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## DESIGN DEVELOPMENT AND EVALUATION OF NOVEL FORMULATION FOR WOUND HEALING ACTIVITY

YELMATE AA<sup>1\*</sup>, SATPUTE KL<sup>1</sup>, EKLINGE SS<sup>2</sup> AND SABNE AA<sup>1</sup>

1: Dayanand College of Pharmacy, Latur, Maharashtra, India

2: IP DRG Medical Coder, GeBBS Healthcare solutions pvt. Ltd., Dayanand College of Pharmacy, Latur,  
Maharashtra, India

\*Corresponding Author: Dr. Yelmate Archana Ashok: E Mail: [archanavelmatel@gmail.com](mailto:archanavelmatel@gmail.com)

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### ABSTRACT

Wound is an anti-inflammatory condition associated with skin rashes, gunfire, flaming objects, surgery, cutting, and piercing substances etc. Wound causes by disruption of skin tissue while it has internal or external in origin. Both internal and external sources of wounds are possible. Internal wounds are typically brought on by poor circulation, neuropathy, or a medical condition. Open or closed wounds caused by an external force or trauma are known as wounds of external origin. There are many possible cure but they associated with disadvantages in the allopathic medicines has need to develop the new formulations from natural remedies with wound healing activity. *Moringa oleifera*, *Annona squamosa*, *Tridax procumbens* found to be efficacious and anti-inflammatory drug with least side effects as compared to synthetic drugs used in treatment of wound. Fundamental to wound research is identifying the basic mechanisms of healing and then controlling these mechanisms to promote quicker healing or prevent undesirable outcomes like infection or scarring. A complex biological process called wound healing occurs inflammation, proliferation, remodeling, and hemostasis. With the aforementioned justification in mind, an effort was undertaken to design and develop herbal nanogel and test it for anti-inflammatory properties. The formulation was evaluated for its in vivo anti-inflammatory efficacy, pH, particle size, viscosity, homogeneity, and consistency.

**Keywords: Anti-inflammatory, Wound, Nanogel, Polyherbal**

## INTRODUCTION

*Moringa oleifera* L. belong to family *Moringaceae* [1]. There are 13 species in the family *Moringaceae* that are currently recognised, among them the locally cultivated *Moringa oleifera* in Afghanistan, the sub-Himalayas (India, Pakistan, and Bangladesh), Caribbean islands, Southeast Asia, Arabia, South America, and Africa. Worldwide, moringa was spread to tropic and subtropical nations. Fast-growing softwood tree *Moringa oleifera* (*Moringaceae*) is native to Northern Indian sub-Himalayan regions [2]. *Moringa oleifera* roots their anti-inflammatory, anti-microbial, wound healing, anti-oxidant properties it also show that *Moringa oleifera* leaves exhibit antidiabetic control and glycemic control [3].

*Annona squamosa* L. belongs to family *Annonaceae*. In India, it is planted up to a height of 900 metres and is widely distributed. In mountainous areas, wastelands, and numerous districts in Andhra Pradesh, Punjab, Rajasthan, Uttar Pradesh, Madhya Pradesh, Bihar, West Bengal, Assam, Gujarat, Maharashtra, Karnataka, Kerala, and Tamil Nadu, it can be seen growing gregariously and widely. It is a native of the West Indies and South America [4]. Stems and leaves of *A. atemoya* examined against both Gram-positive and Gram-negative bacteria. *A.*

*squamosa* shows anti-inflammatory, anti-oxidant, neurological, bronchodilatory, antispasmodic, antihypertensive, and antihistaminic properties. . The anticancer properties of many diterpene isolated from barks against lung and ovarian cancer cells were promising [5].

*Tridax procumbens* L. belongs to family *Asteraceae*. India is home to the weedy wild plant *Tridax procumbens* Linn. Tropical Africa, Asia, and Australia have all adopted the plant as their own. It is originally from tropical America [6]. It is utilized in Ayurvedic medicine to treat heartburn, gastritis, liver conditions, and hepatoprotection. In some areas of India, traditional healers also utilize *Tridax procumbens* to cure boils, blisters, and cuts [7, 19].

## MATERIALS AND METHODS

### Selection of plant material:

Selection of plant material based on Extensive literature survey. Survey reveals that plant *Moringa oleifera*, *Annona squamosa*, and *Tridax procumbens* have preferable wound healing activity as well as other potent activities against diseases.

### Collection of plant material:

Plant materials were collected from botanical garden of Dayanand college campus and from

local farm. After selection some steps are followed:

- Plucking the leaves from plant.
- Picking the clean leaves of plant.
- Leaves are rinse with the water to remove dust particles.
- Washed leaves kept for shed drying on a filter paper at room temperature.
- After drying powdered the leaves with the help of grinder.

