ERNDIM Administration Office
ERNDIM Administration Office
c/o EMQN CIC, Unit 4, Enterprise House,
Manchester Science Park Pencroft Way
Manchester M15 6SE
United Kingdom.
e-mail: admin@erndim.org

Scientific Coordination
Petr Chrastina
Department of Pediatrics and Inherited Metabolic Disorders
General University Hospital in Prague
Ke Karlovu 2
128 08 Prague 2
Czech Republic
Email: petr.chrastina@vfn.cz

Scheme Organisation
1) Alessandro Salemma; **2)** Nicola Braik
CSCQ, Swiss Center for Quality Control
2 chemin du Petit-Bel-Air
CH-1225 Chêne-Bourg
Switzerland
Email: **1)** Alessandro.Salemma@hcuge.ch;
2) Nicola.Braik@hcuge.ch

Published: 26 April 2022[1]

Diagnostic Proficiency Testing

Centre: Czech Republic

Final Report 2021

prepared by
Petr Chrastina

Note: This annual report is intended for participants of the ERNDIM DPT Czech Republic scheme. The contents should not be used for any publication without permission of the Scientific Advisor.
The fact that your laboratory participates in ERNDIM schemes is not confidential, however, the raw data and performance scores are confidential and will only be shared within ERNDIM for the purpose of evaluating your laboratories performance, unless ERNDIM is required to disclose performance data by a relevant government agency. For details, please see the terms and conditions in the ERNDIM Privacy Policy on www.erndim.org.

1. Geographical distribution of participants

Twenty laboratories from 13 countries have participated in the Diagnostic Proficiency Testing scheme in 2021, for details see the below table:

Country	Number of participants	Country	Number of participants
Austria	1	Latvia	1
Croatia	1	Lithuania	1
Cyprus	1	Malaysia	2
Czechia	1	Portugal	1
Denmark	1	Slovakia	2
Finland	1	United Kingdom	1
Germany	6		

2. Design and logistics of the scheme including sample information

The scheme has been designed and planned by Petr Chrastina as Scientific Advisor and coordinated by CSCQ as scheme organiser (sub-contractor on behalf of ERNDIM), both appointed by and according to procedures laid down the ERNDIM Board.
CSCQ dispatches DPT EQA samples to the scheme participants and provides a website for on-line submission of results and access to scheme reports. Existing 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

2 surveys	Round 1: patients A, B and C
	Round 2: patients D, E and F

[1] If this report is not Version 1 for this scheme year, go to APPENDIX 1 for details of the changes made since the last version of this document.

Origin of patients: All six urines were obtained from patients with known diagnoses. Four urine samples have been provided by the scheme organizers and one sample has been provided by Department of Clinical Biochemistry of University Children's Hospital in Bratislava. The common sample was from DPT center Netherlands (distributed in all five DPT schemes).

In 2021 the samples have been heat-treated and apart from the common sample A were re-analyzed in our department after receiving the samples from CSCQ (samples were shipped via courier after 3 days at ambient temperature to mimic possible changes that might arise during transport). In all six samples prepared and checked by us the typical metabolic profiles were preserved after heat treatment and shipment from CSCQ.
Mailing: samples were sent by DHL; FedEx or the Swiss Post at room temperature.

3. Tests

Analyses of amino acids, organic acids, mucopolysaccharides, oligosaccharides and purines/pyrimidines were required in 2021.

4. Schedule of the scheme

Sample distribution by CSCQ	09 February 2021
Start of analysis of Survey 2021/1	08 March 2021
Survey 2021/1 – results submission	29 March 2021
Survey 2021/1 – report	17 May 2021
Start of analysis of Survey 2021/2	07 June 2021
Survey 2021/2 – results submission	28 June 2021
Survey 2021/2 – report	23 August 2021
Annual meeting of participants	07 September 2021
Annual report 2021	December 2021

5. Results

19 of 20 labs returned results for both surveys by the deadline.

	Survey 1	**Survey 2**
Receipt of results	19	19
No answer	1	1

6. Website reporting

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

- Selection of tests: **don't select a test if you will not perform it**, otherwise the evaluation program includes it in the report.
- 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.**
- Recommendations = **advice for further investigation**.
  - Scored together with the interpretative score.
  - Advice for treatment is not scored.
  - **Don't give advice for further investigation in "Comments on diagnosis"**: it will not be included in the evaluation program.

