P-34

Application of metabolomics in differential diagnosis of amino aciduria

Chunhua Zhang[1) 2)], Xuyang Wang[1)], Zonghai Ruan[1)], Mingji Jin[1)], Zhao Ning[1)], Li Fengyun[3)] and Usha Dave[4)]

1) MILS International, Japan
2) Peking Union Medical College Hospital, China
3) MILS(Beijing) Medical laboratory, China
4) MILS International India, India

Background and Purpose

Some types of amino aciduria are caused by various amino acid metabolism disorders leading to elevated blood amino acids, while other types caused by impaired absorption and transport of amino acids in the kidney or intestine leading to increased excretion of amino acids in urine without amino acidemia.

Hence, the precise diagnosis of various amino aciduria mainly relies on the analysis results of blood and urine amino acids and genetic testing. However, the amino acid panels of blood and urine amino acid testing in clinical routine laboratory testing is not comprehensive as some amino acids are not included. Also, gene mutation testing often show VUS results, which makes clinical diagnosis difficult.

We have used blood and urine metabolomics analysis to screen patients with clinically suspected IEM from 2005, and found that the metabolomics expression of more than 10 types of amino aciduria has specific biomarkers and metabolic profile characteristics, which can effectively distinguish diagnosis.

Method

From March 2005 to February 2025, we used a dual mass spectrometry analysis method consisting of urine GCMS metabolomics and blood LCMSMS amino acid analysis to screen 350610 clinically IEM suspected patients from Southeast Asian countries. Based on data analysis, investigated specific amino acid biomarkers corresponding to the following 11 diseases.

Disease	Urine Biomarkers
Sarcosinemia	Sarcosine without other abnormality
Argininosuccinic aciduria (ASA)	Argininosuccinic acid, with/without uracil, orotate
HHH syndrome	Ornithine, uracil, orotate with Homo-Cit
Lysinuric protein intolerance(LPI)	Lysine, Ornithine, uracil, orotate with blood Arg low
Cystinuria	Lysine, Ornithine, Cystine without orotate
Hartnup disease (HND)	Neutral amino acids (valine, serine, phenylalanine, histidine, glutamine, leucine, asparagine, citrulline, isoleucine, threonine, alanine, tyrosine, tryptophan) without proline elevate
Fanconi syndrome	General amino aciduria with high level of glucose, phosphate and uric acid
Prolidase deficiency	glycylproline
Aminoacylase-1 deficiency(ACY1D)	N-acetylated amino acids: Acetylvaline, Acetylglutamine, Acetylalanine, Acetylglycine, Acetylserine
Canavan disease (ACY2D)	N-acetylaspartic acid (NAA)
Pyroglutamic aciduria	Pyroglutamic acid

Results

No	amino aciduria disease	cases
1	Sarcosinemia # 268900	13
2	argininosuccinic aciduria (ASA) # 207900	7
3	HHH syndrome # 238970	5
4	lysinuric protein intolerance (LPI) # 222700	8
5	cystinuria# 220100	1
6	Hartnup disease (HND) # 234500	3
7	Fanconi syndrome# 227650	2
8	prolidase deficiency # 170100	2
9	aminoacylase-1 deficiency(**ACY1D**)# 609924	3
10	Canavan disease (**ACY2D**)# 271900	25
11	pyroglutamic aciduria# 260005 # 266130	22
Total	**11 diseases**	**91**

8 LPI patient clinical information

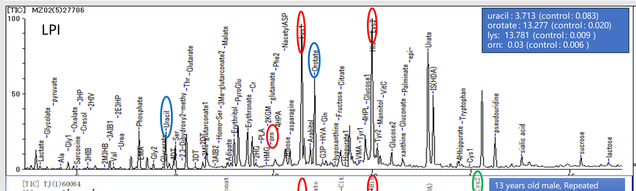
JP: Japanese 2; CH: Chinese 6; Male 5 : Female 3 ; Age 3months ~ 8 years

