International Journal of Pharmaceutics, 61 (1990) 15–25
Elsevier

IJP 02035

Evaluation of viscous ophthalmic vehicles containing carbomer by slit-lamp fluorophotometry in humans

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(Received 11 October 1989)
(Accepted 3 November 1989)

Key words: Slit-lamp fluorophotometry; Viscous eye drop; Ocular retention; Precorneal kinetics; Carbomer; EDTA

Summary

Slit-lamp fluorophotometry was used to evaluate in humans the precorneal kinetics of viscous eye drops containing carbopol 940, disodium EDTA and a fluorescent tracer. The ocular retention of the tracer depends on the concentration of the polymer instilled. The addition of disodium EDTA does not improve the precorneal kinetics significantly.

Introduction

A promising approach to lengthen the ocular contact time and to improve the bioavailability of a therapeutic agent is the addition of water-soluble, natural, semi-synthetic or synthetic viscolysers to ophthalmic vehicles (Benedetto et al., 1975; Hardberger et al., 1975; Saettone et al., 1984).

Besides the viscosity increase of the vehicle, Hui and Robinson (1985) demonstrated the utility of bioadhesion of polymers to reduce the drainage loss after instillation of ophthalmic formulations, hence to improve drug absorption or local action.

Several polyanionic polymers were proposed for ophthalmic formulations (Saettone et al., 1986; Gurny et al., 1987). Consequently, polyacrylate preparations were developed as long-lasting artificial tears for the relief of dry eye syndrome and traumatic injury. Their efficacy was attributed to the mucomimetic, rheological and lubricating properties of these high molecular weight polymers (Alcon Laboratories, 1984; Toko, 1985; Thilo, 1986). Recently gels for antibiotic therapy have been formulated (Tabbara et al., 1989). The claimed advantages of these commercially available products are good tolerance, prolonged contact time on the ocular surface and good miscibility with the lacrimal fluid (Leibowitz et al., 1984; Marquardt and Christ, 1986; Brewitt, 1988).

The aim of the present study is to evaluate the potential of viscous eye drops containing carbomer to prolong the precorneal retention of sodium fluorescein in humans. Moreover, the influence of disodium EDTA on the precorneal kinetics of the fluorescent tracer is also examined. Considering the effect of Ca^{2+} on the viscosity and the tertiary

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structure of mucin, the chelating agent disodium EDTA could influence the strength of the mucoadhesion of polyanionic polymers (Huth et al., 1981; Leung and Robinson, 1988).

A non-invasive method causing minimal disturbance to normal physiological functions is chosen. The fluorescence decay of sodium fluorescein in the precorneal tear film of humans is measured with a slit-lamp fluorophotometer.

Materials and Methods

Materials

Carbomer (Carbopol 940, M_r 4×10^6) henceforth designated as C 940, was obtained from B.F. Goodrich Chemical Co. (Leidschendam, The Netherlands). Sodium fluorescein was obtained from Fluka (Buchs, Switzerland). Mannitol, Na_2EDTA and all other chemicals were of analytical grade. Bovine serum albumin, bovine submaxillary gland mucin, lysozyme and γ-globulin (bovine Cohn fraction II) were purchased from Sigma (München).

Solution preparation

Carbomer was well dispersed in double-distilled water by mechanical stirring at room temperature until a solution free of lumps was obtained. The mixture was then neutralized and adjusted to pH 7.5 with 10% NaOH solution to yield a clear viscous solution. As a stabilizing and tonicity agent, 5% w/w mannitol was used.

Sodium fluorescein and Na_2EDTA were added to the vehicles to final concentrations of 0.05 and 0.01% w/w, respectively.

The solutions, stored in glass containers, were sterilized by autoclaving at 121° C for 20 min. The composition of the various vehicles tested is indicated in Table 1.

Physico-chemical measurements

Osmolality and pH. The osmolality of each solution was determined with a vapor pressure osmometer (model 5500 Wescor, Logan, UT). The pH of the vehicle was measured with a Radiometer PH M63 pH meter (Radiometer, Copenhagen),

Rheological measurements. Rheological measurements on the sterilized vehicles were carried out at 32° C (corneal surface temperature) using a capillary Ostwald viscometer (KPG Viskosimeter Schott Geräte, Mainz) and a rotary viscometer (Rheomat 30; Contraves, Zürich). Dependent on the flow properties of the vehicles different bops and cups are used. The shearing stress was determined as a function of shear rate in the range $D = 2\text{–}200\ s^{-1}$.

The apparent viscosity of vehicles was characterized either by means of complete flow curves or single viscosity values determined at a defined shearing rate.