7. Scoring and evaluation of results

Information regarding procedures for establishment of assigned values, statistical analysis, interpretation of statistical analysis etc. can be found in generic documents on the ERNDIM website.

The scoring system has been established by the International Scientific Advisory Board of ERNDIM. Two criteria are evaluated: 1) analytical performance, 2) interpretative proficiency also considering recommendations for further investigations.

A	Analytical performance	Correct results of the appropriate tests	2
		Partially correct or non-standard methods	1
		Unsatisfactory or misleading	0
I	Interpretative proficiency & Recommendations	Good (diagnosis was established)	2
		Helpful but incomplete	1
		Misleading or wrong diagnosis	0

The total score is calculated as a sum of these two criteria. The maximum to be achieved is 4 points per sample. The scores were calculated only for laboratories submitting results.

Scoring and certificate of participation: scoring is carried by a second assessor who changes every year as well as by the scientific advisor. The results of DPT Czech Republic 2021 have been also scored by George Ruijter, from DPT NL. At the SAB meeting on 26th October 2021, the definitive scores have been finalized.

ERNDIM is now applying the concept of 'critical error' in the scoring of results. In principle this is a category of error that would be unacceptable to the majority of labs and would have a serious adverse effect on patient management.
Examples of such errors could be:
a) Failure to recognise a pre-defined set of diagnoses.
b) Missing a diagnosis when proficiency for that EQA sample is >95% (where proficiency for an EQA sample is the percentage of EQA participants that correctly identified the diagnosis of the sample).
c) Failure to perform a relevant test.
d) Identifying a 'normal' sample as having an IEM when it is clear that the sample was obtained from a patient not suspected of having an IEM and the findings reported were not identified by the rest of the participants and this diagnosis could potentially result in treatment that is harmful for the patient.

When a critical error is established for one or more samples, performance is not acceptable in that year, regardless of the number of points assigned. A critical error needs to be ratified by the ERNDIM Scientific Advisory Board. There were two critical errors in 2021.

A certificate of participation will be issued for participation, and it will be additionally notified whether the participant has received a performance support letter. This performance support letter is sent out if the performance is evaluated as unsatisfactory. Any partial submitters will receive a letter from the ERNDIM Executive Administrator, Sara Gardner.

7.1. Score for satisfactory performance

Performance of the participant that obtained at least 12 points from the maximum of 20 (60%) and more within the calendar year and that did not receive "critical error" mark is considered satisfactory.

From 2022 satisfactory performance will generally be held to be a score of seventeen (71%) or more.

