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Vehicle influence on ocular disposition of sodium cromoglycate in the albino rabbit

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Summary

The influence of vehicle composition on the ocular disposition of sodium cromoglycate, a drug used for the prophylaxis of vernal keratoconjunctivitis, has been studied. The vehicles investigated consisted of a water-soluble base (15% polyvinyl alcohol, PVA, in water); an absorption base (10% acetylated lanolin in a paraffin base); and an oleaginous base (a polyethylene/mineral oil blend). The albino rabbit was chosen as the animal model. The disposition of sodium cromoglycate in tears, conjunctiva, cornea, iris-ciliary body and aqueous humor of the rabbit was monitored at 30, 60 and 240 min post-instillation using radiotracer techniques. At the early time periods, all 3 bases were judged to perform adequately in that they provided approximately the same sodium cromoglycate concentrations in the tear pool, conjunctiva and cornea as the aqueous vehicle. However, after 240 min, the oleaginous polyethylene/mineral oil formulation provided the highest concentrations of drug in the conjunctiva, the target tissue. The water-soluble PVA formulation gave significantly lower levels in the tears, the conjunctiva and the cornea. At this same point, the absorption base containing acetylated lanolin produced the highest drug concentration in the tear pool. Collectively, these data suggest that of the 3 bases studied, the oleaginous base and the absorption base show most promise as vehicles to extend the residence time of sodium cromoglycate in the eye. Obviously, the final choice of vehicle will also be influenced by factors such as the physical and chemical stability of the drug in the formulation chosen and patient acceptance.

Introduction

Sodium cromoglycate has long been used in the form of eye drops for the prophylaxis of vernal conjunctivitis (Easty et al., 1972; Kazdan et al., 1976; Tabbara and Arafat, 1977; El Hennawi, 1980; Leino and Tuovinen, 1980; Nemoto, 1980). Because of the efficient solution drainage process in the eye, the rate of removal of sodium cromoglycate from the pre-corneal area is relatively fast (Lee et al., 1983). Incorporation of the drug into a 5% polyvinyl alcohol solution has been found to reduce the rate at which it is removed by the tears, thereby elevating the concentration of sodium cromoglycate in the conjunctiva, the target tissue, by a factor of 2–4 (Lee et al., 1983). A frequently used alternative or adjunct approach to ophthalmic therapy involves the use of eye ointments and viscous gels. It has been shown (Sieg and Robinson, 1975, 1977, 1979; Saettone et al., 1980) that, depending on the interplay in physicochemical properties between the drug and the base, such preparations can significantly prolong the presence of a drug in various ocular tissues, including the tear chamber. In the case of sodium cromoglycate, such an effect is highly desirable in the management of vernal keratoconjunctivitis, a chronic condition, particularly since the preparation could be used overnight.

The purpose of this study was to evaluate the ability of 3 bases to affect the release of sodium cromoglycate into the tear pool of the albino rabbit eye and its subsequent uptake and retention in the ocular tissues. The vehicles chosen, which have a varying ability to mix with aqueous systems such as tears, were polyvinyl alcohol, a water soluble base; Modulan, an absorption base; and Plastibase, an oleaginous base. Sodium cromoglycate concentrations in various ocular tissues of the rabbit were monitored at 30 and 60 min following application of the vehicle, with a view to evaluating the initial rate of drug release from the base, and at 240 min to assess the duration of drug release.

Materials and Methods

Materials

Three formulations containing 4% sodium cromoglycate spiked with tritiated material were supplied by Fisons plc, Pharmaceutical Division, Loughborough, U.K. The radiochemical purity of the [^{3}H]-labelled sodium cromoglycate was greater than 97%. The 3 bases were: (i) polyvinyl alcohol [1], 15% in water; (ii) acetylated lanolin [2], 10% in a paraffin base; and (iii) a polyethylene and mineral oil blend [3]. The specific activity of each formulation was 59 $\mu Ci \cdot g^{-1}$. The radiochemical homogeneity was verified by measuring the radioactivity among samples taken from various positions within each sample. All formulations were used within 4 weeks of preparation.

