. – Crude quercetin was successfully isolated from onion scales using a rapid, simple, and efficient 4-hour extraction technique including shaking with cold ethyl acetate. The final result is a yellow-colored powder with a purity level of 70%. The yield of the procedure was marginally increased by hot extraction, but the purity of the output was reduced. Crude quercetin was greasy and included up to 78% of impurities after extraction with ethanol with varying amounts of water. It is necessary to further purify such a product.

Determination of Concentration of Aloin, Proanthocyanidin and Quercetin of the powdered extract[12]

An amount of around 50 mg of powdered extract formulations was added to distilled water (50 ml) at a given time and stirred with a magnetic stirrer for 1 hr before filtering the solution. This solution was thoroughly washed with chloroform, with about 40 ml being utilised in a separator funnel to remove non-polar contaminants such as caffeine and pigments. The solution was washed four times with chloroform, and the absorbance was measured at 298 nm with the water phase extract. The water phase extract's absorbance was measured as maximum absorbance of Pure aloin, proanthocyanidin and quercetin were noted at 297nm, 281nm and 258nm using UV- Visible spectroscopy. The results are shown in Table 4 and Figure 1.

METHODS

Preparation of Polyherbal Loaded Microsponge[12-13]

Eudragit RS-100 polymer-based aloe vera extract, grape seed extract and onion extract loaded microsponge were formulated by using the method of quasi-emulsion solvent diffusion. The internal phase consists of polyherbal extract (100-600 mg), ethylcellulose and eudragit RS-100 and tri-ethyl citrate as a plasticizer. They are dissolved in dichloromethane and ethanol (1:1). Addition of the extracts to this solvent by gradually stirring at a speed of 500 rpm. The internal phase was added dropwise into the aqueous external phase. Then continuous stirring up to 2 hr. The removal of dichloromethane and ethanol from the system resulted in the formation of microsponges. Microsponges obtained were filtered and dried at 40°C for 12 hr. The preparation procedure is shown in the Table 1.

Evaluation of Microsponges[13-15]

Particle Size Analysis

An optical microscope was used to evaluate the average particle size of polyherbal microsponge using a calibration ocular and stage micrometre under regular polarised light. The average particle size was obtained by measuring 100 particles of each batch using a little amount of microsponge dispersed on a clean glass slide. Particle size was determined and shown in Table 3 and Figure 2.

Percentage Yield Determination

All batches of manufactured herbal microsponges were weighed properly. The total amount of excipients and drugs utilised in the preparation of the herbal microsponges was divided by the measured weight of the prepared herbal microsponges. The following equation used to be calculated and reports shown in Table 3.

$$\% \text{ Yield} = \frac{\text{Practical yield}}{\text{Theoretical yield (herbal extracts + polymer)}} \times 100$$

Determination of Drug Loading Efficiency

100 mg crushed microsponges were dissolved in a small amount of ethanol in a 100 ml volumetric flask and sonicate for 20 min then the volume was make up to the mark with acetate buffer pH 5.5. 10 ml from the above solution diluted and make up with the mark, absorbance of the drug was measured with a UV-Spectrophotometer

Table 1: Composition of different batches of herbal microsponge formulation.

Formulation	Aloe vera extract (mg)	Grape seed extract (mg)	Onion extract (mg)	Eudragit RS-100 (mg)	Ethylcellulose (mg)	Dichloromethane and ethanol (ml)	Polyvinyl alcohol (g)
HM 1	100	100	100	300	100	5	0.5
HM 2	200	200	200	300	100	5	0.5
HM 3	300	300	300	300	100	5	0.5
HM 4	400	400	400	300	100	5	0.5
HM 5	500	500	500	300	100	5	0.5
HM 6	600	600	600	300	100	5	0.5

Table 2: Composition of different batches of herbal microsponge gel formulation.

Ingredients	Gel 1	Gel 2	Gel 3
Optimized microsponge	500 mg	500 mg	500 mg
Carbopol 940	0.25 gm	0.50 gm	0.75 gm
Propylene glycol	300 mg	300 mg	300 mg
Methylparaben	5 mi	5 ml	5 ml
Propylparaben	1ml	1 ml	1 ml
Methanol	30 ml	30 ml	30 ml
Triethanolamine (surfactant /pH adjuster)	Qs	Qs	Qs
Purified water	Qs	Qs	Qs

Table 3: Percentage yield, percentage drug loading and particle sizes of formulations.

Formulation code	Average particle size (µm)	Percentage yield (%)	Drug loading efficiency (%)
HM 1	60.43 ±2.54	74.98±1.90	56.74±2.30
HM 2	64.94 ±1.76	78.65±2.09	58.12±1.32
HM 3	69.24 ±1.43	81.00±1.23	60.98±1.22
HM 4	70.97 ±1.28	84.80±1.62	61.43±1.74
HM 5	72.87 ±2.03	88.70±1.52	63.79±2.04
HM 6	72.63 ±6.87	87.60±1.23	63.01±1.72

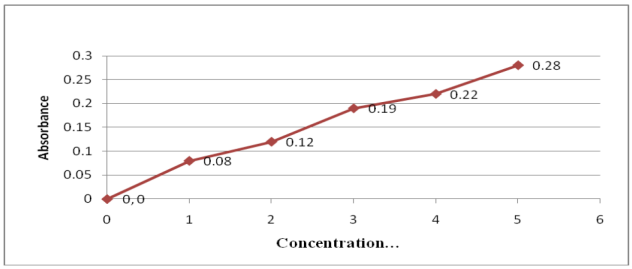

Figure 1: Calibration of Water Phase Extracts.

at 298nm. The following calculation can be used to formulate the microsponges' drug load efficiency and reports shown in Table 3.

