CHAPTER-2

LITERATURE REVIEW
2.1. Literature review on microemulsion for ocular application

Gasco and his coworkers (Gasco et al. 1988) reported timolol microemulsion to enhance corneal permeability by formation of ion pair of timolol with octanoic acid (OA). Owing to the enhanced lipophilicity of ion pair, significantly improved the timolol penetration across corneal surface. These authors utilized the same theory and compared the permeability of timolol octanoate ion pair with timolol without an ion pair in microemulsion and aqueous timolol solution. They reported the stability of timolol microemulsion which suggested excellent thermodynamic stability of microemulsion. The droplet size and viscosity of formulated microemulsion remain unchanged upon ultracentrifugation and freeze thawing. In the performed animal experiments on rabbits, the animals were randomized into three groups each containing eight rabbits. After topical instillation of three timolol formulation, maximum timolol concentration achieved within 15mins. The aqueous humor concentration of timolol was 3 and 4 times higher for the microemulsion and ion pair respectively than for timolol alone. The AUC was 3.5 times higher for microemulsion and 4 times higher for ion pair solution than that of timolol alone. The presence of a physiological substance, egg lecithin, as a surfactant, enabled a therapeutic device to be made with good biocompatibility.

Some of the authors (Gallarate et al. 1993) extended the previous findings (Gallarate et al. 1988), (Gasco et al. 1989) and developed levobunolol octanoate ion pair incorporated in microemulsion. The partition coefficient experiment revealed an increase in apparent permeability of levobunolol upon the increasing molar ratio of OA/levobunolol. Polyethelene glycol 200 (PEG 200) was incorporated to enhance the permeability coefficient of levobunolol octanoate ion pair. In-vitro permeability experiments displayed 6 times higher permeability of levobunolol & OA ion pair through the hydrophilic lipophillic double membrane than that of levobunolol alone. PEG 200 also impacted significantly by the increase in the permeability coefficient of the ion pair. Results of in-vitro permeability experiments were further extended on the hairless mouse skin which confirmed presence of OA as permeation enhancer with improved levobunolol permeability 4 times in case of solution and 2 times in case of microemulsion.

Benita & Levy (Benita and Levy 1993) reported the presence of positive charge on the surface of internal phase that could influence drug absorption through corneal penetration. The supposition was based on negative charge present on the corneal surface which would facilitate binding the
positively charged droplets of the submicron emulsion. Later on the earlier supposition was rechecked (Calvo et al 1996) and a comparative behavior of drug release through colloidal systems namely nanocapsules, nanoparticles and submicron emulsion, the findings of which showed an increased corneal permeation of indomethacin was due to incorporation of the drug in colloidal carriers and not because of the electrostatic attraction between the negatively charged cornea and positively charged drug carrier system. They found that the incorporation of the drug in a colloidal system facilitates uptake of nanoglobules by the corneal epithelium without causing any damage to the cell membrane.

Calvo and his coworkers reported (Calvo et al 1996) that microemulsion made up of lecithin along with esters of fatty acids exhibited a degradative process upon storage indicating change in droplet size, change in pH. The possible mechanism for the degradation was due to the hydrolysis of triglyceride and phospholipid moieties leading to release of free fatty acids. A small decrease in the zeta potential was observed during storage, and this was probably due to reduction in ionization of the anionic phospholipids at the oil-water interface or to the phospholipid degradation resulting in an increased amount of free fatty acid.

In order to validate the extension of the residence time of the microemulsion in the conjunctival sac, (Beilin et al. 1995) some authors added a fluorescent marker to the formulations. After the administration of the eye drops at intervals of 1, 2, 3, 5, 10 and 15 min, 200 ml of saline solution was used to rinse the surface of the eye and the intensity of the fluorescence was determined. It was found that the intensity was higher for the microemulsion dosage form compared to regular eye drop. One minute after the administration, 39.9 ± 10.2% of the fluorescence was measured for the microemulsion as compared to only 6.8 ± 1.8% for regular eye drops. Furthermore, the areas under the curves of the pupillary diameters were 7.5 ± 0.7 mm/h for the microemulsion dosage form and 5.7 ± 0.6 mm/h for regular eye drops. The microemulsion eye drops provide a much more delayed pharmacological action due to a delayed residence time in the conjunctival sac following a more significant contact time with the cornea.

Soon after couple of years two researchers Hase and Keipert (Haße and Keipert, 1997) investigated the mitotic activity of pilocarpine nitrate incorporated in microemulsion in albino rabbits. The developed pilocarpine microemulsion demonstrated physiological acceptability with no signs of irritation, bleeding, vessel lysis and coagulation in the hen's egg chorioallantoic
membrane (HET-CAM) test stating the formulation nonirritating to the mucus membrane. *In vitro* release kinetics of pilocarpine microemulsion performed using hydrophilic membrane as separating membrane between donor and acceptor compartment exhibited a delayed release of the pilocarpine. The microemulsion significantly reduced release of up to 21% less of pilocarpine than the aqueous solution after 6hr. Microemulsion showed miotic effect of pilocarpine lasting up to 5 - 6.5hrs whereas with aqueous pilocarpine remained for upto 3.5 hrs respectively.

Later on some researchers (Fialho & Cunha 2004) developed microemulsion based vehicle for topical ocular delivery of water insoluble drug dexamethasone which is otherwise formulated as a suspension or ointment for ophthalmic use. The microemulsion prepared by titration with cosurfactant and was acceptably tolerated by the rabbit eye without showing any signs of redness or inflammation. The microemulsion remained stable at a temperature of 4°C, 25°C, 37°C for three months without any significant changes in viscosity and droplet size. The animal experiments carried out on male New Zealand white rabbits which showed AUC of concentration of dexamethasone proportional to time of topical application of dexamethasone microemulsion are more than double that of the conventional dexamethasone preparation. With microemulsion achieved peak drug concentration (T\textsubscript{max}) within 30 min in contrast conventional formulation achieved T\textsubscript{max} at 1hour. The C\textsubscript{max} values were 2 fold higher for microemulsion as compared to conventional formulation, thus providing strong evidence of the prolonged effect of the corticosteroid ocular delivery through microemulsion.

