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Single Administration Sustained Release Formats for Delivery of Ophthalmic Medications to the Ocular Surface, Cornea and Anterior Chamber

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Abstract

Post-surgical inflammation and infections are treated with eye-drops containing glucocorticosteroids (Prednisolone, difluprednate, dexamethasone, loteprednol etabonate).

Loteprednol etabonate was designed as a "soft" steroid, whereupon it is rapidly deactivated to inactive metabolites by nonspecific tissue esterases in the ocular tissue. This limits its potential to cause adverse effects such as ocular hypertension and glaucoma, side effects commonly known to occur with steroids.

Loteprednol Etabonate is approved for use in prevention and treatment of post-operative inflammation after cataract surgery (Lotemax™ ointment, Lotemax™ gel), for B.I.D administration.

We have contemplated a sustained release drug product that does not require daily dosing with eye-drops or ointment. We are developing a micro-encapsulated Loteprednol Etabonate dosage form that can be injected in the conjunctival sac, right after surgery. For control of inflammation after cataract surgery, the biodegradable delivery system is modulated to release its payload in 2-3 weeks, with a single injection.

In this study, we utilized PLG as the biodegradable polymer encapsulate. We have investigated the role of MW, structure and size on the release profile of LE. Additionally, we have established injectability as a function of size.

The data generated in this work will be utilized for the development of the final EySite-LE Drug Product.

EySite™ Delivery System

- Proprietary platform drug delivery system for small molecules and large molecules, protected by IP
- Injectable via 27-31G needle
- Enhanced Residence Time
- Precise Release
- Biodegradable
- Disperses; stays on-site

Loteprednol Etabonate

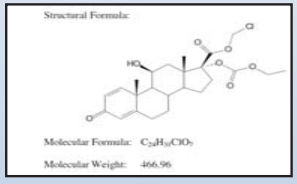

FIG. 1:
Chemical Structure

Materials & Methods

Preparation of Micro-encapsulates:

- PLG microencapsulates were prepared by a proprietary process, to achieve a narrow size distribution, optimum size and injectability. Loteprednol Etabonate was purchased from Sigma, Inc.
- PLG (50:50 L:G) of different molecular weights were utilized to assess effect on in-vitro release.
- A blend of PLG terminated with acid end groups and ester end groups were prepared to assess effect on in-vitro release.

Characterization:

- Particle Size Distribution (PSD): Samples were suspended 20mg /mL in a diluent, diluted with an equivalent volume of water. 500uL of the suspension was dispersed in a dispersal medium (a thirty fold dilution of the diluent in distilled water). Particle size was measured on a Horiba LA-950 Laser Diffraction Particle Analyzer.
- Imaging: Dry encapsulates were characterized by scanning electron microscopy.
- Encapsulation (mg/G): 20 mg of the microencapsulate was dissolved in 1mL of acetonitrile, and 10mL of isopropanol added drop-wise while mixing. 1mL of the slurry was centrifuged (5min; 6000 RPM), and the supernatant was removed for HPLC analysis. HPLC analysis was performed on a RP C18 column.
- Compatibility: The compatibility of PLG and loteprednol etabonate was assessed by comparison of their HPLC profiles.
- *In-Vitro* Burst (%): 30 mgs of the microencapsulates were reconstituted in 1 mL of PBS, pH 7.4 and rotated at 37°C, in a 2 mL polypropylene centrifuge tube. At the 1 hour time-point, the centrifuge tubes were spun down at 15,000 RPM. One mL of the supernatant was removed for analysis by HPLC.
- *In-Vitro* Release: 200-300 mg of microspheres were weighed into 1 mL SpectraPor cassettes (cellulose membranes, MWCO 1000 Kda, 5mL). The tubes were suspended in 45mL and incubated at 50C. Time point samples were obtained after 1 hour and daily over the course of twenty days. The samples were analyzed by HPLC.
- Injectability of the microspheres suspended in the reconstituting fluid were tested both through 23G and 27G needles.

Results

Table 1 tabulates the encapsulation, and PSD correlated with the type of PLGA utilized in preparing the microencapsulates.

