Phosphatidylcholine unilamellar liposomes as vehicles of a 1:2 metal complex dye in wool dyeing: kinetic aspects of the process

*L. Coderch; **S. Serra; *M.A. Manich; *M. Martí; *J.L. Parra and *A. de la Maza

*Departamento de Tensioactivos. Consejo Superior de Investigaciones Científicas (C.S.I.C.). Centro de Investigación y Desarrollo (C.I.D.). Barcelona (Spain).
**Sebastià Serra, Tints i Blanqueig, S.A. Sabadell (Spain)

Introduction

The interaction of weakly polar 1:2 metal-complex dyes with wool has received relatively little attention. In these dyes the metal atom is fully coordinated with the two dye ligands and, as a consequence, coordination of the dye metal ions with ligands such as amino or carboxyl groups in the fibre is not possible. Furthermore, although this dye type is hydrophilic in as much as it possesses anionicity, the dye is predominantly hydrophobic in character, since this relatively small anionicity is present within the large, essentially hydrophobic 1:2 metal-complex structure. Therefore, hydrophobic interactions operating between the dye and hydrophobic domains within the fibre, make an important contribution to substantivity (1).

Dyeing and diffusion properties of wool fibre, in particular, are believed to be governed by the lipid structure of the intercellular spaces that might act as "solvents" for hydrophobic textile chemicals (2). Transmission electron microscopy studies also provide evidence that dyes do, in fact, preferentially diffuse along easily-swollen regions such as the Cell Membrane Complex (CMC) (intercelullar diffusion) rather than through the cuticle cells (transcellular diffusion) (3).

Over the last decade, a number of investigations have been carried out using different carriers capable of reducing the degradative effect brought about in conventional wool dyeing. Thus, the technology of microencapsulation has given rise to a number of innovations utilizing the basic principles of targeting, slow release and protection of this sensitive fibre (4-6). The potential use of liposomes as carriers in wool finishing is based on (i) the similarity existing between the bilayer structure of the CMC and that of the liposomes, (ii) the important role played by the CMC in the processing of chemicals into the fibres and (iii) the relevant importance of the hydrophobic interactions in the structural organization of wool. In this sense, liposomes made with pure phosphatidylcholine (PC) or containing lipids present in the CMC such as cholesterol, have been used as vehicles for aqueous chlorine solutions in the wool chlorination processes (7,8). These applications resulted in an improvement in both the regularity and the homogeneity of these oxidative treatments minimizing wool degradation and facilitating the subsequent treatments in wool processing. Likewise, we investigated the use of liposomes as carriers of commercial milling acid dyes and disperse dyes in the dyeing of untreated wool samples (9-12).

In the present work we seek to extend these investigations by determining the effect caused by PC liposomes in the dyeing of untreated wool using the weakly polar 1:2 metal complex dye C.I. Acid Yellow 129 widely used in the wool industrie. To this end, we describe work on the physical stability of unilamellar liposomes containing this dye at different PC/dye weight ratios. This application has also been examined, focusing on the kinetic aspects of dye adsorption and the dye-fibre bonding forces on wool fibres. This information, together to that obtained for different dyes may allow us in the understanding of the mechanisms involved in the use of liposomes in wool dyeing and in to confirm the suitability of liposomes as carriers of different types of dye in this technological application

– *For summary, figures, tables and references see the spanish text.*

Received: 25/4/2000.

Experimental

Materials

Botany wool fabrics knitted from R64/2 tex (count 2/28) yarns were used. Samples were Soxhlet extracted for 2 hours with methylene chloride and rinsed with water purified by the Milli-Ro system (Millipore) and dried at room temperature. Commercially available Ciba-Geigy 1:2 metal complex dye Yellow Irgalan 2GL KWL (C.I. Acid Yellow 129) was selected; its chemical structure is given in Figure 1.