Table 4: Absorbance of Water Phase Extracts.

Sl.No	Concentration (µg/ml)	Absorbance λ_{max} 298 nm
1	0	0.00
2	1	0.08
3	2	0.12
4	3	0.19
5	4	0.22
6	5	0.28

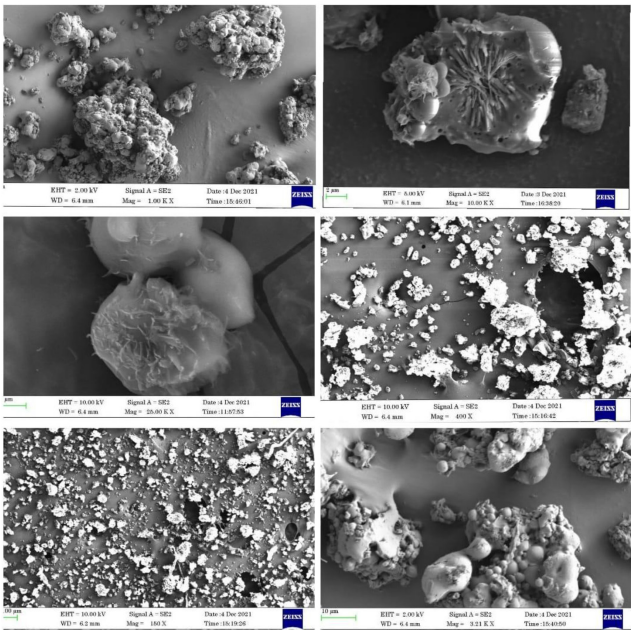

Figure 2: SEM Photographs of Polyherbal Microsponge.

The actual drug content in microsponges

$$\text{Loading efficiency} = \frac{\text{The actual drug content in microsponges}}{\text{Theoretical drug content}} \times 100$$

Fourier Transform Infrared Spectroscopy[13,16]

In an ATR-FTIR spectrophotometer, a pure sample of Aloin, proanthocyanidin, quercetin, and polyherbal loaded microsponges samples was exposed to Fourier

transform infrared spectroscopy utilising KBr pellets (Perkin Elmer spectrum BX II) in the range from 4000 to 400 cm^{-1} and IR spectrums of aloin, proanthocyanidin, quercetin and herbal microsponge the results are shown in Figure 3-6.

Morphology Study Using Scanning Electron Microscope[13,17]

Optimized formulation of HM5 was examine Scanning electron microscopy may be used to the internal and exterior morphology, as well as the surface topography (SEM). After produced herbal microsponges were coated with gold-palladium in an argon atmosphere at room temperature, SEM pictures of herbal microsponges were obtained at the requisite magnification. The structure and function of a shattered microsponge particle may be shown using a SEM picture. SEM photograph is shown in Figure 2.

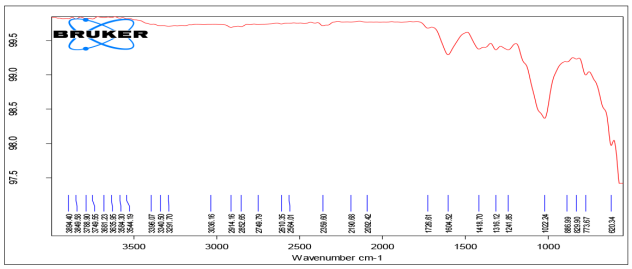

Figure 3: ATR-FTIR Spectrum of Aloin.

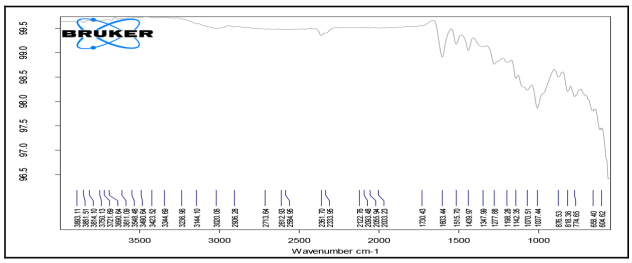

Figure 4: ATR-FTIR Spectrum of proanthocyanidin.

