

# Formulation and Evaluation of *Tylophora indica* Extract Loaded Topical Herbal Microgel for Rheumatoid Arthritis

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## ABSTRACT

This paper focuses on the phytoconstituents contained in herbal plant extracts and presents them in the form of a herbal microgel with an aim to target rheumatoid arthritis. The study starts from the collection, followed by authentication of the selected medicinal plant, extraction through solvents of the whole plant, and simultaneous phytochemical analysis performed with morphological characterization of the selected extract. The consequent phase forms a specialized gel formulation employing synthetic polymers. The prepared herbal microgel is put through comprehensive evaluation that would include the study of short-term stability as per ICH guidelines No: Q1A. This research offers a promising approach towards an effective and natural remedy for rheumatoid arthritis.

**Keywords:** Rheumatoid arthritis, herbal, extract, microgel

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## INTRODUCTION

Rheumatoid arthritis has been recognized for over two centuries. It overwhelmingly presents as an inflammatory polyarthritis with a predilection for the hands and feet. However, the systemic nature of RA may produce complications ranging from neuropathy to lung disease, vasculitis, and subcutaneous nodules. Because the effects are widespread, as are the potential problems of treatment, morbidity from rheumatoid arthritis is enormous. RA is characterized by symmetrical, inflammatory joint inflammation, initially in smaller joints and later progressing to larger ones, affecting not only joints but also vital organs like the heart, lungs, and kidneys. This leads to increased fragility of tendons, ligaments, bones, and cartilage, resulting in deformities and bone erosion, causing substantial pain for the patient. The common symptoms are unexplained weight loss, persistent fatigue, recurring fever, rheumatoid nodules under the skin, and stiff joints-mostly in the morning-which normally takes a long time to fade away.<sup>1</sup> The main goals of rheumatoid arthritis treatment are to decrease the inflammation in joints, to soothe the pain, to enhance the movement of the joints, and to stop or delay further destruction and deformation of the joints. Individualized treatment plans encompass a combination of medication, weight-bearing exercises, patient education, and adequate rest. These plans are tailored based on factors like disease progression, affected joints, age, overall health, occupation, compliance, and patient education.

### Side effects from RA

Early onset of cardiac illness. Furthermore, there is a higher chance of developing diabetes and heart disease in patients with RA, among other chronic disorders. Reducing heart disease risk factors is another important aspect of RA treatment, as it keeps people with RA from developing heart disease. For example, doctors may recommend to RA patients that they stop smoking and lose weight. Obesity is an independent factor that can increase the chances of developing cardiovascular risk factors, such as hypertension and increased cholesterol levels, in patients suffering from rheumatoid arthritis. Additionally, chronic disorders like diabetes and heart disease are more likely to develop in obese people. Finally, people with RA who are fat receive poorer benefit from their medical treatment than those with non-obesity.<sup>2</sup> Work may become difficult if you have RA. Compared to people without RA, adults with RA had a lower employment rate. As rheumatoid arthritis progresses, many report a decline in the performance of activities that were previously manageable. Those with physically demanding work constitute the majority of RA sufferers who lose their jobs. People who work at occupations where they can manage the activities and pace of their workday, or who have positions with less physical demands, tend to lose less time to work.

### Diagnose

X-rays, lab testing, physical examinations, and an examination of the RA symptoms are all conducted. Prior to the disease's progression, such as joint degeneration, it is optimal to diagnose RA within six months of the onset

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Table: 1 Formulation table for *Tylophora indica* herbal microgel.

S. No	Ingredient	F1	F2	F3	F4	F5	F6	F7	F8	F9
1	<i>Tylophora indica</i>	2	2	2	2	2	2	2	2	2
2	Carbopol 934	0.5	1	2	---	---	---	---	---	---
3	Sodium alginate	---	---	---	0.5	1	1.5	---	---	---
4	HPMC	---	---	---	---	---	---	0.5	1	1.5
5	Propylene glycol	1	1	1	1	1	1	1	1	1
6	Methyl paraben	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
7	Triethanolamine	Qs	Qs	Qs	Qs	Qs	Qs	Qs	Qs	Qs
8	Distilled water	Qs	Qs	Qs	Qs	Qs	Qs	Qs	Qs	Qs

Table: 2 Phytochemical test for *Tylophora indica* ethanolic extract.

S. No	Name of the Extract	Phytochemical Active Constituents of <i>TI</i>
1	Petroleum ether extract	Flavonoids, Amino acid, Steroids, Terpenoids, Phenolic compounds
2	Chloroform extract	Alkaloids, Carbohydrates, flavonoids, phenolic compounds, Steroids, Terpenoids
3	Acetone extract	Glycosides, Carbohydrates, flavonoids, tannin, phenolic compounds, steroids, terpenoids
4	Ethanol extract	Carbohydrates, flavonoids, Amino acid, phenolic compounds, steroids, terpenoids
5	Aqueous extract	Steroids, terpenoids, phenolic compounds, Amino acid, flavonoids, Carbohydrates

of symptoms, allowing patients to begin treatment. Most of these undesirable outcomes can be reduced if the RA is diagnosed in time and treated with appropriate drugs, especially those which suppress or regulate inflammation.<sup>3</sup>

