

**DESIGN OF OCULAR CONTROLLED RELEASE OCUSERTS OF BRINZOLAMIDE**

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Corresponding author e-mail:** snehalad22789@gmail.com*Received on: 04-02-2016; Revised on: 25-03-2016; Accepted on: 31-03-2016ABSTRACT**

The objective of research was to formulate and evaluate controlled release drug delivery system like ocuserts of Brinzolamide, an anti-glaucoma agent. Ocuserts were formulated using various polymers and plasticizers by film casting technique as drug reservoir membrane and controlled release polymers like Eudragit as the rate limiting membrane. The ocuserts were evaluated for physical characteristics, pH, uniformity in thickness & weight, swelling index, folding endurance, percent moisture loss, uniformity of drug content, in vitro diffusion studies. The ocusert containing HPMC K4M, HPMC E50 (1:1), Eudragit E100 showed controlled release with $82.00\% \pm 0.594$ at the end of 24 hours. Ex-vivo study showed $80.00 \pm 1.003\%$ of drug permeation. SEM analysis showed drug was in crystal form in the matrix of Ocusert, the surface had pores. IR and DSC studies confirmed no drug-polymer interaction. The optimized formula was sterilized and subjected to stability studies. No ocular irritation was seen in ocular irritancy study.

Keywords: Brinzolamide, Ocuserts, HPMC K4M, HPMC E50, Ocular delivery, controlled release, Eudragit.**INTRODUCTION**

Ocular drug delivery is one of the most challenging tasks faced by pharmaceutical researchers. Many intrinsic barriers, such as the cornea barrier, blood-aqueous barrier and blood-retinal barrier, restrict ocular drug delivery. Tear film acts as a barrier due to high turnover rate & gel-like mucus layer. Reflex stimulation increases lachrymation by 100-fold as compared to normal tear turnover. Mucin, hydrophilic gel layer present in the tear film has a protective role and acts as a barrier not only to microorganisms but also to therapeutic drugs.^[1]

Eye is a unique precise valuable organ, the anatomy, physiology, and biochemistry of the eye is such that it is impervious to foreign substances, therefore, it is a difficult for the formulator to pass through the protective barriers of the eye without causing any permanent tissue damage.^[2] Many ocular diseases can affect the eye and may cause loss of vision. Topical application of drugs to the eye is the most

popular and well-accepted route of administration for the treatment of various ophthalmic disorders. However, the bioavailability of ophthalmic drugs is very poor due to efficient protective mechanisms of the eye. Most commonly available ophthalmic preparations are eye drops and ointments which form about 70% of the ophthalmic dosage formulations in market. These conventional preparations when instilled into the cul-de-sac are rapidly drained away from the ocular cavity due to tear flow and lachrymal nasal drainage. Very little amount of drug is thus available for its therapeutic effect resulting in frequent dosing. So to overcome these problems novel drug delivery systems such as in-situ gels, ocular inserts, nanoparticles, nano-suspensions & micro-emulsions have been developed to increase the bioavailability of the drug in a sustained and controlled manner.^[3]

To increase the therapeutic efficacy of an ocular drug, viscosity enhancers can be added to increase its

contact time with the corneal surface. The contact time can also be increased by formulating water-insoluble ointment formulations.^[4] To maintain the therapeutic drug level, frequent instillations of the eye drops are necessary which can result in adverse effects due to erratic medication dose. To avoid these adverse effects ocular inserts, biodegradable polymeric systems, and collagen shields are coming up with new polymeric systems to attain better ocular bioavailability and sustained drug action.^[5]

Glaucoma is one of the primary causes of blindness and patients with acute glaucoma can develop irreversible vision loss. Glaucoma is a disease where the optic nerve is damaged. Open-angle glaucoma is the most common type in our country. It occurs from blocked aqueous drainage caused by an unidentified dysfunction or microscopic clogging of the trabecular meshwork. Closed-angle glaucoma occurs when the angle between the cornea and iris closes suddenly. Aqueous fluid cannot contact the drainage pathway entirely due to the closure, causing ciliary body ocular pressure to increase rapidly. Current therapy for ocular hypertension and glaucoma is by reducing intraocular pressure (IOP).^[6] Topical beta-blockers, which decrease aqueous humor production at ciliary body are the traditional therapy for patients and have been around for decades. Systemic side effects can occur from nasal absorption, making it especially important to ask patients about history of various diseases like asthma, COPD, and cardiac problems. As first line therapy, physicians are using newer drugs like topical Carbonic anhydrase inhibitors (CAI), alpha-agonists, and prostaglandin analogues, since they have fewer systemic side effects.^[7]

Topical Carbonic anhydrase inhibitors inhibit the production of aqueous humor and reduce the IOP by 16% to 22%. They work by inhibiting the enzyme carbonic anhydrase in the ciliary processes of the eye. Originally, CAIs like acetazolamide & methazolamide were available only in an oral form and were known to induce systemic side effects, such as depression, diarrhea, aplastic anemia metallic taste and kidney stones. Topical formulations of CAIs were developed since the IOP reduction is very effective. Topical preparations noticeably reduced systemic side effects as the inhibition of the carbonic anhydrase enzyme was limited to the eye. Dorzolamide 2% solution was the original drug in this class, followed by brinzolamide 1% (Azopt) suspension. Topical CAIs are an excellent secondary agent, used when the individual's primary drug is effective and tolerated but further IOP reduction is needed.^[8]

Ocular Sustained Release Drug Delivery Systems:

The novel ophthalmic drug delivery systems include: membrane –bound ocular inserts, mucoadhesive dosage forms, filter paper strips, collagen shields, cyclodextrin-based systems, ophthalmic rods, insitu gels, soft contact lens, implants, flexible coils and cotton pledgets as shown in figure 1.

