

Review Article

NON-STEROIDAL ANTI-INFLAMMATORY DRUGS FOR TOPICAL OPHTHALMIC ADMINISTRATION: CONTEMPORARY TRENDS

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ABSTRACT

Topically applied drugs for the treatment of ocular inflammation are the most commonly used formulations due to many reasons: they are simple to use; they can be applied often and provide a high drug concentration; systemic side effects which are associated with oral administration can be avoided. But due to the physiological limitations of the eyes only a small number of anti-inflammatory agents, which have certain physico-chemical properties can be included in appropriate and efficient formulations for treatment of ocular inflammation. To prepare the optimal therapeutic and technological ophthalmic formulation, it is required to know the possibility of enhancing the bioavailability in the ocular tissues and to increase the therapeutic activity of the active substance, by using appropriate technological approaches to create a stable, tolerable and effective ophthalmic drug formulation. In this review, we focus on microemulsions, polymeric NPs, liposomes, SLNs, and nanosuspensions as formulations incorporating non-steroidal anti-inflammatory drugs (NSAIDs) for topical ophthalmic application.

Keywords: NSAIDs, Nanoparticle-drug delivery systems, Topical ophthalmic administration.

INTRODUCTION

The various ophthalmic diseases are usually treated with topically administered drug formulations. Their disadvantage is the short time of contact with the eye, which leads to a low degree of absorption of the active substance [1]. This requires frequent administration, which usually leads to a high systemic exposure. These problems could be solved by looking for appropriate technological methods to increase the efficiency of the ophthalmic formulations. The strict requirements, which they must fulfill, are related to the specific sensitivity of the eye and its anatomical and physiological characteristics [2-5]. To prepare the optimal therapeutic and technological ophthalmic formulation, it is required to know the possibility of enhancing the bioavailability in the ocular tissues and to increase the therapeutic activity of the active substance, by using appropriate technological approaches to create a stable, tolerable and effective ophthalmic drug formulation [6-13].

Inflammation is a cellular and vascular response of the tissues to injury caused by physical or chemical agents, an invasion of pathogens, ischemia, excessive (hypersensitivity) or inadequate (autoimmune) functioning of the immune mechanisms. Inflammation facilitates the immune response and the subsequent removal of the antigenic material and the damaged tissue. From the moment the trauma is recognized by the organism, in the body start mechanisms for locating own and foreign substances and damaged tissues. The response is amplified by the activation of inflammatory cells and the production of chemical mediators such as acidic lipids-prostaglandins, thromboxanes, leukotrienes, vasoactive amines, cytokines and others [14]. Acidic lipids are produced in the arachidonic cascade. Arachidonic acid is released from a phospholipid component of the cell membranes under the influence of phospholipase A2. The produced arachidonic acid takes cyclooxygenase or lipoxygenase pathway. The activation of the cyclooxygenase pathway leads to the formation of prostaglandins and thromboxanes, while the lipoxygenase pathway results in the production of eicosanoids and leukotrienes [14, 15]. The effects of the prostaglandins in the eyes are manifested in different ways [15, 16]. They affect the intra ocular pressure: it can be increased by increasing the local vasodilatation and the water permeability (increased vascular permeability) or reduced by increasing the uveoscleral outflow. Furthermore, prostaglandins have an effect on the smooth muscles of the iris and cause miosis [15, 16].

Potent anti-inflammatory drugs are the corticosteroids and the NSAIDs. They have different mechanisms of action. Corticosteroids

block the phospholipase A2 enzyme and inhibit the release of arachidonic acid. Thus, the production of all prostaglandins, thromboxanes and eicosanoids is suppressed [16]. On the other hand NSAIDs have an anti-inflammatory effect by inhibiting the enzyme cyclooxygenase (COX 1 and COX 2) [14, 15]. Using anti-inflammatory drugs is a basic pharmacological approach for the treatment of inflammation.

Topically applied drugs for the treatment of ocular inflammation are the most commonly used formulations due to many reasons: they are simple to use; they can be applied often and provide a high drug concentration; systemic side effects which are associated with oral administration can be avoided. But due to the physiological limitations of the eyes only a small number of anti-inflammatory agents, which have certain physico-chemical properties can be included in appropriate and efficient formulations for treatment of ocular inflammation. Corticosteroids are usually used as supporting agents for the topical treatment of ocular inflammation [17], but their potent anti-inflammatory effect is offset by serious side effects such as increased intraocular pressure, cataract progression, increased risk of infection and others [18]. NSAIDs have proven their safety and effectiveness and they are an alternative to corticosteroid therapy in the topical treatment of ocular inflammation [19]. Currently this therapeutic group has been widely used in topical formulations for the prevention of operational miosis, postoperative inflammation, treatment of seasonal allergic conjunctivitis, prevention and treatment of cystoid macular edema and for pain control after keratectomy [14, 15, 18, 19]. It has also been proved that NSAIDs are useful in the reduction of bacterial colonization on the lenses and that they participate in the prevention of bacterial adhesion on the epithelial cells of the cornea [20]. Although NSAIDs have a potent cyclooxygenase inhibitory activity their topical administration in ophthalmology is limited to the relatively water-soluble indole acetic acid, aryl acetic acid, aryl propionic acid and derivatives of the enolic acid [21]. Most of the NSAIDs are drugs with weak acidic properties, which ionize at the pH of the tear fluid and thus have a limited permeability through the anionic cornea, which isoelectric point is 3.2 [2-4]. The production of formulations with lower values of pH increases the non-ionized fraction of the drug, improving the drug penetration. The lower pH also causes a discomfort for the patient due to irritation of the cornea and increased excretion of tear fluid. In addition, due to their anionic nature, NSAIDs are susceptible to forming insoluble complexes with the cationic quaternary ammonium preservatives such as benzalkonium chloride [6, 22]. These are major technological

problems and great challenges for the development of ophthalmic formulations.

