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S12.19

Isolation and Characterization of a Glycoprotein Allergen from *Aspergillus fumigatus*

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An important allergen was extracted from the mould *Aspergillus fumigatus* and purified by anion and cation exchange chromatography and gel filtration. The allergen, when examined by SDS-PAGE/immunoblotting, was found to be essentially homogeneous with a molecular weight of ca. 20.000 daltons. The allergen contained 3% protein and 90% carbohydrate. The carbohydrate moiety was composed of mannose, galactose and glucose in the ratio 10:7:0,9. The carbohydrate-protein linkage of the allergen is currently studied. Furthermore, we study how modifications of the molecule affect its allergenic activity.

S12.20

Fucose in α 1-3 Linkage to the N-Glycan Core Forms an Allergenic Epitope that Occurs in Plant and Insect Glycoproteins

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I.) 33 of 46 sera from honeybee venom allergic patients contained IgE reacting with the N-linked carbohydrate moiety of bee-venom phospholipase A2 (PLA). With most sera, this reaction could be inhibited with 1 μ M glycopeptide from pineapple bromelain. Since bromelain glycopeptides devoid of fucose did not bind to IgE-antibody, we conclude that α 1-3 fucosylation of the innermost N-acetylglucosamine-residue (GlcNAc) forms the IgE-reactive determinant.

...-Man α 1-6

Man β 1-4GlcNAc β 1-4GlcNAc

...-Man α 1-3

Fuc α 1-3

II.) This structural element does not only occur in glycoproteins from plants but also from insects as could be shown for honeybee phospholipase A2 and for the membrane-glycoproteins of three insect cell lines. Significant amounts of α 1-3 fucosylated glycans were found in the glycoproteins of Bm-N cells (*Bombyx mori*), predominantly as di-fucosylated structures, i.e. fucoses linked both α 1-3 and α 1-6 to the asparagine-bound GlcNAc-residue. Smaller amounts of these immunogenic structures were found in the cell lines Sf-9 (*Spodoptera frugiperda*) and Mb-0503 (*Mamestra brassicae*). This agrees with the observation that in the latter two cell lines the respective α 1-3 fucosyltransferase activity levels are at the limit of detectability.

S12.21

Antibody Against Sulfated Glycosphingolipids of Peripheral Nerve Myelins Detected in the Patients with Human Cytomegalovirus Infection

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Cytomegalovirus (CMV) infection is one of the frequent antecedent infection associated with autoimmune inflammatory demyelinating neuropathies (1). The proposed autoimmune mechanisms of these diseases involve molecular mimicry or shared viral and self-epitopes. As for CMV, no reports have so far been published about shared antigens with peripheral nervous system (PNS) myelin. In this paper we studied whether CMV infection could induce a production of antibody against PNS glycosphingolipids (GL). Sera from patients with congenital CMV infection were tested for IgM and IgG antibodies against acidic and neutral GL purified from human PNS (2). TLC-immunostaining assay revealed that the CMV infected patients' sera contained antibodies against sulfatide and sulfolucuronyl glycosphingolipids (SGGL), which were PNS myelin associated GL. No reactivity was observed to PNS gangliosides or neutral GL. The antibody also bound to other sulfated glycolipids including seminolipid and LacCer-sulfate, but not to cholesterol-sulfate, suggesting that sulfated sugar chain is required for the binding. Furthermore both anti-sulfatide and anti-SGGL antibodies were absorbed with sulfatide-conjugated octyl-Sepharose column or heparin-Sepharose column, whereas CMV specific IgG titer was not decreased by the absorption of anti-sulfated GL antibody. These results suggested that CMV infection might specifically induce a production of antibody against myelin associated sulfated GL, whereas these antibodies differed from CMV specific antibody.

(1) Winer *et al.*, *J. Neurol. Neurosurg. Psychiat.*, **51**, 613 (1988). (2) Ogawa-Goto *et al.*, *J. Neurochem.*, **55**, 1486 (1990).

S12.22

Structural Analysis of the VI Polysaccharide at Various Degrees of O-Acetylation

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Vi polysaccharide is a linear homopolymer composed of (1 \rightarrow 4) α D-GalpANAc variably O-acetylated at C-3 position. We reported that the immunogenicity of Vi depends mainly on its O-acetyl. In contrast, the carboxyls played a minor role in the immunogenicity of the Vi. The binding of the Vi at various degrees of O-acetylation with different counter ions are studied by potentiometric titration, circular dichroism, and reaction with hydrazine derivatives. We found that the O-acetyl residues hindered the association of carboxyls by chelating ions, such as calcium and magnesium. The difference in the free energy of binding between potassium and calcium (ΔG^{CaK}) was calculated as a function of O-acetylation. We found that ΔG^{CaK} decreases as the degree of O-acetylation increases indicating that when the degree of