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Unlocking the Potential of Eyelid Skin-Based Drug Delivery: A Filipino Pharmaceutical Scientist's Journey

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1. Introduction

In 2015, Dr. See, a lecturer at the University of San Carlos, led the Philippine delegation in a two-week Sakura Science Exchange Program which was hosted by the Laboratory of Drug Safety Management, Josai University (Prof. Yutaka Inoue). It was his first academic immersion in Japan. His brief encounter with the Japanese culture and academic systems deepened his interest for pharmaceutical science research and inspired him to pursue further studies. Leaving the comfort of his home for the next three years was tough yet a fruitful decision.

In spring of the following year, he joined the Laboratory of the renowned dermal scientist Professor Kenji Sugibayashi where they pioneered the technique of delivering ophthalmic drugs through the eyelid skin. Having worked hard together, they produced 4 scientific papers on eyelid-skin based drug delivery, 3 international research presentations, 2 best oral presentation awards, and the first Filipino pharmaceutical scientist to receive the post-doctoral award given by the Academy of Pharmaceutical Sciences and Technology, Japan.

What were the milestones towards these achievements? Let's get to know the details in the succeeding sections.

2. Eyelid Skin as Potential Site for Drug Delivery to Ocular Tissues

For several decades, eye drops have remained as the method of choice in delivering ophthalmic drugs and the most extensively utilized pharmaceutical formulations for various ocular diseases (i.e., the eyeball and surrounding tissues). Currently, eye drops account for about 90% of ophthalmic medicines, primarily due to their ease of administration and good patient compliance. However, several drawbacks are associated with the utilization of eye drops. Eye medications cannot be administered beyond the capacity of the conjunctival sac due to its limited volume. Most eye drops exhibit low bioavailability, poor targeting efficacy, and are virtually impossible to administer during sleep. Anatomical and physiological constrains such as tear turnover, nasolacrimal drainage, reflex blinking, and ocular static and dynamic barriers impede the bioavailability and controlled delivery of drugs administered as eye drops. With these impediments, it is essential to search for an alternative approach to deliver ophthalmic drugs with high targeting ability while simultaneously improving drug absorption into the ocular tissues.

We paid attention to applying formulations and delivering ophthalmic drugs onto the (lower) eyelid skin. The lower eyelid skin exhibits less movement due to blinks compared with the upper eyelid skin, making it a good site for administration. Moreover, it is very interesting for skin researchers to explore the eyelids,

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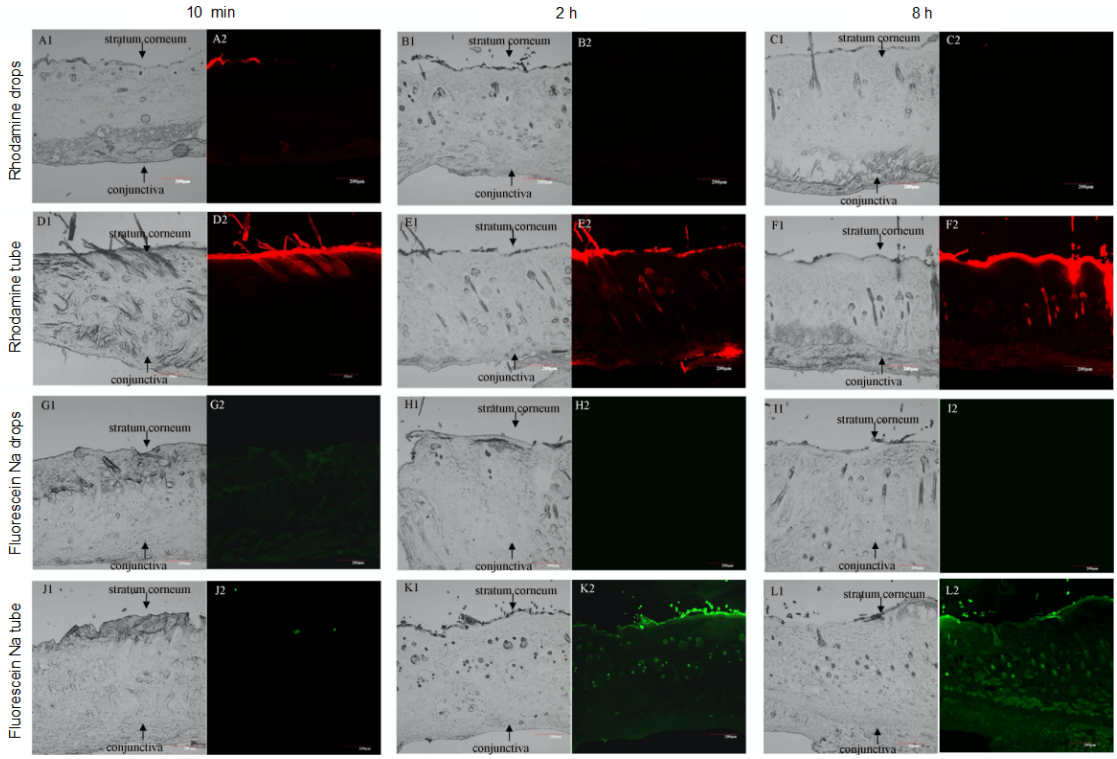

