# *IN VIVO* EVALUATION OF DEXAMETHASONE SODIUMPHOSPHATE NANOPARTICLES FOR POST CATARACT TREATMENT

V.Sankar[a]*, V. Harikrishnan[a], L. Madhura keerthi[a], K.G .Prasanth[a,]
D.Sundar[b], K.Ruckmani[c]

[a] PSG College of Pharmacy, Coimbatore, Tamilnadu
[b] PSG Hospital, Coimbatore, Tamilnadu
[c] Anna University, Tiruchirapalli, Tamilnadu

**Summary**

Dexamethasone sodium phosphate (DSP) nanoparticles were prepared by solvent evaporation process. The prepared nanoparticles were evaluated for drug content uniformity. The drug content uniformity was high in 0.2% methyl cellulose (MC) in 1% sodium alginate (SA) gel & Poly (D, L-lactide-co-glycolide) (PLGA) in 3% SA gel. The amount of drug present in the aqueous humor of New Zealand rabbits after 2 hours of instillation for formulation 0.2% MC in 1% SA gel & Poly (D, L-lactide-co-glycolide) PLGA in 3% SA gel was found to be 75 % and 85%. Hence these ophthalmic gels may be viable alternative to conventional eye drops as it offers increased contact time, decreased frequency of administration and thus improved patient compliance.

**Keywords:** Dexamethasone sodium phosphate, Nanoparticles, Methylcellulose (MC), Poly (D, L-lactide-co-glycolide) (PLGA), Sodium alginate gel (SA).

* Corresponding Author.

## Introduction

Most ocular diseases were treated with topical applications of solutions administered as eye drops. The major deficiencies of this conventional dosage form include poor ocular drug bioavailability, pulse drug entry, systemic exposure due to the nasolacrimal duct drainage and poor entrance to the posterior segments of the eye due to the lens diaphragm. Poor ocular drug bioavailability is the result of ocular anatomical and physiological constrains, which protect the eye and maintain visual functions. After instillation of an ophthalmic drug, most of it is rapidly eliminated from the pre corneal area due to drainage by the nasolacrimal duct, blinking and dilution by the tear turnover (approximately 1µl/min). It has been determined that as much as 90% of the 50 ml dose administered as eye drops is cleared within 2 minutes and only 1-5% of the administered dose permeates to the eye[1]. After instillation, the flow of lachrymal fluid removes instilled compounds from the surface of the eye. Even though the lachrymal turnover rate is only about 1 µl/min the excess volume of the instilled fluid is flown to the nasolacrimal duct rapidly in a couple of minutes. Another source of non-productive drug removal is its systemic absorption instead of ocular absorption. Systemic absorption may take place either directly from the conjuctival sac via local blood capillaries or after the solution flow to the nasal cavity. Anyway, most of small molecular weight drug dose is absorbed into systemic circulation rapidly in few minutes. This contrasts the low ocular bioavailability of less than 5%. Drug absorption into the systemic circulation decreases the drug concentration in lachrymal fluid extensively[2]. Cataract is a degradation of the optical quality of the crystalline lens. The development of cataract is therefore a continuum, extending from minimal changes of original transparency in the crystalline lens to the extreme stage of total opacity [3]. Novel drug delivery system aims to deliver the drug at a rate directed by the needs of the body during the period of treatment. At present, no available drug delivery system behaves ideally achieving lofty goals, but sincere attempts have been made to achieve them through novel approaches in drug delivery systems. In the treatment of postoperative cases of cataract, instillation of eye drops containing corticosteroids with antibiotics, for every hour installation of medicaments is one of the major draw back for getting compliance from the patient. It needs nursing care. To overcome the above drawbacks we wish to formulate an ophthalmic preparation containing Dexamethasone sodium phosphate nanoparticles for the Post cataract treatment with the objectives to enhance contact time of drug in eye, to enhance the bioavailability to corneal epithelium, to provide a sustained action, to reduce the dosing frequency, to improve the patient compliance. Dexamethasone sodium phosphate is a crystalline corticoid that has been used for the treatment of post cataract treatment administered as eye drops. The goal of this work was to formulate and optimize dexamethasone sodium phosphate nanoparticles in gel form by using (1% & 3%) sodium alginate for post cataract treatment.