Phosphatidylcholine (PC) was purified from egg lecithin (Merck, Darmstadt, Germany) according to the method of Singleton (13) and shown to be pure by thin-layer chromatography (TLC). Phosphatidylcholine was stored in chloroform under nitrogen at –20°C until use. Polycarbonate membranes of 400 nm and 800 nm, and membrane holders used for liposome extrusion were purchased from Nucleopore (Pleasanton, CA). The nonionic surfactant Triton X-100 (octylphenol with 10 units of ethylene oxide and an active matter of 100 %) was supplied by Tenneco S.A. (Barcelona, Spain).

Preparation of Unilamellar Liposomes (LUV)

Large unilamellar vesicle liposomes of a defined size (400 nm) the PC concentration in bilayers ranging from 0.25 to 3.0 mmol/l (which corresponds to 0.56 - 6.81 % o.w.f.) and the dye concentration ranging from 0.1 to 1.0 % o.w.f. were prepared by reverse phase evaporation method following a previously described method (9). After preparation liposome suspensions were extruded through 800 and 400 nm polycarbonate membranes to obtain an uniform vesicle size distribution of about 400 nm. (14). The resulting liposome suspensions were then left to equilibrate for 15 minutes and immediately were applied in wool dyeing processes.

Dyeing Procedure

Wool knitted samples were treated with LUV liposome suspensions freshly prepared at PC concentration ranging from 0.56 to 6.81 % o.w.f. (corresponding to 0.25 - 3.0 mmol/l), the dye concentration remaining constant (1.0 % o.w.f.) or varying the dye concentration (from 0.1 to 1.0 % o.w.f.) the PC concentration remaining constant at 2.27 % o.w.f. (which corresponds to 1.0 mmol/l). The complete correspondence of PC concentrations in percentages o.w.f. and in mmol/l has been given in Table I.

The dye was applied at a constant pH 5.5 (controlled addition of acetic acid) and liquor ratio 30:1. Dyeing was started at 50°C and the temperature was raised by 0.9 °C/ min to 90°C. Dyeing was continued for 120 min. Thereafter, samples were rinsed with water for 10 min and dried at room temperature. Laboratory dyeing was carried out in a Multi-Mat dyeing machine (Renigal, Spain). Likewise, untreated wool knitted samples were dyed in the absence of liposome using a dyebath containing 5 % o.w.f. of anhydrous sodium sulphate at a constant pH of 5.5 (acetic acid). Dyeing conditions were similar than those used for dye-liposome systems.

Dyebath exhaustion was determined by spectrophotometry using a Shimadzu UV-265FW spectrophotometer. Dyebath liposome aliquots (0.5 ml) were periodically added to quartz cuvettes filled with 2.0 ml of aqueous solution of Triton X-100 (2.0 % w/v). The interaction between Triton X-100 and liposomes resulted in a complete solubilization of lipid vesicles via mixed micelles formation (15), turning the liposome suspensions into a clear solution. Figure 2 shows the effect of the cleavage of liposomes by Triton X-100 on the absorption spectra of the dye (PC concentration ranging from 2.27 to 6.81 % o.w.f.). It may be seen that the λ_{max} of the dye used in this study did not change in presence of increasing amounts of PC in the lipid/surfactant mixed micelles.

Mean Vesicle Size Distribution and Aggregation Measurements of Liposome Suspensions

Mean vesicle size and polydispersity of the liposome preparations were determined by a photon correlator spectrometer (Malvern Autosizer 4700c PS/MV). The studies of the particle size distribution were made by particle number measurements. Samples were adjusted to the appropriate concentration range. The measurements were made at 25°C with a detection angle of 90°. The aggregation state of the vesicles was estimated as a measure of the physical stability of the liposome suspensions. This was done by monitoring the variation of the mean vesicle size distribution of liposome suspensions as a function of time.

Bilayer Lipid Composition

The lipid concentration of liposomes was determined using the Iatroscan MK-5 TLC-FID analyser. Coupling thin-layer chromatography to an automated detection system based on flame ionization detection (FID) is a recent innovation, which has considerably improved the sensivity of TLC and allows quantitation of materials. This method has been used to quantify most kind of lipids from different sources (16,17). This is a useful method to quantify lipid mixtures even when these lipids are associated forming liposome suspensions in water.

