12th EWGGD ZARAGOZA

The European Working Group on Gaucher Disease

Abstracts Book

Zaragoza · June 29th July 2nd · 2016

Abstract book

The European Working Group on Gaucher Disease, Zaragoza (Spain), on 29th June-2nd July 2016.

12th EWGGD ZARAGOZA

The European Working Group on Gaucher Disease

12th EWGGD ZARAGOZA

The European Working Group on Gaucher Disease

Table of contents

Welcome letter 5

Statements 7

Declaration of scientific interest by the official medical council of Zaragoza

Accreditation as continue education activity by the Aragon's Government

Committees 11

Letter of the Spanish Royal House
Honor Committee
Scientific Committee
Executive Committee
Local Committee

General information 19

Congress venue
Technical Secretariat
Badges
Exhibits
Shuttle bus services
Hotels

Program 23

Scientific Program

Invited Speakers presentation 31

Oral presentations at a glance 39

Poster presentations at a glance 79

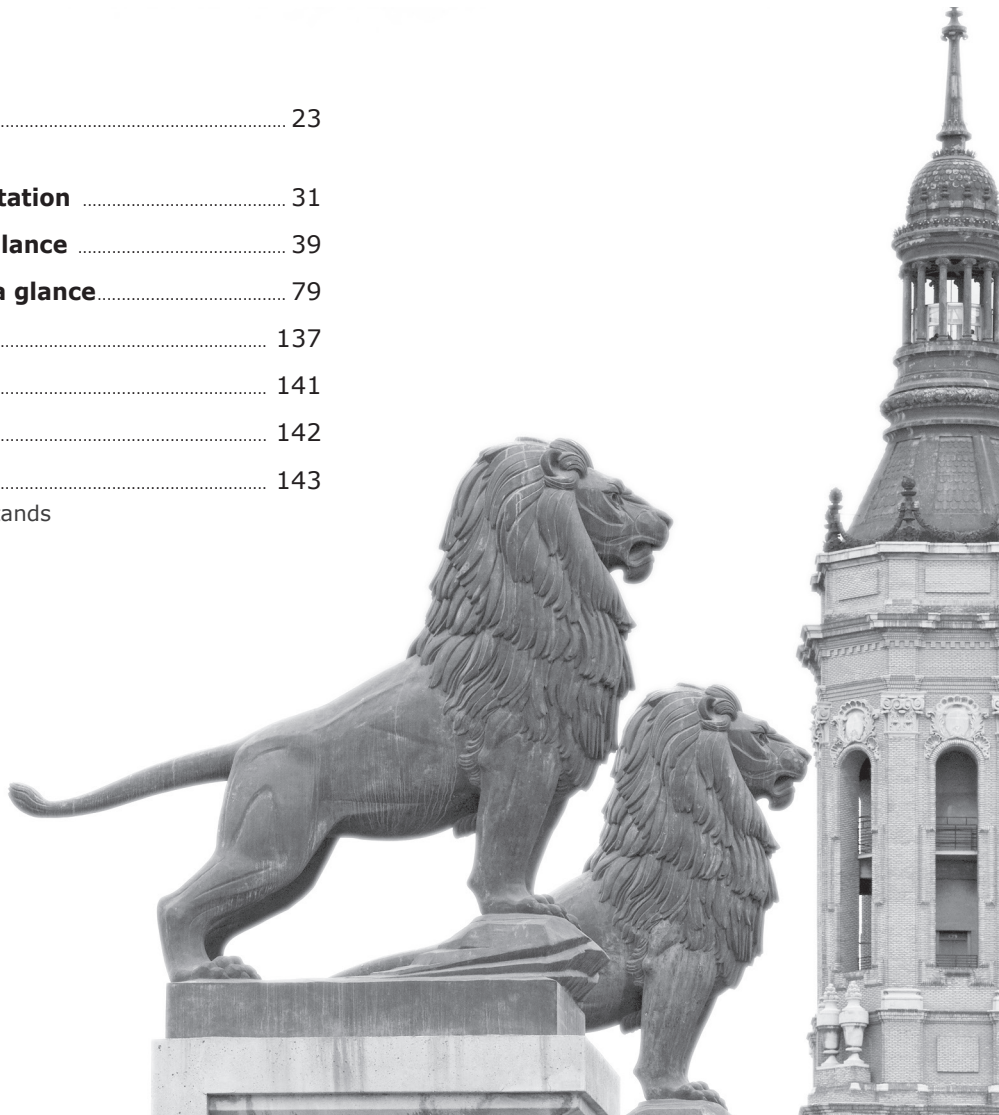
Participants list 137

Acknowledgments 141

Collaborators 142

Map & Floor Plans 143

Plans of Auditorium and Stands
Map of the hotels



Welcome letter

Dear delegates,

On behalf of the local organizing committee of The 12th EWGGD 2016 congress, I have the pleasure to give to all of you a warm welcome to the city of Zaragoza. Spain.

This edition joined more than 250 participants and attendants from around the world: The UK, Germany, Norway, Denmark, Latvia, Macedonia, Slovenia, Croatia, Estonia, Romania, Czech Republic, Italy, Greece, Finland, Sweden, Belgium, The Netherland, Bulgaria, France, Austria, Israel, Russia, Switzerland, Albania, Bosnia Herzegovina, Poland, Moldova Republic, Serbia, Spain, Jordan, Argentina, Brazil, Canada, USA, Paraguay, Mexico, Australia, India, China, Pakistan, South Africa. They will have the opportunity to share and discuss more than 80 works.

For this meeting, the Scientific Committee of EWGGD has carefully selected the most relevant aspects regarding Gaucher disease and the laters advances generated in the last two years in different aspects related to the pathophysiology, genetics, clinical features, associated complications, current and future disease therapies.

Several lectures distributed in eight sessions according to the pathophysiology, inflammation, diagnosis and monitoring, treatment of Gaucher disease, Parkinson's disease, bone disease and a special session related to Humanitarian aid by the initiative of the European Gaucher Alliance. The most relevant speakers on these topics will participate in these presentations and will undoubtedly serve for a good update on the latest basic and clinical research regarding the key processes of complex lysosomal metabolism and their translational projection.