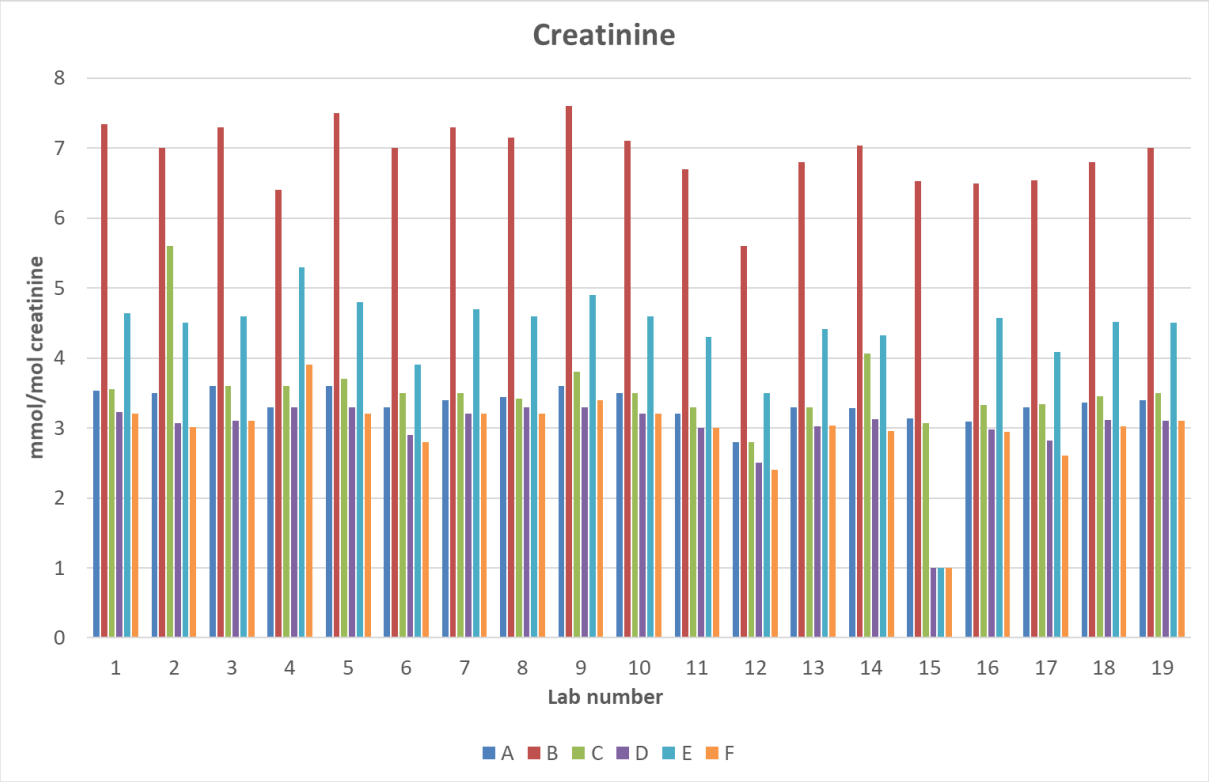
If your laboratory is assigned poor performance and you wish to appeal against this classification, please email the ERNDIM Administration Office (admin@erndim.org), with full details of the reason for your appeal, within one month receiving your Performance Support Letter. Details of how to appeal poor performance are included in the Performance Support Letter sent to poor performing laboratories.

8. Results of samples and evaluation of reporting

8.1. Creatinine measurement for all samples

Creatinine determination was mostly satisfying. Creatinine values are expressed in the figure as the ratio of each measurement over the median of all labs.

Creatinine: ratio to median



Sample	A	B	C	D	E	F
mean	3,35	6,91	3,57	2,98	4,30	2,96
median	3,36	7,00	3,50	3,10	4,52	3,04
SD	0,20	0,47	0,56	0,52	0,89	0,56

8.2. Patient A

alpha-mannosidosis due to alfa-mannosidase deficiency

Patient details provided to participants
A 36 year old male with craniosynostosis, dysmorphic facial features, retardation and deafness.

Patient details
This sample was obtained from a 36 years old man with alpha-mannosidosis due to alpha-mannosidase deficiency, diagnosis was confirmed by enzymatic analysis.

Analytical performance
17 labs performed OLS analysis and all of them reported a correct analytical finding "OLS profile characteristic for alpha-mannosidosis", which was scored with 2 points. The analytical performance was good (89%).

Interpretative proficiency and recommendation
The diagnosis of alpha-mannosidosis due to alfa-mannosidase deficiency was considered correct and scored with 2 points. Confirmation of diagnosis by enzyme assay of alfa-mannosidase activity in plasma/fibroblasts/leucocytes and/or mutation analysis of *MAN2B1* gene were considered helpful. Recommendation to carry out oligosaccharide analysis for those participants who did not perform this analysis was considered also helpful and scored with 1 point. The interpretative proficiency score for this sample was very good (92%).

Critical errors
The failure to carry out oligosaccharide analysis and to recommend this mandatory test is considered by the ERNDIM SAB as a critical error, which would prevent establishing the correct diagnosis; critical error was assigned to one participant in our scheme.

Overall impression
Typical DPT sample with very good proficiency score (91%).

8.3. Patient B

Mucopolysaccharidosis type I due to alpha-L-iduronidase deficiency

Patient details provided to participants
A 6 years old boy was referred for stiff finger joints; rheumatoid arthritis has been excluded. The sample was collected at the age of 18 years on the specific treatment.

Patient details
This sample was obtained from an 18 years old boy with mucopolysaccharidosis type I due to deficiency of alpha-L-iduronidase. Patient received enzyme replacement therapy. The diagnosis was confirmed by enzymatic analysis.

