Journal of Applied Pharmacognosy and Phytochemistry
Volume 4, Issue 1, Year of Publication 2024, Page 32 – 37

Research Article

JOURNAL OF APPLIED PHARMACOGNOSY AND PHYTOCHEMISTRY | JOAPP
www.joapponline.com

DEVELOPMENT AND CHARACTERIZATION OF NOVEL CARBOPOL BASED HYDROGEL FORMULATION CONTAINING EXTRACT OF ECLIPTA PROSTRATA

Monika Bhairam*, Moulik Kumar, Gunjan Kalyani

Article Information
Received: 14th December 2023
Revised: 11th February 2024
Accepted: 9th March 2024
Published: 31st March 2024

Keywords
Hydrogels, Eclipta prostrata, Carbopol-934, Antibacterial, Anticancer

ABSTRACT

The objective of the current work was to create carbomer-based hydrogel formulation that included ethanolic extracts of *Eclipta prostrata*. Hydrogel play a significant role in encapsulation of various bioactive compounds extract. *Eclipta prostrata* is a traditional medicinal herb that has been used for a long time in India to treat ailments like hemorrhagic diseases, skin disorders, respiratory problems, coronary heart disease, hair loss, vitiligo, snake bites, and ailments brought on by liver and kidney deficiencies. However, a number of problems, such as a lack of targeting ability and low bioavailability, have restricted the therapeutic efficacy of herbal extracts and components. Networks of hydrophilic polymers called hydrogels may absorb a lot of liquid. They may allow for continuous medication release and are biocompatible. Because of this, investigations on hydrogels in pharmaceutical formulation have become very popular. The prepared herbal hydrogel formulation contains Carbopol-934, Acacia, Glycerin Sodium benzoate & HPMC. This study aims to investigate the effects of different formulation variables on the release of *Eclipta prostrata* extract from hydrogel with a polymeric composition. The developed herbal hydrogel formulation underwent preliminary testing such as assessments of viscosity, pH, spreadability, rheological investigations, skin irritation tests, homogenous drug content, and accelerated stability stdies. It is possible to draw the conclusion on the basis of evaluation investigation that medicinal herbal hydrogel of *Eclipta prostrata* may be utilised for the treatment of wounds, antibacterial and antifungal action, anticancer, and anti-inflammatory activity.

INTRODUCTION

False daisy is the popular name for *Eclipta prostrata*, which is also known as Bhringraj in Ayurveda and Ecliptaeherba in Chinese. Farmers view it as a typical weed in folklore [1]. However, it has long been used for the treatment of wounds, cuts, high blood pressure, coronary heart disease, vitiligo, diabetes, gastrointestinal disorders, and respiratory illnesses. It also plays a significant part in ethnomedicine. Traditional medicine treats linked illnesses such loose teeth, greying hair, dizziness, tinnitus, and bleeding with the dried aerial parts of the plant, which are regarded as nutritious herbs and used

*Columbia Institute of Pharmacy, Near Vidhan Sabha, Tekari, Raipur, Chhattisgarh, India 493111

***For Correspondence:** **monikab430@gmail.com**

medicinally. Due to its vast range of applications, a scientific community has been paid attention on investigation on its bioactive constituents and a pharmacological action has been done. Even though there have been numerous reviews of *E. prostrata*, they didn't cover all of this plant's characteristics [2].

Many types of chemical compounds have been isolated and identified from *E. prostrata*, including alkenynes, alkaloids, cardiac glycosides, flavonoids, coumestans, lipids, polyacetylene, steroids, saponins, steroidal alkaloids, phytosterol, triterpenes. of these, triterpenes, flavonoids, thiopenes, coumestans, steroids are regarded to be primary constituents and likely future directions that will provide a representative overview of this medicinal plant. For the role of *E. prostrata* in ethnomedicine, pharmacological effects of various extracts and compounds based on traditional applications have been intensively studied in recent decades [3]. To be relevant for its traditional usage, *E. prostrata* exhibits hepatoprotective, anti-osteoporotic, anti-inflammatory, analgesic, hypoglycemic, hypolipidemic, cytotoxic, anti-microbial, neuroprotective, hair growth-promoting, and nephroprotective properties [1].

