ERNDIM
Qualitative Organic acids urine Barcelona ANNUAL REPORT 2018

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1. Introduction

The ERNDIM Qualitative Organic Acids in urine scheme offers urine samples obtained from confirmed patients with confirmed diagnoses to enable laboratories to gain or maintain experience to identify organic acid disorders. The scheme is organised by Dr Judit García Villoria, Barcelona in conjunction with CSCQ, the Swiss organisation for quality assurance in medical laboratories.

As in previous years, samples were sent out to cover the spectrum of what is typically observed in the metabolic laboratory. A mix of clearly diagnostic profiles and some more challenging profiles were provided. As in previous years normal profiles were also sent out. The requirement to interpret a normal profile, as such, is as important as correctly identifying abnormal profiles. Correctly identifying a profile as normal can avoid unnecessary further investigation and distress to the patient and family.

2. Participants

In 2018 74 laboratories from many different countries participated in the QLOU Barcelona scheme. (3 laboratories were educational participants in 2018). They take part in all aspects of the scheme and receive interim reports with scores, but performance is not indicated on the ERNDIM certificate of performance.

The numbers of participants continues to increase and as a result a third organising centre, Barcelona, was added to the QLOU scheme for 2018 and participants were split between the three organising centres. New applicants will distributed between the Barcelona, Sheffield and Heidelberg qualitative urinary organic acid schemes which are run separately. The three organising laboratories each participate in the other's scheme by rotation.

3. Design of the scheme and logistics

As usual, the samples used in 2018 were authentic human urine samples, 6 from affected patients and 3 from a healthy individual. Samples were selected by the Scientific Advisor and tested for suitability in the Scientific Advisor's laboratory. In 2018 CSCQ dispatched the QLOU EQA samples to the scheme participants and provides a website for on-line submission of results and access to scheme reports. Existing QLOU, ACDB, DPT and Urine MPS scheme participants can log on to the CSCQ results submission website at: https://cscq.hcuge.ch/cscq/ERNDIM/Initial/Initial.php

To be able to continue this scheme we need a steady supply of new patient samples. Several laboratories have donated samples to the Urine MPS scheme in the past, for which they are gratefully acknowledged. If you have one or more samples available and are willing to donate these to the scheme, please contact us at admin@erndim.org.
Laboratories which donate samples that are used in the scheme are eligible for a 20% discount on their participation in the QLOU scheme in the following year.

Table 1. Samples included in the 2018 ERNDIM QLOU Barcelona scheme.

Survey, reporting deadline	Sample no.	Sample type
2018.A, 4th June 2018	QLOU-EB-2018-A	Glutaric aciduria type I low excretor
2018.B, 4th June 2018	QLOU-EB-2018-B	Normal profile
2018.C, 4th June 2018	QLOU-EB-2018-C	Phenylketonuria
2018.D, 30th July 2018	QLOU-EB-2018-D	Propionic acidemia
2018.E, 30th July 2018	QLOU-EB-2018-E	Normal profile
2018.F, 30th July 2018	QLOU-EB-2018-F	Valproate treatment
2018.G, 1st October 2018	QLOU-EB-2018-G	Multiple acyl-CoA dehydrogenase deficiency
2018.H, 1st October 2018	QLOU-EB-2018-H	Normal profile
2018.I, 1st October 2018	QLOU-EB-2018-I	Isovaleric acidemia

This was the first year in which Barcelona was used as an organising centre for the QLOU scheme. The scheme format was kept identical to those of previous years for the Sheffield and Heidelberg organising centres. Samples were shipped by courier. Details regarding stability of (reconstituted) samples are provided in the sample package.

Participants submitted results to the CSCQ website https://cscq.hcuge.ch/cscq/ERNDIM/Initial/Initial.php. The due dates for submitting results in 2018 were June 4th, July 30th and October 1st 2018. The website includes a section to specify methods. Method specification is required for correct evaluation of the quantitative results

In 2018 a total of 67 reports were received for survey 1 (samples 2018.A to 2018.C), 68 reports for survey 2 (samples 2018.D to 2018.F) and 71 reports for survey 3 (samples 2018.G to 2018.I). 65 labs submitted results for all three surveys. 1 participant did not submit any report, while 4 other participants submitted one of the three reports and 4 other participants submitted two of the three reports.

