ERNDIM
Qualitative Organic acids Urine
Barcelona ANNUAL REPORT
2019

Scientific Advisor	Website for reporting results	Administration office:
Dr. Judit García Villoria Hospital Clínic de Barcelona Division of Inborn Errors of Metabolism c/Mejía Lequerica s/n Edificio Helios III, pb 08028 Barcelona, Spain e-mail: jugarcia@clinic.cat	Dr. Xavier Albe CSCQ Swiss Center for Quality Control 2 chemin du Petit-Bel-Air CH-1225 Chêne-Bourg Switzerland e-mail : Xavier.Albe@hcuge.ch	ERNDIM Adminsitration Office Manchester Centre for Genomic Medicine 6th Floor, St Mary's Hospital, Oxford Road, Manchester M13 9WL, United Kingdom. e-mail: admin@erndim.org

6th March 2020

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 Judit Garcia Barcelona Scheme 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 2019 seventy four laboratories from many different countries participated in the QLOU *Barcelona* scheme. 2 laboratories were educational participants in 2019 (1 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.

Participants and new applicants will distributed between the Barcelona, Heidelberg and Sheffield qualitative urinary organic acid schemes which are run separately. The three organising laboratories each participate in the other's scheme by rotation.

Table 1: Geographical distribution of participants

Country	*Number of laboratories*		*Country*	*Number of laboratories*
ARGENTINA	4		INDIA	3
BRAZIL	2		ITALY	14
CHILE	1		KINGDOM OF SAUDIA ARABIA	1
CHINA	1		LEBANON	1
COLOMBIA	1		PHILIPPINES	1
CYPRUS	1		PORTUGAL	2
FRANCE	22		REPUBLIC OF SINGAPORE	1
GERMANY	2		SPAIN	9
GREECE	1		URUGUAY	1
HONG KONG	4			

3. Design of the scheme and logistics

As usual, the samples used in 2019 were authentic human urine samples, 6 from affected patients and 3 from healthy individuals.
All samples selected by the Scientific Advisor have been heat-treated and were tested for suitability in the Scientific Advisor's laboratory.
In 2019 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
Labelled copies of chromatograms can be uploaded on the CSCQ website.

4. Schedule of the scheme

Table 2: Time schedule in the 2019 ERNDIM QLOU *Barcelona* scheme.

	1st Submission Round	2nd Submission Round	3rd Submission Round
Sample ID's:	QLOU-EB-2019-A QLOU-EB-2019-B QLOU-EB-2019-C	QLOU-EB-2019-D QLOU-EB-2019-E QLOU-EB-2019-F	QLOU-EB-2019-G QLOU-EB-2019-H QLOU-EB-2019-I
Shipment of samples	February 5th, 2019		
Start of analysis (clinical data available)	May 13th, 2019	July 8th, 2019	September 9th, 2019
Reminder for result submission	May 27th, 2019	July 22th, 2019	September 23th, 2019
Results submission deadline:	June 3rd, 2019	July 31st, 2019	September 30th, 2019
Interim reports available on CSCQ website	*September 2019*	*November 2019*	*January 2020*

To be able to continue this scheme we need a steady supply of new patient samples. Several laboratories have donated samples to the Urine QLOU 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 3: Samples included in the 2019 ERNDIM QLOU *Barcelona* scheme.

Survey	Sample no.	Diagnosis
18-05-OUB	QLOU-EB-2019-A	Mevalonic aciduria
	QLOU-EB-2019-B	2-amino/2-ketoadipic aciduria
	QLOU-EB-2019-C	Normal
18-07-OUB	QLOU-EB-2019-D	Normal
	QLOU-EB-2019-E	MSUD
	QLOU-EB-2019-F	Hyperoxaluria type I
18-09-OUB	QLOU-EB-2019-G	Combined Malonic and Methylmalonic aciduria
	QLOU-EB-2019-H	Normal
	QLOU-EB-2019-I	2-Methylbutyrylglycinuria

The scheme format was kept identical to those of previous years. Samples were shipped by regular mail. Details regarding stability of samples are provided in the sample package.