Ocular Inserts: Ophthalmic inserts help to increase the contact time between the preparation and the conjunctival tissue, to ensure a sustained release suited for topical ocular therapy.^[9]

The advantages of ocular inserts as illustrated in figure 2 over the traditional ophthalmic preparations are summarized as:

Increased ocular residence, prolonged drug activity and higher bioavailability with respect to standard vehicles, release of drugs at a slow, constant rate, accurate dosing (insert contains a precise dose, which is fully retained at the administration site). Reduction of systemic absorption of the drug, better patient compliance, due to reduced frequency of administration and fewer incidences of visual and systemic side-effects. Targeting of internal ocular tissues is achieved through non-corneal routes like conjunctival & scleral, increased shelf life with respect to aqueous solutions, exclusion of preservatives, thus reducing the risk of sensitivity reactions.^[10]

The limitations of Ocusert as mentioned in figure 3 are Solidity as they are felt by the patients as an extraneous body in the eye, Occasional inadvertent loss during sleep or while rubbing the eyes, Difficulty in the placement of ocular inserts, and interference with vision.

MATERIALS AND METHODS

Materials: Brinzolamide was provided by Cipla pharmaceuticals Ltd. and (remove). Various grades of Eudragit like E100, RLPO, S100, L100, and RSPO were a gift sample from Evonik Degussa. Hydroxy Propyl Methyl Cellulose (HPMC K4M & E 50 LV) was a gift sample from Colorcon, Mumbai. PEG 400 was procured from S.D. Fine Chem. Ltd Mumbai India. All other reagents used were of analytical grade.

Standardization of drug: Brinzolamide was standardized as per monograph in USP 2014 & IP 2014.

Solubility: Solubility was checked in water, ethanol, methanol, chloroform, acetone, PEG 400, Tween 80, Dibutyl Phthalate, Glycerine, Propylene glycol and simulated tear fluid at pH 7.4.

Loss on drying: Brinzolamide (1gm) was weighed and dried in an oven at 100°C- 105°C to constant weight for 4 hours. The weight was again recorded.

Identification test: Infrared spectrum of Brinzolamide was investigated using FTIR Infrared Spectrophotometer (IR Affinity1, Shimadzu) using KBr pellet method. Spectrum was scanned over the wave number range 4000-400 cm⁻¹.

PREPARATION OF OCUSERTS:

The Ocuserts were prepared using HPMC K4M & HPMC E50 as drug carrier and Eudragit as rate controlling membrane as given in table 1.

Preparation of rate controlling membrane: The rate controlling films were prepared using hydrophobic polymer (different grades of Eudragit) and plasticizer PEG 400. The solutions were poured into 7 cm diameter glass Petri plate. The solvent was allowed to equilibrate & dry.

Preparation of drug reservoir membrane: The drug reservoir membrane composition was optimized by trying various concentration of HPMC K4M & HPMC E50 from 1%-5% w/w. among all the preparation films containing HPMC K4M HPMC E50 3% w/w (1:1) were optimized and used for further studies of ocuserts.

The optimized ocular film was used as drug containing reservoir film for the preparation of ocusert. The method of preparation is the same as that of the ocular films described earlier.

The drug reservoir solution was poured on the dried rate controlling membrane, and was placed inside an oven maintained at 35 ± 2°C for 3-4 hours. As the reservoir film was about to dry, rate limiting solution was poured above the reservoir membrane so that both the sides of the drug reservoir membrane are covered with rate limiting membrane, forming a tri-layer film. The tri-layer film was completely dried in oven for 2 hours. The dried films thus obtained were cut into required size (8 mm diameter) using stainless steel borer.

EVALUATION OF OCUSERTS

Physical characterization: The Ocuserts were evaluated for their physical characters such as shape, colour, texture, appearance, etc and reported.

pH: The pH (EUTECH instrument) of each of prepared ophthalmic formulations was determined using pH meter. The pH Meter was calibrated before each use with standard pH 4 & 7 buffer solutions.

Uniformity of Thickness: Thickness of ocuserts was measured by a digital Vernier caliper (Mitutoyo Corporation) at five different points on the sides and five different points in the centre of the film. The uniformity of thickness and standard deviation were calculated. ^[11]

Uniformity of Weight: Ten Ocuserts were taken from each batch and their individual weights were determined using electronic balance (Citizon).

Swelling Index: Three ocuserts were weighed and placed separately in beakers containing 4ml of simulated tear fluid. After a period of 5 minutes, ocuserts were removed and the excess water on surface of swollen ocuserts was wiped and weighed. ^[12] The % swelling index was calculated as:

$$\frac{(\text{Weight of swollen insert after time}) - \text{original weight of insert at zero time}}{\text{original weight of insert at zero time}} \times 100$$

Folding Endurance: The ocuserts were folded in the center, between finger and thumb and then opened. This was one folding. The number of times, the ocusert could be folded at the same place without breaking gave the value of folding endurance.

Percentage Moisture loss: The percentage moisture loss was determined to check the integrity of the ocuserts at dry condition. The ocuserts were weighed and kept in desiccator containing anhydrous calcium chloride. After three days, the ocuserts were taken out and reweighed.

Uniformity of Drug Content: Uniformity of drug content was determined by assaying the individual Ocusert. Three ocuserts were taken from each batch and individually dissolved in 10 ml of methanol in a volumetric flask and filtered. The absorbance of each of these solutions was then measured on UV-visible spectrophotometer at 254 nm.