Dye Extraction

The dye bonded superficially to the fibres by non polar forces (hydrophobic interactions, van der Waals

forces and hydrogen bonds) was extracted with pure ethanol at 25°C for 60 min (9). Subsequent extractions with ammonia solution (0.5 % v/v at 60°C for 15 min) stripped the dye diffused inside the fibre and not substantively bonded by ionic links (18). The concentration of extracted dye was evaluated in all cases by spectophotometry.

The percentages of bonded dye in wool fibres are expressed by the following equation:

$$C_B = [(C_A - C_E) / C_A] \cdot 100 \qquad [1]$$

where C_B is the relative percentage of bonded dye (%), C_A is the amount of adsorbed dye (dyebath exhaustion given in mg dye per g wool) and C_E is the total amount of extracted dye with pure ethanol and ammonia solutions (mg dye per g wool).

Optical Microscope Observations

Some representative yarns and fibres were isolated from the wool knitted samples dyed by LUV liposomes at different lipid concentration (PC concentration ranging from 0.56 to 6.81 % o.w.f.) or using the conventional dyeing procedure in the absence of liposomes. Samples were directly observed using a Reichert Polyvar optical microscopy (Leica, Wien, Austria). A 100 W low-voltage halogen lamp was used for illumination of dyed samples. The range of magnification used to determine the regularity in the distribution of dye in wool surface was from 200 to 1000 magnifications. In order to assess the changes in the dye diffusion into the fibre due to the presence of different liposomes, dyed fibres were embedded in Imedio resin (Perfecta Chemie B.V. Goes, Holland) and cross-sections were cut using a Cuenca wool microtome (Ulloa Optico Oftalmología S.A. Madrid, Spain). The sections were mounted onto glass slides and examined with a Reichert Polivar 2 microscope using a 100 W low-voltage halogen lamp for illumination of dyed samples.

Results and discussion

Stability of Liposome Suspensions

The possible aggregation of liposome suspensions during dyeing was monitored by measuring the variations in the vesicle size distribution and polydispersity of these suspensions in the absence of wool samples, using a quasi-elastic light scattering method (19). The results obtained for PC concentrations ranging from 0.56 to 6.81 % o.w.f. (corresponding to 0.25-3.0 mmol/l) and constant dye concentration (1.0 % o.w.f.) are given in Figure 3-A and 3-B respectively. There was a initial fall in the size of vesicles (minimum obtained approx at 30 min of dyeing) followed by a small rise in the size of vesicles during the succesive dyeing steps. The polydispersity index (PI) defined as a measure of the width of the particle size distribution obtained from the "cumulative analysis" remained after treatments always lower than 0.15. indicating that the liposome suspensions showed a homogeneous size distribution during the dyeing process. The presence of increasing amounts of PC in liposomes enhanced slightly their stability with respect to the aggregation, reducing both the mean particle size values and the polydispersity indices during the overall process.

Dyeing Kinetics

We carried out kinetic studies on the dyebath exhaustion of the dye-liposomes systems on untreated wool samples at different PC/dye weight ratios. To this end, a series of treatments were carried out varying the PC concentration (from 0.56 to 6.81 % o.w.f., which corresponds to 0.25 -3.0 mmol/l), the dye concentration remaining constant (1.0 % o.w.f.). The results obtained are plotted in Figure 4-A. Using liposome suspensions the wool dyeing resulted in a inhibition of dyebath exhaustion with respect to that obtained in the dyeing in the absence of liposomes (•). Moreover, dyebath exhaustion increased as the PC concentration in bilayers increased, the maximum being reached for 3.97 % o.w.f. (95 %), which corresponds to 1.75 mmol/l. Increasing concentration of PC in bilayers resulted in a progressive fall in dyebath exhaustion.