#### **Standardization of selected plant material:**

The quality of the initial material is crucial for producing a final product of reproducible quality. As a result, the standardization of the plant samples used in the study was carried out in accordance with the detailed instructions of WHO monographs. The following specifications were used to standardize. Ash value, Acid insoluble ash, Water soluble ash, Total ash value, Loss on Drying/ Moisture determination, Foreign Matter Determination, and Determination of Extractive Values.

#### **Trace Element Determination in Medicinal Plant Samples by ED-XRF Analysis:**

ED-XRF method is used to measure the concentration of trace elements in specific medicinal plants that are used to treat skin conditions. We analyze trace element in *Moringa oleifera*, *Annona squamosa*, and *Tridax procumbens* by ED-XRF method. We

found element like Si, P, S, Cl, K, Ca, Ti, V, Mn, Fe, Cu, Zn, Br, Rb, Sr, Ag, Re, Cr, Zr, Ni. Quantity of element K and Ca shown maximum Concentration.

The calcium ion has been demonstrated to function as a key cue, directing the cellular processes of various types of cells during wound healing in addition to being an important coagulation factor during hemostasis. For keratinocytes and fibroblasts, calcium serves as both an intracellular second messenger and an external signaling molecule. It is well known that calcium ions control the intracellular signals that control a variety of cellular functions. Chapman first showed that the activation of calcium channels near the cell membrane was accompanied by changes in the calcium gradient following chemical or physical stimuli in 1983. Although 100 times less sensitive to extracellular calcium than keratinocytes, fibroblasts nonetheless respond to it. Calcium is primarily used by fibroblasts intracellularly for contraction, and this contraction is crucial for minimizing the size of the wound during wound healing. In mouse embryonic hind limbs, potassium channels play a function in limb wound healing and regeneration [11, 12, 20].

#### **Extraction of plant:**

Here, cold maceration method used for extraction. The *Moringa oleifera* powdered leaves (500g) were air-dried, ethanol-extracted (95%v/v), and concentrated on a water bath to produce ethanol extract (32%). The 500g of powdered *Annona squamosa* leaves, which had been air dried, was extracted with ethanol (95%v/v), concentrated on a water bath, and then yielded ethanol extract (26.4%). *Tridax procumbens* leaves that had been air dried and powdered (500g) were extracted with ethanol (95%v/v) and concentrated over a water bath to produce ethanol extract (28.3%).

**Phytochemical screening:** The prepared herbal extract were subjected for qualitative chemical test.

#### **Pre-formulation studies:**

Following are the Preformulation studies performed before formulation: Estimation of extract by U.V Spectrophotometer:

Dissolve 13.872g of potassium dihydrogen phosphate and 35.084g of disodium hydrogen phosphate in sufficient water to produce 1000ml of 0.2M pH 6.8 phosphate buffer. PH check by digital pH meter before use.

#### **Preparation of standard calibration curve:**

To prepare the stock solution of 1000 g/ml, 50 mg of accurately weighed extract was diluted in 25 ml of ethanol and made up in 50 ml with phosphate buffer pH 6.8 in a 50 ml volumetric

flask. To create 100 gm/ml solutions, 5 ml of the stock solution was pipetted out and diluted up to 50 ml with buffer. 0.5, 1, 1.5, 2, 2.5, and 3 ml were taken out of it and diluted to a final volume of 10 ml with buffer to produce a concentration in range of 2–10 ug/ml. Through the use of a UV spectrophotometer, the absorbance of the solutions was calculated in nm. Concentration and absorbance were plotted on a graph. Multiple extracts with different wavelengths.

#### **FTIR of drug extract:**

#### **Formulation:**

#### **Solvent dispersion method:**

“A dispersion of one or more active ingredients in an inert carrier or matrix of solid state prepared by melting (fusion), solvent or melting solvent method” is the definition of a solid dispersion. Coprecipitates and melts are other names for solid dispersion. This method reduce particle size, improve porosity, improve wettability, stabilize unstable drug.

#### **Preparation of nanogel:**

##### **Phase I**

0.4 gm of carbopol 940 was taken in a large beaker and dispersed in 30 ml of distilled water. The beaker containing Carbopol 940 and water kept on mechanical heating plate. Slightly heat it to dissolve Carbopol 940. This solution kept aside to cool.

