

Screening neonatale allargato

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Meyer**



Topics

-Expanded Newborn Screening, today

-Future Perspectives: LSDs and SCIDs



Mass Spectrometry, Clinical
Chemistry, Farmacology Lab.

Newborn Screening Center

Metabolic Unit, Meyer Children's
Hospital

CLINICAL LAB.



**Interdepartmental Mass Spectrometry
Center,**

University of Florence

<http://www.cism.unifi.it/>

RESEARCH LAB.



MEYER CHILDREN'S HOSPITAL NEWBORN SCREENING STAFF





Newborn Screening

It identifies biochemical or other inherited conditions that may produce mental retardation, other disabilities and/or death.

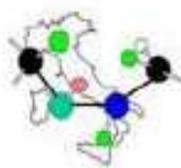
Babies are screened for these conditions during the newborn period.

These conditions are identified using tests on blood collected from a heel stick onto filter paper

Criteria of a screening programme

- **The condition being screened for should be an important health problem**
- **The natural history of the condition should be well understood**
- **There should be a detectable early stage**
- **Treatment at an early stage should be of more benefit than at a later stage**
- **A suitable test should be devised for the early stage**
- **The test should be acceptable**
- **Intervals for repeating the test should be determined**
- **Adequate health service provision should be made for the extra clinical workload resulting from screening**
- **The risks, both physical and psychological, should be less than the benefits**
- **The costs should be balanced against the benefits**

Wilson-Jungner criteria; World Health Organisation 1968



SISMME



SISN

**Società Italiana Studio Malattie
Metaboliche Ereditarie**

Società Italiana Screenings Neonatali

**LINEE GUIDA PER LO SCREENING
NEONATALE ESTESO E LA CONFERMA
DIAGNOSTICA
2008**

DOCUMENTO REDATTO A CURA DELLA COMMISSIONE AD HOC DELLA SISMME E SISN

APPROVATO DAI DIRETTIVI SISMME E SISN

APPROVATO DAI PRESIDENTI DELLA SISMME E SISN , Proff. A. Burlina e R. Cerone

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Dr. Giancarlo La Marca, Università di Firenze

Dr. Elisabetta Pasquini, Università di Firenze

SOME PRINCIPLES OF ITALIAN GUIDELINES FOR NEWBORN SCREENING

1: Lo screening neonatale è oggi considerato una *responsabilità essenziale del sistema di salute pubblica* ed è ritenuto di importanza critica per migliorare la salute dei bambini affetti.

“ Today newborn screening is considered to be a main responsibility of the public health system. It is fundamental to improve the health of affected babies”.

2: La politica di sviluppo dello screening neonatale viene oggi considerata *primariamente diretta all'interesse dei neonati affetti* mentre sono secondari gli interessi dei neonati sani, delle famiglie, degli operatori sanitari e del pubblico.

3: Le raccomandazioni per l'implementazione dello screening neonatale possono oggi essere esclusivamente *basate sulla evidenza scientifica e sull'opinione degli esperti*.

4: Per essere inclusa nel pannello di screening secondo i criteri odierni una condizione deve: essere identificabile in epoca preclinica, possibilmente entro le 48 ore di vita, deve essere disponibile un test sensibile e specifico e debbono essere evidenti i benefici di un precoce intervento, che non si limitano al trattamento efficace ma che consistono nel miglioramento della qualità della vita anche in presenza di sintomi clinici e nel consiglio genetico alla famiglia.

“ ... early detection is necessary and it should be, possibly, before the 48 hours of life; there should be a simple, safe, precise and validated screening test; the benefits of an early detection have to be intended not only to get a resolute treatment but also to improve the quality of life (even if clinical symptoms are present) and to give genetic counseling to the family”.

5. Il programma di screening deve *comunque riportare ogni altro eventuale rilievo di potenziale significato clinico*.

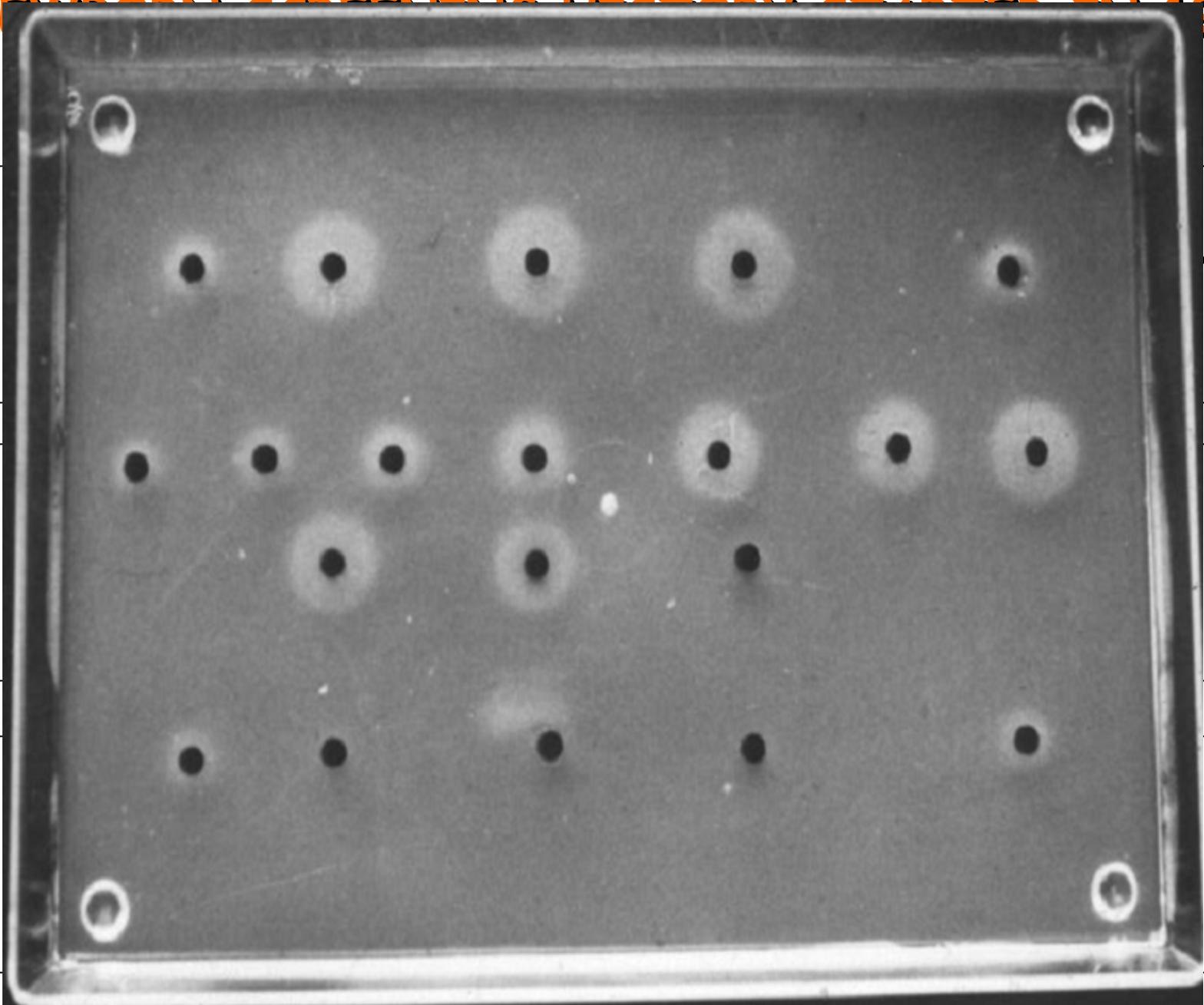
*** INCREMENT OF LIFE EXPECTANCY**

*** IMPROVEMENT OF THE QUALITY OF LIFE**

*** POSSIBILITY OF SUITABLE GENETIC COUNSELLING**

*** PRENATAL DIAGNOSIS**

NUMBER OF ZONAL PATTERNS OF DISK #1961:



NURIA

Clinica Pediatrica Università di Firenze

**Organizzazione del servizio di Screening
per la diagnosi delle encefalopatie dismetaboliche
congenite sulle urine dei neonati del comune di Firenze ***

S. BORGHERESI - E. ZAMMARCHI

Estratto da:

SIMPOSIO SU PROBLEMI ENDOCRINO-METABOLICI IN PEDIATRIA

Milano - 18-19-20 Ottobre 1973

Estratto da:

RIVISTA DI NEUROBIOLOGIA

Volume XXVII - fasc. 3-4 1981

CLINICA PEDIATRICA I - UNIVERSITÀ DI FIRENZE

E. ZAMMARCHI, M. R. BARDINI, M. A. DONATI e A. SAVELLI

LO SCREENING PER LE AMINOACIDOPATIE NEI NEONATI
DEL COMUNE DI FIRENZE E COMUNI LIMITROFI NEGLI ANNI
1973-1980

MODELLO ORGANIZZATIVO

DELIBERAZIONE REGIONALE 301 DEL 2/6/1981
"AVVIO DI ATTIVITÀ PER L'ACCERTAMENTO
DELL' IPOTIROIDISMO E FENILCHETONURIA..."

ISTITUZIONE DELLO SCREENING FIBROSI CISTICA

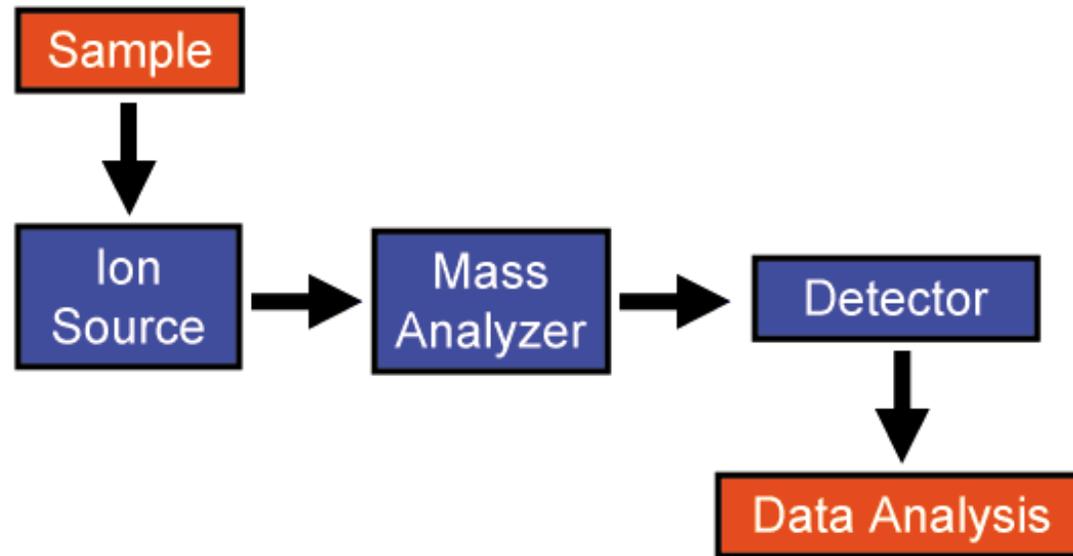
ITALIAN LAW N. 104/1992

...nei primi giorni di vita, ancora in ospedale, il bimbo viene sottoposto al cosiddetto "screening neonatale", una serie di esami che permettono di individuare precocemente alcune malattie congenite (cioè presenti alla nascita), ma che si manifestano in genere più tardivamente. Grazie a questo test, che deve essere eseguito dopo quarantotto ore di vita, è possibile individuare e curare precocemente queste malattie, che possono, altrimenti, avere gravi conseguenze sullo sviluppo psicomotorio e sull'accrescimento del bambino. Dal 1992 (legge-quadro n. 104 del 5-5-1992) questo esame deve essere eseguito su tutti i neonati italiani (la prima legge che ne ha sancito l'importanza e quella della regione Liguria del 17-8-1973).

*Lo "screening neonatale" viene effettuato per identificare alcuni disturbi molto seri, che se vengono individuati precocemente possono essere curati con ottimi risultati. Queste malattie sono congenite, presenti cioè già dalla nascita, ma nei primi giorni di vita non si manifestano e, se non viene eseguito il test, possono essere individuate solo più tardi. **I disturbi individuabili con questo esame sono tre: la fenilchetonuria, una malattia ereditaria che provoca problemi nell'assimilazione di una sostanza, la fenilalanina (monitorando il dosaggio di quest'ultima); l'ipotiroidismo congenito, un problema della tiroide, la ghiandola che regola lo sviluppo e la crescita (in base al dosaggio del TSH o ormone tireotropo) e la fibrosi cistica, una malattia respiratoria molto seria (verificata tramite la concentrazione di un enzima la tripsina).***

IN ITALY THE SCREENING IS MANDATORY ONLY for PKU, CHT, CF

SCREENING NEWBORN USING LC-MS/MS (1990-3)



Short Communication

Tandem Mass Spectrometry: A New Method for Acylcarnitine Profiling with Potential for Neonatal Screening for Inborn Errors of Metabolism

D. S. MILLINGTON, N. KODO, D. L. NORWOOD and C. R. ROE
Division of Pediatric Genetics and Metabolism, Box 3028, Duke University Medical Center, Durham, NC 27710, USA
J. Inher. Metab. Dis. 13: 321-324; 1990

La spettrometria di massa



Che cosa è?



Quali sono le sue applicazioni?



Quali sono i vantaggi?

Che cosa è?

E' una tecnica di analisi e

di rivelazione

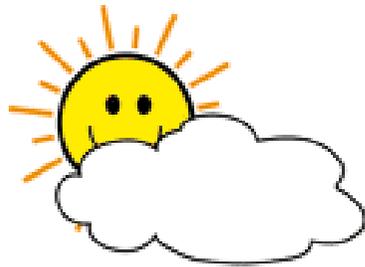
quali-quantitativa delle molecole (ioni)

Principali campi di applicazioni

- **Chimica farmaceutica e farmacologia:** caratterizzazione di farmaci e loro impurezze, metabolismo, farmacocinetica e farmacodinamica, etc.
- **Biochimica clinica:** determinazione quantitativa di sostanze endogene ed esogene
- **Biotechnologie:** monitoraggio on-line di processi di fermentazione, caratterizzazione dei relativi prodotti etc.
- **Chimica agroalimentare:** analisi di alimenti, di componenti residui, caratterizzazione e determinazione di additivi legali ed illegali
- **Chimica ambientale:** identificazione, caratterizzazione e determinazione quantitativa di inquinanti idrici polari, di pesticidi, tensioattivi etc., e/o di loro metaboliti e prodotti di degradazione.
- **Chimica Organica e Organometallica:** caratterizzazione strutturale di prodotti e intermedi di sintesi, controllo dei prodotti di partenza etc.

VANTAGGI

- VERSATILITA'
- VELOCITA'
- SENSIBILITA'
- SELETTIVITA'



SVANTAGGI

- NON FACILE UTILIZZO
- COSTI ELEVATI

Quantità da rilevare	Peso (g)	Numero di molecole	Tecnica analitica chimico-fisica
Milligrammo (mg)	10^{-3}	10^{18}	Titolazione, NMR
Microgrammo (μg)	10^{-6}	10^{15}	Cromatografie + Spettrofotometria
Nanogrammo (ng)	10^{-9}	10^{12}	
Picogrammo (pg)	10^{-12}	10^9	MS
Femtogrammo (fg)	10^{-15}	10^6	MS
Attogrammo (ag)	10^{-18}	10^3	MS
Zeptogrammo (zg)	10^{-21}	10^0	?

NBS (Test di Guthrie)
(Un Test-Un Difetto)



NBS mediante MS/MS
(Multiplex Testing)

PILOT PROJECT



01/10/2001 - 31/10/2004

in

Florence, Prato and Pistoia areas

42371 screened

Regional Legislative Action no. 800 (3/8/2004):

since 01/11/2004

“..... the program must screen all babies born in Tuscany starting from **November 2004** (approximately 35000/year) for selected acylcarnitines and amino acids....



... ”

35000
newborns/year

since 01/01/2010

Florence has performed the expanded newborn screening also for Umbria Region



8000
newborns/year

PILOT PROJECT

01/01/2002-31/10/2004

3 provinces of Tuscany = 13,500/year

42,371 newborns

REGIONAL LEGISLATIVE ACTION No. 800 (3/8/2004)

... the program must screen all babies born in Tuscany starting from **November 2004** (approximately 40000/year) for selected acylcarnitines and amino acids...

