as being very malignant, increased fucose content was detected even when the overall tumour burden was small. The presence of breast cancer did not change the concentration of the other sugars. It is concluded that the binding of Hp to lotus lectin in cancer is due to increased fucose content. This change could be caused by cytokines that are released during tumour growth affecting the activity of fucosyl transferases in the liver, or by direct release of fucosyl transferases from the tumour.

S15.23

A Reliable Anti-Ganglioside Antibody Assay, Capable of Monitoring IgM Anti-GM1 Antibodies

E. O. Bech and T. F. *Ørntoft*
Dept Clin Chemistry, Aarhus University Hosp., DK.

Circulating IgM anti-bodies against ganglioside GM1 occur in some neurological diseases, especially in patients having motor neuron involvement. The lack of a reliable assay to monitor these antibodies have yielded conflicting results, as well as reduced the clinical value of the test. In order to improve the clinical value of anti-GM1 antibodies we have developed an ELISA assay, using a high titer serum as calibrator, and Cholera toxin (B subunit) and a high titer patient serum as Controls.

The validation of the assay has shown the following:

Regression on the Standard curve is linear and results are calculated using $f(x) = aX + C$ obtaining slope (a) and intersept (C) from Sigmaplot. This optimised and standardised assay enables us to reproduce data with a between-day variation coefficient (cv) ranging from 5.5% to 16% (N = 14). Intra assay cv are 4% – 6% within the detection range, (N = 32). Sensitivity is high, detecting anti-GM1 antibodies in normals at dilution 1:500, hereby reducing possible matrix effect. Recovery, estimated by mixing high and low titer sera is 93% – 98%. The assay is reliable and can be used for longitudinal monitoring of IgM anti-GM1 antibodies in sera and CSF.

In patients with Guillian-Barré-Syndrome the serum titer corresponds very well with disease intensity. In patients with lung cancer, paraneuroplastic neurological syndromes can be detected with the present assay.

S15.24

Monoclonal Antibody Defined Carbohydrate Structures as Circulating Tumor Markers for Cancer of Rectum and Sigmoid

T. F. Ørntoft and N. Langkilde
Dep. of Clinical Chemistry, Aarhus Kommunehospital, Dk 8000 Aarhus C, Denmark; and Anil Singhal, Biomembrane Inst., Seattle, WA, USA.

The structural glycosylation is known to aberrate dramatically in tumor tissue compared to normal tissue, leading to production of tumor associated carbohydrate structures that are shed into the circulation. On preoperatively collected serum samples from 182 patients with carcinomas of the rectum and sigmoid, the carbohydrate structures Le^x, Sialosyl-Le^x, Sialosyl-Le^a, and Sialosyl-Tn were quantitated by ELISA, and related to survival. The patient follow-up time was >5 years. The patient distribution on Dukes' classification was: A: 12; B: 59; C: 95; D: 16. A conservative estimate of % cancer death distributed on Dukes classification showed: A: 0%; B: 38%; C: 60%; D: 100%. Kaplan-Meyer plots were constructed for the total population and for the Dukes C group for the first five years of observation. The 90% percentile value (calculated on 132 serum samples from Dukes A patients) of each carbohydrate structure was used as cut-point. Preoperative S-concentrations above the cut-points were significantly associated with death (Log rank test) in case of Le^x ($p<0.0017$) and Sialosyl-Le^x ($p<0.0068$), and highly significant in the case of Sialosyl-Le^a ($p<0.0001$), and Sialosyl-Tn ($p<0.0009$). Dukes C patients could be divided into two groups with low and high survival rate by both Sialosyl-Le^a ($p<0.003$) and Sialosyl-Tn ($p<0.009$). There was no correlation between serum level of the various carbohydrate epitopes, and between these and CEA. The S-level of these carbohydrate structures may form a rational basis for selection and intensity of therapy in colorectal carcinomas.

S15.25

Immunoassays for Determining the Concentration of a Carcinoma-Specific Antigen in the Blood of Cancer Patients

J. F. Codington, S. Haavik, Z. Hui, S. Matson, Z. Wu and X. Chen
Boston Biomedical Research Institute; Department of Biological Chemistry and Molecular Biology, Harvard Medical School, Boston, Mass., USA; and Department of Pharmacognosy, Institute of Pharmacy, University of Oslo, Oslo, Norway.

Immunoassays have been developed for the quantitative determination of a human carcinoma-specific antigen (HCA) in the blood of patients with epithelial-derived cancers. These assays were made possible by the finding that the HC antigen shares a common epitope with the mouse mucin-type glycoprotein, epiglycanin, present at the surface of the mammary carcinoma ascites cell, TA3-Ha, and present in the blood of mice bearing this tumor in ascites form. The assays have utilized both polyclonal antibodies prepared in the rabbit, as well as monoclonal antibodies prepared from the B cells of immunized mice. Sandwich assays employing monoclonal and polyclonal antibodies, as well as lectins, have been developed. In addition, competitive binding assays have been used. These include radioimmunoassays in which the antigen is radioactively labeled, and the inhibition of the formation of a radioactive antigen-antibody complex, when human sera are used, is a measure of the concentration of the HC antigen. Competitive binding assays have employed antigen immobilized on solid supports. The concentration of antibody bound to the antigen is determined by the use of a second antibody labeled with an enzyme such as alkaline phosphatase or horseradish peroxidase. A description of some of these procedures and the results obtained will be presented. (This work was supported by Epigen, Inc., Wellesley, MA, USA.)

S15.26

Quantitation of Polysaccharide in Haemophilus Type B Conjugate Vaccines by High-Performance Anion-Exchange Chromatography with Pulsed Amperometric Detection