Topical route represents a safer administration. Therefore a major challenge to the scientists is to overcome the ocular barriers and reach the tissue target. In order, to overcome these problems, nanotechnology involving drug-loaded nanocarriers has been proposed as ophthalmic drug delivery systems that may control drug release and maintain therapeutic levels over a prolonged period of time. The success of nanocarriers for ocular drug delivery may depend on optimizing lipophilic-hydrophilic properties of the carrier-drug system, optimizing rates of biodegradation and safety. Polymers used for the preparation of nanocarriers should be mucoadhesive and biocompatible. The choice of polymer plays an important role in the release kinetics of the drug from a nanocarrier.

Classification of NSAIDs

NSAIDs is a heterogeneous group of compounds with different structural classes, which do not include a steroid nucleus derived

biosynthetically from cholesterol in their chemical structure. Fig. 1 shows the general chemical structure of NSAIDs [23].

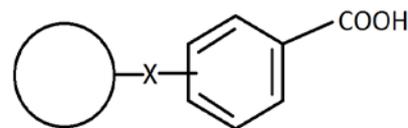


Fig. 1: General structure of NSAIDs

A classification of some popular representatives from this medicine's group according to their chemical structure is presented in table 1 [23, 24]. The classification, based on selective/non-selective action towards COXs is popular and commonly used (table 2) [24]. The classification according to their chemical structure is used in this review.

Table 1: Classification of some NSAIDs according to their chemical structure

General chemical group	Sub-group	Representatives
Carboxylic acids	<i>Salicylic Acids & Esters:</i>	Acetylsalicylic Acid, Diflunisal, Choline Magnesium Trisalicylate, Sodium Salicylate, Olsalazine, Salsalate, Sulfasalazine
	<i>Salicylates</i>	
	<i>Acetic Acids:</i> Alkanones	Nabumetone
	<i>Acetic Acids:</i>	Diclofenac, Aceclofenac, Bromfenac
	<i>Phenylacetic Acids</i>	
	<i>Acetic Acids:</i>	Indene and Indole Acetic Acids: Sulindac, Indomethacin, Etodolac;
Enolic Acid derivatives	<i>Aryl and Hetero-aryl-acetic Acids</i>	The Pyrrole Acetic Acids: Tolmetin, Ketorolac;
	<i>Propionic Acid Derivatives</i>	Ibuprofen, Dexibuprofen, Fenoprofen, Carprofen, Flurbiprofen, Ketoprofen, Dexketoprofen, Naproxen, Loxoprofen, Oxaprozin
	<i>Anthranilates</i>	Mefenamic Acid, Meclofenamate
COX-2 Selective Inhibitors	<i>Oxicams</i>	Piroxicam, Meloxicam, Tenoxicam, Droxicam, Lornoxicam
	<i>Phenylpyrazolones</i>	Phenylbutazone, Oxyphenbutazone
Anilides		Celecoxib, Rofecoxib, Valdecoxib, Etorixocib, Lumiracoxib, Firocoxib, Parecoxib
Sulfonanilides		Acetaminophen, Phenacetin
		Nimesulid

Some of the drugs shown in table 1 have not yet found application in ophthalmology. Poor aqueous solubility, stability of the drug, a low degree of absorption and irritating effect on the cornea are some of the reasons.

Table 2: Classification of some NSAIDs according to their COXs selectivity

Selectivity	Drugs
Weak COX inhibitors	Choline Magnesium Trisalicylate, Sodium Salicylate, Olsalazine, Salsalate, Sulfasalazine
COX-1/COX-2 inhibitors	Piroxicam, Sulindac, Indomethacin, Tolmetin, Ketorolac, Ibuprofen, Dexibuprofen, Fenoprofen, Carprofen, Flurbiprofen, Ketoprofen, Dexketoprofen, Naproxen, Loxoprofen, Oxaprozin, Diclofenac
COX-2 preferential inhibitors	Nimesolid, Meloxicam
COX-2 selective inhibitors	Celecoxib, Rofecoxib, Valdecoxib, Etorixocib, Lumiracoxib, Firocoxib, Parecoxib
COX-3 inhibitors	Acetaminophen

NSAID-loaded nano-and microcarriers for topical ophthalmic administration

Carboxylic acids

Salicylic acids and esters

Acetylsalicylic acid (ASA) is the main representative of the sub-group of the salicylic acids and their esters. ASA (2-acetoxybenzoic acid) is slightly soluble in water and freely soluble in alcohol [6, 22]. By varying ASA: albumin ratios from 0.06 to 1.0 stable ASA loaded albumin nanoparticles (NPs) have been obtained by coacervation. The NPs were with sizes from 46.8 nm to 190.8 nm respectively and had low polydispersity. NP formulations have released ASA at a sustained rate for prolonged duration (50% total cumulative percentage at the end of 20 hrs, 90% at 72 hrs). Thus produced NPs could be applied as intraocular release agents for diabetic retinopathy [25].

Acetic acids

The main representatives of this sub-group, which find application in topical ophthalmic formulations, are derivatives of phenylacetic acids and carbo-and hetero-cyclic acetic acids.

i) Phenylacetic acids

Diclofenac (2-(2,6-dichloranilino) phenylacetic acid) is an aryl-acetic acid derivative in the group of NSAIDs. It is mainly employed for the inhibition of intraoperative miosis and post-operative acute and chronic nonbacterial inflammations of the anterior part of the eyes in cataract surgery. Diclofenac is used as either the sodium or potassium salt [6, 22].

Agnihorti *et al.* have prepared polymeric NP suspensions from poly (lactide-co-glycolide) and poly(lactide-co-glycolide-leucine) loaded with diclofenac sodium (DS), with the aim of improving the ocular availability of the drug [26]. The NP system has shown an interesting size distribution suitable for ophthalmic application. Polymer NPs seemed to be devoid of any irritant effect on cornea, iris and conjunctiva for as long as 24 hours after application according to the modified Draize test. *In-vitro* release tests have shown an extended-release profile of DS from the NPs. Thus apparently the NP suspensions from poly (lactide-co-glycolide) and poly (lactide-co-glycolide-leucine) polymers are suitable inert carrier for ophthalmic drug delivery of DS.

