ERNDIM DPT Center Czech Republic

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Proficiency Testing Centre Czech Republic
Annual Report 2016

1. Introduction

In 2016 proficiency testing in our centre was running as a regular ERNDIM scheme.

2. Geographical distribution of participants

Twenty laboratories from 14 countries have participated in the Diagnostic Proficiency Testing scheme in 2016, for details see the below table:

Country	Number of participants
Austria	1
Croatia	1
Cyprus	1
Czech Republic	1
Denmark	1
Finland	1
Germany	5
Kingdom of Saudi Arabia	1
Latvia	1
Lithuania	1
Malaysia	1
Poland	1
Portugal	1
Slovakia	3
in total	**20**

3. Logistics of the scheme

- ✓ Two surveys: 2016/1 – samples A, B and C
  2016/2 – samples D, E and F

  Origin of samples: Five urines were obtained from patients with known diagnoses (samples were provided by the DPTC participants and by the organizers) + a common sample was from the DPTC Netherlands (distributed in all five DPT schemes).
- ✓ In 2016 the samples without addition of thiomersal have been heat-treated and with the exception of the common sample A were re-analyzed before distribution to participants after

the receipt of control shipment from CSCQ. In all five samples the typical metabolic profiles were preserved after undergoing this treatment.

- ✓ The samples for Diagnostic Proficiency Testing scheme were distributed via CSCQ in Geneva. On 1st February 2016 the urinary samples were distributed to the participants at ambient temperature using the courier. Based on the report of the courier all parcels were delivered within 3 days.
- ✓ The following protocol for heat inactivation is used (please, note that since 2016 no thiomersal is added prior to heating): Heat urine to 56 °C for one hour in water bath, make sure that this temperature is achieved in the entire urine sample and not only in the water bath. The urinary samples have to be frozen until shipment.
- ✓ Tests required in 2016: amino acids, organic acids, mucopolysaccharides, oligosaccharides and purines/pyrimidines

4. Schedule of the scheme in 2016

Sample distribution	February 1, Monday
Start of analysis of Survey 2016/1	February 22, Monday
Survey 2016/1 – results submission	March 14, Monday
Survey 2016/1 – report	May 23, Monday
Start of analysis of Survey 2016/2	May 23, Monday
Survey 2016/2 – results submission	June 13, Monday
Survey 2016/2 – report	August 15, Monday
Annual meeting of participants	September 06, Tuesday
Annual report 2016	December 07, 2016

5. Submission of results

	2016/1	**2016/2**
in time	19	19
no report	1	1

6. Samples

Sample A (common sample)

Clinical picture provided with the sample: At the age of 5 years this boy was referred for the first time to a pediatric nephrologist, because of urolithiasis. At ages 7 and 10 years, renal stones were found again. At the time of the urine collection, he was 10 years old and in good health. He used no medication, had a normal diet and adequate renal function.

The common sample provided by the DPTC Netherlands was obtained from a 10-year old patient with hyperoxaluria type 2. The diagnosis was confirmed by molecular genetic analysis.

Analytical performance: All participants analyzed organic acids and all participants observed the increased excretion of glycerate; such analytical finding was considered correct and scored by 1 point. 15 participants detected also elevated excretion of oxalate, such analytical finding was also considered correct and scored by 1 point. The proficiency score for this sample was good (89%).

Interpretative proficiency and recommendation: Hyperoxaluria type 2 was considered the correct diagnosis. Confirmation of diagnosis by mutation analysis 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 very good proficiency score.

Sample B

Clinical picture provided with the sample: A 33 years old woman who presented with cachexia and malnutrition. The sample was obtained at the age of 34 years when the patient was receiving a non-specific therapy.

The sample was obtained from a patient with thymidine phosphorylase deficiency (MNGIE syndrome). The diagnosis was established by demonstrating enzyme deficiency in lymphocytes and completed by molecular genetic analysis. The sample was obtained from our repository.

Analytical performance: All participants analyzed organic acids, only 15 participants performed analysis of purines and pyrimidines. The presence of thymine and/or uracil only was considered a partially correct analytical result and scored by 1 point, elevated concentration of thymidine and/or 2'-deoxyuridine (observed by all participants analyzing purines and pyrimidines) was also a correct analytical result and scored by 1 point. The analytical performance was good (89%).