**Herbal microgel**

Herbal microgels are a specific kind of microgel that contains natural plant-derived substances like herbal

extracts. It is made to deliver these herbal components' advantages in a controlled and effective way. In addition to various pharmaceutical and medical applications, herbal microgels are frequently employed in skincare and cosmetic goods. The ability of herbal microgels to encapsulate and transport bioactive substances from herbs and plants, like antioxidants, vitamins, and phytochemicals, in a stable and sustained way, is one of their key properties. This enables better skin penetration, absorption, and extended release of these healthy compounds.<sup>4</sup> Herbal microgels are frequently employed for moisturising, calming, anti-aging, or the treatment of particular skin diseases. They are well-liked in the natural and organic skincare sector because of their formulation, which makes them lightweight, non-greasy, and simple to apply to the skin. Depending on the targeted therapeutic or cosmetic benefits, an herbal microgel's plant selection and specific formulation may change. Herbal extracts or natural product active properties are formulated as microgels to be used in various cosmetic and skincare formulations, such as creams, lotions, gels, and serums. These herbal ingredients can be contained and shielded by the microgel structure, enabling regulated release and improved penetration into the skin. Due to their capacity to combine the benefits of microgel technology, such as improved product stability and controlled release, with the potential therapeutic properties associated with herbal extracts, such as soothing, moisturising, or antioxidant effects, herbal microgels are popular in the cosmetics and skincare industry. They follow the general trend of personal care products employing natural and plant-based components.<sup>5</sup>

**Synonyms**

*Indian ipecac, Indian ipecacuahna, Dam-ni-vel*

**Plant taxonomy**

Kingdom: Plantae  
 Phylum: Vascular plant  
 Order: Gentianales



Figure: 1 *Tylophora indica* herbal microgel formulation

Table: 3 Percentage yield of *Tylophora indica* extract.

S. No	Parts used	Plant Name	Solvent and % yield of plant extract		
			Pet. Ether	Ethanol	Aqueous
1	Plant entire part	<i>Tylophora indica</i>	14.29%	15.8%	12.38%

Familia: Apocynaceae  
Genus: *Tylophora*  
Species: *Tylophora indica* (Burm.f.) Merr.

Table: 4 Physical description and organoleptic properties of extract.

S.No	Name of Extract	Colour	Odour	Nature
1	<i>Tylophora indica</i>	Dark green to brownish green	Odourless	Solid

Table: 5 Solubility studies of ethanolic extract of *Tylophora indica*.

S. No	Media	Solubility ( $\mu\text{g/ml}$ ) of <i>Tylophora indica</i>
1	Distilled water	Soluble(0.248 $\mu\text{g/ml}$ )
2	Ethanol	Soluble (0.531 $\mu\text{g/ml}$ )
3	Hydrochloric acid (pH 1.2)	Sparingly Soluble (0.0157 $\mu\text{g/ml}$ )

Table: 6 HPLC peak and time for extract.

S.No	Retention peak	Time
1	Peak 1	2.449
2	Peak 2	2.994
3	Peak 3	3.761

## MATERIALS AND METHODS

### Method for extraction

For 72 hours, *Tylophora indica*(250g) was extracted using the solvents Petroleum ether, Ethanol, and Aqueous (60-80°C). After extraction, the defatted extracts were hot-filtered using Whatman filter paper (No. 10) to remove any potential impurities. The vacuum distillation was carried out in order to reduce the volume to one-tenth. Transfer the concentrated extract into a 100 mL beaker and evaporate the residual solvent on a water bath. The extract was dark brownish in colour. After that, the concentrated extract was placed in a desiccator to eliminate excess moisture. The dried extract was then kept in an airtight glass container for future research that may be conducted.

### Identification of Phytochemical Active Constituents

The obtained extracts (Petroleum ether, Ethanol, Aqueous solvents) were test for alkaloids, carbohydrates, flavonoids, glycosides, Amino acid, saponins, tannins, Phenolic compounds, terpenoids, steroids were performed

Table: 8 pH for *Tylophora indica* herbal microgel.

Formulation Code	pH
F1	5.8
F2	5.7
F3	6.0
F4	5.8
F5	5.7
F6	5.9
F7	5.8
F8	6.1
F9	5.9

6.

### Percentage yield

The yield percentage was calculated by comparing the mass of the dried extract with the mass of the dried plant material<sup>7</sup>

The percentage yield was calculated using the formula: (mass of the dried extract divided by the mass of the dried plant sample) multiplied by 100.

### ATR- FTIR Spectra analysis

The wavelength range was from 4000 to 400  $\text{cm}^{-1}$ . An ATR-FTIR spectrophotometer was used to get the IR spectrum after directly placing the sample into the cavity of the sample holder.<sup>8</sup>

### Particle size (SEM Analysis)

The Scanning Electron Microscope is among the powerful analytical techniques using a focused electron beam for scanning on the surface of the sample in order to get high-resolution images providing detailed information related to the morphology, composition, and microstructure of solid inorganic materials. It is usually known as SEM analysis or SEM microscopy. Its applications are vast in various types of materials in regard to microanalysis and failure analysis.<sup>9</sup>

### Zeta potential

A Zeta potential measuring system is made up of six basic components. A laser is used to illuminate the particles in the sample, which is then separated into an incident beam and a reference beam for zeta potential measurements.<sup>10</sup>

### Preformulation studies

#### Physical description and organoleptic characteristics

The organoleptic characteristics of the drug refer to the

Table: 7 Visual inspection for *Tylophora indica* herbal microgel.