Ahuja *et al.* [27] have studied the influence of the pharmaceutical factors on the absorption of diclofenac from an experimental model and from an aqueous ophthalmic formulation which is commercially available, using cornea from goat.

Solid lipid NPs (SLNs) have been prepared with a combination of homolipid from goat (goat fat) and phospholipid, and evaluated for DS delivery to the eye using bio-engineered human cornea, produced from immortalized human corneal endothelial cells (HENC), stromal fibroblasts and epithelial cells *CEPI 17 CL 4* [28]. Encapsulation efficiency has been high and sustained release of DS and high permeation through the bio-engineered cornea has been achieved. Results obtained by Attama *et al.* have shown that permeation of DS through the cornea construct has been improved by formulation as SLN modified with phospholipid.

Also enhancement in ocular delivery of DS and sustained drug release for 6 h *in vitro* have been achieved with DS loaded SLNs using a microemulsion technique [29].

DS-loaded Eudragit RS 100-based nanosuspensions have been fabricated by employing oil-in-water emulsion solvent evaporation technique [30]. The prepared NPs have revealed a mean particle size in a narrow range of 145.2 to 215.3 nm, high values of the zeta potential (ZP) of +32.6 to +49.6 mV, and also high drug entrapment efficiency in the range of 64.55% to 98.50%. The nanosuspensions have been observed to provide sustained *in vitro* drug release behaviour.

The absence of irritation towards the ocular structures has been verified by applying placebo nanosuspensions to the rabbit eye and a modified Draize test has been performed. Polymeric NPs seemed to be devoid of any irritant effect on cornea, iris and conjunctiva for as long as 24 hrs after application. It has been observed that the drug release have matrix diffusion controlled and the release mechanism was non-Fickian. Eudragit RS 100 nanosuspensions loaded with DS may represent an efficacious vehicle for controlled ophthalmic delivery.

In another study, the ocusert has been prepared as membrane permeation controlled devices with the drug loading sufficient enough for 'Once a day' therapy. DS ocusert has been prepared using the polymers hydroxy propyl methyl cellulose (HPMC) and hydroxy propyl cellulose (HPC) and dibutyl phthalate (DBP) as plasticizer by solvent casting technique [31]. The prepared DS ocusert (thickness 0.217 mm, weight uniformity 21.53 mg, and drug content about 0.25 mg) has shown prolonged zero order release and increased contact time. The rabbit eyes subjected to ocular toxicity test have not shown any signs of irritations or inflammations.

In a similar study ocular inserts of DS using different polymers such as HPMC, Eudragit L100 at various concentrations and combinations using DBP as plasticizer have been prepared [32]. The optimized formulation has shown anomalous diffusion predominantly with first order kinetics.

These are just some of the many examples in the literature which show conclusively that the incorporation of diclofenac into various carriers such as polymer NPs, SLNs, nanosuspensions or intraocular inserts could lead to increased contact time, high permeation through the cornea and provide sustained *in vitro* drug release behaviour, without any signs of ocular toxicity.

Acetoclofenac (2-[2-[2-(2,6-dichlorophenyl)amino]phenyl]acetyl] oxy acetic acid) is the glycolic acid ester of diclofenac.

Acetoclofenac loaded PLGA NPs were prepared by spontaneous emulsification solvent diffusion method (SESD) [33]. *In-vitro* release studies in phosphate buffer (pH 7.4) showed extended drug release and fitted the theoretical target release profile. Most formulations exhibited Fickian diffusion drug release profiles. The optimized formulation, prepared using 94.07 mg acetoclofenac, 400 mg poly(lactic-co-glycolic acid) (PLGA) and 4.72% poly(vinyl alcohol) (PVA), had entrapment efficiency 67.72%, a mean particle size of 593 nm and exhibited a prolonged release ($t_{50\%}=5.23$ h).

The SLN gel formulations of the dispersions were compared to the SLN dispersions and with the marketed gel of acetoclofenac [34]. The SLNs were prepared by high speed homogenization and ultrasonication method with the fixed amount of acetoclofenac (10%) and pluronic F68 (1.5%). Permeation rate and controlled release property of xanthan gum loaded SLN gel formulations and SLN dispersion was studied through excised pig skin for 24 hrs. The drug release of SLN gel formulations was better controlled as compare to SLN dispersions. *In vivo* anti-inflammatory study showed that the action of acetoclofenac was enhanced through SLN dispersion and gel formulations.

Eudragit RL 100-based NPs of acetoclofenac obtained by nanoprecipitation have shown sustained *in vitro* drug release which followed the Higuchi square-root kinetics [35]. In the *in vitro* permeation study, the NP formulation showed 2-fold higher permeation of the drug through excised cornea compared to an aqueous solution of the drug with no signs of corneal damage. The *in vivo* studies involving arachidonic acid-induced ocular inflammation in rabbits revealed significantly higher inhibition of polymorphonuclear leukocytes migration ($p<0.05$) and lid closure scores by the NP formulation compared with the aqueous solution. The formulation was quite stable to ensure two year shelf life at room temperature.

Bromfenac is another representative of this group. The high degree of penetration and potency of bromfenac can be attributed to the halogenation of the molecule: by adding a bromine moiety the NSAIDs becomes highly lipophilic which allows rapid, sustained drug levels in the ocular tissues.

