

type of inflammation-induced change in glycosylation for one of these proteins, α -acid glycoprotein (AGP). This regards the induction on AGP of NeuAc α 2 \rightarrow 3Gal β 1 \rightarrow 4(Fuc α 1 \rightarrow 3)-GlcNAc-R, the determinant for bloodgroup SLe^x (sialyl-Lewis X). Variations in the degree of α 1 \rightarrow 3-fucosylation (Lewis X) were detected by changes in the affinity of AGP towards *Aleuria aurantia* lectin (AAL) in crossed affinoimmuno-electrophoresis (CAIE) of patient sera. Five forms of AGP could be distinguished differing in reactivity towards AAL (A0: non-reactive with AAL, A1-A4: reactive with AAL). A strong transient increase in the serum levels of the forms A3 and A4 was detected after laparotomy, severe burning and primary *sectio caesarea*. A constitutive elevated level of these two forms was detected in sera of patients suffering from rheumatoid arthritis or mammary tumours relative to control sera. The acute-phase-induced increase in fucosylation of AGP, at least partly represented an increased expression of sialyl-Lewis-X determinants on AGP. This was revealed by the staining of AGP with the monoclonal antibody CSLEX-1 on Western blots, after isolation of the various AAL-reactive fractions of AGP from patient sera by immunoaffinity chromatography and fractionation by preparative CAIE with AAL. A direct correlation between the expression of sialyl-Lewis-X groups on AGP, the reactivity with AAL and the degree of fucosylation was established. Because the interaction between leukocytes and inflamed endothelium is mediated via sialylated Lewis-X structures on leukocytes and the endothelial selectin ELAM-1, we postulate that the inflammation-induced increase in the AAL-reactive fractions of AGP represent a physiological feed-back response of the inflammatory reaction.

S15.17

A New Isolation Procedure of Differently Fucosylated Forms of α 1-Acid Glycoprotein by Preparative Crossed-Affinity Electrophoresis

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In acute and chronic inflammation human α 1-acid glycoprotein (AGP) is subject to marked changes in its biantennary glycan content. Recently we have described a new type of inflammation-induced change in glycosylation: an increase in the degree of α 1 \rightarrow 3-fucosylation, leading to an elevated expression of Sialyl Lewis-X structures on AGP [1]. This was detected by the affinity of AGP to the fucose-specific *Aleuria aurantia* lectin (AAL) in crossed affinoimmuno-electrophoresis (CAIE). Five forms of AGP could be distinguished differing in their AAL reactivity (A0: non reactive with AAL, A1-A4: reactive with AAL). Especially the forms A3 and A4 of AGP were transiently increased during acute inflammatory reactions.

To be able to perform biological studies with, and to study the oligosaccharide structures occurring on each of the molecular forms of AGP, these forms have to be isolated from each other in sufficient amounts from human sera. A procedure was developed making use of immunoaffinity chromatography followed by preparative CAIE with the lectines AAL and Con A. A 5-mm thick lectin-containing

agarose gel was used from which the fractions were collected electrophoretically in a DEAE-Sephacel-containing slot of the gel. A small intermediate gel containing sugar was used to dissociate lectin-glycoprotein complexes. The DEAE-Sephacel was replaced at appropriate times and subsequently transferred to a small column. The AGP-forms were recovered from the DEAE-Sephacel column by elution with a NaCl-containing buffer. In this way AGP could be fractionated in high yields in different glycosylated forms with respect to their degree of α 1-3 fucosylation and their biantennary glycan content.

1. De Graaf, T. W., Van der Stelt, M. E., Anbergen, M. G., Van Dijk, W., *J. Exp. Med.* 177, in press (March 1993).

S15.18

A Unique Glycosylation Pattern in Placental Vessels?

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Endothelial carbohydrates (CH) are probably involved in various endothelial functions. The placenta is a highly vascular tissue with angiogenesis and is an excellent model for the study of vascular functions. The purpose of this work was to study the expression of various CH by means of monoclonal antibodies in placental vessels and to compare with CH on maternal vessels (placental bed). Biopsies from 10 normal human placentas at term and their corresponding placental beds were studied immunohistochemically. The endothelium of the fetal placental vessels expressed various CH in a pattern not previously reported for endothelia. Expression of CH on the uteroplacental vessels was also very different from other myometrial (maternal) vessels. For example, the uteroplacental vessels strongly expressed the sialyl-Le^x oligosaccharide which is involved in various adhesion phenomena. These apparently unique expressions of CH in placental vessels compared to maternal tissue suggest an important functional role for these structures in the placenta.

S15.19

Protein Glycation and its Metabolic Consequences

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The exposure of proteins to glucose, leads over time to nonenzymatic binding of the sugar to reactive amino groups located on lysine side chain and N-terminal amino acid residues of proteins, forming early glycosylation products. In a complex series of chemical rearrangements these products become irreversible AGEs (advanced glycosylation end-products). The accumulation of AGEs in proteins is accelerated in aging and in people with diabetes (1). A macrophage-monocyte receptor system for AGE moieties mediate the uptake of AGE-modified proteins with concomitant induction of growth factors and cytokines (TNF, IL-1). These cytokines are able to activate the hepatic acute phase response. It was found previously (2), that AGEs may modulate via cytokines liver protein synthesis, contributing to the changes observed in serum protein level in diabetes and in