S. No	Physical Characters	F1	F2	F3	F4	F5	F6	F7	F8	F9
1	Colour	Light green	Light green	Light green	Light green	Light green	Light green	Light green	Light green	Light green
2	Odour	Pleasant	Pleasant	Pleasant	Pleasant	Pleasant	Pleasant	Pleasant	Pleasant	Pleasant
3	Nature	Opaque	Opaque	opaque	Opaque	Opaque	Opaque	Opaque	Opaque	Opaque

odor, color, and other sensory properties of the drug. The first step in study comprises determination of such organoleptic properties, which helps in preliminary identification of the natural product and aids in assessing

the possible acceptability of the raw material's odor, appearance, and nature by the patients, an important consideration for its inclusion in the final dosage form.<sup>11</sup>  
**Solubility study**

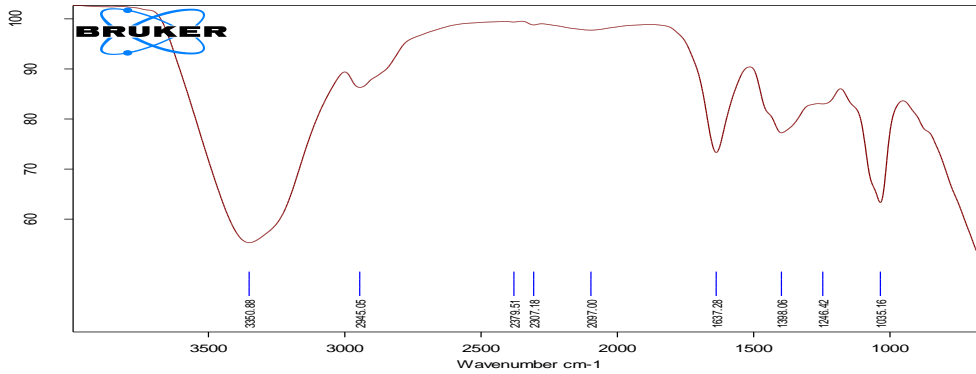


Figure: 2 FTIR spectrum of *Tylophora indica* plant extract

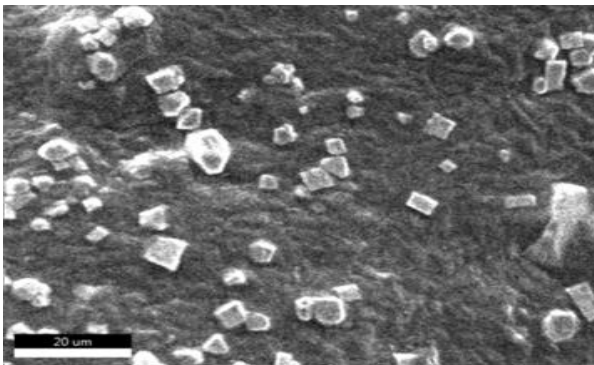


Figure: 3 SEM viewing for *Tylophora indica* plant extract

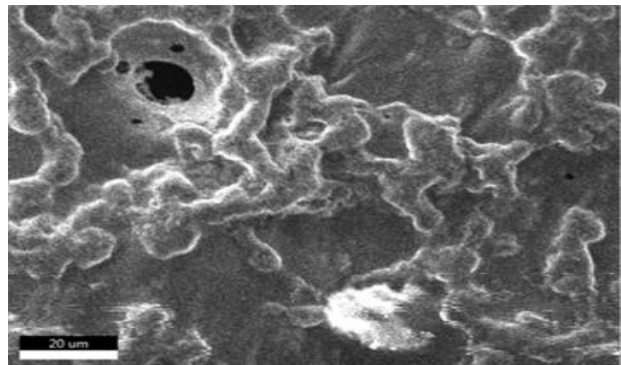


Figure: 4 SEM viewing for *Tylophora indica* herbal microgel

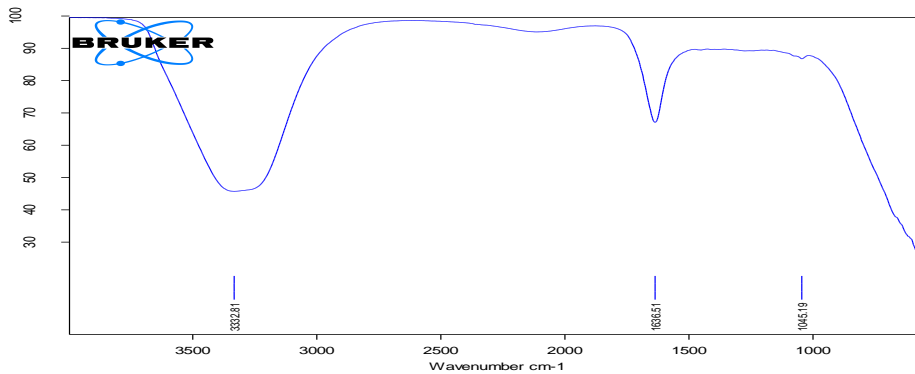


Figure: 5 ATR- FTIR Spectrum of *Tylophora indica* herbal microgel.