Analytical performance
17 participants analysed glycosaminoglycans (GAG) in urine and 14 participants performed GAG fractionation. This was a challenging sample because the patient received enzyme replacement therapy and GAG excretion was normal until borderline. Only 3 of 17 labs reported elevated excretion of GAG. Elevated excretion of glycosaminoglycans without report on dermatan sulphate elevation was considered as partially correct and scored with 1 point. Increased proportion of dermatan sulphate was scored as correct analytical result with 2 points. Analytical performance slightly suboptimal (68%).

Interpretative proficiency and recommendation
The diagnosis of mucopolysaccharidosis type I was considered correct (2 points), while suspicion for MPS (other types of MPS or non-specified MPS) was considered helpful but incomplete (1 point). Confirmation of diagnosis by measurement of alfa-L-iduronidase in leukocytes/fibroblasts and/or mutation analysis of *IDUA* gene were considered helpful. Recommendation to carry out GAG fractionation for those participants that did not perform this analysis was considered also helpful (1 point). The interpretative proficiency score for this sample was slightly suboptimal (68%).

Critical errors
No critical error for this sample.

Overall impression
Challenging DPT sample with slightly suboptimal proficiency score (68%).

8.4. Patient C

3-methylcrotonyl-CoA carboxylase deficiency

Patient details provided to participants
A 2 years old boy presented with horseshoe kidney and short stature. The sample was collected at the age of 2 years.

Patient details
This sample was obtained from a 5 months old boy with 3-methylcrotonyl-CoA carboxylase deficiency. The diagnosis was confirmed by molecular genetic analysis.

Analytical performance
All participants analysed organic acids and all of them reported elevated excretion of 3-hydroxyisovalerate and 3-methylcrotonylglycine. Such analytical finding was considered correct result and scored with 2 points. The proficiency score for this sample was excellent (100%).

Interpretative proficiency and recommendation
3-methylcrotonyl-CoA carboxylase deficiency was considered correct diagnosis and scored with 2 points. Confirmation of diagnosis by enzymatic assay of 3-methylcrotonyl-CoA carboxylase and/or mutation analysis of *MCCC1* and *MCCC2* gene was considered helpful. The proficiency score for this sample was excellent (100%).

Critical errors
No critical error for this sample.

Overall impression
Easy DPT sample with excellent proficiency score (100%).

8.5. Patient D

No IEM

Patient details provided to participants
A 5 months old boy presented with microcephaly, dysmorphia and epilepsy. The sample was collected at the age of 5 months.

Patient details
This sample was obtained from a 5 months old boy without any evidence of an inherited metabolic disorder after extensive metabolic screening.

Analytical performance
Nineteen labs performed analysis of organic acids and 16 of them reported elevated excretion of 3-hydroxybutyrate, such analytical finding was considered correct and scored by 1 point. 16 participants analysed minimally 3 of the 5 required methods and they reported normal profile except for organic acids, such analytical finding was considered correct and scored by 1 point. The analytical performance was good (87%).

Interpretative proficiency and recommendation
We considered the report of “no IEM”, “non-specific finding”, 3-hydroxybutyrate treatment and ketonuria due to malnutrition a good diagnosis, which was scored with 2 points. Other diagnosis we scored with 0 points. The interpretative proficiency score for this sample was good (84%).

Critical errors
No critical error for this sample.

Overall impression
Typical DPT sample with good proficiency score (86%).

8.6. Patient E

Hyper IgD syndrome

Patient details provided to participants

This female patient was referred at the age of 8 years with suspicion for juvenile idiopathic arthritis. Since the age of 6 years, repeated febrile illness were observed. The sample was collected at the age of 8 years during a febrile illness.

Patient details

This sample was obtained from an 8 years old girl with hyper-IgD syndrome due to mevalonate kinase deficiency. The diagnosis was confirmed by molecular genetic analysis.

Analytical performance

All participants analysed organic acids and 16 of them reported elevated excretion of mevalonolactone or mevalonic acid. Such analytical finding was considered correct result and scored by 2 points. The proficiency score for this sample was good (84%).

Interpretative proficiency and recommendation

Hyper-IgD syndrome or mevalonate kinase deficiency was considered correct diagnosis and scored by 2 points. The diagnosis of Hyper-IgD syndrome based on clinical information was scored with 1 point. Confirmation of diagnosis by mutation analysis of *MVK* gene was considered helpful. The proficiency score for this sample was good (89%).