Fig. 1 Photomicrographs of rat eyelid skin under CLSM after *in vivo* absorption studies. Transmission Images (A1–L1); Confocal laser scanning images (A2–L2)[1].

which is the thinnest skin layer on the human body (<1 mm). Generally, the thinner the stratum corneum, the greater the drug permeation. In addition, drug administration through the skin provides several advantages because it generally facilitates avoidance of premature metabolism, decreased toxicity, fewer side effects as well as greater patient compliance.

With the limited number of studies investigating the delivery of drugs into the conjunctiva through eyelid skin and the paucity of data describing eyelid drug permeation, we were prompted to study hydrophilic and lipophilic model compounds and to compare permeability characteristics through the eyelid and the abdominal skin. Pilocarpine hydrochloride, tranilast, antipyrine, diclofenac sodium, aminopyrine, and lidocaine were used to evaluate their skin permeation; the former two were used to evaluate the skin concentration, and two fluorescent dyes (fluorescein sodium and rhodamine B) were used to determine the *in vivo* distribution of the drugs in the conjunctiva.

Drug permeation through the eyelid skin was higher than through the abdominal skin independent of the lipophilicity of the applied drugs. Figure 1 shows the drug distribution in skin after application of its solution in eye drops or by topical application onto the lower eyelid skin. The presence of rhodamine B and fluorescein sodium administered as eye drops as markedly observed in the conjunctiva, as indicated by the fluorescence in the images 10 min after administration, whereas these were absent at 2 and 8 h. For topical administration onto the eyelid skin, a stronger intensity of the fluorescence given off by the model compounds was observed over time, which indicated greater amounts of dye compound in the conjunctiva.

The eyelid skin is known to be thinner than the conventional skin administration sites, and it offers higher drug permeation, as demonstrated by the results of this study. Developing formulations dedicated to eyelid administration such as an eyelid patch is recommended. In fact, ointments are often used for night time application but are associated with discomfort due to greasiness, tear film instability, and uncertain drug disposition after eye shutting while asleep. Thus, eyelid skin application is seen to be beneficial even during sleep when the use of liquid and semi-solid eye preparations is deemed impossible.

3. Enhancement of Drug Delivery into the Eyeball through Eyelid Skin by Iontophoresis

In the previous section, we established that drugs applied to the lower eyelid skin could migrate to the conjunctiva, and the drug concentration was maintained in the conjunctival sac for a long duration com-

