Appendix II

Analysis of drug diffusion through holes in cornea made by microneedles

Residual holes in the cornea made by microneedle insertion could increase corneal permeability to drugs, which could be of interest to improve topical drug delivery efficiency, or could increase corneal permeability to other xenobiotics, which could be of concern for safety. To address this scenario, we compared the rate of fluorescein transport across intact cornea to the rate of transport through a hole in cornea created by microneedle insertion.

The fluorescein delivery rate across intact cornea ($M_{\text{cornea}}$) can be estimated as:

$$M_{\text{cornea}} = P \cdot C \cdot A_{\text{cornea}}$$  \hspace{1cm} (6)

where $P$ is the corneal permeability to fluorescein, which has previously been measured as $5.0 \times 10^{-6} \text{cm/s}$; $C$ is the applied drug concentration, which was set as the 300ng/ml fluorescein, and $A$ is the surface area of rabbit cornea, which is $1.6 \text{cm}^2$.

The fluorescein delivery rate through a hole created in cornea by microneedle insertion ($M_{\text{mn}}$) can be estimated as

$$M_{\text{mn}} = \frac{D_{fl} \cdot C \cdot A_{\text{hole}}}{d_{epi}}$$  \hspace{1cm} (7)

where $D_{fl}$ is the diffusivity of fluorescein in water, which is $6.4 \times 10^{-6} \text{cm}^2/\text{s}$; $A_{\text{hole}}$ is the cross-sectional area of the hole created by microneedle insertion, which is assumed to be equal to the cross-sectional area of the microneedle shaft, $8.5 \times 10^{-5} \text{cm}^2$, and $d_{epi}$ is the thickness of corneal epithelium, which is $45 \mu\text{m}$ in the rabbit. This analysis assumes that the corneal epithelium is the rate limiting barrier for transcorneal transport and that there is no hindrance to diffusion in the hole created by microneedle insertion, such that fluorescein diffusivity within the hole is equal to that in free solution.
Guided by this approach, we revisited the analysis of the in vivo data on delivery of fluorescein (Figs. 4–5) and pilocarpine (Figs. 6–7). In contrast to the proposed mechanism of drug delivery, in which microneedles deposit drug within the cornea for subsequent diffusion deeper into the eye, it is possible that the microneedle coating largely washed off in the tear fluid and the increased bioavailability was due to increased transcorneal fluorescein diffusion through holes made by microneedles, which effectively increased corneal permeability. We believe, however, that this alternative mechanism is unlikely, in part because the extended release of fluorescein over many hours after microneedle delivery cannot easily be explained by transcorneal diffusion from the tear film, which renews itself every 2.5 to 10 min in the rabbit eye.\(^5\)

We extended this analysis by calculating the increase in corneal permeability that would result from microneedle penetration. Using Eq. 6 in combination with an experimental value for corneal permeability to fluorescein,\(^1\) the rate of fluorescein delivery across rabbit cornea is estimated as 8.6 ng/h, assuming a constant fluorescein concentration of 300 ng/ml in the tear film. In contrast, Eq. 7 predicts that the rate of fluorescein delivery through a hole in the cornea created by microneedle insertion is expected to be 0.13 ng/h, which is 66 times less than the flux through the rest of the cornea. This finding indicates that a single microneedle insertion by itself has little impact on corneal permeability and that insertion of 66 microneedles should be needed just to double corneal permeability. This is because even though holes made by microneedles are locally very permeable, a microneedle hole is microscopically small and covers just 0.005% of corneal surface area.

Along these same lines, one might ask why most of the drug deposited within the cornea diffused into the aqueous humor and not back out into the tear film. This is probably because diffusion through the corneal stroma and endothelium into the anterior chamber
encounters less resistance than diffusion across the corneal epithelium into the tear film. Intact epithelium is known to have much lower permeability to fluorescein than stroma or endothelium, and the above analysis indicates that the presence of a microneedle hole should not change that.

References


