Available online at www.sciencedirect.com
SCIENCE DIRECT®
International Journal of Pharmaceutics 254 (2003) 59–64
international journal of pharmaceutics
www.elsevier.com/locate/ijpharm

Note

Influence of methyl-β-cyclodextrin and liposomes on rheological properties of Carbopol® 974P NF gels

Laïla Boulmedarat, Jean Louis Grossiord, Elias Fattal*, Amélie Bochot

UMR CNRS 8612, School of Pharmacy, Faculté de Pharmacie, Université Paris-Sud, 5 rue JB Clément, 92296 Châtenay-Malabry Cedex, France

Received 16 September 2002; accepted 11 November 2002

Abstract

The influence of positively-charged and sterically stabilized liposomes and/or methyl-β-cyclodextrin on rheological properties of Carbopol® 974P NF hydrogels was investigated. All formulations have displayed a shear-thinning behavior of Carbopol® gels, and the rate stress as a function of the shear rate was fitted using the Cross equation. An important loss of viscosity was observed when 1.5% Carbopol® gels were formed in Hepes/NaCl buffer or in a 5% aqueous solution of methyl-β-cyclodextrin. Nevertheless, when methyl-β-cyclodextrin was dissolved in buffer at 5% there was no additional effect on gel viscosity reduction. The incorporation of positively-charged and sterically stabilized liposomes at 2 mM of lipid concentration had no incidence on rheological properties of the Carbopol® gels, whereas gel viscosity was significantly increased in the presence of positively-charged liposomes at 10 mM of lipid concentration. Finally, the viscosity of hydrogels containing both liposomes and methyl-β-cyclodextrin tended to be close to control gels, remaining high and relevant for a topical delivery.

Keywords: Liposomes; Polyacrylate hydrogels; Methyl-β-cyclodextrin; Cross fitting

Due to their hydrophilic nature and bioadhesive characteristics, crosslinked poly (acrylic acid) polymers such as Carbopol® polymers are among the most widely used excipients for thickening topical vehicles (lotions, creams, and gels). Methyl-β-cyclodextrin is a modified cyclodextrin which is an oligosaccharide that can be used in topical gel formulation to act as well as solubilizer for lipophilic drugs by their complexation and as penetration enhancer by improving drug absorption (Loftsson and Masson, 2001). Recently, particulate systems such as liposomes have been considered for topical delivery (Niesmann, 1992; Schreier and Bouwstra, 1994). They can be added in a gel formulation to protect the encapsulated drugs against the degradation and to provide their controlled and sustained release (Bochot et al., 1998a; Ruel-Gariépy et al., 2002; Glavas-Dodov et al., 2002). In the present work, the rheological properties of Carbopol® 974P NF hydrogels, which are particularly interesting for pharmaceutical and cosmetic formulation, in the presence of methyl-β-cyclodextrin and/or liposomes (cationic and sterically stabilized liposomes) were investigated.

Multilamellar SPC:CHOL:SA (60:30:10 mol%) and SPC:CHOL:PEG_{2000}-DSPE (64:30:06 mol%) liposomes were prepared in Hepes/NaCl (10 mM/145 mM, pH 7.4) buffer by the film hydration method (Bangham et al., 1965). Liposome suspensions were extruded

* Corresponding author. Tel.: +33-1-46835568; fax: +33-1-46619334.
E-mail address: elias.fattal@cep.u-psud.fr (E. Fattal).

0378-5173/02/$ – see front matter © 2002 Elsevier Science B.V. All rights reserved.
doi:10.1016/S0378-5173(02)00683-X

through polycarbonate membranes with diameters of 0.4 and 0.2 μm consecutively. Oligolamellar vesicles were formed with a mean diameter, evaluated by Laser Light Scattering, around 185 nm and Zeta potential values of $+56 \pm 1.7$ and -39.9 ± 1.6 mV, respectively for SPC:CHOL:SA and SPC:CHOL:PEG_{2000}-DSPE liposomes. The total lipid content in liposome suspensions was evaluated by measuring the phosphatidylcholine amount using an enzymatic assay (Phospholipides enzymatiques PAP 150, Biomerieux, France).