## Phase II

Take a beaker plastic beaker, add 10ml of ethanol, add measured quantity of eudragit, polaxomar, methyl paraben, propyl paraben by continuous stirring. Add solubilized extract of three plant. The solution kept for sonication in ultrasonic sonicator for 10 minutes. Phase II is ready.

## Mixing of phase I and phase II

By using homogenizer add phase II in phase I drop by drop with the help of syringe. After completely mixing of phase add propylene glycol and glycerine drop by drop. Keep the prepared mixture for homogenization for 1 hour. Add a few drop of triethanolamine by continuous stirring to adjust pH. Make up the volume by adding distilled water. Use 8000rpm speed of homogenizer.

## Formulation batches of nanogel:

Table 1: Formulation of polyherbal nanogel

Sr. No.	Ingredient	Formulation Quantity taken per 50 gram			Use
		F1	F2	F3	
1.	<i>Moringa oleifera</i> ethanolic extract	0.5gm	1gm	1.5gm	API
2.	<i>Annona squamosa</i> ethanolic extract	0.5gm	1gm	1.5gm	API
3.	<i>Tridax procumbens</i> Ethanolic extract	0.5gm	1gm	1.5gm	API
4.	Carbopol 940	0.4gm	0.4gm	0.4gm	Gelling agent
5.	Polaxomar F-407	0.2gm	0.2gm	0.2gm	Surfactant
6.	Eudragit RS-100	0.3gm	0.3gm	0.3gm	Permeability enhancer
7.	Methyl paraben	0.2gm	0.2gm	0.2gm	Preservative
8.	Propyl paraben	0.02gm	0.02gm	0.02gm	Preservative
9.	Propylene glycol	5ml	5ml	5ml	Penetration enhancer
10.	Glycerine	5ml	5ml	5ml	Co solvent
11.	Triethanolamine	q.s	q.s	q.s	Maintain pH
12.	Ethanol	q.s	q.s	q.s	Solvent
13.	Distilled water	q.s upto 50gm	q.s upto 50gm	q.s upto 50gm	Vehicle

## Evaluation parameters:

All developed polyherbal nanogel formulations were evaluated physicochemical evaluation in the current investigation. To suit the needs of the intended polyherbal nanogel formulation, the following factors were investigated.

## Physical appearance:

All three prepared polyherbal formulation were evaluated physico chemically.

**Color:** The formulation consistency was examined against a white background.

**Odor:** The odor was checked by smelling the nanogel.

**Greasiness:** check by applying on skin.

**Consistency:** Determined by applying on skin.

**Skin irritation study:** check by skin response after some time of application

**pH:**

Using a digital pH meter, the pH of produced gel compositions was assessed. 100 ml of distilled water were used to dissolve 1g of gel, which was then set aside for 1 hour. Take reading simultaneously.

**Viscosity:**

Viscosity of polyhedral gel determined by Brookfield viscometer Model LV (DV-E) at 2.4 rpm using spindle no. 64.

Take a beaker filled of nanogel dip spindle in beaker keep it stable until reading of viscometer remain constant.

**Spreadability:**

The equipment, which consists of a wooden block with a pulley at one end, was used to determine spreadability. By using this technique, spreadability was assessed based on the gels' properties of slip and drag. On this ground slide, extra gel (approximately 2 gm) was used for the experiment. The gel was then placed in a sandwich between this glass slide and another glass slide with a hook and a fixed ground slide dimension. To remove air and create a consistent gel film between the slides, a one kg weighted was placed on top of the two slides for five minutes. The borders were scraped clean of extra gel. After then, an 80 gram pull was applied to the top plate. With

the aid of the thread that is fastened to the hook, calculate how long (in seconds) it takes the top slide to travel 7.5 cm [17, 21, 22].

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

Where, S= Spreadability,

M= weight in the pan (tied to upper slide),

L= Length moved by the slide,

T= Time (in sec.)

**Extrudability:**

The collapsible aluminium tubes with standard caps were filled with the gel compositions, and the ends were crimped shut to seal. The tube weight were noted. The tubes were clamped after being positioned between two glass slides. The slides were covered with 500 gram, and the cap was then taken off. The extruded gel's volume was gathered and weighed. Calculated percentages of the extruded gel include >90% excellent, >80 percent good, and >70 percent fair extrudability [17].

**Washability:**

Skin care products were administered, and a manual check was made to see how much the skin had been washed. When washed with water, all of the formulations demonstrated excellent wash ability and left no residue on the skin.