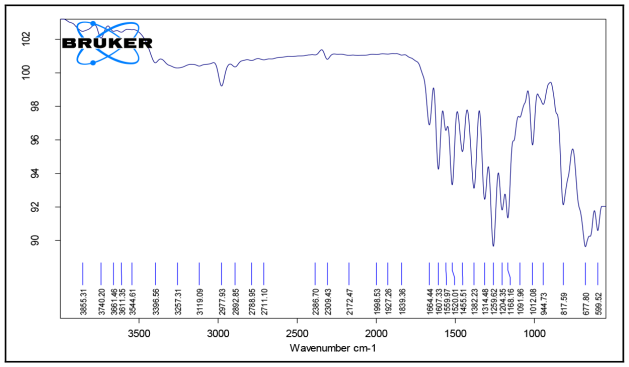

Figure 5: ATR-FTIR Spectrum of quercetin.

Scanning Electron Microscope

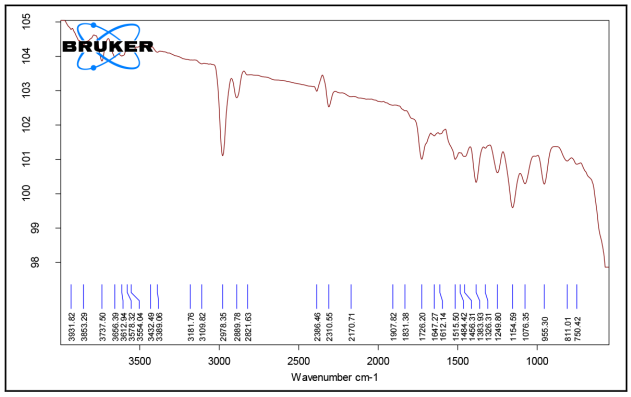

Figure 6: ATR-FTIR Spectrum of microsponge formulation.

PREPARATION OF POLYHERBAL LOADED MICROSPONGES GEL[12,18]

Step-I Preparation of Carbopol gel base

A weighed quantity of Carbopol 940 was gently added to distilled water and swirled continuously for 1 hr using a mechanical stirrer. Preservatives were also added to the gel base, which was then mixed with a mechanical stirrer once more. To prevent microbiological development, Methyl Paraben and Propyl Paraben were used as preservatives.

Step-II Preparation of Gel Formulation

Weighed amount of herbal microsponge powder formulation was solubilized in an appropriate amount of ethanol and addition of propylene glycol. The mixture was stirred gradually 1 hr. Mixture of herbal microsponges was transferred to aqueous dispersion of carbopol gel base. Prepared gel were degassed with ultrasonication. Finally pH was adjusted with Triethanolamine. The pH was adjusted 6-7. The preparation procedure are shown in Table 2.

Evaluation Of Gel Containing Microsponge[12,13,14,19]

After a visual inspection for consistency, colour, and homogeneity, the gel was assessed for the following characteristics.

pH Determination

By placing the electrode tip into the prepared gel and recording the result after 2 min, the pH of the gel was determined using a pH metre (standardised using buffer, pH 7 before use). The pH of the formulation was measured three times and the mean value was obtained and reports shown in Table 5.

Spreadability

The spreadability of the produced microsponge gel was determined by spreading 0.5 g of the gel on a

Table 5: Determination of Visual Inspection, pH, Spreadability, Viscosity and percentage Drug content.

Parameter	HMG 1	HMG 2	HMG 3
Visual Inspection	Slightly lemon yellow colour and slightly viscous gel.	Lemon yellow colour and viscous gel.	Dark Lemon yellow colour and viscous gel.
pH	4.5±0.05	4.6±0.24	4.8±0.20
Spreadability (g.cm/sec)	5.5±1.21	5.7±1.31	5.9±2.91
Viscosity(cps)	1935±1.98	2298±1,93	2309±2.54
% Drug content	85.9±1.13	88.7±1.65	89.6±1.03

circle premarked on a glass plate with a diameter of 2 cm, followed by a second glass plate. For 5 min, a half kilogramme of weight was allowed to lie on the top glass plate. The circle's diameter was measured after the gel had been spread out. The diameter of the gels grew as they spread. Spreadability values are reported in Table 5.

$$S = M.L / T$$

M- Weight tied to the upper slide
L - Length moved on the glass.
T - Time Taken

Viscosity

The viscosity was measured using a Brookfield digital viscometer. The sample was placed in the Brookfield Digital Viscometer's detachable sample holder, which was then put into a flow jacket installed on the viscometer. The viscosity of the preparations was measured using a tiny sample adapter (spindle) rotating at a speed of 20 rpm. The gel formulation's viscosity was determined. The results are shown in Table 5.

Drug content

1 g polyherbal loaded microsponge gel was weighed and dissolved in ethanol, then sonicated for 10-15 min and made up to the mark in a 100 ml volumetric flask with acetate buffer pH 5.5. To reach a concentration within Beer's range, 10 ml was pipetted out and diluted to 100 ml using acetate buffer pH 5.5 and the final dilution was produced with distilled water. The absorbance was measured at 298 nm using a UV spectrophotometer against a blank gel that had been prepared in the same way as the sample. The results are shown in Table 5.

Irritancy test[20-22]

Apply prepared gel on the backside of the left hand. Then the area of gel applied and time is taken into consideration. Irritation on applied area, eczema, other rashes are observed within 24 hr after the application of the gel.

