ERNDIM QC scheme for CDG screening

Nijmegen, 01-2012

Please find enclosed the results of the QC scheme 2011 for CDG screening. The current scheme included 57 participants from many countries around the world. We have applied a scoring of 2 points per sample for the method & technical interpretation of the results (category C) and 2 points per sample for the suggestions for further diagnostics (category D). In total, a maximum of 24 points could be scored.

One of the main concerns for future rounds of this QC scheme will be the access to patient material (about 2.5 ml plasma/serum per patient), especially since we increased the number of samples to eight and increased the quantity to 50 micro liters for laboratories using HPLC in 2012. We would like to ask you if you could provide material for future rounds of this QC scheme. Please, send samples (~2.5 ml) to my address below, including information about age, sex, and a brief clinical description on first visit of the patient.

In case of any questions, please do not hesitate to contact us.

With kind regards, also on behalf of Cas Weykamp,

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ERNDIM QC scheme for CDG screening 2011

General comments

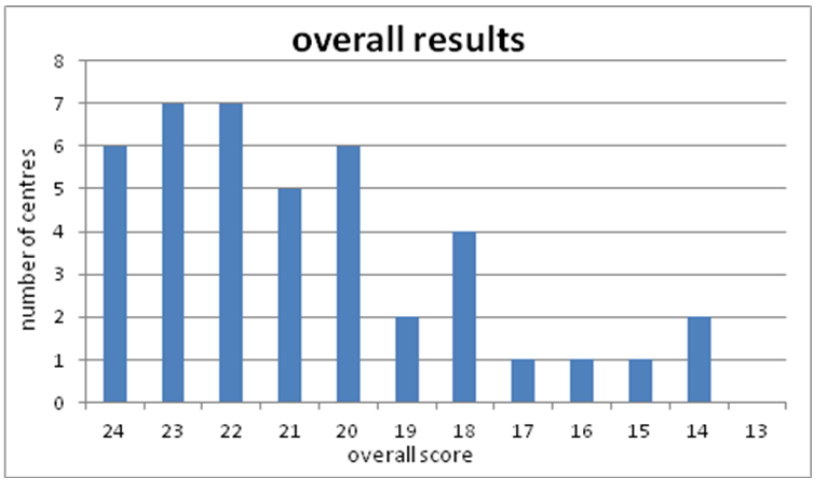
We have received 42 report forms, 15 centres were unable to respond (~75% response). In total 342 plasma/serum samples were shipped after lyophilisation in the presence of a cryoprotectant. We had to resend a few samples due to shipment or handling problems. No sample degradation was reported, and no interference was reported of the lyophilisation procedure in any of the methods used. Isofocusing was employed most often (28), followed by HPLC (7), CE (5), and Mass spectrometry (2).

Results

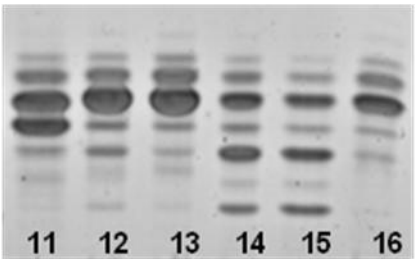
In this round of the QC scheme, we have been unable to arrange true patient samples for all cases. For sample 012, we had to "mimic" a mild type I profile by mixing sufficient quantities of a control and a more severe type I case. The clinical information is from a mild CDG type I patient that presents with the same abnormality as present in sample 012. For all other cases, the clinical information was as described on the first patient request form that we obtained.

Sample	Clinical information (sex, age, phenotype)	Diagnosis
CDG 011	F, 60 yrs, elevated transaminases and CK, mild motor retardation	Protein polymorphism
CDG 012	F, 15 yrs, abnormal CT-scan, non-progressive ataxia	Mild PMM2-CDG (CDG-Ia)
CDG 013	M, 3 yrs, facial dysmorphism, mental and motor retardation, hypotonia	Control
CDG 014	F, 4 yrs, gastrointestinal problems, protein-losing enteropathy, coagulopathy	PMI-CDG (CDG-Ib)
CDG 015	F, 3 months, hypoglycemia, facial dysmorphism, extrapyramidal symptoms, vomiting, failure-to-thrive	PMM2-CDG (CDG-Ia)
CDG 016	M, 11 yrs, mental retardation, coagulopathy, wrinkled skin	Control

In the graph, the overall score is shown for all centres. In general, proper identification and assignment of the profile was correct for 92% of the responses, while a 80% score was obtained for the suggestions for further diagnostics. Below, we have summarized the results from all participants per sample.



Isofocusing profiles



ERNDIM CDG011
This patient is not suffering from a CDG. When using methods like isoelectric focusing or CE, an increase of trisialotransferrin was observed, more or less in equal quantities as tetrasialotransferrin. In most cases, this has been recognized as a possible polymorphism in the transferrin protein. Indeed, incubation of the sample with neuraminidase showed two bands, which is in agreement with a protein polymorphism. Alternative ways to exclude a polymorphism include analysis of parental samples. This polymorphism did not disturb interpretation of HPLC or mass spec profiles.

ERNDIM CDG012
A mild abnormality can be found in this sample with a slightly elevated disialotransferrin fraction. Secondary causes for a CDG-I profile should be considered (like alcohol abuse). A clinical phenotype of ataxia in combination with mild (or even normal) type I CDG screening result should be seen as indication for PMM2-CDG. This has been reported for several patients with (mild) CDG-Ia (PMM2-CDG). Subsequent enzyme analysis (phosphomannomutase) and molecular genetic confirmation (*PMM2* gene) leads to the diagnosis of PMM2-CDG.
Identification of this type of mild abnormalities can be quite challenging; 12 centres report a normal result, randomly divided over the different methods. For HPLC, the limited amount of material (in patient diagnostics mostly 100-200 microliter is requested) could complicate the identification of mild abnormalities. A critical look at the isofocusing results shows that the abnormality is visible, however, possibly overlooked as a possible diagnosis of CDG-I. In only few cases, the method itself was insufficient to observe an abnormality.

ERNDIMCDG013
A normal profile was identified by almost all centres.

ERNDIMCDG014
A clear CDG type I profile was obtained in this patient. A polymorphism was excluded (profile is not very suggestive), the clinical phenotype is not highly suggestive of either galactosemia or fructosemia as secondary causes of CDG type I, however, is highly suggestive for CDG-Ib (PMI-CDG). Especially in view of the possible treatment options, suggestions for further diagnostics should include direct enzyme analysis of phosphomannose isomerase.
Almost all centres correctly assigned this as an abnormal profile corresponding to a CDG type I and most centres suggested appropriate work-up for reaching the final diagnosis. However, not all centres suggested CDG-Ib on basis of the clinical symptoms.

ERNDIMCDG015
A clear CDG type I profile was observed in this patient. A polymorphism was excluded (profile is not very suggestive) and the clinical phenotype is not highly suggestive of either galactosemia or fructosemia as secondary causes of CDG type I. Subsequent enzyme analysis (phosphomannomutase and phosphomannose isomerase) and subsequent molecular genetic analysis (*PMM2* gene) led to a diagnosis of CDG-Ia (PMM2-CDG).
Almost all centres reported an abnormal profile with the correct assignment as type I and the appropriate further diagnostics to arrive at a diagnosis CDG-Ia.

ERNDIMCDG016
A normal profile was identified by almost all centres.