293,900 newborns

PREVALENCE 1:1750

166 DIAGNOSES



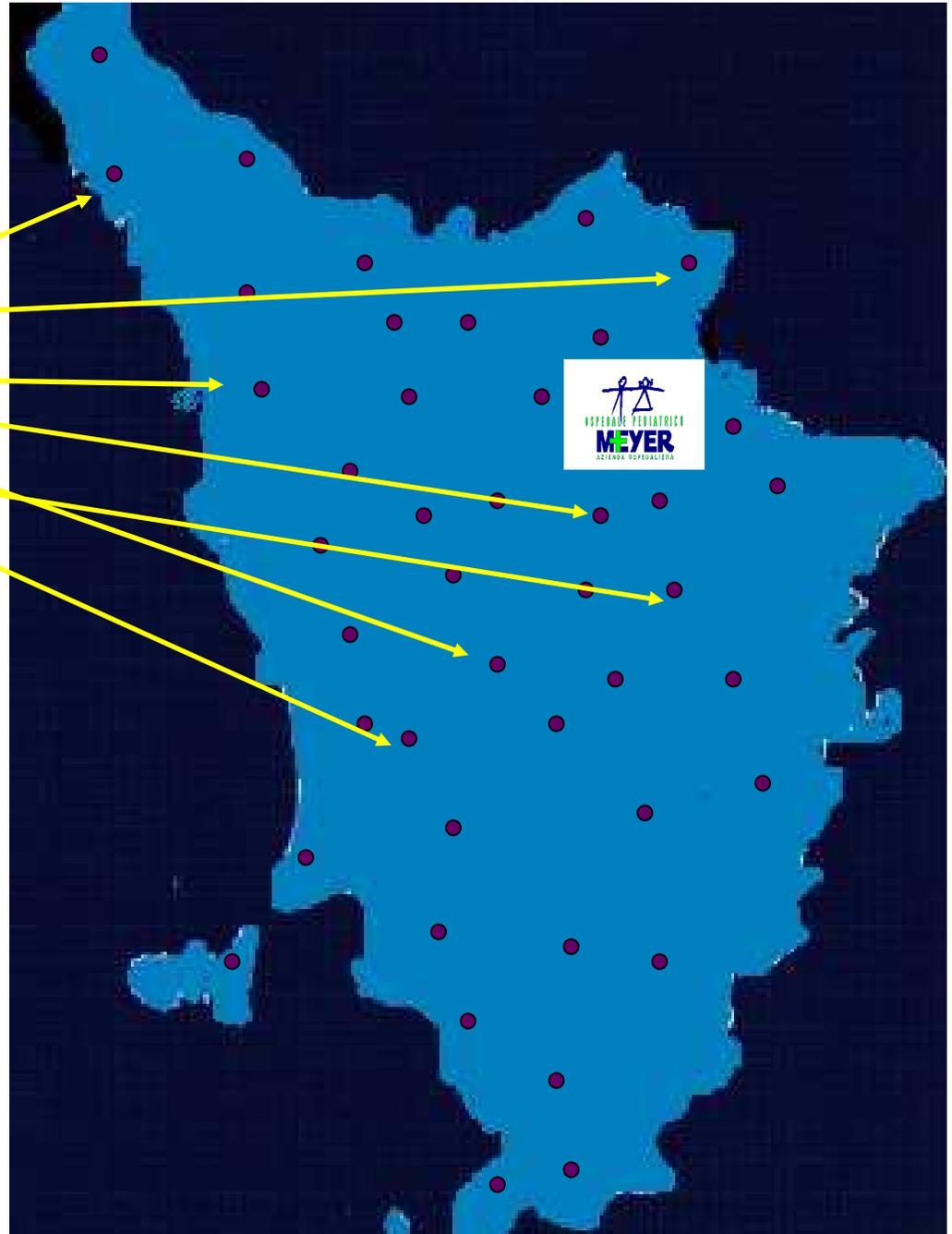
PREANALYTICAL PHASE

THE DRAWING OF THE BLOOD





**THE LAW REQUIRES THAT
THE DRAW OF THE BLOOD
MUST BE COLLECTED
BETWEEN 48 AND 72 HOURS
OF LIFE**



Pick-up time: between 16-17 P.M from 39 nurseries

Delivery to the NS centre: before 9 A.M

DBSs ARE SENT TO THE LAB BY COURIER (from MONDAY TO SATURDAY)



ANALYTICAL PHASE

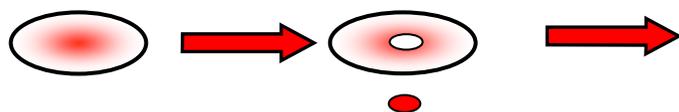
FROM ANALYSIS TO RESULT



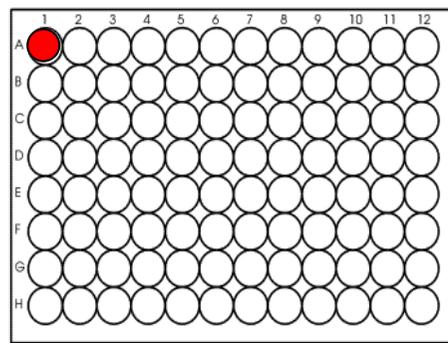
METHOD



dried blood spot (DBS)



Diameter of
circle 3.2 mm



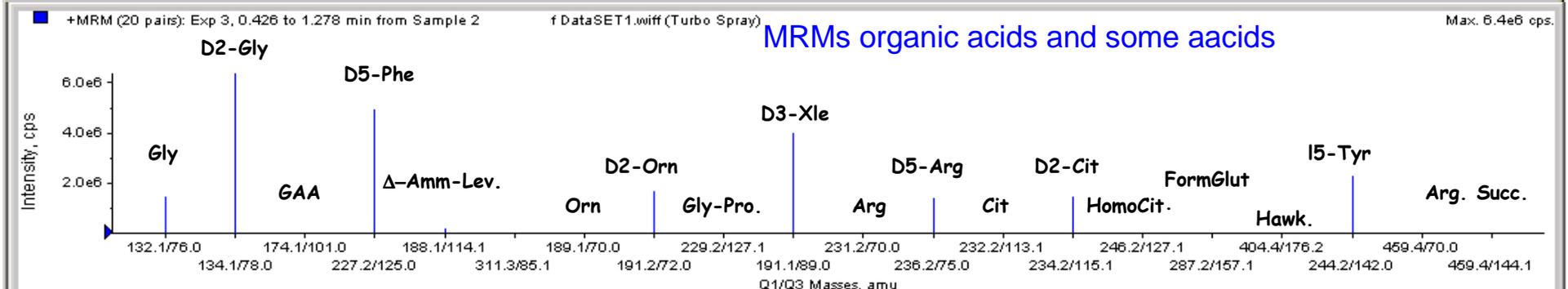
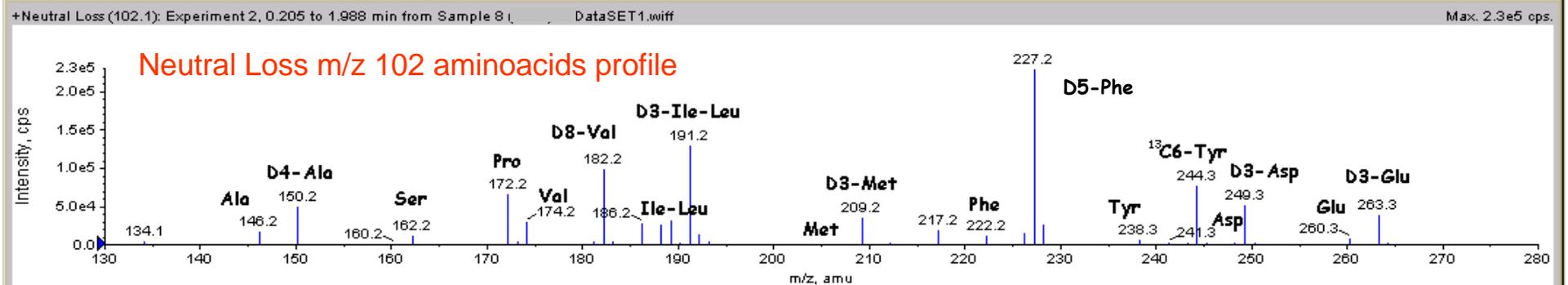
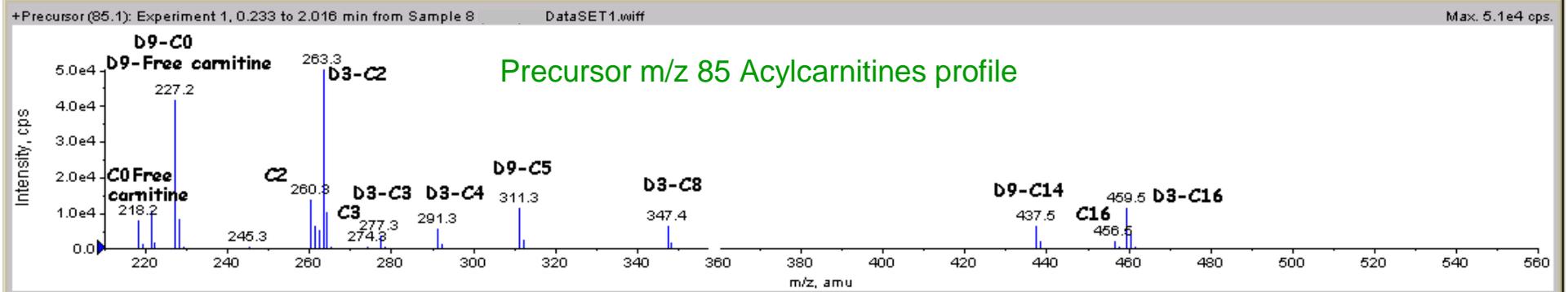
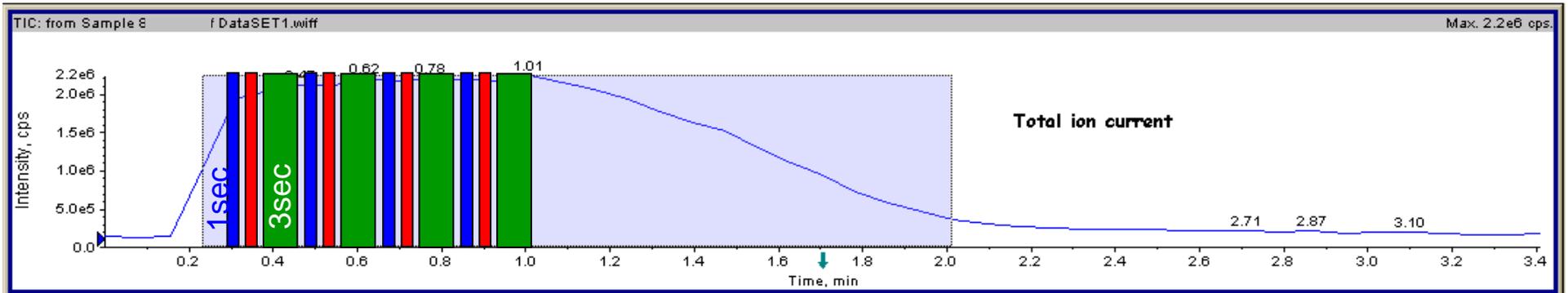
200 μ L of MeOH +
labelled internal
standards and

100 μ L of
hydrazine 3mmol/L
25 min 37°C

Evaporation
under nitrogen
flow 55°C



ACN/Water +0.05% formic acids 70:30
Flow Injection Analysis (LC-MS/MS)



C	I	J	K	L	M	N	O	P	Q	R
Sample	01b-C0	02b-C2	03b-C3	04b-C3:1	05b-C3DC	06b-C4	07b-C4DC	08b-C4OH	09b-C5	10b-C5DC
	8	6.3	0.2	0.01	0.01	0.01	0.04	0.01	0.01	0.01
	45	48	5.65	1.5	0.25	0.92	0.54	0.55	0.56	0.21
24775	20.33	21.26	0.84	0.03	0.09	0.08	0.18	0.18	0.04	0.03
	11b-C5:1	12b-C5OH	13b-C6	14b-C6:1	15b-C6DC	17b-C8	18b-C8:1	20b-C10	21b-C10:1	22b-C10-OH
	0.01	0.05	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01
	0.33	0.57	0.25	0.14	0.13	0.31	0.3	0.36	0.29	0.36
	0.05	0.09	0.05	0.13	0.09	0.10	0.12	0.08	0.05	0.06
	23b-C12	24b-C12:1	25b-C12-OH	26b-C14	27b-C14:1	28b-C14:2	29b-C14-OH	30b-C16	31b-C16:1	32b-C16-OH
	0.01	0.01	0.01	0.05	0.01	0.01	0.01	0.48	0.02	0.01
	0.69	0.39	0.11	0.57	0.44	0.2	0.11	5.6	0.52	0.1
	0.20	0.10	0.03	0.20	0.12	0.03	0.04	2.23	0.16	0.03
	33b-C18	34b-C18:1	35b-C18:2	36b-C18-OH	37b-C18:1-OH	38b-C20	39b-C20:1	40b-C22	41b-C22:1	42b-C24
	0.2	0.3	0.01	0.01	0.01	0.01	0.001	0.06	0.06	0.01
	1.77	2.43	0.41	0.08	0.1	0.08	0.17	0.78	1.85	0.92
	0.55	0.74	0.05	0.01	0.03	0.02	0.03	0.46	1.21	0.26
	43b-C24:1	44b-C26	45b-C26:1	46b-C28	47b-C28:1	48b-C16DC	49b-C18DC	80b-C2/C0	81b-C3/C0	82b-C3/C4
	0.08	0.08	0.01	0.01	0.01	0.01	0.01	0.5	0.03	1.1
	0.52	0.28	0.32	0.28	0.28	1.05	0.65	2.12	0.13	12.5
	0.33	0.10	0.17	0.05	0.15	0.31	0.14	1.05	0.04	10.13
	83b-C5/C3	84b-C5/C4	85b-C5/C8	86b-C3/C16	87b-C5DC/C4	88b-C5DC/C8	89b-C5DC/C12	90b-C8/C6	91b-C8/C10	92b-C16DC/C16
	0.01	0.04	0.01	0.11	0.04	0.19	0.12	0.85	0.33	0.01
	0.29	1.36	4.2	1.6	0.5	1.11	0.84	3	1.6	0.51
	0.05	0.53	0.44	0.38	0.41	0.34	0.17	2.22	1.32	0.14
	93b-C18DC/C18	93b-C14:1/C16	94b-C26/C22	104b-C14:1/C5	96b-CPTI	95b-CPTII	55b-Ala	56b-Val	57b-XIe	58b-Met
	0.01	0.05	0.07	0.01	1.6	0.05	59	57	60	7
	0.7	1.48	0.81	3.69	15	0.21	284	210	195	46
	0.25	0.74	0.22	2.69	2.78	0.14	118.42	80.78	110.48	13.61
	59b-Phe	60b-Tyr	61b-Asp	62b-Glu	63b-Gly	64b-Orn	65b-Arg	66b-Cit	67b-ArgSuc	68b-GAA
	18	10	4	80	110	2	1	3	0.01	0.01
	100	200	69	420	721	85	12	25	0.5	3
	53.55	58.50	34.84	215.07	412.43	35.87	5.88	8.19	0.04	0.59
	69b-ForGlu	70b-AmmLev	71b-GlyPro	72b-HomoCit	73b-Hawk					
	0.01	1	1	0.2	0.01					
	2	65	15	2.8	0.2					
	0.16	14.69	14.80	0.68	0.04					

Acylcarnitines

β -oxidation of fatty acids defects

- Short Chain Acyl-CoA Dehydrogenase Deficiency (SCAD)
- Medium Chain Acyl-CoA Dehydrogenase Deficiency (MCAD)
- Multiple Acyl-CoA Dehydrogenase Deficiency (MADD)
- Very Long Chain Acyl-CoA Dehydrogenase Deficiency (VLCAD)
- Long Chain 3-OH-Acyl-CoA Dehydrogenase Deficiency (LCHAD)/
Mitochondrial Trifunctional Protein Defect
- Carnitine Transporter Defect
- Carnitine-Acylcarnitine Translocase Deficiency
- Carnitine Palmitoyl Transferase Deficiency (CPT I and II)

Organic acidurias

- Propionic Aciduria
- Methylmalonic Acidurias
- Isovaleric Aciduria
- Methylcrotonyl-CoA Carboxylase Deficiency
- Holocarboxylase Synthetase Deficiency
- Glutaric Acidurias type I
- 3-OH-3-Methylglutaryl-CoA Liase Deficiency
- β -Ketothiolase Deficiency

Aminoacids

Aminoacidopathies

- HyperPhenylalaninaemias
- Tyrosinaemia type I and II
- Leucinoses
- Non ketotic Hyperglycinaemia
- Citrullinaemia type I and II
- Argininsuccinic aciduria
- Argininaemia
- Homocystinuria
- HyperOrnithinemia

Progress in expanded newborn screening for metabolic conditions by LC-MS/MS in Tuscany: Update on methods to reduce false tests

G. la Marca • S. Malvagia • B. Casetta • E. Pasquini • M. A. Donati • E. Zammarchi

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Summary We report on our 6-year experience of expanded newborn screening by tandem mass spectrometry in Tuscany (Italy), the first Italian Region to screen all newborns for more than 40 inborn errors of metabolism: organization, diseases observed and updates on methods to reduce false-positive and false-negative tests are described. Blood collection is recommended between 48 and 72 h of life. Blood spots are sent daily by courier to laboratory. When a positive result occurs, two subsequent procedures are followed: for disorders with possible acute metabolic decompensa-

tion, the baby is immediately recalled and clinical examinations and confirmatory tests are performed; for the other disorders, the nursery provides for a second blood spot. If the test is positive, clinical examinations and confirmatory tests are performed. In both cases, if confirmatory tests are positive, a treatment and a follow-up programme are started. Up to now, spots from 160 000 infants have been analysed and 80 affected patients have been identified (disorders of amino acids, organic acids and fatty acids metabolism). We describe adjustments to cut-off values, the introduction of a second-tier test for propionic acidaemia and for methylmalonic aciduria, the inclusion of succinylacetone in the panel of metabolites, and protocols for premature infants and for newborns on parenteral nutrition or

Communicating editor: Bridget Wilcken

Our panel contains 22 primary targets and 22 secondary targets

J Inherit Metab Dis. 2008 Oct 27



Aminoacidopathies

AMINOACIDOPHATIES

58 HYPERPHENYLALANINEMIAS

Prevalence 1:5000

13 classical PKU Prevalence 1:22000

1 DHPR Defect

5 Citrullinemias type I

2 Tyrosinemia type I

2 Citrullinemias type II

1 Argininosuccinic acidemia

2 MAT III

4 Hyperhydroxyprolinemias

1 OCT

PHENYLKETONURIA

(Occurrence 1:15000)