Hydrogels have a hydrophilic, three-dimensional, polymeric network that allows them to absorb large volumes of water and biological fluids. In comparison to other synthetic bio-material formulations, they closely resemble natural living tissues due to their high water content, soft softness, and porosity. Chemically stable hydro-gels can also dissolve and disintegrate. This hydrogel's ability to adapt or remain stable depends on how different functional groups display their structural elements. When charged hydro-gels are exposed to electric fields, they can alter in form and consistency in addition to swelling when the pH is exceptional [4]. The chemical formulation of hydrogels typically involves one of two approaches: either three-dimensional polymerization, in which hydrophilic monomers are polymerized in the presence of a multifunctional cross-linking agent, or direct coupling of hydrogel forms to water-soluble polymers. Natural polymers like collagen and gelatin, as well as polysaccharides like starch and alginate, are used to create hydrogel [5]. Chemical polymerization techniques are typically used to create hydrogels from synthetic polymers.

The objective of the current study was to create a stable hydrogel using carbomer as a gel-forming agent. It also investigated into the feasibility of adding very potent ethanolic herbal extracts into the polymeric matrix while maintaining the product's medicinal properties [6].

MATERIAL AND METHODS

Dried powder of leaves of Eclipta prostrate (herbs were collected between Dec- Jan from village– mandhar), Polyethylene glycol 400 (Sigma-Aldrich) as plasticizer, and Carbapol 934 ((Sigma-Aldrich) was used as a gelling agent. All reagents were of analytical grade.

Herbal Extracts Preparation

The bioactive compound-loaded extracts were obtained utilising a solvent extraction technique with ethanol as a reagent. For this, dried leaf powder was mixed with 70% v/v ethanol and allowed to macerate for 10 days at room temperature. The proportion of solvent to herbal powder was 1/10 (g/mL). After utilising a rotary evaporator to concentrate the samples, they were cooled down, centrifuged for 15 minutes at 4500 rpm, and the supernatant was collected [7].

Preparation of the Hydrogel

To begin with, some pilot studies (data not given) were conducted to determine the best hydrogel composition for incorporating the ethanolic plant extracts. Further, the formulations were prepared by dissolving the Carbapol and PEG 400 into water, adding the herbal extracts. Glycerine and sodium benzoate, the last two ingredients, were then added. The Mixtures went through a sonication process [8]. The prepared hydrogel was put in the appropriate container. Composition of formulation is mentioned in table 1 & process is shown in fig 1

Table 1: Composition of Hydrogel formulation

S No	Ingredients	Quantity (mg)
1.	Carbapol	500
2.	PEG 400	250
3.	Acacia	250
4.	Glycerine	15ml
5.	sodium benzoate	100
6.	Herbal extract of *Eclipta prostrata*	100 mg
7.	Double distilled water	Up to 15ml

Phytochemical screening

Phytochemical characterisation was performed in accordance with practice guidelines.

Test for reducing agent

The extract (1ml) was added to a test tube filled with boiling Fehling's solution (A and B) (5ml). Chemical reactions were observed in the solution.

Test for anthraquinones

Take 5 ml of extract with 10 ml of sulphuric acid, boil it, and filter it while still hot. Then, 5 ml of chloroform was added to the filtrate and shaken. After the chloroform layer was pipetted into another test tube, 1 ml of diluted ammonia was added. Chemical reactions were observed in the solution.

Test for terpenoids

Take 5 ml of extract with 2 ml of ethanolic extract. Progressively add 3 ml of concentrated sulphuric acid, and observe the colour change.

Test for flavanoids

We took a 5ml extract with 5ml diluted ammonia. Then, we gradually added 1ml of concentrated sulphuric acid. The yellow color determines the presence of flavonoids.

Test for saponins

Take 2 ml extract with 5 ml of distilled water in a test tube. Shake vigorously. After that, observe stable, persistent froth on the top of the test tube. Mix the frothing with 3 drops of olive oil and shake vigorously, after which observe the formation of an emulsion.

Test for tannins

2 ml of the extract is first added to a test tube with 10 ml of purified water, boiled, and then filtered. Following the addition of 0.1% of Fecl3, the presence of tannin is indicated by a colour change to brownish green and blue-black.