Carbopol® 974P NF was used as a vehicle for the incorporation of liposomes and/or methyl-β-cyclodextrin (MeβCD). Gels (final concentration of 1.5%, w/v) were prepared by dispersing under stirring Carbopol® resin in distilled water or in Hepes/NaCl buffer in which methyl-β-cyclodextrin (2 or 5%, w/v) was previously or not added. Mixtures were kept before use during 24 h at +4 °C to ensure complete humectation of the polymer chains. The dispersions were neutralized to pH 7 with 18% NaOH solution. After dilution in Hepes/NaCl buffer, liposomes were incorporated under shaking with a vortex into Carbopol® vehicle (volume ratio 1:3) prepared in Hepes/NaCl buffer containing or not methyl-β-cyclodextrin (final concentrations of 2 or 5%, w/v). Lipid concentrations in the gels (final Carbopol® concentration of 1.5%, w/v) were 2 and 10 mM. Dispersions were neutralized as described above.

Carbopol® hydrogels containing methyl-β-cyclodextrin and/or liposomes were tested for basic rheological properties. Flow properties of 1.5% Carbopol® 974P NF hydrogels with incorporated liposomes (SPC:CHOL:SA and SPC:CHOL:PEG_{2000}-DSPE at 2 and 10 mM) and methyl-β-cyclodextrin (2 and 5%) were determined on a RotoVisco 1 RV1 rheometer (Haake, Rheo, France). Measurements were performed at 37 ± 0.5 °C with a shear rate ranged from 0 up to 200 s^{-1} for 2 min using the cone-plate C 60/1° (diameter: 6 cm, cone angle: 1°) measuring system. Flow properties of liposome-free and methyl-β-cyclodextrin-free control gels (i.e. gels prepared in water or in buffer) were examined in the same conditions.

All hydrogel formulations (Fig. 1A) studied have shown a shear-thinning behavior according to the Cross model with an excellent fitting ($R > 0.998$) described by the equation reported below (Table 1).

Table 1
Rheological parameters of Cross model for 1.5% Carbopol® 974P NF hydrogels at 37 °C

Sample (Gel made in)	η_0 (Pa s)	$\dot{\gamma}_b$ (s^{-1})	R
Water	45	12	0.9986
Hepes/NaCl buffer	8.1	11	0.9994
MeβCD water solution at 2%	30	18	0.9990
MeβCD water solution at 5%	20	16	0.9990
MeβCD buffer solution at 2%	5.8	13	0.9995
MeβCD buffer solution at 5%	4.6	10	0.9995

η_0: zero-shear rate viscosity (Pa s) (upper Newtonian plateau), $\dot{\gamma}_b$: characteristic shear rate (s^{-1}), and R: correlation coefficient.

$$\tau = \dot{\gamma}\left[\eta_\infty + \frac{(\eta_0 - \eta_\infty)}{1 + (\dot{\gamma}/\dot{\gamma}_b)^n}\right] \quad (1)$$

where τ represents the shear stress (Pa); $\dot{\gamma}$ the shear rate (s^{-1}); η_0 is the zero-shear rate viscosity (Pa s) (upper Newtonian plateau) acquainting with the gel structure "at rest"; η_∞ is the infinite-shear rate viscosity (Pa s) (lower Newtonian plateau); $\dot{\gamma}_b$ is a characteristic shear rate (s^{-1}) for which the viscosity value is $\eta = (\eta_0 + \eta_\infty)/2$ when $\dot{\gamma} = \dot{\gamma}_b$, informing about gel sensitivity to the shear stress; and n is a dimensionless exponent.

Carbopol® 974P NF gels formed in Hepes/NaCl buffer have shown an important loss of viscosity compared to the hydrogel prepared in distilled water (Fig. 1B). Modifications in the viscosity of Carbopol® 974P NF gel made in Hepes/NaCl buffer was due to the electrolytes (sodium ions Na^+) which are well known to reduce the thickening efficiency of Carbopol® polymers (Dittgen et al., 1997; Singla et al., 2000; Pavelic et al., 2001). However, the viscosity of 1.5% Carbopol® 974P NF gels still remained high under these conditions.