**In vitro diffusion study:**

Utilizing a diffusion cell setup with an open ended cylindrical tube, in-vitro diffusion tests

were performed on all formulations. The dialysis membrane-70 (Hi-Media) was coated with a weighed amount of formulation equal to 1gm of the gel formulations, which was then slightly submerged in 100ml of receptor medium (phosphate buffer pH 6.8) while being stirred continuously on a magnetic stirrer and kept at a temperature of 37°C. At intervals of 5, 10, 15, 30, and 60 minutes, samples of 1ml were taken from each system. The same quantity of media was then reintroduced in the diffusion cell to maintain the sink condition before the drug content was examined using a UV spectrophotometer [18, 19].

#### **Particle size:**

The nanometer range was identified by the particle size study for the Nanogel. The homogenization duration and Carbopol 940 concentration both had an impact on the size of the nanoparticles. Malvern Master Sizer 2000 MS was used to measure and record the average sizes of nanogel.

#### **Screening of Antimicrobial study of Polyherbal formulation:**

**Principle:** antibiotic- impregnated discs with a known concentration on an agar plate that has been uniformly infected (or seeded) with a culture of the bacterium under investigation, discs are positioned. The plate is incubated at 37°C for 18–24 hours. The antibacterial

ingredient diffuses into the agar during this time and may stop the growth of organisms. The diameter of the inhibitory zone around the discs has a direct relationship with the effectiveness of susceptibility. Those organisms that reach the disc's edge are resistant.

**Procurement of microbial strains:** All the microorganism strains were obtained from NCIM. Polyherbal nanogel formulations containing ethanolic extracts of *Moringa oleifera* Linn and *Annona squamosa*, *Tridax procumbens* Linn were tested against *Staphylococcus aureus* (ATCC:6538), *Bacillus subtilis* (ATCC:10231), and *Pseudomonas aeruginosa* (ATCC:9027).

**Making distinct Concentrations of Formulation:** Using 10% DMSO, three distinct conc. of the plant's aqueous and ethanol extracts—25 mg/ml, 50 mg/ml, and 100 mg/ml were made.

**Culture media preparation:** Media and common anti-microbial medications (discs) were acquired from Hi-Media Laboratories Ltd. in India. According to the instructions, all the media were made on sterile glass Petri plates with a 4 mm thickness [15].

#### **Procedure:**

By using the agar well diffusion and agar disc diffusion methods, the anti-microbial activity of ethanolic extracts of *Moringa oleifera* Linn,

*Annona squamosa* Linn., and *Tridax procumbens* Linn. Including polyherbal nanogel formulations was evaluated against various bacteria. To determine the antimicrobial activity of the formulation, the zone of inhibition was assessed using the agar well and agar disc diffusion methods.

To prevent contamination during the investigation, all processes involved in this preparation were carried out in a strictly aseptic environment. Prior to use, all glassware—including beakers, volumetric flasks, droppers, measuring cylinders, pipettes, and laboratory glass bottles was autoclaved at 121 °C for 20 minutes. By using the agar well diffusion and agar disc diffusion methods, polyherbal nanogel formulations including ethanolic extracts of *Moringa oleifera* Linn., *Annona squamosa* Linn., and *Tridax procumbens* Linn. were evaluated against *Staphylococcus aureus*, *Bacillus subtilis*, and *Pseudomonas aeruginosa*.

A 10% DMSO solvent was used to create the extract solutions. The culture of was made on nutrient agar medium slants and incubated for

24 hours in an aerobic environment at 37 °C. Using a sterile 8 mm cork borer, an agar plate was punctured, and 25 L of polyherbal nanogel formulations ethanolic extracts of different concentrations (25 mg/mL, 50 mg/mL, and 100 mg/mL) were pipetted into each well. 30 minutes were given for the plates to stand. After that, plates underwent an aerobic incubation period of 24 hours at 37 °C. Additionally, the positive control Streptomycin discs (100 g/mL) were utilised. The diameter of the zone of inhibition in mm was used to calculate the antibacterial activity of topical polyherbal nanogel formulations [16].

## RESULT AND DISCUSSION:

### Collection and authentication of plant material:

In the month of January, leaves of *Tridax procumbens*, *Annona squamosa*, and *Moringa oleifera* were collected in the Latur area. Authentication was done by Dr.C.S Swami, Head of the department of Botany, Dayanand Science College, Latur.

