ORIGINAL RESEARCH PAPER

Volume-8 | Issue-3 | March-2019 | PRINT ISSN No 2277 - 8179

INTERNATIONAL JOURNAL OF SCIENTIFIC RESEARCH

# PREPARATION AND EVALUATION OF DICLOFENAC SODIUM NANOPARTICLES FOR OCULAR DRUG DELIVERY

Pharmaceutical

**G.Harini Kumari** Asst Professor, East Point College of Pharmacy,

**V. Chandrakala*** Professor, East Point College of Pharmacy, *Corresponding Author

## ABSTRACT

Diclofenac sodium (DS) nanoparticles were prepared by using a polymer polycaprolactone (PCL) at different concentration and combinations using polaxmer 188 as stabilizing agent. The DS nanoparticles were prepared by using Double Emulsion Solvent Diffusion Technique. The prepared nanoparticles were evaluated by Entrapment efficiency and morphology of DS-PCL particles were studied using TEM. The *in vitro* release profile of DS nanoparticles containing different ratios of PCL were studied and the release rate were studied. NP1 consist of DS (20mg), PCL(1%), Ethyl acetate(3ml), polaxmer188 (1or2 drops) and polyvinyl alcohol(10ml) was selected as the optimized formulation with sufficient drug-polymer ratio and *in-vitro* correlation. Nanoparticles of the optimized formulation provided an effective drug concentration at the site of action for appropriate period of time to improve the ocular bioavailability of drug by increasing the pre-corneal residence time.

## KEYWORDS

Double Emulsion Solvent Diffusion Technique, Entrapment Efficiency, poly dispersity index

## INTRODUCTION

Topical application of drugs to the eye is the well established route of administration for the treatment of various eye diseases like dryness, conjunctiva, eye flu etc. which can be affected to the eye and also eye vision. The protective mechanisms of the eye such as blinking, drainage and reflux lacrymation decrease the bioavailability of drug and also help to remove rapidly foreign substances from the surface of eye [1]. The marketed ophthalmic preparations such as drops and ointments when instilled into eye are rapidly drained away from the ocular surface due to blinking, tear flow and lachrymal nasal drainage of the eye. Only small amount of the drug is available for its therapeutic effect resulting in frequent dosing application to the eye. So to overcome these problem newer pharmaceutical ophthalmic formulation have been developed in the last three decades to increase the bioavailability of the drug [2-9].

Nanoparticles are solid colloidal particle with diameters ranging from 1 to 1,000nm. Nanoparticles have excellent tissue penetration and persistence for drug delivery, more targeted dosing and longer intervals between doses are beneficial to patients [10,11].

In ocular drug delivery system, the main problem is extensive loss of drug and to overcome from this we need an optimize ocular drug delivery system by prolonged contact time of drug and appropriate reological properties and concentration of viscolyser [12,13]. The main advantages of controlled ocular drug delivery system is to provide targeting within the ocular globe, so as to prevent the loss to other ocular tissues and improve therapeutic performance of drug.

## MATERIALAND METHODS:

The drug was obtained as gift sample from Devi Laboratories, Ltd, Hyderabad, India. Polymer were gifted by Sigma Aldrich chemicals, USA and Polaxmer from Merck Specialties Private Limited and all other chemicals used were of analytical grade.

**Differential scanning calorimetry (DSC):** Thermo gram of the drug was taken by using DSC (Model TA-60 WS shimadzu). 4 mg of the Drug sample and Drug + Polymer mixture was weighed on an electronic balance. The sample was placed on the aluminum pans and then sealed. The instrument was calibrated using indium. On one side the sample pan was placed and on the other side an empty aluminum pan was placed. The sample was heated between 50- 200°C at the rate of 10°C/Minute. Nitrogen gas was introduced at a pressure of 2 bars and a flow rate of 50 ml/minute maintained. The DSC of drug and drug polymer mixture is given in figure 1.

**Preparation of standard curve**

100mg of the drug was accurately weighed and transferred to 100ml volumetric flask. The volume was made up with simulated tear fluid. Ten milliliters of this solution was pipette out and transferred to another 100-ml volumetric flask. The volume was made with simulated tear fluid resulting in to stock solution 100 µg per ml. Different volumes of the stock solution were transferred to different 10-ml volumetric flasks and volumes made up with simulated tear fluid to get concentration of 1, 2, 3, 4, 5, 6, 7, 8, 9 and 10µg per ml respectively. The absorbance's of the resulting different concentrations of the drug solutions were measured at 276 nm against simulated tear fluid as blank using UV- Visible double beam spectrophotometer.