Bromfenac encapsulated chitosan/sodium alginate mucoadhesive NPs for sustained ocular application, optimized using experiments by employing a 3-factor, 3-level Box-Behnken statistical design, have exhibited a biphasic drug release profile with an initial burst followed by a very slow drug release [36]. The ocular pharmacokinetics of NPs and marketed formulation has been evaluated in rabbits. The NPs exhibited significant mucin adhesion. In comparison to the marketed suspension, the NP formulation has exhibited significant enhancement of AUC (0- ∞) (~4.02-fold) and clearance has been significantly decreased (~5.5-fold). Thus, mucoadhesive bromfenac-loaded chitosan/alginate NPs could be considered useful approach aiming to sustained ocular residence and reduce dosing frequency.

In another study microspheric *in-situ* gel for ocular drug delivery system of bromfenac sodium has been prepared [37] for the treatment of post-operative cataract surgery. The microspheric *in-situ* gel of optimized formulation has shown drug release of 77.989% at the end of 24 hrs. Eye irritancy test has performed on albino rabbits. The results of the ocular irritation studies have indicated that the formulation have been non-irritant. The stability data recorded over a 3 months period according to ICH guidelines have observed that formulations have been stable. Hence, it could be concluded that microsphere *in-situ* gels are a viable alternative to conventional eye drops by providing sustained release of medicaments in the eye.

There are some NSAIDs approved by the FDA for the treatment of post-operative inflammation after cataract surgery (ketorolac, flurbiprofen, bromfenac, diclofenac and nepafenac). Nepafenac is the only nonsteroidal anti-inflammatory prodrug that requires conversion to its active form, amfenac, through intraocular enzymatic hydrolysis [38]. In a clinical study comparing aqueous humor concentrations of four NSAIDs after administration in patients having cataract surgery, nepafenac showed significantly greater ocular bioavailability than the others [39].

ii) Carbo-and hetero-cyclic acetic acids

Indomethacin and ketorolac are the most studied representatives of this group for inclusion in nanocarriers for eye administration.

Indomethacin (IMC), 2-{1-[(4-Chlorophenyl) carbonyl]-5-methoxy-2-methyl-1H-indol-3-yl} acetic acid, is used in ophthalmology as an aqueous solution of sodium salt and tromethamine salt [6, 22]. It is practically insoluble in water, unstable in alkaline medium and poorly soluble in acidic medium. IMC can be used topically in eye drops with concentration from 0.1% to 1% (w/v) for the prevention of miosis during cataract surgery and for cystoid macular edema prevention [6, 22].

To overcome the technological problems associated with the insolubility and the instability of IMC in an aqueous medium and its low bioavailability after topical administration, several models of drug delivery systems have been developed and studied.

Poly (ϵ -caprolacton) NPs, nanocapsules and lecithin nanoemulsions with IMC with an average size of 225 nm, obtained by surface deposition, nanoprecipitation and spontaneous emulsification have been studied. *In vitro* comparison between colloidal carriers and commercial eye drops has shown a threefold higher concentration of the drug from the colloidal systems. Moreover, *in vivo* assessment of these colloidal systems in rabbit's eyes has shown three fold higher concentration of IMC in the aqueous humor 0.5 hrs after administration and 300% increase in drug availability compared to commercial formulations [40].

Furthermore, chitosan coated poly (ϵ -caprolacton) NPs with IMC have demonstrated a twofold increase in the ocular bioavailability [41]. In another study, an IMC-chitosan-nanoemulsion has shown more efficient healing of corneal chemical ulcer compared to NPs preparation and the high level of IMC in inner ocular structure thereby increasing drug delivery efficiency [42].

Chetoni *et al.* [43] have developed a model of an aqueous solution of IMC (0.1%, w/v). Poloxamer-407 was used as a co-solvent. The solution was stable and showed a higher level in the aqueous humor and faster disappearance of the inflammation symptoms in a model of immunogenic uveitis compared to the commercial formulation.

Andonova *et al.* have demonstrated for the first time the possibility of an effective *in-situ* incorporation of IMC into homopolymer poly(vinyl acetate) (pVAc) and polystyrene NPs and NPs of copolymers of vinyl acetate (VAc) with 3-dimethyl (methacryloyloxyethyl) aminopropyl sulfonate (DMAPS) (p (VAc-co-DMAPS)) as well as mixtures of NPs of these polymers with the hydrophilic chitosan, Carbopol® and p(DMAPS) by one-stage emulsion polymerization without using an emulsifier [44-47]. The rate and degree of release of IMC from these nanosized polymer carriers included in ophthalmic formulations could be adjusted by the composition of the copolymers of the polymer mixtures, as well as the conditions for the preparation of the NPs. The established biocompatibility of pVAc [48], the stabilizing role on the *in-situ* included IMC [49] and the research on its release from nanosized carrier at pH 7.4, have given grounds for the authors to recommend the NPs from pVAc homo-and copolymers and their mixtures with hydrophilic and biocompatible chitosan, Carbopol® and p (DMAPS) as a drug delivery system in ophthalmic formulations [50, 51].

An ophthalmic solution of ketorolac is available and is used to treat eye pain and to relieve the itchiness and burning of seasonal allergies. The ophthalmic formulation could be used in cases where a raised intraocular pressure (glaucoma) is to be avoided [22]. Ketorolac ((\pm)-5-benzoyl-2,3-dihydro-1H-pyrrolizine-1-carboxylic acid) is an isostere of ketoprofen, more precisely, it is a dihydro pyrrolizine carboxylic acid derivative structurally related to indomethacin. Ketorolac loaded chitosan NPs have been obtained by ionic-co-precipitation technique. The drug loading-release studies have been very promising for an alternative to treat ocular diseases such as pseudophakic cystoid macular edema according to the obtained data [52].