Evaluation of results was performed using Excel with the submitted results extracted from the database by the website manager.

4. Scoring of results

A scoring system was developed in 2012 and approved by the ERNDIM Scientific Advisory Board. Similar to other qualitative (proficiency testing) ERNDIM schemes, the maximum score for a sample is 4 points. Scores are allocated to different elements of the results reported (Table 3).
Qualitative results and diagnostic proficiency of the 2018 samples were scored using the criteria given in Table 4 and 5. These criteria have been set by the Scientific Advisor, approved by the Scientific Advisory Board. The final decision about scoring of the scheme is made in the Scientific Advisory Board (SAB) during the Autumn meeting (November 29th-30th, 2018 for the 2018 scheme).

A note on scoring of diagnostic proficiency and the use of check boxes and the comment box:

To indicate the most likely diagnosis check boxes must be used to facilitate evaluation of results. The use of the 'comments' box in the website form is recommended to explain your interpretation of results. Comments will be taken into account to score interpretation.

Table 2. General criteria used to score results

Satisfactory	4	Helpful but incomplete	3
Not helpful	2	Slightly misleading	1
Misleading	0		

Starting with the 2014 schemes the concept of 'critical error' is introduced to the assessment of the qualitative schemes. Labs failing to make a correct diagnosis of a sample considered eligible for this category will be deemed not to have reached a satisfactory performance even if their total points for the year is sufficient according to the requirement set by the SAB. The classification of samples to be judged for critical error was undertaken at the SAB meeting held on November 29th-30th, 2018.

Samples QLOU-EB-2018-B, QLOU-EB-2018-C, QLOU-EB-2018-G and QLOU-EB-2018-I were eligible for critical error. Amongst the reports of regular participants 4 critical errors were identified in 2018. Details are given under item 7 'Results of individual samples and evaluation of reporting'.

We are required to define "Participation" for the purpose of the ERNDIM Annual Certificate which covers all ERNDIM schemes. For this urinary organic acid scheme we have defined "Participation" as requiring at least two returns during the year. Failure to meet this requirement will result in the certificate of participation showing 'non-submitter' rather than 'satisfactory' or 'unsatisfactory'.

In 2018 the satisfactory performance score has changed from 61% to 70% which equates to 25/36 for 3 returns and 17/24 for two returns.

5. Communication of results

Interim reports with diagnoses, summaries of the results submitted and interim scores were made available August 23, 2018 (survey 2018-1), October 8, 2018 (survey 2018-2) and November 12, 2018 (survey 2018-3).

ERNDIM provides a single certificate for all its schemes with details of participation and performance.

Three Performance Support letters *were* sent for the 2018 surveys. *None* of these *three* participants have also received a performance support letter in *2017* or *2016*. Unsatisfactory performance (either due to overall score or due to critical error) within an EQA scheme for at least 2 out of 3 years that the participant has subscribed for will result in a notification letter of unsatisfactory performance to the quality manager or head of department.

6. Proficiency of the 2018 surveys

In 2018, 65 participants submitted 3 reports including *1* educational participant. From the *74* ordinary (non-educational) participants *65* (*87,8*%) achieved satisfactory performance (score ≥*25 for 3 returns or* ≥*17 for two returns*, no critical error). *9* participants did not accomplish satisfactory performance, including *5* due to incomplete submission of results (i.e. no report or 1 survey report submitted instead of 2 reports).
Overall proficiencies of each sample are depicted in Table 6.

Table 6. Overall proficiencies of the 2018 surveys.