In vitro diffusion study: The *in vitro* diffusion of drug from the ophthalmic inserts was explored using Franz diffusion cell. Dialysis membrane No.150 (HiMedia Laboratories, Mumbai) was tied to one end of open cylinder, which acts as a donor compartment. The dialysis membrane was considered as corneal epithelium. The entire surface of the membrane was in contact with receptor compartment containing 22 ml of STF pH 7.4. An ophthalmic insert was placed inside the donor compartment. The content of the receptor compartment was stirred continuously using a magnetic stirrer at 200 rpm and temperature was maintained at 37 ± 2°C. Aliquots (2ml) were withdrawn at 10, 20, 30, 45, 60 minutes, & 2, 3, 4, 5,

6, 8, & 24 hours and replaced by equal volume of fresh solution each time. The samples were analyzed spectrophotometrically at 254 nm against reference standard using STF as blank. [13]

Ex-vivo Corneal Permeation Studies: The *ex vivo* permeation studies were carried out on optimized formulations using Franz diffusion cell. Goat cornea was mounted onto a Franz-diffusion cell in such a way that corneum side continuously remained in an intimate contact with ocusert in the donor compartment. The receptor compartment was filled with simulated tear fluid pH 7.4 at $37 \pm 2^\circ\text{C}$. The receptor medium was stirred magnetically at 200 rpm. Aliquots (2ml) were withdrawn at relevant time intervals upto 24 hrs & analyzed spectrophotometrically at 254nm. [14]

Drug-excipient compatibility studies: Drug-excipients compatibility studies were investigated using Fourier transform infrared spectroscopy (FTIR) and Differential scanning calorimetry.

FTIR: IR spectra of blends of drug and HPMC K4M, HPMC E50, PEG 400, Eudragit E100, poloxamer F excipients were recorded on a FTIR spectrophotometer (IR Affinity1, Shimadzu) in the range of $4000\text{--}400\text{ cm}^{-1}$ using potassium bromide discs. Individual samples as well as the mixture of drug and excipients were ground, mixed thoroughly with potassium bromide for 3-5mins in a mortar and compressed into disc by applying a pressure in hydraulic press. The concentration of sample in potassium bromide should be in the range of 0.2% to 1%. The pellets were placed in light path and spectrum was obtained and reviewed for evidence of any interactions. [15]

DSC: A drug-excipients compatibility study of API & formulation was investigated using Differential scanning calorimetry (METTLER-TOLEDEO DSC1). Thermal analysis of Brinzolamide was carried out employing Differential Scanning Calorimeter. Samples were accurately weighed into aluminium pans and sealed. All samples were run at a heating rate of $10^\circ\text{C}/\text{min}$ over a temperature range $25\text{--}400^\circ\text{C}$ in atmosphere of nitrogen and thermograms were obtained.

SEM analysis: The morphology of inserts was studied using a Quanta 200 ESEM scanning electron microscope (SEM). The samples were prepared by freezing the inserts in liquid nitrogen. Next, the surfaces of the inserts were analyzed. The devices were analyzed at suitable acceleration voltages using

varying magnification for each sample. Representative micrographs were taken. [16]

Sterilization of ocular insets: The optimized ocuserts were sterilized by gamma radiation using the Cobalt-60 isotope as source of radiation at ISOMED at Trombay, Mumbai. These sterile formulations were used for ocular irritancy study.

Ocular Irritancy Test: The ocular irritation study was performed on rabbits using optimized formulations (Ocusert of Brinzolamide). After administration of the formulation, the rabbit eyes were inspected visually at specific time intervals. Approval of the Institutional Animal Ethic Committee was obtained prior to the commencing of the study. The approval number is CPCSEA/IAEC/BNCP/P-47/2014. Ocusert of Brinzolamide was instilled in right eye into the lower cul-de-sac and left eye considered as control. Both eyes of rabbit under test were examined periodically for erythema, edema, and lacrimation and for any sign of irritation before treatment and 30 min, 1hr, 24 hrs, 48hrs, 72 hrs, and 1 week after administration.

Stability Studies: The stability studies on optimized ocusert were conducted according to ICH guidelines. The ocular inserts were packed in Aluminium foil & stored in containers similar to contact lens. The formulations were stored at $8^\circ\text{C} \pm 2^\circ\text{C}$, $25^\circ\text{C} \pm 2^\circ\text{C}$ / $60 \pm 5\%$ RH, $40^\circ\text{C} \pm 2^\circ\text{C}$ / $75 \pm 5\%$ RH in incubator (LABLINE), $25^\circ/60\%$ RH, 40°C for 3 months. Samples were withdrawn on days 0, 30, 60 and 90 and analyzed for any changes in physico-chemical properties, average thickness, average weight, pH, drug content and diffusion profiles.

RESULTS & DISCUSSION

Preformulation studies:

Solubility: Brinzolamide was found to be readily soluble in methanol, ethanol, soluble in PEG 400, Tween 80 and propylene glycol, soluble in simulated tear fluid, poorly soluble in water, insoluble in Dibutyl phthalate & glycerin.

Loss on drying: Loss on drying of Brinzolamide was $0.175\% \pm 0.006$ (n=3). The USP limit of the loss on drying of Brinzolamide is not more than 0.5 %.

FTIR: Figure 4 shows the IR spectrum of Brinzolamide with the following peaks- 3313 cm^{-1} N-H stretch of primary amines, 3095 cm^{-1} Aromatic ring C-H stretch, 1335 & 1354 cm^{-1} Asymmetric stretch of sulfones, 1607 cm^{-1} N-H bends in primary amines, 1528 cm^{-1} N-H bends of secondary amines, 1465 cm^{-1}

¹ Characteristic bending of methylene groups, 1014 cm⁻¹ C-O stretch of ethers, 721 cm⁻¹ Bending (rocking) associated with 4 or more CH₂ groups in an open chain

EVALUATION OF OCUSERTS:

Ocuserts of Brinzolamide were optimized by using different grades of Eudragit in rate limiting membrane. The physical characteristics of the formed Ocuserts were same, therefore Ocuserts were optimized on the parameters like percent drug diffused, drug content, thickness, weight, swelling index, folding endurance, percent moisture loss as per the data in table 2 & 3.

