A New Preparation Method for Ophthalmic Drug Nanoparticles

Noriaki Nagai and Yoshimasa Ito*
Faculty of Pharmacy, Kinki University, 3-4-1 Kowakae, Higashi-Osaka, Osaka, 577-8502, Japan

Abstract
The most challenging task in ophthalmic therapy has long been the formulation of suitable ocular drug delivery systems due to the unique structure of the eye that restricts entry of the drug molecule at the site of action. Recently, the use of nanotechnology in the ophthalmic field has gained much attention, since nanoparticulate drug delivery is considered to be one of the most promising technologies to overcome poor drug stability and the difficulties in delivering drugs across biological barriers (improvement of bioavailability). This review demonstrates the usefulness of ophthalmic formulations containing drug nanoparticles. Furthermore, in this review, we introduce a new method established in our laboratory for the preparation of drug solid nanoparticles. This information provides significant information that can be used to design further studies aimed at developing less toxic eye drops.

Keywords: Nanoparticle; Ophthalmic formulations; Corneal toxicity; Transcorneal penetration; Drug delivery

Introduction
The eye is a small and complex organ that is separated from the rest of the body by multiple layers of biological barriers. Moreover, the internal ocular structures and tissues are protected from the external environment by the tight junctions of the corneal epithelium and the mucosal surface. The ophthalmic application of drugs is the primary route of administration for the treatment of various eye diseases, and is well-accepted by patients; however, in traditional formulations, only small amounts of the administered drug penetrate the cornea to reach the desired intraocular tissue due to corneal barriers and dilution caused by lacrimation [1-3]. Consequently, there is a need for the frequent instillation of concentrated solutions to obtain the desired therapeutic effect in both the anterior and posterior hemispheres of the eye. However, the frequent administration of drugs can cause corneal damage as well as undesirable side effects resulting from the systemic absorption of drugs through the nasolacrimal duct [4,5]. Therefore, the primary challenge for ocular drug delivery is how to circumvent these protective barriers in order to achieve therapeutically effective concentrations of drugs in the intraocular tissues. In this regard, it is very important to increase the effectiveness of drugs by enhancing their bioavailability [6]. In order to overcome these problems and increase ocular drug bioavailability, several strategies including the preparation of viscous solutions, micro/nanoparticles and hydrogels have been developed and investigated [1,4,7-10]. In the case of viscous solutions, numerous studies have demonstrated that they do not possess sufficient mechanical strength to resist the ocular clearance mechanism, and offer only a transient improvement in ocular residence time [11,12]. On the other hand, it has been reported that the capability of drugs to penetrate across the cornea can be significantly improved by decreasing the particle size using nanoparticles [3,9,13-15]. Ophthalmic formulations containing drug nanoparticles present a possible solution to the limitations surrounding ocular drug penetration [16-19], and it is known that decreasing direct cellular stimulation and reducing the amount drug used by increasing its bioavailability are useful ways to circumvent the side effects related to drug delivery [6]. It is expected that ophthalmic drug systems using nanoparticles may provide an alternative strategy for increasing ocular drug penetration [16-19]. This review addresses the usefulness of ophthalmic formulations containing drug nanoparticles. In addition, we introduce the new method for the preparation of drug solid nanoparticles.

Design Considerations for Ocular Drug Nanomaterials
The size of a particle influences its functionality in terms of its uptake, residence in circulation, adherence, degradation, as well as clearance [20-24]. The fate of particles inside the body has been reported as follows: ≥ 100 nm, escape from blood vessels through the endothelial lining; ≥ 200 nm, trapped inside liver cells; ≥ 2 µm, filtered in the spleen; ≥ 100 nm, escape from blood vessels through the endothelial lining. Thus, size governs the movement of nanoparticles inside tissues. In the ophthalmic field, nanoparticles in sizes ranging from 10 to 1000 nm allow for the improved topical passage of large, water insoluble molecules through the barriers of the ocular system [25]. Superficial barriers impede direct and systemic drug access to the specific site of action. Drug loaded nanoparticles show favorable biological properties including prolonged residence time for eye drops, decreased toxicity, and increased ability of the drug to penetrate into the deeper layers of the ocular structure and aqueous humor thus minimizing precorneal drug loss caused by rapid tear fluid turnover [6,26]. Techniques were planned to transform nanoparticles from lipophilic to hydrophilic and to down-regulate irritation to the eye. Preparations that include nanoparticles could be very useful for the extended delivery of ophthalmic drugs [2,27,28]. Preparations that include nanoparticle systems have been used to deliver ophthalmic drugs to target sites in the treatment of many eye diseases as summarized in Table 1 [13,29]. An ideal ocular drug delivery system should possess key properties that include: (I) a controlled and sustained release profile to maintain a therapeutic concentration of the drug over a prolonged period of time to reduce the frequency of administration; (II) specific targeting and prolonged retention in the diseased tissues to improve therapeutic efficiency and mitigate side effects; and (III) patient-friendly delivery routes that minimize or eliminate side effects resulting directly from these administration methods. At present, nanoparticle-based ocular drug delivery systems appear to be the most promising tool to meet the...
primary requirements of an ideal ocular delivery system.