Sample ID	Sample type	Proficiency (%)
QLOU-EB-2018-A	Glutaric aciduria type I low excretor – **Educational sample**	49%
QLOU-EB-2018-B	Normal profile	99%
QLOU-EB-2018-C	Phenylketonuria	99%
QLOU-EB-2018-D	Propionic acidemia	99%
QLOU-EB-2018-E	Normal profile	97%
QLOU-EB-2018-F	Valproate treatment	88%
QLOU-EB-2018-G	Multiple acyl-CoA dehydrogenase deficiency	77%
QLOU-EB-2018-H	Normal profile	100%
QLOU-EB-2018-I	Isovaleric acidemia	99%

7. Results of individual samples and evaluation of reporting

- **Sample QLOU-EB-2018-A:**

Patient details: 29 year-old male with spastic-dystonic tetraparesis.

Diagnosis: Glutaric aciduria type I low excretor.

Analytical details: The organic acid profile was the characteristic of low excretor patients showing only a slight increase of 3-hydroxyglutarate, glutarate was within the control values. 27/67 participants (43%) reported a slight increase of 3-hydroxyglutarate; 6 laboratories (9%) reported elevated excretion

of glutarate; 30 laboratories (45%) reported an increase of lactate, that could be slightly elevated in this disease; 7 laboratories (10%) reported an increase of 3-methylglutaconate.

Interpretation: The correct diagnosis was performed by 33/67 of the laboratories. The overall diagnostic performance was only 49%. 21 laboratories (31%) reported a normal profile. The remaining laboratories 13 (16%) established other diagnosis.
Some mutations in *GCDH* have been associated to low excretors. The characteristic profile was a slight increase of 3-hydroxyglutarate with normal or slightly increased excretion of glutarate (C. Busquets et al. Pediatr. Res 2000; 48:315-322), which is the biochemical phenotype of this patient.

SAB considered this sample as educational. Therefore the score obtained in the interim report of the first round is not taken into account.

- **Sample QLOU-EB-2018-B:**

Patient details: 42 year-old female with muscle weakness.

Diagnosis: Normal sample, obtained from a healthy adult.

Analytical details: No abnormalities were reported by 56/67 participants (84%); 11 laboratories found different elevated metabolites.

Interpretation: 66/67 laboratories reported a normal sample. The overall diagnostic performance was 99%. One laboratory reported the diagnosis of PKU, probably due to a cross between sample B and C, **and it was elected as critical error by SAB.**

- **Sample QLOU-EB-2018-C:**

Patient details: 48 year-old female with severe mental retardation and obesity**.**

Diagnosis: Phenylketonuria

Analytical details: 36/67 participants (54%) detected the 4 key metabolites: phenyl-lactate, phenyl-pyruvate, phenyl-acetate, 2-hydroxy-fenyl-acetate. The remaining laboratories only mentioned the increase of some of them. The metabolite most frequently detected was phenyl-lactate and the less frequently detected was phenypyruvate.

Interpretation: 66/67 laboratories (99%) reported the correct diagnosis. The remaining laboratory failure the diagnosis due to a presumable crossing between sample C and sample B, and **it was elected as a critical error by SAB.**

- **Sample QLOU-EB-2018-D:**

Patient details: 29 year-old female with severe intellectual deficit and recurrent epilepsy

Diagnosis: Propionic acidemia.

Analytical details: 65/68 participants (96%) reported elevated excretion of all the key metabolites: 3-hydroxypropionate, 2-methylcitrate and propionylglycine; 62 laboratories (93%) reported an increase of tiglylglycine, which is also characteristic of this disease. In addition, increased excretion of fumarate has been reported by 13 laboratories (19%).

Interpretation: The correct diagnosis was performed by 67/68 laboratories. The diagnostic performance was 99%. One laboratory established the diagnosis of multiple carboxylase deficiency.

- **Sample QLOU-EB-2018-E:**

Patient details: 37 year-old female with challenging behaviour.

Diagnosis: Normal sample, obtained from a healthy adult.

Analytical details: No abnormalities were reported by 46/68 participants (67%); 18 laboratories (26%) found elevations of different metabolites.

Interpretation: 60/68 participants reported correctly a normal sample. The overall diagnostic performance was 97%; 3 laboratories established other diagnosis (Hyperprolinemia type II, Partially treated Phenylketonuria, urea cycle defect).