In-vitro Drug Release Studies of Polyherbal Microsponge Formulations[20,21]

Franz diffusion cells with cellulose membranes were used to test the gel *in vitro*. The cellulose membranes were soaked in distilled water for 1 hr at room temperature before being mounted in Franz-type diffusion cells. Acetate buffer pH 5.5 was used to fill the receptor compartments. A Franz diffusion cell with a 30 ml receptor compartment and an effective area of 4.52 cm2 was put on a thermostatic magnetic stirrer and kept at 37°C for the duration of the experiment. For the diffusion investigation using the diffusion cell, herbal microsponge gel (HMG1, HMG2, HMG3) was employed. The donor compartment received each gel formulation (20 mg/cm2). At 0 hr, 1 hr, 2 hr, 3 hr, and 8 hr intervals, aliquots of 1 ml volume were removed and replaced with an equivalent amount of receptor medium. The aliquots were diluted with the receptor media to the desired concentration. Samples were withdrawn and absorbance was measured using a UV spectrophotometer set found to Aloin λ_{max} at 297 nm, Proanthocyanidin λ_{max} at 281 nm and Quercetin λmax at 258 nm. The results are shown in Table 6,7,8 and Figure 7,8,9.

In vitro UV Study[23-24]

The Sun Protecting Factor (SPF) is a comparison of the amount of UV radiation necessary to cause a minimal erythemal dose (MED) in protected vs. unprotected skin. Screening the product's absorbance between 290 and 320 nm at 5 nm intervals is a simple and quick way to determine the SPF *in vitro*. The following formulae may be used to calculate the Sun Protecting Factor (SPF). (Equation of Mansur): The results are shown in Table 9,10,11 and 12.

$$\% \text{ Yield} = \frac{\text{Practical yield}}{\text{Theoretical yield (herbal extracts + polymer)}} \times 100$$

CF - Correction Factor (10)
EE - Erythmogenic effect of radiation with a wavelength
Abs - spectrophotometric absorbance values at a wavelength
EE, x, and I are constants.

Stability Study[12,25]

ICH criteria were used to examine the stability of herbal microsponges gel under accelerated settings. For three months, the microsponge compositions were maintained at 40°C and 75% relative humidity. Microsponges were examined for physical appearance, pH, spreadability, *in vitro* drug release, and SPF value after three months. The results are shown in Table 13.

RESULTS

To make polyherbal loaded microsponges, the quasi-emulsion solvent diffusion technique was applied. These microsponges were coated with carbopol gel. Different characteristics were used to characterise the microsponges, and the gel was assessed using various approaches.

Evaluation of Polyherbal Microsponge

Different process parameters were evaluated of different formulations like percentage yield, percentage drug loading, and particle sizes of formulations, as per below given Figure 6 and other results are shown in Table 3.

Determination of Concentration of Aloin, Proanthocyanidin and Quercetin in Formulations

Determination of Absorbance of Water Phase Extracts of aloin, proanthocyanidin and quercetin in the formulations, resulted in Table 4 and linearity were noted in Figure 1.

Fourier Transform Infrared Spectroscopy

Evaluation of Polyherbal Microsponge Gel

Visual inspection, pH, spreadability, viscosity, and percentage were all tested as process factors for diverse herbal microsponge gel compositions. Table 5 shows the drug content findings.

Irritancy test

During irritancy tests, the formulation exhibited no redness, oedema, inflammation, or irritation. These products are safe to use on the skin.

In vitro Drug Release

In vitro Drug Release (Aloin at the λ_{max} of 297 nm)

Table 6 shows the percentage drug release *in vitro* (aloin), and Figure 7 shows the percentage drug release *in vitro* (aloin).

In vitro Drug Release: (Proanthocyanidin at the λ_{max} of 281 nm)

Table 7 shows the percentage drug release *in vitro* (proanthocyanidin), and Figure 8 shows the percentage drug release *in vitro* (proanthocyanidin).

Table 6: *In vitro* drug (aloin) release.

Time (hours)	Gel 1	Gel 2	Gel 3
0	00.00	00.00	00.00
1	12.98±1.27	14.76±1.65	17.76±1.63
2	21.49±1.72	19.43±2.12	22.64±1.83
3	24.12±2.02	28.86±1.83	36.98±2.62
4	30.98±1.65	40.87±2.74	51.99±1,91
5	39.94±1.824	49.86±1.43	59.87±1.02
6	44.98±1.92	56.09±1.72	67.93±1.98
7	50.86±1.65	69.98±1.78	80.71±2.03
8	68.98±1.11	74.98±1.48	89.09±2.47

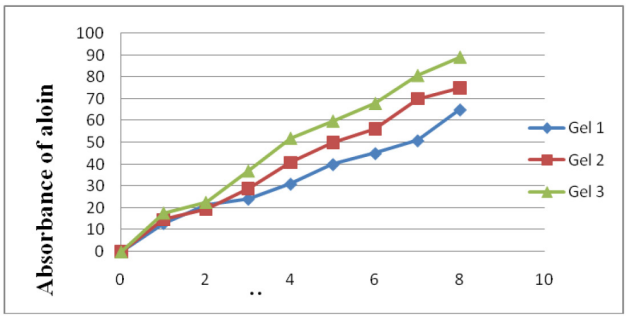

Figure 7: *In vitro* drug (aloin) release.