Physical characterization: Various other physical parameters were also evaluated as follows: Shape: Circular, Colour: clear & transparent, Texture: Smooth & Uniform, Edge: Smooth & Uniform as given in figure 5

pH: The ocuserts were found to exhibit pH in the range of 6.1 to 7.8. Ophthalmic formulations must be in the pH range between 4.5 and 8. The pH of all formulations was within the range and hence no eye irritation was expected.

Uniformity of Thickness: The average thickness of Ocuserts was between 0.17mm to 0.23mm. There were no marked variations in the thickness of Ocuserts within each formulation indicating uniform behavior of film throughout the process.

Uniformity of Weight: The average weights of Ocuserts were found to be in the range of 11mg to 14.72 mg. The uniformity of weight of Ocusert indicated good distribution of the drug, polymer and plasticizer.

Swelling Index: Swelling index was found in the range of 92- 120%. This showed that ocuserts hydrated quickly. This indicates the water retaining capacity of HPMC.

Folding Endurance: The folding endurance was measured for all formulations manually. It was found in the range of 75 to 98. This test reflects the flexibility of ocuserts. This test ensures that the prepared ocuserts were suitable for large scale manufacture to produce long, continuous film without breaking or tearing.

Percentage Moisture loss: Percentage moisture absorption was observed from 3% to 10%. There was no change in integrity at high humid and dry

conditions which was observed by physical appearance.

Uniformity of Drug Content: Drug content of Ocuserts was in the range of 85-102%. The results indicated that the drug was uniformly dispersed.

In vitro diffusion study: *In vitro* diffusion studies of Brinzolamide ocuserts were carried out using dialysis membrane No.150 (HiMedia Laboratories, Mumbai) in STF pH 7.4 solutions. The apparatus was designed with the objective of mimicking the conditions of ocular cavity to certain extent. The release data is given table 4 for formulation E1 to E5 with graphical presentation in figure 6.

The drug diffused from formulation E2 was found to be lowest about 50% and that for E3, E4 & E5 was 57%, 68% & 72% respectively. The drug diffused from E1 was found to be about 91%.

Ex-vivo Corneal Permeation Studies: Ex-vivo study was carried out on goat cornea using Franz diffusion cell for 24hrs and drug retention on the cornea was calculated as shown in figure 7.

Ex-vivo studies showed about 75-79% of the drug permeated through the cornea in 24 hours for optimized Brinzolamide ocuserts; E1.

Drug retention study: 10-13% of the drug retained on the corneal membrane. This indicates good ocular permeation.

Drug-excipient compatibility studies:

FTIR: Figure 8 & 9 IR spectrum of brinzolamide and excipients HPMC K4M & HPMC E50 respectively. Figure 10 shows IR spectrum of physical mixture of ocusert(E1).

The peaks of pure drug (Brinzolamide) corresponded with those obtained in peaks of mixtures of drug and excipients indicated absence of an interaction between drug and excipients.

DSC: DSC analysis was performed using a Shimadzu differential scanning calorimeter (METTLER-TOLEDEO DSC1). Samples (3–4 mg) were placed in flat-bottomed aluminum pan and heated at a constant rate of 10°C/min in an atmosphere of nitrogen in a temperature range of 20–250°C.

DSC thermogram of Brinzolamide was typical of a crystalline substance as shown in the figure 11, exhibiting a sharp endothermic peak at 133°C relative to its melting point, with onset of the peak at 130.98°C and endset at 138.95°C. The thermograms of the formulation ocusert (E1), figure 12 did not show the endothermic peak of brinzolamide at 133°C. The drug sharp characteristic peak was completely broadened and hardly detected in the

DSC thermogram of the formulation which indicates the suppression of the drug crystallinity in the formulations. This is an indication of complete drug amorphization and/or well distribution of brinzolamide ocusert matrices.

SEM analysis: On observing SEM images as shown in figure 13 in high magnification of formulation there were drug crystals from which we can conclude drug was in crystal form in the matrix of Ocusert, the surface showed there were pores in the matrix from which it can be concluded that diffusion of drug was due to these pores.

Sterilization: The optimized ocuserts were sterilized by gamma radiation before *in vivo* study using the Cobalt-60 source.

Ocular Irritancy Test: Ocular irritation study was performed to determine whether the developed formulation might cause irritation and pain. It was performed on rabbits using optimized E1 ocusert. There was no sign of redness and non-continuous blinking of the eye. Thus it was concluded that the formulation was non irritant to rabbit eye.

Stability Studies: The stability data conducted for 3 months at $8^{\circ}\text{C} \pm 2^{\circ}\text{C}$, $25^{\circ}\text{C} \pm 2^{\circ}\text{C}$ / $60 \pm 5\%$ RH, $40^{\circ}\text{C} \pm 2^{\circ}\text{C}$ / $75 \pm 5\%$ RH is given in the table 5. The results showed that the formulations were stable during storage.

CONCLUSION

Ocuserts of Brinzolamide were prepared by using the drug, HPMC K4M, HPMC E50 and Eudragit E100 with PEG 400 as plasticizer. The drug reservoir membrane of Ocuserts were prepared similar to films as film casting method with Eudragit E100 (4% w/w) as the rate limiting membrane. The prepared Ocuserts

were evaluated for various parameters. In vitro drug release studies using diffusion membrane and ex vivo permeation studies using goat cornea were carried out on Franz diffusion cell. The optimized formulation (E1) was subjected to sterilization, ocular irritancy study and stability study.