[1] Polyviol W 20/90, Wacker Chemicals, U.K.
[2] Modulan, Amerchol U.K., U.K.
[3] Plastibase, E.R. Squibb, U.K.

Male albino rabbits [4], weighing 2.2–2.5 kg, were used throughout the study. They were fed a regular diet with no restrictions on food or water consumed.

Administration of formulation

During the experiments, all rabbits were kept in restraining boxes in a normal upright posture. Both eyes of the rabbit were used, but the dosing times were staggered so that each animal was used for two time points. Individual doses, of approximately 25 mg of formulation, were weighed on an analytical balance immediately before application to the center of the lower eyelid of an albino rabbit with a microspatula. During dosing, care was taken not to irritate the eye or touch the corneal surface with the spatula. Immediately following dosing, the lower eyelid was gently moved upward to spread the dose over the corneal surface and then released. No other manipulative technique was used during the run to distribute the formulation in the precorneal area.

Sodium cromoglycate concentration–time profiles in the tear film and ocular tissues

Approximately 5 s prior to sacrifice of the rabbit by marginal ear vein injection of a 30% sodium phenobarbital solution, 1 μl of tears was collected from the center of the lower tear strip using a disposable glass capillary pipet [5]. Tears were also collected at 120 and 180 min following application of each formulation to those rabbits designated for the 240 min time point. Pipets containing tear samples were transferred to vials [6] containing 4 ml of prerefrigerated scintillation cocktail [7] and counted in a liquid scintillation spectrometer [8] after storage in the dark for 24 h. The presence of glass capillaries in the scintillation cocktail did not alter the counting efficiency or affect the results in any way.

Following sacrifice of the rabbit its corneal and conjunctival surfaces were thoroughly rinsed with normal saline and blotted dry in order to remove any residual vehicle. The anterior segment tissues — conjunctiva, aqueous humor, cornea and iris-ciliary body — were obtained in that order. The surgical procedures on each eye were completed within 5 min of sacrificing the rabbit so that any errors due to redistribution of drug during the time required to obtain ocular tissue samples were minimized. The aqueous humor samples were transferred to vials [6] containing 4 ml of prerefrigerated scintillation cocktail [7].

Each of the tissue samples was digested at 55°C for 18 h in 1.5 ml of a tissue solubilizer [9] contained in a glass scintillation vial [5] followed by decolorization with 100 μl of hydrogen peroxide and addition of 10 ml of a scintillation cocktail [10]. All samples were stored in the dark for 24 h prior to counting in a liquid scintillation spectrometer [8]. After correcting for background and quenching effects, the data in

[4] ABC Rabbitry, Pomona, CA, U.S.A.
[5] Curtin Matheson Scientific, Fountain Valley, CA, U.S.A.
[6] BioVials, Beckman, Irvine, CA, U.S.A.
[7] Aquasol-2, New England Nuclear, Boston, MA, U.S.A.
[8] Beckman model 7500, Irvine, CA, U.S.A.
[9] Protosol, New England Nuclear, Boston, MA.
[10] Econofluor, New England Nuclear, Boston, MA.

counts per minute were converted to μg sodium cromoglycate/g of tissue through the use of spiked tissue standards. The density of aqueous humor was assumed to equal 1.0. It was assumed that, as is the case with the systemic administration of sodium cromoglycate (Ashton et al., 1973), none of the drug was metabolized in the ocular tissues.

Results and Discussion

Drug release from ointment and gel bases can involve one or more of several processes, including partitioning, diffusion, dissolution and facilitated release. The latter process refers to the mechanical rupture of dispersed droplets in emulsion systems (Sieg and Robinson, 1979). With the bases used in this study, drug release from the water-soluble PVA base probably only involves diffusion since the sodium