For Carbopol® 974P NF gels prepared in methyl-β-cyclodextrin water solutions, a change in the viscosity was observed for a concentration of 2% ($\eta_0 = 30$ Pa s versus $\eta_0 = 45$ Pa s for the control), whereas in the same conditions, 5% of methyl-β-cyclodextrin induced a more pronounced loss of gel viscosity ($\eta_0 = 20$ Pa s versus $\eta_0 = 45$ Pa s for the control) (Table 1). Cyclodextrins, are cyclic (α-1–4)-linked oligosaccharides of α-D-glucopyranose characterized by the presence of a hydrophobic central cavity and hydrophilic outer surface due to a large number of

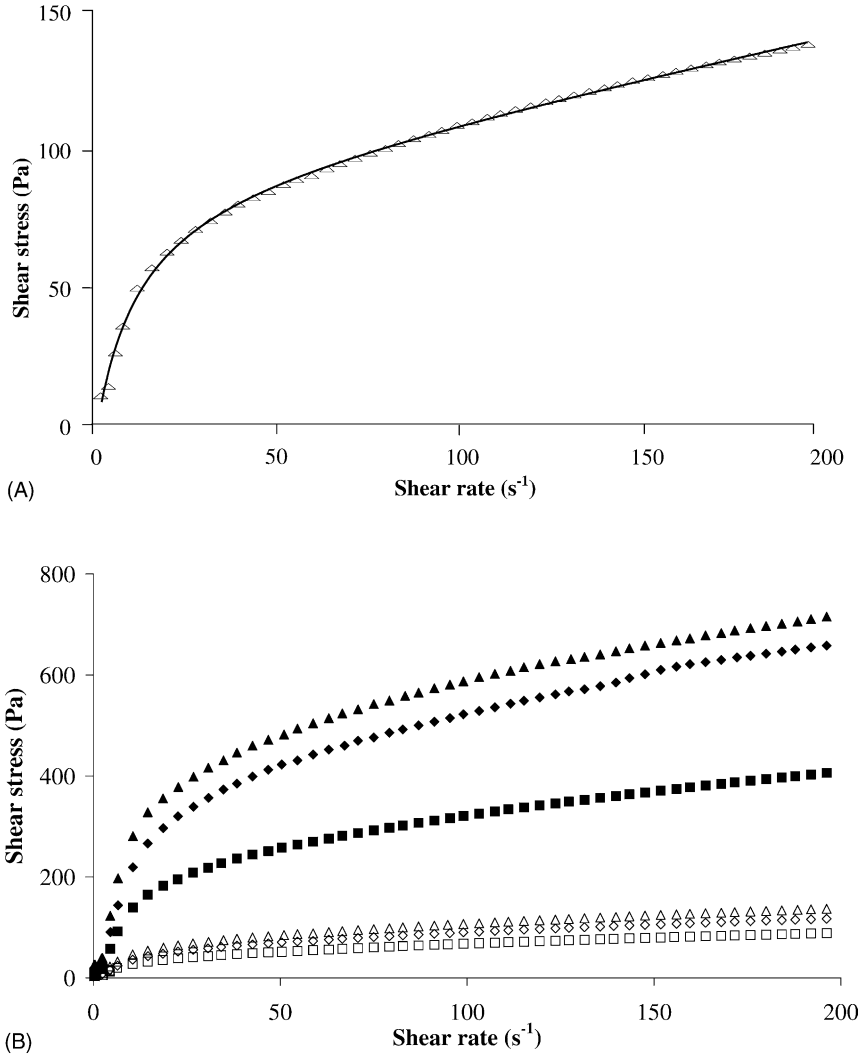

Fig. 1. Flow plots and Cross model correlation (—) for 1.5% Carbopol® 974P NF gels prepared in Hepes/NaCl (10 mM/145 mM) buffer pH 7.4 (▵) (A). Flow plots for 1.5% Carbopol® 974P NF gels prepared in water (▲), in Hepes/NaCl (10 mM/145 mM) buffer pH 7.4 (Δ), and in MeβCD water solutions at 2% (◆) and 5% (■), w/v or in methyl-β-cyclodextrin solutions at 2% (◇) and 5% (□), w/v prepared in Hepes/NaCl buffer (B).