## Materials and Methods

### Materials

Methyl cellulose (MC), Poly (vinyl alcohol) (PVA), Sodium alginate (SA), Poly (D, L-lactide-co-glycolide) (PLGA) (85:15), Phosphotidyl Choline, Dexamethasone sodium phosphate (DSP), Dichloromethane, Benzalkonium chloride, Polysorbate 80, EDTA.

### Solvent Evaporation method

The particulate solution was prepared by solvent evaporation method. Dexamethasone sodium phosphate (0.1% by weight) was added to dichloromethane and sonicated for 3 mins (PCI, 50 Hz, Chennai). The organic phase was added to corresponding aqueous phase like Methylcellulose (0.1 - 0.4 %), or Poly (vinyl alcohol) PVA (0.25%), then magnetically stirred (Remi instruments, Mumbai) at 1200 rpm at room temperature to evaporate dichloro methane (about 4 h). The particulate solution was obtained [4, 5].

### Dispersion of particulate solution in gels

The ophthalmic particulate solutions were taken and mixed with the specified quantity of Sodium Alginate (1% or 3%) correspondingly, and triturating was continued for 1 hour till the particulate solution was dispersed and to get a gel consistency [6].

### Drug content Uniformity

The vials containing the preparations were shaken for a few minutes and 100 µl of the preparations were transferred to 25 ml volumetric flasks using a Micro Pipette. Phosphate buffer (pH 7.4) was added in small portions (5 ml), shaken to dissolve the contents, volume was adjusted to 25 ml, and the solutions were assayed for Dexamethasone sodium phosphate content at 242 nm [7,8].

$$\text{Drug Content Uniformity} = \frac{\text{Concentration} \times 1 \times \text{Dilution Factor}}{1000}$$

### *In vivo* Studies

The animal ethical committee (PSG Institute of Medical Sciences and Research, Reg no: 158/ 1999/ CPCSEA, dated on 5th Jan 2009) had given permission for the *in vivo* study. Twelve male healthy albino rabbits weighing 1.5 to 2 kg each were divided into 3 groups. The animals with any ocular abnormalities were excluded after thorough examination. The animals were housed in individual cages with husk bedding and were fed with rodent pellet diet and water as much as required. The rabbits were anesthetized with intramuscular injection of Ketamine (40 mg/kg) [9].To the first group drug in solution was applied in the left eye. To the second group 0.2% Methyl cellulose (MC) in 1% sodium alginate was applied in the left eye and for the third group Poly (D, L-lactide-co-glycolide) (PLGA) in 3% sodium alginate was applied in the left eye. Aqueous humor (50 µl) was collected by using a syringe connected to a 30- Gauge needle in the sclero-corneal limbus [10]. The aqueous humor was collected at every 30 mins interval for 2 hour. The absorbance of the withdrawn sample was measured after suitable dilution with phosphate buffer and assayed for drug content by UV spectrophotometer method at 242 nm.

## Results and discussion

### Formulation of ophthalmic particulate solution

The ophthalmic nanoparticulate formulation was formulated using solvent evaporation method. The particles in the particulate solutions were found to be good and visible in the formulation prepared with 0.2% Methyl cellulose (MC), and Poly (D, L-lactide-co-glycolide). Hence, these formulations were selected for the further investigations.

Table1: Formulation of Nanoparticule Solution with Methyl Cellulose

| S.No | INGREDIENTS | METHYL CELLUOSE | | | |
|---|---|---|---|---|---|
| | | **0.1%** | **0.2%** | **0.3%** | **0.4%** |
| 1. | Dexamethasone Sodium Phosphate | 0.01gm | 0.01gm | 0.01gm | 0.01gm |
| 2. | Methyl Cellulose | 0.01gm | 0.02gm | 0.03gm | 0.04gm |
| 3. | Sodium Edetate | 0.001gm | 0.001gm | 0.001gm | 0.001gm |
| 4. | Benzalkonium chloride | 0.001gm | 0.001gm | 0.001gm | 0.001gm |
| 5. | Polysorbate 80 | 0.005gm | 0.005gm | 0.005gm | 0.005gm |
| 6. | Phosphate buffer (qs) | 10 ml | 10 ml | 10 ml | 10 ml |