Surface tension measurement. The surface tension was measured at 22 ± 1.0° C by the Wilhelmy plate method, using a Cahn electrobalance. Details of the procedure have been described previously (Ludwig and Van Ooteghem, 1989).

TABLE 1

Characteristics of the viscous solutions

Composition	pH	η (mPa s)	Osmolality (mosmol/kg)	σ (mN/m)
(1) mannitol (5%)	8.99	0.93	283	71.1
(2) + C-940 (0.10% w/w)	7.56	40	308	71.0
(3) + C-940 (0.15% w/w)	7.45	144	–	70.3
(4) + C-940 (0.20% w/w)	7.55	329	–	70.0
(5) + C-940 (0.10% w/w) + EDTA	7.72	29	308	71.0
(6) + C-940 (0.15% w/w) + EDTA	7.79	102	313	70.8
(7) + C-940 (0.20% w/w) + EDTA	7.85	270	–	70.5

Interaction of carbomer with the tracer and components of the lacrimal fluid

Tracer. The possibility of complex formation between the tracer and carbomer was examined by equilibrium dialysis of 0.12% lysozyme.

Cleaned Visking cellulose membrane was stretched firmly over the end of a glass tube and tied. Then 2 ml of each carbomer solution was placed in the glass tube, then immersed in a beaker containing 20 ml of a 5% w/w mannitol solution and the same amount of NaOH used to neutralize the carbomer.

Dialysis was carried out at room temperature and a state of equilibrium was reached after 48 h or several days depending on the viscosity of the polymer solution examined. The concentration of sodium fluorescein was determined spectrophotometrically at 490 nm.

Ions. Several studies pointed to the effect of electrolytes on the viscosity of carbomer gels (Testa and Etter, 1973; Toko, 1985). In order to obtain a rough estimate of the viscosity of the C 940 solution after instillation in the conjunctival sac and mixing with the resident lacrimal fluid, rheological measurements were performed after dilution of the vehicles with simulated lacrimal fluid (SLF).

Based on literature data, the following composition of SLF was selected: K, 24 mmol; Ca, 0.4 mmol, Mg, 0.5 mmol; Na, 134 mmol; HCO_3, 26 mmol; Cl, 134 mmol/l; adjusted to pH 7.5 with 0.1 N HCl (Van Haeringen, 1981). The osmolality of SFL was 277 mosmol/kg. Considering that 10 μl of each vehicle is applied and that normally the volume of the lacrimal fluid is about 7 μl, the carbomer solutions were diluted in the following manner: 10 : 7, 10 : 10.5, 10 : 14 with SLF solution (Mishima et al., 1966).

The flow curves of different dilutions were determined at 32 °C with a Rheomat 30 rotational viscometer.

Proteins. To investigate the effect of tear proteins on the carbomer solutions, 0.39% albumin, 0.50% mucin, 0.12% lysozyme and 0.16% w/v γ-globulin were added to the SLF solution (Frauch, 1978). The same dilutions and rheological measurements were performed as described in the previous section.

Slit-lamp fluorophotometry

The influence of the viscous vehicles on the precorneal kinetics of sodium fluorescein was examined using a slit-lamp fluorophotometer. Technical details of the apparatus have been described in previous papers (Ludwig and Van Ooteghem, 1986, 1987).

Six adult volunteers, from whom informed consent was obtained, participated in the study, which was carried out under medical supervision.

The solution (10 μl) was instilled in the lower conjunctival sac of the left eye. The fluorescence decay curve (fluorescence intensity vs time) was recorded. Details of the instillation procedure and fluorescence monitoring are given in a previous paper (Ludwig and Van Ooteghem, 1989a).

Results and Discussion

Physical characteristics

All solutions tested exhibit an isohydric pH after sterilization. The apparent viscosity data reported in Table 1 were obtained with the rotational viscometer at a shear rate of $D = 200\ s^{-1}$. The viscosity of the mannitol solution, however, was measured with the capillary viscometer. The addition of disodium EDTA to the vehicles decreased the viscosity of the C 940 solutions.

Because of the high apparent viscosity of some vehicles, no reproducible osmolality measurements could be performed. From the results reported the concentration of the polymer does not seem to have a significant influence on the osmolality of the vehicles.

Carbomer solutions do not exhibit surface activity.

Rheological behaviour

As shown in Fig. 1 the rheograms of the carbomer solutions exhibit a pseudoplastic flow with no hysteresis. This indicates that no structural change occurred under these shearing conditions.