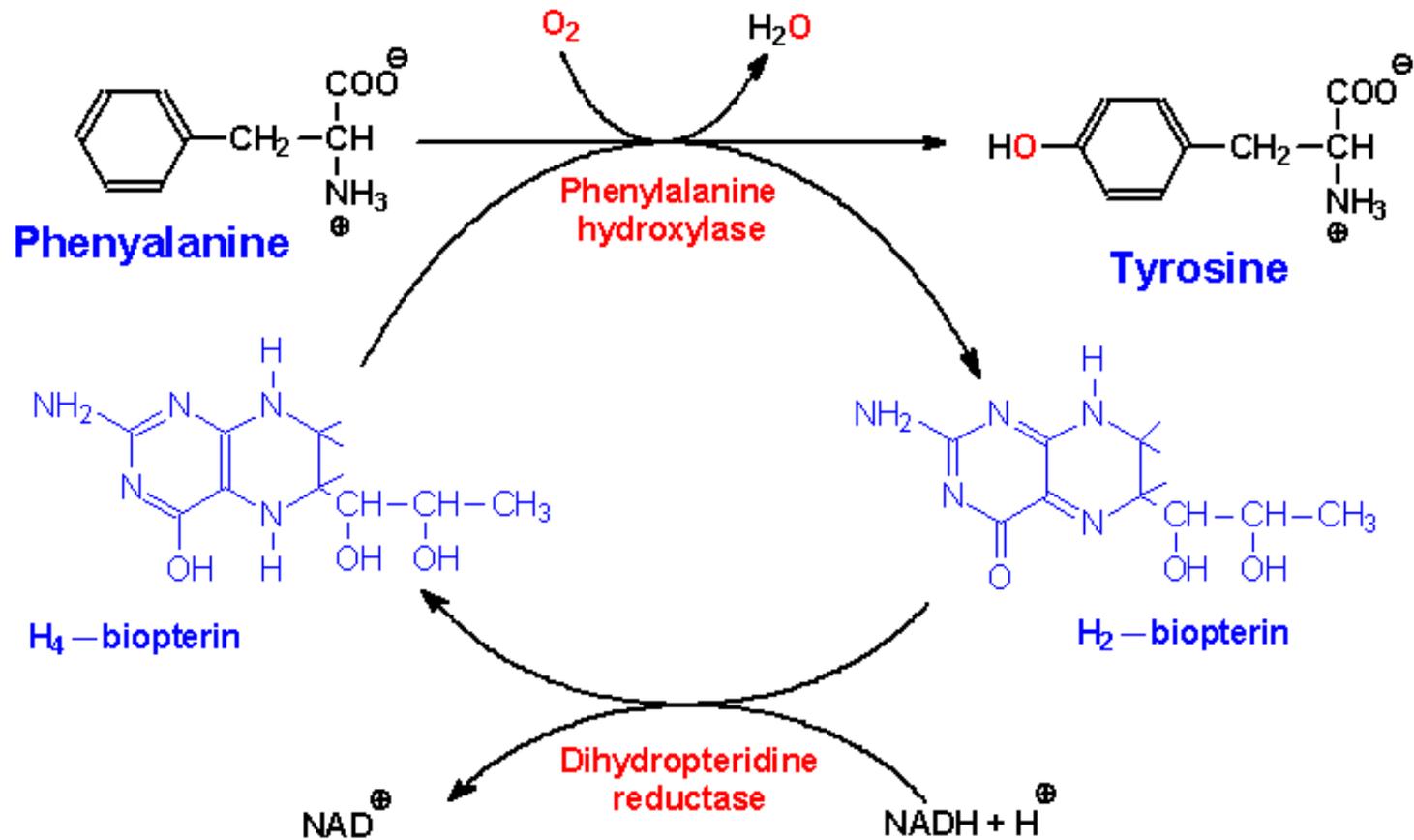
Phenylalanine Hydroxylase (PKU)

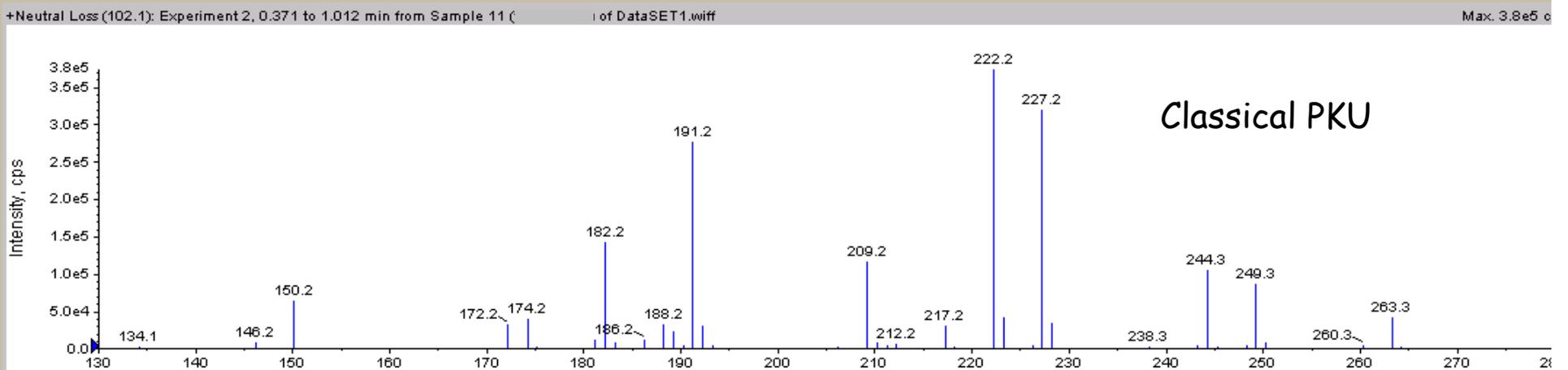
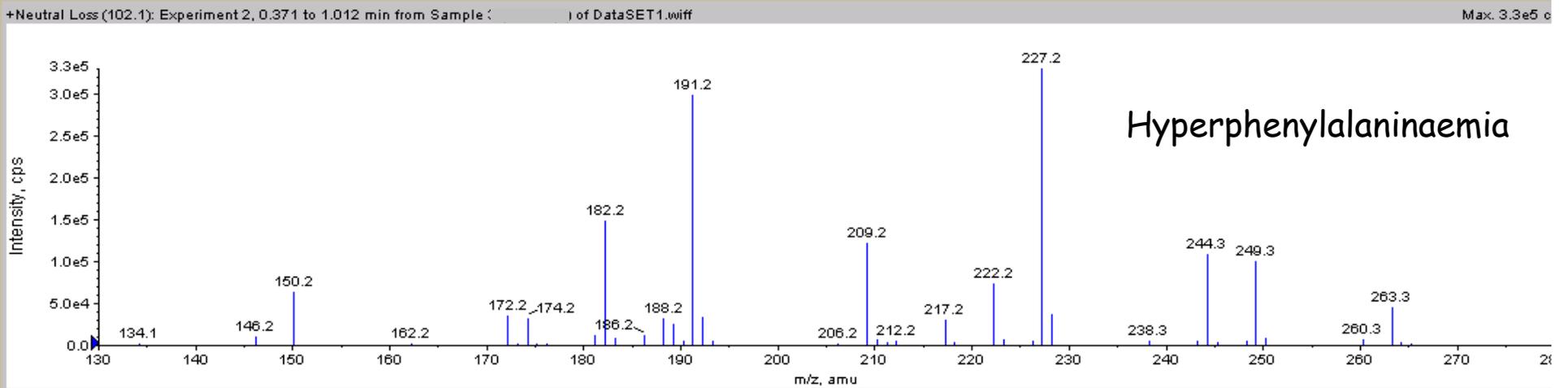
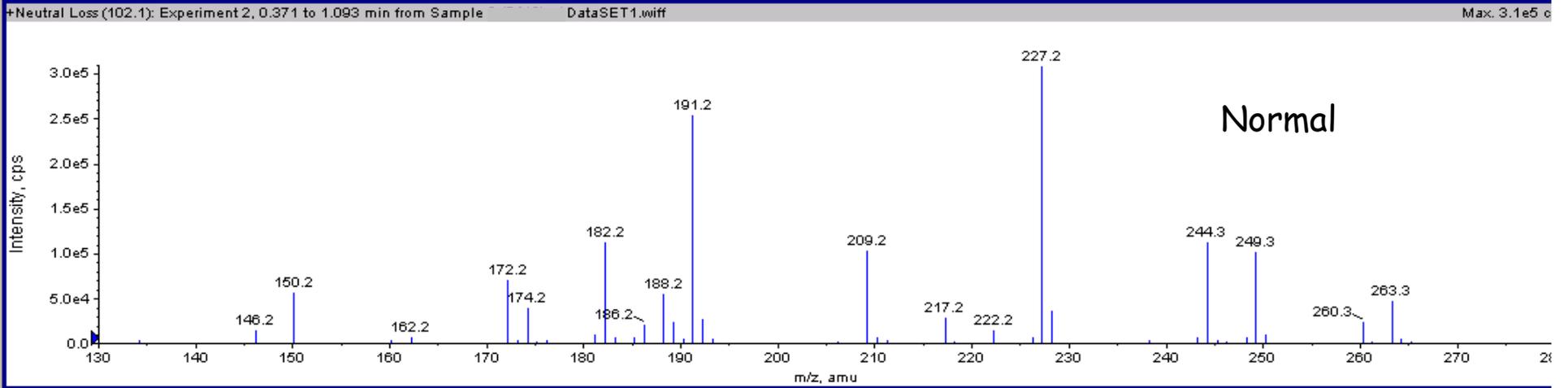
or co-factor BH_4 Defect

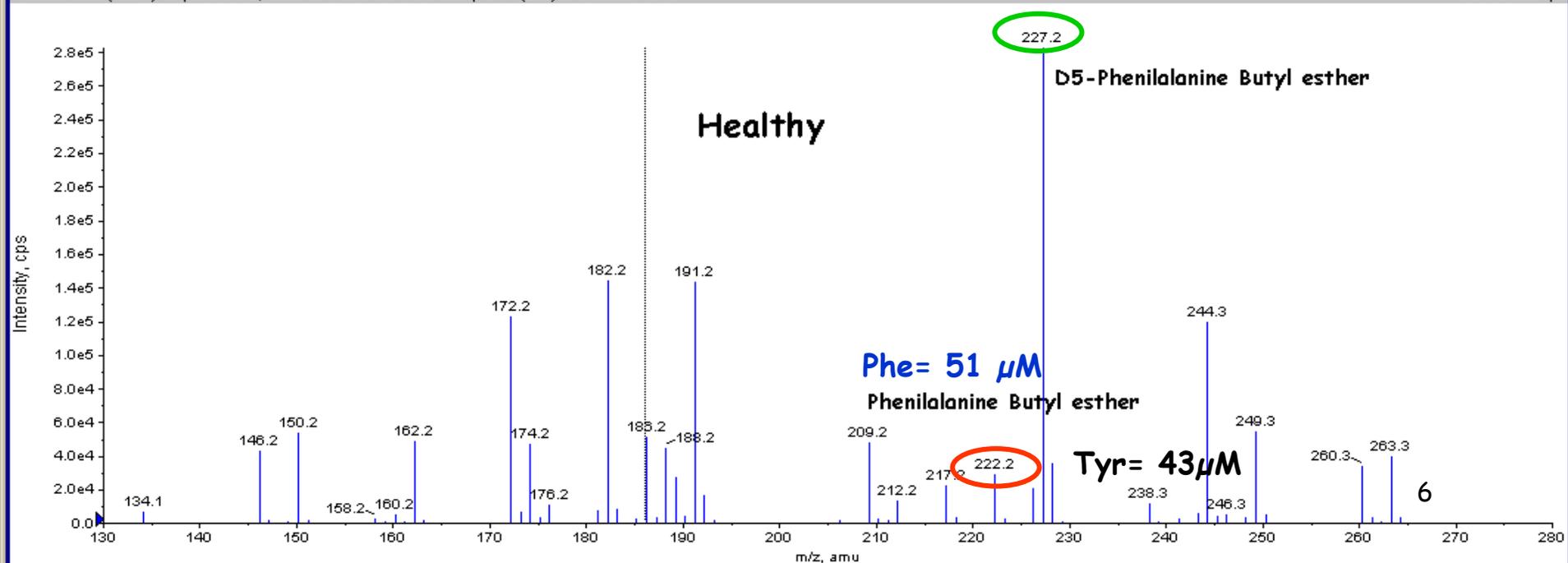
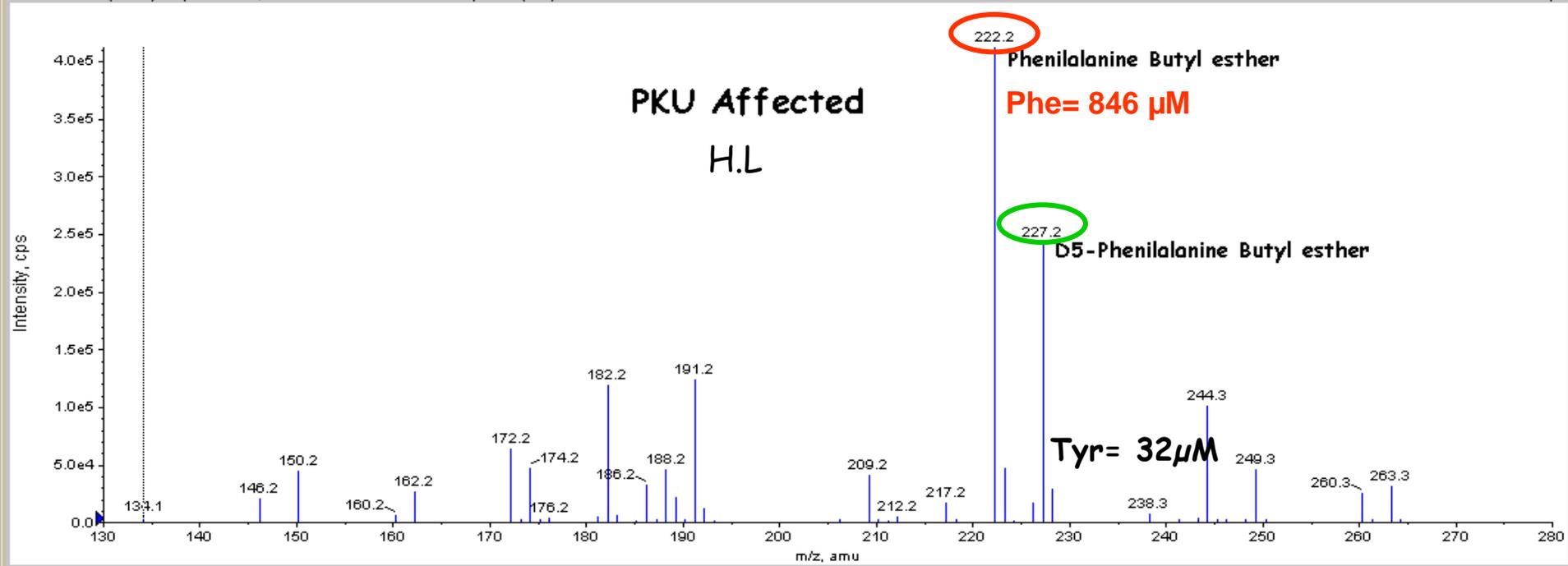
Toxic accumulation of Phenylalanine
(SNC)

Mental retardation, epilepsy,
depigmentation

Hyperphenylalaninaemia







49 ORGANIC ACIDURIAS

3 Propionic Acidemias

1 Glutaric Aciduria tipo I

3 3-Me-Crotonyl Glycinurias (maternal transmission)