hydroxyl groups that could interact with poly (acrylic acid) polymers. Compared to data obtained with hydroxyl donors: polyols (such as glycerine, propylene glycol, and polyethylene glycol), sugar alcohols such as mannitol, which were added (10–20%) to uncoil Carbopol® polymers in water systems (Barry and Meyer, 1979), methyl-β-cyclodextrin exhibited a different behavior. These reagents form hydrogen bond with the polymer molecule causing its unfolding. When methyl-β-cyclodextrin was replaced in the formulation by the natural γ-cyclodextrin, the viscosity was slightly increased compared to the control gel in water (data not shown). Methyl-β-cyclodextrin is a chemically modified cyclodextrin with a degree of methylation of 1.8, which is more lipophilic than natural cyclodextrins. Hydrophobic interactions could occur between the polymer chains and the methyl-β-cyclodextrin resulting in a reduction of the

polymer chains unfolding. Consequently, it may modify the polymer affinity for the hydration medium, hence decreasing its swelling.

Surprisingly, when Carbopol® 974P NF gels were made in methyl-β-cyclodextrin Hepes/NaCl solutions, no significant change in gel viscosity, even with 5% of methyl-β-cyclodextrin, was obtained compared to control gel prepared in the same buffer as shown by η_0 values (Table 1). The results indicated that cationic electrolytes and methyl-β-cyclodextrin added in the same preparation did not induce additional effects on viscosity decrease.

Table 2 showed the rheological parameters of Carbopol® 974P NF containing sterically stabilized vesicles (SPC:CHOL:PEG_{2000}-DSPE) or positively-charged liposomes (SPC:CHOL:SA) at different lipid concentrations (2 and 10 mM). The addition of liposomes whatever the composition into the gels at low lipid concentration (2 mM) did not change the rheological behavior and the viscosity (η_0 values in range of 5–8 Pa s) of the system compared to the control gel (no incorporated liposome hydrogel made in Hepes/NaCl buffer) (Table 2). In contrast, the rheological assays performed with positively-charged liposomes at 10 mM indicated that a high concentration of cationic lipid could significantly increase the viscosity of the preparation (Fig. 2B). This effect was not observed with SPC:CHOL:PEG_{2000}-DSPE liposomes at the same high lipid concentration (Fig. 2A). In most of the studies, liposomes incorporated within hydrogel (Poloxamer 407, Bochot et al., 1998b; Carbopol® 974P NF, Pavelic et al., 2001) did not modify the rheological properties of the systems. However, Ruel-Gariépy et al., 2002 have noted that negatively-charged liposomes were able to interact with thermosensitive chitosan-based hydrogel to decrease slightly the gelation rate and the gel strength. In the present study, stearylamine might be responsible for polymer chains reorganization probably due to the interaction between the cationic charges of the lipid and the carboxylic groups of the polymer.

Table 2
Rheological parameters of Cross model for 1.5% Carbopol® 974P NF hydrogels made in Hepes/NaCl buffer with incorporated liposomes at 2 and 10 mM of lipid concentration, containing or not methyl-β-cyclodextrin at 37 °C

Sample	η_0 (Pa s)	$\dot{\gamma}_b$ (s^{-1})	R
Hepes/NaCl buffer	8.1	11	0.9994
SPC:CHOL:PEG_{2000}-DSPE 2 mM			
0% MeβCD	5.6	15	0.9994
2% MeβCD	4.4	15	0.9993
5% MeβCD	4.5	9.4	0.9995
SPC:CHOL:PEG_{2000}-DSPE 10 mM			
0% MeβCD	5.0	15	0.9995
2% MeβCD	8.3	7.0	0.9995
5% MeβCD	5.5	9.8	0.9995
SPC:CHOL:SA 2 mM			
0% MeβCD	5.7	10	0.9991
2% MeβCD	5.7	6	0.9994
5% MeβCD	7.6	2.8	0.9995
SPC:CHOL:SA 10 mM			
0% MeβCD	18	10	0.9982
2% MeβCD	9.4	9.1	0.9990
5% MeβCD	5.4	10	0.9994

η_0: zero-shear rate viscosity (Pa s) (upper Newtonian plateau), $\dot{\gamma}_b$: characteristic shear rate (s^{-1}), R: correlation coefficient.