The apparent viscosity of the carbomer solutions increased rapidly with concentration. This demonstrates strong interaction between macro-

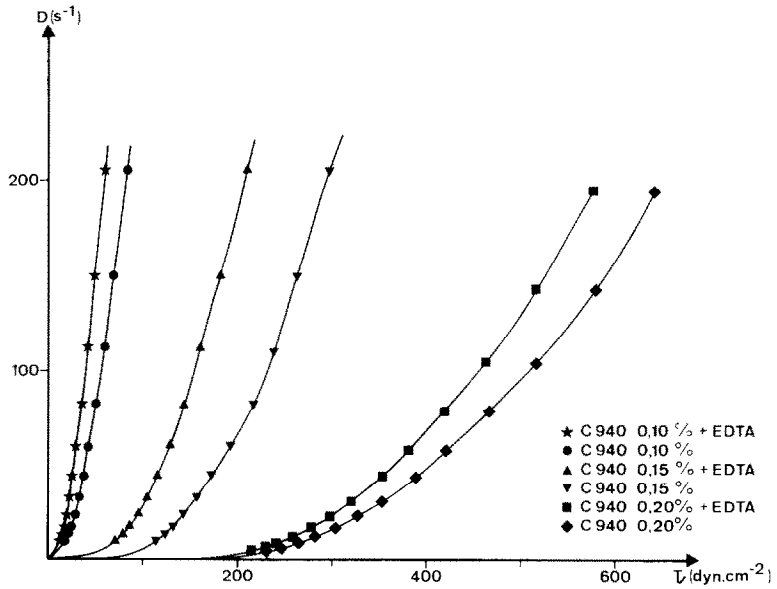

Fig. 1. Rheograms of the viscous solutions tested.

molecules. During neutralization of the aqueous carbomer dispersion the addition of NaOH causes dissociation of the carboxyl groups present in the polymer chain, resulting in electrostatic repulsion between the charges and uncoiling of the flexible macromolecules into an extended structure. Bardet et al. demonstrated that this increase in interaction between polymer chains results in the formation of an irregular, loose network without strong cohesion (Bardet and Alain, 1975, Bardet and Cabrol, 1978; Alain and Bardet, 1982).

The flow curves of the C 940 solutions containing disodium EDTA are similar to the corresponding carbomer solutions, but show a marked decrease in apparent viscosity. The addition of EDTA and consequently of Na^+, shields the negative charges, reduces the expansion forces and decreases the entanglement of the polymer (Testa and Etter, 1973; Alain and Bardet, 1982).

In vivo the shear rate during blinking is estimated to be about 20000 s^{-1}, therefore the shear thinning effect is expected to distribute the vehicle evenly on the eye (Van Ooteghem, 1987).

Interaction of carbomer with the tracer and components of the lacrimal fluid

Tracer. The difference between the calculated concentration at equilibrium and the measured fluorescein concentration is less than 2% for each of the solutions examined. Therefore, no strong binding between sodium fluorescein and C 940 exists and thus the tracer diffuses freely in the viscous vehicle.

Ions and proteins. The apparent viscosity values at $D = 700^{-1}$, except for C 940 0.20% ($*D = 200\ s^{-1}$), after dilution with SLF, are given in Table 2.

The increase in ionic strength of the medium after dilution with SLF results in a marked decrease in viscosity. The addition of cations neutralizes the carboxyl groups, resulting in a signifi-

TABLE 2

Apparent viscosity values (mPa s)

	C 940 concentration		
	0.10%	0.15%	0.20%
No dilution	20.56	72.29	(368*)
+ SLF dilution 10 : 7	4.23	4.88	5.80
10 : 10.5	4.06	4.77	4.77
10 : 14	3.82	4.43	4.60
+ SLF with proteins			
dilution 10 : 7	4.47	6.14	7.16
10 : 10.5	4.16	5.49	6.48
10 : 14	4.13	5.29	5.80

