as mechanical obstruction and surfactant displacement by free fatty acids have been suggested. Recently the Human Lung Surfactant Protein SP-A has been characterized as one of several C-type lectins, which in the presence of Ca^{2+}-ions bind specificially to certain carbohydrates. To test our hypothesis of specific lectin-carbohydrate interactions as part of the pathogenesis in meconium aspiration syndrome we set up to test the binding of native human SP-A towards meconium glycosphingolipids.

Our data are in general agreement with recently published data (Childs, RA *et al.* (1992) *J. Biol. Chem.*, 267, 9972 – 9979) and show a specific and Ca^{2+}-related binding to two neutral glycosphingolipids present in meconium extract from all individuals tested; Galactosylceramide and Lactosylceramide. As was noted before, when using the thin layer chromatogram binding technique, the binding of SP-A to lactosylceramide species was strongest to those containing α-hydroxy fatty acids and phytosphingosine. In order to optimize a microtiter well binding assay and possibly reveal some factors of biological significance we have used a reduced factorial design of experimental approach. This includes in all 34 variables testing 69 different buffers with various concentrations of $CaCl_2$, NaCl, EDTA, pH, bovine serum albumin, NaN_3 and Tween 20 at different steps of the assay as well as various concentrations, incubation times and incubation temperatures of blocking reagent, native SP-A, biotinylated anti SP-A antibody, ALP-coupled avidin and PNPP substrate. The use of this experimental design will be discussed.

S15.14

Enhanced Binding of Enterotoxigenic *Escherichia coli* K99 to a Receptor Analogue Obtained by Chemical Modification of NeuGc-GM_3

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The K99-fimbriated *E. coli* can cause severe diarrhoea in some young domestic animals in which the natural receptor is the ganglioside NeuGc-GM_3 (NeuGcα3Galβ4GlcβCer)[1]. Significant anti-adhesive effects have been obtained by treatment of infected calves with oligosaccharides released from glycoproteins of bovine plasma[2]. in an attempt to reveal the detailed structural requirements of the K99 adhesin we have modified NeuGc-GM_3 chemically. Five different sialic acid C(1)-amides were produced in addition to the C(1)-methylester and the C(1)-primary alcohol. The products were purified and their structures analysed by negative-FAB mass spectrometry. Bacterial binding to the products was investigated by incubation of ^{35}S-labelled bacteria with thin layer plates on which the glycolipids were chromatographed. Modification of the carboxyl group to C(1)-amide enhances the binding between the bacterial adhesin and the glycolipid at least fivefold. Some strengthening of the binding is also obtained with the analogue methylamide as well as the reduced compound, C(1)-alcohol. Ethylamide binds with about the same strength as the native NeuGcGM_3, while propylamide and benzylamide show decreased binding. The C(1)-methylester of NeuGcGM_3 was found not to be stable enough to withstand overlay experiments on thin layer plates where water is present, and reverted to the acid.

1) Teneberg *el al.*, 1990, *FEBS Lett.*, **263**, 10 – 14, and references therein.
2) Mouricout *et al.*, 1991, *Infect. Immun.*, **58**, 98 – 106.

S15.15

Structural Changes in the Sugar Chains of Serum IgG in HTLV-I Transgenic Mice

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Rheumatoid arthritis (RA) is considered to be an autoimmune disease which produces autoantibodies directed to the Fc portion of serum IgG. Interestingly, patients with RA have an abnormally high percentage of IgG which lack terminal galactose in its *N*-linked sugar chains (1). This galactose deficiency in RA is limited to the sugar chains of IgG molecules. However, the etiology and pathogenesis of this disease are poorly understood. Generally, it is believed that either exogenous infectious agent and/or endogenous factors, such as connective-tissue proteins and altered immunoglobulins, are the initiating inflammation which leads to the development of this complex disease. Although various etiologic agents including viruses, bacteria, and mycoplasma have been suggested as causes of RA in humans, no conclusive data has been obtained. Therefore, establishment of an animal model of RA will be useful to understand the cause and the pathogenesis of this disease. Recently, Iwakura *et al.* reported (2) that HTLV-1-transgenic mice develop chronic arthritis similar to RA, and suggested that HTLV-1 may be one of the etiologic agents of chronic arthritis in humans. Accordingly, it will be of interest to determine whether less galactosylation occurs in the IgG of HTLV-1-transgenic mice or not. IgGs were highly purified from the sera of HTLV-1 transgenic and nontransgenic mice. Comparative studies of the sugar chains released by hydrazinolysis revealed that their structures of transgenic IgG are quite different from those of nontransgenic IgG. Although both IgGs contained biantennary complex-type oligosaccharides, transgenic IgG had less galactosylated forms (45%) than those from nontransgenic IgG (28%), just as was found in patients with RA. These transgenic mice should provide a useful model to investigate the relationship between the galactosylation of IgG and the development of RA.

(1) Parekh, R. B. *et al. Nature,* **316**, 452, 1985. (2) Iwakura, Y. *et al. Science,* **253**, 1026, 1992.

S15.16

Expression of (Sialyl)-Lewis X Groups on Human α1-Acid Glycoprotein in Acute and Chronic Inflammation

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The plasma levels and the glycosylation of several serum glycoproteins are subject to marked changes during acute and chronic inflammation. This phenomenon is the result of cytokine-induced alterations in the hepatic synthesis of these so-called acute-phase glycoproteins. We now report on a new