Table 7: *In vitro* drug (proanthocyanidin) release.

Time (hours)	Gel 1	Gel 2	Gel 3
0	00.00	00.00	00.00
1	13.51±1.42	15.87±1.83	16.98±1.92
2	19.87±1.72	21.98±1.82	23.97±1.73
3	25.54±1.87	29.59±1.91	33.87±1.09
4	31.09±1.65	36.97±1.07	49.00±1.53
5	42.02±2.05	49.98±1.92	60.87±1.38
6	54.11±2.04	60.09±2.82	69.98±1.72
7	61.51±1.62	67.59±2.10	76.45±1.95
8	69.29±1.53	74.76±1.92	82.73±1.73

In vitro Drug Release (Quercetin at the λ_{max} of 258 nm)

Table 8 shows the percentage drug release *in vitro* (quercetin), and Figure 9 shows the percentage drug release.

In vitro UV Study (Determination of SPF factor)

Sample Preparation

Gel-3 was weighed at 1g and transferred to a volumetric flask, volume makeup with ethanol, and the solution sonicated for up to 5 min to thoroughly dissolve, with a final volume of 100 ml ethanol. This stock solution was then used to make the 0.1 percent solution. In the range

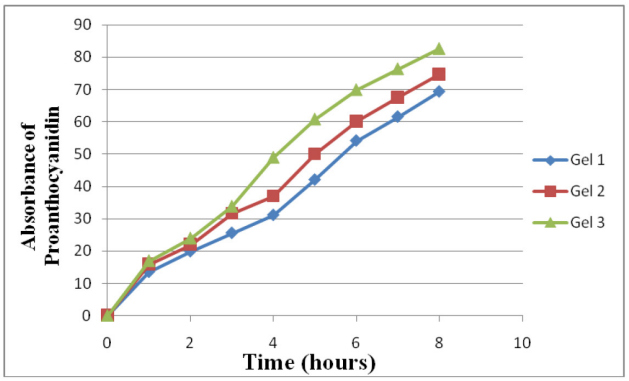

Figure 8: *In vitro* drug (proanthocyanidin) release.

Table 8: *In vitro* drug (quercetin) release.

Time (hours)	Gel 1	Gel 2	Gel 3
0	00.00	00.00	00.00
1	12.76±1.54	13.54±1.45	15.64±2.92
2	19.87±1.56	19.23±2.12	21.76±3.12
3	24.41±0.91	29.99±1.41	34.65±3.24
4	31.51±1.03	40.81±1.29	48.09±1.65
5	42.89±1.53	50.87±1.62	58.04±2.99
6	54.61±1.48	61.91±1.81	69.98±4.61
7	62.72±2.41	74.59±2.91	79.12±2.53
8	74.50±2.16	85.60±3.62	89.72±2.93

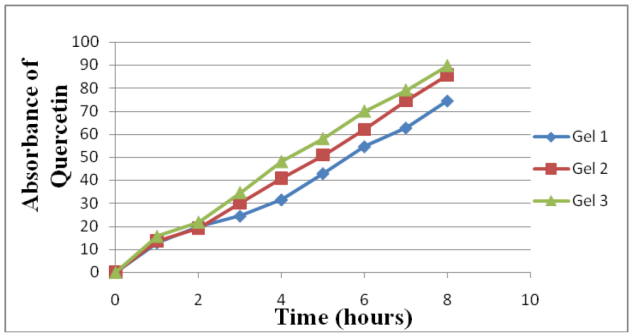

Figure 9: *In vitro* drug (quercetin) release.

290-320, absorption was measured every 5 nm, with three scans at each wavelength, followed by the Mansur equation and SPF calculation.

$$\text{SPF (spectrometry)} = CF \; x \sum_{290}^{320} EE(\lambda) \; x \; I(\lambda) \; x \; abs(\lambda)$$

Absorption was measured at every 5 nm intervals in the range 290-320 nm and performed at each wavelength, followed by the Mansur equation and SPF values were found to be HMG1 - 27.42, HMG2 - 29.71, HMG3 - 36.25 respectively. HMG3 gel was high production of SPF value.

Table 9: Determination of SPF Factor on HMG 1.

Sl.No	Wavelength (λ nm)	Absorbance	EE x I (Constant)	Abs x (EE x I)
1	290	3.156	0.0150	0.0473
2	295	2.941	0.0817	0.2402
3	300	2.841	0.2874	0.8165
4	305	2.741	0.3278	0.8984
5	310	2.593	0.1864	0.4833
6	315	2.532	0.0839	0.2124
7	320	2.473	0.0180	0.0445
		Total:		**2.7426**

Table 10: Determination of SPF Factor on HMG 2.

Sl.No	Wavelength (λ nm)	Absorbance	EE x I (Constant)	Abs x (EE x I)
1	290	3.465	0.0150	0.0519
2	295	3.302	0.0817	0.2697
3	300	3.185	0.2874	0.9153
4	305	2.902	0.3278	0.9512
5	310	2.743	0.1864	0.5112
6	315	2.692	0.0839	0.2258
7	320	2.601	0.0180	0.0468
		Total:		**2.9719**

Table 11: Determination of SPF Factor on HMG 3.