Analyze the alkaloids

First, dilute 2ml of extract alcohol with 5ml of alcohol. Filter the extract afterward, then add 2 ml of diluted ammonia with 5 ml of the filtrate. The alkaloid base was extracted by adding 5 ml of $CHCl_3$. The layer of chloroform was extracted with 10 ml of acetic acid. First, there was a section for Mayer's reagent, then Draggendorff's reagent. Alkaloids were considered present if a gel with Mayer's reagent and a reddish brown precipitate with Draggendorff's reagent formed [9].

Physicochemical characterization

Loss of weight on drying

The percentage LOD was determined using the gravimetrical approach, in which 5g of drug was precisely weighed using the standard method. The test substance was dried in an oven between 100°C and 110°C until two subsequent weighs did not differ by more than 5 mg.

Ash value

Ash is what remains after incineration. 5g of the crude drug was precisely weighed using a treated silica cross. The crude drug was incinerated gradually and thoroughly until it was cool and carbon-free. The amount of total ash was measured by weight and calculated as a percentage of total ash.

Extractive value

5g of crude drug were precisely weighed and macerated with 100ml of ethyl alcohol. Then, for six hours, the extract was shaken. The solution was filtered and gathered quickly. A 25 ml of alcoholic extract evaporated to dryness in a shallow dish with a flat bottom. The extract was weight-consistently dried. Finally, the extraction value of alcohol by source for air-dried medicines [10].

Chromatography

It is a separation technique for non-volatile mixtures. It is operated on a plastic or glass sheet, which is coated with a thin layer of adsorbent material used in silica gel. After the sample has been applied to the plate, a solvent or solvent mixture (known as the mobile phase) is drawn up the plate via capillary action because different analytes rise, and the chromatography plate separation is achieved at different rates [11].

Evaluation of herbal *Eclipta Prostrata* Hydrogel

Appearance

The prepared herbal extract hydrogel had Carbopol and PEG400 and was milky white and yellowish transparent, respectively.

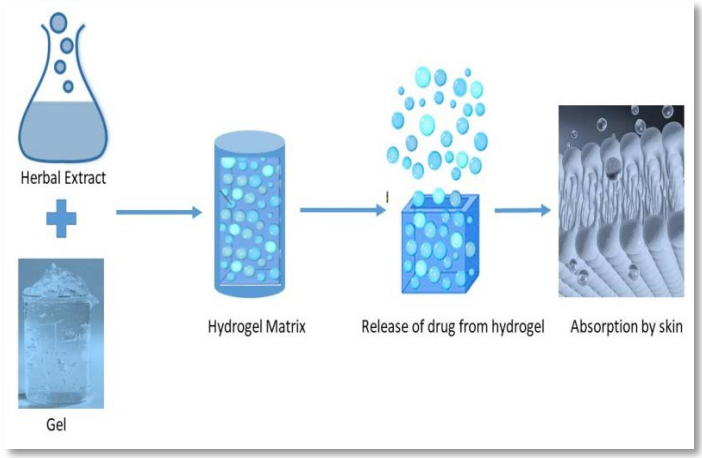

Fig 1: Whole process of preparation and application

Determination of pH

A pH meter was used to determine the formulation's pH. This technique cleaned the electrode with double-distilled water, dried it using tissue paper, and dipped it in a 10ml hydrogel formulation. At ambient conditions, the hydrogel formulations' average pH was 6.4.

Spreadability test

Hydro-gel is applied on a flat surface to test its spreadability, and its grittiness is then assessed.

Viscosity assessment

Viscosity is an important feature in determining the resistance to flow of a hydrogel formulation. It also determines the spreadability of gel on the skin. This study used a Brookfield viscometer (spindle 2) to determine the viscosity.

Drug content

20 mL of phosphate buffer pH 7.4 and 1 g of the formulation were combined appropriately, then let to stand for 30 minutes. The final combination was put through a membrane filter with a pore size of 0.45 m. At 277 nm, the sample underwent spectrophotometric analysis.

