Thermochimica Acta 613 (2015) 71–76

Contents lists available at ScienceDirect

Thermochimica Acta

journal homepage: www.elsevier.com/locate/tca

The assembly of dialkyldimethylammonium bromide cationic lipids as vesicles or monolayers in presence of poly(ethylene glycol)

Renata D. Adati, Eloi Feitosa*

Department of Physics, Sao Paulo State University, Sao Jose do Rio Preto, SP, Brazil

ARTICLE INFO

Article history:
Received 20 January 2015
Received in revised form 26 May 2015
Accepted 28 May 2015
Available online 1 June 2015

Keywords:
Differential scanning calorimetry
Dialkyldimethylammonium bromide salts
Poly(ethylene glycol)
Langmuir films
Polymer–surfactant interaction

ABSTRACT

The interaction between polymer and lipid vesicles may result in stabilization and formation of highly ordered networks, which can mimic biological membranes and be used as drug delivery system. Here we used differential scanning calorimetry (DSC) and Langmuir–Blodgett (LB) techniques to describe the effect of the nonionic water-soluble poly(ethylene glycol), PEG 35 kDa, on vesicles or monolayers from the cationic dialkyldimethylammonium bromide, D_nDAB (n = 12–18). Based on DSC data, up to 10 wt%, PEG plays minor role on the thermal behavior of D_nDAB, thus preserving the bilayer structure. Above 10 wt% PEG, there is no bilayer for n = 12–16, while for n = 18 there remains bilayer structures even in presence of 30 wt% PEG. The effect of PEG on the Langmuir monolayer of $D_{18}DAB$ depends on the amount of PEG in the sub-phase. In presence of up to ca 1 wt%, PEG yields more compressible films, while at higher polymer concentration the film is more extended and the collapse pressure is lower, most probably due to lipid solubility by the polymer solution. The PEG–D_nDAB complexes have potential application in controlled drug release by microgel nanoparticles.

1. Introduction

The self-assembly of synthetic cationic lipids from quaternary ammonium salts, such as dialkyldimethylammonium bromide (D_nDAB, n = 12, 14, 16, 18), into bilayer (*e.g.*, vesicles) or monolayer structures, are promising colloidal systems for industrial applications, with the advantage that they are low cost compounds [1,2]. One striking characteristics of these lipids is their melting temperature (T_m), which delineates the gel-to-liquid crystalline (LC) transition, which increases with the lipid chain length [1]. The reverse transition usually occurs at a lower temperature (T'_m), thus characterizing a thermal hysteresis the extent of which being characteristic of the lipids. The thermal behavior of D_nDAB bilayers [1], together with their monolayer characteristics [3], can be used to monitor the PEG–lipid interaction either in solution or at surface.

Cationic lipid vesicles are promising systems for drug delivery studies, especially after the pioneer work of Felgner et al. on the application of cationic lipid vesicles in nucleic acid delivery [4]. The interaction of cationic lipids with the anionic DNA or siRNA for lipoplex formation and transfection has been investigated [5,6].

In spite of optimizing the DNA cell transfection, polymer–lipid conjugates are widely used in the field of drug delivery to provide a polymer coat to confer favorable characteristics to the cell transfection. Poly(ethylene glycol), PEG, is a neutral polymer soluble in water as well as in some non-polar solvents, such as tetrahydrofuran, chloroform, dimethylsulfoxide or methanol [7]. The solubility in water makes PEG highly suitable for use in countless different applications in chemical, cosmetic and pharmaceutical industries [8].

Besides, PEG has been shown to stabilize the lamellar phase of lipoplexes, determining the efficiency in cell transfection [9,10]. Reports are available concerning preparation methods and physical stability of organized systems from didodecyldimethylammonium bromide ($D_{12}DAB$) and dioctadecyldimethylammonium bromide ($D_{18}DAB$) [11–13], while the characterization of bilayer structures from ditetradecyldimethylammonium bromide ($D_{14}DAB$) and dihexadecyldimethylammonium bromide ($D_{16}DAB$) has received minor attention [1,2].

Herein we focus on the interaction between neutral polymer and vesicle forming cationic lipids and describe the structural, thermal and kinetic behaviors of D_nDAB vesicles (n = 12–18) in the presence of PEG 35 kDa. The weak interaction between these compounds allows formation of vesicles in gel solution of PEG with potential use in controlled drug delivery.

* Corresponding author at: Department of Physics, São Paulo State University, Rua Cristovao Colombo, 2265, 15.054-000 Sao Jose do Rio Preto, SP, Brazil. Tel.: +55 17 3221 2238.
E-mail address: eloi@ibilce.unesp.br (E. Feitosa).

http://dx.doi.org/10.1016/j.tca.2015.05.022
0040-6031/

2. Experimental

2.1. Materials and methods

High purity >98% D_nDAB lipids were used: $D_{12}DAB$ (462.65 g mol^{-1}), $D_{14}DAB$ (518.74 g mol^{-1}), $D_{16}DAB$ (574.87 g mol^{-1}), and $D_{18}DAB$ (630.95 g mol^{-1}) were supplied by Sigma–Aldrich (St. Louis, USA) and used without further purification. Poly(ethylene glycol) 35 kDa (PEG) was purchased from Fluka (Buchs, Switzerland). Ultra-pure Milli-Q® water quality (resistivity 18.2 MΩ cm) was used in sample preparation and in the Langmuir film experiments.

2.2. Vesicle preparation

Dispersions were obtained from D_nDAB at 5.0 mM prepared in water in the absence or in presence of up to 30 wt% PEG. Mixtures of D_nDAB/water or D_nDAB/PEG solution were gently stirred magnetically for 1 h at a temperature safely above the melting temperature (T_m) from the lipids [1]. Accordingly, $D_{12}DAB$ dispersions were prepared at 25 °C, $D_{14}DAB$ and $D_{16}DAB$ at 35 °C, and $D_{18}DAB$ at 60 °C.

