ERNDIM DPT Center Prague

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Proficiency Testing Centre Czech Republic
Annual Report 2013

1. Introduction

In 2013 proficiency testing in our centre was running as a regular ERNDIM scheme.

2. Geographical distribution of participants

Twenty laboratories from 15 countries have participated in our Diagnostic Proficiency Testing scheme in 2013, for details see the below table:

Country	Number of participants
Austria	1
Croatia	1
Cyprus	1
Czech Republic	1
Denmark	1
Finland	1
Germany	4
Greece	1
India	1
Latvia	1
Malaysia	1
Poland	1
Portugal	1
Slovakia	3
UK	1
in total	**20**

3. Logistics of the scheme

- ✓ Two surveys: 2013/1 – samples A, B and C
  2013/2 – samples D, E and F
  Origin of samples: Five urines obtained from patients with known diagnoses (samples were provided by the DPTC participants and by the organizers) + a common sample from DPTC Czech republic (distributed in all five DPT schemes).
- ✓ The samples with addition of thiomersal have been heat-treated and were re-analyzed in our Institute after receiving the samples from CSCQ that were shipped via courier 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 undergoing this treatment.
- ✓ This year the samples for the 2013 Diagnostic Proficiency Testing scheme were distributed via CSCQ in Geneva. On 15th April 2013 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 being used: Thiomersal 100 mg/l of urine is added and urine is heated at 56 °C for one hour in water bath (this temperature is checked in urinary sample and not only in the water bath). The urinary samples have been frozen until shipment.
- ✓ Tests required in 2013: amino acids, organic acids, mucopolysaccharides, oligosaccharides and purines/pyrimidines

4. Schedule of the scheme in 2013

Sample distribution	April 15, Monday
Start of analysis of Survey 2013/1	April 29, Monday
Survey 2013/1 – results submission	May 17, Friday
Survey 2013/1 – report	July 8, Monday
Start of analysis of Survey 2013/2	June 10, Monday
Survey 2013/2 – results submission	June 28, Friday
Survey 2013/2 – report	August 23, Friday
Annual meeting of participants	September 3, Tuesday
Annual report 2013	April 2014

5. The receipt of samples and results

Date of receipt of samples (samples sent on April 15, 2013)

date of receipt (reported by participants)	number of participants	date of receipt (reported by courier service)	number of participants
1 day	8	1 days	14
2 days	2	2 days	4
3 days	2	3 days	2
4 days	2		
7 days	1		
8 days	1		
12 days	1	-	-
not indicated	3	-	-

Submission of results

	2013/1	2013/2
in time	19	17

6. Samples

Sample A

The sample was obtained from a 14-year old boy with thymidine phosphorylase deficiency (MNGIE syndrome). The diagnosis was established by demonstrating enzyme deficiency in lymphocytes and completed by molecular analysis. The sample was obtained from our repository.

Analytical performance: All participants performed analysis of organic acids, but only 14 participants performed analysis of purines and pyrimidines. The presence of thymine and/or uracil only was considered a partially correct analytical result. All labs, which have analyzed purines and pyrimidines, reported elevated concentration of thymidine and/or 2'-deoxyuridine, and of thymine and/or uracil, such analytical finding was also considered correct and scored by 2 points. The analytical performance was slightly suboptimal (79%).

Interpretative proficiency and recommendation: Thymidine phosphorylase deficiency was considered correct diagnosis. Confirmation of diagnosis by enzymatic assay and/or mutation

analysis was considered helpful. The proficiency score for this sample was slightly suboptimal (79%).
Tentative critical errors: The failure to detect any of the following analytes: thymine, uracil, thymidine or 2'-deoxyuridine would be considered a critical error that would prevent establishment a correct diagnosis.
Overall impression: Typical DPT sample with slightly suboptimal proficiency score.

Sample B
Patient: This sample came from a 36 years old woman with very long-chain acyl-CoA dehydrogenase (VLCAD) deficiency. The diagnosis was confirmed by enzymatic analysis. This sample was contributed by Dr. Wanda Gradowska from the Children´s Memorial Health Institute in Warsaw.
Analytical performance: All participants performed analysis of organic acids. Elevated excretion of saturated and unsaturated medium-chain dicarboxylic acids was considered correct analytical results. Many laboratories also reported absence of glycine conjugates of dicarboxylic acids. The analytical performance was very good (95%).
Interpretative proficiency and recommendation: The diagnosis of VLCAD deficiency or fatty acids oxidation disorders were considered correct. Diagnosis of long-chain 3-hydroxyacyl-CoA dehydrogenase/mitochondrial trifunctional protein deficiency was incorrect as 3-hydroxydicarboxylic acids were not present in the sample; similarly MCAD deficiency was considered incorrect as the urine did not contain appreciable amounts of glycine conjugates of dicarboxylic acids. Confirmation of diagnosis by enzymatic assay and/or mutation analysis was considered helpful. The proficiency score of 47% was below the usual performance of our group.
Tentative critical errors: The failure to detect any dicarboxylic aciduria would be considered a critical error leading to an incorrect diagnosis.
Overall impression: Moderately difficult DPT sample with suboptimal proficiency score.