Regarding the educational session with Professor Mistry and other members of EWGGD, they will be providing valuable information for specialists who are starting in their pathway of this rare entity and a refreshment for all.

As you can see, this city without losing its tradition of "ancient and immortal", maintains a mixture of cultures and monuments that characterize the city past history, and the various people who left their traces here and are part of our history (Jews, Muslims, French, American). Nevertheless, the city has been able to adapt to changes and it has generated infrastructure and architectural spaces that give it a special charm. Easy access to the city from different points of the Peninsula and the traditional friendliness and hospitality of the inhabitants will contribute to the participant enjoyment during the days of this congress.

It is our desire, as in previous editions the expectations deposited in this biennial congress, will be met therefore: organizers, delegates and exhibitors have placed their hope and desire that the congress will satisfy all participants.

Enjoy these days of the EWGGD Congress.

Pilar Giraldo
Host of EWGGD 2016

12th EWGGD ZARAGOZA

The European Working Group on Gaucher Disease

Statements

Declaration

Declaration of scientific interest by the official medical council of Zaragoza
(Declaración Interés Científico ICOMZ)

Declared of Scientific Interest

Accreditation

Accreditation as continue education activity by the Aragon's Government
(Actividad Formación Continua Gobierno de Aragón)

Accredited activity by the Ministery of Health and Ministery of Education and General Council of Oficial Medical College (B.O.E. Núm. 97,Sec. I. Pág. 36000) whit 1,8 credits

12th EWGGD ZARAGOZA

The European Working Group on Gaucher Disease

Committees


©VIRGILI

Letter

Letter of the Spanish Royal House (Carta de la Casa Real).

CASA DE S. M. EL REY

CREDENCIAL

Nº 145/2016

Su Majestad la Reina, accediendo a la petición que tan amablemente Le ha sido formulada, ha tenido a bien aceptar la

PRESIDENCIA DEL COMITÉ DE HONOR

del **«12º CONGRESO DEL GRUPO EUROPEO DE ENFERMEDAD DE GAUCHER (EWGGD)»**, que se celebrará en Zaragoza del 29 de junio al 2 de julio próximos.