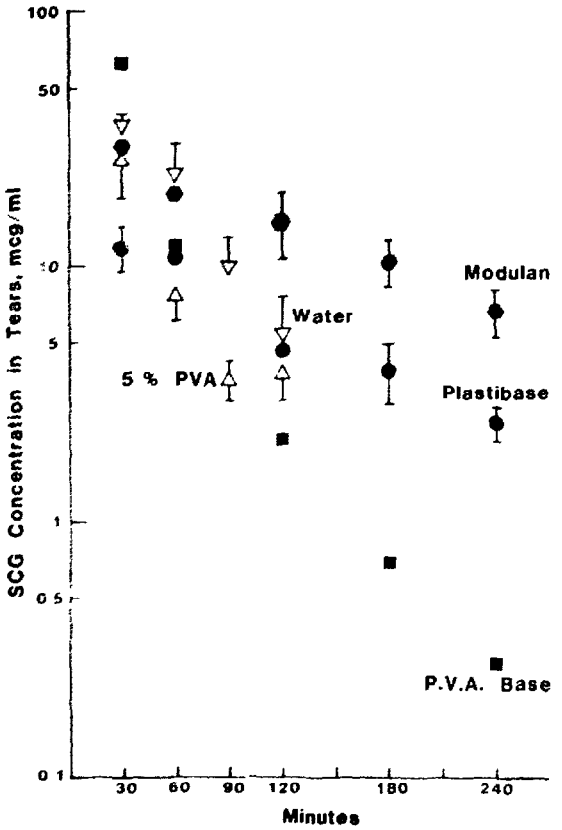

Fig. 1. Sodium cromoglycate concentration, expressed as μg of drug/ml of tears and normalized to amount of drug applied, in tears of albino rabbits following topical application of 25 mg of 4% ointments or 25 μl of 2% solutions (Lee et al., 1983). Between 8 and 12 eyes were used for each time point. Error bars represent standard error of the mean; where not shown, they were omitted for clarity. Key: ▽, aqueous solution; △, aqueous solution containing 5% PVA; ■, ointment containing 15% PVA; ●, ointment based on acetylated lanolin (Modulan) and paraffins; ●, ointment based on polyethylene/mineral oil blend (Plastibase).

cromoglycate will be in solution in the base. In contrast, drug release from the Modulan-containing base (an absorption base) and Plastibase (an oleaginous base) probably involves the penetration of water into the base, dissolution of drug and then outward diffusion to the tear pool. In the case of Modulan, some emulsion formation may occur and lead to facilitated release. Each of these mechanisms can be expected to affect the rate and duration of drug release and, in turn, the drug concentrations attained in the tears and ocular tissues. Because drugs administered in ointments and gels are not immediately available to the tear film, it is likely that the initial drug concentration in tears will be lower than in the case of aqueous solutions. This was found to be the case with the oleaginous base Plastibase (Fig. 1). In contrast, the PVA and Modulan bases were either superior or equivalent, respectively, to the aqueous vehicles in providing drug to the tear pool 30 min after administration. Such a trend correlates with the increasing lipophilic characteristics of these 3 bases, and has been noted by Bottari et al. (1979) in their evaluation of the influence of ointment base composition on the duration of corneal anesthesia attainable from lidocaine and benzocaine. At 60 min there was no significant difference between the PVA and Plastibase preparations, while the Modulan was superior to both but not significantly different from the aqueous solution studied previously (Lee et al., 1982).

In making comparisons between sodium cromoglycate levels in the tear pool derived from these bases and aqueous solutions, it should be borne in mind that at these earlier time points (30 and 60 min) the source of the detected drug may differ between these two types of formulation. Thus, since almost all of an instilled aqueous solution is lost within 5 min of instillation (Chrai et al., 1973), the drug found in the tear film at 30 and 60 min is likely to have been derived from back diffusion of the drug already in the conjunctiva and/or cornea, particularly if the drug is distributed in favor of the epithelial layer of these tissues. The vehicles studied in the present work are not subject to such rapid drainage and will remain in the precorneal area longer than aqueous solutions. The sodium cromoglycate detected in the tear film at 30 and 60 min may therefore be assumed to have come primarily from the vehicle, rather than totally from back diffusion from the conjunctiva and/or cornea.