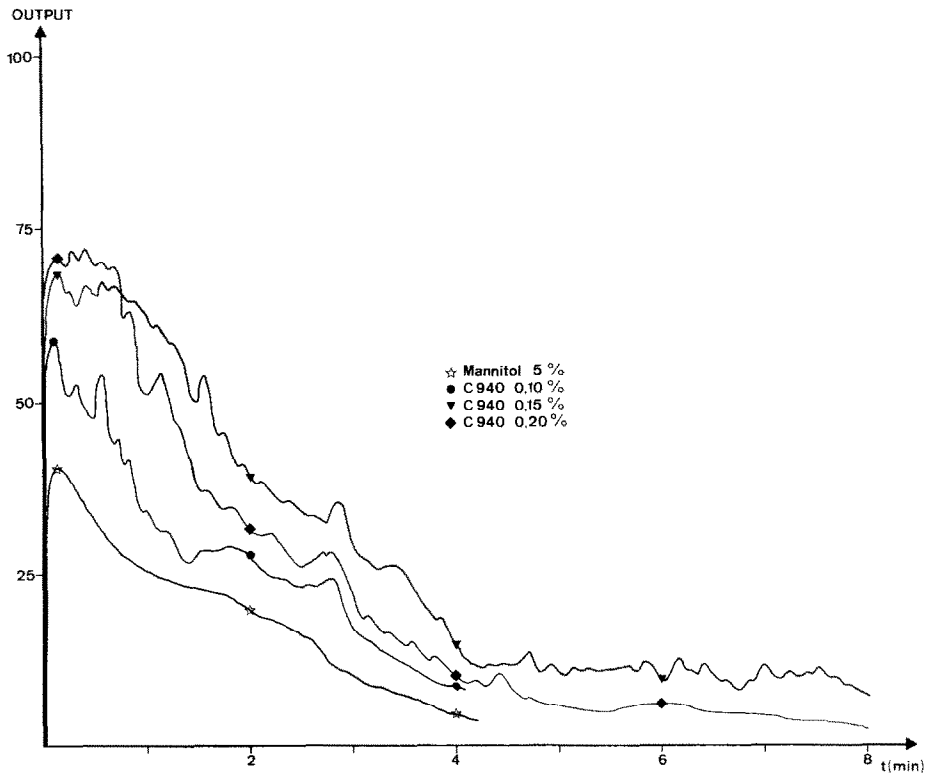

Fig. 2. Fluorescence decay curves of C 940 solutions (volunteer no. 4).

cant decrease in electrostatic repulsion. The uncoiled, extended macromolecule curls up and could even be partly desolvated by the divalent ions (Ikegami, 1964).

The viscosity data suggest that some weak interactions between carbomer macromolecules and proteins might exist.

Fluorescence decay curves

The standard procedure in previous studies consists of the registration of the decay curve during 4 min post-instillation (Ludwig and Van Ooteghem, 1989a). During the present experiments the registration period was extended to 8 min, considering the slow elimination of the tracer after instillation of the 0.15 and 0.20% C 940 vehicles.

Schematic plots of the fluorescence signal of the tracer as a function of time are shown in Figs 2 and 3. The output of the signal is expressed in arbitrary units. The plots of Figs 2 and 3 originate from experiments with volunteer no. 4.

The decay curves indicate that the addition of carbomer to the vehicle decreases the rapid fluorescence decay seen after instillation of the mannitol solution.

The instillation of the 0.15 and 0.20% C 940 solutions causes a 2.5-fold higher initial fluorescence signal with respect to the mannitol solution. These high fluorescence signals indicate an influence of the polymer on the tear film thickness, as the fluorescence intensity approximates tracer concentration × film thickness. In comparison with polyvinyl alcohol and cellulose derivatives, the effect of carbopol is smaller (Ludwig and Van Ooteghem, 1989b).

The carbomer solutions do not mix easily with the lacrimal fluid. An intensely fluorescent tear strip is observed, which means that part of the solution instilled remains in the conjunctival sac.

