Colloids and Surfaces A: Physicochem. Eng. Aspects 415 (2012) 461–467

Contents lists available at SciVerse ScienceDirect

Colloids and Surfaces A: Physicochemical and Engineering Aspects

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

Unique concentration dependence on the fusion of anionic liposomes induced by polyethyleneimine

Kazuma Yasuhara*, Manami Tsukamoto, Yasutaka Tsuji, Jun-ichi Kikuchi*

Graduate School of Materials Science, Nara Institute of Science and Technology, 8916-5 Takayama, Ikoma, Nara 630-0192, Japan

ARTICLE INFO

Article history:
Received 3 October 2011
Received in revised form 12 January 2012
Accepted 13 January 2012
Available online 24 January 2012

Keywords:
Membrane fusion
Lipid vesicle
Cationic polymer
Lipid mixing
FRET

ABSTRACT

We investigated the interaction of branched polyethyleneimine (PEI) with anionic vesicles formed with 1,2-dioleoyl-*sn*-glycero-3-phospho-L-serine (DOPS) by spectroscopic measurements and microscopic observations. PEI induced the fusion of the vesicles only over a specific concentration range, which was found to be below the transition of vesicular zeta-potential from negative to positive value. Above the concentration window of membrane fusion, charge inversion of DOPS vesicles due to the further adsorption of PEI onto the surface of vesicle, inhibited the membrane fusion events. The PEI with low molecular weight induced the membrane fusion in a wider range of concentration than high molecular weight PEIs. Our results suggest that the mechanism of PEI interaction with the lipids depends on the molecular weight and stoichiometry of the polycation to negatively charged lipids.

1. Introduction

Currently, a variety of cationic polymers have been designed and synthesized for the development of functional materials. In particular, various biological and medicinal applications of cationic polymers have been developed, including non-viral gene carriers [1,2] and disinfectants [3,4]. Polyethyleneimine (PEI) is a classic polycation with dense amino groups on the main chain of the polymer under physiological conditions [5]. Many biological applications of PEI have been suggested such as cell adhesion [6,7] and gene transfection [8,9]. However, the cytotoxicity of PEI significantly limits its practical use [10,11]. Some classes of cationic polymers including PEI are believed to disrupt cell membranes through strong interaction and are therefore toxic [12].

In general, most cell membranes display a negatively charged surface due to the presence of acidic lipids in contrast to the fact that cationic membranes are rarely found in nature. For example, the membrane of *Escherichia coli* is enriched in phosphatidylglycerol [13] whereas the membranes of erythrocytes include phosphatidylserine as part of the inner leaflet [14]. Thus, it is reasonable to assume that the membrane interacting activity of cationic polymers partly relies on the electrostatic interaction between the polymer and the negatively charged surface of the membrane. Liposomes or phospholipid vesicles are excellent model systems that mimic cell membranes for physicochemical studies examining the interaction of membrane-active agents with lipid bilayers [15]. Previously, the action mechanisms of several cationic polymers or polypeptides have been investigated using a liposomal system. Such polycations were found to display various effects on the structure or property of lipid membranes including the flip-flop rates of lipids [16,17], membrane fusion [18–21], domain formation in the lipid membrane [22–24] and pore formation [25,26]. The interaction of polymers or polypeptides with the lipid membranes listed above consequently results in various biological functions of the polycations. Thus, a physicochemical study using liposomes should provide valuable information that describes the interaction mechanism of biologically active polymers with lipid membranes.

In this report, the interaction of branched PEI with negatively charged vesicles was investigated from a physicochemical viewpoint. An anionic lipid, 1,2-dioleoyl-*sn*-glycero-3-phospho-L-serine (DOPS), was used for the preparation of the vesicles as model cell membranes. The following series of techniques were combined to clarify the action mechanism of the PEI on a lipid membrane: dynamic light scattering (DLS), a lipid mixing assay by fluorescence measurements and zeta-potential measurements. Additionally, cryogenic transmission electron microscopy (cryo-TEM) was used for the direct visualization of vesicular morphology. Finally, we discuss the unique effect of the concentration and molecular weight of PEIs on the interaction mode with DOPS membranes. This discussion provides insight into the mechanism of action of PEIs on biomembranes.

* Corresponding authors. Tel.: +81 743 72 6093; fax: +81 743 72 6099.
E-mail addresses: yasuhara@ms.naist.jp (K. Yasuhara), jkikuchi@ms.naist.jp (J.-i. Kikuchi).

0927-7757/$ – see front matter © 2012 Elsevier B.V. All rights reserved.
doi:10.1016/j.colsurfa.2012.01.024