- **Sample QLOU-EB-2018-F:**

Patient details: 5 year-old male with epilepsy.

Diagnosis: No abnormalities detected except for valproate metabolites.

Analytical details: 31/68 laboratories (46%) detected valproate metabolites. Some of them included the specific name of these metabolites; 2 laboratories identified drug metabolites or anticonvulsivant metabolites without specific mention to valproate theraphy.

Interpretation: 2 points were given both, for the diagnosis of valproate treatment (32/68 laboratories) or normal profile (28/68). The overall diagnostic performance was 88%. Other 5 laboratories reported other diagnosis (2-hydroxyglutaric aciduria, 2-ketoglutarate dehydrogenase, 3 methylglutaconic aciduria, Biotinidase deficiency, mitochondrial stress/dysfunction).

Although the detection of valproate metabolites is not relevant for the diagnosis, it is important to detect such metabolites as this drug interferes with the metabolism and alters the excretion of certain organic acids: 3- hydroxy-isovalerate, 2-methyl-3-hydroxy-butyrate, acylglycines and dicarboxylic acids, which can lead to diagnostic difficulties. The presence of valproate metabolites also generates extra peaks in the chromatogram that can interfere in the detection of other organic acids.

- **Sample QLOU-EB-2018-G:**

Patient details: 18 year-old female with mild cognitive retardation, decompensations for fasting and / or intercurrent infections

Diagnosis: Multiple acyl-CoA dehydrogenase deficiency.

Analytical details: Increased ethylmalonate and 2-hydroxyglutarate lactone were reported by 65/71 laboratories (93%); Isobutyrylglycine, isovalerylglycine, hexanoylglycine and suberylglycine were detected by less than 67 % of the laboratories. The increase of adipate, suberate, unsaturated suberate, sebacate and unsaturated sebacate.was detected by less than 29% of the participants.

Interpretation: Correct diagnosis was performed by 55/71 laboratories, the overall diagnostic performance was 77%; 6 laboratories (8%) established the diagnosis of medium chain acyl-CoA dehydrogenase deficiency and other 5 laboratories (7%) reported the diagnosis of 2-hydroxyglutaric aciduria. One lab reported lactic acidosis due to biotinidase deficiency, it **was elected as critical error by SAB.** The 4 remaining laboratories established other diagnosis (2 Ethylmalonic encephalopathy, 1 Brown-Vialetto-Van Laere and Fazio Londe syndrome mimiking mild MADD and 1 Isobutyryl-CoA dehydrogenase deficiency).

- **Sample QLOU-EB-2018-H:**

Patient details: 6 year-old female. Autistic spectrum disorder.

Diagnosis: Normal sample, obtained from a healthy child.

Analytical details: No abnormalities were reported by 71/71 participants (100%).

Interpretation: All laboratories 71/71 reported correctly as normal sample.The overall diagnostic performance was 100%.

- **Sample QLOU-EB-2018-I:**

Patient details: 41 year-old male with developmental delay and feeding refusal, under treatment.

Diagnosis: Isovaleric acidemia

Analytical details: 66/71 laboratories (94%) reported the increase of isovalerylglycine and only 13 laboratories (19%) detected the increase of isovalerylglutamate, wich is a key metabolite of this disease.

Interpretation: The correct diagnosis was performed by 70 laboratories, the overall diagnostic performance was 99%. One laboratory reported the diagnosis of multiple acyl-CoA dehydrogenase deficiency (MADD), **it was elected as critical error by SAB.**

8. Preview of the scheme in 2019

The format of the QLOU 2019 scheme will be similar to that of previous years.

2018 was the first year for electronic reporting via the CSCQ website. We will continue to evaluate results via the website for 2019.

Changes planned for 2019: Interim reports are intended to be produced automatically by a software developed by CSCQ.

This is already working in the proficiency testing schemes and has to be adopted to the QLOU requirements.

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Please note:
This annual report is intended for participants of the ERNDIM QLOU scheme. The contents should not be used for any publication without permission of the scheme advisor.