Interpretative proficiency and recommendation: Thymidine phosphorylase deficiency was considered a correct diagnosis. Confirmation of diagnosis by enzymatic assay and/or mutation analysis was considered helpful. The diagnosis of dihydropyrimidine dehydrogenase deficiency (thymine-uraciluria) was considered helpful but incomplete and scored with 1 point. The proficiency score for this sample was very good (95%).

Critical errors: No critical error for this sample.

Overall impression: Easy DPT sample with good total proficiency score.

Sample C

Clinical picture provided with the sample: This boy was referred at the age of 12 years. He had macrocephaly and motoric neurological symptoms. The sample was obtained at the age of 30 years when the patient was receiving specific treatment.

The sample was obtained from a man with glutaric aciduria type I. The diagnosis was confirmed by molecular genetic analysis. The sample was obtained from our repository.

Analytical performance: All participants analyzed organic acids, 17 of them reported elevated excretion of 3-hydroxyglutarate. Such analytical finding was considered correct result and scored by 2 point; failure to detect elevated 3-hydroxyglutarate is a critical error. In addition, elevated excretion of glutarate was observed by some participants; however, due to borderline concentrations in this low excretor this analyte was not scored and failure to detect elevated glutarate excretion was not considered a critical error. The analytical performance was good (89%).

Interpretative proficiency and recommendation: Glutaric aciduria type I was considered correct diagnosis. Confirmation of diagnosis by enzymatic assay and/or mutation analysis was considered helpful. The proficiency score for this sample was good (89%).

Critical errors: The failure to recognize abnormal excretion of 3-hydroxyglutarate in a patient with typical clinical picture is considered by the ERNDIM SAB as a critical error which would prevent establishing the correct diagnosis; critical error was assigned to two participants in our scheme.

Overall impression: Difficult DPT sample with suboptimal proficiency score obtained from a low glutarate excretor.

Sample D

Clinical picture provided with the sample: A 4 year old girl was repeatedly admitted with vomiting, severe metabolic acidosis and ketonuria. The sample was obtained at the age of 40 years when the patient was receiving specific treatment.

The sample was obtained from a woman with isovaleric acidemia, diagnosis was confirmed by molecular genetic analysis. The sample was obtained from our repository.

Analytical performance: All participants analyzed organic acids and reported elevated excretion of isovalerylglycine. Such analytical finding was considered a correct result and scored by 2 point. The analytical performance was excellent (100%).

Interpretative proficiency and recommendation: Isovaleric acidemia was considered correct diagnosis. Confirmation of diagnosis by enzymatic assay and/or mutation analysis was considered helpful. The proficiency score for this sample was excellent (100%).

Critical errors: **N**o critical error for this sample.
Overall impression: Easy DPT sample with an excellent proficiency score.

Sample E

Clinical picture provided with the sample: A 45 year old man with spinal stenosis, mild dysmorphia, semiflection position of hands and with sternum deformity. The sample was collected at the age of 45 years; patient did not receive any therapy.
The sample was obtained from a man with mucopolysaccharidosis type VI due to deficiency of arylsulfatase B, diagnosis was confirmed by molecular genetic analysis. The sample was obtained from our repository.
Analytical performance: Elevated excretion of glycosaminoglycans and an increased proportion of dermatan sulfate were considered a correct analytical result. Increased excretion of GAGs or increased proportion of dermatan sulfate only was scored as partially correct. Analytical performance was good (82) %.
Interpretative proficiency and recommendation: The diagnosis of mucopolysaccharidosis type VI was considered correct while suspicion for MPS (other types of MPS or non-specified MPS) was considered helpful but incomplete. Confirmation of diagnosis by enzyme assay of arylsulfatase B activity in fibroblasts/leucocytes and/or mutation analysis of ARSB gene were considered helpful. Recommendation to carry out analysis of GAG fractionation for those participants that did not perform this analysis was considered also helpful. The proficiency score for this sample was good (87%).
Critical errors: No critical error for this sample.
Overall impression: Typical DPT sample with good proficiency score.

Sample F

Clinical picture provided with the sample: A 4 years old girl with premature loss of primary teeth and waddling gait. The urine sample was collected at the age of 18 years.
The sample was obtained from a woman with hypophosphatasia, diagnosis was confirmed by molecular genetic analysis. The sample was acquired with help of VKS (the Dutch patient organization) and provided by the DPTC Netherlands.
Analytical performance: All participants analyzed amino acids and reported elevated excretion of phosphoethanolamine. Such analytical finding was considered correct result and scored by 2 points. The analytical performance was excellent (100%).
Interpretative proficiency and recommendation: Hypophosphatasia was considered the correct diagnosis. Confirmation of diagnosis by enzymatic assay, and/or PLP determination and/or mutation analysis of the *ALPL* gene were 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.