The Ocuserts were found to be smooth, transparent, flexible and clear. The thickness and weight of the Ocuserts were uniform and clearly indicates the accuracy of the preparation. The percentage moisture absorption was not so significant and the integrity of the ocuserts was stable throughout the study duration. Folding endurance was satisfactory for all the ocuserts since the crack was observed in these strips after more number of foldings. Among the prepared formulations, the formulation (E1) containing HPMC K4M & HPMC E50 (1:1), Eudragit E100 as rate limiting membrane showed controlled release with $82.00\% \pm 0.594$ at the end of 24 hours. Ex vivo study showed $80.00 \pm 1.003\%$ of drug permeation and 10-13% of drug retention. SEM in high magnification of formulation there were drug crystals from which we can conclude drug was in crystal form in the matrix of Ocusert, the surface showed there were pores in the Matrix from which it can be concluded that diffusion of drug was due to these pores.

The developed formulations are viable alternatives to conventional eye drops by virtue of their ability to enhance bioavailability through longer precorneal residence time and ability to sustain drug diffusion. Ocuserts can also decrease the frequency of administration resulting in better patient acceptance.

ACKNOWLEDGEMENT

It is a great pleasure to acknowledge "SVKM's Dr. Bhanuben Nanavati College of Pharmacy", for providing the necessary infra-structural facilities to carry out the research work successfully.

Table 1: Composition of Ocuserts containing Brinzolamide:

Formulation code	RATE LIMITING MEMBRANE			DRUG RESERVOIR MEMBRANE					
	Eudragit grade	Eudragit grade	PEG 400	Drug	HPMC K4M (w/v)	HPMC E50 (w/v)	PEG 400	Ethanol	Purified water
E1	E 100	120 mg	0.5ml	1%	3%	3%	0.5ml	2ml	q.s 10ml
E2	S 100	120 mg	0.5ml	1%	3%	3%	0.5ml	2ml	q.s 10ml
E3	L 100	120 mg	0.5ml	1%	3%	3%	0.5ml	2ml	q.s 10ml
E4	RS PO	120 mg	0.5ml	1%	3%	3%	0.5ml	2ml	q.s 10ml
E5	RL100	120 mg	0.5ml	1%	3%	3%	0.5ml	2ml	q.s 10ml

Table 2: Evaluation parameters of ocuserts

FORMULATION CODE	pH	**AVERAGE THICKNESS (mm)	**AVERAGE WEIGHT (mg)	*SWELLING INDEX
		AM ± SD	AM ± SD	MEAN %SW ± SD
E1	6.7	0.23 ± 0.042	16.93 ± 1.57	79.00 ± 2.31
E2	6.5	0.176 ± 0.022	14.23 ± 1.40	82.30 ± 2.98
E3	6.1	0.189 ± 0.055	13.88 ± 1.78	102.46 ± 1.11
E4	7.8	0.196 ± 0.049	14.72 ± 1.79	105.97 ± 0.76
E5	7.1	0.238 ± 0.030	11.19 ± 1.34	116.23 ± 1.56

* Average of three determinations

** Average of ten determinations

Table 3: Evaluation parameters of ocuserts

FORMULATION CODE	*Folding endurance	*% moisture loss	*Amount of drug present (mg)	% Drug content
	MEAN ± SD	AM ± SD	AM ± SD	
E1	97.3 ± 2.08	4.12 ± 0.18	0.526 ± 0.007	105.28
E2	88 ± 1.73	8.34 ± 0.05	0.462 ± 0.003	92.41
E3	83.6 ± 1.53	9.58 ± 0.08	0.498 ± 0.005	99.54
E4	73.3 ± 2.08	7.89 ± 0.14	0.447 ± 0.005	89.42
E5	75.3 ± 3.21	4.67 ± 0.15	0.426 ± 0.007	85.28

* Average of three determinations

** Average of ten determinations

Table 4: In-vitro % drug diffused of Ocuserts (E1, E2, E3, E4, and E5)

TIME	% drug diffused				
	E1	E2	E3	E4	E5
5	6.74	1.84	2.50	1.47	0.66
10	14.83	4.40	7.57	4.39	2.38
20	23.27	9.74	9.67	14.42	6.57
30	29.98	20.14	11.94	21.05	27.37
45	44.13	24.84	17.49	30.86	31.34
60	51.67	25.37	21.83	35.72	36.90
120	54.69	28.23	27.31	39.23	40.91
180	64.33	26.46	27.50	37.24	46.62
240	69.52	38.04	33.26	41.33	47.30
300	73.79	36.52	41.11	39.17	53.48
360	79.03	39.75	46.29	42.60	50.72
480	80.18	37.07	48.00	40.08	61.94
1320	82.59	50.28	57.89	68.44	72.67
1440	83.95	55.31	62.27	69.23	90.00

Table 5: Stability study of optimized Ocuserst:

Evaluation parameter/ Stability condition		Appearance	pH	Average Thickness (mm)	Average Weight	Drug content (%)	% drug diffused
8°C ± 2° C	1month	Clear smooth transparent	6.7	0.23 ± 0.042	16.93 ± 1.57	105.28 ± 0.023	88.23 ± 0.26
	2month	Clear smooth transparent	6.2	0.26 ± 0.45	17.03 ± 1.28	102.8 ± 0.56	87.13 ± 0.54
	3month	Clear smooth transparent	5.9	0.24 ± 0.05	16.5 ± 0.9	101.8 ± 0.09	90.13 ± 0.78
25°C ± 2° C / 60 ± 5 % RH	1month	Clear smooth transparent	6.4	0.21 ± 0.75	19.93 ± 1.45	101.28 ± 0.45	87.23 ± 0.11
	2month	Clear smooth transparent	6.1	0.22 ± 0.58	18.66 ± 1.89	101.1 ± 0.79	86.13 ± 0.89
	3month	Clear smooth transparent	5.8	0.20 ± 0.41	17.06 ± 0.95	100.6 ± 0.29	85.13 ± 0.9
40°C ± 2° C / 75 ± 5 % RH	1month	Clear smooth transparent	5.8	0.18 ± 0.52	17.93 ± 0.25	102.2 ± 0.59	86.89 ± 0.11
	2month	Clear smooth transparent	5.6	0.19 ± 0.24	16.66 ± 0.98	101.41 ± 0.97	86.60 ± 0.34
	3month	Clear smooth transparent	5.4	0.17 ± 0.85	15.36 ± 0.74	100.3 ± 0.97	85.40 ± 0.95

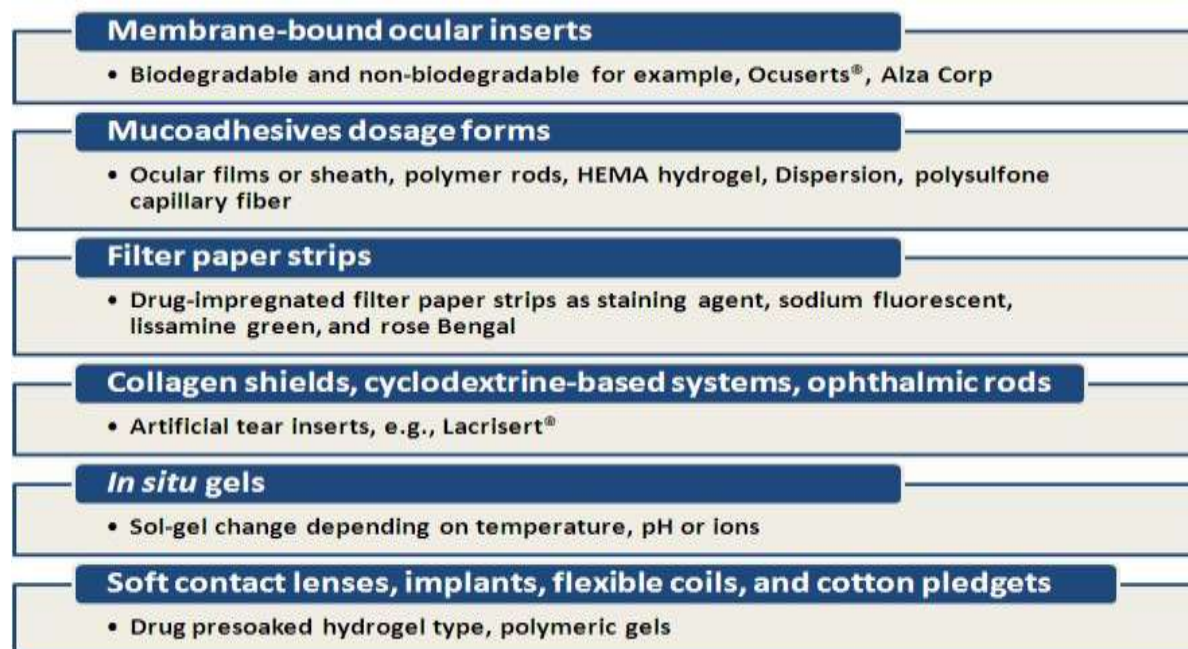
**Figure 1: Schematic representation of novel ophthalmic drug delivery systems include**