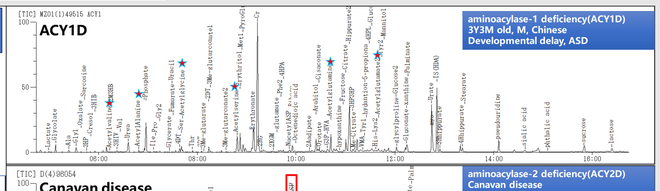
Analysis No.	Patient	Sex	Age	Diagnosed	Clinical note	Blood AA	Genetic result
Hoka2199-2023	I K(JP)	M	2Y10M	2023/6/13	hyperammoniemia, UCD?	Arg low	SLC7A7 deficiency
Hoka2210-2023	**YK (JP)**	M	1Y	2023/7/5	hyperammoniemia, hepatomegaly, developmental delay, blood Arg decreased	Arg low 17 (40-90)	SLC7A7 deficiency
MZ02-27843	ZYT (sisitr) (CN)	F	3Y10M13D	2022/7/19	Autoimmune disease?	Arg low 8.36	SLC7A7 c.625+1G>A(splicing) Pathogenic homo
MZ02-27706	ZYX (yonger sister) (CN)	F	1Y5M26D	2022/7/6	Fever, cough, blue spot, abnormal liver function, Vasculitis and skin abnormalities, Autoimmune disease?	Arg low 3.96	SLC7A7 c.625+1G>A(splicing) Pathogenic homozygous
D86253	DYC(CN)	M	0Y4M13D	2020/12/7	Feeding difficulties, abnormal liver function, hepatomegaly, Difficulty breathing, sepsis, poor growth, recurrent vomiting, yellowish fur, Genetic test result LPI suspected	Arg low	SLC7A7 del/c.724T>C, like Pathogenic/VUS heterozygous
MZ01-20431	WXJ(CN)	F	8Y7M9D	2019/5/16	language retardation, developmental delay, abnormal liver function, hepatomegaly, Splenomegaly and abnormal renal function, hyperammoniemia, hypoimmunity.	Arg low 8.45	SLC7A7 heterozygous
MZ96-00003	FYR(CN)	M	0Y3M1D	2024/12/10	Recurrent pneumonia in the neonatal period, abnormal liver function, blood Cit, C3DC high, MMA and PPA suspected	Cit, C3DC high	SLC7A7 like Pathogenic/LP
D104026	LHR(CN)2nd	M	3Y8M14D	2025/3/17	hyperammoniemia, abnormal liver function	Gly high	SLC7A7 C235 G>A/ c.625+1G>A LP/LP?
D92205	LHR(CN)1st	M	0Y10M0D	2022/5/24	Genetic result LP, confirm test		

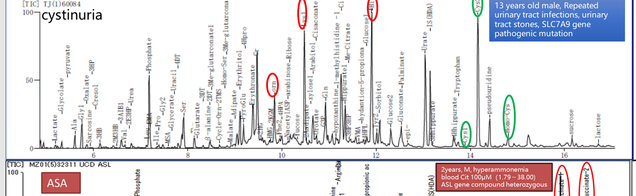
25 Canavan disease patient clinical information

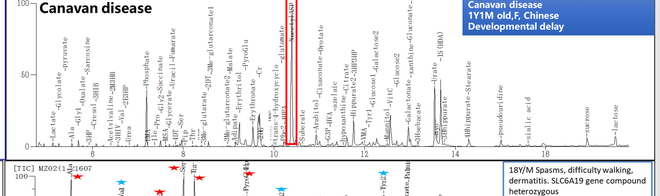
Age range 2months-11yearsF13:M12,Chinese 24, Indian 1