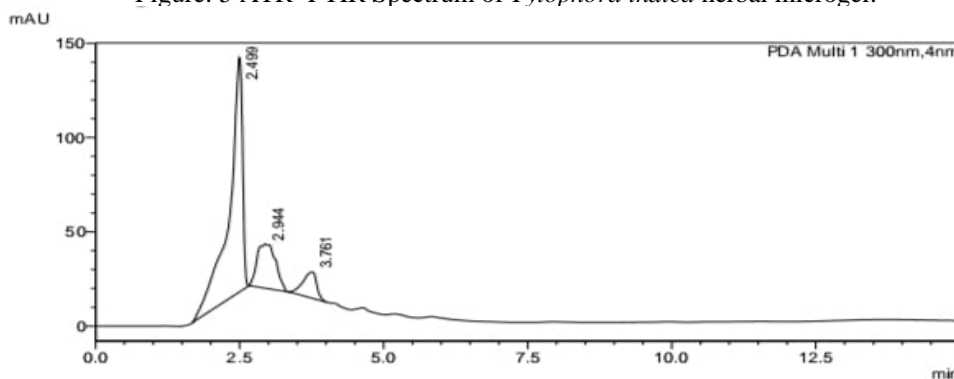


Figure: 6 HPLC for *Tylophora indica* plant extract.

Table: 9 Viscosity for *Tylophora indica* herbal microgel.

Formulation Code	Viscosity (cps)
F1	12000
F2	14000
F3	16000
F4	11000
F5	14000
F6	15000
F7	12000
F8	11000
F9	13000

Table:10 Zeta potential for *Tylophora indica* herbal microgel.

Sample Name	Zeta potential distribution (mV)	Zeta deviation	Conductivity	Result quality
<i>Tylophora indica</i> herbal micro gel (Optimized Formulation)	-29.3 Mv	5.10	0.225	Good

A solubility study was performed in order to determine the profile of the drug substance regarding its solubility in various solvents. The method consists of adding the extract to the respective solvent media and agitating the mixtures for 2 h. The presence or absence of undissolved material was observed to find out whether the saturation point is achieved or not. The resulting slurry was filtered, and then samples were assayed by a UV-visible spectrophotometer at a wavelength between 252 to 255 nm.<sup>12</sup>

**Drug-Excipients compatibility study**

A drug-excipient compatibility study was conducted using an Attenuated Total Reflectance Fourier-Transform Infrared (ATR-FTIR) spectrometer. Spectral data were collected over the wavelength range of 4000 to 400 cm<sup>-1</sup>. A precisely measured amount of extract was thoroughly mixed with excipients in a mortar to achieve homogeneity. The resulting mixture was then transferred to the sample holder cavity for analysis. The FTIR spectrum was subsequently recorded.<sup>13</sup>

**HPLC**

The HPLC system was configured with ECTO technology and a photodiode array detector. A SHIM-pack G15 analytical column (C18, 5 μm, 4.6 x 250 mm) was utilized, with the mobile phase comprising a mixture of 0.5 M NaOH and HPLC-grade water (35%) from pump A, and acetonitrile (65%) from pump B. The column temperature was kept constant at 40°C, and the flow rate was set at 1 mL/min. The detection wavelengths were programmed to start at 190 nm and end at 800 nm.<sup>14</sup>

***Tylophora indica* herbal microgel formulation**

**Methodology for Herbal Gel Formulation**

The process begins with the precise measurement of Carbopol 934, which is then dispersed in 50 mL of purified water. This mixture is left undisturbed for a half-hour to allow the Carbopol to fully hydrate. Subsequently, the solution is agitated using a laboratory stirrer at 1200 rotations per minute. In a separate vessel, a specific quantity of the herbal extract is combined with 1.5 mL of propylene glycol. Concurrently, another container is

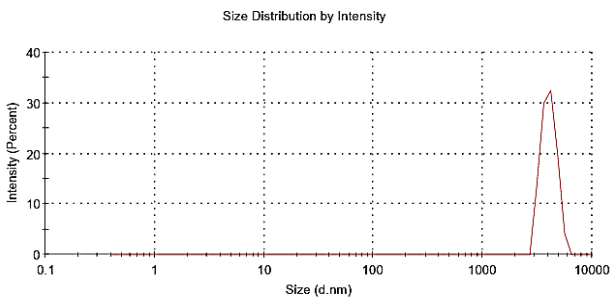


Figure: 7 Globule size measurement for *Tylophora indica* herbal microgel.

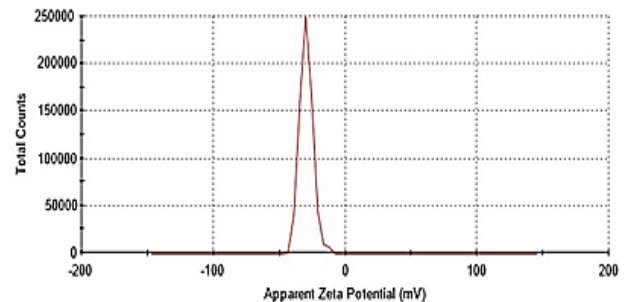


Figure: 8 Zeta potential for *Tylophora indica* herbal microgel.