7. Scoring of results

Two criteria are evaluated: analytical and interpretative proficiency. The recommendations pertaining to further investigations are scored as a part of interpretative proficiency. The summary of scoring criteria is given below.

A	Analytical performance	Correct results of the appropriate tests	2
		Partially correct or non-standard methods	1
		Unsatisfactory or misleading (in some instances will be evaluated also as a critical error)	0
I	Interpretative proficiency	Good (diagnosis was established and appropriate further tests were recommended)	2
		Helpful but incomplete	1
		Misleading/wrong diagnosis (will be most likely evaluated also as a critical error)	0

The total score is calculated as a sum of these two criteria. The maximum that can be achieved is 4 points per sample, i.e. 12 points per survey and 24 points in 2016. Provisional scores assigned by the organizers were reviewed by an independent advisor from another DPT Centre and final scoring was approved by the ERNDIM Scientific Advisory Board on November 30, 2016.

Normal samples are usually not eligible for Critical Error. The main argument is that one cannot be absolutely certain that a sample is normal. The patient could, for example, have an IEM that we did not know at the time of analysis, but did result in subtle metabolite abnormalities that the majority of the participants were not aware of. However, 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 then this diagnosis could potentially result in treatment that is harmful for the patient and the findings could constitute a critical error. With effect from 2017, the SAB will determine critical errors on a case by case basis.

8. Score of participants for individual samples

Lab no	Sample A			Sample B			Sample C		
	A	I	T	A	I	T	A	I	T
1	2	2	**4**	2	2	**4**	2	2	**4**
2	2	2	**4**	2	2	**4**	2	2	**4**
3	2	2	**4**	1	2	**3**	2	2	**4**
4	2	2	**4**	2	2	**4**	2	2	**4**
5	2	2	**4**	2	2	**4**	2	2	**4**
6	2	2	**4**	2	2	**4**	2	2	**4**
7	2	2	**4**	2	2	**4**	2	2	**4**
8	1	2	**3**	2	2	**4**	2	2	**4**
9	1	2	**3**	2	2	**4**	2	2	**4**
10	2	2	**4**	2	2	**4**	2	2	**4**
11	2	2	**4**	2	2	**4**	2	2	**4**
12	1	2	**3**	1	1	**2**	2	2	**4**
13	2	2	**4**	1	2	**3**	2	2	**4**
14	1	2	**3**	2	2	**4**	2	2	**4**
15	2	2	**4**	1	1	**2**	2	2	**4**
16	2	2	**4**	1	2	**3**	2	2	**4**
17	2	2	**4**	2	2	**4**	0*	0	**0**
18	2	2	**4**	2	2	**4**	0*	0	**0**
19	2	2	**4**	2	2	**4**	2	2	**4**
20	-	-	**-**	-	-	**-**	-	-	**-**
Lab no	**Sample D**			**Sample E**			**Sample F**		
	A	**I**	**T**	**A**	**I**	**T**	**A**	**I**	**T**
1	2	2	**4**	2	2	**4**	2	2	**4**
2	2	2	**4**	2	2	**4**	2	2	**4**
3	2	2	**4**	1	2	**3**	2	2	**4**
4	2	2	**4**	2	2	**4**	2	2	**4**
5	2	2	**4**	1	1	**2**	2	2	**4**
6	2	2	**4**	2	2	**4**	2	2	**4**
7	2	2	**4**	2	2	**4**	2	2	**4**
8	2	2	**4**	1	2	**3**	2	2	**4**
9	2	2	**4**	2	2	**4**	2	2	**4**
10	2	2	**4**	2	2	**4**	2	2	**4**
11	2	2	**4**	2	2	**4**	2	2	**4**
12	2	2	**4**	0	1	**1**	2	2	**4**
13	2	2	**4**	1	1	**2**	2	2	**4**
14	2	2	**4**	2	2	**4**	2	2	**4**
15	2	2	**4**	2	2	**4**	2	2	**4**
16	2	2	**4**	2	2	**4**	2	2	**4**
17	2	2	**4**	1	1	**2**	2	2	**4**
18	2	2	**4**	2	1	**3**	2	2	**4**
19	2	2	**4**	2	2	**4**	2	2	**4**
20	-	-	**-**	-	-	**-**	-	-	**-**