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

5. Results

Table 4: Receipt of results in the 2019 ERNDIM QLOU *Barcelona* scheme.

Survey	In time	Late	Total
19-05-OUB	69	0	69
19-07-OUB	69	0	69
19-09-OUB	69	0	69

Table 5: Returned results in the 2019 ERNDIM QLOU *Barcelona* scheme.

Submissions	Number of laboratories	%
3	66	89
2	4	5
1	1	1
0	3	4

6. Website reporting

The website reporting system is compulsory for all centres. Please read carefully the following advice:

- Results
  - Give quantitative data as much as possible.
  - Enter the key metabolites with the evaluation **in the tables** even if you don't give quantitative data.
  - If the profile is normal: enter "Normal profile" in "Key metabolites".
  - **Don't enter results in the "comments" window**, **otherwise your results will not be included in the evaluation program.**
- ***Diagnosis***
  - **Don't enter the diagnosis in the "comments" window**, **otherwise your results will not be included in the evaluation program.**
- Recommendations = **advice for further investigation**.
  - Scored together with the interpretative score.
  - Advice for treatment are not scored.
  - **Don't give advice for further investigation in "Comments on diagnosis"**: it will not be included in the evaluation program.

7. 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.
Qualitative results and diagnostic proficiency of the 2019 samples were scored using the criteria given in Table 6. 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 21st, 2019).

Table 6: 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 21st, 2019.

Table 7: Samples eligible for critical errors in the 2019 ERNDIM QLOU *Barcelona* scheme.

Sample	Critical errors
QLOU-EB-2019-E	3 for failing to identify MSUD

Details are given under item 9 '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'.

Satisfactory performance is defined as **70% of maximum score** which equates **25/36** points for three returns and **17/24** points for two returns.

8. Proficiency of the 2019 surveys

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

In 2019, 66 participants submitted 3 reports including 1 educational participant. From the 65 ordinary (non-educational) participants 60 (91%) achieved satisfactory performance (score ≥ *25 / 17*, no critical error). 6 participants did not accomplish satisfactory performance, including 1 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 8.

Table 8: Overall proficiencies of the 2019 surveys.

Sample ID	Sample type	Proficiency (%)
QLOU-EB-2019-A	Mevalonic aciduria	99%
QLOU-EB-2019-B	2-amino/2-ketoadipic aciduria	67%
QLOU-EB-2019-C	Normal	95%
QLOU-EB-2019-D	Normal	89%
QLOU-EB-2019-E	MSUD	93%
QLOU-EB-2019-F	Hyperoxaluria type I -– **Educational sample**	44%
QLOU-EB-2019-G	Combined Malonic and Methylmalonic aciduria	71%
QLOU-EB-2019-H	Normal	93%
QLOU-EB-2019-I	2-Methylbutyrylglycinuria	87%

7 Performance Support letters will be sent for the 2019 surveys. 1 of these *7* participants have also received a performance support letter in 2018 or 2017. 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.
For the 2018 scheme 8 Performance Support letters were sent.

9. Results of individual samples and evaluation of reporting

- **Sample QLOU-EB-2019-A:**

Diagnosis: **Mevalonate kinase deficiency**. The overall proficiency for the 69 laboratories that submitted their results was 99%.

Analytical details: The organic acid profile showed pathological excretion of mevalonate lactone and unsaturated mevalonate. This last acid only was reported by 3 laboratories, for that reason the mass spectrum was added in interim report. The scoring was 2 points for the detection of the increase of mevalonate lactone and/or mevalonate.

Interpretation: The correct diagnosis was performed by 68 laboratories (99%). The scoring was 2 points for the correct diagnosis of mevalonate kinase deficiency; 1 point for the diagnosis of hyper-IgD syndrome with any other alternative diagnosis.

- **Sample QLOU-EB-2019-B:**

Diagnosis: **2-amino/2-ketoadipic aciduria**. The overall proficiency for the 69 laboratories that submitted their results was 67%.

