Phase separation in binary mixtures of lactosylceramide and DPPC .

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Introduction

We have been investigated the relation between phase separation in binary mixtures of ganglioside GM3 and dipalmitoylphosphatidylcholine(DPPC) and physiological properties of GM3 in biomembrane. In this report we focused on binary mixtures of lactosylceramide(LacCer) and DPPC. LacCer is widely distributed in epithelial tissues and it may be the receptor for disseminated disease such as certain kinds of cryptococcus and candida [1] and also involves in human aortic smooth muscle cell proliferation [2].

As shown in Fig.1, the difference between LacCer and GM3 is whether acetyl neuraminic acid is present or not in oligosaccharide portion. Thus, it is expected that x-ray diffraction study of LacCer/DPPC system gives some information about the role of neuraminic acid in the mechanism of phase separation in GM3/DPPC system.

Materials and Methods

LacCer was prepared from equine red blood cells. DPPC was purchased from Avanti Polar Lipids, Inc. (Birmingham, AL). Mixtures of LacCer and DPPC dissolved in chloroform-methanol were dried under vacuum and then were hydrated with phophate buffer at 55 ^{o}C to prepare multilamellar vesicles.

X-ray diffraction measurements were carried out at BL-15A. The diffraction patterns were detected by imaging plates (Type BAS-III, Fuji Photo Film Co., Ltd., Japan).

Results

Fig. 2 displays x-ray diffraction profiles of DPPC containing different molar ratio of LacCer at 50 ^{o}C. These samples were in the liquid crystalline phase (the L_α phase) because measurements of x-ray diffraction profiles during temperature scanning have exhibited that the main transition temperature was located near 41 ^{o}C. The peaks from 0.14- 0.16 nm^{-1} and those from 0.28-0.32 nm^{-1} were the 1st and the 2nd order diffraction peaks due to the lamellar repeat distance, respectively.

In 0.5 and 1.0 mol% LacCer content, coexistence of two kinds of lamellar structures was observed exhibiting phase separateion. This coexistence was observed in 0.5-1.7 mol% LacCer content (data not shown). Above 2.0 mol% LacCer content, the phase separation did not occur. As increasing LacCer content, the diffraction peak shifted to the wider-angle region due to the decrease of lamellar repeat distance.

In DPPC containing GM3 prepared from equine red blood cells, the phase separation was observed in 4-7 mol% GM3 content. Thus lack of acetyl neuraminic acid in oligosaccharide portion affects the property of phase separation.

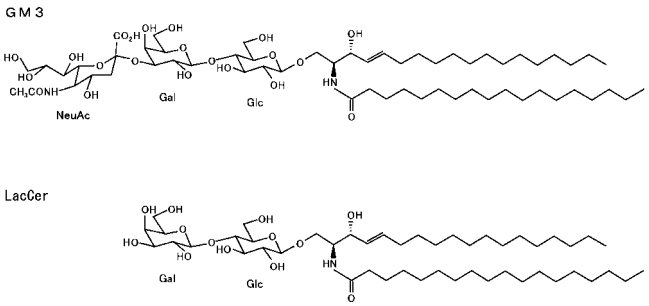
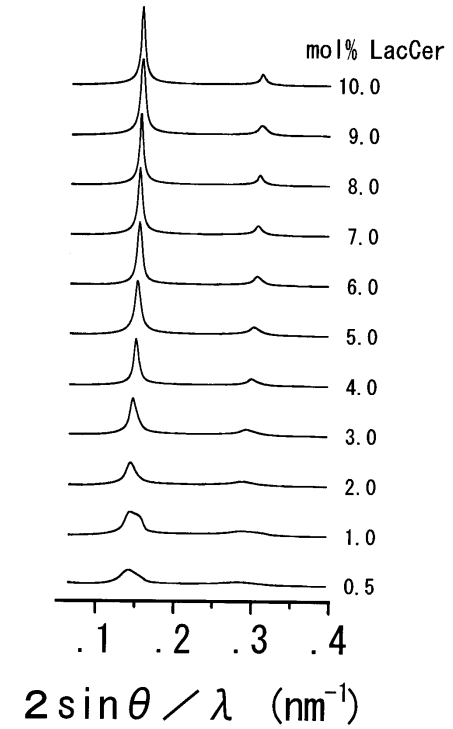


Fig. 1 Structures of ganglioside GM3 and lactosylceramide(LacCer)

Fig. 2 X-ray diffraction profiles of multilamellar vesicles of DPPC containing different molar ratio of LacCer in the L_α phase.



References

[1] V. Jimenez-Lucho et al., Infection & Immunity. 58, 2085 (1990).
[2] A.K. Bhunia et al., J. Biol. Chem. 272, 15642 (1997)

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