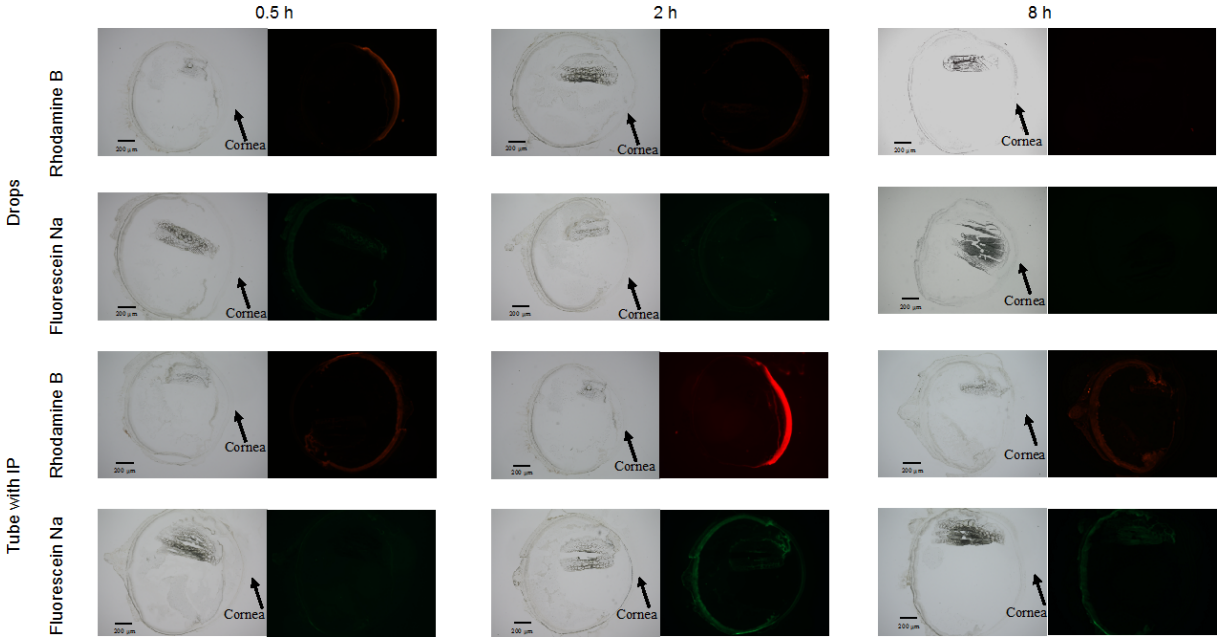

Fig. 2 Photomicrographs of rat eyeballs after *in vivo* distribution studies. All observations were performed without changing observation conditions for each drug[2].

pared with eye drop application. However, the migration of drugs to the eyeball after eyelid skin application was considered to be lower than eye drops because drugs, in the case of eye drops, were delivered directly onto the eyeball. In this case, we considered the necessity of utilizing physical approaches, such as iontophoresis (IP), to further promote percutaneous absorption and delivery to the eyeball via the lower eyelid skin.

IP is a non-invasive technique to increase the penetration of ionized compounds into the body across the epithelial surface, including the skin and ocular surface. The basic principle of IP is that ions with opposing charges attract and ions with the same charge repel. Moreover, ionized substances are driven into the tissue by electrorepulsion and electroosmosis. This method has the potential to control the penetration of drugs depending upon the applied current density and application time. Because of its recognizable advantages, such as the ease of application, non-invasiveness, and increased drug penetration directly into the target tissue, IP has been used to deliver drugs across various barriers such as the skin and eyes. Moreover, topical preparations utilizing IP have already been put to practical use in ocular drug delivery by direct application of an IP device to the cornea or the eyelids but none have been done on the lower eyelid skin.

In vivo study reveals that the concentration of pilocarpine in the eyeball is significantly increased by 3-fold with IP application over 0.5 h. For the 2-h application, however, it did not significantly increase with IP, suggesting that the steady state of pilocarpine had been reached within 0.5 h after IP application. *In vivo* distribution studies of two fluorescent dyes further supported the findings of our *in vivo* skin permeation studies. Topical administration with IP showed that corneal permeation of the both fluorescein sodium and rhodamine B was observable 0.5 h after administration (Fig. 2).

The eyelid skin is known to be thinner than most known application skin sites, and offers higher drug permeation into target tissues. With an aid of physical permeation enhancement methods, it is expected that permeation rates will exceed conventional drug delivery methods. IP application on the eyelid skin allowed the delivery of pilocarpine to the eyeballs. The concentration of pilocarpine after eye drop is important and the measurement of its pharmacokinetic profile as well as the disposition in the eye will be considered in succeeding studies.