Moreover, ketorolac has been successfully entrapped in polymeric micelles made of copolymer of *N*-isopropylacrylamide (NIPAAm),

vinyl pyrrolidone (VP) and acrylic acid (AA) having cross-linkage with *N,N'*-methylene bis-acrylamide (MBA). The NP formulations with an average diameter of 35 nm at 25 °C and drug entrapment about 80% have been stable for 8-10 days at room temperature. The drug release in aqueous buffer (pH 7.2) from the polymeric micelles at 25 °C has been 20% and 60% after 2 and 8 hrs respectively and has been temperature and pH dependent. *In vitro* corneal permeation studies through excised rabbit cornea have indicated a twofold increase in the ocular availability with no corneal damage compared to an aqueous suspension containing the same amount of drug as in the NPs. The formulation has shown significant inhibition of lid closure up to 3 hrs and PMN migration up to 5 hrs compared to the suspension containing non-entrapped drug, which did not show any significant effect [53].

Propionic acids

Flurbiprofen ((RS)-2-(2-fluorobiphenyl-4-yl) propanoic acid), ibuprofen ((RS)-2-(4-(2-methylpropyl) phenyl) propanoic acid) and naproxen ((+)-(S)-2-(6-methoxynaphthalen-2-yl) propanoic acid) are the exploited members of this group for inclusion in NPs as drug delivery systems in ophthalmic formulations.

Poly (D,L-lactide-co-glycolide) nanospheres with incorporated flurbiprofen have been prepared by the solvent displacement technique for the purpose of assessing drug-polymer physicochemical interactions, flurbiprofen release from the polymer matrix and eye permeation of the drug formulated in the colloidal system [54]. The *ex vivo* corneal permeation study has shown that flurbiprofen-loaded nanospheres enhanced drug penetration by about twofold over commercial eye drops containing PVA and by about fourfold over flurbiprofen in pH 7.4 phosphate buffer. The corneal hydration level of each cornea has been determined in order to evaluate potential corneal damage.

Flurbiprofen has been also included in Eudragit RS 100® and Eudragit RL 100® for prevention of miosis during extracapsular cataract surgery [55]. The drug included in these polymers has shown prolonged release and increased bioavailability.

Moreover, flurbiprofen has been successfully included in nanostructured lipid carriers. Gonzales-Mira *et al.* have implemented a 2⁴ full factorial design based on 4 independent variables to plan the experiments, namely, the percentage of stearic acids with regard to the total lipid, the flurbiprofen concentration, the stabilizer concentration, and the storage conditions (i.e. storage temperature) [56]. Optimization of the process has been achieved and the best formulation corresponded to the nanostructured lipid carriers formulation composed of 0.05 (wt %) flurbiprofen, 1.6 (wt %) Tween® 80 and a 50:50 ratio of stearic acid to castor oil, with an average diameter of 288 nm, polydispersity index (PI) 0.245 and zeta-potential (ZP) of -29 mV. The developed systems have shown physico-chemical stability with high tolerance after ophthalmic application.

With the aim of improving the availability of sodium ibuprofen at intraocular level, ibuprofen-loaded polymeric NP suspensions have been made from Eudragit RS 100 [57]. *In vitro* dissolution tests have indicated a controlled release profile of ibuprofen from NPs. *In vivo* efficacy has been assessed on rabbit's eye after induction of an ocular trauma (paracentesis). An inhibition of the miotic response to the surgical trauma has been achieved, comparable to a control aqueous eye-drop formulation, even though a lower concentration of free drug in the conjunctival sac has been reached from the NP system. Drug levels in the aqueous humour have been also higher after application of the nanosuspensions. The NPs did not show any damage to the ocular tissues and thus these polymers have proven to be inert carriers.

Ibuprofen nanostructured lipid carriers (NLCs) have been prepared by melted-ultrasonic methods; gelucire 44/14 has been selected as one of the solid lipid matrix materials due to the good particle size dispersion and excellent contribution to the corneal permeability of the model drug [58]. The corresponding apparent permeability coefficients (P (app)) have been 1.28 and 1.36 times higher than that of the control preparation. Ibuprofen NLCs have displayed

controlled-release properties. The AUC of the optimized formulation of ibuprofen NLCs has been 3.99 times greater than that of ibuprofen eye drops.

The NPs of naproxen with Eudragit RS 100 have been formulated using the solvent evaporation/extraction technique (the single emulsion technique) [59]. The effect of several process parameters i.e. drug/polymer ratio, aqueous phase volume and speed of homogenization on the size of the nanoformulations has been considered. All the prepared formulations using Eudragit RS 100 have resulted in nano-range size particles with relatively spherical smooth morphology. The NPs have displayed a slowed release pattern with the reduced burst release in comparison with the intact drug powder and the physical mixtures of drug and polymer. According to these findings, the formulation of naproxen-Eudragit RS 100 NPs has been able to improve the physicochemical characteristics of the drug and possibly could increase the anti-inflammatory effects of the drug following its ocular or intra-joint administration.

Javadzadeh *et al.* formulated NPs of naproxen with PLGA using single emulsion technique [60]. Drug/polymer ratio, aqueous phase volume and speed of homogenization have been considered as process parameters to achieve optimal preparation conditions. The study suggested the feasibility of formulating NPs of PLGA with satisfactory physicochemical characteristics and increasing the anti-inflammatory effects of the drug following its ocular or intra-joint administration.

Enolic acids

The derivatives of enolic acid are subdivided into pyrazolones and oxicams according to their structure. Piroxicam (4-hydroxy-2-methyl-N-(2-pyridinyl)-2H-1,2-benzothiazine-3-carboxamide 1,1-dioxide) is the main representative of the oxicams sub-group included in nanocarriers as drug delivery systems in topical eye formulations.

To investigate the anti-inflammatory impact of piroxicam nanosuspension, Eudragit RS100 nanoformulations have been used to control inflammatory symptoms in rabbits with endotoxin-induced uveitis (EIU). The NPs of piroxicam: Eudragit RS100 has been formulated using the solvent evaporation/extraction technique. Kinetically, the release profiles of piroxicam from NPs appeared to best fit the Weibull model and diffusion has been the superior phenomenon. The *in vivo* examinations have revealed that the inflammation could be inhibited by the drug: polymer nanosuspension more significantly than the microsuspension of the drug alone in rabbits with EIU. Upon these findings, the authors have proposed that the piroxicam: Eudragit RS100 nanosuspensions may be considered as an improved ocular delivery system for local inhibition of inflammation [61].