Sample C
Patient: This sample came from an 8 years old boy with α-mannosidosis due to α-mannosidase deficiency. The diagnosis was confirmed by enzymatic analysis. This sample was contributed by Dr. Elena Gregová from the F. D. Roosevelt Hospital in Banská Bystrica.
Analytical performance: 16 participants performed analysis of OLS. The pattern of OLS characteristic for α-mannosidosis was considered a correct analytical finding. Abnormal OLS pattern without specified diagnosis was considered partially correct. The analytical performance was slightly suboptimal (74%).
Interpretative proficiency and recommendation: The diagnosis of α-mannosidosis (α-mannosidase deficiency) was considered correct. Fourteen laboratories reached correct diagnosis. Confirmation of diagnosis by enzyme assay of α-mannosidase activity preferably in plasma/fibroblasts/leucocytes and/or mutation analysis of *MAN2B1* gene were considered helpful. Recommendation to carry out analysis of OLS for those participants that did not perform OLS analysis was considered also helpful. The proficiency score for this sample was slightly suboptimal (72%).
Tentative critical errors: The failure to perform and/or recommend OLS analysis, or failure to recognize abnormal OLS pattern would be considered a critical error which would prevent establishing the correct diagnosis.
Overall impression: Typical DPT sample with suboptimal proficiency score.

Sample D (common sample)
The sample was obtained from a 17-years old boy with lysinuric protein intolerance. This sample was contributed by Dr. Jeannette Klein from Charité-Campus Virchow – Klinikum in Berlin.
Analytical performance: The presence of dibasic hyperaminoaciduria and simultaneously of orotic aciduria were considered a correct analytical result and scored by 2 point. All 17 participants detected dibasic hyperaminoaciduria but one participant missed to report elevated excretion of orotic acid. The analytical performance was very good (97%).

Interpretative proficiency and recommendation: Lysinuric protein intolerance was considered the correct diagnosis. Confirmation of diagnosis by mutation analysis was considered helpful. The proficiency score for this sample was good (88%).
Tentative critical errors: The failure to detect orotic acid would be a critical error leading to an incorrect diagnosis.
Overall impression: Typical DPT sample with good proficiency score.

Sample E
Patient: This urinary sample was obtained from a patient without any known inborn error of metabolism who suffered from diabetes mellitus type 1. Extensive metabolic screening including plasma and urinary amino acids, organic acids, purines and pyrimidines, galactitol and plasma carnitine did not reveal any specific abnormality. The sample was obtained before the beginning of diabetes-specific therapy. The sample was taken from our repository.
Analytical performance: All participants performed analysis of amino acids. 16 participants observed hyperaminoaciduria, such analytical finding was considered correct and scored by 1 point. All participants detected elevated excretion of glucose, such analytical finding was also considered correct and scored by 1 point. The analytical performance was very good (94%).
Interpretative proficiency and recommendation: Scoring of diagnoses was quite difficult due to large variability of conclusions, we considered the report of "no IEM", non-specific finding or diabetes mellitus a good diagnosis. The diagnosis of non-ketotic hyperglycinemia was scored with 0 points. The diagnosis of Fanconi syndrome, Fanconi-Bickel syndrome or mitochondrial disorder was scored with 1 point. The proficiency score for this sample was slightly suboptimal (71%).
Tentative critical errors: No submitted data met the criteria of tentative critical error.
Overall impression: Moderately difficult DPT sample with good proficiency score.

Sample F
Patient: The sample was obtained from a 3 years old boy suffering from mucopolysaccharidosis type VI due to deficiency of arylsulfatase B. The diagnosis was confirmed by enzymatic analysis. This sample was contributed by the Dr. Darina Behulova, Department of Clinical Biochemistry from University Children's Hospital in Bratislava.
Analytical performance: Elevated excretion of glycosaminoglycans and increased proportion of dermatan sulphate were considered a correct analytical result. Increased excretion of GAGs without report on dermatan sulphate elevation was scored as partially correct. Analytical performance was slightly suboptimal (79) %.
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. Seven laboratories reached correct diagnosis. 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 suboptimal (68%).
Tentative critical errors: The failure to perform and/or recommend GAG analysis would be considered a critical error which would prevent establishing the correct diagnosis.
Overall impression: Typical DPT sample with slightly suboptimal proficiency score.

