



ISSN (E): 2320-3862  
ISSN (P): 2394-0530  
NAAS Rating 2017: 3.53  
JMPS 2017; 5(5): 300-302  
© 2017 JMPS  
Received: 10-07-2017  
Accepted: 11-08-2017

**Shweta Padher**  
Department of Pharmaceutics,  
AISSMS College of Pharmacy,  
Pune, Maharashtra, India

## ***In-vitro* skin permeation and biological evaluation of Gingerol and Piperine cream**

**Shweta Padher**

### **Abstract**

Rheumatoid Arthritis (RA) is a chronic, inflammatory autoimmune disease causing synovial proliferation and joint destruction. Piperine and Gingerol have been reported to be effective in reduction of Tumor Necrosis Factor-TNF- $\alpha$ , the key inflammatory mediator in RA and have an action on synovial fibroblasts. Essential oils from similar families of oils; such as the florals, herbs, citrus and those that have some similar constituents will normal work together well. Synergy, as used in aromatherapy, is the working together of two essential oils that result in an effect greater than the sum of their individual effects. Thus topical formulation i.e cream containing essential oils containing piperine and gingerol were prepared and evaluated. These formulations showed high retention (60-72%) at the site of action i.e. the dermis, where the fibroblasts are located which was demonstrated by ex-vivo permeation studies. The drug was topically applied to the tibiotarsal joint to evaluate the paw volume was measured up till 28th day, there was less paw edema as measured by plethysmometer.

**Keywords:** Rheumatoid arthritis, polyherbal, cream, synergism

### **Introduction**

Rheumatoid arthritis (RA) is a chronic, progressive inflammatory disease of the joints characterized by synovial proliferation as well as inflammatory and immunological processes which lead to irreversible changes in the articular cartilage and juxta-articular bone, commonly affecting wrists, fingers, knees, feet, and ankles. (Marie-Christophe Boissier 2012) [5] NSAIDs are used as first choice drugs for RA. However, since these agents alone do not change the course of the disease or prevent joint destruction drugs belonging to the class of DMARDs (disease modifying anti-rheumatoid drugs) are often given in low doses as first-line therapy in combination with NSAIDs to treat the disease early and aggressively. Intervening early in the treatment with DMARD agents has shown to alter the disease course and improve radiographic outcomes. Synovial fibroblasts which play an important role in triggering inflammatory reactions in RA, reside in the dermis. Many phytoconstituents such as Piperine and 6-Gingerol are reported to possess DMARD action (Shivaprasad H 2011, A. Subramoniam 2013) [6, 1] due to TNF  $\alpha$  inhibition and prevention of proliferation of synovial fibroblasts (Bhalekar MR 2015, Subhash Yende 2010, Vinod K.R 2011, Janet L. Funk 2016) [2, 8, 9, 4] Topical drug delivery of various anti-rheumatoid agents such as corticosteroids, NSAIDs has widely reported, recently we have reported topical delivery of chloroquine (Yasuka Isa 2008) [10]. This is because of the fact that the fibroblasts which regulate the TNF  $\alpha$  secretion and reside in dermal layers can be inhibited by topical administration and it saves the systemic load of the drug. The objective of present work is to formulate and evaluate topical formulation i.e cream of phytoconstituents. Piperine and 6-Gingerol so as to achieve localization at the site of inflammation thereby increasing the effectiveness of the therapy. Apart from performance properties such as pH, spreadability the ex vivo permeation was studied and the polyherbal formulation was demonstrated to exhibit significantly higher accumulation in dermal layers. These formulations were further studied in a CFA- induced pharmacodynamics model in rats and evaluated using paw edema. The topical treatment demonstrated significant reduction in paw-edema compared to standard treatment (chloroquine dispersion)

### **Results & Discussion**

#### **Measurement of hind paw volume**

Paw volumes of both the hind limbs were recorded from day 7 to day 28 at 4-day interval using plethysmometer.

### **Correspondence**

**Shweta Padher**  
Department of Pharmaceutics,  
AISSMS College of Pharmacy,  
Pune, Maharashtra, India

A significant reduction in the paw volume was seen in polyherbal cream treated group when compared to the individual cream treated group and arthritic control indicating enhanced action due to synergism. Statistical analysis by two-way ANOVA followed by Bonferroni multiple comparison test using Graph Pad Prism Software, \*P < 0.05, \*\*P < 0.01 and \*\*\*P < 0.001 attributed all values to be significant.