**Preparation of nanoparticles:**[14,15]

Nanoparticles were prepared by double emulsion solvent diffusion technique. 1ml of 20mg/ml of drug solution is suspended in 3ml of Ethyl acetate containing polycaprolactone(NP1-1%,NP2-1.5%,NP3-2%) and polaxmer 188, by sonication using probe sonicator at 20 % amplitude for 90 seconds. The so prepared primary emulsion was then added in to10ml polyvinyl alcohol solution and was sonicated for 90 seconds at 50 % amplitude. Organic solvent was evaporated by stirring the emulsion on magnetic stirrer over night with constant magnetic stirring speed of 1500 rpm approximately. The resulting nanoparticles dispersion was collected and subjected for purification and concentration.

**Purification and resuspension of nanoparticles:**

The suspension was washed by adding 10ml of doubled distilled water and subjecting to centrifugation. Nanoparticles were washed for 3 times. Purified nanoparticles were taken and again re-suspended in purified water containing 0.5% polyvinyl alcohol. The so formed purified nanoparticles were used for further studies.

**Physicochemical characterization:**

**Determination of drug entrapment efficiency:** The drug Entrapment efficiency for the free drug content was determined spectrophotometrically by measuring the absorbance at 276nm. This is done by measuring the non-entrapped drug in the external aqueous solution. In case of nanoparticles the external aqueous solution was obtained after the centrifugation and purification of the colloidal suspension in the centrican tubes at 1800 rpm for 20 minutes. The entrapment efficiency of nanoparticles was determined by subtracting free drug amount from initial added amount of drug. The entrapment efficiency (EE %) could be calculated by following equation.

$$\text{Entrapment efficiency(EE \%)} = \frac{\text{initial drug} - \text{free drug}}{\text{Initial drug}} \times 100$$

**Particle size and Morphology :**

The mean particle size for the formulations was determined by photon correlation spectroscopy (PCS) with a Zetasizer Nano ZS-90 (Malvern Instruments Ltd, Worcestershire, UK). The reading was carried out at 90° angle to the incident beam at 25°C using proper diluted with filtered water (0.5 micrometer filter). All experiments were done in triplicate.

***In-vitro drug release*:**

The *in vitro* drug release study of nanosuspension was performed in modified dissolution apparatus containing a two sided open glass

cylinder. A pre-soaked dialysis membrane (Himedia, Mumbai) was adapted to the terminal portion of glass cylinder. DS nanosuspension was accurately placed into glass cylinder from open side and fixing this cylinder to the burette stand. The cylinder was suspended in 200 ml dissolution simulated tear fluid (pH 7.4) medium maintained at 37 $^0$C ± 5$^0$C at 100 rpm, so that the dialysis membrane fixed cylinder end just touched to the receptor medium surface. The samples were withdrawn at specified time intervals with volume replacement. The withdrawn samples were analyzed after proper dilution using simulated tear fluid pH 7.4 for drug content by measuring absorbance at 276 nm in UV/visible spectrophotometer. All the experiments were conducted in triplicate.

## RESULTS AND DISCUSSION:

**DSC analysis** DSC thermographs of the physical mixture of the drug along with polymer and excipients have been shown in figures (Fig 1A and 1B). The DSC analysis of the physical mixture of pure drug shows melting point at 284.31$^0$C, and polymer drug melting point was 56.11 & 287$^0$C respectively.

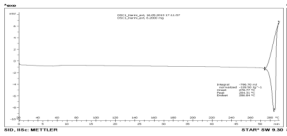


**Fig 1A: DSC sample for DS**

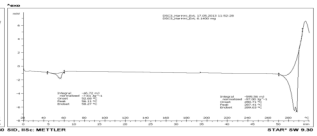


**Fig 1B: DSC sample for drug DS and PCL**

### Preparation of standard curve

The standard curve was obtained by plotting absorbance vs. concentration of drug. The standard calibration curve obeyed Beer's law at the given concentration range of 0 to 10mg/ml in tears fluid pH 7.4. The value of regression coefficient was found to be 0.997 which showed linear relationship between absorbance and concentration.

### Entrapment Efficiency:

Np1 formulation showed maximum entrapment compared to NP2 and NP3. The ratio of drug and polymer was equal to 1, hence NP1 exhibited maximum drug encapsulation (table 1)

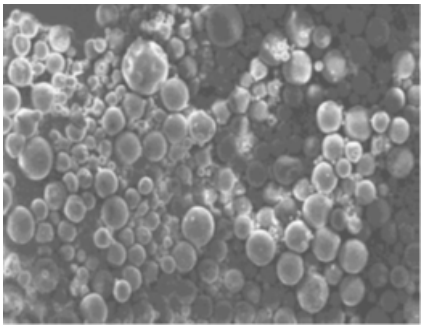
**Table1: Entrapment efficiency, Particle size and % Drug Released for formulations**

| S.NO | FORMULATION CODES | EE+ S.D | Particle size+S.D | % Drug Release (6hrs) |
|---|---|---|---|---|
| 1 | NP1 | 75.63+ 0.08 | 62.90+ 2.16 | 89.52 |
| 2 | NP2 | 72.74+ 0.14 | 66.44+ 0.49 | 92.94 |
| 3 | NP3 | 73.14+ 0.07 | 74.83+ 0.16 | 84.9 |