Over a 4 h period it is obvious that drug formulated in the PVA gel base disappears rapidly from the tear film when compared with that in the Modulan and Plastibase vehicles (Fig. 1). The first-order disappearance rate constant for drug in the PVA base (determined from the terminal slope of the concentration–time profile in Fig. 1) was found to be 0.025 min^{-1}, a value close to that for sodium cromoglycate administered as a solution in water or a solution in 5% PVA, namely 0.022 min^{-1} (Lee et al., 1983). In contrast, the rate constants for the non-hydrophilic bases, Modulan and Plastibase, were calculated to be 0.007 and 0.008 min^{-1}, respectively. However, it is possible that these rate constants were for release of drug from the ointment rather than for its disappearance from the tear pool. Even so, the Modulan vehicle produced significantly higher levels of drug in the tear pool than the Plastibase vehicle after 4 h.

The rate and extent of drug release from the vehicles to the tear pool will

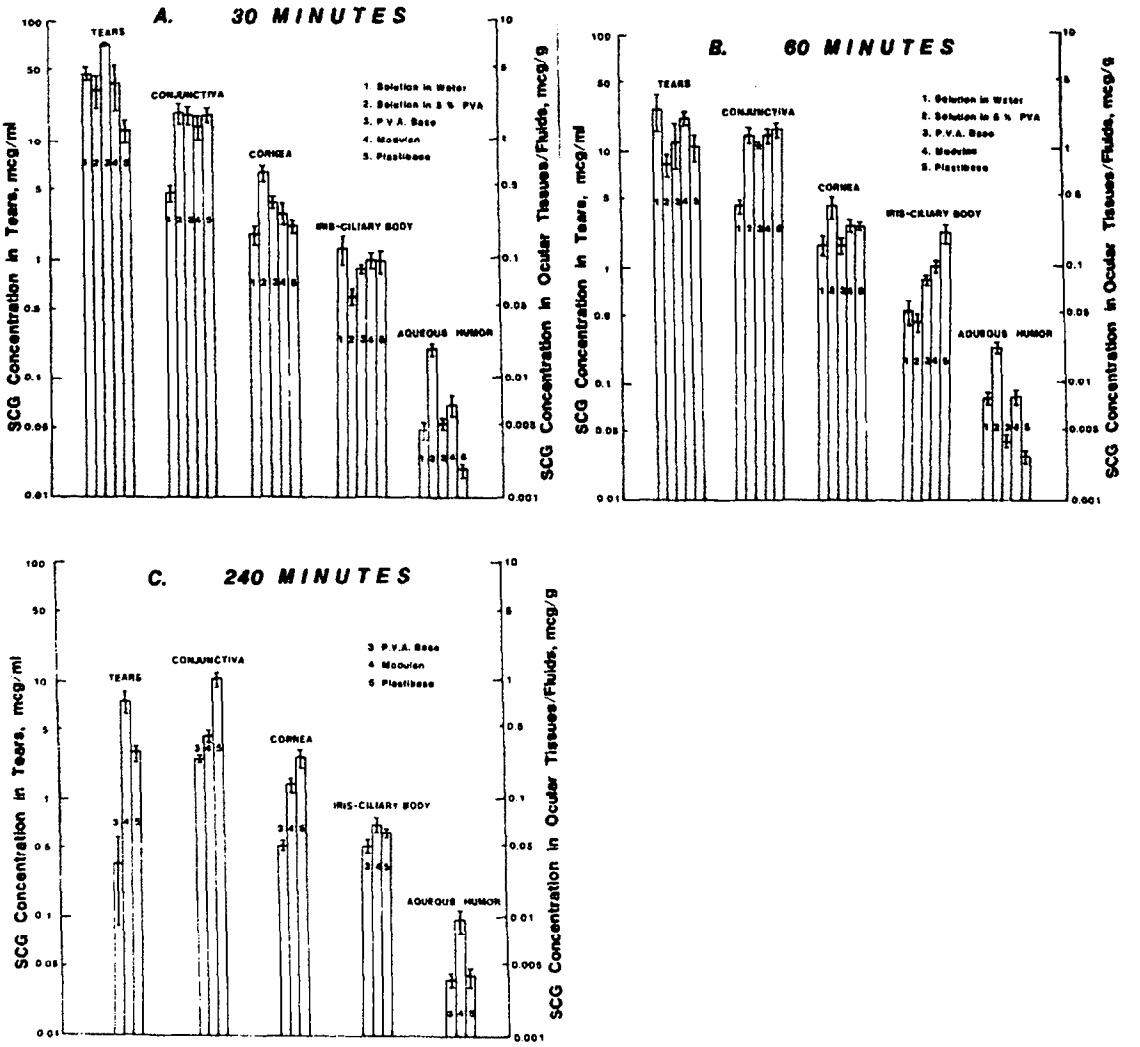

Fig. 2. Sodium cromoglycate concentration in various ocular tissues and fluids at 30, 60 and 240 min following application of drug in various vehicles. The concentration is expressed as $\mu g \cdot ml^{-1}$ for tears and aqueous humor and as $\mu g \cdot g^{-1}$ for conjunctiva, cornea and iris-ciliary body. Note the use of different axes. All concentrations are normalized for the amount of drug applied. Between 8 and 12 eyes were used for each tissue or fluid. Error bars represent standard error of the mean. Key: A, 30 min; B, 60 min; C, 240 min.

ultimately affect the drug concentration attained in the conjunctiva, cornea, iris-ciliary body and aqueous humor. As with solutions in water and 5% PVA (Lee et al., 1983), the uptake of sodium cromoglycate from the tear pool with the 3 vehicles studied was greatest in the conjunctiva (the target tissue), followed by the cornea, the iris-ciliary body and the aqueous humor in that order. During the first 60 min of drug administration, the concentration of drug achieved in the conjunctiva, one of the two tissues in direct contact with the tear pool, was not statistically different among the 3 bases, despite some significant differences in drug concentration in the tear pool (Fig. 2A and B), as already noted. In the cornea, the second tissue in