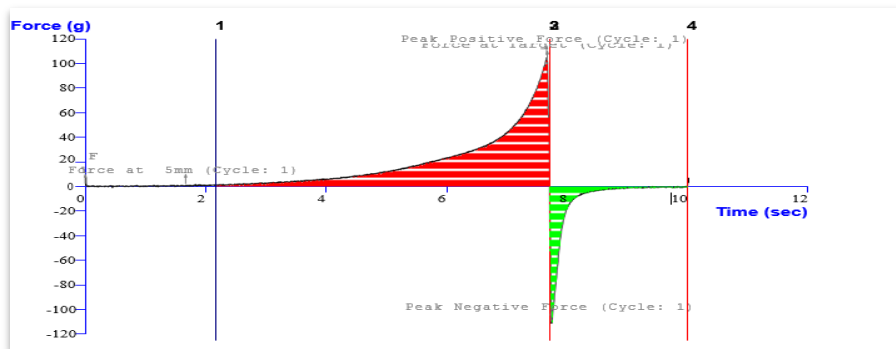


Figure: 9 Spreadability for *Tylophora indica* herbal microgel.

Table:11 Spreadability for *Tylophora indica* herbal microgel.

Formulation Batch	Spreadability (gm.cm/s)
F1	25.18
F2	20.13
F3	19.31
F4	24.21
F5	21.03
F6	20.34
F7	25.15
F8	22.19
F9	21.15

Table: 12 Extrudability for *Tylophora indica* herbal microgel.

Formulation Batch	Extrudability
F1	30.12
F2	24.32
F3	20.94
F4	29.00
F5	27.57
F6	22.22
F7	32.15
F8	26.21
F9	23.12

prepared by thoroughly blending 1.5 mL of propylene glycol with 2.5 g of methyl paraben. The next step involves the integration of the Carbopol dispersion with 2 g of the herbal extract and the previously prepared preservative mixture. This combination is subjected to continuous agitation to ensure homogeneity. The formulation is then brought to a final volume of 100 mL through the addition of purified water. To achieve a skin-compatible pH range of 6.8-7.0, triethanolamine is carefully introduced in small increments. This final adjustment results in the completed gel preparation at the intended concentration

**Evaluation of herbal microgel of whole plant extracts of *Tylophora indica***

**Determination of pH**

The acidity or alkalinity of the gel preparations was assessed using an electronic pH measurement device. A sample of each formulation, weighing precisely 1 gram, was introduced into 100 mL of purified water. This solution was then left undisturbed for a period of 120 minutes to ensure complete dissolution and stabilization. For duplicates of each extract, the pH values were recorded three times to ensure accuracy and reliability. The arithmetic mean of the measurements then was calculated to obtain average pH values for the respective gel formulations examined in the study.<sup>15</sup>

**Viscosity**

The incubation period was at least 16 hours at a temperature of 25 degrees Celsius. Then the specimens were submitted to a rheological study, through a rotational viscometer after the incubation. To ensure the right measurements within the optimal range of torque, which varied between 10% and 90%, the instrument was used at various rotational speeds. For instance, the revolution speed of the spindle was varied in order to reach 0.5, 1.0, 2.5, and 5 rpm. This allowed for the thorough checking up of the viscosity profile of sample within specific shear rates.<sup>16</sup>

**Spreadability** Determination of spreadability of gel formulations Using a special set-up consisting of a wooden block with a pulley attached at one end, the apparatus was designed to estimate the slip and drag properties of the gels. Spreadability S can be calculated using the following:

$$S = M \times L / T,$$

where S means the spreadability,

M is the mass attached to the upper slide,

Table: 13 anti-inflammatory activities for *Tylophora indica* ethanolic extract and *Tylophora indica* herbal microgel.

Standard	Concentration (µg/ml)	OD at 660 nm	% of Inhibition
Control	-	0.787	
Diclofenac	6.25	0.725	7.88
	12.5	0.661	16.01
	25	0.545	30.75
	50	0.368	53.24
	100	0.125	84.12
IC50	49.68 µg/ml		
Sample code	Concentration (µg/ml)	OD at 660 nm	% of Inhibition
Control	-	0.864	-
<i>Tylophora indica</i> Ethanolic extract	6.25	0.821	4.98
	12.5	0.765	11.46
	25	0.649	24.88
	50	0.560	35.19
	100	0.514	40.51
Expected IC 50	200	0.450	47.92
	222.27		
	6.25	0.807	6.60
	12.5	0.746	13.66
<i>Tylophora indica</i> herbal microgel	25	0.632	26.85
	50	0.528	38.89
	100	0.479	44.56
	200	0.403	53.36
IC 50	162.76		

L represents distance travelled by the gel, and T indicates the time taken by the upper slide to completely detach from the lower slide.

