GLYCOPROCESSING IN CLASSICAL GALACTOSAEMIA

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Thesis submitted for the degree of
Doctor of Medicine
in
The University of Adelaide
(Faculty of Medicine)

April 1997

Table of Contents

Abstract

Classical galactosaemia is a disorder of galactose metabolism that results from a deficiency of galactose-1-phosphate uridyltransferase. Infants with galactosaemia usually present with an acute toxicity syndrome that is characterised by vomiting, failure to thrive, hepatitis, jaundice, and sepsis. The acute symptoms are resolved by removing galactose from the diet and this treatment has formed the basis of management of galactosaemia for over 60 years. However, as a group, the galactosaemic patients develop longer term problems that include impaired intellectual development, learning disabilities, poor motor function, ovarian failure, and occasionally ataxia and tremor. These long-term complications develop independent of either the severity of initial illness, the age at which diet was started, or the success of dietary control.

If the long-term complications are not related to the acute toxicity of galactosaemia, then other explanations must be sought in alterations of the cellular environment that occur secondary to transferase deficiency. The serum in untreated galactosaemia contains hyposialylated isoforms of several glycoproteins. This suggests a possible disturbance in glycoprotein synthesis, which could contribute to the long-term complications. The aim of this thesis was to examine N-glycosylation in transferase-deficient skin fibroblasts to determine what abnormalities of glycoprotein synthesis occur in galactosaemia. When the mature complex N-linked oligosaccharides from galactosaemic fibroblasts were examined by size-exclusion chromatography and anion-exchange high performance liquid chromatography, they were structurally complete. However, the galactosaemic fibroblasts consistently incorporated less [2-^{3}H]-mannose into protein than normal controls. This occurred whether the incorporation was corrected to the cell protein or to the incorporation of [^{35}S]-methionine. The decrease was observed in cellular and secreted proteins and was proportional to the concentration of galactose in the culture medium. The galactosaemic fibroblasts also incorporated less [2-^{3}H]-mannose into dolichol-linked oligosaccharides. This was accompanied by an accumulation of $Man_{3-5}GlcNAc_2$ dolichol-linked intermediates, and predominantly $Man_5GlcNAc_2$-sized oligosaccharides were transferred to protein. These abnormalities are very similar to those reported in fibroblasts from patients with the carbohydrate-deficient glycoprotein syndrome type I, and in cells starved of glucose.

It is proposed that galactose-1-phosphate interferes with the conversion of glucose to mannose intermediates that are required for the synthesis of dolichol-linked oligosaccharides. This may be one mechanism for hyposialylation of serum glycoproteins in untreated galactosaemia. It remains to be determined whether abnormal N-glycosylation contributes to the long-term complications in patients.