Percentage Moisture Content

The standard method assessed the manufactured formulation's moisture percentage (%) loss. 50gm of calcium chloride anhydrous were contained in a desiccator and two gm of precisely weighed formulation. The mixture was weighed three days later. The following formula was used to calculate the moisture percentage (%) loss:

$$Moisture\ (\%)loss = \frac{Initial\ weight - Final\ weight}{Final\ weight} \times 100$$

Transparency

The 10ml (BorosilR) test tube that contained the 5ml gel formulation was then visually examined for transparency.

Smoothness

The gel formulation was determined by rubbing it between the fingers to determine whether it was smooth, clumped, homogeneous, or rough.

Relative density

The formulation's relative density (or weight per ml) was calculated by weighing 10ml of the formulation and 10ml of distilled water using an RD bottle.

Accelerated stability studies

According to ICH guiding principles, the accelerated stability test was conducted on the optimized prepared formulation. The prepared, tested formulation was placed in aluminum tubes and put through stability testing according to ICH standards for three months at a temperature of 40 ± 2ºC and relative humidity of 75 5%. Over three months, samples were obtained at regular 1-month intervals to check for changes in spreadability, pH, viscosity, and medication content [12-15].

RESULT & DISCUSSION

Phytochemical Screening

All tests were positive for the phytochemical screening of the ethanolic extract of Eclipta prostrata except for anthrax quinones. Table 2 depicts all the results.

Table 2: Phytochemical screening of extract of *E. prostrata*

S No	Phytochemical screening test	Result
1.	Test for reducing agent (Fehling test)	Present
2.	Test for anthraquinones	Absent
3.	Test for terpenoids	Present
4.	Test for flavanoids	Present
5.	Test for saponins	Present
6.	Test for tannins	Present
7.	Analyze the alkaloids	Present

Table 3: Physicochemical evaluation of *Eclipta prostrata*

S No	Particular	*E. prostrata* (%)
1.	Loss of weight on drying	1.01
2.	Total ash	25
3.	Extractive value- Alcohol	10
4.	Solvent system -Chloroform: Alcohol (8:2)	Rf- 0.54

Table 4: Hydrogel characterization results

S No	Characterization	Results
1.	Appearance	Green-yellowish transparent
2.	pH	6.4± 023
3.	Spreadability	slightly Gritly in nature
4.	Viscosity	1014μ
5.	Drug content	99%
6.	Moisture Content	94%
7.	Transparency	Translucent
8.	Smoothness	Smooth
9.	Relative density	1.23 g/cm^3

Physicochemical characterization

The percentage (%) of weight loss owing to drying, total extractive, and ash value were investigated using a traditional method. All the results are depicted in Table 3. The leaf powder's extractive principle percentage in alcohol solvent systems was also determined.

Thin Layer Chromatography profile (TLC)

This study was carried out on the ethanolic extracts of Eclipta prostrata leaf. The silica gel G used for TLC was heated to 100°C in a hot air oven for one hour. The thin layer plate was marked on the line for the sample application at 2 cm from the base. The sample was spotted on the line using a capillary tube and left to dry. The developing chamber containing the mobile phase was filled with the plate. The hit-and-trial method was used to choose the mobile phases. The spots were seen, and the Rf value was computed using various solvent system ratios. After viewing the spots, a solvent system was chosen. The solvent system was used to run TLC plates.

Study on accelerated stability

The tested hydrogel did not show significant modifications. After being exposed to an accelerated humidity and temperature environment for three months, the tested formulation was found to be stable. All the results of the hydrogel characterization and accelerated stability data are mediation in Tables 4 and 5, respectively.

Table 5: Physical parameters after accelerated stability study of formulation

Characteristics	pH	Viscosity (cps)	Drug content (%)
Initial	6.4± 023	1014±0.12	99± 0.23
After1 month	6.4± 023	1014±0.22	99± 0.22
After 2 months	6.4± 021	1014±0.23	99 ± 0.22
After 3 months	6.4± 012	1014±0.25	99± 0.21

CONCLUSION

However, several issues, such as a lack of targeting ability and low bioavailability, have hindered the therapeutic efficacy of herbal extracts and ingredients. Networks of hydrophilic polymers called hydrogels may absorb a lot of liquid. They are highly biocompatible due to various characteristics, including their high water content and low surface tension. Some hydrogels have a high drug loading efficiency as well, which has drawn extensive research in pharmaceutical development. In recent years, pharmacologists and other researchers from the medical and paramedical fields have begun to examine ayurvedic medicines. Polymer was used to create the topical hydrogel for *Eclipta prostrata.*

The current hydrogels showed good physico-chemical characteristics when compared to commercial formulations in terms of transparency, smoothness, density, moisture content, and pH. These findings point to the viability of topical hydrogel for effective, safe treatment of skin irritability and skin disorders. Additionally, it may be said that *Eclipta prostrata* herbal hydrogel may be employed for wound healing, anticancer, antibacterial, and antifungal action and many more.