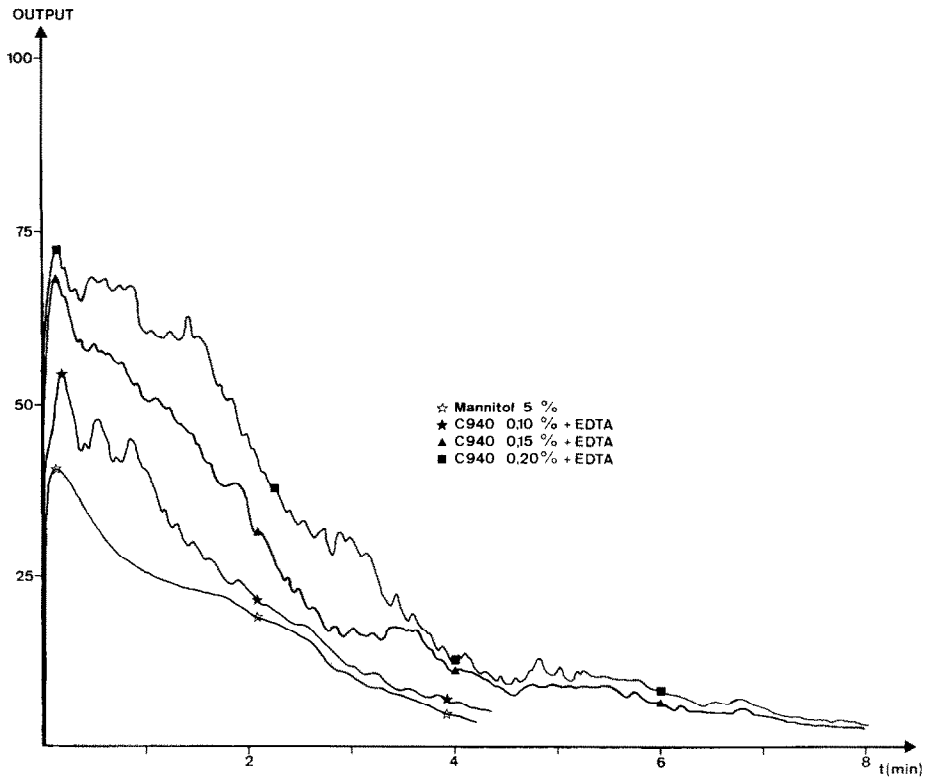

Fig. 3. Fluorescence decay curves of C 940 solutions with EDTA (volunteer no. 4).

In the case of the highly viscous C 940 solution, a uniform fluorescent film is observed on the surface of the eye only after a few minutes. This is unexpected because of the pseudoplastic behaviour and the high surface tension of the solutions. Very irregular decay curves are observed, as illustrated in Fig. 4. The amount of tracer in contact with the cornea depends partly on the blinking force, which squeezes the tracer out of the polymer network into the tear film. Therefore, the differences in profiles of the decay curves of the six volunteers are mainly due to the mixing efficacy of the lid movements during blinking.

From the decay curve (Fig. 4) it can be deduced that the elimination of the tracer does not obey first-order kinetics during the first part of the fluorescence monitoring. A homogeneous spreading of the solutions seems to occur only after sufficient dilution by basal lacrimation, resulting in a more regular fluorescence decay profile.

In general, mixing problems are observed with the 0.15% C 940 and especially with the sticky and sometimes irritating 0.20% C 940 solutions. Moreover, the volunteers complained of blurred vision not immediately, but 1–3 min post instillation. This is probably due to the interaction of the resident lacrimal fluid with the instilled solution, the disruption of the polymer network and the steric rearrangement of the polymer chain.

A pain sensation at the inner canthus was also reported by some volunteers. This is attributed to some drainage difficulties for the high molecular weight polymer through the puncta into the lacrimal sac.

The C 940 solutions with disodium EDTA exhibit the same phenomena as the corresponding C 940 solutions, but the discomfort is less pronounced.

Tear elimination coefficient and AUC value

When the decay profile obeyed first-order kinetics fairly well, the data were subjected to