Analytical details: The organic acid profile showed pathological excretion of 2-ketoadipate and 2-hydorxyadipate. Less than 65% of laboratories detected these metabolites, for that reason the mass spectrum of both compounds were included in the interim report. The scoring was 2 points for the detection of increased 2-ketoadipate and 2-hydorxyadipate. 1 point if any of these metabolites was missed.

Interpretation: the correct diagnosis was performed by 43 laboratories (62%). The scoring was 2 points for the correct diagnosis of 2-amino/2-ketoadipic aciduria.

- **Sample QLOU-EB-2019-C:**

Diagnosis: **Normal sample**, obtained from a healthy adult. The overall proficiency for the 69 laboratories that submitted their results was 95%.

Interpretation: The majority of laboratories, 65 (94%), reported correctly as normal sample. The other 4 laboratories reported as diagnosis a beta-oxidation defect due to clinical description of recurrent episodes of rhabdomyolysis and myoglobinuria.

The maximum score of this sample is for the interpretation of normal profile.

- **Sample QLOU-EB-2019-D:**

Diagnosis: **Normal sample**, obtained from a healthy adult. The overall proficiency for the 69 laboratories that submitted their results was 89%.

Interpretation: The majority of laboratories, 59 (86%), reported correctly as normal sample.
3 laboratories reported the diagnosis of Glycerol kinase deficiency, due to slight increase of glycerol and by the clinical presentation with anorexia and autistic behavior. The remaining 6 laboratories reported individual diagnosis.

The maximum score of this sample is for the interpretation of normal profile.

- **Sample QLOU-EB-2019-E:**

Diagnosis: **Maple Syrup Urine Disease (MSUD).** The overall proficiency for the 69 laboratories that submitted their results was 93%.

Analytical details: 60 laboratories (87%) reported elevated excretion of 2-hydroxy-isovalerate. Less than 55 % of laboratories reported the elevation of 2-Hydroxy-3-methylvalerate and 2-

hydroxy-isocaproate and less than 11% detected the increase of N-acetyl-valine, N-acetyl-isoleucine, N-acetil-alloisoleucine. For that reason the mass spectrum of these three last metabolites were included in the interim report. P-hydroxy-phenyl-lactate was also increased. In addition, if oximation was used an increase of 2-Keto-3-methyl-n-valerate and 2- ketoisocaproate could be found. The scoring was 2 points for the detection of an increase of 2-hydroxy-isovalerate and 1 point when the alteration of some metabolite is reported, if this is normal and creates confusion for the definitive diagnosis.

Interpretation: The correct diagnosis was performed by 64 laboratories (93%). The scoring was 2 points for the correct diagnosis of Maple Syrup Disease (MSUD) and 1 point for the diagnosis of Dihydrolipoamide dehydrogenase (Subunit E3) deficiency.

It was decided at the November SAB critical error meeting that the failure to identify MSUD in sample **QLOU-UB-2019-E** and reporting the alterations secondary to hepatic failure or Tyrosinemia would also be **classed as a critical error.** Three participants were awarded critical error.

- **Sample QLOU-EB-2019-F:**

Diagnosis: **Hyperoxaluria type I**. The overall proficiency for the 69 laboratories that submitted their results was 44%.

Analytical details: 36 laboratories (52%) indentified an increase of glycolate, but only 16 laboratories (23%) reported an increase of oxalate. 8 laboratories (12%) reported only hyperoxaluria and 1 lab hyperoxaluria type 2.23 laboratories (33%) reported this sample as normal.

Interpretation: Only 31 laboratories (45%) reported the correct diagnosis of hyperoxaluria type 1. Eight laboratories (12%) reported only hyperoxaluria and one lab hyperoxaluria type 2. The remaining laboratories (33%) reported this sample as normal.

It is known that the detection of oxalic acid using the conventional extraction from organic acids in urine is not the best method. However, in order to improve the diagnosis of hyperoxaluria type 1, in the cases that the labs find an increase of glycolic acid the lab should recommend the quantification of oxalic acid by other specific methods.