Flow properties of Carbopol® 974P NF hydrogels with co-incorporated SPC:CHOL:PEG_{2000}-DSPE liposomes (2 and 10 mM) and methyl-β-cyclodextrin (2 and 5%) were almost the same than those observed for the control gel prepared in buffer. The addition of methyl-β-cyclodextrin at 2 and 5% into Carbopol® 974P NF hydrogels with incorporated SPC:CHOL:SA liposomes at 10 mM of lipid, induced a decrease of viscosity reaching viscosity values of the control gel, even less in the case of methyl-β-cyclodextrin at 5% (Table 2).

Independently from liposome formulation, zero-shear rate η_0 values were not modified by the temperature (20 and 37 °C) and after gel storage for 30 days at +4 °C (data not shown). Furthermore, despite viscosity reduction, hydrogels were slightly sensitive to the shear stress as notified at 37 °C by the $\dot{\gamma}_b$ relatively high values for all formulations (Tables 1 and 2). These results indicated that both liposomes and methyl-β-cyclodextrin were not incompatible with Carbopol® 974P NF hydrogels.

Carbopol® 974P NF hydrogels with incorporated liposomes and methyl-β-cyclodextrin have shown a shear-thinning behavior according to the Cross model with a good correlation. To formulate Carbopol® hydrogels in water in presence of methyl-β-cyclodextrin (5%), it is important to take into account the decrease of gel viscosity. In addition, the incorporation of li-