2 Isovaleric Acidurias

1 Methylglutaconic aciduria

12 Isobutyric Acidurias

9 Formimino Trasferase defects

18 Methylmalonic Acidurias (3 maternal; mothers were vegetarian)

23 β -OX FATTY ACIDS DEFECTS

10 MCAD (1:26600)

7 SCAD

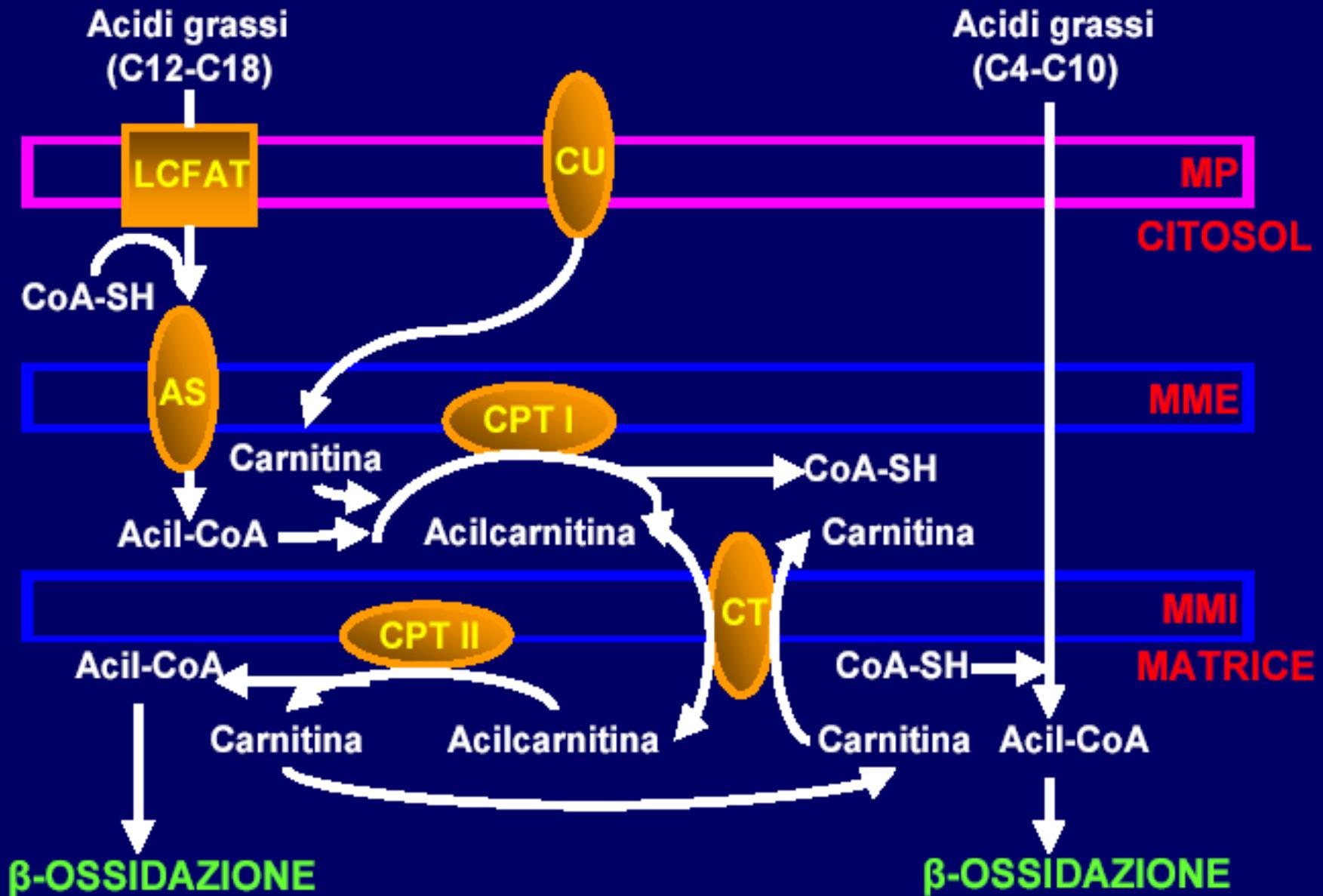
2 VLCAD

4 CARNITINE DEFICIENCIES (2 maternal)



β -oxidation of fatty acids defects

MITOCHONDRIAL CARNITINE PATHWAY

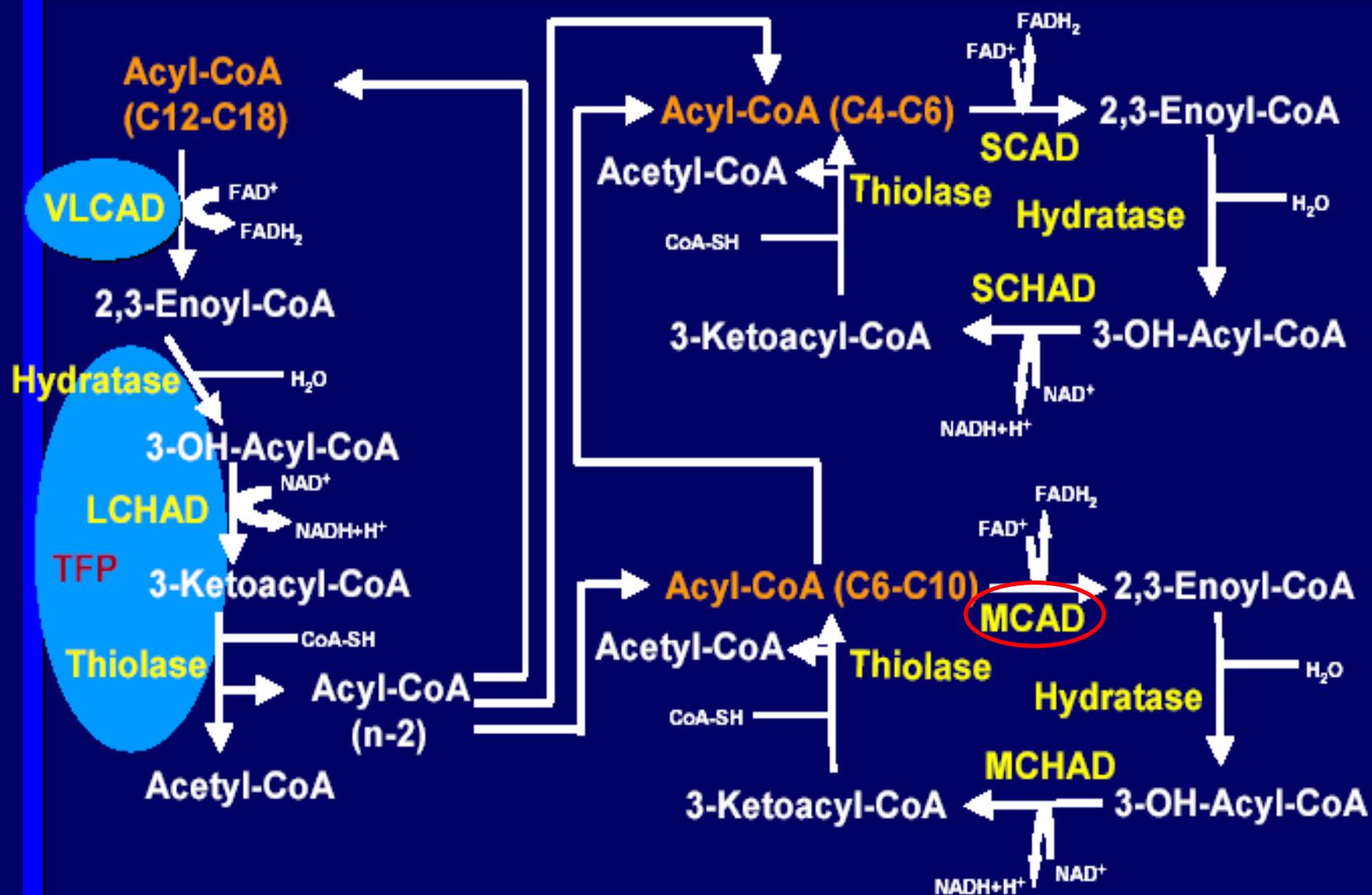


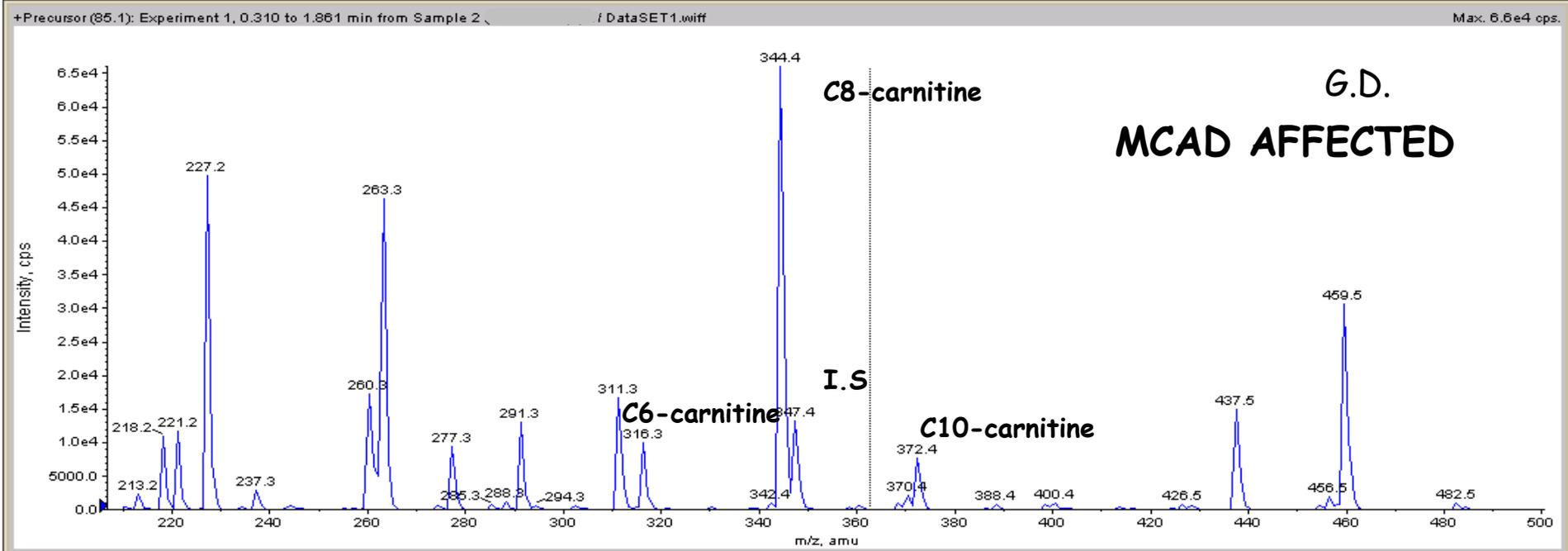
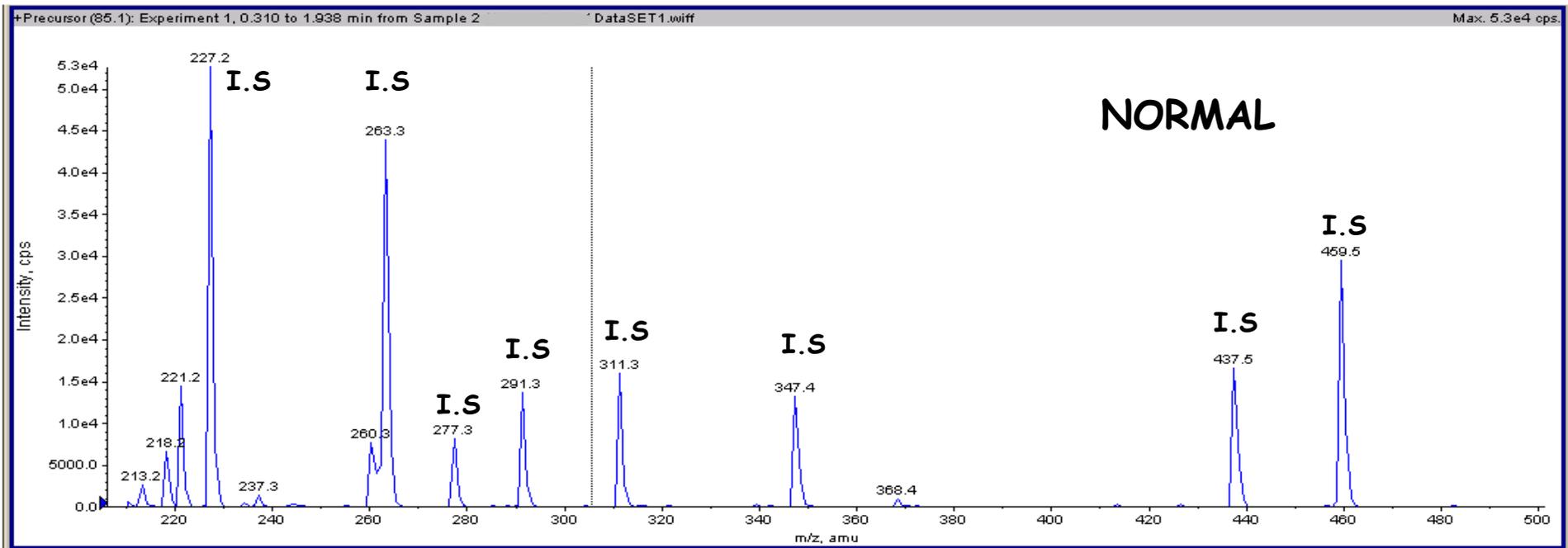
MCADD: Medium-Chain Acyl CoA
Dehydrogenase Deficiency

AUTOSOMAL RECESSIVE

Estimated occurrence ~1:15000
Caucasian births

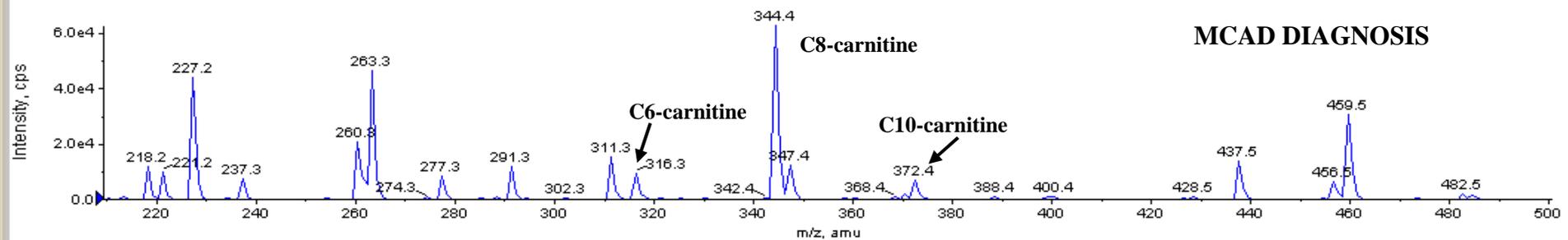
β -oxidation of long, medium and short chain fatty acids



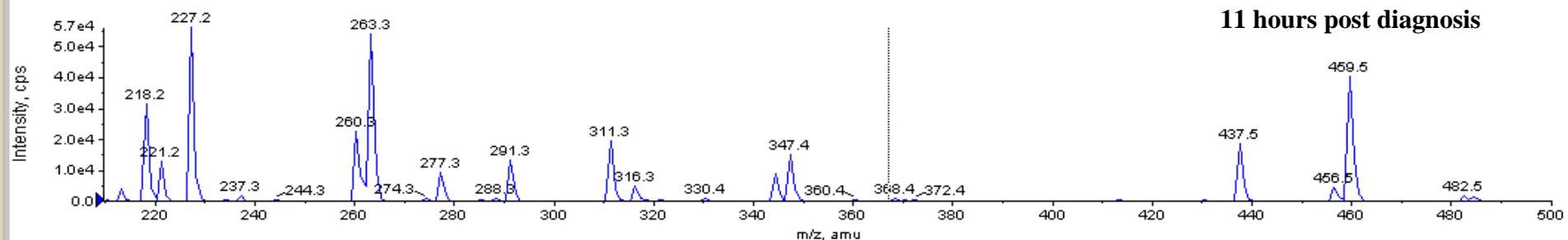


G. la Marca et al, Rapid Comm Mass Spectrom, 17, 2003

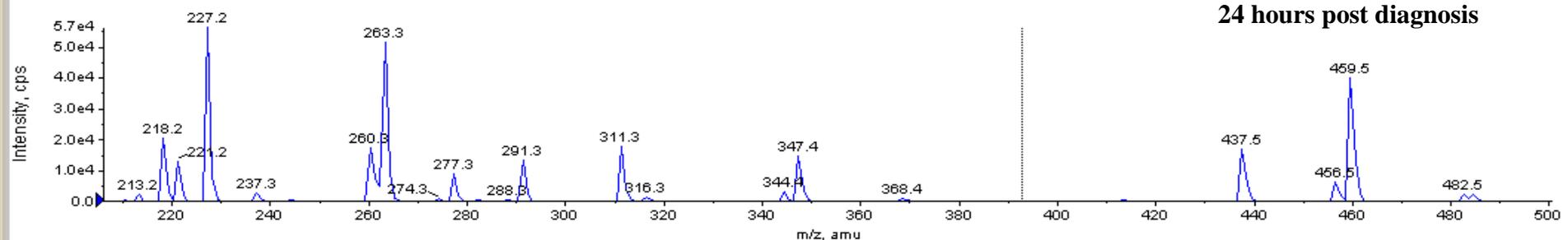
+Precursor (85.1): Experiment 1, 0.310 to 1.706 min from DataSET1.wiff Max. 6.3e4 cps.



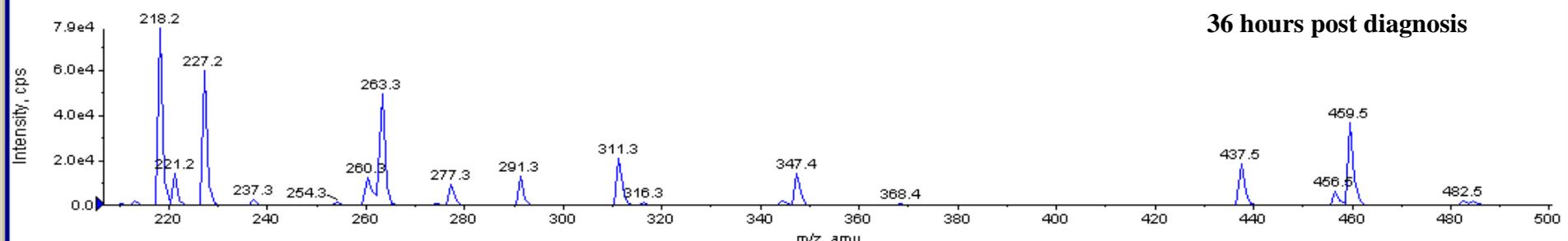
+Precursor (85.1): Experiment 1, 0.233 to 1.783 min from Sample 2 ((eb) of DataSET1.wiff Max. 5.7e4 cps.