Piroxicam-loaded pectin microspheres have been prepared by a spray-drying technique [62]. Piroxicam loaded in the pectin microspheres has shown a faster *in vitro* dissolution rate compared to solid micronized drug. The precorneal retention of fluorescein-loaded microspheres has been evaluated *in vivo* in albino rabbits. An aqueous dispersion of fluorescent microspheres has shown a significantly increased residence time in the eye (2.5 vs. 0.5 hr) when compared to a fluorescein solution. *In vivo* tests in rabbits of dispersions of piroxicam-loaded microspheres have also indicated a significant improvement of the drug bioavailability in the aqueous humour (2.5-fold) when compared to commercial piroxicam eye drops.

Cox II Inhibitors

Celecoxib and valdecoxib are the representatives of the group of Cox-II inhibitors. A challenge is to improve the dissolution rate of drugs and thus to optimize the bioavailability in the ocular tissues. Data on the inclusion of meloxicam in nanocarriers as drug delivery systems in topical eye formulations were not found.

Several celecoxib-loaded NPs have been prepared by emulsification solvent diffusion technique using different polymers including chitosan, sodium alginate, PCL, PLA and PLGA [63, 64]. In one of the

studies, selected preparations have been loaded with celecoxib and incorporated in three different ophthalmic preparations (eye drops, *in-situ* gelling system and gel) [64]. *In vitro* release of celecoxib from different ophthalmic formulations has been sustained over a period of 24 hrs and belonged to non-Fickian Higuchi diffusion model. A cytotoxicity experiment has shown that the tested formulations are biologically safe and non-toxic. Gels containing celecoxib-loaded PCL-NPs and chitosan-NPs have proven to be the most physically and chemically stable formulations which determined the *in vivo* study in order to evaluate the ocular bioavailability of celecoxib in NP-loaded ophthalmic formulations. Gels containing NPs have shown a higher ocular bioavailability than the control preparation which indicated higher AUC₀₋₂₄, AUC_{0-∞} and C_{max} values and several folds increase in the relative bioavailability. These formulations have also shown more extended release of celecoxib that lasts for more than one day. The gel containing chitosan-NPs has had a higher bioavailability and more extended release of the drug than those containing PCL-NPs due to the bio adhesiveness and penetration enhancement of the polymer.

In an attempt to improve ocular bioavailability, NP formulation of valdecoxib with HP β CD has been evaluated [65]. As anticipated, levels of valdecoxib in the cornea and conjunctiva have been significantly higher in NP-treated rabbit's eyes compared to control.

CONCLUSION

The nano carriers applied as ocular drug delivery systems offer undoubted advantages such as higher solubility, higher area available for dissolution and higher dissolution rate. Another advantage of the nano systems is the higher corneal penetration. They could combine ophthalmic prolonged action with the ease of the application of liquid eye drops. They could offer a high stability of the drug, protecting it from the negative environmental impact and also provide controlled release and target action. Ophthalmic drug delivery may benefit to a full extent from the characteristics of nano-sized drug particles. In conclusion, a multi-disciplinary approach from pharmacology to ophthalmology and from biomaterial science to pharmaceutical science will bring to clinical use these innovative NSAIDs-loaded nano systems for the pharmacological management of sight-threatening eye diseases.

CONFLICT OF INTERESTS

Declared None

REFERENCES

1. Kumar A, Malviya R, Sharma PK. Recent trends in ocular drug delivery: a short review. *Eur J Appl Sci* 2011;3:86-92.
2. Harry J, Mission G. Clinical ophthalmic pathology, Principles of Diseases of the Eye and Associated Structures. Oxford (UK): Butterworth Heinemann; 2001.
3. Foster JB, Lee WB. The tear film: anatomy, Structure and Function. In: Holland J, Mannis MJ, Lee WB. editors. *Ocular Surface Disease: Cornea, Conjunctiva and Tear Film*. saunders; 2013.
4. Tomlinson A, Khanal S. Assessment of tear film dynamics: quantification approach. *Ocul Surf* 2005;3:81-95.
5. Kulkarni VS. *Handbook of Non-Invasive Drug Delivery Systems*. Elsevier; 2010.
6. AHFS. Drug information American Society of Health-System Pharmacists; 2001.
7. Mainardes RM, Urban MCC, Cinto PO, Khalil NM, Chaud MV, Evangelista RC, *et al.* Colloidal carriers for ophthalmic. *Drug Delivery Curr Drug Targets* 2005;6:363-71.
8. Alonzo MJ. Nanomedicine for overcoming biological barriers. *Biomed Pharmacother* 2004;58:168-72.
9. Patel PB, Shastri DH, Shelat PK, Shukla AK. Ophthalmic drug delivery systems: challenges and approaches. *Syst Rev Pharm* 2010;1:113-20.
10. Kreuter J. Nanoparticles-a historical perspective. *Int J Pharm* 2007;331:1-10.
11. Janoria KG, Gunda S, Boddur SHS, Mitra AK. Novel approaches to retinal drug delivery. *Expert Opin Drug Delivery* 2007;4:371-88.
12. Das S, Suresh PK. Drug Delivery to Eye: Special Reference to Nanoparticles. *Int J Drug Delivery* 2010;2:12-21.