Critical errors

The failure to recognize abnormal excretion of mevalonolactone and to recommend mevalonolactone analysis based on clinical information is considered by the ERNDIM SAB as a critical error, which would prevent establishing the correct diagnosis; critical error was assigned to one participant in our scheme.

Overall impression

Typical DPT sample with good proficiency score (87%).

Figure 1: Organic acids profile (GC/MS) in urine of patient 2021E (heat-treated urine after 3 days at RT)

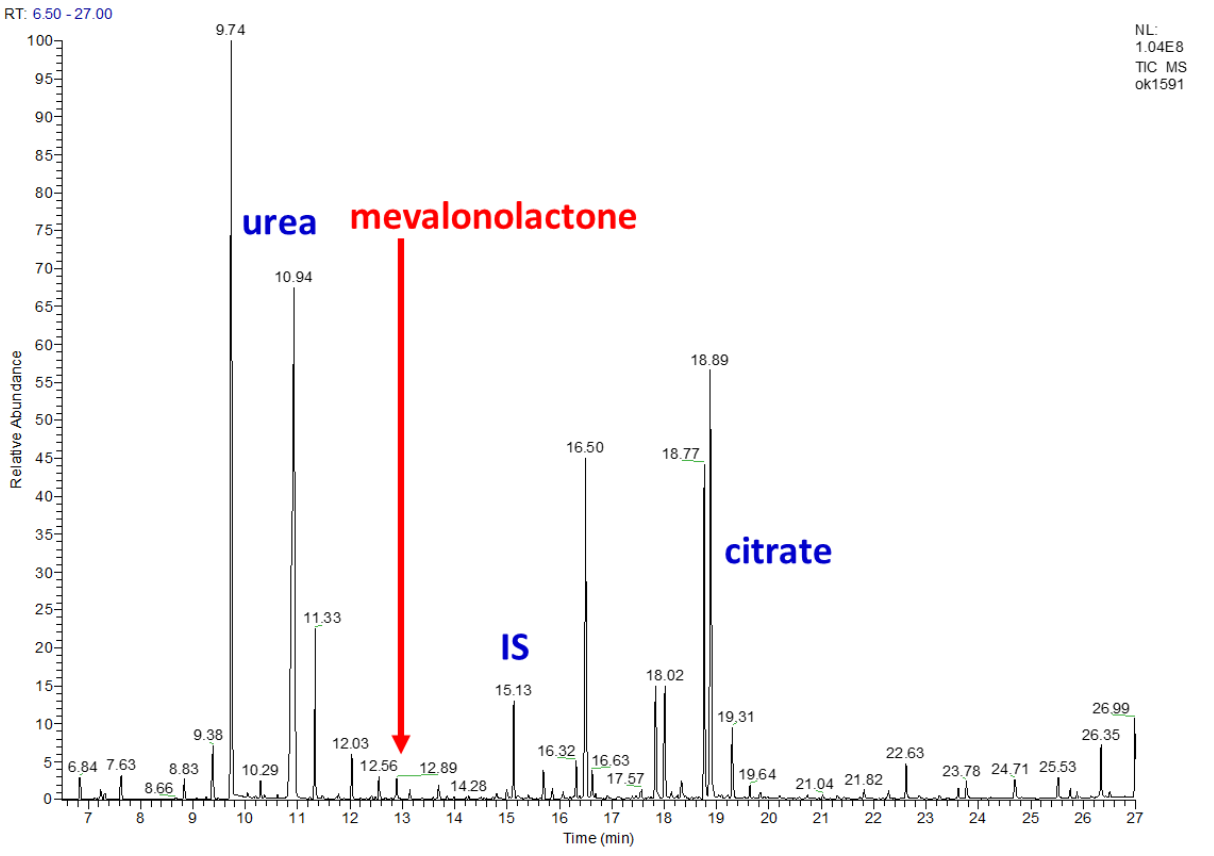
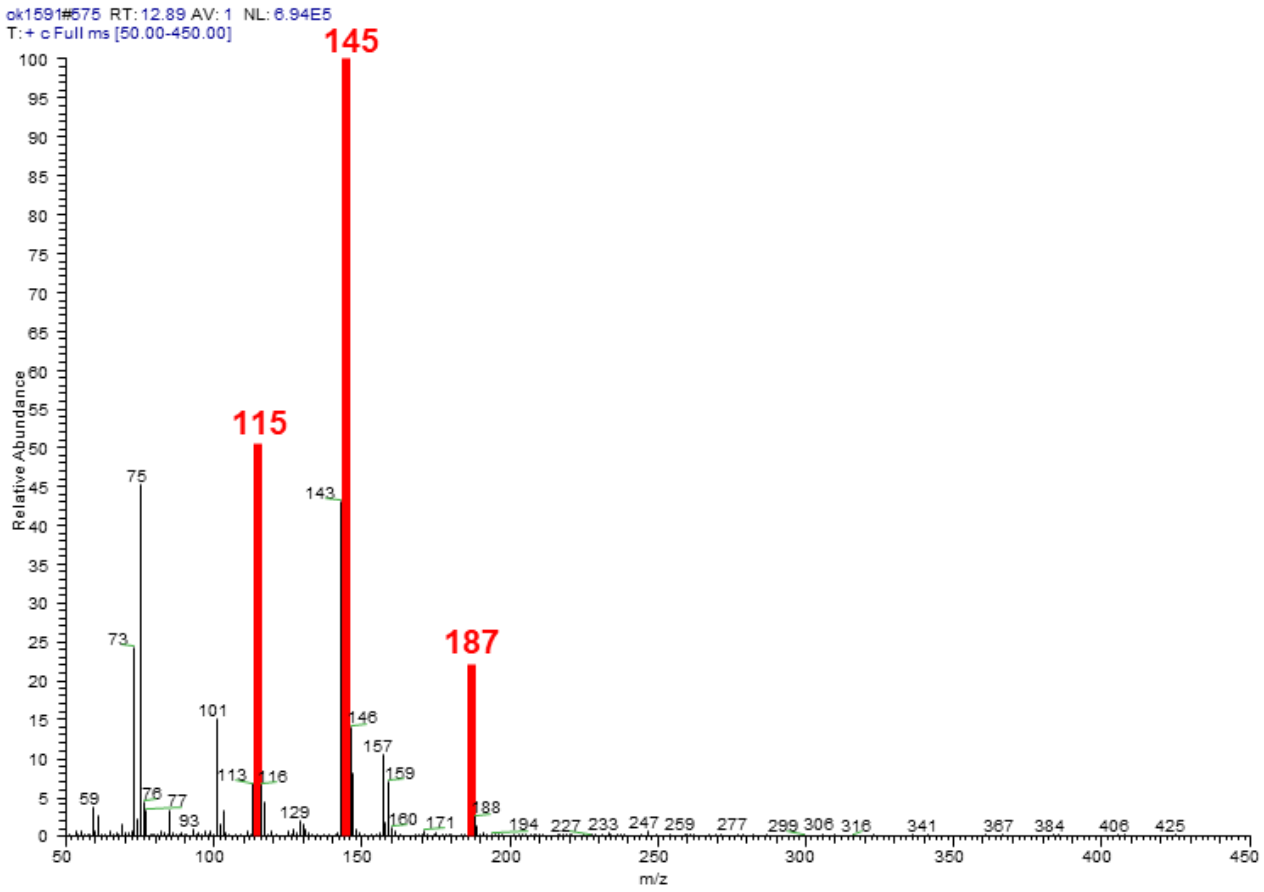