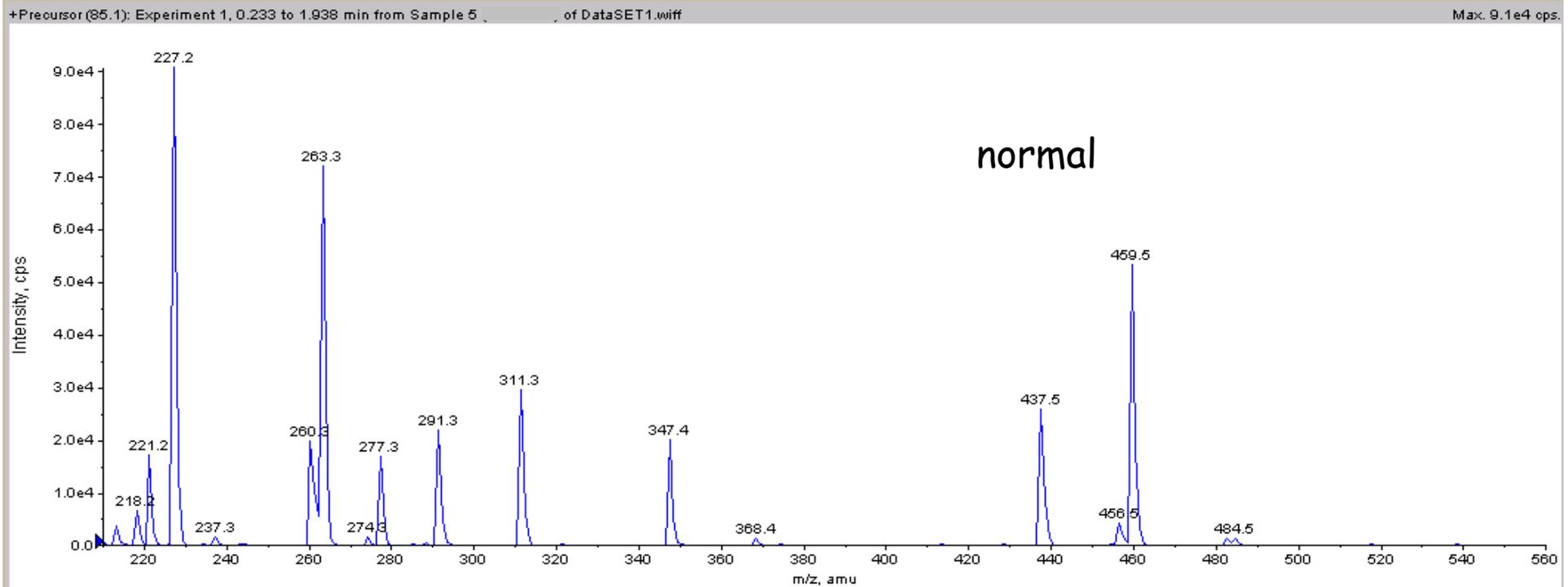
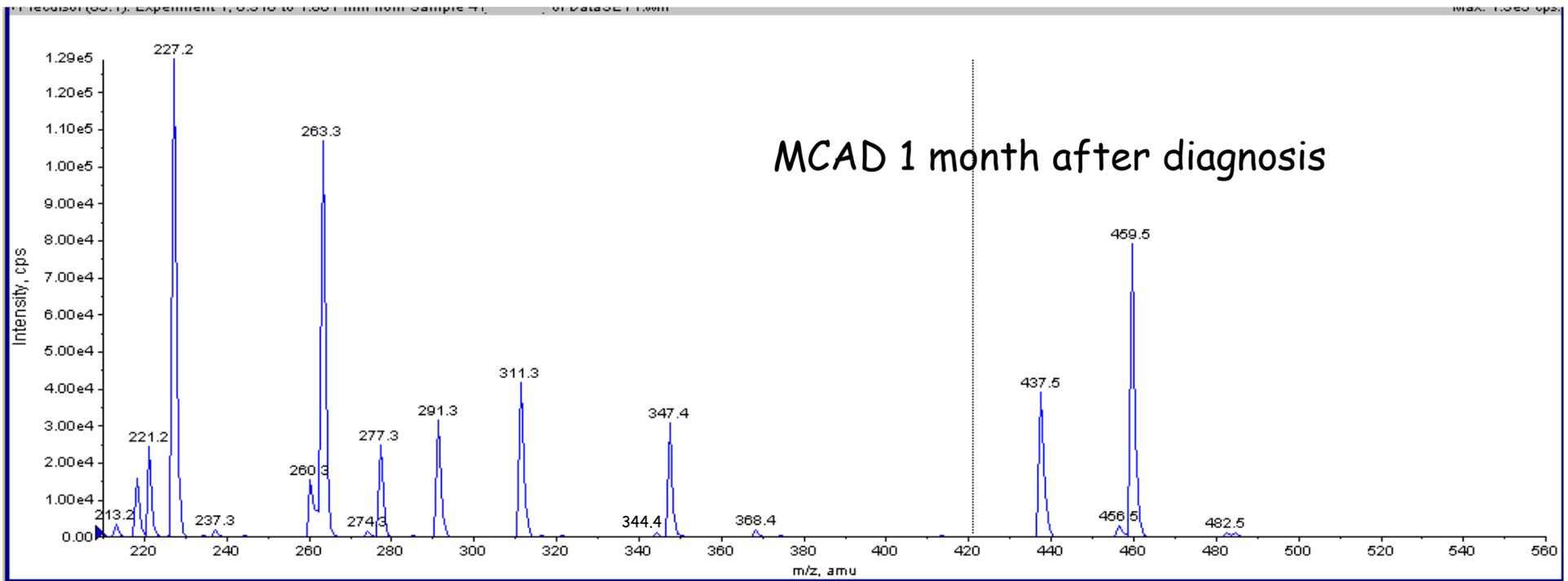


+Precursor (85.1): Experiment 1, 0.310 to 1.861 min from Sample 3 ((eb) of DataSET1.wiff Max. 5.7e4 cps.



+Precursor (85.1): Experiment 1, 0.310 to 1.551 min from Sample 1 ((eb) of DataSET1.wiff Max. 7.9e4 cps.





MCAD DEFICIENCY

- hypoketotic hypoglycaemia
- Liver failure
- Reye like manifestations

- After 8-16 hours fasting
- During ordinary illness
- After surgery

POST-SYMPTOMATIC DIAGNOSIS (120 patients.)

Coma	84%
Residual neurologic damage	40%
Mortality	20%

NEWBORN SCREENING by LC-MS/MS

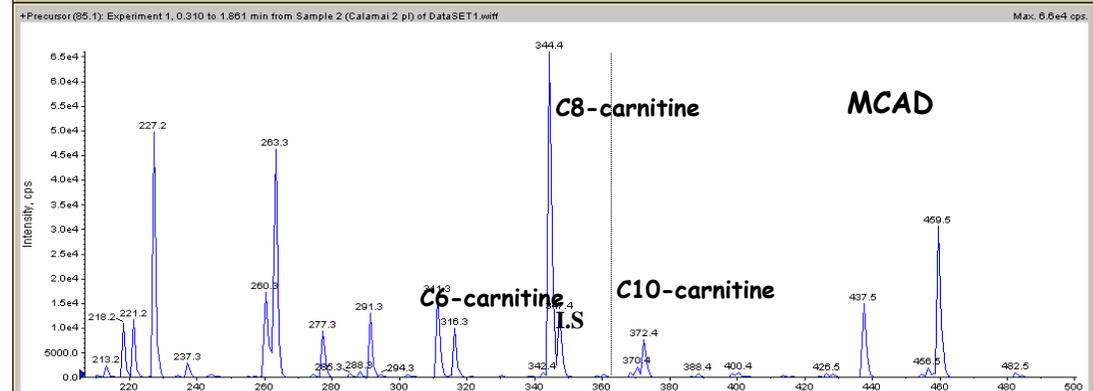
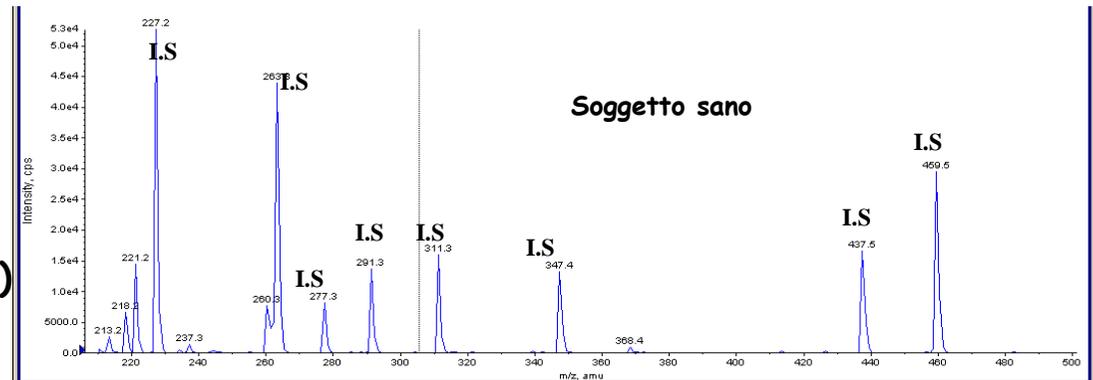
PRE-SYMPTOMATIC DIAGNOSIS (62 pat.)

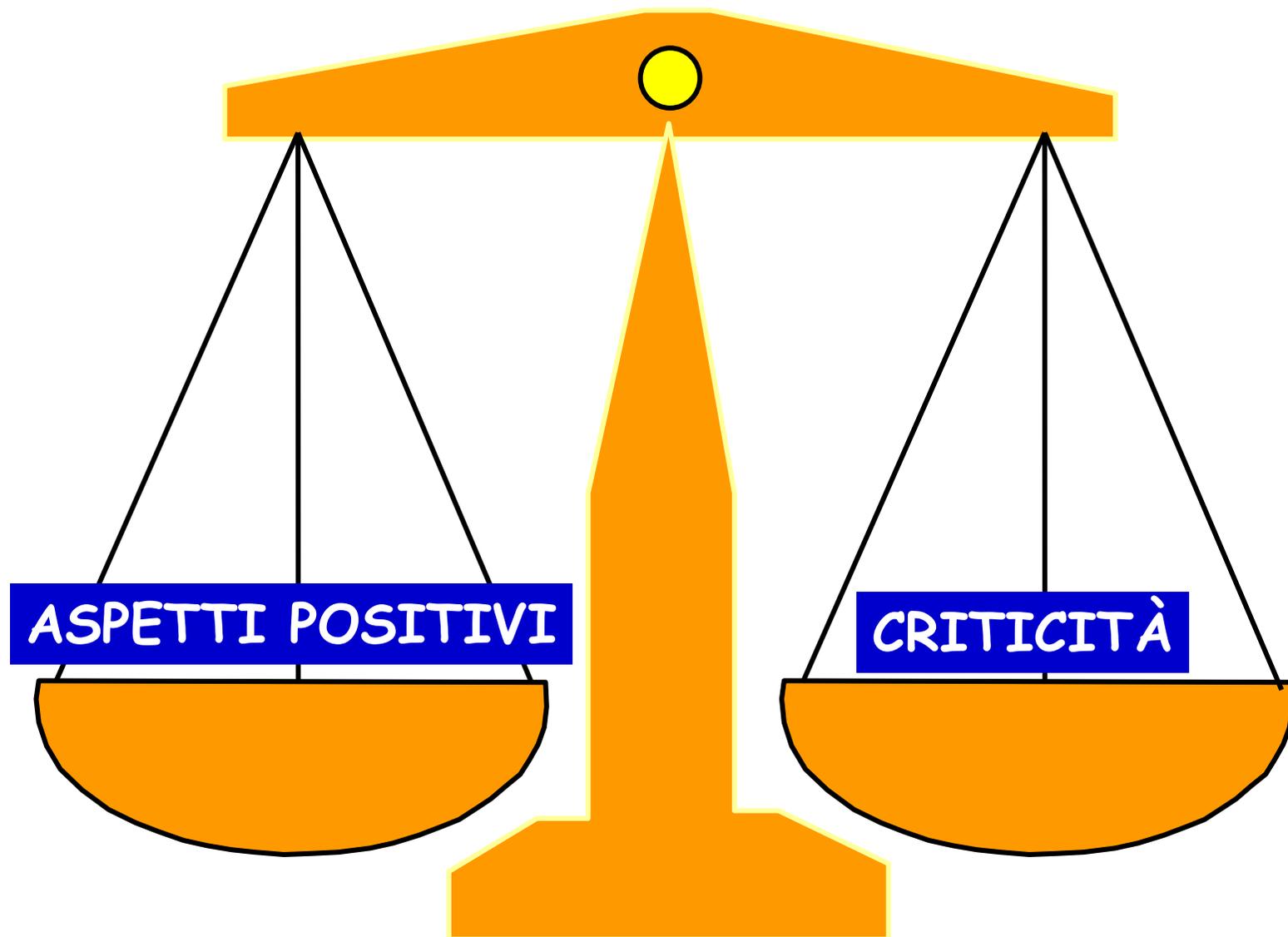
Coma 0%

Mental Retardation 0%

Mortality 0%

Muntau 1999-2003





ASPETTI POSITIVI

CRITICITÀ

SCREENING BY MS/MS

POSITIVE ASPECTS

- EXPANDED SCREENING
- PRECLINICAL DIAGNOSIS
- EARLY THERAPY
- BEST PROGNOSIS
- QUANTITATIVE RESULTS
- MONITORING DURING THERAPY

CRITICISMS

- FALSE POSITIVES, NEGATIVES
- ASYMPTOMATIC FORM
- NO RESOLUTIVE THERAPY IN SOME CASES
- FAMILIAR STRESS
- COSTS

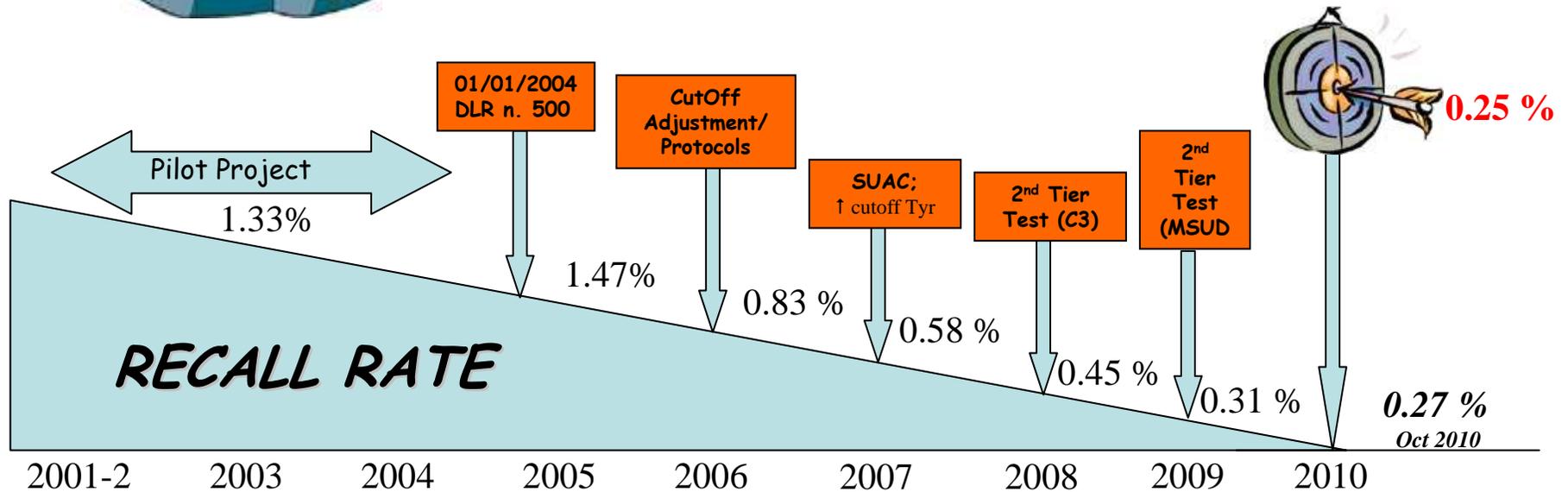
COSTS and BENEFITS

LARGE NUMBER OF FALSE POSITIVE TESTS

ELEVATED NUMBER OF RECALLS



PARENTAL STRESS



Italy has
32
newborn
screening
centers

Firenze



**Padova/
Verona**

Genova

Roma

Milano

Napoli

**Catania/
Palermo**

Newborn screening for Severe Combined Immunodeficiencies

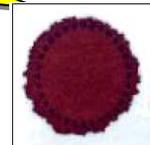
FUTURE PERSPECTIVES

Newborn screening for Lysosomal Storage Disorders

***NEXT FUTURE PERSPECTIVES OF
NEWBORN SCREENING IN TUSCANY***



**Newborn Screening for Severe
Combined Immunodeficiency**

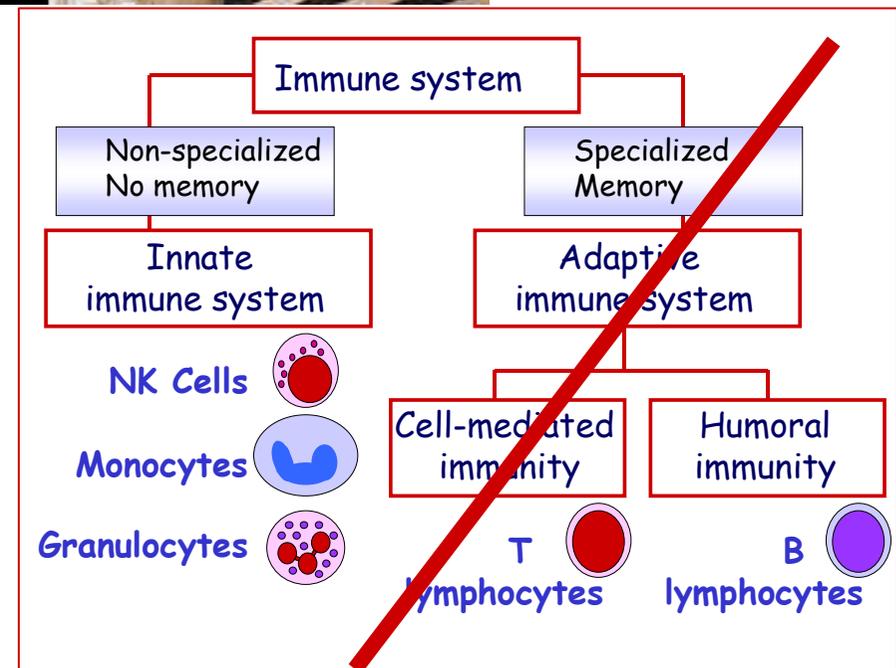


**A pilot project for ADA SCID
has started January 1st 2011**

Among all immunodeficiencies,
Severe Combined Immunodeficiencies (SCID)
are undoubtedly the most severe

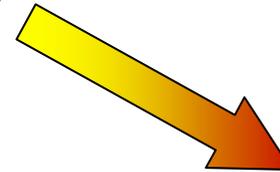


Children with SCID lack
all adaptive
immune responses



**A prompt diagnosis in the first days of life
allows to plan and start
the correct therapy**

IMMEDIATELY



**SEVERE
COMPLICATIONS
CAN BE PREVENTED**

Therapy

Antibiotics

Antiviral

Immunoglobulins e.v.

Enzyme therapy i.m.



How was up to now diagnosis performed?

**When a clinical suspicion was present...
toxic metabolites of ADA or PNP
were evaluated in urine samples**



**or ADA activity was
evaluated on blood
samples**

**Both methods are
late and expensive**

The idea....