### Drug Content uniformity:

The drug content uniformity in 1% sodium alginate gel with Poly (D, L-lactide-co-glycolide) (PLGA) and 0.2% Methyl cellulose (MC) was found to be high as 87.57% and 77.36% & where as 3% sodium alginate gel with Poly (D, L-lactide-co-glycolide) (PLGA) and 0.2% Methyl cellulose (MC) was 75.27% and 79.17%.[Table 3]

Table 2: Formulation of Nanoparticule Solution with PLGA

| S.No | INGREDIENTS | PLGA |
|---|---|---|
| 1. | Dexamethasone Sodium Phosphate | 0.01 gm |
| 2. | PLGA | 0.025 gm |
| 3. | Phosphotidyl choline | 0.2 gm |
| 4. | Poly vinyl alcohol | 0.025 gm |
| 5. | Sodium Edetate | 0.001 gm |
| 6. | Benzalkonium chloride | 0.001 gm |
| 7. | Polysorbate 80 | 0.005 gm |
| 8. | Phosphate buffer (qs) | 10 ml |

Table 3: Drug Content Uniformity.

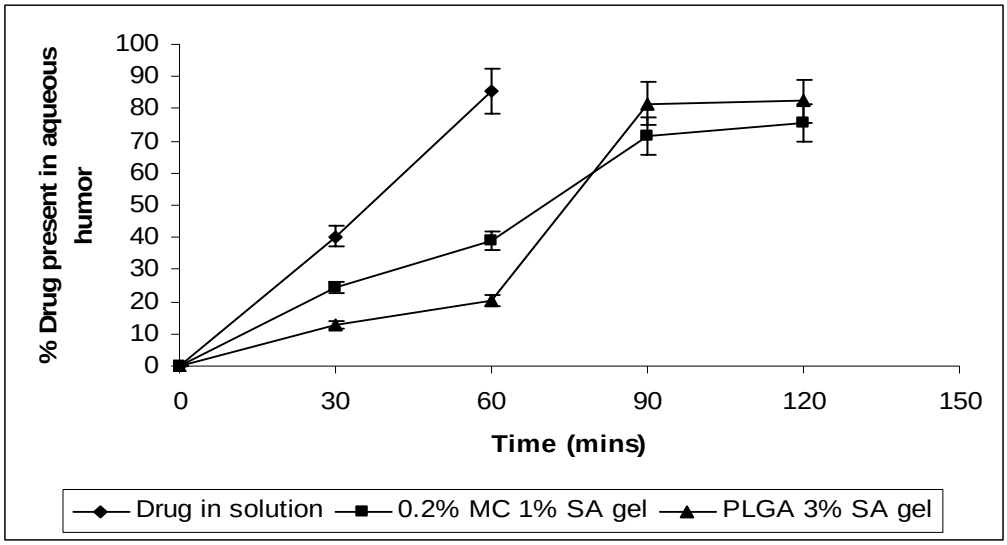
| S.No | GEL FORMULATION | CONTENT | % UNIFORMITY |
|---|---|---|---|
| 1. | 1% Sodium alginate gel | 0.2% MC | 77.36 |
| 2. | | PLGA | 87.57 |
| 3. | 3% Sodium alginate gel | 0.2% MC | 79.17 |
| 4. | | PLGA | 75.27 |

***In vivo* studies**

Percentage of drug present in the aqueous humor from drug in solution was 85.51% at 60 mins, where as 0.2% Methyl cellulose (MC) in 1% sodium alginate gel & PLGA in 3% sodium alginate gel was 75.53% & 82.42% at 120 mins. Both the formulations 0.2% Methyl cellulose (MC) in 1% sodium alginate gel and Poly (D, L-lactide-co-glycolide) (PLGA) in 3% sodium alginate gel were found to be effective in extending the drug release (Fig-1). The drug concentration in aqueous humor was found to increase significantly ($P < 0.001$) with drug in solution when compared with formulation 0.2% MC in 1% sodium alginate gel and Poly (D, L-lactide-co-glycolide) (PLGA) in 3% sodium alginate gel.