FINANCIAL ASSISTANCE

Nil

CONFLICT OF INTEREST

The authors declare no conflict of interest.

REFERENCES

[1] Lai WF, Rogach AL. Hydrogel-Based Materials for Delivery of Herbal Medicines. *ACS Appl Mater Interfaces* **9**, 11309-20 (2017)

[2] Pandey R, Bhairam M, Shukla SS, Gidwani B. Colloidal and vesicular delivery system for herbal bioactive constituents. *Daru* **29**, 415-38 (2021)

[3] Ali A, Garg P, Goyal R, Kaur G, Li X, Negi P, Valis M, Kuca K, Kulshrestha S. A Novel Herbal Hydrogel Formulation of Moringa oleifera for Wound Healing. *Plants (Basel)* **10**, 25 (2020)

[4] Kurre PK, Ghritlahare SK, Jangdey MS. Formulation and Evaluation of Hydro-gel containing Eclipta alba extract. *Pharmaceutical and Biosciences Journal* **8(5)**, 27-32 (2020).

[5] Gidwani B, Bhattacharya R, Shukla SS, Pandey RK. Indian spices: past, present and future challenges as the engine for bio-enhancement of drugs: impact of COVID-19. *J Sci Food Agric* **102**, 3065-77 (2022)

[6] Jales STL, Barbosa RdM, de Albuquerque AC, Duarte LHV, da Silva GR, Meirelles LMA, da Silva TMS, Alves AF, Viseras C, Raffin FN, et al. Development and Characterization of *Aloe vera* Mucilaginous-Based Hydrogels for Psoriasis Treatment. *Journal of Composites Science* **6(8)**, 231 (2022)

[7] Chan ES, Yim ZH, Phan SH, Mansa RF, Ravindra P. Encapsulation of herbal aqueous extract through absorption with ca-alginate hydrogel beads. *Food and Bioproducts Processing* **88(2-3)**, 195-201 (2010)

[8] Yadav RNS, Agarwala M. Phytochemical analysis of some medicinal plants. *Journal of Phytology* **12**,10-14 (2011)

[9] Gavan A, Colobatiu L, Hanganu D, Bogdan C, Olah NK, Achim M, Mirel S. Development and Evaluation of Hydrogel Wound Dressings Loaded with Herbal Extracts. *Processes* **10(2)**, 242 (2022)

[10] Hoque R. Phytochemical analysis and TLC studies of *Eclipta alba* and *Scoparia dulcis* plant extract-hot extraction. *Int J Drug Dev & Res* **11**, 12-5 (2019).

[11] Bhairam M, Roy A, Bahadur S, Banafar A, Turkane D. Standardization of herbal medicines–an overview. *Journal of Applied Pharmaceutical Research* **1**, 14-21 (2013).

[12] Ahmed EM. Hydrogel: Preparation, characterization, and applications: A review. *Journal of Advanced Research* **6**, 105-21 (2015).

[13] Aswathy SH, Narendrakumar U, Manjubala I. Commercial hydrogels for biomedical applications. *Heliyon* **4**, 1-13 (2020)

[14] Gul K, Gan RY, Sun CX, Jiao G, Wu DT, Li HB, Kenaan A, Corke H, Fang YP. Recent advances in the structure, synthesis, and applications of natural polymeric hydrogels. *Crit Rev Food Sci Nutr* **62**, 3817-32 (2022)

[15] Fathi M, Ahmadi N, Forouhar A, Hamzeh Atani S. Natural Hydrogels, the Interesting Carriers for Herbal Extracts. *Food Reviews International* **27**, 1-25 (2021)