**Looking for toxic metabolites
of ADA (adenosine and deoxyadenosine)
or PNP (deoxyinosine, deoxyguanosine, guanosine)
at birth on dried blood spots**

**Including diagnosis of
ADA and PNP SCID in the
newborn screening panel**

Features of SCIDs due to deficiency of

- ✦ Adenosine-deaminase (ADA-SCID) or**
- ✦ Purine-nucleoside phosphorilase (PNP-SCID)**

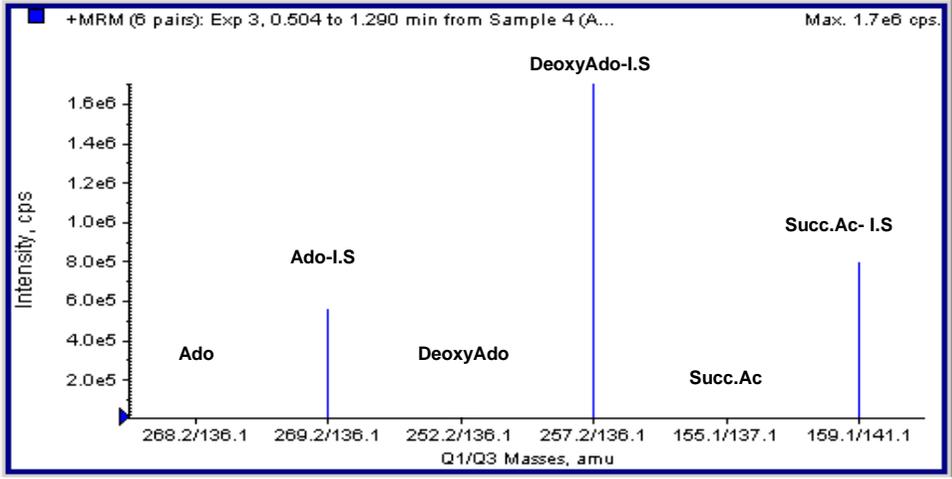
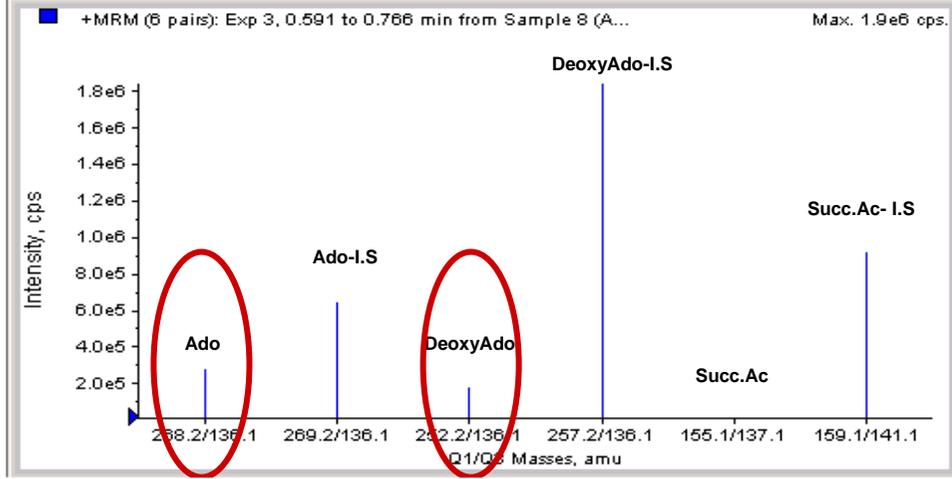
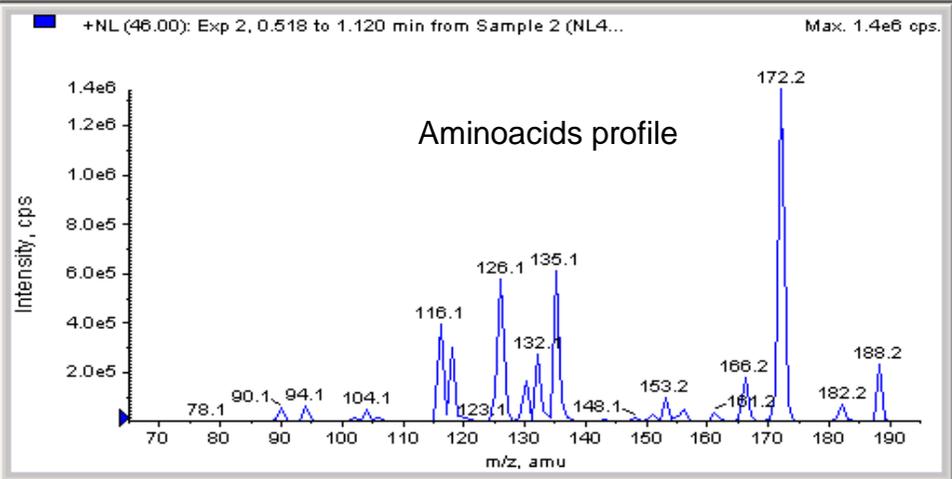
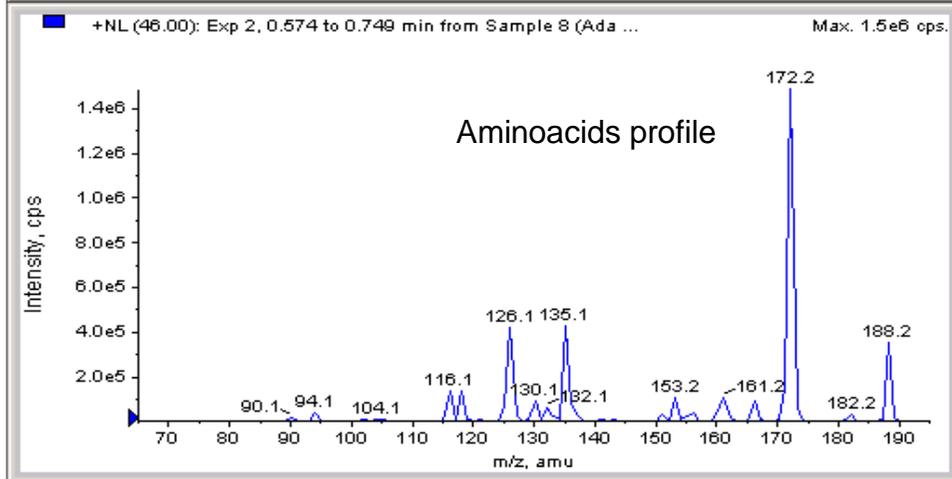
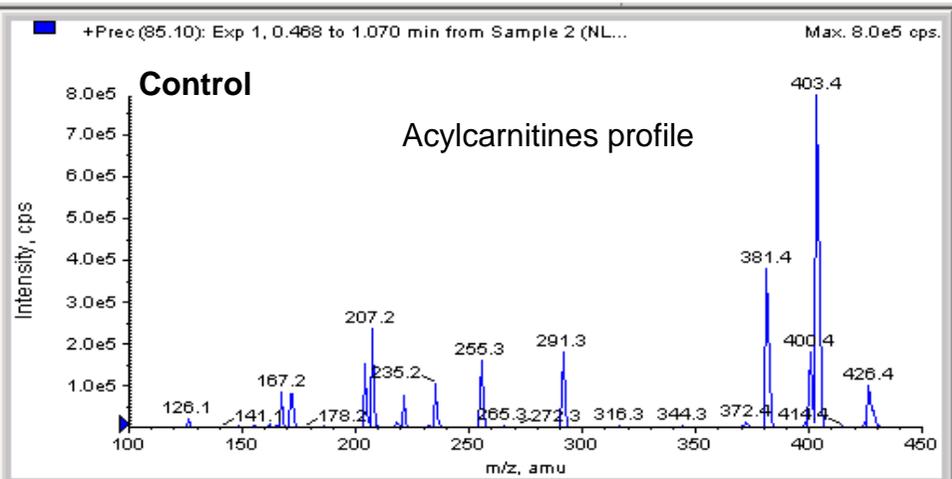
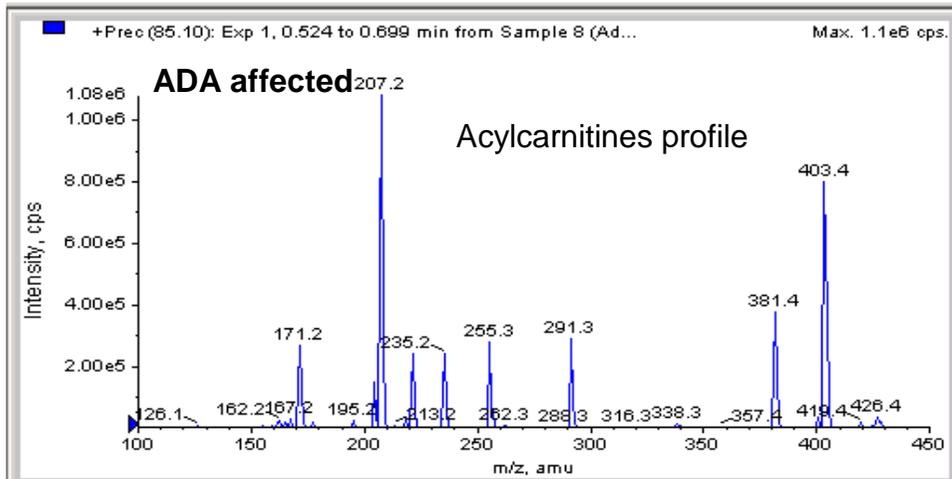
**...are those considered
as necessary criteria to
include that diagnosis
in a screening program**

Availability of diagnostic tests

Low cost per test

Availability of specific therapies

Clinical improvement if early diagnosis is performed



The experiments....

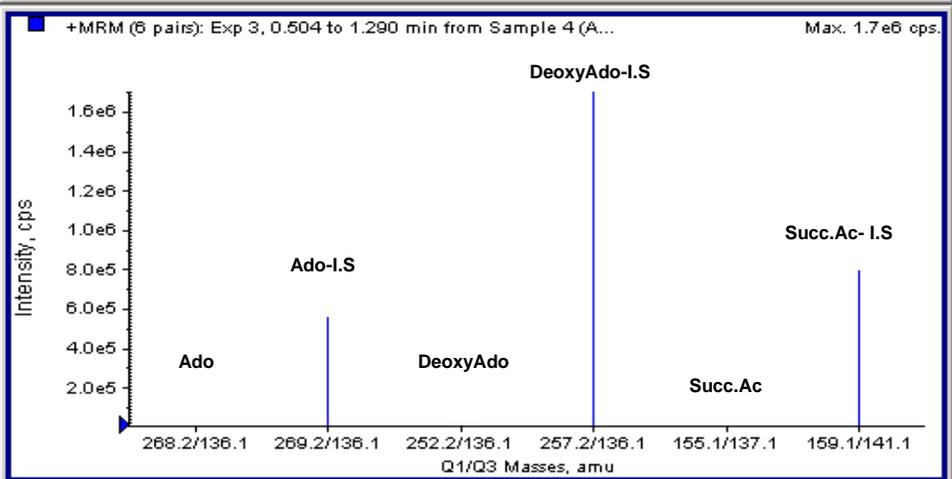
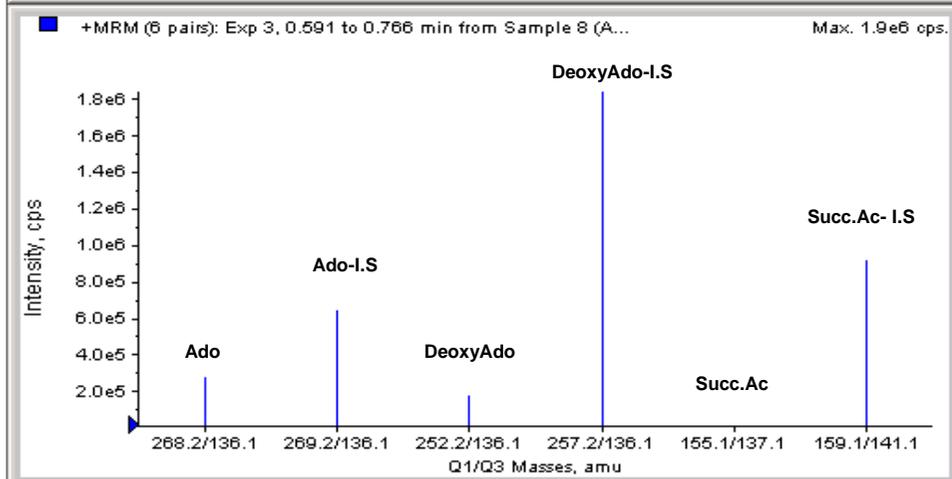
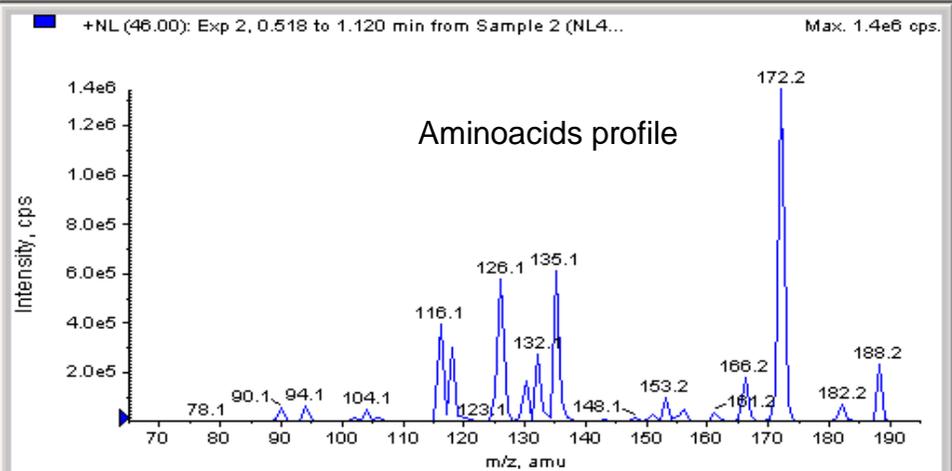
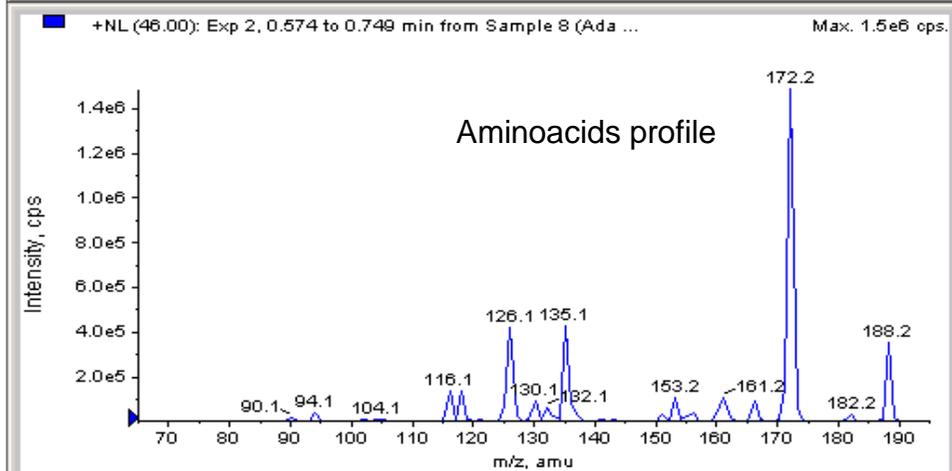
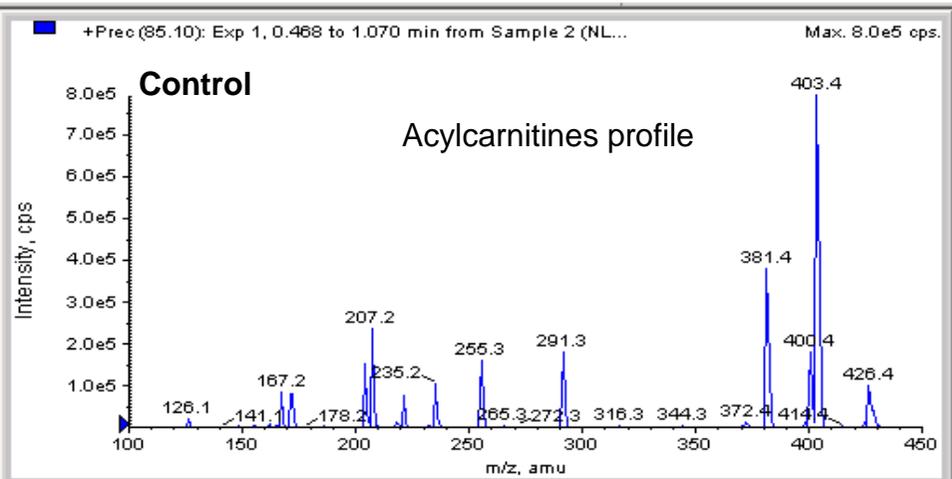
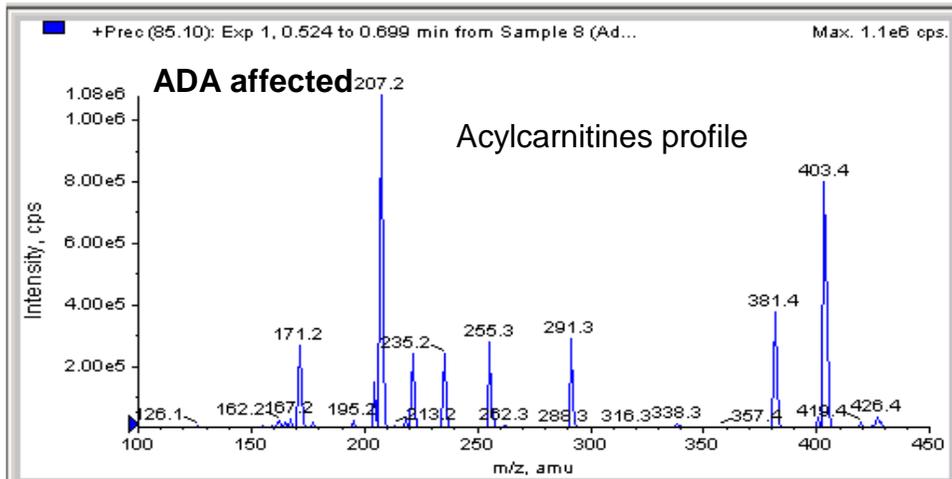