**Extrudability**



The gel formulations were dispensed to standard collapsible aluminum tubes, which were then sealed by capping and crimping. The weight of each filled tube was recorded. The tubes were then placed between two glass slides, and the amount of gel extruded was measured. Extrudability was calculated as a percentage, with the results categorized as follows: >90% extrudability—excellent, >80%—good, and >70%—fair<sup>18</sup>.

**Globule size measurement**

A drop of the microgel was placed on a glass slide, covered with a coverslip, and examined under a microscope using a 10X objective lens to measure the particle size of the optimized microgel batch. Built-in cameras in the digital microscope allow it to be plugged directly into a computers USB port. The software on the computer was used to view live images and measure globule size, after which the images were saved<sup>19</sup>.

**Anti-inflammatory activity study**

The anti-inflammatory efficacy was assessed through an in vitro protocol employing a reaction medium composed of a 3% bovine serum albumin (BSA) solution in aqueous form. This solution, measuring 0.4 mL, was combined with varying concentrations of the test compound to achieve a total volume of 0.5 mL. The resulting mixtures underwent incubation at 37°C for a duration of 20 minutes. Subsequently, 2.5 mL of phosphate-buffered saline (PBS), adjusted to pH 6.3, was introduced to each experimental vessel. The samples were then subjected to thermal stress by heating at 80°C for a 10-minute interval. Spectrophotometric analysis was performed to measure the absorbance of the solutions at a wavelength of 660 nm. To quantify the anti-inflammatory effect, the degree of protein denaturation inhibition was determined. This was accomplished by applying a mathematical formula to calculate the percentage of inhibition based on the spectrophotometric data obtained.<sup>20,21</sup>

Table no: 14 Stability study for Tylophora indica herbal microgel.

S.No	Tests	Initial	1 Month (40±2°C/75±5% RH)	2 Month (40±2°C/75±5% RH)	3 Month (40±2°C/75±5% RH)
1	Description	Light green	Light green	Light green	Light green
2	Odour	Pleasant	Pleasant	Pleasant	Pleasant
3	Nature	Opaque	Opaque	Opaque	Opaque
4	Ph	6.0	6.0	5.8	5.7
5	Viscosity	16000	15500	13000	14000
6	Spreadability	19.31	22.1	21.01	20.22
7	Extrudability	20.94	845.36	943.13	958.27

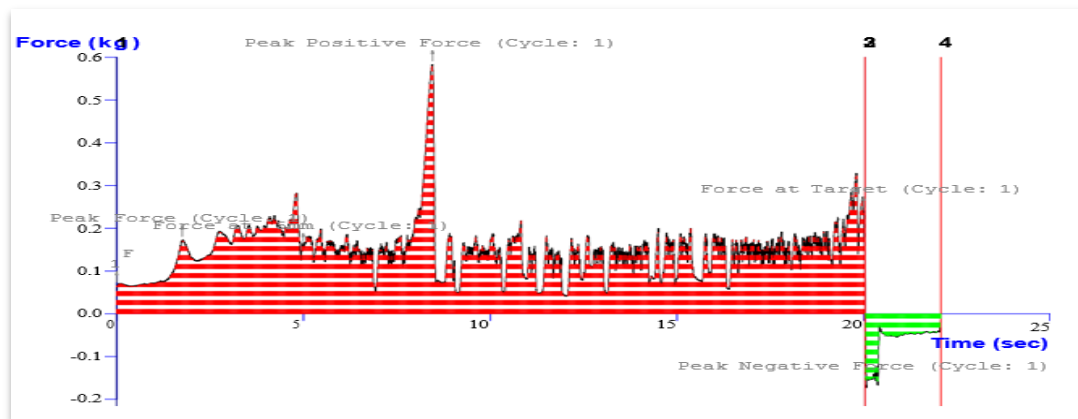


Figure: 10 Extrudability for Tylophora indica herbal microgel.

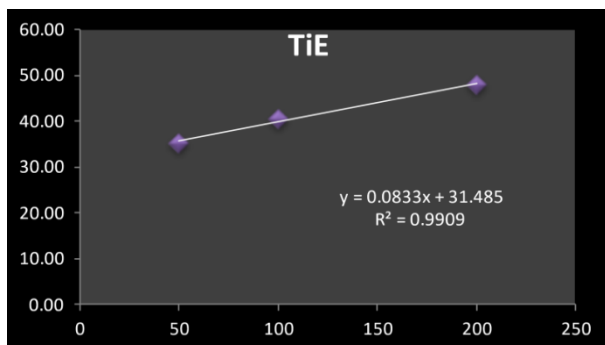


Figure:11 Tylophora indica extract for anti-inflammatory activity.

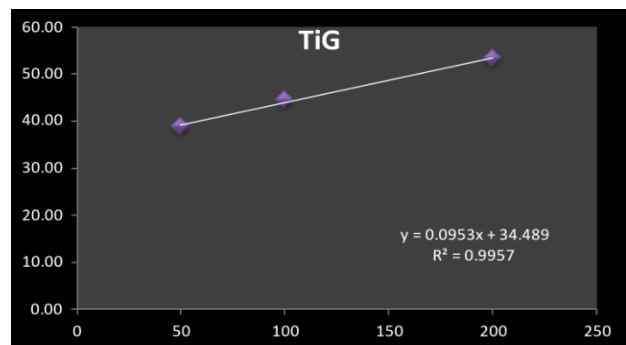


Figure:12 Tylophora indica herbal microgel for anti-inflammatory activity.