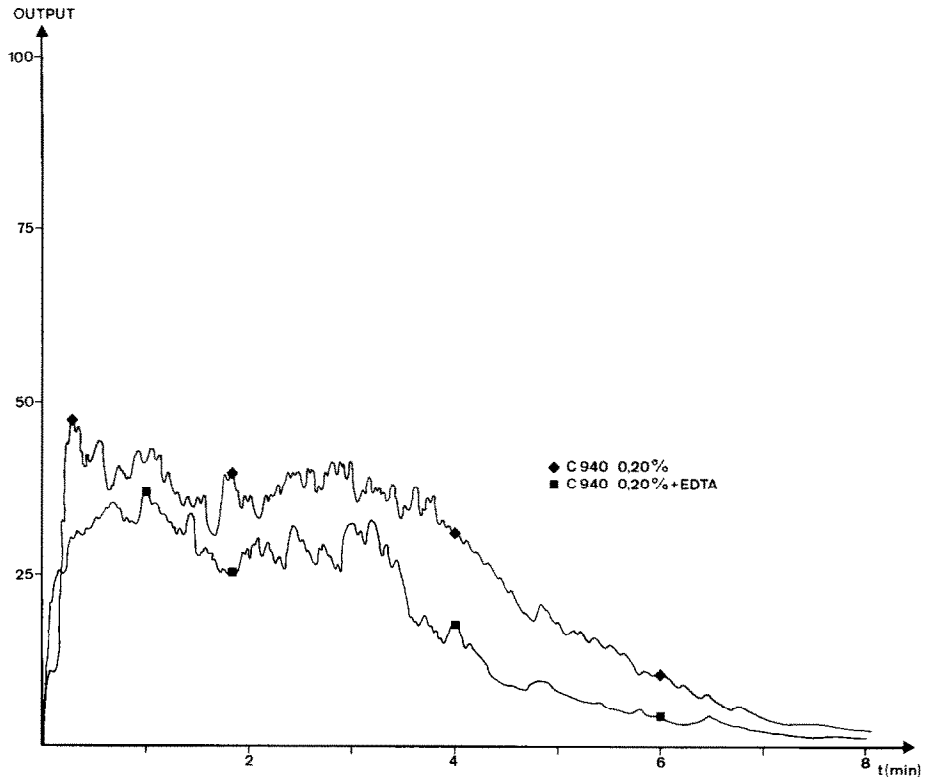

Fig. 4. Fluorescence decay curves of C 940 (0.20%) and C 940 (0.20%) with EDTA (volunteer no. 2).

linear regression analysis and the first-order rate constant, tear elimination coefficient k, was determined. The areas under the decay curve (AUC) were calculated using the trapezoidal rule.

From the recorded decay curve tear elimination coefficients k_1 (from 0 to 4 min) and k_2 (from 4 to 8 min post-installation) and also the corresponding AUC_1 and AUC_2 values were calculated. Each solution was tested six times on each volunteer. The mean values of each series and the SD values are summarized in Table 3.

All polymeric vehicles are capable of increasing the ocular retention of the tracer. When comparing the k values, k_1 and even k_2 values of the viscous vehicles are always smaller than the k_1 value of the mannitol solution. Besides, the AUC_2 values of the carbomer solutions are similar or larger than the AUC_1 value of the aqueous standard vehicle. Thus, carbomer seems to be a useful viscolyser for the formulation of sustained ocular drug delivery.

Significant inter- and intra-subject variations were observed. The marked differences between AUC values are due to the differences in efficacy of the blinking movement to spread the vehicle over the ocular surface.

Influence of the viscosity

To investigate the influence of the viscosity of the carbomer solutions the mean k_1 and AUC_1 of the various viscous solutions were compared, for each volunteer separately, with the k_1 and AUC_1 value of the mannitol solution. The elimination rate decrease and AUC increase, expressed as %, are given in Table 4. The data reported in Table 4 show that the elimination decreased when the carbomer concentration was increased. Non-parametric statistical analysis revealed that a concentration of 0.15% or 0.20% carbomer is necessary to increase the residence time significantly.

Considering the dramatic viscosity decrease after dilution with SLF solutions, smaller dif-

TABLE 3

Tear elimination coefficient k_1 and k_2 (s^{-1}) ($\times 10^{-3}$) and AUC_1 and AUC_2 values of the decay profiles