Figure 2: EI mass spectrum of mevalonolactone 1TMS



8.7. Patient F

Tyrosinemia type I

Patient details provided to participants
A 2 months old boy was admitted to hospital for hepatic failure. The sample was collected at the age of 13 years on the specific treatment.

Patient details
This sample was obtained from a 13 years old boy with tyrosinemia type I. The diagnosis was confirmed by molecular genetic analysis.

Analytical performance
All participants analysed organic acids and all of them reported elevated excretion of tyrosine metabolites (4-hydroxyphenylpyruvate, 4-hydroxyphenyllactate, and 4-hydroxyphenylacetate). Such analytical finding was considered correct result and scored by 1 point. All participants analysed amino acids and 18 of them reported elevated excretion of tyrosine. Such analytical finding was considered correct result and scored by 1 point. The proficiency score for this sample was very good (97%).

Interpretative proficiency and recommendation
The diagnosis of tyrosinemia type I was considered correct (2 points), while suspicion for other types of tyrosinemia was considered helpful but incomplete (1 point). Confirmation of diagnosis by mutation analysis of *FAH* gene was considered helpful. The proficiency score for this sample was very good (97%).

Critical errors
No critical error for this sample.

Overall impression
Typical DPT sample with very good proficiency score (97%).

9. Scores of participants

All data transfer, the submission of data as well as the request and viewing of reports proceed via the DPT-CSCQ results website. The results of your laboratory are confidential and only accessible to you (with your username and password). The anonymous scores of all laboratories are accessible to all participants and only in your version is your laboratory highlighted in the leftmost column.

Detailed scores – Round 1

Lab n°	Patient A alpha-mannosidosis			Patient B Mucopolysaccharidosis type I			Patient C 3-methylcrotonyl-CoA carboxylase deficiency			
	A	I	Total	A	I	Total	A	I	Total	Total
1	2	2	4	2	2	4	2	2	4	12
2	2	2	4	2	2	4	2	2	4	12
3	2	2	4	2	2	4	2	2	4	12
4	2	2	4	0	0	0	2	2	4	8
5	2	2	4	2	2	4	2	2	4	12
6	2	2	4	2	2	4	2	2	4	12
7	2	2	4	0	0	0	2	2	4	8
8	2	2	4	2	2	4	2	2	4	12
9	2	2	4	2	2	4	2	2	4	12
10	2	2	4	0	0	0	2	2	4	8
11	0	1	1	0	1	1	2	2	4	6
12	2	2	4	1	1	2	2	2	4	10
13	2	2	4	2	2	4	2	2	4	12
14	2	2	4	0	0	0	2	2	4	8
15	0	0	0	2	2	4	2	2	4	8
16	2	2	4	2	1	3	2	2	4	11
17	2	2	4	1	1	2	2	2	4	10
18	2	2	4	2	2	4	2	2	4	12
19	2	2	4	2	2	4	2	2	4	12
20	--	--	--	--	--	--	--	--	--	0

Detailed scores – Round 2

Lab n°	Patient D No IEM			Patient E Hyper IgD syndrome			Patient F Tyrosinemia type I			
	A	I	Total	A	I	Total	A	I	Total	Total
1	2	2	4	2	2	4	2	2	4	12
2	2	2	4	2	2	4	2	2	4	12
3	2	2	4	0	1	1	2	2	4	9
4	2	2	4	0	1	1	2	2	4	9
5	1	2	3	2	2	4	2	2	4	11
6	2	2	4	2	2	4	2	2	4	12
7	2	2	4	2	2	4	2	2	4	12
8	2	0	2	2	2	4	2	2	4	10
9	2	2	4	2	2	4	2	2	4	12
10	1	2	3	2	2	4	2	1	3	10
11	1	2	3	2	2	4	2	2	4	11
12	2	2	4	2	2	4	2	2	4	12
13	1	0	1	2	2	4	2	2	4	9
14	2	2	4	2	2	4	2	2	4	12
15	2	2	4	0	0	0	2	2	4	8
16	1	2	3	2	2	4	2	2	4	11
17	2	2	4	2	2	4	1	2	3	11
18	2	0	2	2	2	4	2	2	4	10
19	2	2	4	2	2	4	2	2	4	12
20	--	--	--	--	--	--	--	--	--	0

Total scores

Lab n°	A	B	C	D	E	F	Cumulative score	Cumulative score (%)	Critical error
1	4	4	4	4	4	4	24	100	
2	4	4	4	4	4	4	24	100	
3	4	4	4	4	1	4	21	88	
4	4	0	4	4	1	4	17	71	
5	4	4	4	3	4	4	23	96	
6	4	4	4	4	4	4	24	100	
7	4	0	4	4	4	4	20	83	
8	4	4	4	2	4	4	22	92	
9	4	4	4	4	4	4	24	100	
10	4	0	4	3	4	3	18	75	
11	1	1	4	3	4	4	17	71	
12	4	2	4	4	4	4	22	92	
13	4	4	4	1	4	4	21	88	
14	4	0	4	4	4	4	20	83	
15	0	4	4	4	0	4	16	67	CE
16	4	3	4	3	4	4	22	92	
17	4	2	4	4	4	3	21	88	
18	4	4	4	2	4	4	22	92	
19	4	4	4	4	4	4	24	100	
20	--	--	--	--	--	--	0	0	