13. Canavan N. New Insights into Ophthalmic Drug Delivery. PFO; 2011.
14. Schoenberger SD, Kim SJ. Nonsteroidal anti-inflammatory drugs for retinal disease. *Int J Inflammation* 2013. doi: 10.1155/2013/281981. [Article in Press]
15. Avunduk AM, Avunduk MC, Vornell ED, Kaufman HE. The comparison of efficacies of topical corticosteroids and nonsteroidal anti-inflammatory drops on dry eye patients: a clinical and immunocytochemical study. *Am J Ophthalmol* 2003;136:593-602.
16. Pflufelder SC. Anti-inflammatory therapy for dry eye. *Am J Ophthalmol* 2004;137:337-42.
17. Udoetuk JD, Dai Y, Ying GS, Gangaputra S, Rosenbaum JT, Suhler EB, *et al.* Risk of corticosteroid-induced hyperglycemia requiring medical therapy among patients with inflammatory eye diseases. *Ophthalmology* 2012;119:1569-74.
18. Schalnus R. Topical nonsteroidal anti-inflammatory therapy in ophthalmology. *Ophthalmologica* 2003;217:89-98.
19. Bandare BM, Sankaridurg PR, Willcox MD. Non-steroidal anti-inflammatory agents decrease bacterial colonization of contact lenses and prevent adhesion to human corneal epithelial cells. *Curr Eye Res* 2004;29:245-51.
20. Rathore MS, Majumdar DK. Effect of formulation factors on *in vitro* transcorneal permeation of gatifloxacin from aqueous drops. *AAPS Pharm Sci Tech* 2006;7:57.
21. Ahuja M, Dhake AS, Sharma SK, Majumdar DK. Topical ocular delivery of NSAIDs. *AAPS J* 2008;10:229-41.
22. Sweetman CS. *Martindale: The Complete Drug Reference*. 34th ed. London: Pharmaceutical Press; 2005.
23. Deruiter J. Non-steroidal anti-inflammatory drugs (nsaids), Principles of drug action 2: Tulane University Fall; 2002.
24. Sánchez-Borges M. Clinical management of nonsteroidal anti-inflammatory drug hypersensitivity. *WAO J* 2008;1:29-33.
25. Das S, Banerjee R, Bellare J. Aspirin loaded albumin nanoparticles by coacervation: implications in drug delivery. *Trends Biomaterials Artificial Organs* 2005;18:203-12.
26. Agnihotri SM, Vavia PR. Diclofenac-loaded biopolymeric nanosuspensions for ophthalmic application. *Nanomed Nanotechnol Biol Med* 2009;5:90-5.
27. Ahuja M, Dhake AS, Majumdar DK. Effect of formulation factors on *in vitro* permeation of diclofenac from experimental and marketed aqueous eye drops through excised goat cornea. *Yakugaku Zasshi* 2006;126:1369-75.
28. Attama AA, Reichl S, Muller-Goymann CC. Diclofenac sodium delivery to the eye: *in vitro* evaluation of novel solid lipid nanoparticle formulation using human cornea construct. *Int J Pharm* 2008;355:307-13.
29. Sahu V, Solanki H, Pandey AK. Development and characterization of solid lipid nanoparticle of diclofenac sodium in the treatment of ocular pain after photorefractive keratectomy. *Int J Nanomater Nanotechnol Nanomed* 2015;179:80.
30. Swamy NGN, Abbas Z, Santosh Kumar IH. Eudragit RS 100 nanosuspensions for the controlled ophthalmic delivery of diclofenac sodium. *Thai J Pharm Sci* 2013;37:157-70.
31. Amkanth S, Madhusudhana Chetty C, Alagusundaram M, Angalaparameswari S, Thiruvengadarajan VS, Gnanaprakash K. Design and evaluation of diclofenac sodium ocusert. *Int J Pharm Tech Res* 2009;1:1219-23.
32. Ara T, Sharma S, Bhat SA, Bhandari A, Deva AS, Rathore MS, *et al.* Preparation and evaluation of ocular inserts of diclofenac sodium for controlled drug delivery. *Pharm Lett* 2014;6:93-9.
33. Khowessah O, Amin M, Basalious EB. Development and optimization of aceclofenac nanoparticles utilizing a full factorial design. *Proceedings of the 5th International Conference on Nanotechnology: Fundamentals and Applications Prague, Czech Republic; 2014. p. 300.*
34. Dasgupta S, Ghosh SK, Ray S, Mazumder B. Solid lipid nanoparticles (SLNs) gels for topical delivery of aceclofenac *in vitro* and *in vivo* evaluation. *Curr Drug Delivery* 2013;10:656-66.
35. Katara R, Majumdar DK. Eudragit RL 100-based nanoparticulate system of aceclofenac for ocular delivery. *Colloids Surf B* 2013;103:455-62.
36. Boddada B, Ratna JV. Mucoadhesive nanoparticles for sustained ocular delivery of bromfenac: *in vitro* and pharmacokinetic studies. *J Bioequivalence Bioavailability* 2014;6:5.
37. Vengurlekar P, Singh A, Rathod S. Microspheric in-situ gel for ocular drug delivery system of bromfenac sodium. *IJPSR* 2014;5:179-85.
38. Souto EB, Doktorovova S, Gonzalez-Mira E, Egea MA, Garcia ML. Feasibility of lipid nanoparticles for ocular delivery of anti-inflammatory drugs. *Curr Eye Res* 2010;35:537-52.
39. Walters T, Raizman M, Ernest P. *In vivo* pharmacokinetics and *in vitro* pharmacodynamics of nepafenac, Amfenac, Ketorolac, and Bromfenac. *J Cataract Refractive Surg* 2007;33:1539-45.
40. Calvo P, Vila-Jato JL, Alonso MJ. Comparative *in vitro* evaluation of several colloidal systems, nanoparticles, nanocapsules and nanoemulsions as ocular drug carriers. *J Pharm Sci* 1996;85:530-6.
41. De Campos AM, Sanchez A, Gref R, Calvo P, Alonso MJ. The effect of a PEG versus a chitosan coating on the interaction of drug colloidal carriers with the ocular mucosa. *Eur J Pharm Sci* 2003;20:73-81.
42. Badawi AA, El-Laithy HM, El Qidra RK, El Mofty H, El Dally M. Chitosan based nanocarriers for indomethacin ocular delivery. *Arch Pharm Res* 2008;31:1040-9.
43. Chetoni P, Panichi L, Buralgassi S, Bendli U, Seattone MF. Pharmacokinetics and anti-inflammatory activity in rabbits of a novel indomethacin ophthalmic solution. *J Ocul Pharmacol Ther* 2000;16:363-72.
44. Andonova V, Georgiev G, Toncheva V, Kassarova M. Preparation and study of poly (vinyl Acetate) and Poly (styrene) nanosized latex with indometacin. *Pharmazie* 2012;67:601-4.
45. Andonova V, Georgiev G, Toncheva V, Petrova N, Karashanova D, Penkov D, *et al.* Indomethacin loading and *in vitro* release properties from vinyl acetate homo- and co-polymer nanoparticles, coated with polyzwitterion and carbopol@ shells. *Int J Pharm Pharm Sci* 2014;6:691-9.
46. Andonova V, Georgiev G, Toncheva V, Karashanova D, Katsarov P, Kassarova M. Carbopol@ and chitosan coated nanoparticles with *in-situ* loaded indomethacin. *Am J Pharm Tech Res* 2014;4:664-78.
47. Andonova V, Georgiev G, Toncheva V, Kassarova M. Influence of some technological factors on the preparation of polymeric nanoparticles with indomethacin. *Open Access Sci Rep* 2013; 2. doi:10.4172/scientificreports.703:1-4. [Article in Press]
48. Andonova V, Draganov M, Feodorova Y, Georgiev G, Kassarova M. Biological activity of indomethacin-loaded nanoparticles. *World J Pharm Res* 2014;4:85-101.
49. Andonova V, Georgiev G, Dimitrova S, Katsarova M, Kassarova M. Characterization, *In vitro* evaluation and stability studies of indomethacin-loaded polyzwitterionic copolymer nanoparticles. *Int J Drug Delivery Technol* 2015;5:16-23.
50. Andonova VY, Georgiev GS, Georgieva VT, Petrova NL, Kassarova M. Indomethacin nanoparticles for applications in liquid ocular formulations. *Folia Med* 2013;55:76-82.
51. Andonova V, Zagorchev P, Katsarov P, Kassarova M. Eye drops with nanoparticles as drug delivery systems. *Int J Pharm Pharm Sci* 2015;7:431-5.
52. Aşık MD, Uğurlu N, Yülek F, Tuncer S, Türk M, Denkbaş EB. Ketorolac tromethamine loaded chitosan nanoparticles as a nanotherapeutic system for ocular diseases. *Hacettepe J Biol Chem* 2013;41:81-6.
53. Gupta AK, Madan S, Majumdar DK, Maitra A. Ketorolac entrapped in polymeric micelles: preparation, Characterisation and ocular anti-inflammatory studies. *Int J Pharm* 2000;209:1-14.
54. Vega E, Gamisans F, García ML, Chauvet A, Lacoulonche F, Rgea MA. PLGA nanospheres for the ocular delivery of flurbiprofen: drug release and interactions. *J Pharm Sci* 2008;97:5306-17.
55. Pignatello R, Bucolo C, Spedalieri G, Maltese A, Puglisi G. Flurbiprofen-loaded acrylate polymer nanosuspensions for ophthalmic application. *Biomaterials* 2002;23:3247-55.
56. Gonzales-Mira E, Egea MA, Garcia ML, Souto EB. Design and ocular tolerance of flurbiprofen loaded ultrasound-engineered NLC. *Colloids Surf B* 2010;81:412-21.

