J Inherit Metab Dis (2007) 30:818
DOI 10.1007/s10545-007-0461-x

SHORT REPORT

Combination of enzyme analysis, allele-specific PCR and sequencing to detect mutations in the *GALT* gene

F. R. O. Calderon · L. Nelson · P. Dobrowolski · I. Sinitsyna · A. Phansalkar · N. Longo · M. Pasquali · R. Mao

Received: 29 August 2006 / Submitted in revised form: 5 June 2007 / Accepted: 8 June 2007 / Published online: 17 September 2007

Online citation: JIMD Short Report #084 (2007) Online

Summary Newborn screening can identify patients with classical galactosaemia, and their diagnosis needs to be confirmed with assay of the activity of galactose-1-phosphate uridyltransferase (GALT). Unfortunately, in many cases the results can be ambiguous and further testing is required. Here we report a combination of biochemical analysis of GALT enzyme activity and mutation analysis of the most common mutations in the corresponding gene. Samples (n=243) submitted for confirmatory testing for classical galactosaemia were analysed simultaneously for GALT enzyme activity and allele-specific PCR/fragment analysis for seven mutations and two polymorphisms in the *GALT* gene (mutations IVS2–2A>G, p.S135L, p.T138M, p.L195P, p.K285N, p.Q188R, p.Y209C; polymorphisms p.N314D, p.L218L). Mutation detection accorded with biochemical analysis in 93% of samples. Subsequently, a total of 34 samples with either discordant results between the above methods or low enzyme activity were fully sequenced, identifying previously reported pathogenic mutations and seven novel variations (p.P185H, p.R201C, p.E220K, p.R223S, p.I278N, p.L289F and p.L218X) in the *GALT* gene. This approach further increased concordance between genetic and biochemical analysis to 99% of all alleles tested. Our results indicate that DNA testing can help to verify biochemical enzymatic data and improve distinction of borderline enzyme activities where a patient may still benefit from treatment.

Communicating editor: Gerard Berry

F. R. O. Calderon · L. Nelson · A. Phansalkar · N. Longo · M. Pasquali · R. Mao
ARUP Institute for Clinical and Experimental Pathology, Salt Lake City, UT, USA

P. Dobrowolski · I. Sinitsyna · N. Longo · M. Pasquali · R. Mao (✉)
ARUP Laboratories, 500 Chipeta Way, Salt Lake City, UT 84108-1221, USA
e-mail: rong.mao@aruplab.com

N. Longo · M. Pasquali · R. Mao
Department of Pathology, University of Utah Health Sciences Center, Salt Lake City, UT, USA

Electronic Supplementary Material
The online version of this article (doi:10.1007/s10545-007-0461-x) contains supplementary material, which is available to authorized users.