ERNDIM QC pilot for CDG screening

Nijmegen, 01-10-2009

Dear colleagues,

Hereby, you will find the results of the QC scheme for CDG screening, for the first time run as a pilot under ERNDIM. After initial QC schemes in the Euroglycanet program with about 25 participants, the current scheme involves 46 participants, mainly from Europe, but also including 10 centres from other continents. In the coming month, a decision will be made whether CDG screening will be established as an official ERNDIM QC scheme.

For future rounds of this QC scheme, we will try to increase the volume to 40 microL for centres using CE/HPLC and to increase the number of samples to 6 per round. However, we have almost run out of sufficient numbers and amounts of plasma/serum samples of patients with abnormal CDG screening results and would urgently ask you to provide material for future rounds. Please, send samples (2-2.5 mL) to our institute, including information about age, sex, and a brief clinical description on first visit of the patient.

A scoring system has been applied for interpretation of the results (4 points for correct identification and assignment of the profile type, and 2 points for proper suggestions for further diagnostics). We tried to keep the scoring as objective as possible, although we realize that in some cases this remains difficult. In case of any questions, please do not hesitate to ask. The individual scores will be send to you by email.

With kind regards,

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ERNDIM QC scheme for CDG screening 2009

General comments

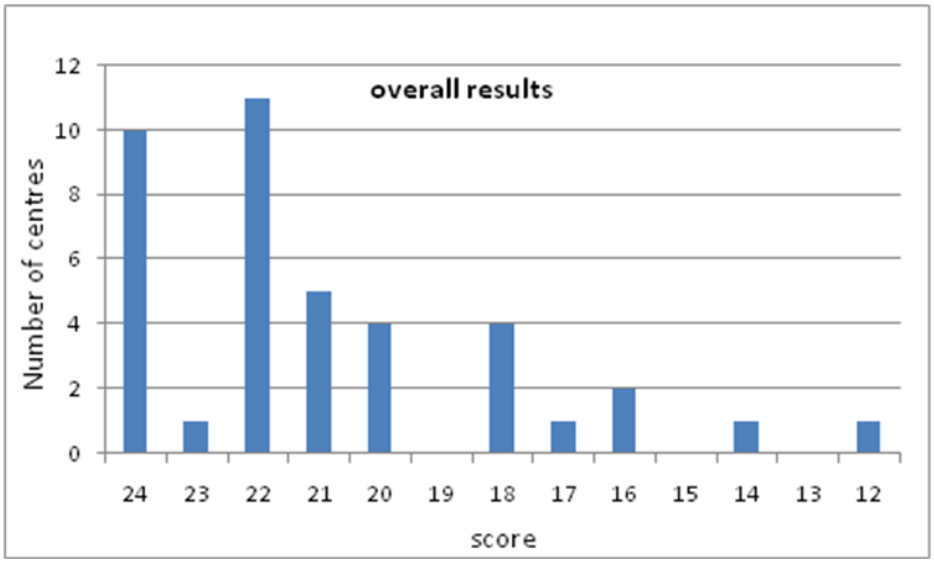
We have received 40 report forms, six centres were unable to respond. A total of 184 serum samples was shipped, lyophilised in the presence of a cryoprotectant. Five samples had to be resend due to problems with sample handling, no sample degradation was reported, and no interference was reported of the lyophilisation procedure in any of the methods used. Isofocusing was employed most often (27, mostly using Multiphor or Phast System), followed by CE/HPLC (11) and Western Blotting (1) or Mass spectrometry (1). The final results for the samples in this round were similar for the different methods used. In general, the quantity of the samples was sufficient for a proper analysis, although some of the centres (using CE/HPLC) indicated that a volume of 20 microL is lower than routinely used (about 100 microL).

Results

In this second round of the QC scheme we have send you the following samples, the clinical information was just as described on the first patient request form that we obtained:

	Clinical information	Patient data	Final diagnosis
ERNDIMCDG001	Protein-losing enteropathy, coagulation problems, epilepsy	F, 3 years	CDG-Ia
ERNDIMCDG002	Mental retardation, increased transaminases	F, 40 years	No known CDG
ERNDIMCDG003	Congenital myopathy, mental retardation	M, 20 years	Transferrin polymorphism
ERNDIMCDG004	Cerebellar ataxia	F, 22 years	CDG-Ia

In the graph, the overall score is shown for all centres. In general, proper identification and assignment of the profile was correct for 95% of the responses, while a 72% score was obtained for the suggestions for further diagnostics. Below, we have summarized the results from all participants per sample.



ERNDIMCDG001

After exclusion of a polymorphism and secondary causes of CDG type I, enzyme analysis (phosphomannomutase and phosphomannose isomerase) and subsequent molecular genetic analysis (*PMM2* gene) led to the diagnosis of CDG-Ia.

The average score for this sample was 5.1 (max 6). All centers correctly assigned this as an abnormal profile corresponding to a CDG type I and almost all centers suggested appropriate work-up for reaching the final diagnosis. Not every centre did suggest the exclusion of secondary causes. Although part of the clinical phenotype might suggest CDG-Ib, these features can be observed in other CDG-I subtypes as well.

ERNDIMCDG002

A normal profile was identified by almost all centres, leading to an average score of 5.75 (max 6).

ERNDIMCDG003

Isoelectric focusing of transferrin showed an abnormal profile with increased trisialotransferrin. The more or less equal levels of Transf-3 and Transf-4 should suggest a protein polymorphism. Incubation of the sample with neuraminidase (or analysis of parent samples) leads to identification of a protein polymorphism.

Most centres correctly identified an abnormal profile of type II CDG (or corresponding to a protein polymorphism). Some centres performed neuraminidase incubation on the sample. Not all centres suggested the proper methods to arrive at the correct diagnosis. Obviously, centres using Mass spectrometry or Western blotting of transferrin found a normal profile. Among the group using CE/HPLC, different profiles were reported, ranging from normal profiles to the presence of additional peaks at the pentasialo- or trisialotransferrin position. Average score for this sample: 5.4 (max 6).

ERNDIMCDG004

A relatively mild abnormality was found with increased disialotransferrin (some centres also reported increased asialotransferrin). Secondary causes for a CDG-I profile should be suggested (concerning age of patient at least alcohol abuse). Subsequent enzyme analysis (phosphomannomutase and phosphomannose isomerase) and subsequent molecular genetic analysis (*PMM2* gene) led to the diagnosis of CDG-Ia. A clinical phenotype of cerebellar ataxia in adult CDG-Ia patients has been reported by several groups, even with normal transferrin isofocusing profiles.

Almost all centres reported an abnormal profile, most of those also with the correct assignment as type I. Not all centres suggested the appropriate further diagnostics to arrive at a diagnosis CDG-Ia. Average score: 4.7 (max 6).