In another study involving (Lv et al. 2006) cosurfactant free microemulsion based on non-Ionic food grade surfactants and evaluated for improved stability of chloramphenicol incorporated in the developed microemulsion. Chloramphenicol drug undergoes hydrolytic degradation into glycols in aqueous solution. In microemulsion chloramphenicol molecule gets incorporated into the oil core or in the palisade of hydrophilic part of surfactant group. The accelerated stability experiments showed 27.11% w/w glycols released after 3 months in aqueous solution. However 14.38% w/w of glycols was released in the microemulsion. This indicated strong evidence that microemulsion improved stability of chloramphenicol remarkably.

More recently some authors (Alany et al. 2006) put focus on the ocular irritation potential of some non ionic surfactants sorbitan mono laureate, polyoxyethylene sorbitan mono-oleate using a HET-CAM test (Luepke 1985) and developed microemulsion formulation with and without use
of cosurfactant 1-butanol (Leighton et al. 1985). The usefulness of the HET-CAM test to assess the irritation potential of surfactant based hydro-alcoholic formulations is well established and it is reported that protocols with short exposure times seem to provide more accurate results compared to others which require longer exposure times (Herzinger et al. 1985). The non-Ionic surfactants, sorbitan mono laureate and polyoxyethylene sorbitan mono-oleate with ethyl oleate were found to be practically non-irritant when applied to the surface of the CAM.

Chan and his coworkers (Chan et al. 2007) extended the previous finding of ocular irritation potential of non-ionic surfactants and oils (Alany et al. 2006) and collaborated the phase transition phenomenon with the ocular retention characteristics of water in oil type microemulsion based phase transition systems of pilocarpine hydrochloride. The w/o microemulsion undergoes phase transition after instilling in the eye. A cascade of events takes place after instillation of the w/o microemulsion. Initially it gets diluted with lacrimal fluid resulting in the structuring of the system & formation of liquid crystals of high viscosity. It is remarkably seen that liquid crystal phase offers matrix like a reservoir of drug sustaining the drug release. The proposed mechanism is based on the adsorption of the nanodroplets representing the internal phase of the microemulsion, which constitutes a reservoir of the drug on the cornea and limit their drainage. The in-vitro results highlighted the occurrence of phase transition of microemulsion after topical application to the eye by dilution of tears.

However the natural defense mechanism of the eye allows the secretion of upto 400µl of tears for removal of any foreign substance from the ocular surface. In the phase transition microemulsion drug delivery involves dilution of microemulsion by reflux tear secretion. As a result repeated administration of phase transition microemulsion might hamper the natural defense mechanism of the eye which is designed for removal of foreign particles however is potentially exploited for therapeutic purpose. Thus it needs to be further investigated for the long term effects on the ocular tear kinetics upon ocular therapeutic utilization of such formulation.

Earlier many workers have reported microemulsion composed of surfactant lecithin, isolecithin, poloxamers, polyethylene glycols, brij 96, sorbitan esters of fatty acid with the oily portion composed of esters of fatty acids such as isopropyl myristate, isopropyl palmitate, ethyl oleate, medium chain triglycerides. The perceived ocular toxicity was minimal, drug release and corneal penetration were significantly higher than the aqueous solution of the drug. Lecithin was claimed
to be highly promising and biocompatible surfactant ingredient of the microemulsion. Its ocular toxicity and irritation potential was found to be negligible making it highly suitable candidate as a surfactant in the microemulsion.

Later on a group of researchers (Baspinar et al, 2008) prepared ocular microemulsion based on non ionic surfactants and triacetin bearing everolimus for preventing corneal-graft rejection. The permeation rate of the model drug everolimus through a freshly isolated pig cornea was determined ex-vivo. Authors concluded that, prepared microemulsion is a promising ocular formulation for preventing corneal-graft rejection.

Sergio and his co workers filed patent (Sergio et al, WO 154985A1, 2011) in which they claimed to developed o/w microemulsion for encapsulation of water insoluble drugs for topical ophthalmic application. The developed microemulsion carrier remained stable for period of 6 months displaying a particle size of 15nm without any signs of instability or separation.

Gobel (Gobel, European patent EP- 2485714A1, 2012) filed another patent for developing a transparent o/w microemulsion for delivery of immunosuppressant agent tacrolimus and is subjected to HET-CAM test thereof claimed to be free from signs of irritation. The particle size range varied from 5-100nm. Additionally the tacrolimus microemulsion was found to penetrate efficiently the stratum corneum tissue and reach the dermis due to presence of lymphocyte, which is the target for the active ingredient.

Carli and coworkers filed patent (Carli et al., US Patent US 8414904B2, 2013) for o/w microemulsion composed of prostaglandin formulated with two non ionic surfactant and one oily component that displayed a particle size not more than 700nm and a low zeta potential of +2 to -2 due to use of non ionic surfactants as emulsifying agents. The formulation was claimed to be free from any signs of irritation on rabbits eye. The microemulsion remained stable for a period of 12 months

Recently some researchers (Kesavan et al, 2013) developed mucoadhesive chitosan coated cationic microemulsion for treatment in conditions of chronic uveitis. The average globule size was less than 200 nm with a positive surface charge. The developed microemulsion revealed
stability for 3 months. The \textit{in-vivo} studies evidenced marked improved therapeutic effect of the incorporated steroid.
[BIBLIOGRAPHY]


