Physico-chemical characteristics of triacyl lipid A partial structure OM-174 in relation to biological activity

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The triacylated lipid A partial structure OM-174 was characterized in detail using a variety of physical and biological techniques.The temperature T_c of the gel to liquid-crystalline phase transition of the hydrocarbon chains lies at 0 °C and, with that, considerably lower than for hexaacyl lipid A (Fig. 1, top), from which a high fluidity of the acyl chains at 37 °C can be deduced. From the X-ray diffraction spectra (Fig. 1, bottom), exhibiting a periodicity of approximately 8.78 nm and three further reflections at 8.78/ $\sqrt{3}$, 8.78/$\sqrt{4}$, and 8.78/ $\sqrt{7}$, it can be concluded that OM-174 aggregates assume, under near physiological conditions, the micellar H_I structure. In this structure, the lipid molecules are packed with their backbone on a cylindrical surface with the acyl chains directed inward. The cylinders themselves are hexagonally packed. From the occurrence of the $\sqrt{7}$-reflection, the existence of a cubic phase can be excluded. Theoretically, also the inverted hexagonal structure H_{II} would match the indexing. However, from considerations of the molecular geometry as well as from diffraction patterns of the H_{II}-structure observed for lipid A [1], which display a completely different intensity ratio of the reflections, this possibility can be excluded.

Conformational analyses of the behaviour of the various functional groups of OM-174 as compared to hexaacyl lipid A suggest an altered hydration of the phosphate charges and unusually strong hydration of the ester groups, which is probably connected with the high accessibility of these groups to water in the micellar aggregate structure. With fluorescence resonance energy transfer spectroscopy [2], OM-174 was shown to intercalate into a phospholipid membrane corresponding to the macrophage membrane within seconds in the presence and within minutes to hours in the absence of lipopolysaccharide-binding protein (LBP). In the limulus amebocyte lysate (LAL) assay, the triacyl lipid A is more than 10^5-fold less active than hexaacyl lipid A, but only 10-fold less active in inducing interleukin-6 in human mononuclear cells, and equally active in inducing nitric oxide in murine macrophages. These findings allow an interpretation of the mechanism of the lipid A-induced cell activation.

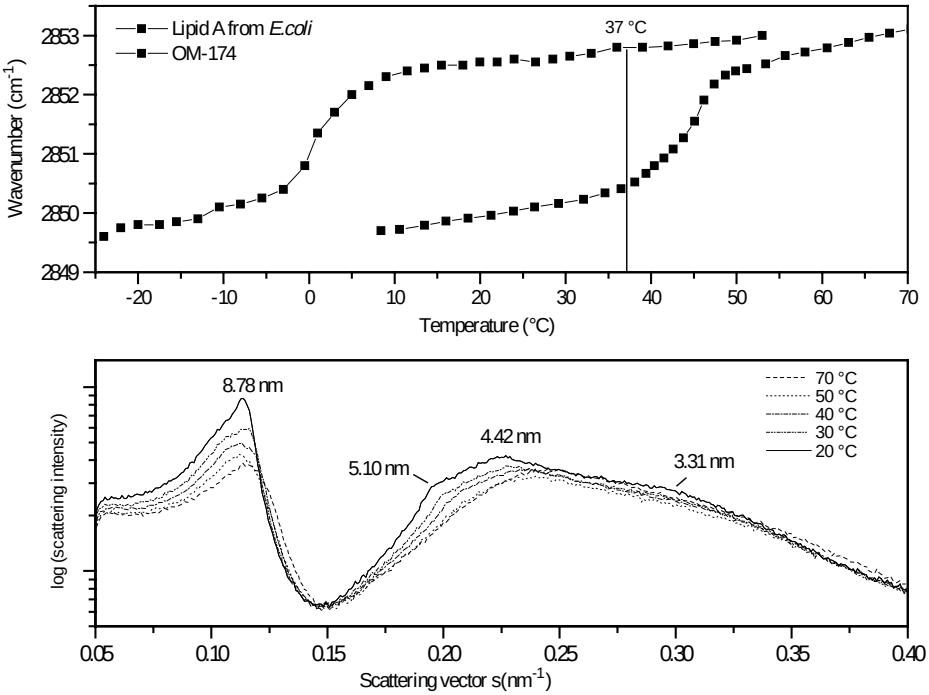

Figure 1: Peak position of the symmetric stretching vibration of the methylene groups versus temperature of OM-174 as compared with hexaacyl lipid A (top) and

Small-angle X.ray diffraction patterns of OM-174 at 85 % water content between 20 and 70 °C (bottom)

References

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