A series of treatments were carried out varying the dye concentration (from 0.1 to 1.0 % o.w.f.), the PC concentration remaining constant (2.27 % o.w.f., which corresponds to 1.0 mmol/l), to determine the influence of dye concentration on the dyebath exhaustion of these systems. The results obtained are plotted in Figure 4-B. It may be seen that dyebath exhaustion increased as the dye concentration in liposomes increased, reaching the highest value (approx 94 %) for a dye concentration ranging between 0.5-0.6 % o.w.f. Increasing amounts of dye in liposomes also resulted in a decrease in the dyebath exhaustion in the relative range of PC and dye concentrations investigated. It is interesting to note that the maximum dyebath exhaustion was attained in both cases approx at the PC/dye weight ratio of 4. This means that a PC concentration four times higher (in weight) than that of dye was needed in the dye/liposome systems to obtain the maximum dyebath exhaustion in all cases.

Influence of PC/dye weight ratio in liposomes on the bonding of dye to wool fibres

In order to find out whether lipid bilayers as carriers of a weakly polar 1:2 metal complex dye caused changes to dye-fibre bonding forces after dyeing, extractions by pure ethanol (9) and ammonia solutions (18) were performed on samples dyed at different PC/dye weight ratios. The results corresponding to the adsorbed dye (dyebath exhaustion given in mg dye/g wool), the

extracted dye by pure ethanol and ammonia (mg dye/g wool) and bonded dye (percent values derived by eqn 1) for the application of liposomes at PC concentrations ranging from 0.56 to 6.81 % o.w.f. (which corresponds to 0.25-3.0 mmol/l) at a constant dye concentration (1.0 % o.w.f.) are indicated in Table I. As aforementioned, the PC concentrations have been given in this Table both in percentages o.w.f. and in mmol/l in order to facilitate correlation of these data. Furthermore, the results corresponding to the application of liposomes at constant PC concentration (2.27 % o.w.f., which corresponds to 1.0 mmol/l), varying the dye concentration from 0.1 to 1.0 % o.w.f. are also given in Table II.

In general terms, the amounts of extracted dye from samples dyed in the absence of liposomes (given in the first line on Table I and indicated as 0 % PC concentration) were higher that those extracted from samples dyed in the presence of these structures. Furthermore, the amounts of extracted dye using ammonia solutions were always higher that those extracted with pure ethanol, which showed very small values in all cases.

The amounts of extracted dye in both Tables were closely connected with the PC/dye weight ratios in liposomes. Thus, the higher the concentration of PC in bilayers the lower the amounts of extracted dye in both extractions (Table I) and the higher the concentration of dye in liposomes the higher the amounts of extracted dye in these extractions (Table II). It is noteworthy that the amounts of extracted dye versus PC concentration (Table I) were in all cases lower that those reported for similar extractions in wool samples dyed with milling acid dyes via unilamellar and multilamellar liposomes (9). These findings are consistent with the higher level of fixation in wool exhibited by the 1:2 metal complex dyes with respect to that reported for milling acid dyes (1).

The decreasing amounts of stripped dye with ammonia versus PC concentration of liposomes could be attributed to the increasing contribution of the nonpolar interaction in the dye-fibre bonds. This finding underlines the important role played by PC in the hydrophobic interactions between the dye and hydrophobic regions within the fibre, which, as aforementioned, are essential in the substantivity of this type of dye.

Tables I and II also show the percentages of dye bonded to the fibre (derived by eqn 1). The presence of increasing amounts of PC enhanced the percentages of total bonded dye until achieve a maximum percentage for the higher PC concentration (Table I). Likewise, Table II shows that a maximum in the percentage of bonded dye was achieved for a dye concentration ranging from 0.5 to 0.6 % o.w.f., despite the fact that increasing dye concentrations in bilayers increased the amount of extracted dye. This percent range corresponds approximately to the PC/dye weight ratio of 4.