**Preparation of Ocular Drug Nanomaterials**

Previous research over several decades focused on two major approaches to the design of nanocarriers: bottom-up synthesis and a top-down approach. Bottom-up synthesis, which is based on self assembly and emulsion systems, has been studied extensively in the past, and a variety of potential nanocarriers have been developed using this method, for example, polymeric nanoparticles, micelles, liposomes, nanoemulsions, dendrimers, biodegradable and non-biodegradable carriers, solid lipid nanoparticles, magnetic nanoparticles etc. A majority of these carriers are colloidal systems governed by different forces such as hydrophobic interactions, Van der Waals forces, hydrogen bonding, and ionic interactions. With this approach, high polydispersities are often exhibited, and the systems developed sometimes present certain limitations. In the in vivo drug release profiles, the physicochemical and degradation kinetics of these carriers are difficult to evaluate and reproduce as they are variable. On the other hand, major advancements have been made recently in the fabrication technology by the introduction of the “top-down” approach for micro and nanofabrication systems using electromechanical techniques. This approach exhibits the potential for designing nanoparticles with precision in terms of particle shape and size. Such an approach can provide control over particle size, functionality, and precise particle geometry. This approach may also resolve the limitations of the bottom-up approach in research.

We have also designed ophthalmic formulations containing drug nanoparticles obtained by mill methods [30,31]. Ophthalmic solutions containing indomethacin (IMC) solid nanoparticles (IMCnano) were prepared using zirconia beads and Bead Smash 12 (a bead mill, Wakenyaku Co. Ltd, Kyoto, Japan) (Figure 1). Briefly, zirconia beads (diameter: 2 mm) were added to IMC microparticles (solid) containing BAC, mannitol and methylcellulose (MC), and the mixtures were crushed with the bead mill for 30 sec (3,000 rpm, 4°C). The mixtures were dispersed in saline with or without 5% 2-Hydroxypropyl-β-cyclodextrin (HPβCD), and crushed again with the bead mill (5,500 rpm, 30 sec×15 times, 4°C) using smaller zirconia beads (diameter: 0.1 mm). The compositions of the dispersions containing IMC are shown in Table 2. 0.5% IMC is equivalent to 14.0 mM IMC; the pH was 6.5 for both ophthalmic dispersions containing IMC micro- or nanoparticles. The IMC particle size reached the nano order by the bead mill method while those instilled with IMCnano showed 48.1% healing 12 hr after preparation. Furthermore, the IMCnano preparations showed high antimicrobial activity approximately equal to that of a 0.001% BAC solution, a preservative used in the ophthalmic field.

**Usefulness of Ophthalmic Formulations containing Drug Solid Nanoparticles obtained by Bead Mill Methods**

It is very important to elucidate the corneal toxicity and permeability of ophthalmic solutions containing IMC solid nanoparticles (IMCnano). Therefore, we evaluated the transcorneal penetration of IMCnano and its effects on corneal damage using human corneal epithelial cells (HCE-T), and rat and rabbit corneas [30]. The corneal wounds of rat eyes instilled with commercially available IMC eye drops (INDOMELOL® eye drops, Senju Pharmaceutical Co., Ltd, Osaka, Japan) showed 21.6% healing while those instilled with IMCnano showed 48.1% healing 12 hr after corneal epithelial abrasion. In addition, the viability of HCE-T cells...
treated with IMCnano was significantly higher than that of cells treated with commercially available IMC, pranoprofen (NIFLAN® eye drops, Senju Pharmaceutical Co., Ltd, Osaka, Japan), diclofenac (DICLOD® eye drops, WAKAMOTO Co., Ltd., Tokyo, Japan), bromfenac (BRONUK® eye drops, Senju Pharmaceutical Co., Ltd, Osaka, Japan) or nepafenac (NEVANAC® eye drops, ALCON Japan Ltd., Tokyo, Japan), and the accumulation of IMC in HCE-T cells treated with IMCnano was less than that in cells treated with commercially available IMC. On the other hand, the penetration of IMC from IMCnano was higher than from commercially available IMC eye drops in HCE-T cell monolayers. Furthermore, we have demonstrated the corneal penetration of ophthalmic solutions containing solid nanoparticles using rabbit corneas, which include corneal epithelial, stromal and endothelial cells (in vitro and in vivo transcorneal penetration experiments). In the in vitro study, the penetration coefficients through the cornea and the cornea/preparation partition coefficients were higher, and the diffusion constants within the cornea lower than those for commercially available IMC eye drops. The IMC penetration rate from IMCnano (18.9 ± 1.95 nmol/cm²/h, n=5) was significantly higher in comparison with commercially available IMC eye drops (5.43 ± 2.59 nmol/cm²/h, n=5). In the in vivo study, no IMC from commercially available IMC eye drops was detected in the aqueous humor until 50 min after administration; however, the lag time from IMCnano eye drops was ca. 25 min. The penetration rate from IMCnano was higher than those from commercially available IMC eye drops, and the penetration rate from IMCnano and the area under the IMC concentration-time curve (AUCnano) were approximately 10.7 times greater in comparison with commercially available IMC eye drops. These results show that nanoparticle formulations provide a reduction in corneal toxicity, and may make it possible to decrease the amount of drug used via an increase in bioavailability, thus resulting in a reduction in drug toxicity.