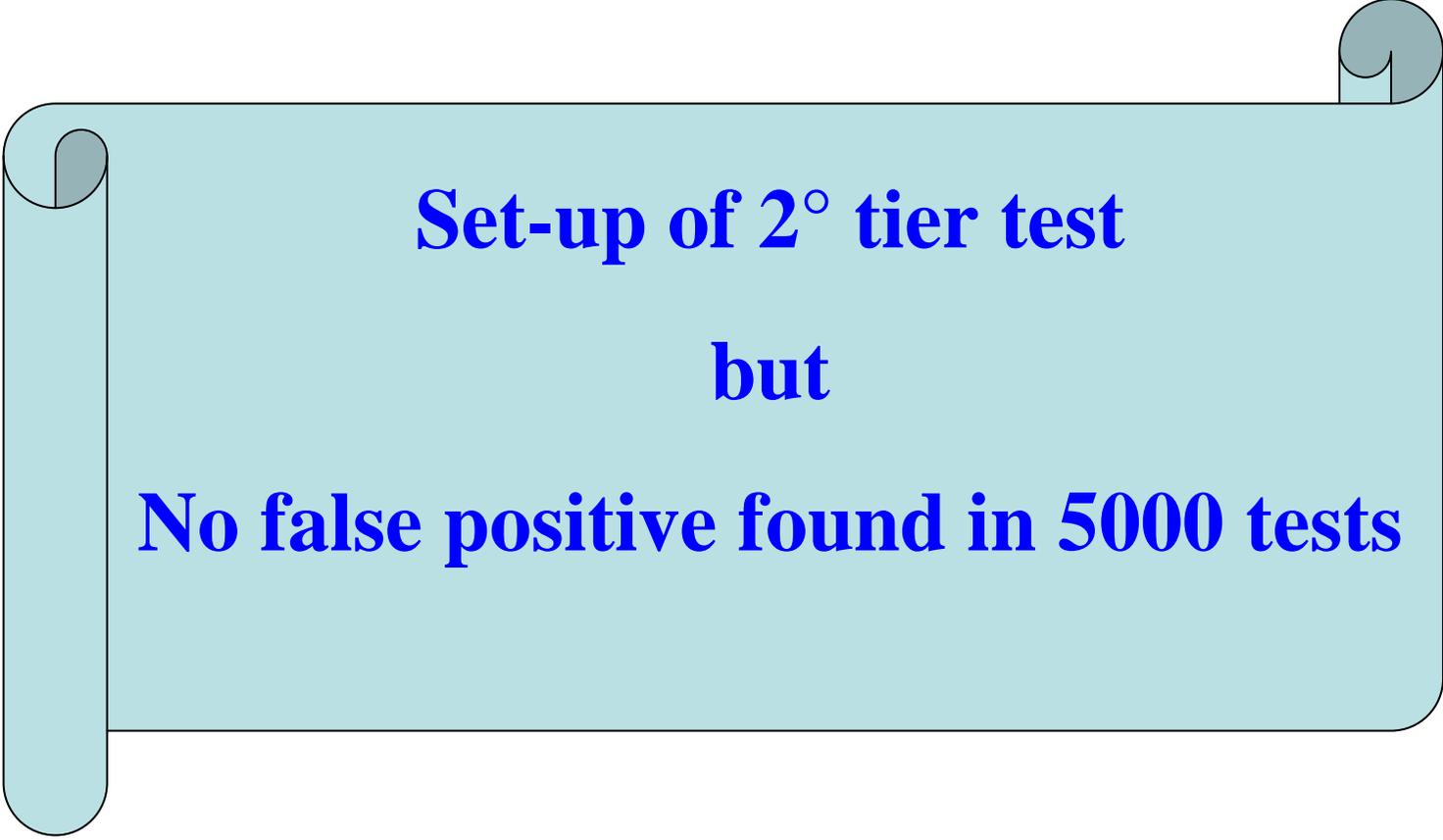
Sensitivity

Analysis of dried blood spots taken at birth from 4 newborns with confirmed (genetic) diagnosis of ADA