Sl.No	Wavelength (λ nm)	Absorbance	EE x I (Constant)	Abs x (EE x I)
1	290	3.812	0.0150	0.0571
2	295	3.753	0.0817	0.3066
3	300	3.732	0.2874	1.0725
4	305	3.621	0.3278	1.1869
5	310	3.494	0.1864	0.6512
6	315	3.481	0.0839	0.2920
7	320	3.299	0.0180	0.0593
		Total:		**3.6256**

Table 12: SPF Value of Herbal Microsponge Gel.

Formulation	Gel	SPF Value (Spectrophotometrically)	Effect
Polyherbal microsponge gel formulation	HMG1	27.42± 2.13	Medium protection
	HMG2	29.71± 2.02	Medium production
	HMG3	36.25± 2.93	High production

Stability Study

In the stability studies, HMG 3 formulation found that there are no changes in appearance, spreadability, pH, *in vitro* release and SPF value after storing in ICH conditions for about 3 months.

Table 13: Stability Parameter of Optimized Formulation.

Parameters		Initial	After one month (40°C/75% RH)	After two month (40°C/75% RH)	After three month (40°C/75% RH)
Appearance		Yellow colour	Yellow colour	Yellow colour	Yellow colour
Spreadability(g.cm/sec)		5.92±0.85	5.89±0.98	5.69±1.09	5.59±0.76
pH		5.8±0.16	5.8±0.14	5.9±0.65	5.9±0.54
In vitro release	Aloin	89.09±2.47	88.97±2.32	88.12±2.91	87.34±3.91
	proanthocyanidin	82.73±1.73	82.32±2.71	81.74±2.47	81.01±2.91
	Quercetin	89.72±2.93	88.03±2.91	87.38±3.12	87.16±2.47
SPF value		36.25±2.87	36.15±2.54	36.02±1.98	35.75±1.89

DISCUSSION[26,27]

Evaluation of Polyherbal Microsponge

Different process parameters were evaluated for different formulations like percentage yield, percentage drug loading, and particle sizes of formulations. Visual inspection of all batches done using an optical microscope for particle size analysis. Particle size ranges from 60.43 ±2.54 to 72.63 ±6.87 and increased particle size with an increase in extracts: polymer ratio. Microsponges were prepared and their Percentage yield was calculated. They were found to be in the range of 74.98% to 87.60%. It shows increasing the drug:polymer ratio increased the percentage yield. Percentage Loading efficiency ranged from 56.74 to 63.01%. The highest loading efficiency was found for the formulations HM5 and HM6. This shows that increasing the drug:polymer ratio increased loading efficiency.

Determination of Concentration of Aloin, Proanthocyanidin and Quercetin in Formulations

Determination of Absorbance of Water Phase Extracts of aloin, proanthocyanidin and quercetin in the formulations by using UV- spectrophotometric analysis. The absorbance of the drug at 298 nm showed good linearity.

Fourier Transform Infrared Spectroscopy

Polyherbal extracts were tested for compatibility with the other excipients in preformulation tests. Individual IR spectra of polyherbal extracts and another excipient, as well as combined spectra of polyherbal extracts and polymer The interaction of the polyherbal with the excipients in the formulation was investigated.

Scanning Electron Microscope

Scanning electron microscopy was used to examine the internal and exterior morphology and surface topography of the improved HM 5 formulation (SEM). Surface morphology of Polyherbal microsponges as seen in SEM. these images of microsponges are primarily spherical and include small sponges.

Evaluation of Polyherbal Microsponge Gel

Visual inspection, pH, spreadability, viscosity, and percentage were all tested as process factors for diverse herbal microsponge gel compositions. The pH values of the gel were found in the range of 4.5±0.05, 4.6±0.24, 4.8±0.20, which was expected since the carbopol was formulated with pH ranges of 5-5.5 because these values were sufficient to obtain a good viscosity. The spreadability of the HMG3 was considered high by having a low spread of time. The gel spreading helps in the uniform application of the gel to the skin HMG3 has good spreadability and satisfies the ideal quality in the sunscreen application. The viscosity of prepared microsponge gels HMG 1 to HMG 3 ranges from 1935±1.98 to 2309±2.54. from these, the formulation HMG 3 has high viscosity 2309±2.54 cps. The percentage of drug content for the formulation HMG 1 to HMG 3 ranges from 85.9±1.13 to 89.6±1.03. from the result, the HMG 3 formulation has 89.6±1.03 which shows a high percentage of drug content compared with other formulations.

Irritancy Test

From the prepared gel formulation HMG1 to HMG3 were obtained skin irritancy test probably would not produce any skin irritation.