2.3. DSC measurements

A VP-DSC Microcalorimeter (Microcal Inc., Northampton, MA, USA) was used to collect data and the Origin® 7.0 software (supplied by the manufacturer) was used to record and to analyze the data. The experiments were performed by heating and cooling the sample and reference at the desired scan rate (60 or 20 °C/h, respectively for the shorter or the longest lipid chain length) in the temperature range of 1–65 °C. The thermograms were recorded as change in heat capacity at constant pressure, as a function of temperature. The transition enthalpies, ΔH(kJ mol^{-1}), were calculated from the area under the peaks, and the temperature at the peak maximum was taken as the transition temperature. The shape of the curve gives additional information about the phase transition and the width at half-height ($\Delta T_{1/2}$) is related to the cooperatively of the transition, which is higher the narrower is the peak. The DSC scans were obtained in duplicate to check reproducibility. More details about the experimental setup can be found elsewhere [11].

2.4. Langmuir films

The surface pressure–area (π–A) isotherms of the Langmuir films were obtained using a KSV-NIMA Langmuir balance. The monolayers were formed by spreading 50 μL of a 0.30 mg mL^{-1} $D_{18}DAB$ chloroform solution on the air/water or air/aqueous solution of up to 10 wt% PEG in the sub-phase. The solvent was allowed to evaporate for 10 min prior to sweeping the surface with the movable barriers. The π–A isotherms were recorded using a barrier speed of 10 mm min^{-1}. All experiments were carried out at room temperature (25 ± 1 °C).

3. Results and discussion

3.1. $D_{12}DAB$/PEG

Fig. 1 shows DSC thermograms for 5 mM (0.23 wt%) $D_{12}DAB$ dispersions in the absence and presence of up to 30 wt% PEG obtained in the heating mode using a 12 h pre-scan time. This rather long pre-scan time was used because of the slow (LC-to-gel) cooling transition [14–16]. The gel-to-LC transition temperature of $D_{12}DAB$ is well defined giving a very sharp single endotherm around $T_m \approx 16.0$ °C, with peak width $\Delta T_{1/2} < 1$ °C, in the DSC trace,

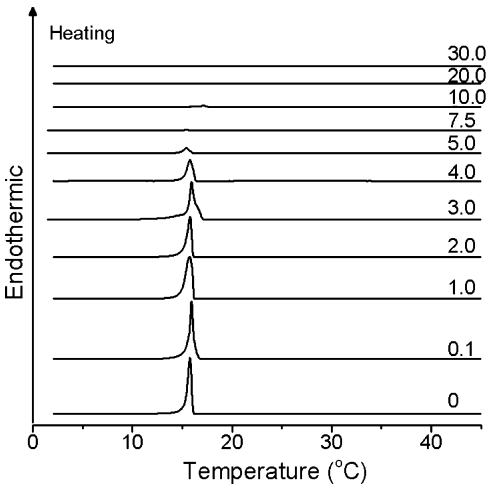

Fig. 1. Heating DSC thermograms for $D_{12}DAB$ aqueous dispersions at 5.0 mM in the absence and presence of up to 30 wt% PEG. Input DSC parameters: pre-scan time 12 h and scan rate 20 °C/h.

indicating high cooperativity of this transition. The reverse LC-to-gel transition, however, has not been detected directly by DSC (or any other technique) because of its very slow kinetics even though it has been detected indirectly as a broad exotherm around $T_m \approx 9.5$ °C [16]. In water $D_{12}DAB$ thus displays a thermal hysteresis of 6.5 °C.

By adding PEG, T_m tends to decrease indicating $D_{12}DAB$-PEG interaction, leaving the $D_{12}DAB$ bilayer more fluid. In presence of up to 4 wt% PEG, the T_m value does not change appreciably ($T_m \approx 16$ °C), but it decreases slightly to a minimum of 13.3 °C when PEG concentration attains 5.0 wt% (Fig. 2).

The enthalpy change (ΔH_m) related to the gel-to-LC transition decreases steeply from 54 to 1.5 kJ mol^{-1} when PEG concentration was raised from 4.5 to 7.5 wt% (Fig. 2). Above this concentration there is no vesicle. We ascribe the decrease in enthalpy to the reduction of the relative amount of vesicles. The lipids sequestered from the vesicles are probably complexing with PEG molecules despite the reported weak affinity of PEG to micelle-forming cationic surfactants [17]. The structure determination of these complexes is beyond the scope of the present work which focus on the vesicle structure in presence of PEG.

3.2. $D_{14}DAB$/PEG

Vesicles from $D_{14}DAB$ (together with $D_{16}DAB$) are the less investigated from the D_nDAB series most probably because of their low stability in water. The reason these vesicles are more unstable

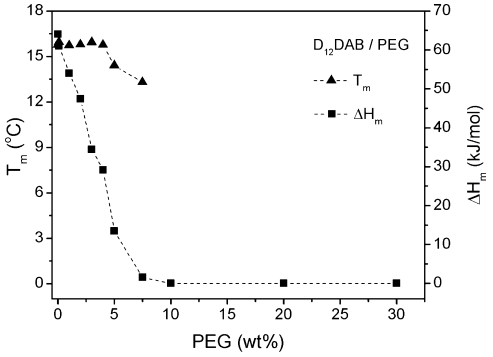

Fig. 2. Effect of PEG concentration on the melting temperature (T_m) and the enthalpy (ΔH_m) of $D_{12}DAB$ at 5 mM.