Lot#	Polymer Name	Intrinsic Viscosity	Encap.	PSD (d_{90}/d_{50})
KB-05-01	Purasorb 5004A; 50/50 L:G	0.39 dl/g	187.9 mg/g	40.4/67
KB-05-25A	Purasorb 5002; 50/50 L:G	0.2 dl/g	19.8 mg/g	15/41.9
KB-05-25B	Purasorb 5002; 50/50 L:G	0.2 dl/g	29.4 mg/g	16.7/37.2
KB-05-28	Purasorb 5002A; 50/50 L:G	0.21 dl/g	47.5 mg/g	11.6/24.3
KB-05-34	Mix: A 50/50 combination of Purasorb 5002 and 5002A, both 50/50 L:G	0.21 dl/g	14.9 mg/g	9.3/21

Results

Process Produces Solid Microstructures

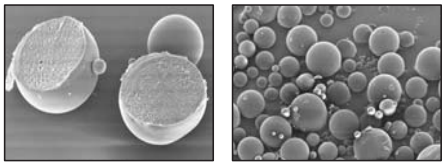

FIG.1: Micro-encapsulates have solid interior (all batches)

Scaled-up Process Yields High Encapsulation Efficiencies

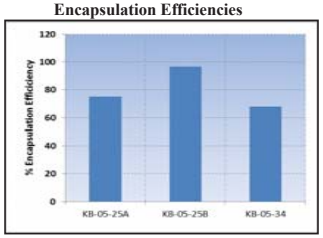

FIG.2: Micro-encapsulates have solid interior (all batches)

Blend of Acid-end group and Ester End group PLG (KB-05-34) demonstrates best LE release

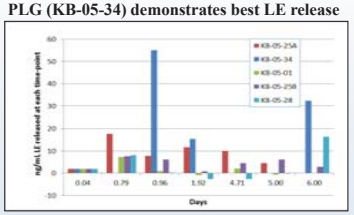

FIG 3: Accelerated *in-vitro* release, all batches

Smallest size (KB-05-34) and lowest MW (KB-05-34) demonstrates best LE release

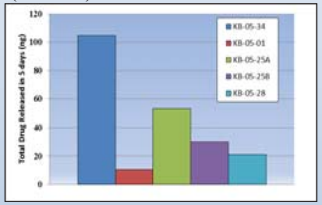

FIG 4: Accelerated *in-vitro* release

Results

Injectability:

- All batches were deemed injectable through 23G needles.
- All batches were deemed injectable through 27G needles.

Compatibility:

- The HPLC assay profile of the loteprednol etabonate standard and extracted loteprednol etabonate looked comparable.

In-Vitro Burst:

- The in-vitro burst for all prototypes were minimal (<0.001%).

Discussion and Conclusions

- This data set demonstrated that loteprednol etabonate could be encapsulated successfully in PLG-based microencapsulates. The size of the microencapsulates can be modulated by control of process conditions, with a range of sizes between 9.3-40.4 microns (d_{50}).
- An accelerated in-vitro release test was developed to enable fast screening of prototypes to assess difference in in-vitro release of drug produced as a function of size, polymer structure and MW. Through this test, it was ascertained that PLG of higher molecular weight and larger size (KB-05-01) released very slowly, as opposed to a blend prototype of ester end and acid end PLG (50:50 L:G) (KB-05-34).
- In comparing the release rate of acid end group (KB-05-28) and ester end group PLG (KB-05-25B) at the same MW (0.21 dl/g), it was expected that more drug would be released with the acid end group prototype, which was not the case. This result will be investigated further.
- All batches were injectable through 27G needles for the entire size range of the prototypes, which renders it feasible for injection in ocular tissue.
- PLG polymers and loteprednol etabonate were deemed compatible.
- All PLG microencapsulates were designed to have a solid microstructure. Future experiments will explore the effect of "honey-combed" microstructures on loteprednol etabonate release profiles.
- ***The results presented herein will provide the basis of further development of the EyeSite-LE Drug Product.***

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