Performance

	Number of labs	% total labs
Satisfactory performers (≥ 60 % of adequate responses)	18	90
Unsatisfactory performers (< 60 % adequate responses and/or critical error)	1	5
Partial and non-submitters	1	5

Overall Proficiency

Sample	Diagnosis	Analytical (%)	Interpretation (%)	Total (%)
DPT-CP-2021-A	alpha-mannosidosis	89	92	91
DPT-CP-2021-B	Mucopolysaccharidosis type I	68	68	68
DPT-CP-2021-C	3-methylcrotonyl-CoA carboxylase deficiency	100	100	100
DPT-CP-2021-D	No IEM	87	84	86
DPT-CP-2021-E	Hyper IgD syndrome	84	89	87
DPT-CP-2021-F	Tyrosinemia type I	97	97	97

10. Annual meeting of participants

The annual meeting of participants of the Proficiency Testing Centre Czech Republic was held online on 7th September 2021 instead of a face-to-face meeting as international travel restrictions were in place due to Covid-19.

- This year we encountered one major analytical difficulty, namely absent annotation of mevalonolactone in sample E.

We remind you that attending the annual meeting is an important part of the proficiency testing. The goal of the program is to **improve** the competence of the participating laboratories, which includes the critical review of all results with a discussion about improvements.

11. Information from the Executive Board and the Scientific Advisory Board

- **Training**: SSIEM Academy training courses.
  - A 2 days course will be been organized on Monday and Tuesday 27 and 28 June 2022 in Amsterdam. The program includes:
    - Aminoacidopathies
    - Hyperammonaemia
    - Homocystinurias & remethylation
- **Urine samples**: we remind you that every year, each participant must provide to the scheme organizer at least 300 ml of urine from a patient affected with an established inborn error of metabolism or “normal” urine, together with a short clinical report. If possible, please collect 1500 ml of urine: this sample can be sent to all labs participating to one of the DPT schemes. Each

urine sample must be collected from a single patient (don't send urine spiked with pathological compounds). Please don't send a pool of urines, except if urine has been collected on a short period of time from the same patient. For "normal" urine, the sample must be collected from a symptomatic patient (don't send urine from your kids!). As soon as possible after collection, the urine sample must be heated at 56 °C for 30 minutes. Make sure that this temperature is achieved in the entire urine sample, not only in the water bath. Please send us an e-mail about possible samples and we will organize transport with your cooperation.

12. Reminders

We remind you that to participate to the DPT-scheme, you must perform at least:

- Amino acids
- Organic acids
- Oligosaccharides
- Mucopolysaccharides

If you are not performing one of these assays, you can send the samples to another lab (cluster lab) but you are responsible for the results.
Please send quantitative data for amino acids and, as much as possible, for organic acids.

13. Tentative schedule and fee in 2022

Sample distribution	2 February 2022
Start of analysis of Survey 2022/1 Website open	March 14
Survey 2022/1 - Results submission	March 28
Survey 2022/1 - Reports	April
Start of analysis of Survey 2022/2	June 6
Survey 2022/2 – Results submission	June 28
Survey 2022/2 - Reports	July
Annual meeting of participants	August 30/31, Freiburg SSIEM
Annual Report 2022	December

14. ERNDIM certificate of participation

A combined certificate of participation covering all EQA schemes will be provided to all participants who take part in any ERNDIM scheme. For the DPT scheme this certificate will indicate if results were submitted and whether satisfactory performance was achieved in the scheme.

Date of report, 2022-01-30
Name and signature of Scientific Advisor

Petr Chrastina
Department of Pediatrics and Inherited Metabolic Disorders
General University Hospital in Prague
Ke Karlovu 2
128 08 Prague 2
Czech Republic
Tel: +420 224 947 161
Email: petr.chrastina@vfn.cz

APPENDIX 1. Change log (changes since the last version)

Version Number	Published	Amendments
1	26 April 2022	2021 annual report published

END