A – Analytical score, I – Interpretative score, T – Total score

9. Total score of participants for individual surveys and their performance in 2016

Lab no	Survey 2065/1 [points]	Survey 2016/2 [points]	Total point 2016
1	12	12	**24**
2	12	12	**24**
3	11	11	**22**
4	12	12	**24**
5	12	10	**22**
6	12	12	**24**
7	12	12	**24**
8	11	11	**22**
9	11	12	**23**
10	12	12	**24**
11	12	12	**24**
12	9	9	**18**
13	11	10	**21**
14	11	12	**23**
15	10	12	**22**
16	11	12	**23**
17	8	10	**18***
18	8	11	**19***
19	12	12	**24**
20	-	-	**-**

* Critical error assigned to the participant

10. Score summary in 2016

Sample	Diagnosis	Analytical [%]	Interpretatative and recommendations [%]	Total [%]	Number of critical errors
A	***Hyperoxaluria type 2***	89	100	95	0
B	***MNGIE***	87	95	91	0
C	***Glutaric aciduria type 1***	89	89	89	2
D	***Isovaleric academia***	100	100	100	0
E	***MPS VI***	82	87	84	0
F	***Hypophosphatasia***	100	100	100	0

"Easy" and "difficult" samples were included in the surveys. The analytical performance was good for 5 samples; however there were 2 critical errors due to analytical mistakes. The interpretative performance was adequate for samples with adequate results of analytical investigations.

11. Satisfactory performance

The participants who obtained more than 14 points within the calendar year and did not receive "critical error" scoring are considered to be performing satisfactory. Seventeen laboratories returning the results achieved a satisfactory performance of more than 14 points without critical error. In 2 instances a serious mistake considered as a critical error has been observed in a total of two participating laboratories (although both laboratories achieved more than 14 points and would otherwise be considered as having adequate performance). Participants not achieving satisfactory performance will obtain a Performance Support letter in due course.

12. Annual meeting of the participants

The annual meeting of participants of the Proficiency Testing Centre Czech Republic took place during the ERNDIM Meeting 2016 in Rome on 6th September 2016, 15 participants from 11 laboratories were represented. The following items were discussed during the annual meeting of our DPT centre:

1. Information
    - ERNDIM is aiming at accrediting its activities
    - VK informed participants about news from Executive Committee and SAB
2. Tests required for to 2017
    - amino acids, organic acids, mucopolysaccharides, oligosaccharides and purines/pyrimidines
3. Discussion of results of samples A-F
    - scoring of 2016 results proposed by DPTC Czech Republic organizers has been subsequently evaluated by a second reviewer from an independent DPT center
    - Analytical difficulties in 2016 surveys
        - Interpretation of thymine and uracil in organic acid profile in sample B, some participants suggested that in MNGIE lactate may be possibly elevated in contrast to normal lactate in DHPD deficiency
        - Low excretion of glutaric acid in sample C, participants agreed that lack of detection of 3-OH-glutarate would be a critical error
4. Varia
    - Participants requested instructions on heat-treatment preparation as a separate file
    - Participants requested a guidance from ERNDIM on recommended/standardized methods to analyze GAGs

13. Tentative schedule of DPT scheme and fee in 2017

Sample distribution	February 6, Monday
Start of analysis of Survey 2017/1	February 20, Monday
Survey 2017/1 – results submission	March 13, Monday
Survey 2017/1 – report	May 29, Monday
Start of analysis of Survey 2017/2	May 22, Monday
Survey 2017/2 – results submission	June 12, Monday
Survey 2017/2 – report	August 21, Monday
Annual meeting of participants-tentatively Manchester	Tentatively November 21-22
Annual report 2017	December 2017

Since there will be no SSIEM Symposium in 2017 (and ICIEM Symposium will take place in the Brazil) the next Annual Meeting of the DPT Center Czechia will be tentatively organized in Manchester on November 21-22, 2017, the date and location will be confirmed by ERNDIM in due course.

The Executive Board and Board of Trustees of ERNDIM determined the DPT fee for 2017 in the amount of 416 €.

14. Certificate of participation and performance in Proficiency Testing for 2016

Results of DPT Scheme are included in the Certificate of participation and performance, which are issued by ERNDIM.

Prague, February 23, 2017

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