Vehicle	Volunteer					
	1		2		3	
	$k_1 \pm$ SD	$k_2 \pm$ SD	$k_1 \pm$ SD	$k_2 \pm$ SD	$k_1 \pm$ SD	$k_2 \pm$ SD
Mannitol 5%	14.04 ± 3.73	–	9.34 ± 3.04	–	11.37 ± 2.28	–
+ C 940 0.10%	10.14 ± 4.01	–	7.35 ± 2.24	–	5.11 ± 1.01	–
+ C 940 0.15%	9.30 ± 1.56	–	4.83 ± 0.94	–	3.27 ± 0.87	–
+ C 940 0.20%	N	6.96 ± 2.67	N	4.83 ± 1.52	N	3.12 ± 1.46
+ C 940 0.10% + EDTA	10.35 ± 1.73	–	8.79 ± 2.03	–	5.76 ± 1.46	–
+ C 940 0.15% + EDTA	8.26 ± 1.04	–	4.77 ± 1.15	4.94 ± 0.97	4.94 ± 1.55	3.75 ± 1.53
+ C 940 0.20% + EDTA	5.67 ± 2.77	3.18 ± 2.45	2.30 ± 0.87	5.13 ± 2.31	3.89 ± 1.60	4.42 ± 1.30
	Volunteer					
	4		5		6	
	$k_1 \pm$ SD	$k_2 \pm$ SD	$k_1 \pm$ SD	$k_2 \pm$ SD	$k_1 \pm$ SD	$k_2 \pm$ SD
Mannitol 5%	10.07 ± 2.57	–	5.15 ± 1.95	–	19.55 ± 6.00	–
+ C 940 0.10%	8.22 ± 2.57	–	3.55 ± 1.01	–	19.47 ± 7.03	–
+ C 940 0.15%	6.95 ± 1.91	–	3.82 ± 1.07	–	12.27 ± 4.72	–
+ C 940 0.20%	9.18 ± 4.33	3.99 ± 1.75	2.46 ± 0.89	2.39 ± 0.99	N	2.39 ± 1.11
+ C 940 0.10% + EDTA	7.32 ± 1.75		4.89 ± 0.96		6.65 ± 1.33	
+ C 940 0.15% + EDTA	6.49 ± 1.28	4.71 ± 0.98	3.08 ± 1.76	3.98 ± 1.76	4.92 ± 0.65	4.61 ± 1.52
+ C 940 0.20% + EDTA	7.13 ± 2.28	6.65 ± 2.58	2.05 ± 0.72	2.46 ± 0.92	N	1.90 ± 0.81
	Volunteer					
	1		2		3	
	$AUC_1 \pm$ SD	$AUC_2 \pm$ SD	$AUC_1 \pm$ SD	$AUC_2 \pm$ SD	$AUC_1 \pm$ SD	$AUC_2 \pm$ SD
Mannitol 5%	18.61 ± 3.06	–	16.38 ± 5.35	–	8.28 ± 1.39	–
+ C 940 0.10%	51.19 ± 11.42	–	52.33 ± 12.30	–	12.57 ± 6.19	–
+ C 940 0.15%	41.80 ± 18.32	–	43.39 ± 6.08	–	11.03 ± 2.56	–
+ C 940 0.20%	41.34 ± 16.34	6.86 ± 1.51	57.12 ± 21.48	27.69 ± 11.06	13.66 ± 6.05	11.05 ± 9.11
+ C 940 0.10% + EDTA	28.98 ± 9.07	–	28.89 ± 8.46	–	11.65 ± 1.30	–
+ C 940 0.15% + EDTA	34.37 ± 7.81	–	35.66 ± 10.70	12.60 ± 4.68	13.00 ± 2.70	5.38 ± 1.08
+ C 940 0.20% + EDTA	50.46 ± 1.23	14.15 ± 5.23	55.63 ± 8.40	21.18 ± 9.54	20.32 ± 6.58	9.59 ± 2.57
	Volunteer					
	4		5		6	
	$AUC_1 \pm$ SD	$AUC_2 \pm$ SD	$AUC_1 \pm$ SD	$AUC_2 \pm$ SD	$AUC_1 \pm$ SD	$AUC_2 \pm$ SD
Mannitol 5%	41.64 ± 14.64	–	28.95 ± 9.40	–	13.71 ± 5.10	–
+ C 940 0.10%	60.72 ± 10.44	–	25.50 ± 8.67	–	14.23 ± 6.76	–
+ C 940 0.15%	71.36 ± 15.56	–	20.48 ± 2.27	–	26.10 ± 8.85	–
+ C 940 0.20%	64.80 ± 10.09	11.79 ± 10.04	19.27 ± 5.63	16.58 ± 5.28	27.52 ± 3.58	25.07 ± 8.31
+ C 940 0.10% + EDTA	38.47 ± 9.00	–	23.30 ± 2.01	–	23.62 ± 6.73	8.67 ± 2.45
+ C 940 0.15% + EDTA	71.42 ± 17.37	20.01 ± 5.70	26.03 ± 6.28	11.18 ± 3.96	22.15 ± 7.87	9.71 ± 4.01
+ C 940 0.20% + EDTA	76.86 ± 18.78	18.99 ± 6.43	43.43 ± 22.69	29.18 ± 11.80	27.71 ± 6.46	22.76 ± 4.99

ferences between the carbopol solutions could be expected. Therefore, the rate and extent of the interaction in vivo are probably not the same for each solution and each volunteer.

The highest AUC improvement for almost all volunteers occurs after instillation of the C 940 0.20% solution with EDTA. From the statistical analysis it could be deduced that the differences

TABLE 4

k_1 decrease (%) and AUC increase (%)

	Vehicle	Volunteer					
		1	2	3	4	5	6
(k_1 increase)	C 940 0.10% w/w	28	21	55 [b]	18	31 [a]	0
	C 940 0.15% w/w	34 [a]	48 [b]	70 [b]	31 [a]	26	37
	C 940 0.20% w/w	N	N	N	8	52 [a]	N
	C 940 0.10% w/w + EDTA	26	6	49 [b]	27	5	66 [b]
	C 940 0.15% w/w + EDTA	41 [b]	49 [b]	57 [b]	36 [a]	40 [a]	75 [a]
	C 940 0.20% w/w + EDTA	60 [b]	75 [b]	66 [b]	29 [a]	60 [b]	N
(AUC increase)	C 940 0.10% w/w	175 [a]	219 [b]	52	47 [a]	−11	4
	C 940 0.15% w/w	125 [b]	164 [b]	33	73 [a]	−29	90
	C 940 0.20% w/w	122 [a]	249 [b]	65 [a]	57 [a]	−33	101 [b]
	C 940 0.10% w/w + EDTA	56 [a]	76 [a]	41 [a]	−7	−20	72 [a]
	C 940 0.15% w/w + EDTA	85 [a]	118 [b]	57 [b]	73 [a]	−10	62 [a]
	C 940 0.20% w/w + EDTA	171 [b]	240 [a]	145 [b]	86 [a]	50 [a]	102 [a]