57. Pignatello R, Bucolo C, Ferrara P, Maltese A, Puleo A, Puglisi G. Eudragit RS100® nanosuspensions for the ophthalmic controlled delivery of ibuprofen. *Eur J Pharm Sci* 2002;16:53-61.
58. Li X, Nie SF, Kong J, Li N, Ju CY, Pan W. A controlled-release ocular delivery system for ibuprofen based on nanostructured lipid carriers. *Int J Pharm* 2008;363:177-82.
59. Adibkia K, Javadzadeh Y, Dastmalchi S, Mohammadi G, Niri FK, Alaei-Beirami M. Naproxen-eudragit RS100 nanoparticles: preparation and physicochemical characterization. *Colloids Surf B* 2011;83:155-9.
60. Javadzadeh Y, Ahadi F, Davaran S, Mohammadi G, Sabzevari A, Adibkia K. Preparation and physicochemical characterization of naproxen-PLGA nanoparticles. *Colloids Surf B* 2010;81:498-502.
61. Adibkia K, Siahishadbad MR, Nokhodchi A, Javadzadeh A, Barzegar-Jalali M, Barar J, *et al.* Piroxicam nanoparticles for ocular delivery: physicochemical characterization and implementation in endotoxin-induced uveitis. *J Drug Targeting* 2007;15:407-16.
62. Giunchedi P, Conte U, Chetoni P, Saetone MF. Pectin microspheres as ophthalmic carriers for piroxicam: evaluation *in vitro* and *in vivo* albino rabbits. *Eur J Pharm Sci* 1999;9:1-7.
63. Ibrahim MM. Formulation and evaluation of certain drugs for ophthalmic therapy. PhD Thesis. Mansoura University; 2014.
64. Ibrahim MM, Abd-Elgawad A-EH, Soliman OA-E, Jablonski MM. Nanoparticle-based topical ophthalmic formulations for sustained celecoxib release. *J Pharm Sci* 2013;102:1036-53.
65. Kararli TT, Bandyopadhyay R, Singh SK, Hawley LC. Ophthalmic Formulation of a Selective Cyclooxygenase-2 Inhibitory Drug. WO02005815; 2002.