Table 2: Morphological characteristics of plant part used

Sr no.	Evaluation Parameter	Name of plants		
		<i>Moringa oleifera</i>	<i>Annona squamosa</i>	<i>Tridax procumbens</i>
1.	Color	Green leaves	Green leaves	Green leaves
2.	Odour	Earthy, Nutty	Floral scent	Characteristics
3.	Taste	Bitter	Bitter	Slightly bitter
4.	Size	30-60cm (11.8 to 23.6in).	10-15cm long, and 3-5cm wide.	The lamina is 2-6cm in length and 2-4cm in width
5.	Shape	Pale green, compound, tripinnate	The leaves are oblong-lanceolate	Oval to lanceolate

**Standardization of selected plant:**

As a consequence, the standardization of the *Tridax procumbens*, *Annona squamosa*, and *Moringa oleifera* leaves utilized in the study

was completed in accordance with the precise guidelines of WHO monographs. It was evaluated that:



Figure 1: Some pictures during standardization

Values recorded during standardization of plants mentioned under study

Table 3: Values recorded during standardization of plant leaves

Sr.no.	Evaluation Parameters	Name of plants		
		<i>Moringa oleifera</i>	<i>Annona squamosa</i>	<i>Tridax procumbens</i>
1.	Ash value	8.7	0.21	15.7
2.	Acid insoluble ash	0.98	0.29	1.2
3.	Water soluble ash	19.4	5.2	4.2
4.	Total ash value	9.5	6.20	12.5
5.	Loss on drying	5.33	6	4
6.	Foreign matter determination	0.2	0.1	0.3

**Preparation of ethanolic extract: Percentage yield:**

Table 4: Percentage yield of plant extract

Sr.no.	Solvent	Color	Odour	Consistency	% yield w/w
1.	<i>Moringa oleifera</i>	Green	No	Semisolid sticky mass	31.5%
2.	<i>Annona squamosa</i>	Green	No	Semisolid sticky mass	28%
3.	<i>Tridax procumbens</i>	Green	No	Semisolid sticky mass	30%

**Preliminary phytochemical screening of extract:**

Result of phytochemical screening depends upon chemical constituents in extract. Either

result are positive or negative. Results are given in **Table 5**.

Sr.no	Test	<i>Moringa oleifera</i>	<i>Annona squamosa</i>	<i>Tridax procumbens</i>
1.	Carbohydrate	+	+	-
2.	Flavonoids	+	+	+
3.	Alkaloids	+	+	+
4.	Glycoside	-	+	-
5.	Saponin	-	-	-
6.	Terpenoid	-	-	-
7.	Phenol	-	+	+
8.	Steroid	+	+	+
9.	Tannin	-	+	+
10.	Protein	+	-	+
11.	Anthocyanin	-	+	-
12.	Reducing sugar	-	+	-

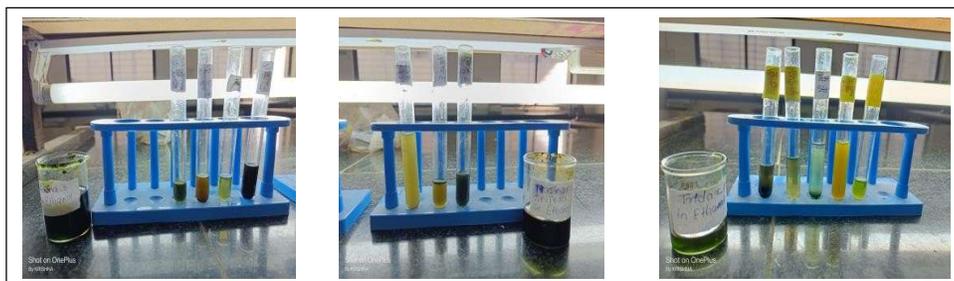


Figure 2: Phytochemical screening of Plant extract

#### FORMULATIONS STUDIES:

**Physical appearance:** The Polyherbal nanogel formulation was slightly blackish green in colour transparent in nature result are show in table no.

**pH:** pH of polyherbal nanogel formulation was found to be 6.2 to 6.96. As shown in **Table 6**

Photograph showing determination pH of formulation by using Digital pH meter

**Viscosity:** Utilizing a Brookfield viscometer, the viscosity of the Polyherbal nanogel was measured, and the results of the formulation are displayed in **Table 7**.

**Spreadability:** Spreadability was checked by glass slide and wooden block result was

recorded in the **Table 7**.

**Extrudability:** Collapsible aluminium tube was used to check extrudability. Results are given in **Table 7**.

**Washability:** Manually checked by after application washed with water. Wash ability shown in **Table 7**.

**In- vitro diffusion study:** In-vitro diffusion study was performed by using Franz diffusion cell and dialysis membrane to check percentage drug release of polyherbal nanogel. A result has been recorded as follows.

**Particle size:**

**Determination of particle size of polyherbal nanogel formulation:**

Using the Particulate System NanoPlus, the mean size of the nanogels was determined.