In vitro Drug Release (Aloin at the λ_{max} of 297 nm, Proanthocyanidin at the λ_{max} of 281 nm and Quercetin at the λ_{max} of 258 nm)

In vitro drug release from the gel formulations (HMG1, HMG2, HMG3) in the absorbance ranges for aloin at 68.98±1.11, 74.98±1.48, 89.09±2.47 respectively, proanthocyanidin at 69.29±1.53, 74.76±1.92, 82.73±1.73 respectively and quercetin at 74.50±2.16, 85.60±3.62, 89.72±2.93 respectively. The three formulations of HMG 3 show a satisfactory effect and a maximum drug release was seen. So this formulation is selected for further SPF determination and stability studies.

In vitro UV Study (Determination of SPF factor)

Absorption was measured at every 5 nm intervals in the range 290-320 nm and 3 scans were performed at each wavelength, followed by the Mansur equation and SPF values were found to be HMG1 - 27.42, HMG2 - 29.71, HMG3 - 36.25 respectively. HMG3 gel was high production of SPF value.

Stability Study

The stability studies conducted for optimized formulation HMG 3 found that there are no changes in appearance, spreadability, pH, *in vitro* release and SPF value after storing in ICH conditions for about 3 months.

SUMMARY AND CONCLUSION

The microsponge was generated using polyherbal extract utilising a quasi-emulsion solvent diffusion technique, with eudragit RS-100, ethylcellulose, and PVA as the polymers. The drug-polymer interaction and solvent diffusion rate were investigated on the basis of solubility. The solvents utilised to make microsponges are dichloromethane and ethanol. Drugs in dichloromethane, ethanol, and PVA solution in water are utilised as internal and exterior phases in various formulations. Then, all microsponge formulations were evaluated for drug content, and it was discovered that the formulation with the lowest SD value had consistent drug content and can be reproduced in all preparations. The loading efficiency of each microsponge preparation was found to be consistent. Also, utilising the Franz diffusion cell and standards such as aloin, proanthocyanidin, and Quercetin, a comparative study was conducted for various concentrations of microsponge-loaded polyherbal extract gel and their release profiles were compared. According to the findings, the microsponge GEL3 has a superior medication release profile as well as a higher SPF rating for sunscreen application.

ACKNOWLEDGEMENT

The authors are thankful to Dr B.Jaykar, Professor and Registrar, Vinayaka Mission's Research Foundation (Deemed to be a University) and Vinayaka Mission's College of Pharmacy, Salem, Tamil Nadu for extending their support and facilities for this research.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

ABBREVIATIONS

ATR-FTIR: Attenated Total Reflectance- Fourier Transform Infrared; **HMG1:** Herbal Microsponge gel 1; **HM:** Herbal Microsponges; **PVA:** Poly Vinyl Alcohol; **UVA:** Ultra Violet A; **UVB:** Ultra Violet B.

Disclaimer

In our field of study and country, the goods employed in this study are commonly and majorly used. Because we do not intend to use the items as a tool for litigation, but rather to enhance knowledge, there is no conflict of interest between the authors and the manufacturers of the products. Furthermore, the research was funded solely by the writers' own efforts, rather than by the production company.

Ethical Approval

Written ethical approval has been gathered and retained by the author in accordance with international or university standards.

REFERENCES

1. Yuan H, Ma Q, Ye L, Piao G. The traditional medicine and modern medicine from natural products. Molecules. 2016;21(5). doi: 10.3390/molecules21050559, PMID 27136524.
2. Li FS, Weng JK. Demystifying traditional herbal medicine with modern approach. Nat Plants. 2017;3:17109. doi: 10.1038/nplants.2017.109, PMID 28758992.
3. Singhvi G, Manchanda P, Hans N, Dubey SK, Gupta G. Microsponge: An emerging drug delivery strategy. Drug Dev Res. 2019;80(2):200-8. doi: 10.1002/ddr.21492, PMID 30456763.
4. Gangadharappa HV, Gupta NV, Prasad M SC, Shivakumar HG. Current trends in microsponge drug delivery system. Curr Drug Deliv. 2013;10(4):453-65. doi: 10.2174/1567201811310040010, PMID 22974222.
5. Bens G. Sunscreens. Sunlight, Vitamin D and skin cancer. 2014:429-63.
6. Saewan N, Jimtaisong A. Natural products as photoprotection. J Cosmet Dermatol. 2015;14(1):47-63. doi: 10.1111/jocd.12123, PMID 25582033.
7. Rodrigues LR, Jose J. Exploring the photo protective potential of solid lipid nanoparticle-based sunscreen cream containing *Aloe vera*. Environ Sci Pollut Res Int. 2020;27(17):20876-88. doi: 10.1007/s11356-020-08543-4, PMID 32249384.
8. Martincigh BS, Ollengo MA. The photostabilizing effect of grape seed extract on three common sunscreen absorbers. Photochem Photobiol. 2016 Nov;92(6):870-84. doi: 10.1111/php.12652, PMID 27759892.
9. George D, Maheswari PU, Begum KMMS. Synergic formulation of onion peel quercetin loaded chitosan-cellulose hydrogel with green zinc oxide nanoparticles towards controlled release, biocompatibility, antimicrobial and anticancer activity. Int J Biol Macromol. 2019;132:784-94. doi: 10.1016/j.ijbiomac.2019.04.008, PMID 30951778.
10. Liu C, Cui Y, Pi F, Cheng Y, Guo Y, Qian H. Extraction, purification, structural characteristics, biological activities and pharmacological applications of acemannan, a polysaccharide from *Aloe vera*: A review. Molecules. 2019;24(8):1554. doi: 10.3390/molecules24081554, PMID 31010204.
11. de Souza RC, Machado BAS, Barreto GA, Leal IL, Anjos JPD, Umsza-Guez MA. Effect of experimental parameters on the extraction of grape seed oil obtained by low pressure and supercritical fluid extraction. Molecules. 2020;25(7):1634. doi: 10.3390/molecules25071634, PMID 32252316.
12. Dutta D, Goyal N, Kumar Sharma D. Formulation and development of herbal microsponge sunscreen gel. J Cosmet Dermatol. 2022;21(4):1675-87. doi: 10.1111/jocd.14274, PMID 34087952.