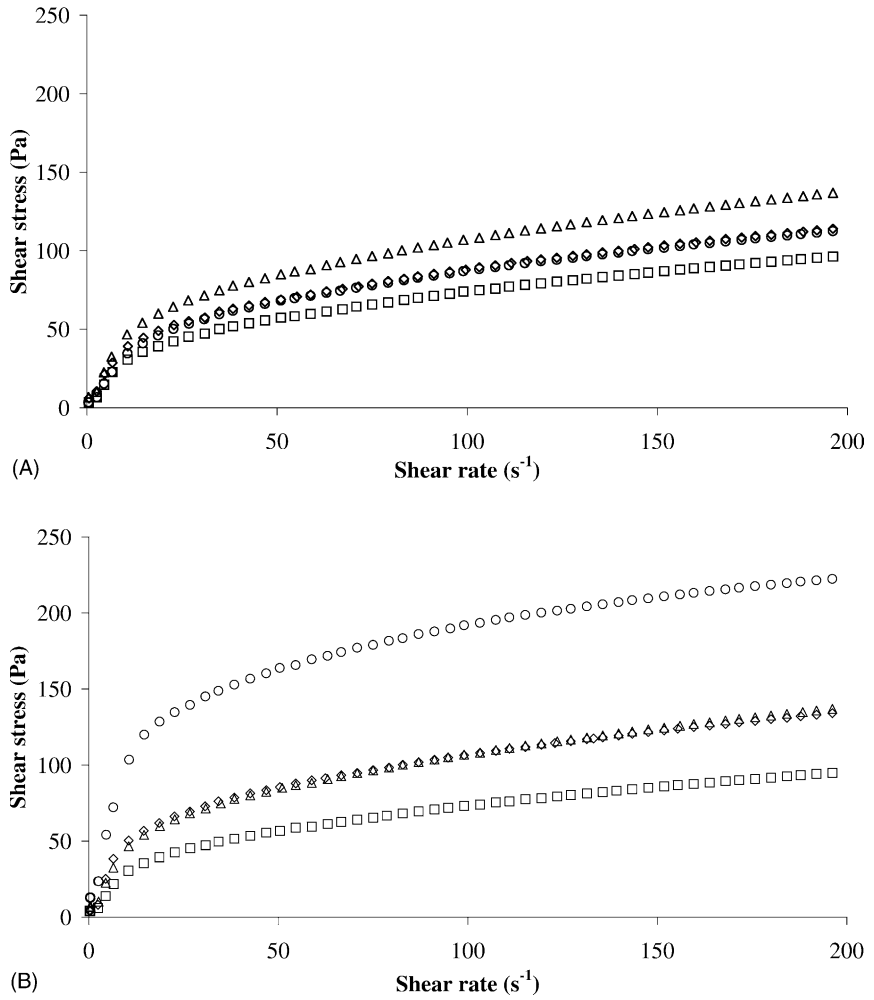

Fig. 2. Flow plots for 1.5% Carbopol® 974P NF gels with incorporated SPC:CHOL:PEG_{2000}-DSPE (A) and SPC:CHOL:SA (B) liposomes at 10 mM of lipid concentration and with methyl-β-cyclodextrin in buffer solutions at 2% (◇) and 5% (□), w/v. Control 1.5% Carbopol® 974P NF gels were prepared in Hepes/NaCl (10 mM/145 mM) buffer pH 7.4 with incorporated liposomes at 10 mM (○) of lipid concentration and methyl-β-cyclodextrin-free; methyl-β-cyclodextrin-free and liposome-free (Δ).

posomes at high lipid concentration and according to their composition could dramatically change the gel viscosity. However, the viscosity of hydrogels containing both liposomes and methyl-β-cyclodextrin tended to be close to the control gels, remaining high and desirable for a topical delivery. Further experiments, such as viscolelastic measurements, are needed in order to investigate the mechanisms of interaction between the polymer chains and methyl-β-cyclodextrin and stearylamine.

References

Bangham, A.D., Standish, M.M., Watkins, J.C., 1965. Diffusion of univalent ions across the lamellae of swollen phospholipids. J. Mol. Biol. 13, 238–252.

Barry, B.W., Meyer, M.C., 1979. The rheological properties of Carbopol gels I. Continous shear and creep properties of Carbopol gels. Int. J. Pharm. 2, 1–25.

Bochot, A., Fattal, E., Grossiord, J.L., Puisieux, F., Couvreur, P., 1998b. Characterization of a new ocular delivery system based on a dispersion of liposomes in a thermosensitive gel. Int. J. Pharm. 162, 119–127.

Bochot, A., Fattal, E., Gulik, A., Couarraze, G., Couvreur, P., 1998a. Liposomes dispersed within a thermosensitive gel: a new dosage form for ocular delivery of oligonucleotides. Pharm. Res. 15, 1364–1369.

Dittgen, M., Durrani, M., Lehman, K., 1997. Acrylic polymers: a review of pharmaceutical applications. S.T.P. Pharm. Sci. 7, 403–437.

Glavas-Dodov, M., Goracinova, K., Mladenovska, K., Fredro-Kumbaradzi, E., 2002. Release profile of lidocaine HCl from topical liposomal gel formulation. Int. J. Pharm. 242, 381–384.

Loftsson, T., Masson, M., 2001. Cyclodextrins in topical drug formulations: theory and practice. Int. J. Pharm. 225,15–30.

Niesmann, M.R., 1992. The use of liposomes as drug carriers in ophthalmology. Crit. Rev. Ther. Drug. Carrier Syst. 9, 1–38.

Pavelic, Z., Skalko-Basnet, N., Schubert, R., 2001. Liposomes gels for vaginal drug delivery. Int. J. Pharm. 219, 139–149.

Ruel-Gariépy, E., Leclair, G., Hildgen, P., Gupta, A., Leroux, J., 2002. Thermosensitive chitosan-based hydrogel containing liposomes for the delivery of hydrophilic molecules. J. Control. Release 82, 373–383.

Schreier, H., Bouwstra, J., 1994. Liposomes and niosomes as topical drug carriers: dermal and transdermal drug delivery. J. Control. Release 30, 1–15.

Singla, A.K., Chawla, M., Singh, A., 2000. Potential applications of carbomer in oral mucoadhesive controlled drug delivery system: a review. Drug. Dev. Ind. Pharm. 26, 913–924.