The result found as below:

**Batch F1**

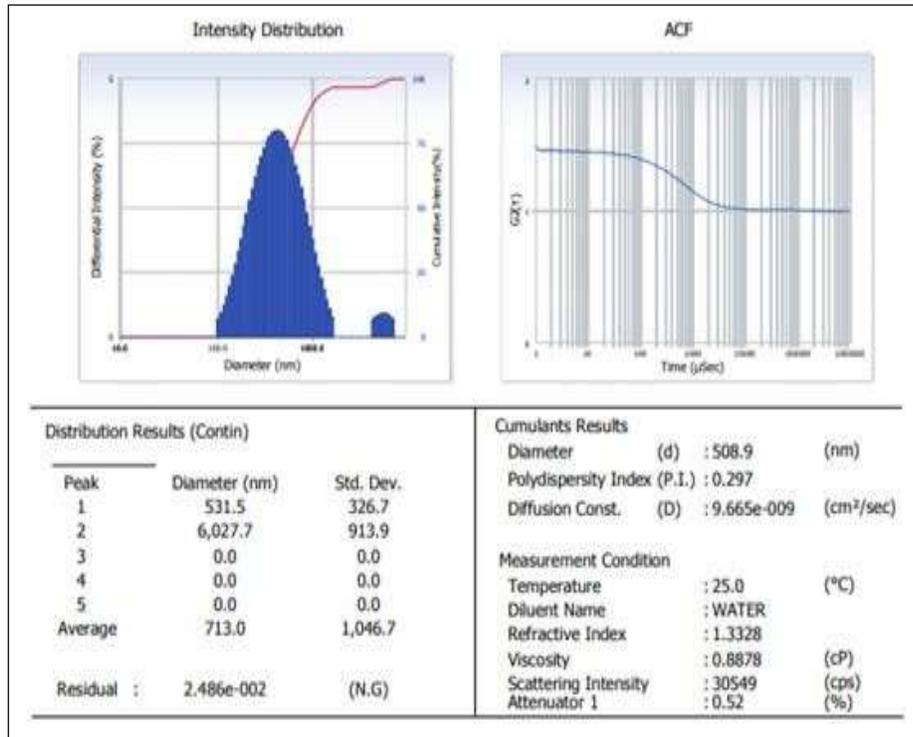


Figure 3: Evaluation of particle size of batch F1

**Batch F2**

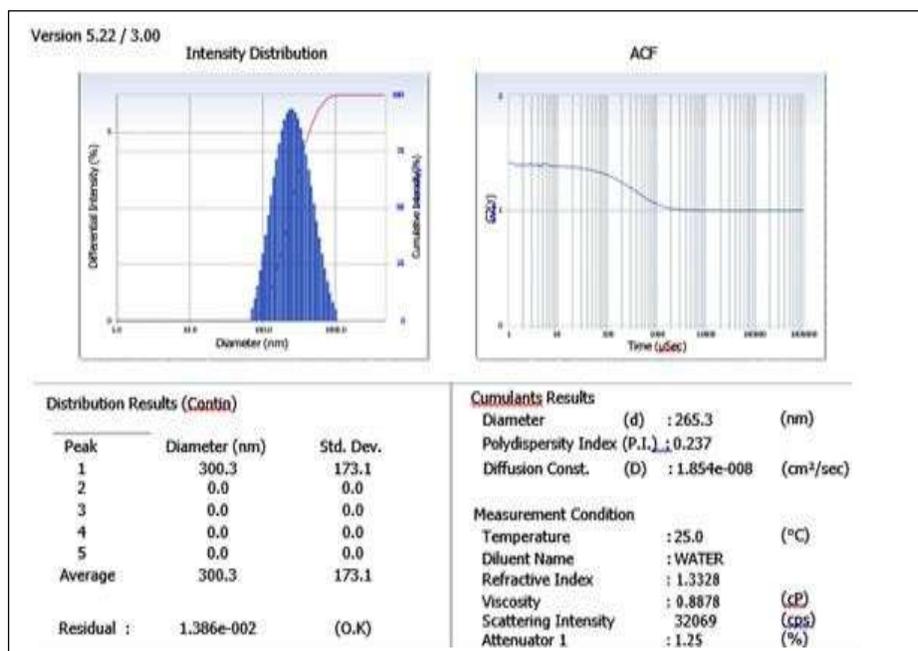
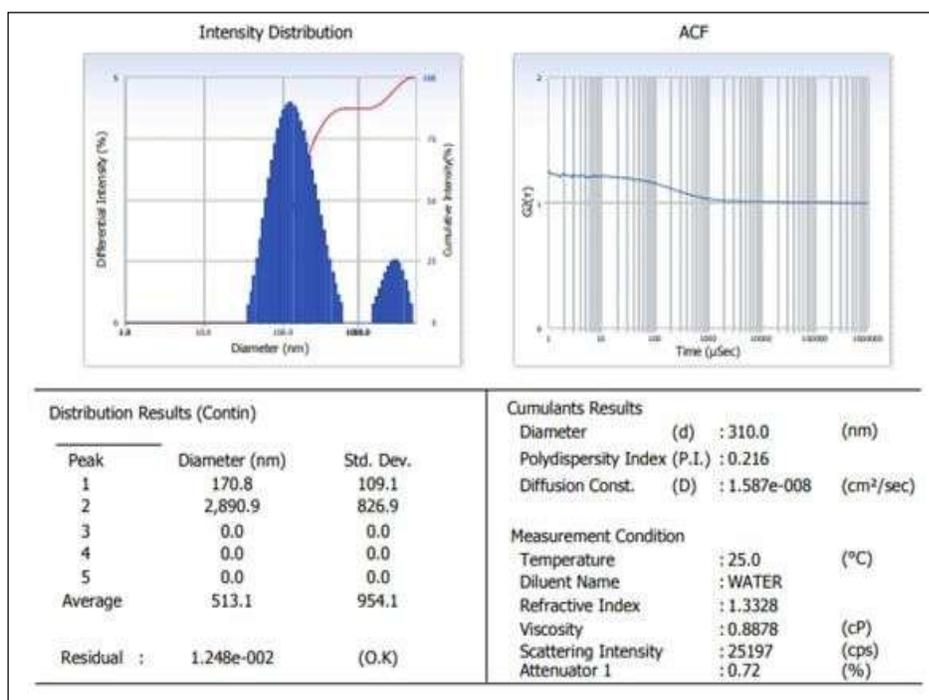