Figure 2: Advantages of ocular insert

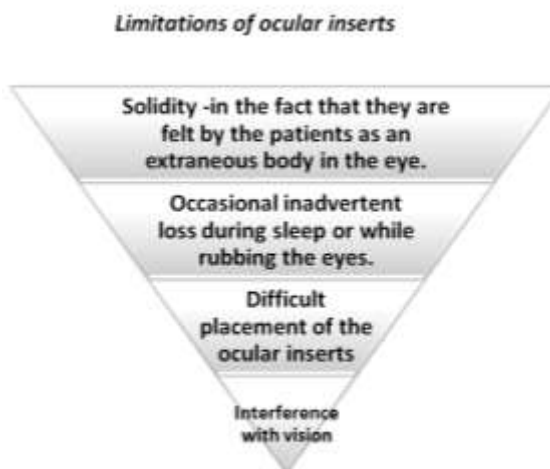


Figure 3: Limitations of ocular inserts

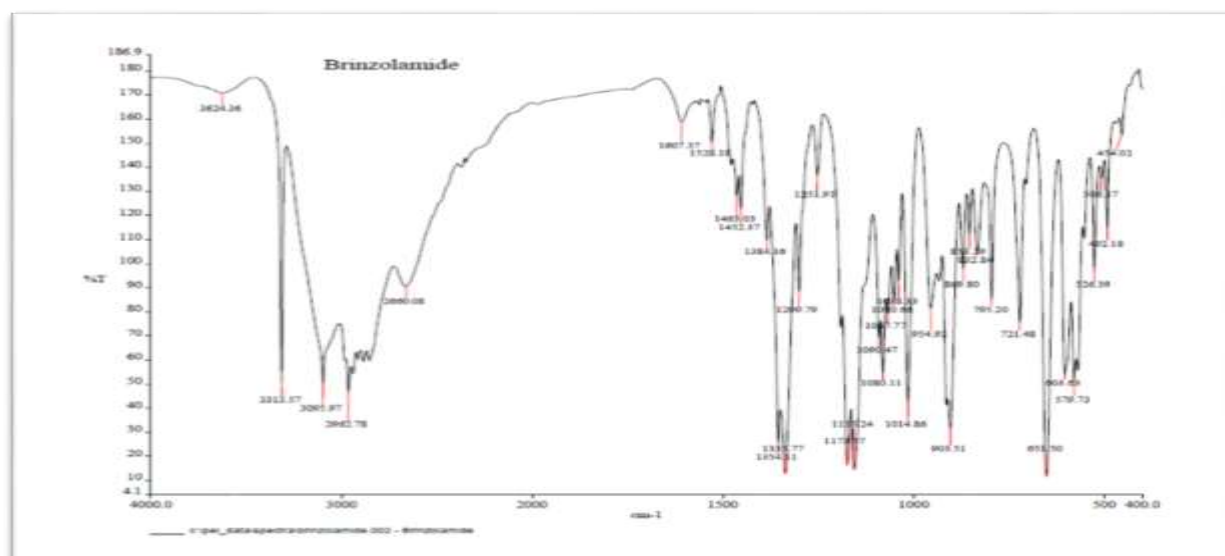


Figure 4: IR spectrum of Brinzolamide



Figure 5: Photograph of the prepared ophthalmic ocuserts (E1)

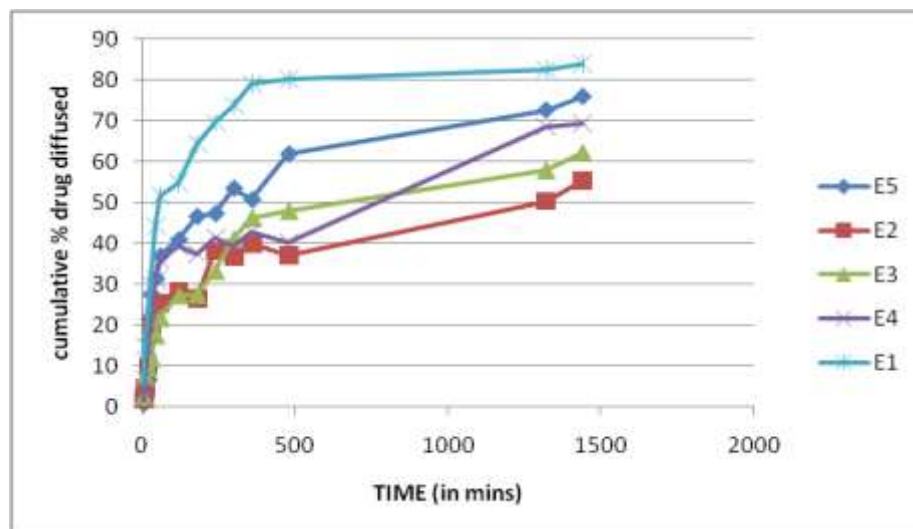


Figure 6: Graphical representation of Time (in minutes) Vs in-vitro cumulative % drug diffused

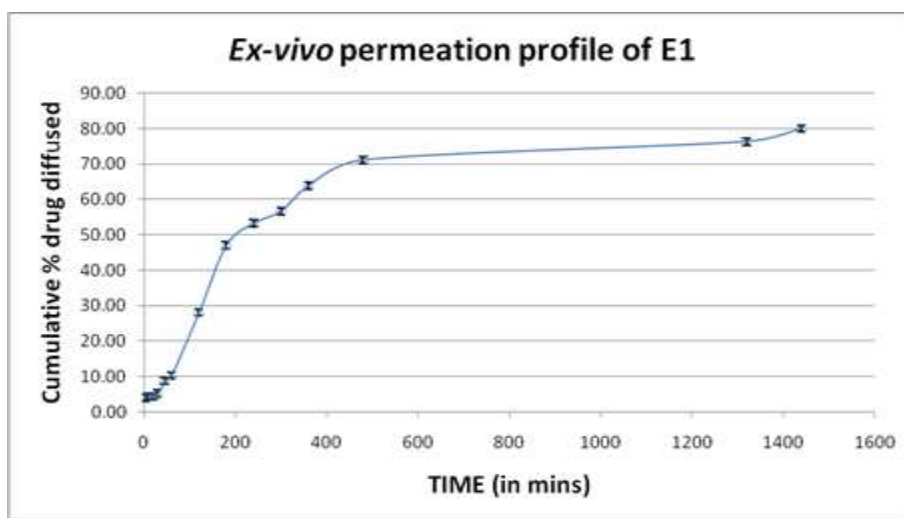


Figure 7: Graphical representation of Time (in minutes) Vs ex-vivo cumulative % drug diffused