Specificity

Analysis of dried blood spots taken at birth from 15000 healthy newborns





Set-up of 2^o tier test

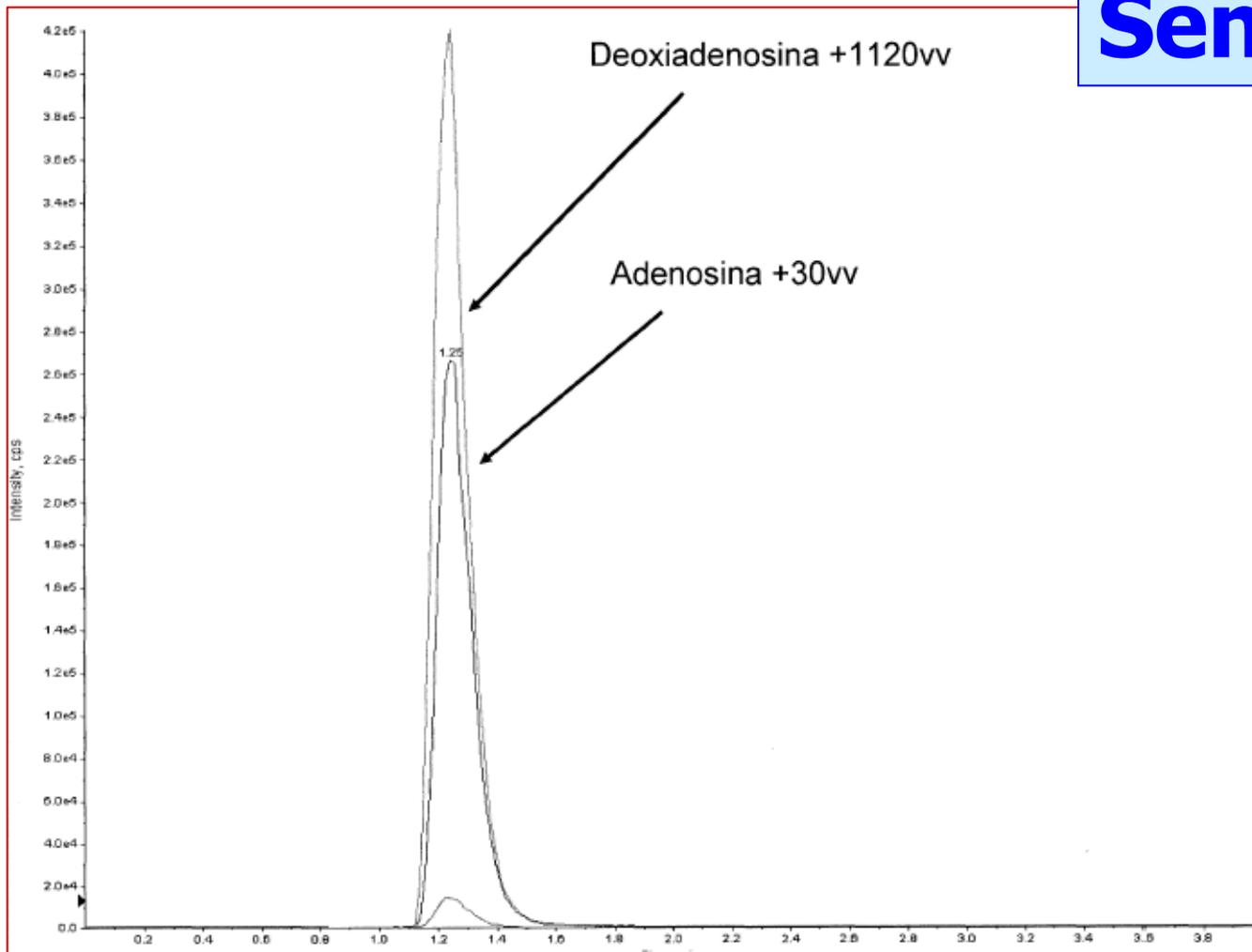
but

No false positive found in 5000 tests

The results

1

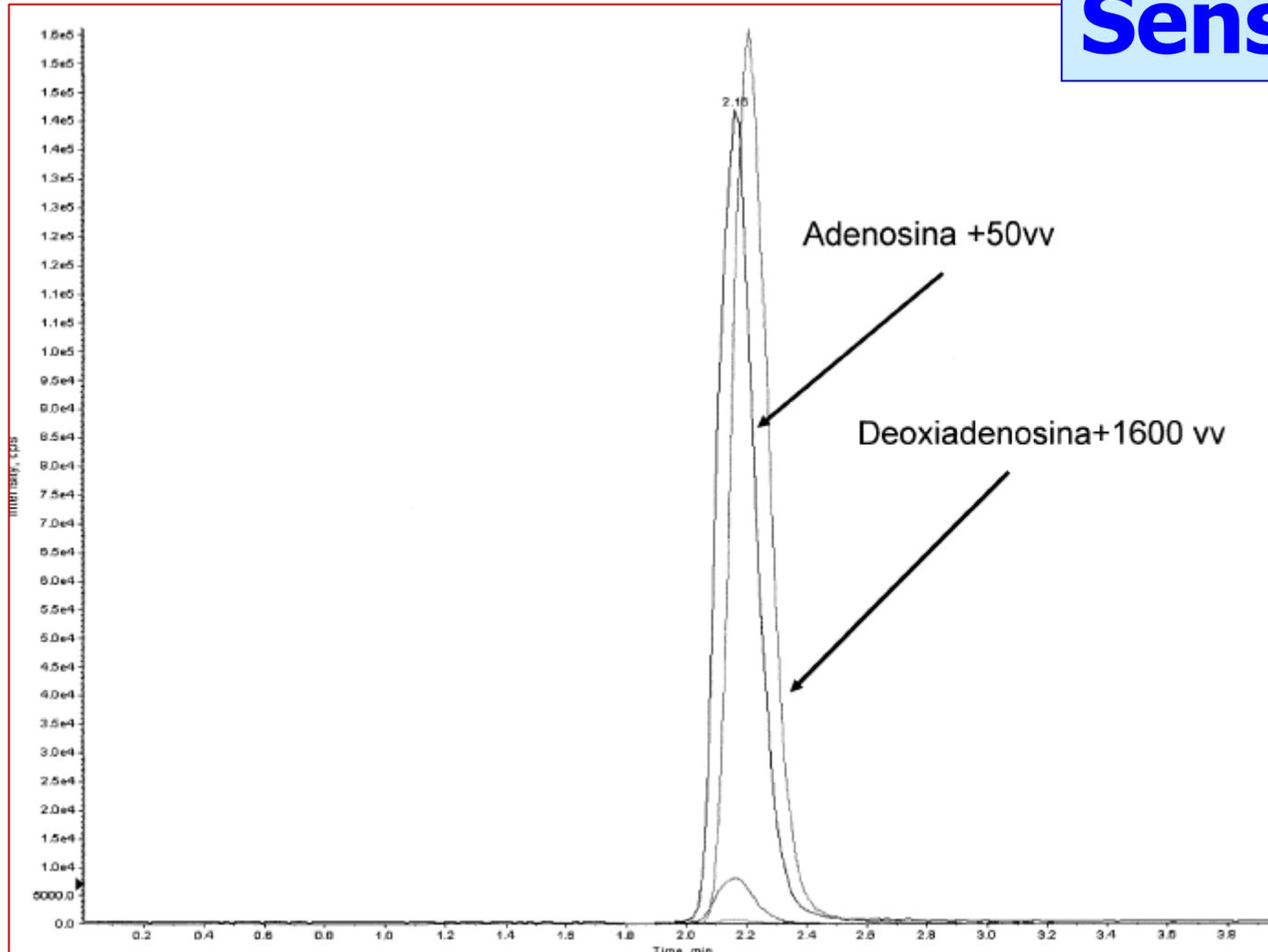
Sensitivity



The results

2

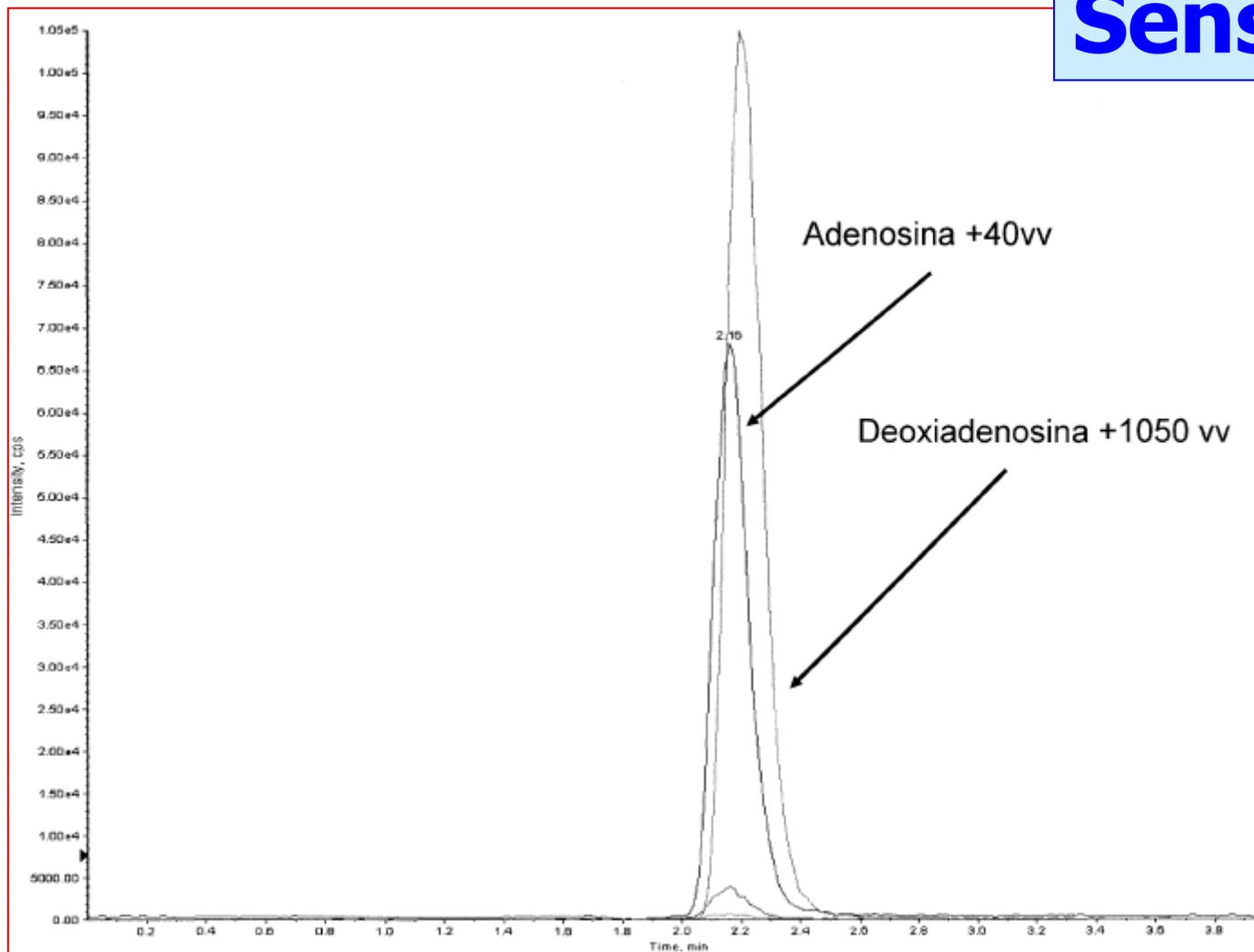
Sensitivity

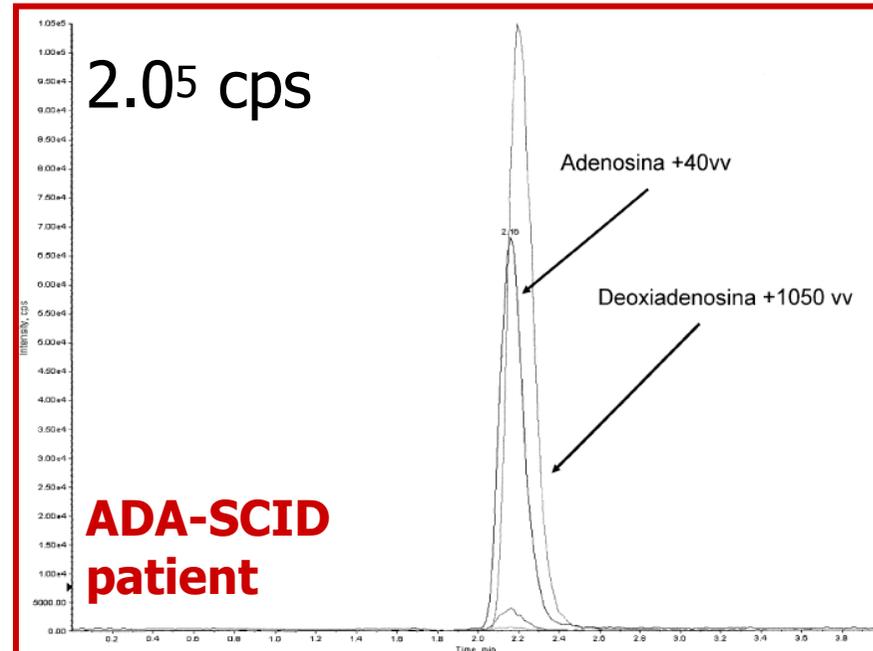
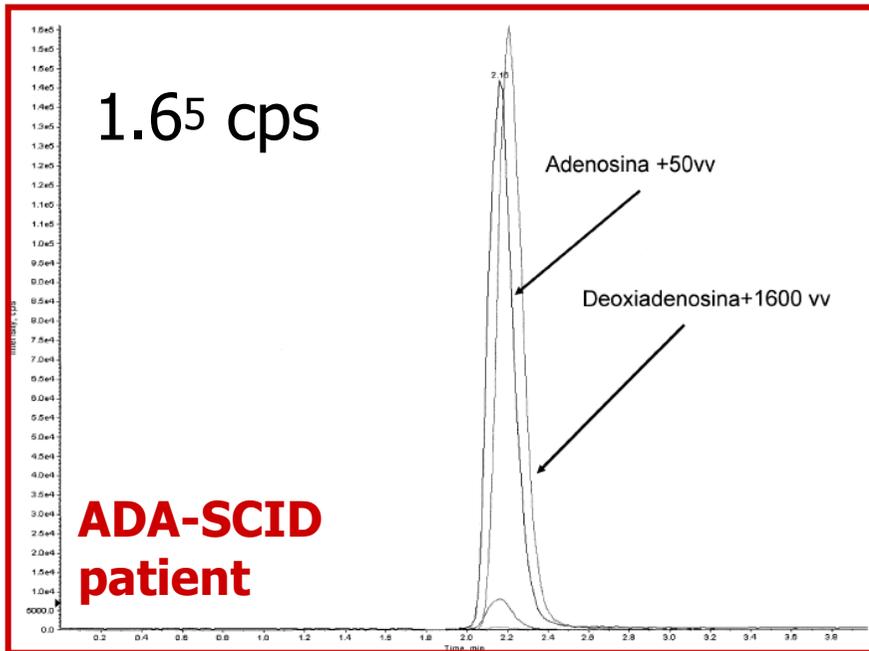
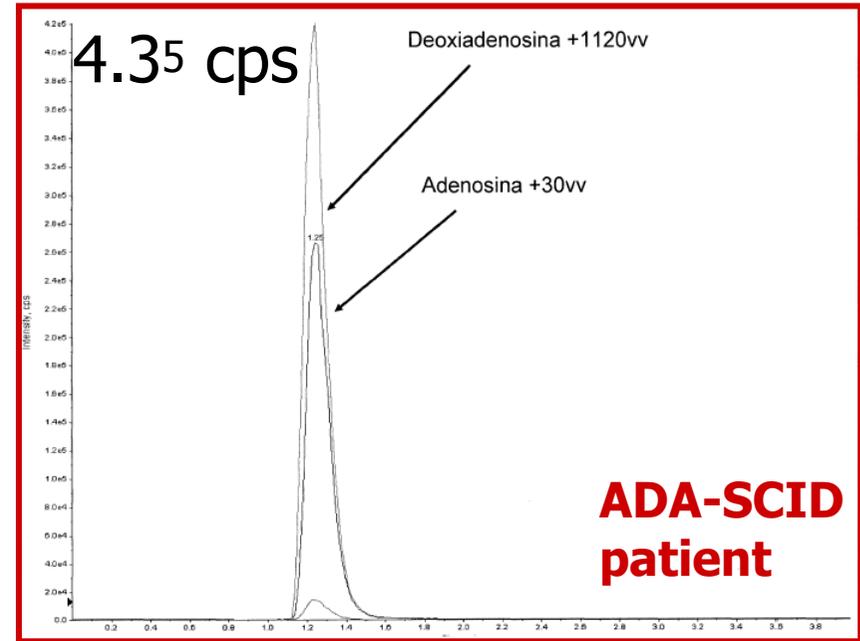
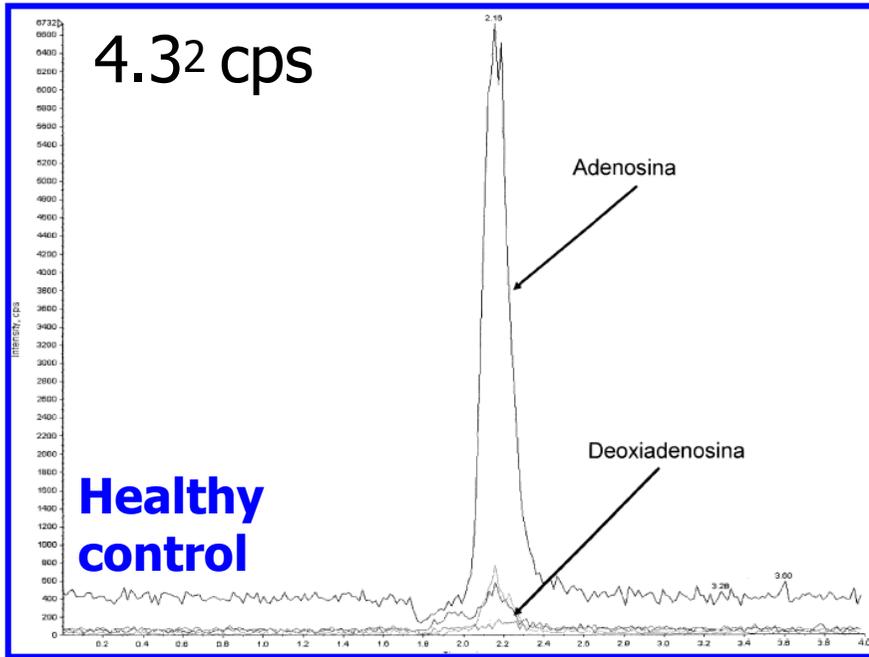


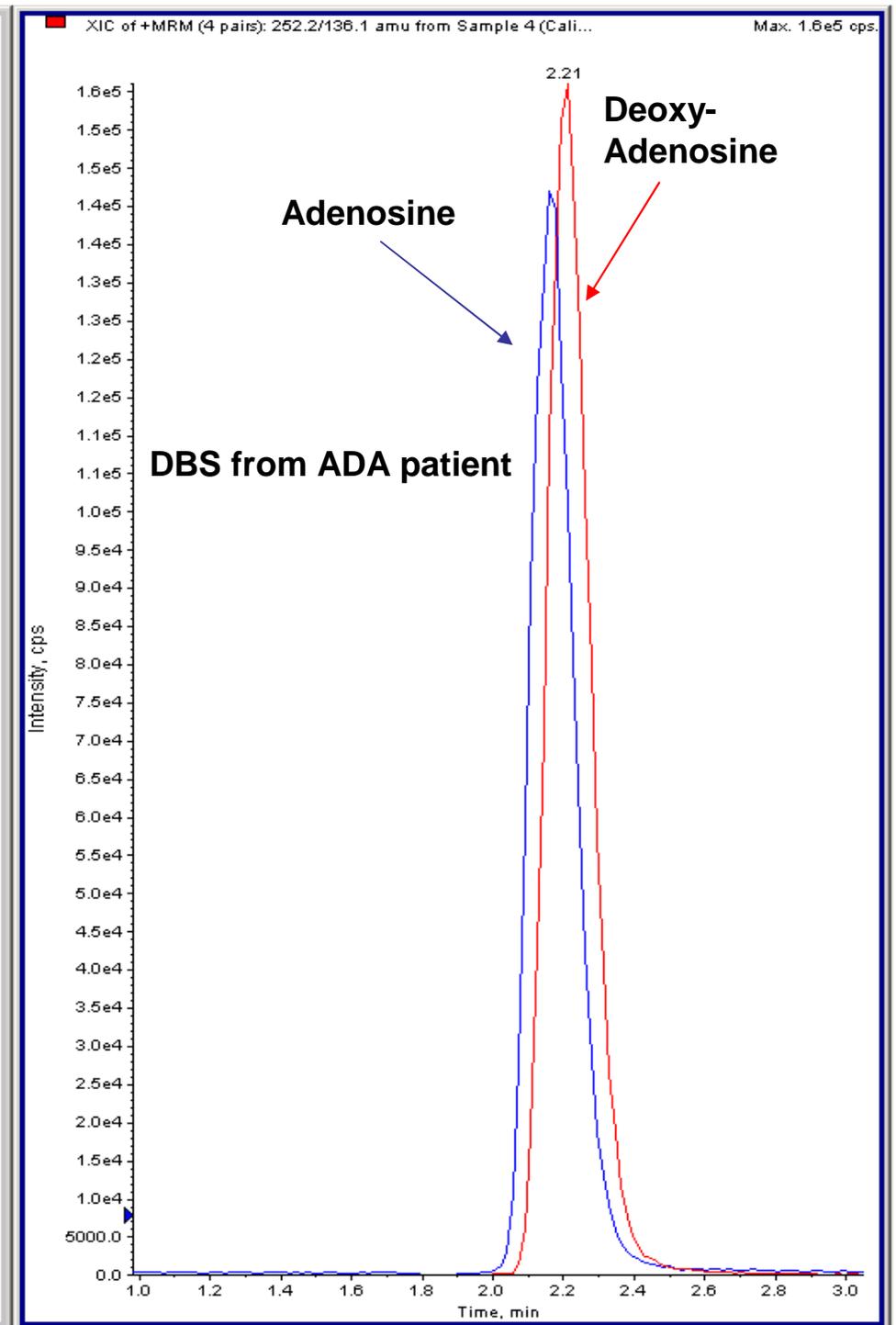
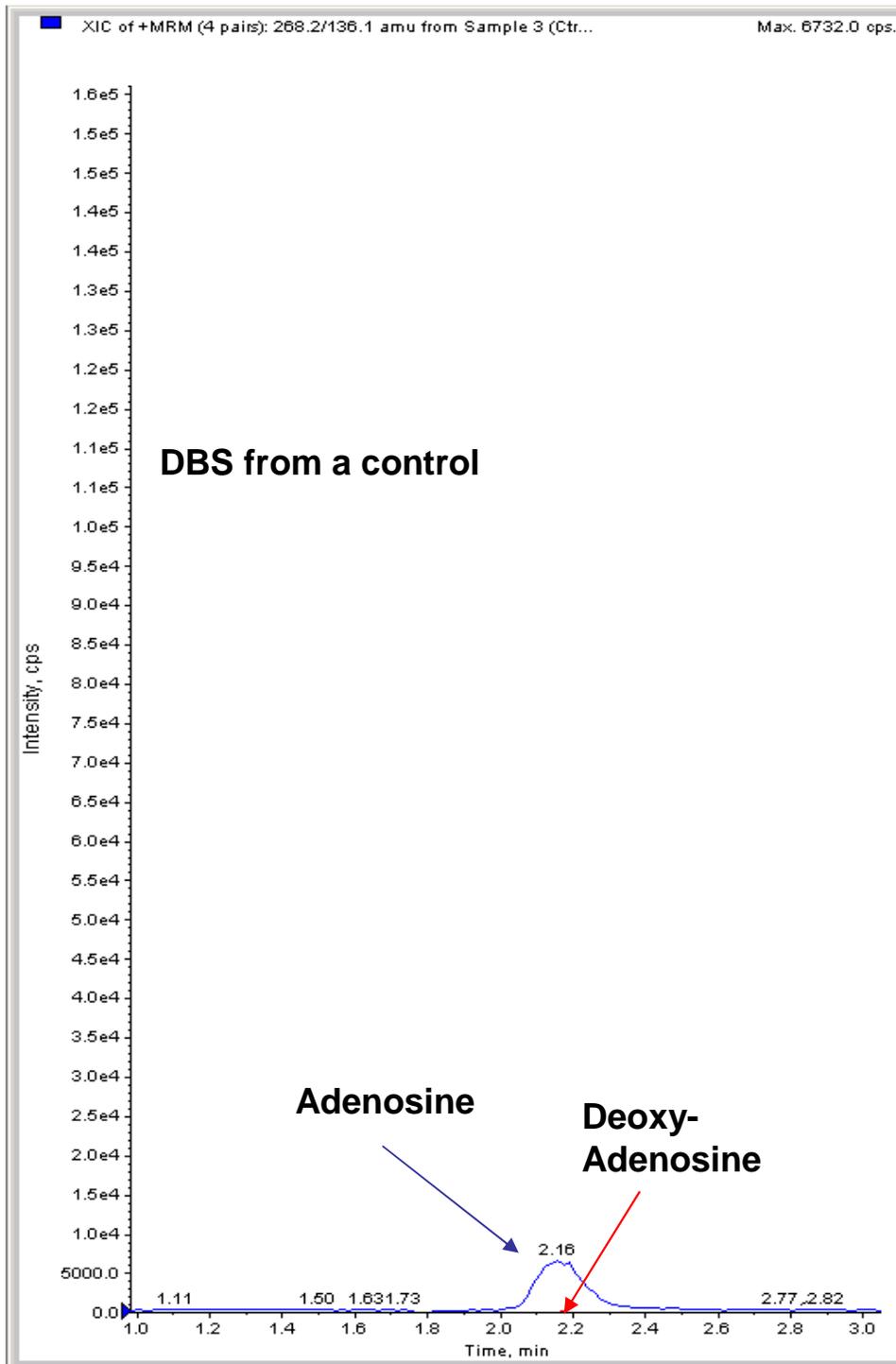
The results

3

Sensitivity







Secretary's Advisory Committee on Heritable Disorders in Newborns and Children

WASHINGTON – January 27, 2010



**In a historic vote on January 21, 2010, the Secretary's
Advisory Committee for Heritable Disorders in
Newborns and Children (ACHDNC)**

unanimously agreed

**to recommend the addition of Severe Combined
Immunodeficiency (SCID) to the
uniform newborn screening panel.**

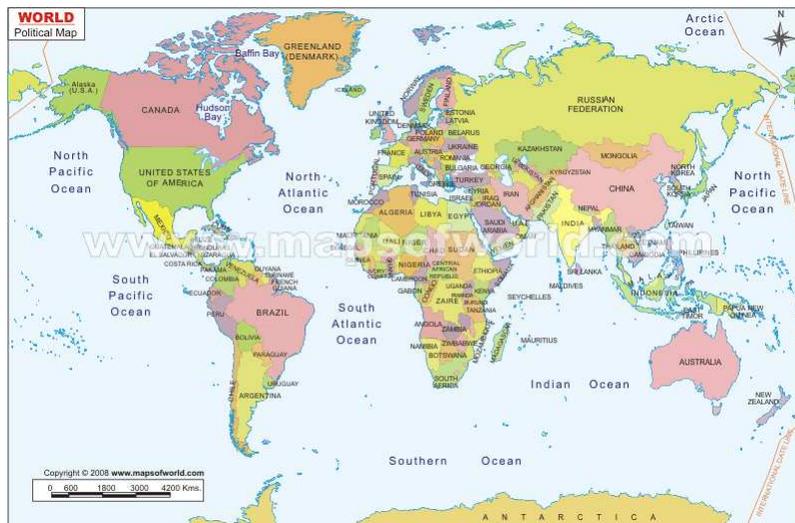
(Cost per test: not <10-15\$)

The cost

The cost per test is below 0.05 €

No extra-equipment is required

A commercial kit could be distributed in all countries where neonatal screening are performed



The cost

The frequency of ADA-SCID is now hypothesized around 1:1,000,000.

It is largely underestimated because of deaths in the first year of life due to severe infections before diagnosis of SCID

Preliminary data from Tuscany suggest an incidence of more than **1:50,000**



The frequency of other metabolic diseases included in the screening is in a similar range

Neonatal screening for severe combined immunodeficiency caused by an adenosine deaminase defect: A reliable and inexpensive method using tandem mass spectrometry

Chiara Azzari, MD, PhD,^{a,b} Giancarlo Ia Marca, PharmSc,^{b,c} and Massimo Resti, MD^{a,b} *Florence, Italy*

Background: Adenosine deaminase (ADA)-severe combined immunodeficiency (SCID) is an SCID caused by a defect in the enzyme adenosine deaminase. It is usually fatal in infancy because of severe recurrent infections. When diagnosis is made, permanent damage caused by infections or by metabolites is often present. Gene therapy, bone marrow transplantation, or enzyme therapy might be effective if performed early. ADA-SCID complies with all the criteria for inclusion in a newborn screening program. However, screening methods are still expensive or provide a non-negligible number of indeterminate results.

Objective: The aim of the present study was to develop a simple, reliable, and inexpensive method for diagnosis of ADA-SCID by using dried blood spot (DBS) samples taken at birth. Cost per test was calculated, including the cost for reagents, equipment, and operators.

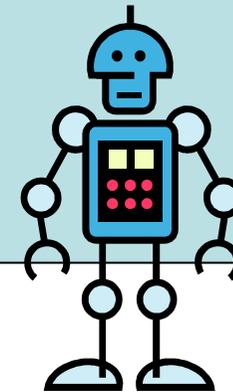
Methods: DBS samples from 4 patients with genetically confirmed ADA-SCID and 12,020 DBS samples from healthy newborns were examined. Adenosine and 2'-deoxyadenosine were tested by using tandem mass spectrometry (PCT EP2010/070517).

Abbreviations used

ADA: Adenosine deaminase
DBS: Dried blood spot
SCID: Severe combined immunodeficiency
TREC: T-cell receptor gene excision circle

Severe combined immunodeficiency (SCID) is a group of severe diseases that affect the immune system. Infants with SCID are healthy at birth but die of severe infections in infancy unless adequate therapy is provided.¹⁻³ Unfortunately, almost all infants with SCID are not identified in the preinfection period; when the diagnosis is hypothesized, usually a severe infection has just occurred. At that time, however, even though a correct therapeutic intervention is started, damages caused by the severe infections (eg, meningitis, encephalitis, and severe pneumonia) can already be present,¹⁻³ and permanent sequelae can be an important burden both for patients and families, as well as for the society. Moreover,

**NEW
PERSPECTIVES
FOR THE NEXT
FUTURE**



**The inclusion of
additional SCIDs
in NS panels
(PNP, Bruton etc)**



*THIS IS MATTEO LA MARCA,
WHO NEVER SLEEPS
(UNFORTUNATELY)*

THANK YOU!