Comparison of Figures 4-A and 4-B with Table II shows that, both the maximum dyebath exhaustion and the maximum percentage of bonded dye were attained at a PC/dye weight ratio in liposomes of about 4. These findings confirm the importance of this ratio both in the transport and adsorption of the dye on wool and in the subsequent bonding of dye into the fibre.

The wash fastness of dyed samples in the absence or the presence of liposomes was determined using the IWS TM 193 test method (corresponding to ISO 105:C06 (1978) and UNE 40-120-81). The samples having the higher percentages of bonded dye (Tables I and II) also showed better levels of fastness (4 or higher). Thus, in general, the higher the PC concentration in bilayers the better the colour fastness of dyed samples. In addition to that, the samples dyed at the PC/dye weight ratio of 4 (corresponding to the maximum dyebath exhaustion, diffusion and bonding into the fibres) for each PC concentration investigated also showed the best level of fastness. The samples that showed the best wash fastness also attained a good level of xenon arc fastness (6 or higher)(ISO/R 105 (V), Part 2a, corresponding to UNE 40-187-73). The wash fastness and xenon arc fastness levels of dyed samples are also indicated in Tables I and II.

In order to abtain additional information about the distribution and diffusion of dye into the fibre optical microscopy observations were carried out of samples dyed in the absence or in the presence of liposomes using different PC concentrations at a different PC/dye weight ratios. These observations revealed that the use of these bilayer structures resulted in more regular distribution of dye on the surface of wool fibres. The presence of increasing amounts of PC in liposomes increased the regularity in the distribution of dye on the surface of fibres. In addition to that, the use of liposomes at the PC/dye weight ratio of 4 resulted in the best regularity results. The analysis of cross-sections of fibres also showed that the presence of increasing concentrations of PC in bilayers slightly rose the diffusion of dye into the fibre as well as the regularity in the distribution of dye on the wool surface. In a parallel way, the use of liposomes at PC/dye weight ratio of 4 also resulted in the best diffusion of dye into the fibre. As a consequence, these findings confirm the suitability of this specific weight ratio in the improvenent of the diffusion of dye into the wool fibre.

The main contribution of this paper is to extend the application of liposomes in wool dyeing to the 1:2 metal complex dyes, widely used in the textile industrie, using a lipid concentration similar to that used for conventional additives (ammonium sulphate, glauber's salts, albegal, etc). This method modulates the dyebath exhaustion on untreated wool samples and improves both the dye-fibre bonding forces, in particular at the PC/dye weight ratio of about 4, also improving the dye distribution on the wool surface and its diffusion into the fibres. Under a practical viewpoint a CRAFT project has been undertaken by different european textile industries specialized in wool tecnology in order to apply this tecnological innovation at industrial level (20).

Conclusions

An alternative method of wool dyeing using the weakly polar 1:2 metal complex dye (C.I. Acid Yellow 129) via LUV liposomes in presence of increasing amounts of PC and for different PC/dye weight ratios was suitable for modulation of dyebath exhaustion improving the dye-fibre bonding forces, the dye distribution on wool surface and the diffusion into wool fibres.

Unilamellar liposomes containing the C.I. Acid Yellow 129 dye were stable to the aggregation during the dyeing process at pH 5.5. This stability was directly dependent on the PC concentration in bilayers.

The transport and exhaustion of dyebath on untreated wool, the diffusion of dye into the wool fibres and the percentage of total bonded dye were also dependent on the PC concentration and on the PC/dye ratio in bilayers, the maximum being achieved at the weight ratio of both components of about 4. This effect may be considered as an important factor in modulating the dyeing kinetics and improving the regularity of dye distribution on wool surface and its intercellular diffusion into the fibres, bearing in mind the wide application of the 1:2 metal complex dyes in the textile industrie.

Acknowledgements

This work was supported by founds from D.G.I.C.Y.T. (Dirección General de Investigación Científica y Técnica)(Proyecto Nº PB94-0043), Spain and CRAFT Project BE-ST 2213 CEE. Authors acknowledge the expert technical assistance of Mr. G. Knorring.