Figure 4: Evaluation of particle size of batch F2

**Batch F3**



**Figure 5: Evaluation of particle size of batch F3**

Nano particle size ranges between 200 to 500nm. After the evaluation F1 formulation recorded 508.9nm particle sizes range. F2 formulation recorded 265.3 particle size and

F3 formulation recorded 310.0nm particle size. Hence observed that F2 batch is optimized batch.

**Table 6, 7: Physicochemical evaluation of Polyherbal nanogel formulation batches F1, F2,F3**

Sr.no.	Batch	Appearance	pH	Homogeneity	Spreadability (g.cm/Sec)
1.	F1	Green transparent	6.9±0.2	Good	25.68
2.	F2	Brown transparent	6.8±0.2	Good	30.72
3.	F3	Green transparent	6.9±0.2	Good	26.33

Sr.no.	Batch	Extrudability	Viscosity(cps)	%Drug release	Particle size(nm)
1.	F1	82.08	4042	83.3	508.8
2.	F2	89.80	4899	88.01	265.3
3.	F3	80.58	3359	91.8	310.0

**Screening of Antimicrobial activity of Polyherbal nanogel formulations:**

After 24 hours, the plates were examined. By

measuring the smallest dimension of the area around the patch where no bacterial growth occurred, the zone of inhibition is estimated.