### **Inhibitory effect calculation**

% inhibition = [(Abs. control – Abs. test)/ Abs. control] x100

Where, Abs. = Absorbance

### **Determination of short time stability studies as per ICH guidelines**

Stability assessments are integral to evaluating the temporal alterations in drug substance or product quality, considering the impact of environmental variables including temperature, moisture content, and illumination. The findings from these investigations inform the determination of appropriate storage parameters, intervals for re-evaluation, and product longevity. Since degradation processes at ambient conditions are often not detectable within short time frames, the conducted study used an accelerated stability protocol. The optimized formulation was evaluated using controlled conditions of elevated temperature  $40^{\circ}\text{C} \pm 2^{\circ}\text{C}$  and relative humidity  $75\% \pm 5\%$  RH for a period of three months. This approach is used to simulate long-term storage effects within a compressed time frame of acceptable periods.<sup>22</sup>

## **RESULTS AND DISCUSSION**

Different chemical tests were conducted to identify the phytoconstituents of plant extracts. The largest amount of the present phytoconstituents in the extract was steroids, Flavonoids, saponins, phenolic compound, Amino-acids, glycosides, terpenoids, and Carbohydrates. The extraction efficiency of the herbal plant material was tested by using different solvents with different polarities. The extractive process used the petroleum ether as well as ethanol and an aqueous medium to extract the phytoconstituents of *Tylophora indica*. Each extraction yield resulted in various differences among the solvent used. Extraction yield with a non-polar solvents using petroleum ether was 14.29%. In contrast, the more polar ethanol solvent exhibited higher extractability, extracting 15.8% of the plant material. The aqueous extraction—the most polar solvent system—showed a yield of 12.38%. These observations then gave an idea about the extractability dependent on the solvent system used in this experiment with regard to the components of the plant material, which varied in polarities of the phytochemicals found in *Tylophora indica*. Such variability in yields itself presents an imperative, for the selection of the right solvents would help in optimising the yield from this herbal source.

### **ATR- FTIR Spectra analysis**

No new peak or disappearance of an existing peak was noted in ATR-FTIR spectral analysis of *Tylophora indica* ethanolic extract and formulation excipients, which indicates that the pure plant extract and incorporated excipients do not interact with each other significantly.

### **Particle size determination (SEM)**

SEM technique was adopted for morphological characterization of *Tylophora indica*. The rationale for the use of surface analytical technique lies in the potential of creating a comparison of the surface features of the raw herbal extract with that of the formulated microgel. This information obtained from the SEM analysis will reveal the topographical characteristics and structural attributes

of the *Tylophora indica* samples in both forms, which will provide valuable knowledge about the physical properties of the material before and after incorporation in the microgel formulation. SEM was used for the explanation of the surface morphology features of the samples. From the SEM images obtained, it may be concluded that the size of particles ranges within the interval of 1 to 2 micrometers and shows a randomly distributed spherical morphology.

### **Pre formulation studies**

Based on overall sensory analysis, the physical attributes of the extract underwent an evaluation. The investigation that was conducted included its chromatic characteristics; its olfactory properties; and the general physical nature of the extract. From these investigations, the outcome of the analyses was generally found to be acceptable, and this may imply that its sensory profile is unlikely to compromise patient compliance or cause severe discomfort during its application. These observations thus suggest that the physical properties of the extract are acceptable to the patients and, therefore, not likely to compromise the acceptability of the regimen.

### **Solubility study**

Different solvents were used to perform the solubility test on the extracts. Results indicate that the ethanolic extract of *Tylophora indica* is soluble in distilled water, ethanol, and hydrochloric acid; values obtained are  $0.248\mu\text{g/ml}$ ,  $0.531\mu\text{g/ml}$ , and  $0.0157\mu\text{g/ml}$ , respectively.

### **Chemical compatibility studies**

Fourier-transform infrared spectroscopy with attenuated total reflectance (ATR-FTIR) was utilized to assess the interactions between the botanical extract and auxiliary substances in the formulation of herbal microgels. The spectral analysis of both the plant-derived extract and the synthesized microgel was conducted across a wavenumber spectrum spanning from  $4000$  to  $400\text{ cm}^{-1}$ . This has been confirmed in the study, because no extra peaks had been found in the physical mixture, therefore the extract is compatible with the excipients. The ATR-FTIR test of compatibility pointed out that excipients do not interact with the extract.

### **HPLC**

Among this studied formulation of ethanolic extract was range carried out from 300nm and size 4nm.

### **Evaluation of herbal microgel of entire part plant extract of *Tylophora indica***

#### **Visual inspection of Gel**

Color, texture, and appearance of the prepared herbal microgel were observed. All the formulae showed a light greenish color and a smooth and viscous texture, confirming good consistency without the presence of lumps. The color, odor, and features of the formulations were investigated by ocular inspection. The observations demonstrated that all the formulations had presented uniform status in their transformation and stated that the extract-loaded topical herbal microgel formulation may be well tolerated without serious impairment for the patients.