### Particle size and Morphology :

The morphology of these DS-PCL particles were spherical structures as resolute by using Transmission electron microscope (TEM). Figure 2 is the structure of DS-PCL particles . The surfaces of the particles were rough and rounded. In this studies by maintaining the pH, uniform particles are formed in the nanoparticles. Particle size for various formulations were reported in table 1

**Fig 2: Surface structure of Diclofenac sodium and PCL**



### *In-vitro release profile:*

The *in vitro* results for three formulations are given in figure 2. The amount of drug released in 2 hrs for the formulations were 83.87 % for 1%, 49.04 for 1.5% and 46.79 for 2%. 85.78 % for 1%, 67.11 for 1.5% and 65.76 for 2% of the drug in 4 hrs and 89.52% for 1%, 92.94% for 1.5% and 84.90% for 2% of the drug in 6 hrs.

## CONCLUSION

Double emulsion solvent diffusion technique was able to engineer Diclofenac sodium(DS) nanoparticles to reach the target size. The smallest size was obtained by applying sonication method. On the other hand, agitation speed of homogenization depend on dispersing agent. effect of increasing dispersing agent content, in contrast to literatures, caused bigger particle size. In terms of phase volume, increasing continuous phase volume caused smaller particle size. The formulation NP1 was selected as the optimized formulation with sufficient drug-polymer ratio and *in-vitro* correlation.

It was concluded that preparation of nanoparticles for ocular drug delivery was one of the alternative routes of administration to avoid first pass effect and improve the bioavailability of diclofenac sodium and enhance the release of drug for extended period of time. In addition, these formulation reduce the need of frequent administration and enhance patient compliance

## REFERENCES


1. Lang J C. Ocular drug delivery conventional ocular formulation. Advanced drug delivery review 1995;16:39-43.
2. K Basavaraj, Nanjawade, Manvi FV and Manjappa AS: In situ-forming hydrogels for sustained ophthalmic drug delivery. Journal of Controlled Release2007; 122: 119–134.
3. Sahoo KS, fahima SAD, kumar K: Nanotechnology in ocular drug delivery, Drug delivery today 2008; 13: 144-151.
4. Weidener J: Mucoadhesive ocular inserts as an improved delivery vehicle for ophthalmic indications. Drug Discovery Today 2003; 8: 906– 907.
5. Lang JC, Roehrs RTE and Jani R: Ophthalmic preparations, Edition 21, vol-1; Lippincott Williams and Wilkins, 2005.
6. Andrews GP, Laverty TP and Jones DS: Mucoadhesive polymeric platforms for controlled drug delivery Review article. European Journal of Pharmaceutics and Biopharmaceutics 2009; 71: 505–518.
7. Alany RG, Rades T, Nicoll J, Tucker IG and Davies NM: W/O microemulsions for ocular delivery: Evaluation of ocular irritation and precorneal retention.Journal of Controlled Release 2006; 111: 145-152.
8. Binstock EE and Domb AJ: Iontophoresis: A non-invasive ocular drug delivery. Journal of Controlled Release 2006; 110: 479–489.
9. de la Fuente M, Raviña M, Paolicelli P, Sanchez A, Seijo B,Alonso MJ. Chitosan-based nanostructures: A deliveryplatform for ocular therapeutics. Adv Drug Deliv Rev 2010;62:100-17.
10. Bucolo C, Maltese A, Drago F. When nanotechnology meetsthe ocular surface. Expert Rev Ophthalmol 2008; 3:325-32.
11. Wang X, Zheng C, Wu Z, Teng D, Zhang X, Wang Z, Li C.Chitosan-NAC nanoparticles as a vehicle for nasal absorptionenhancement of insulin.Biomed Mater Res B ApplBiomater 2009; 88:150-61.
12. David quintanar-Guerrero,Eric allemannn,Hatem fessi and Eric doelker devolped Preparation techniques and mechanisms of formation of biodegradable nanoparticles from preformed polymers.1998,vol.24,No12,Pages1113-1128.
13. Yi-yan yang,Tai-shung chung,Ngee ping Ng;Fabricated drug distribution and in vitro release profiles of biodegradable polymeric microspheres containing protein fabricated by double emulsion solvent extraction/evapouration method.Institute of material engineering,No3 research link,National university of Singapore.
14. Medina C, Santos-Martinez MJ, Radomski A, Corrigan OI,Radomski MW. Nanoparticles: pharmacological andtoxicological significance. Br J Pharmacol 2007; 150:552-8.
15. Sahoo SK, Dilnawaz F, Krishnakumar S. Nanotechnology inocular drug delivery. Drug Discov Today 2008; 13:144-51.