Conclusion

In this review, we introduce the applications of nanoparticles in the ophthalmic field. Future work should focus on investigating combinations of additives to enhance the favorable properties of nanoparticles. It will be interesting to observe what benefits these innovative drug preparations can provide to patients. In addition, we describe a new method for preparing drug solid nanoparticles established by us [30-32]. The particle size of our drug solid nanoparticles is of high quality (particle size, approximately 60-80 nm). Dispersions containing these drug nanoparticles are tolerated better by human and rat corneal epithelial cells than commercially available eye drops, since the accumulation of drug in nanoparticles is lower than from solutions. Furthermore, the state of the dispersions containing drug solid nanoparticles does not affect the antimicrobial activity of BAC against E. coli, and the corneal penetration of drug solid nanoparticles is significantly better than that from commercially available eye drops. The cost-effectiveness of the new preparation method is also very high, since IMCnano can be prepared in within 2 hours, and this cost for preparation is only reagent cost. It is expected that ocular drug delivery systems using drug nanoparticles may expand their usage for therapy in the ophthalmologic field.

References

Submit your next manuscript and get advantages of OMICS Group submissions

Unique features:
1. User friendly/feasible website-translation of your paper to 50 world’s leading languages
2. Audio Version of published paper
3. Digital articles to share and explore
4. Quality and quick editorial, review and publication processing
5. 30,000 editorial team
6. 350 Open Access Journals
7. Digital articles to share and explore
8. Audio Version of published paper
9. User friendly/feasible website-translation of your paper to 50 world’s leading languages
10. Quality and quick editorial, review and publication processing
11. 30,000 editorial team
12. 350 Open Access Journals
13. Digital articles to share and explore
15. User friendly/feasible website-translation of your paper to 50 world’s leading languages
16. Quality and quick editorial, review and publication processing
17. 30,000 editorial team
18. 350 Open Access Journals
19. Digital articles to share and explore
20. Audio Version of published paper
21. User friendly/feasible website-translation of your paper to 50 world’s leading languages
22. Quality and quick editorial, review and publication processing
23. 30,000 editorial team
24. 350 Open Access Journals
25. Digital articles to share and explore
26. Audio Version of published paper
27. User friendly/feasible website-translation of your paper to 50 world’s leading languages
28. Quality and quick editorial, review and publication processing
29. 30,000 editorial team
30. 350 Open Access Journals
31. Digital articles to share and explore
32. Audio Version of published paper
33. User friendly/feasible website-translation of your paper to 50 world’s leading languages
34. Quality and quick editorial, review and publication processing
35. 30,000 editorial team
36. 350 Open Access Journals
37. Digital articles to share and explore
38. Audio Version of published paper
39. User friendly/feasible website-translation of your paper to 50 world’s leading languages
40. Quality and quick editorial, review and publication processing
41. 30,000 editorial team
42. 350 Open Access Journals
43. Digital articles to share and explore
44. Audio Version of published paper
45. User friendly/feasible website-translation of your paper to 50 world’s leading languages
46. Quality and quick editorial, review and publication processing
47. 30,000 editorial team
48. 350 Open Access Journals
49. Digital articles to share and explore
50. Audio Version of published paper

Special features:
1. 350 Open Access Journals
2. 30,000 editorial team
3. 21 days rapid review process
4. Quality and quick editorial, review and publication processing
5. Indexing at PubMed (partial), Scopus, EBSCO, Index Copernicus and Google Scholar etc
6. Sharing Option: Social Networking Enabled
7. Authors, Reviewers and Editors rewarded with online Scientific Credits
8. Better discount for your subsequent articles

Submit your manuscript at: http://www.editorialmanager.com/virology


Pharm Anal Acta
ISSN: 2153-2435 PAA, an open access journal
Volume 5 • Issue 7 • 1000305