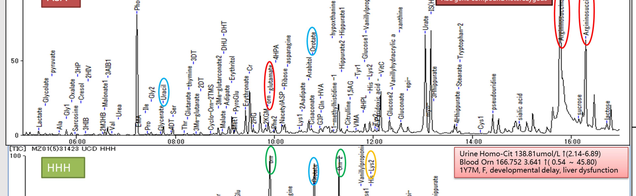
Case No	Data No	Sex	diagnosed age	Clinical note
1	D2819	F	4M	IEM suspected
2	D10659	M	11Y11M	IEM suspected
3	D12842	F	6M	IEM suspected
4	D15399	F	11M	IEM suspected
5	D24148	M	0Y2M26D	IEM suspected
6	D24247	M	0Y3M3D	Abnormal muscle tone, epilepsy, cerebral injury
7	D60936	F	1Y11M9D	developmental delay
8	D63298	F	0Y6M1D	IEM suspected
9	D63422	M	0Y4M3D	IEM suspected
10	D67765	F	0Y5M1D	developmental delay, mental retardation, MRI abnormal
11	D96054	F	1Y1M24D	developmental delay
12	BCH18330	M	1Y1M23D	developmental delay, mental retardation
13	MZ01-07403	M	4M4D	developmental delay, High muscle tension
14	MZ01-25326	F	0Y5M3D	developmental delay, Nystagmus
15	MZ02-06402	M	5M29D	developmental delay, mental retardation, MRI abnormal
16	MZ02-12529	M	7M10D	developmental delay
17	BCH8791	F	7M	IEM suspected
18	BCH15868	F	4M	IEM suspected
19	BCH23937	F	1Y7M	IEM suspected
20	BCH55801	M	4Y5M	IEM suspected
21	CN7191	M	2Y2M	IEM suspected
22	CN10073	F	11M	IEM suspected
23	Me-1741	F	8Y	IEM suspected
24	Me-1936	M	9M	IEM suspected
25	PLC-10211	M	7M30D	IEM suspected

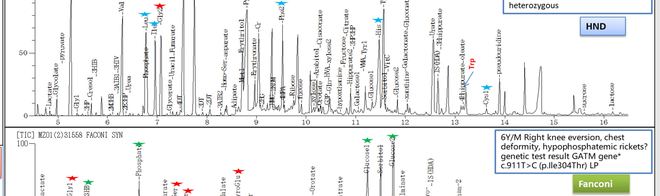


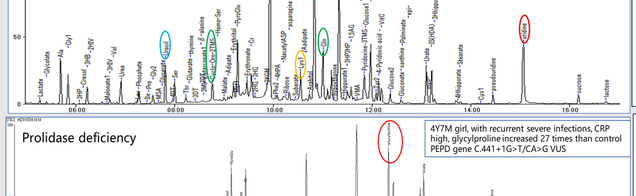


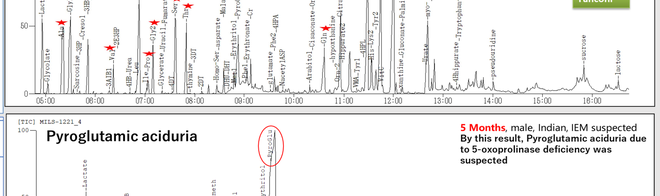


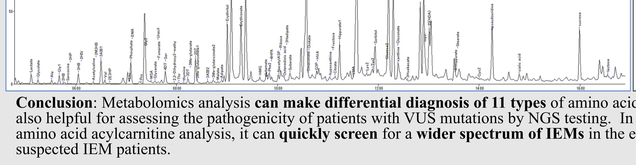


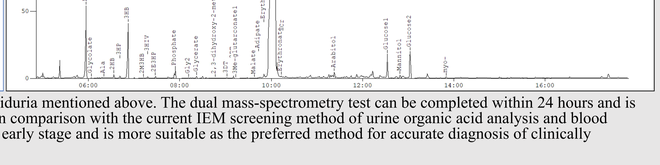












Conclusion: Metabolomics analysis **can make differential diagnosis of 11 types** of amino aciduria mentioned above. The dual mass-spectrometry test can be completed within 24 hours and is also helpful for assessing the pathogenicity of patients with VUS mutations by NGS testing. In comparison with the current IEM screening method of urine organic acid analysis and blood amino acid acylcarnitine analysis, it can **quickly screen** for a **wider spectrum of IEMs** in the early stage and is more suitable as the preferred method for accurate diagnosis of clinically suspected IEM patients.