#### **Globule size measurement**

By using a digital microscope with a 10X magnification objective lens, microgel images revealed spherical shape



globules with size range of Globule size by digital microscope. The globule size measurement of the herbal microgel. By using a digital microscope with a 10X magnification objective lens, microgel images revealed spherical shape globules with size range. F3 showed better rheological properties, forming a microgel that could be spread more easily with less application of shear force. This is noted as a potentially plagiarized section after the content verification process. The enhanced consistency of formulation F3 relies on the possibility of structuring and conducting optimization of the mechanical features of the microgel. Improved low shear spreadability of the microgel suggests an optimum between viscoelastic properties, including storage and loss moduli, and thixotropic behavior. This improved profile is most presumably set by the careful selection and manipulation of the compositional and processing parameters of the formulation.

#### **pH**

The acidity levels of the synthesized botanical hydrogel formulations were quantified using an electronic pH measurement device. The procedure involved submerging the glass sensing probe entirely within the gel matrix, ensuring comprehensive contact between the electrode and the sample. This method facilitated accurate determination of the hydrogen ion concentration in the prepared phytotherapeutic colloidal system. An analysis of the acidity levels was conducted across all engineered gel compositions, which incorporated varying concentrations of polymeric substances. The formulations F1 to F9 had a pH ranging between 5.6 to 6.0. When the results were analyzed, it was found that the F3 preparation had a more favorable profile in terms of pH compared with others.

#### **Viscosity**

The viscosities of various extract-loaded herbal microgel formulations, that is, F1 to F9, were measured by a rotational viscometer, LMDV-100 model, using an RV-7 spindle. Measurement was done at 20 revolutions per minute. In each case, 100 gram of microgel formulation was taken in a beaker, and the spindle was immersed into the sample. The stirring was continued for 5 minutes to get a stable and representative viscosity reading for all the formulations. These results point out that the viscosity of all formulation fell within the range of 11,000 to 16,000 cps. Viscosity in a microgel preparation depends on the concentration of its gelling agent. So, formulation F3 showed better consistency with a viscosity of 16,000 cps.

#### **Zeta potential and Size distribution**

In this investigation, the colloidal stability and dimensional characteristics of the formulation were assessed through zeta potential and particle size distribution analyses. The measurement of zeta potential serves as a predictive indicator for the stability of the botanical extract in question. Generally, zeta potential values spanning from -30 mV to +30 mV are considered significant for colloidal stability. In the case of the extract derived from *Tylophora indica*, the observed zeta potential was determined to be -29.3 mV.

#### **Spreadability**

The viscosity-dependent spreadability of the microgel was calculated to be within the range of 18.18–19.31 g·cm/sec for all formulations. Formulation F3, being more cohesive, exerted higher spreadability with minimum shear force.

#### **Extrudability**

An evaluation of the extrusion characteristics was conducted for the series of gel formulations, which were prepared using varying concentrations of polymeric agents. The quantitative assessment of extruded material was performed on preparations labeled F1 through F9. The results revealed that the extruded mass ranged from 27.57 to 30.12 gm·cm<sup>-2</sup>. Analysis of these findings indicates that all engineered gel compositions fall within the acceptable parameters as stipulated by pharmacopoeial standards.

#### **Anti-inflammatory activity**

The anti-inflammatory activity was evaluated by the *Tylophora indica* ethanolic extract and *Tylophora indica* herbal microgel.

#### **Assessment of Accelerated Stability Parameters in Accordance with International Harmonization Protocols**

Stability testing of F3, the optimized microgel formulation, was carried out following the ICH guideline. The accelerated stability assessment was conducted under controlled environmental conditions, with the samples exposed to an elevated temperature of 40°C and a relative humidity of 75% for a duration of one quarter of a year. This stability study demonstrates that the physical and chemical characteristics of the microgel did not change profoundly within the testing period.

The findings have established the stability profile of the F3 formulation under accelerated storage conditions according to the ICH guidelines. Consequently, insignificant changes in the physicochemical properties of the microgel during the three-month period of evaluation presuppose the ability of the formulation to retain its quality and performance characteristics during environmental stresses, such as elevated temperature and humidity. The optimized formulation F3 underwent an accelerated stability assessment over a three-month period. This evaluation was conducted under controlled environmental conditions, specifically at an elevated temperature of 40°C (with a tolerance of ±2°C) and a relative humidity of 75% (with a tolerance of ±5%). The results showed that no significant variation in all parameters was observed during the stated period, and all parameter values were within the acceptable limit.

#### **CONCLUSION**

The present study was designed to develop and evaluate a herbal microgel containing *Tylophora indica* extract.. To prepare extract by using hot percolation method. So, considering the above results it was found that the formulation F3 was found to be optimized formulation from the data obtained for all studies of Scanning electron microscope (SEM) analysis, ATR-FTIR, HPLC, pH, viscosity, Spreadability, extrudability, zeta potential, globule size measurement was optimal range compare to other formulations. Stability studies in room temperature 40°C/75% maintained for three months herbal microgel

any physical and chemical changes. Topical herbal microgel drug penetration in drug release of specific site in action. It should be advantageous for patients suffering, providing better patient compliance and an effective mode of treatment.

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