But when the formulations 0.2% Methyl cellulose (MC) in 1% sodium alginate gel and Poly (D, L-lactide-co-glycolide) (PLGA) in 3% sodium alginate gel were compared 0.2% Methyl cellulose (MC) in 1% sodium alginate gel was seemed to be significant ($P<0.001$) at 60 mins. . Sodium alginate 1% gel formulation with 0.2% Methyl cellulose (MC) was seemed to be the best formulation than Poly (D, L-lactide-co-glycolide) (PLGA) in 3% sodium alginate gel. The difference in drug release from the formulation may be due to the polymeric concentration in the particulate formulation, diffusion of drug particulates from the formulation into the gel and permeation of gel through the vitreous membrane.

Fig1: Comparative *In vivo* Release of Ophthalmic Gels




## Conclusion

In conclusion, ophthalmic gel formulation prepared with 0.2% Methyl cellulose (MC) in 1% sodium alginate and Poly (D, L-lactide-co-glycolide) (PLGA) in 3% sodium alginate gives promising results in drug content uniformity and *in vivo* studies. These ophthalmic gels may be a viable alternative to the conventional eye drop as it offers increased contact time, decreased frequency of administration and thus improved patient compliance.

## Acknowledgment

Authors wish to thank Tamilnadu State council for science and technology (project code MS-009) to provide grant for this study. And also thank the management of PSG College of Pharmacy for encouragable support and providing necessary facilities to carry out this research work.

## References

1. ChrystleLeBourlais, LilianeAcar, HoseinZia, Pierre A Sado, Thomas Needham, Roger Leverge. Ophthalmic drug delivery systems recent advances. Prog Ret & Eye Res;17:33-58.
2. Eugen Barbu, Liliana Verestiuc, Thomas G. Nevella, John Tsibouklis. Polymeric materials for ophthalmic drug delivery: trends and perspectives. J Mater Chem 2006;6:3439–3443.
3. Loganathan VM. Atlas of clinical. 2007;2nd Edition, Jaypee
4. Martin, Dong-Hwan Kim, David C. Sustained release of Dexamethasone from hydrophilic matrices using PLGA nanoparticles for neural drug delivery. Biomaterials 2006;27:3031–3037
5. Neeraj Kumar, Jay Prakash Jain AJ, Domb, Sweta Modi. Copolymers of Pharmaceutical Grade Lactic Acid and Sebacic Acid: Drug Release Behavior and Biocompatibility. Eur J Pharm 2006;64:277-286.
6. Johannes Kdri Kristinsson, Hafrun Fridriksddttir, Sigridur Thorisdottir, Anna M. Sigurdardottir, Einar Stefdnsson F, Thorsteinn Lofisson: Dexamethasone – Cyclodextrin - Polymer Co-complexes in Aqueous Eye Drops. Aqueous Humor Pharmacokinetics in Humans. Invest Opthalmol Vis Sci 1996;37:1199-1203
7. Pandit JK, Bharathi D, Srinatha A, Ridhurkar DN, Singh S. Long Acting ophthalmic formulation of Indomethacin: Evaluation of alginate gel systems. Int J Pharm 2007;69:37-40.
8. Pandit JK, Shri Kant, Jagdish Balasubramaniam. In vitro and in vivo evaluation of the Gelrite gellan gum-based ocular delivery system for Indomethacine. Acta Pharm 2003;53:251–261.
9. Abdullahi Elsa, Stephen Ubandawak. Ketamine anesthesia following premedication of rabbits with vitamin C. J Vet Science 2005;6:239-241.
10. Sheng Lim K, Sanjeewa S Wickremasinghe, Francesca Cordeiro M, Catey Bunce, Peng T Kham. Accuracy of Intraocular Pressure Measurements in New Zealand White Rabbits. Inv Ophthal and Vis Sci 2005;46:2419-2423**.**