13. Bhatia M, Saini M. Formulation and evaluation of curcumin microsponges for oral and topical drug delivery. Prog Biomater. 2018;7(3):239-48. doi: 10.1007/s40204-018-0099-9, PMID 30242738.
14. Osmani RA, Aloorkar NH, Ingale DJ, Kulkarni PK, Hani U, Bhosale RR, *et al*. Microsponges based novel drug delivery system for augmented arthritis therapy. Saudi Pharm J. 2015;23(5):562-72. doi: 10.1016/j.jsps.2015.02.020, PMID 26594124.
15. Tripathi PK, Gorain B, Choudhury H, Srivastava A, Kesharwani P. Dendrimer entrapped microsponge gel of dithranol for effective topical treatment. Heliyon. 2019;5(3):e01343. doi: 10.1016/j.heliyon.2019.e01343, PMID 30957038.
16. Song Y, Cong Y, Wang B, Zhang N. Applications of Fourier transform infrared spectroscopy to pharmaceutical preparations. Expert Opin Drug Deliv. 2020;17(4):551-71. doi: 10.1080/17425247.2020.1737671, PMID 32116058.
17. Khattab A, Nattouf A. Optimization of entrapment efficiency and release of clindamycin in microsponge based gel. Sci Rep. 2021;11(1):23345. doi: 10.1038/s41598-021-02826-7, PMID 34857863.
18. Pandit AP, Patel SA, Bhanushali VP, Kulkarni VS, Kakad VD. Nebivolol-loaded microsponge gel for healing of diabetic wound. AAPS Pharm Sci Tech. 201;18(3):846-54. doi: 10.1208/s12249-016-0574-3, PMID 27357423.
19. Pawar AP, Gholap AP, Kuchekar AB, Bothiraja C, Mali AJ. Formulation and evaluation of optimized oxybenzone microsponge gel for topical delivery. J Drug Deliv. 2015;2015:261068. doi: 10.1155/2015/261068, PMID 25789176.
20. Shah VP, Elkins J, Hanus J, Noorizadeh C, Skelly JP. *In vitro* release of hydrocortisone from topical preparations and automated procedure. Pharm Res. 1991;8(1):55-9. doi: 10.1023/a:1015826205930, PMID 2014209.
21. Montenegro L, Puglisi G. Evaluation of sunscreen safety by *in vitro* skin permeation studies: Effects of vehicle composition. Pharmazie. 2013;68(1):34-40. PMID 23444778.
22. Thiyagarasaiyar K, Goh BH, Jeon YJ, Yow YY. Algae metabolites in cosmeceutical: An overview of current applications and challenges. Mar Drugs. 2020;18(6):323. doi: 10.3390/md18060323, PMID 32575468.
23. He H, Li A, Li S, Tang J, Li L, Xiong L. Natural components in sunscreens: Topical formulations with sun protection factor (SPF). Biomedicine and pharmacotherapy= Biomedecine and pharmacotherapie. 2020;134:111161-.
24. Ricci T, Marra A, Rauen K, Caswell M. Bias in sunscreen SPF testing: A review of published data. J Cosmet Sci. 2020;71(5):351-60. PMID 33022201.
25. Das S, Wong ABH. Stabilization of ferulic acid in topical gel formulation via nanoencapsulation and pH optimization. Sci Rep. 2020;10(1):12288. doi: 10.1038/s41598-020-68732-6, PMID 32703966.
26. Bhuptani RS, Patravale VB. Starch microsponges for enhanced retention and efficacy of topical sunscreen. Mater Sci Eng C Mater Biol Appl. 2019;104:109882. doi: 10.1016/j.msec.2019.109882, PMID 31500041.
27. Dutta D, Goyal N, Sharma DK. Preclinical evaluation from *in-silico* to *in-vivo* study of polyherbal microsponge sunscreen gel. J Cosmet Dermatol. 2021. doi: 10.1111/jocd.14674, PMID 34962064.

Cite this article: Muruganantham V, Venkateswarlu BS, Chinraj V. Design and Evaluation of Isolated Herbal Extracts of Aloin, Proanthocyanidin, Quercetin Loaded Microsponge as Sunscreen Gel. Asian J Biol Life Sci. 2022;11(2):356-66.