Difference of acyl chain length of GM3 affects phase behaviors of binary mixtures of GM3 and DPPC

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Introduction
We have been reported phase behavior of binary mixtures of 1,2-dipalmitoyl-L-phosphatidylcholine (DPPC) and native ganglioside GM3 prepared from equine red blood cells. In this report, we observed phase behaviors of binary mixtures of DPPC and GM3 with a C18:1 sphingoid base and a 24:0 acyl chain (GM3(18,24)) and those of DPPC and GM3 with a 18:0 acyl chain (GM3(18,18)) by x-ray diffraction (see Fig.1). The former GM3 is one of the major components of equine GM3. The latter has chain length similar to that of DPPC and is comparably minor component in equine GM3. By comparing phase behaviors of these two system, we investigated the effect of acyl chain length of GM3 on phase behaviors in binary mixtures of DPPC and GM3.

Materials and Methods
Ganglioside GM3(18,24) and GM3(18,18) was purchased from Wako Pure Chemical Industries, Ltd. (Osaka, Japan). DPPC was purchased from Avanti Polar Lipids, Inc. (Birmingham, AL). Mixtures of GM3 and DPPC dissolved in chloroform-methanol were dried under vacuum and then were hydrated with phosphate buffer at 55 °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
X-ray diffraction profiles of binary mixtures of GM3(18,24) and DPPC exhibited coexistence of two lamellar structure from 3 mol% to 7 mol% as shown in Fig.2a. This indicates that the formation of GM3-rich phase took place above 3 mol% and coexisted with GM3-poor phase from 3 mol% to 7 mol% and became dominant above 10 mol%. In GM3(18,18) / DPPC system, only the single lamellar structures were seemingly observed at least below 18mol% GM3 content (see Fig. 2b). However, lamellar repeat distance as a function of GM3(18,18) content (Fig.3) showed biphasic indicating formation of another phase which will be the GM3-rich phase. If this is the case, GM3-rich phase became dominant above 6 mol% GM3(18,18), which was different from the case of GM3(18,24).

The formation of GM3-rich phase will relate to biological functions such as cell adhesion and reactivity of antibody. It may be possible that cells regulate biological functions by varying acyl chain component of GM3 in the plasma membranes.

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