O-acetylation is high, calcium binding with the Vi is mainly electrostatic, similar to the binding of sodium. As the degree of *O*-acetylation is lowered, calcium binds to the Vi as a chelating agent, different from monovalent counter ions. Circular dichroism measurement showed conformational difference between K^+ and Ca^{++} when the degree of *O*-acetylation is low. The results are consistent with our proposed structure of Vi, based on molecular modeling, that the surface location of the bulky *O*-acetyl groups blocks the accessibility of carboxyls and explains the immunodominant role of the *O*-acetyl groups.

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S12.23

Expression of α2→6Sialyltransferase and CDw75 in Human Myeloid Cell Line During Differentiation Into Monocytic Lineage

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α2→6sialyltransferase is expressed in B cells and suggested to be involved in B cell-B cell interaction. In addition, α2→6sialyltransferase is required for CDw75 production in B cells. Among hematopoietic cells and cell lines, only mature B cells have been thought to express CDw75. Further, expression of α2→6sialyltransferase messages, activities and its products has not been fully analyzed. Here we present the systematic analyses in human lymphoid and myeloid cell lines. Moreover, the results showed that not only B cell lines but also myeloid cell lines during differentiation into monocytic lineage expressed α2→6sialyltransferase and CDw75-positive glycoprotein.

Among B cell lineage, only mature B cell lines expressed high levels of α2→6sialyltransferase messages, activities, and cell surface epitope, Neuα2→6Galβ1→4GlcNAc. In addition, only mature B cell lines exhibited cell surface CDw75 and 63 kDa CDw75-positive glycoprotein. Pre-B and myeloma cell lines did not have such characteristics. By contrast, myeloid cell lines showed little activities and messages of α2→6sialyltransferase and expression of CDw75 and CDw75-positive glycoprotein. In monocytic differentiation, however, α2→6sialyltransferase activities and messages were up-regulated and CDw75-positive glycoprotein appeared in a time-dependent manner. Furthermore, CDw75-positive cells were observed by immunofluorescence staining.

These results suggest that CDw75-positive glycoprotein plays some role not only in lymphoid cells but also in myeloid cells during differentiation.

S12.24

Embrional Albumin-Like Protein is Glycosylated

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Human embrional glycoprotein (GP66) was isolated from fetal serum. Serum was treated with 0.0125 M NH_4OAc, pH 6.2, the extract chromatographed on DEAE-Toyo-Pearl 650S column and subjected to SDS-EF in 12% PAAG followed by transfer to Immobilon PVDF membrane and immunoblot with rabbit antibodies to fetal serum proteins. Two immunoreactive proteins, GP72 (α-fetoprotein, AFP) and GP66 were found by immunoblotting. Only GP72 reacted with mAbs to AFP.

GP66 isolated by chromatography on Toyo-Pearl HW-55F and Nucleosil-300μC_4 showed 4 isoforms, pI from 4.8 to 5.1 in IEF. Amino acid sequence released the same 35 *N*-terminal amino acid residues as in HSA [1], whereas fluorescence and UV-adsorption spectra of GP66 and HSA were different showing the presence of 3-5 Trp residues in GP66 but absence of Trp in HSA. Besides, GP66 was glycosylated. Monosaccharide composition determined by HPLC of aminomethylcoumarine derivatives [2], was the following: Gal/GlcNAc/Man/GalNAc/Neu5Ac/Fuc 4:5:3:0.8:1:0.2.

Inhibitory activity of GP66, neuraminidase desialylated protein and HSA towards tumor necrose factor (TNF) cytotoxic activity was determined. Only GP66 inhibited cytotoxic activity of TNF as was shown with L929 cell line. Detailed study of structure and immunomodulating activity of this albumin-like glycoprotein is in progress.

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S12.25

Immunochemical Studies on Peptidorhamnomannan of *Sporothrix schenckii* Yeast Forms

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Previous studies showed that the rhamnomannans isolated by alkaline extraction from *S. schenckii* cultures contain single-unit α-L-rhamnopyranosyl and (1-2)-linked di-α-L-rhamnopyranosyl side chains attached to 3 on a (1-6)-linked α-D-mannan, and are the main antigenic determinant in such molecules (1).

Recently (2), we have isolated and chemically characterized *O*-linked neutral tri-saccharide: Rha(α1-3)Man(α1-2)Man; and acidic tetra and pentasaccharide: Rha(α1-4)GlcA(α1-2)Man(α1-2)Man; Rha(α1-4) [Rha(α1-2)]GlcA(α1-2)Man(α1-2)Man, released from yeast forms of *S. schenckii* peptidorhamnomannan by mild alkaline borohydride treatment (β-elimination). We verified the antigenic involvement of these molecules using antiserum raised in rabbits by injecting acetone-dried *S. schenckii* cells. The ability of the purified