#### Stability data for creams

The cream was evaluated for appearance, pH and particle size. It is evident that the formulation did not show any sign of instability and was stable over the testing period

#### Drug content

The drug content of the formulations was found to be in the range of 98% to 99%. Hence uniformity of drug content was found to be satisfactory.

#### Viscosity

Viscosity values for creams are totally attributable to the concentration of cetyl alcohol.

#### Spreadability

The observations for spreadability of all formulations are listed in table 2. The spreadability of the formulations is a characteristic derived from its more basic property i.e. viscosity. The greater the viscosity, longer is the time taken for spreading. In the study it was found that as changes in spreadability time are proportional to the changes in viscosity for different formulation.

#### Ex-vivo permeation study

*Ex-vivo* permeation study data is presented in table 2. Since the globules were localized in the dermis, the % drug in receptor medium was to a lesser extent. The amount of drug remaining on the surface is that, that is either not yet released from the gel base or un-diffused. The production of interleukin-1(IL-1) and tumor necrosis factor-(TNF- $\alpha$ ) by monocytes is central to the inflammatory process. In particular, IL-1 is responsible for stimulating prostaglandin E<sub>2</sub>, while TNF- $\alpha$  is a key component in activating matrix proteinases. The globules get entrapped in the dermal layer of skin from which the drug is released and the drug moiety is proposed to be acting on the fibroblasts residing in the dermis which plays a vital role in triggering inflammation and subsequent bone and cartilage damage. This is the rationale behind localizing the drug in the dermal layers of the skin. The results clearly indicate that % drug localized in skin viscosity are strongly related. Piperine being bio-enhancer enhances the diffusion of gingerol in the polyherbal cream as compared to plain gingerol cream.

#### Experimental (Harry Ralph)

Black pepper oil and ginger oil were purchased from R.K Aroma, Mumbai. Cetyl alcohol, Sodium lauryl sulphate, methyl and propyl paraben from Cosmo chemical, Pune Cetyl alcohol and glyceryl monostearate and oil blend were melted at 60 °C to form oil phase. Propyl paraben was added in the same phase. Sodium lauryl sulfate and methyl paraben was dissolved in water heated at 60 °C. The phases were stirred together till cream was formed. Individual creams (with one essential oil at a time) with polyherbal cream (containing blend of essential oils) were prepared.

**Appearance:** All formulations were observed for appearance, color and consistency.

**pH determination:** pH of all formulations was determined by using pH meter. 1% dispersion formulation was stirred in distilled water and pH of the dispersion was measured.

**Drug content evaluation:** % drug content was calculated by HPTLC.

**Viscosity:** Brookfield digital viscometer, equipped with a ULC adapter and spindle S-92 was used to determine viscosity (cp) of the formulations. The viscosity was measured at 10 rpm after 30 seconds. Measurements were performed at ambient temperature and in triplicate.

**Spreadability:** The spreadability of the formulation was determined using an apparatus Texture analyzer CEB Texture analyzer, Brookfield Engineering Labs, Inc., Model Texture Pro CT V1.4 Build 17.

**Ex-vivo permeation studies:** Rat skin was prepared by cleaning any soil from the surface with a mild skin cleanser, removing any hair with clippers and removing subdermal fat and fascia. The excised rat skin after hydrating in water for 1 hour was mounted on the Franz diffusion cell (with effective diffusion area 3.14 cm<sup>2</sup> and 7 ml cell volume) with stratum corneum facing upwards. The receptor compartment was filled with phosphate buffer pH 7.4, and the assembly was maintained at 37°C  $\pm$  0.5 under constant magnetic stirring. With reference to SUPAC (Scale-Up and Postapproval Changes) guidelines laid by FDA, 300 mg of cream was applied to the membrane on the donor compartment and then covered with aluminum foil to prevent drying out. Aliquots were withdrawn at predetermined time intervals over a period of 8 hours and analyzed by HPTLC method (Shrimanker Mitali 2012)<sup>[7]</sup>.

**Pharmacodynamic study:** The pharmacodynamic study was carried out on Male Wistar rats weighing 180-220 gm. The experimental protocol was approved by the Institutional Animal Ethical Committee constituted as per the rules of the Committee for the Purpose of Control and Supervision of Experiments on Animals, India (CPCSEA/IAEC/PT-02/02-2K16).