The use of IP enhanced the penetration and delivery of drugs into the eyeball after topical application onto the lower eyelid skin. Higher concentrations of pilocarpine were detected in the eyeball after IP application. Histomorphological evidence confirmed the migration of dyes into the posterior chamber of the eyeball after topical administration onto the lower eyelid skin with IP.

4. Pharmacokinetics and Tissue Distribution of Pilocarpine Following Eyelid Skin Delivery

Extending the delivery of drugs into the eyes while reducing systemic bioavailability is of utmost impor-

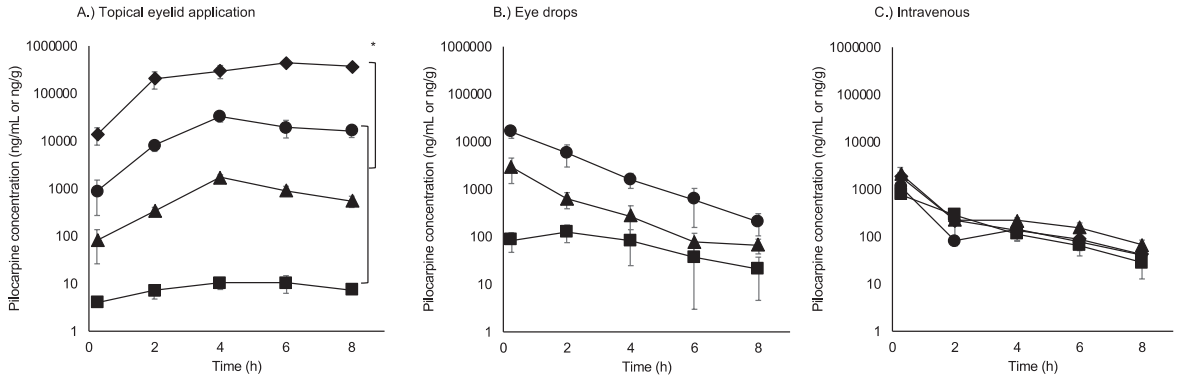

Fig. 3 Disposition of pilocarplne after topical eyelid, eye drops, and intravenous administration. Eyelid skin (◆); Conjunctiva (●); Eyeball (▲); Plasma (■). Each value represents the mean ± S.D. ($n=4$) (*$p<0.05$)[3].

tance, especially for the management of chronic ocular diseases. The direct and sustained delivery of drugs into the eyes is seen to be a practical strategy in the management of these diseases. Furthermore, the eyelid skin is proximate to the conjunctiva, which could function as a reservoir for ocular drugs. In this case, the drug may not be eliminated rapidly because it is localized in the vasculature, allowing sustained delivery of the drug into the eyeball from the eyelid skin. Of note, the conjunctiva is 2 to 30 times more permeable to drugs than the cornea, and it has a direct relation with the eyeball, the target site in most chronic ocular diseases.

We selected pilocarpine as a model drug in this investigation. Pilocarpine hydrochloride, a nonselective muscarinic receptor agonist, has long been used in the treatment of glaucoma by increasing trabecular outflow leading to reduced intraocular pressure. In ciliary smooth muscle cells, pilocarpine binds to and activates muscarinic M_3 receptors and stimulates the contraction of longitudinal ciliary muscle manifested as miosis. Current delivery systems, eye drops and ocular inserts, are available for the transport of pilocarpine into the eyes, but they are linked with disadvantages. Pilocarpine drops deliver high dose of drugs, which initially cause side effects of headache, dimmed vision due to miosis, and myopia.

There have been no investigations whether topical application onto the lower eyelid skin may be useful for the treatment of chronic eye diseases. The only evidence is in our previous work, wherein the eyelid skin was found to be more permeable to drugs. To further elucidate the value of delivering drugs onto the eyeball through the eyelid skin, pharmacokinetic studies of pilocarpine were conducted. Our pharmacokinetic results affirmed our hypothesis, and these results were verified using a direct pharmacodynamic study of pilocarpine in rats.