contact with tears, the Modulan and Plastibase vehicles showed no significant differences at 30 and 60 min even though, again, significant differences were observed in the tear pool. Such data could be taken to suggest that the transfer of sodium cromoglycate from the tear pool into the conjunctiva and cornea is rate-limiting in the overall absorption process. More extensive studies would be needed to investigate this possibility. After 4 h had elapsed, the Plastibase preparation had provided the highest concentration of drug in these two tissues (Fig. 2C), in contrast to the tear pool where the Modulan preparation had been superior at this time interval. The levels produced by the PVA preparation were clearly below those from the other two products.

In summary, the ocular disposition of sodium cromoglycate is influenced by the lipophilic characteristics of the base housing the drug. Not surprisingly, the PVA base behaved in a manner similar to the 5% PVA aqueous solution examined previously in that it provided drug in the tear film most readily but failed to significantly prolong the release of drug. The lipophilic bases, Modulan and Plastibase, by comparison were much more capable of maintaining a high drug concentration in the tear film, conjunctiva and cornea over a 4 h period, even though they produced somewhat lower concentrations at the earlier times. As early as 30 min post-dosing, all 3 bases were capable of providing cromoglycate to the conjunctiva at a concentration higher than that resulting from an aqueous solution. Of the 3 bases studied, that using Plastibase was the most effective in maintaining drug concentration in the conjunctiva over the period studied. Paradoxically, the Modulan-based preparation resulted in the highest drug concentrations in the tear pool after 4 h. Studies with pilocarpine (Sugaya and Nagataki, 1978) suggest that data obtained with the rabbit eye can be extrapolated to man. More recently, Saettone et al. (1982) have compared 4 different vehicles containing tropicamide in humans and rabbits. The mydriatic effect was found to be more pronounced in humans. Taken together, these studies suggest that effects noted in the rabbit eye will be of at least equal significance in man. Accordingly, Plastibase and, to a lesser extent, Modulan should be viable candidates for further evaluation in developing an effective ointment delivery system for sodium cromoglycate in the prophylaxis of vernal keratoconjunctivitis. Quite obviously, the choice of vehicle will also be influenced by other factors, such as patient acceptance, the potential for visual disturbances and the physical and chemical stability of the product.

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