**Induction of arthritis:** Arthritis was induced by a single injection of 0.1ml Complete Freund's Adjuvant (CFA) containing heat killed and dried Mycobacterium tuberculosis into subplantar region of the right hind paw on day one. Animals were divided into six groups with six rats in each group. Treatment was initiated after the onset of arthritis (day 7) and continued once daily until the 28th day of the experiment. The standard group was treated with oral dose of chloroquine (dose equivalent to 15mg).

**Measurement of hind paw volume:** Paw volumes of both the right hind limbs were recorded from day 7 to 28 at 4-day interval using water displacement plethysmometer.

**Stability studies:** Formulations were subjected to accelerated stability study at 40 °C 75% RH for 3 months. After completion of stability testing period, test samples were analyzed for appearance, particle size and pH.

**Table 1:** Experimental protocol for pharmacodynamics studies

Groups (n=6)	Treatment	Dose
Group I	Positive control	-
Group II	Arthritic control	-
Group III	Arthritic animals treated with standard	Solution of Chloroquine phosphate equivalent to 15 mg
Group IV	Arthritis animals treated with Polyherbal Cream	Applied topically to the joints
Group V	Arthritis animals treated with Piperine cream	Applied topically to the joints
Group VI	Arthritis animals treated with Gingerol cream	Applied topically to the joints

**Table 2:** Evaluation parameters for cream

	Viscosity (cp)	Hardness (g)	% drug localized in skin	% drug in receptor medium
Polyherbal cream	3050	51	Piperine-60.2 Gingerol-59.2	Piperine-10.616 Gingerol-13.385
Piperine cream	2500	53.3	79.75	4.526
Gingerol cream	2450	64	81.001	4.32

### Conclusions

Topical preparations which would bypass gastrointestinal contact and exhibit a potential for reducing systemic toxicities were developed. Topical cream systems were developed and characterized. Formulation containing the essential oil blend showed the highest retention of drug in the dermal layers of skin where the fibroblasts are located. To evaluate the arthritic protective potential, the paw volume was measured up till 28 days. It was observed that the polyherbal cream formulation showed a greater protective potential than the individual creams and standard. Thus the polyherbal cream was effective in controlling inflammation associated with arthritis due to synergistic action of the two oils. Thus it was concluded that further development in the formulation prospects can yield a better product for clinical investigation.

### References

- Subramoniam V, Madhavachandran A. Gangaprasad: Annals Medicinal plants in the treatment of arthritis of Phytomedicine. 2013; 2(1):3-36.
- Bhalekar MR, Upadhaya PG, Nalawade SD, Madgulkar AR, Kshirsagar SJ. Anti- rheumatic activity of Chloroquine-SLN gel on wistar rats using complete Freund's adjuvant (CFA) model. Ind j rheumatol. 2015
- Harry Ralph. Harry's Cosmeticology'.The principles & practices of modern cosmetics. Sixth edition, 54-55.
- Janet Funk L *et al.* Anti-inflammatory effects of the essential oils of ginger (*Zingiber officinale* Roscoe) in experimental Rheumatoid Arthritis, Pharma Nutrition. 2016; 4(3):123-131.
- Marie-Christophe Boissier *et al.* Rheumatoid arthritis: From autoimmunity to synovitis and joint destruction Journal of Autoimmunity. 2012; 39:222-228.
- Shivaprasad H, Venkatesha Brian M, Berman Kamal D. Moudgil: Herbal medicinal products target defined biochemical and molecular mediators of inflammatory autoimmune arthritis. Bioorganic & Medicinal Chemistry. 2011; 19:21-29.
- Shrimanker Mitali *et al.* Hptlc Quantification Method of Piperine and Zingiberene in A Veterinary Polyherbal Formulation International Journal of Current Research. 2012; 4(12):477-479.
- Subhash Yende R *et al.* Antirheumatoid activity of aqueous extract of *Piper longum* on freunds adjuvant-induced arthritis in rats. Indian journal of pharmaceutical and scientific research. 2010; 1(9):129-133.
- Vinod KR, Santhosha D. Formulation and evaluation of piperine cream a new herbal dimensional approach for vitiligo patients. International Journal of Pharmacy and Pharmaceutical Sciences. 2011; 3:29-33.
- Yasuka Isa *et al.* 6-Shogaol and 6-gingerol, the pungent of ginger, inhibit TNF-a mediated downregulation of adiponectin expression via different mechanisms in 3T3-L1 adipocytes. Biochemical and Biophysical Research Communications. 2008, 429-434.