Among the three methods of application (Fig. 3), topical eyelid application proved to be superior in delivering pilocarpine into the eyes for an extended period. It was evident that the mean residence time (MRT) of pilocarpine in the ocular system was significantly longer compared with eye drops and the intravenous route. A longer residence time corresponded to a slower absorption rate and consequently a slower elimination process, which allowed the drug to accumulate in the specific ocular tissues for a longer period of time and exhibit its therapeutic efficacy. Thus, high drug concentrations were detected in the conjunctiva and eyeball after topical eyelid application. Notably, pilocarpine plasma concentrations were lower after topical application onto eyelid skin, suggesting ocular absorption and specific accumulation of the drug in the conjunctiva driven by a high concentration gradient in the eyelid skin and then diffusing into the eyeball. Very high concentrations of the drug in the application site, eyelid skin, remained for the whole observation time. However, the concentration started to decline in the conjunctiva and eyeball from 4 h after drug instillation. A possible depletion of drug from the application tube may have occurred, since our previous *in vitro* study showed that about 5–10% of the applied dose permeated over 4 h. Hence, a longer half-life time and MRT after eyelid skin administration in comparison to eye drop instillation are good indicators for a longer duration of therapeutic response, possibly resulting to a lower dosing frequency.

When drugs were delivered as eye drops, a very different situation was observed. Drugs penetrate the eye by absorption across the cornea from the precorneal tear film. The kinetic behavior of drugs in the tears has a direct bearing on the efficiency of drug absorption by the eye. Our study demonstrated that drugs following

eye drops rapidly entered the eyeball and exerted a pharmacologic effect, but this was short-lasting due to the short MRT and particularly fast elimination rate from the application site. Eye drops typically remain on the ocular surface for a very short period of time before being washed away by the tears into the nasolacrimal duct, a gateway for the systemic circulation. Moreover, high plasma drug concentration, as shown in this study, after eye drops indicated non-specific accumulation of the drug in the non-target organs. This phenomenon emphasizes the possible occurrence of unwanted side effects associated with pilocarpine when given as eye drops. Furthermore, in the case of intravenous route, although pilocarpine in the systemic circulation reached the ocular tissues, high doses were essential, which may lead to unwanted side effects. Thus, the ocular kinetics of pilocarpine after eye drops application is more like that after intravenous dose.

There is extensive evidence indicating differences in the permeability of skin to chemical substances depending upon the application site. Based on the previous experiments, it was shown that the skin permeability of pilocarpine was 2.6-fold higher in the case of eyelid skin than for the abdominal skin of hairless rats. In order to prove the results obtained from *in vitro* experiments in *in vivo* conditions, topical application on the abdominal skin was performed. The results of this experiments confirmed the trend observed *in vitro*, where skin permeability for pilocarpine was low for abdominal skin, whereas the eyelid skin was permeable to a greater extent. The eyelid skin, through the conjunctiva, behaves like a reservoir wherein a drug absorbed after topical application remains in the specific target organ and exerts a specific pharmacologic effect. The higher tissue-to-plasma concentration indicates that drug accumulation takes place in the specific target organ following topical application.

Direct pharmacologic observation was conducted to further confirm the results of the pharmacokinetic data. Pupil size reduction, miosis, is attributed to the pharmacologic effect of pilocarpine. Our findings showed that pupil size reduction increases over time after topical eyelid application indicating a prolonged pharmacologic effect of pilocarpine. This further confirmed localized drug action with sustained release features.

Topical eyelid skin application was shown to have the highest mean residence time and a higher drug concentration in the conjunctiva and eyeball, suggesting localized drug absorption and specific drug accumulation in the ocular tissues. Consequently, a longer duration of therapeutic response may permit less-frequent dosing.

5. Summary and Acknowledgements

In summary, the eyelid skin allows drugs to permeate at a higher rate regardless of drug lipophilicities which is attributed to the thinner size of the stratum corneum and lesser amount of neutral lipids. Topical application onto the lower eyelid skin resulted to higher drug penetration into the eyeball with the application of iontophoresis. Topical application onto the lower eyelid skin allows extended delivery of drugs into the ocular region compared to eye drops based on pharmacokinetics and pharmacodynamics data. Hence, it is rational to prepare the topical formulations directed to the eyelid skin, which is suitable for drugs requiring long-term treatment.

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