7. Scoring of results

ERNDIM are being encouraged by the European Society of Human Genetics to harmonise scheme performance assessments with the other European genetic laboratory EQA providers. The principal points of difference lie with the allocation of points in the scoring systems and the adoption of the concept of 'critical errors'. 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. We have provisionally indicated critical error for the DPT scheme 2013 (presence of a critical errors was not

scored as unsatisfactory performance), critical errors will be evaluated as a part of performance assessment in 2014 DPT schemes.
This year only two criteria were evaluated: analytical and interpretative proficiency. The recommendations pertaining to further investigations were 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 three criteria. The maximum that can be achieved is 4 points per sample, i.e. 12 points per survey and 24 points in 2013. There is a new procedure for scoring DPT Scheme; scores assigned by organizer and agreed at the Annual Meeting have been reviewed by independent advisor from another DPT Centre and scoring is finalized after any possible discrepancies had been resolved at the March 2014 ERNDIM Scientific Advisory Board meeting.

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

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

9. Total score of participants for individual surveys and their performance in 2013

Lab no	Survey 2013/1 [points]	Survey 2013/2 [points]	Total point 2013
1	11	11	**22**
2	12	12	**24**
3	12	9	**21**
4	8	5	**13**
5	12	12	**24**
6	10	9	**19**
7	10	10	**20**
8	10	9	**19**
9	8	12	**20**
10	10	11	**21**
11	10	10	**20**
12	8	10	**18**
13	11	10	**21**
14	3	7	**10**
15	10	11	**21**
16	10	11	**21**
17	0	0	**0**
18	12	10	**22**
19	0	0	**0**
20	2	0	**2**

10. Score summary in 2013

Sample	Diagnosis	Analytical [%]	Interpretatative [%]	Total [%]	Number of tentative critical errors
A	***MNGIE syndrome***	79	79	79	2
B	***VLCAD deficiency***	95	47	71	1
C	***α-mannosidosis***	74	74	72	4
D	***Lysinuric protein intolerance***	97	88	93	1
E	***Diabetes mellitus type 1***	94	71	82	0
F	***MPS type VI***	79	68	74	1

"Easy" and "difficult" samples were included in the surveys. The analytical performance was good to very good for most diagnoses. The interpretative performance was slightly suboptimal for most diagnoses.

11. Satisfactory performance

The participants who obtained more than 14 points within the calendar year are considered to achieve satisfactory performance. Fifteen laboratories returning the results achieved a satisfactory performance of more than 14 points while four laboratories did not reach this threshold. In 9 instances a serious mistake considered tentatively as a critical error has been observed in a total of five participating laboratories.

12. Annual meeting of the participants

The annual meeting of participants of the Proficiency Testing Centre Czech Republic took place during the ERNDIM Meeting 2013 in Barcelona on 3rd September 2013, six 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
   - changes in DPT (sample recruitment and distribution, web based system at CSCQ)
   - SAB is developing a new concept (similar to other genetic disciplines) of the critical error for evaluation of performance; participants will be informed in advance about this changes
2. Tests required for to 2014
   - amino acids, organic acids, mucopolysaccharides, oligosaccharides and purines/pyrimidines
3. Discussion of results of samples A-F
   - scoring of 2013 results proposed by DPTC Czech Republic organizers has been subsequently evaluated by a second reviewer from an independent DPT center

13. Tentative schedule of DPT scheme and fee in 2014

Sample distribution	March 31, Monday
Start of analysis of Survey 2014/1	April 7, Monday
Survey 2014/1 – results submission deadline	April 25, Friday
Survey 2014/1 – report	May 30, Friday
Start of analysis of Survey 2014/2	June 9, Monday
Survey 2014/2 – results submission deadline	June 27, Friday
Survey 2014/2 – report	August 15, Friday
Annual meeting of participants	September 2, Tuesday
Annual report 2014	April 2015

The annual meeting of participants will take place on September 2nd during the SSIEM Annual Symposium in Innsbruck, Austria.

The Executive Board and Board of Trustees of ERNDIM determined the DPT fee for 2014 in the amount of 353 €.

14. Certificate of participation and performance in Proficiency Testing for 2013

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

Prague, March 24, 2013

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