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-2019-G:**

Diagnosis: **Combined malonic and methylmalonic aciduria**. The overall proficiency for the 69 laboratories that submitted their results was 71%.

Analytical details: The majority of laboratories, 67 (97%), reported an increase of methylmalonate, but only 31 labs (45%) detected the elevation of malonate. The scoring was 2 points for the detection of increased methylmalonate and malonate. 1 point if any of these metabolites was missed.

Interpretation: Only 26 laboratories (38%) reported the correct diagnosis of combined malonic aciduria and methylmalonic aciduria. The remaining labs reported the diagnosis of vitamin B12 deficiency (41 %) or methylmalonic aciduria (14%). The scoring was 2 points for the correct diagnosis of combined malonic aciduria and methylmalonic aciduria . 1 point for the diagnosis of methylmalonic aciduia or Vitamin B12 deficiency. 0 points for the diagnosis of malonic aciduria or normal profile.

- **Sample QLOU-EB-2019-H:**

Diagnosis: **Normal sample**, obtained from a healthy adult. The overall proficiency for the 69 laboratories that submitted their results was 93%.

Interpretation: The majority of laboratories, 65 (94%), reported correctly as normal sample. The remaining 4 laboratories reported individual diagnosis.

The maximum score of this sample is for the interpretation of normal profile.

- **Sample QLOU-EB-2019-I:**

Diagnosis: 2-Methylbutyrylglycinuria. The overall proficiency for the 69 laboratories that submitted their results was 87%.

Analytical details: 59 laboratories (86%) indentified an increase of 2- methylbutyrylglycine. 4 laboratories identified this metabolite as isovalerylglycein, for that reason the mass spectrum of both acylglycine were included in the interim report. The scoring was 2 points for the detection of increased 2- methylbutyrylglycine.

Interpretation: 57 laboratories (83%) reported the correct diagnosis of 2-methylbutyryl-CoA dehydrogenase deficiency. The remaining labs reported other individual diagnosis. The scoring was 2 points for the correct diagnosis of 2-methylbutyryl-CoA dehydrogenase deficiency or 2-methylbutyrylglycinuria.

10. Scores of participants

Table 9 presents detailed scores and performance data for all participants.

Scores and performance data were confirmed by the Scientific Advisory Board meeting in November 2019.

The anonymous data are accessible to all participants. Individual data are only visible to your laboratory.

Lab no	A	B	C	sum	D	E	F*)	sum	G	H	I	sum	Total score	Performance
1	4	4	4	12	4	4		8	2	4	4	10	30	
2	4	4	4	12	2	4		6	4	4	4	12	30	
3														NO RETURN
4					4	0		4	2	4	4	10	14	2 RETURN PP CE
5	4	4	4	12	4	4		8	3	4	4	11	31	
6	4	4	4	12	4	4		8	4	4	2	10	30	
7	4	4	4	12	4	4		8	4	4	4	12	32	
8	4	0	4	8	0	4		4	2	4	4	10	22	

Lab no	A	B	C	sum	D	E	F*)	sum	G	H	I	sum	Total score	Performance
9	4	4	4	12	4	4		8	4	4	4	12	32	
10	4	0	4	8	0	4		4	2	4	4	10	22	
11	4	3	4	11									11	1 RETURN
12	4	0	4	8	4	4		8	4	4	4	12	28	
13	4	4	4	12	2	4		6	2	4	4	10	28	
14	4	0	4	8	4	4		8	1	4	4	9	25	
15	4	0	4	8	2	4		6	2	4	4	10	24	
16	3	0	2	5	4	4		8	2	4	4	10	23	
17	4	4	4	12	2	4		6	2	4	4	10	28	
18	4	2	4	10	4	4		8	2	4	4	10	28	
19	4	1	4	9	4	2		6	2	4	4	10	25	
20	4	4	4	12	4	4		8	4	4	4	12	32	
21	4	4	4	12	4	4		8	4	4	4	12	32	
22					4	4		8	4	4	4	12	20	2 RETURNS
23	4	3	4	11	4	4		8	4	4	4	12	31	
24	4	2	0	6	4	4		8	2	2	4	8	22	
25	4	3	4	11	4	0		4	2	2	4	8	23	CE
26	4	0	4	8	4	3		7	4	4	0	8	23	
27	4	4	4	12	4	4		8	2	4	0	6	26	
28	4	4	4	12	4	4		8	4	4	4	12	32	
29	4	4	4	12	4	4		8	2	4	4	10	30	
30	4	4	4	12	4	3		7	2	4	4	10	29	
31	4	0	4	8	2	4		6	4	4	4	12	26	
32	4	1	4	9	4	4		8	4	4	4	12	29	