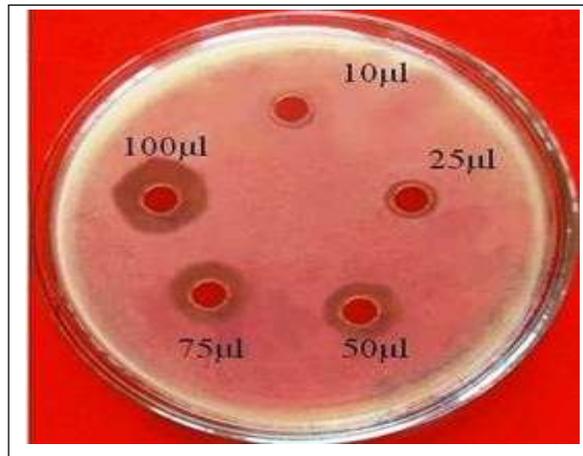


Figure 6: Antibacterial activity against *Bacillus subtilis*

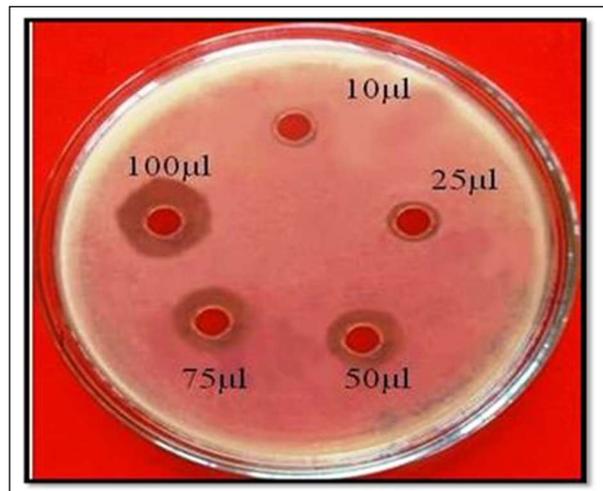


Figure 7: Antibacterial activity against *Staphylococcus aureus*

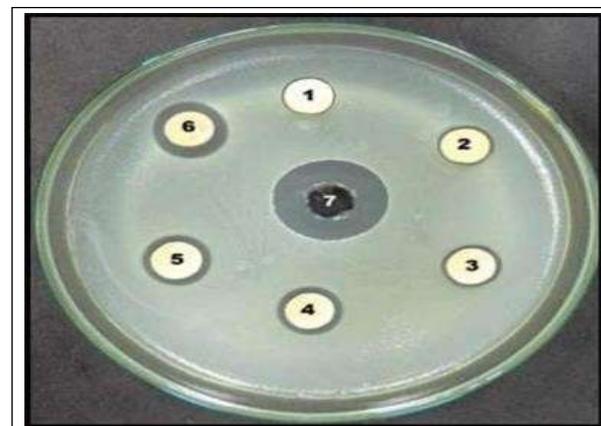


Figure 8: Antibacterial activity against *Pseudomonas aeruginosa*

**Experimental Conditions:**

**Method used:** Agar well diffusion and Agar disk-diffusion method.

**Organisms used:** *Staphylococcus aureus* (ATCC: 6538), *Bacillus subtilis* (ATCC: 10231), *Pseudomonas aeruginosa* (ATCC:9027)

**Media used:** Nutrient agar media.

**Test used:** Polyherbal nanogel formulation F1, F2, F3.

**Standard:** Streptomycin

**Solvent:** 10% DMSO

**Table 8: Antimicrobial activity showing zone of inhibition**

Sr.no	Name of Bacteria	Zone of inhibition in mm					
		Sample					
		F1	F2	F3	Standard	Solvent control	Standard Used
1.	<i>StaphylococcusAureus</i>	1.5	6	5	10	Nil	Streptomycin
2.	<i>Bacillus subtilis</i>	1.5	8	5	10	Nil	Streptomycin
3.	<i>Pseudomonas aeruginosa</i>	2	14	10	12	Nil	Streptomycin

Formulation F1 bacterial plate containing *Staphylococcus aureus* shows 1.5mm zone of inhibition, the plate containing *Bacillus subtilis* shows 1.5mm zone of inhibition and bacterial plate containing *Pseudomonas aeruginosa* shows 2mm zone of inhibition.

Formulation F2 bacterial plate containing *Staphylococcus aureus* shows 6mm zone of inhibition, the plate containing *Bacillus subtilis* shows 8mm zone of inhibition and bacterial plate containing *Pseudomonas aeruginosa* shows 14m zone of inhibition.

Formulation F3 bacterial plate containing *Staphylococcus aureus* shows 5mm zone of inhibition, the plate containing *Bacillus subtilis* shows 5mm zone of inhibition and plate containing *Pseudomonas aeruginosa* shows 10mm zone of inhibition. Hence,

observed that batch F2 have more inhibition than F1 and F3.

**CONCLUSION:**

A number of medicinal plants have been tested for their antibacterial activity as a result of the increasing failure of therapeutic agents due to antibiotic resistance by pathogenic microbes.

The prepared formulations tested for antibacterial efficacy against selected microorganisms. The results demonstrate that a polyherbal nanogel formulation shows good results against *Staphylococcus aureus* as compared to *Bacillus subtilis* and *Pseudomonas aeruginosa*. The antimicrobial activity of nanogel (F2) shows highest zone of inhibitions against *Staphylococcus aureus* as compared to *Bacillus subtilis* and

*Pseudomonas aeruginosa*. Our optimized formulation is F2 based on the zone of inhibitions obtained from the antibacterial investigation and the findings from the physicochemical evaluation of the polyherbal gel formulation with a pH of 6.9, good homogeneity, spread ability, and extrudability, the optimized formulation performs well. F2 has a measured viscosity of 4899 cP. The zones of inhibition recorded against *Bacillus subtilis*, *Pseudomonas aeruginosa* and *Staphylococcus aureus* were 6 mm, 8 mm, and 14 mm, respectively.

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