Statistically significant difference between the mannitol solution and the carbomer solution at [a] $P < 0.05$ and [b] $P < 0.01$, using Mann-Whitney test.
N, no first-order kinetics.

between the AUC values of the 0.15 and 0.20% C 940 solutions with disodium EDTA were significant for volunteers 1, 2, 3 and 5. However, the differences between the AUC values of the 0.15 and 0.20% C 940 solutions are not significant.

In the case of volunteer no. 5, paradoxical results were noted. The elimination rate was retarded as the carbomer concentration increased, but also the AUC_1 values were lower with respect to the mannitol solution. This means that the solution remained mostly in the conjunctival sac. With respect to formulation, the low AUC value may not be a negative factor per se in the case of a therapeutic agent with local action.

Compared to previous studies with Newtonian cellulose solutions, a similar k_1 decrease and AUC_1 increase are observed to those for the 7.5 mPa s solutions. Notwithstanding blurring of vision, carbomer solutions are better tolerated than hydroxypropylcellulose and hydoxypropylmethylcellulose solutions (Ludwig and Van Ooteghem, 1989a,b).

Influence of EDTA addition

The k_1 and AUC_1 ratio of the vehicles containing the same amount of carbomer were calculated, the results being summarized in Table 5. In general, the addition of disodium EDTA did not cause significant differences among k_1 or AUC_1 values. A possible reason could be the low concentration of EDTA used. However, this small amount was selected to avoid a deleterious effect

TABLE 5

k_1 ratio C 940/C 940 + EDTA solution and AUC_1 ratio C 940/C 940 + EDTA solution

	Volunteer	Concentration		
		0.10%	0.15%	0.20%
k_1 ratio	1	0.98	1.13	–
	2	0.84	1.01	–
	3	0.89	0.68	–
	4	0.89	1.07	1.29
	5	1.12	1.24	1.20
	6	0.73 [a]	2.49 [b]	–
AUC_1 ratio	1	1.77 [a]	1.22	0.82
	2	1.81 [b]	1.22	1.03
	3	1.08	0.85	0.67
	4	1.58 [a]	1.00	0.84
	5	1.09	0.79 [a]	0.44 [b]
	6	0.60 [a]	1.18	0.99

Statistically significant difference between the C 940 and the C 940 + EDTA solution at [a] $P < 0.05$ and [b] $P < 0.01$ using Mann Whitney test.

on the morphology and permeability of the cornea (Grass and Robinson, 1988), although the amount of EDTA incorporated has a positive effect on the stability of the formulations (Unlü et al., to be published).

The effect of disodium EDTA on the bioadhesion of the viscous solutions instilled is questionable. Good bioadhesion was obtained when intimate contact occurred after wetting, swelling and interpenetration of the polymer chains with the mucus network (Duchêne et al., 1988). Under the present conditions, the ocular mucus layer is very small (about 0.05 μm), thus the polymer has less opportunity to interact. Moreover, the mucoadhesive properties of polyacrylic acid occur mainly because of hydrogen bonding, while hydrophobic interactions with mucin are not significant (Leung and Robinson, 1988). But in the precorneal area the neutral pH value of the tears and the shielding of the carboxyl groups by cations diminish the interactions of carbomer with functional groups of mucin.

Compared to a 7.5 mPa s Newtonian polyvinyl alcohol solution, which is considered not to be bioadhesive, C 940 solutions show smaller k_1 and higher AUC_1 values (Ludwig and Van Ooteghem, 1989a). This could indicate that the sustained effect is either not only based on viscosity or that the viscosity decrease due to mixing with resident lacrimal fluid does not occur immediately.

Conclusions

This study demonstrates the relative effectiveness of carbomer C 940 at increasing the ocular retention of sodium fluorescein. The polymer is a useful viscolyser for the formulation of sustained ocular delivery systems. However, one should take into account the striking differences in acceptability of viscous solutions by the patients as far as blurred vision and mixing problems are concerned.

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