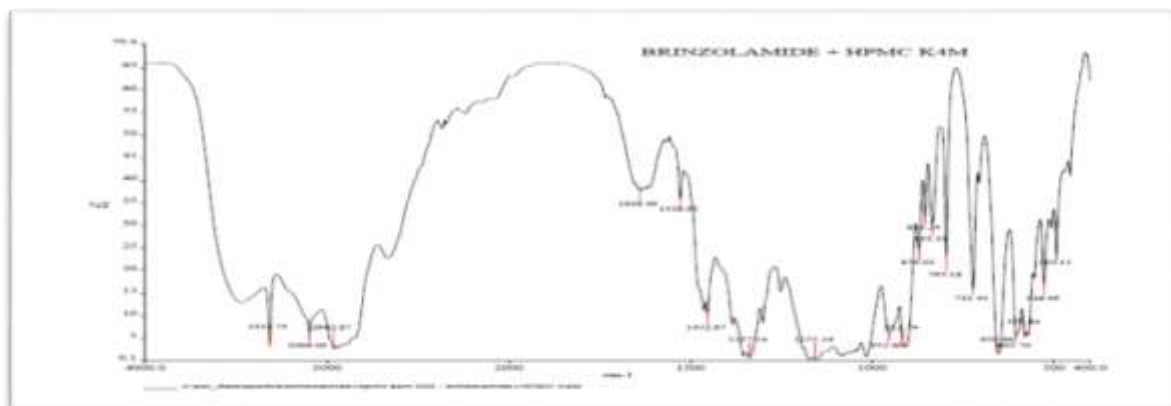


Figure 8: FTIR spectra of Brinzolamide and HPMC K4M

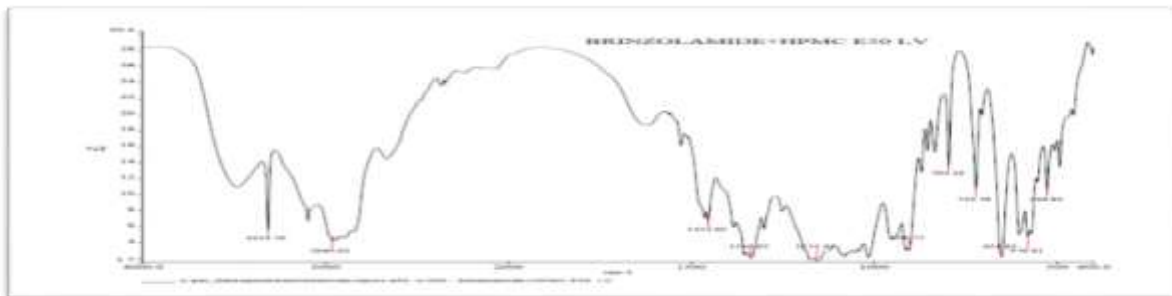


Figure 9: FTIR spectra of Brinzolamide and HPMC E50

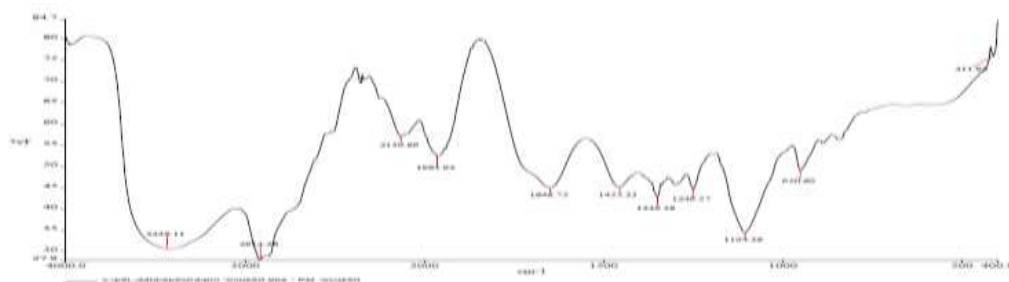


Figure 10: FTIR spectra of physical mixture of OCUSERT (E1)

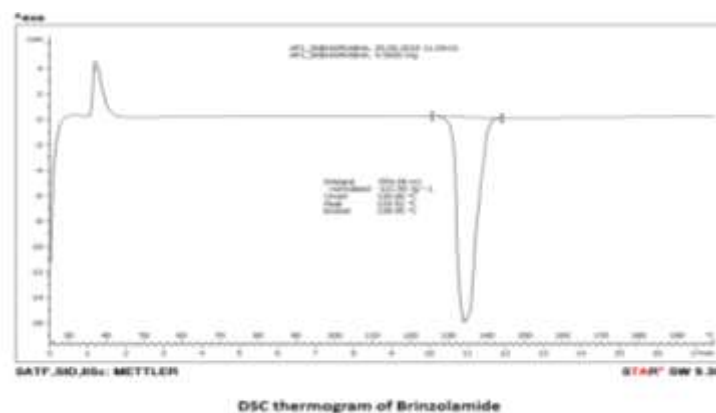


Figure 11: DSC thermogram of Brinzolamide



Figure 12: DSC thermogram of Ocusert (E1) of Brinzolamide

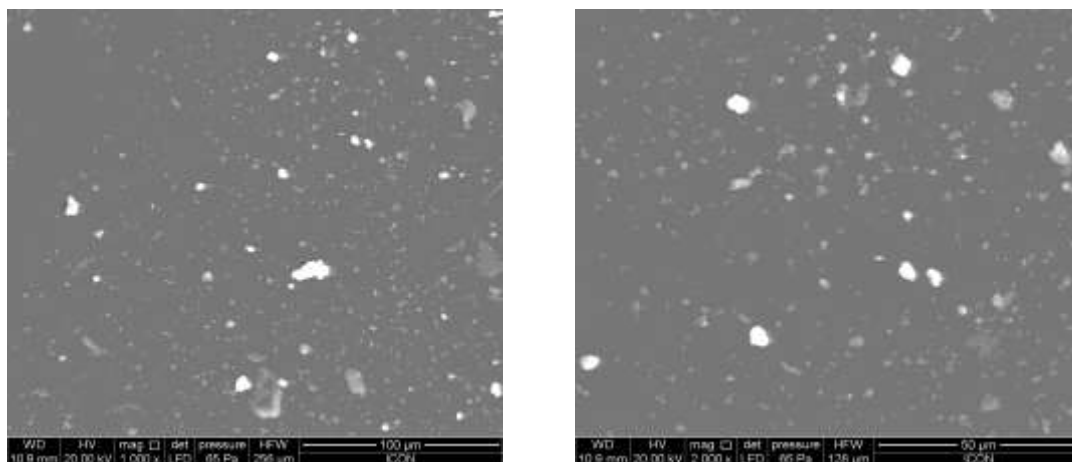


Figure 13: Scanning electron photomicrographs of the Formulation E1,

A): 1000 X, B): 2000 X

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