Lab no	A	B	C	sum	D	E	F*)	sum	G	H	I	sum	Total score	Performance
33	4	4	4	12	4	4		8	2	4	4	10	30	
34	4	4	4	12	4	4		8	2	4	4	10	30	
35	4	4	4	12	4	4		8	4	4	4	12	32	
36	4	0	0	4	4	4		8	2	1	4	7	19	PP
37	4	4	4	12	4	4		8	0	4	4	8	28	
38	4	4	4	12	4	4		8	2	4	4	10	30	
39	4	4	4	12	4	4		8	2	4	4	10	30	
40	4	4	4	12	4	4		8	4	4	4	12	32	
41	4	4	4	12	4	4		8	4	4	2	10	30	
42	4	4	4	12	4	4		8	2	4	4	10	30	
43	4	4	4	12	4	4		8	2	4	0	6	26	
44	4	4	4	12				0	4	4	4	12	24	2 RETURNS
45	4	4	4	12	4	4		8	4	4	4	12	32	
46	4	1	4	9	4	0		4	2	4	4	10	23	CE
47	4	4	4	12	4	4		8	2	4	4	10	30	
48	4	0	4	8	4	4		8	4	4	4	12	28	
49	4	4	4	12	4	4		8	4	4	4	12	32	
50	4	4	2	10	4	3		7	3	2	4	9	26	
51	4	0	4	8	4	4		8	4	4	4	12	28	
52														NO RETURN
53	4	3	4	11	4	4		8	4	4	2	10	29	
54	4	0	4	8	4	4		8	2	4	4	10	26	
55	4	4	4	12	4	4		8	4	4	4	12	32	
56	4	4	4	12	4	4		8				0	20	2 RETURNS

Lab no	A	B	C	sum	D	E	F*)	sum	G	H	I	sum	Total score	Performance
57	4	4	4	12	4	4		8	2	4	4	10	30	
58	4	4	4	12	4	4		8	2	4	4	10	30	
59	4	4	4	12	4	4		8	2	4	4	10	30	
60	4	4	4	12	4	4		8	3	4	4	11	31	
61	4	4	4	12	4	4		8	4	4	0	8	28	
62	4	1	4	9	4	4		8	2	4	4	10	27	
63	4	4	4	12	2	4		6	2	4	2	8	26	
64	4	4	4	12	0	4		4	2	4	4	10	26	
65	4	4	4	12	4	4		8	4	4	4	12	32	
66	4	4	4	12	4	4		8	4	0	2	6	26	
67	4	0	4	8	4	4		8	2	4	4	10	26	
68	4	2	4	10	4	4		8	4	4	4	12	30	
69	4	0	4	8	0	4		4	2	4	4	10	22	
70	4	0	4	8	4	4		8	2	4	4	10	26	
71	4	0	4	8	4	4		8	4	2	2	8	24	
72	4	1	2	7	2	2		4	2	4	0	6	17	PP
73														NO RETURN
74	4	4	4	12	4	4		8	4	4	4	12	32	

*) Educational sample
CE: Critical error
PP: Poor performance (on score)

Figure 1: Boxplot presentation of all scores
Outliers result from contributing less than three submissions

11. Preview of the scheme in 2020

The format of the QLOU 2020 scheme will be similar to that of previous years.

Changes planned for 2020:
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.

Judit García Villoria
January 2020

Judit Garcia
Scientific Advisor

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