Lo que me complace participarle para su conocimiento y efectos.

PALACIO DE LA ZARZUELA, 10 de mayo de 2016

EL JEFE DE LA CASA DE S.M. EL REY,

SEÑORA PRESIDENTA DEL COMITÉ ORGANIZADOR DEL CONGRESO.

ZARAGOZA

Honor Committee

President:

S.M. la Reina Doña Leticia

Committee members:

Excmo. Sr. D. Francisco Javier Lambán Montañés
President of the Autonomous Community of Aragon

Excmo. Sr. D. Pedro Santisteve Roche
Mayor of the City of Zaragoza

Excmo. Sr. D. Sebastián Celaya Pérez
Health Minister of the Aragon Government

Excmo. Sr. D. José Antonio Mayoral Murillo
Rector of the Zaragoza University

Sr. D. Ángel Lanas Arbeloa
Scientific Director of the Institute for Health Research Foundation Aragon (IIS Aragon)

Sra. Dña. Sandra García Armesto
Director of the Aragon Institute of Health Sciences (IACS)

Sr D. Pablo Lapunzina
Director Científico del CIBER de Enfermedades Raras (CIBERER)

Sr. D. Carlos Pérez Caseiras
Rector of San Jorge University

Sr. D. Juan Carrión Tudela
President of the Spanish Federation of Rare Diseases

Sra. Dña. Pilar Giraldo Castellano
President of the Spanish Foundation for the Study and Therapy of Gaucher Disease and Other Lysosomal

Scientific Committee

President:

Professor Stephan vom Dahl
Department of Gastroenterology,
Hepatology and Infectious Diseases,
University Hospital Dusseldorf (Germany)

Committee:

Professor Pilar Giraldo
Instituto Investigación Sanitaria Aragón,
CIBERER, Zaragoza (Spain)

Professor Mia Horowitz
Department of Cell Research and Immunology,
Tel Aviv University, Ramat Aviv (Israel)

Helen Michelakakis, PhD
Department of Enzymology and
Cellular Function, Institute
of Child Health, Athens (Greece)

Executive Committee

Prof. Stephan vom Dahl
University Hospital Dusseldorf
(Germany)

Helen Michelakakis
Institute of Child Health, Athens
(Greece)

Prof. Hans Aerts
Academic Medical Center, University of Amsterdam
(The Netherlands)

Nadia Belmatoug
Assistance Publique- Hôpitaux de Paris
(France)

Prof. Mia Horowitz
Tel Aviv University
(Israel)

Jeremy Manuel, OBE
European Gaucher Alliance
(EGA)

Hanna Rosenbaum
Center for Gaucher disease for the north of Israel, Haifa
(Israel)

EWGGD 2016 host: Pilar Giraldo, PhD
Instituto Investigación Sanitaria Aragón, Zaragoza
(Spain)

Marieke Biegstraaten
Academic Medical Center, University of Amsterdam
(The Netherlands)

Local Committee

President:
Pilar Giraldo Castellano

Vicepresident:
Mercedes Roca Espiau

Coordinator**:**
Marcio Andrade Campos

Vocals:
Jose Luís Capablo Liesa
Jorge Cebolla Sanz
Ignacio de Blas Giral
Beatriz Escuder Azuara
Carlos Gómez Notario
Pilar Irún Irún
Paz Latre Martinez
Laura López de Frutos
Blanca Medrano Engay
Concepción Pérez Valero
Teresa Pérez Valero
Ana Roy Yaiza
Belén Tisner Giraldo
Daniel Toro Begue

12th EWGGD ZARAGOZA

The European Working Group on Gaucher Disease

General Information

Congress venue

Auditorio de Zaragoza

C/ Eduardo Ibarra nº 3 - 50009 Zaragoza
Teléfono: 976 72 13 00
www.auditoriozaragoza.com

Technical Secretariat

Viajes El Corte Ingles S.A.

División de Congresos, Convenciones e Incentivos
Avda. Cesar Augusto nº 4 - 50004 Zaragoza
Teléfono: 976 46 96 28
EWGGD2016@viajeseci.es

Badges

The use of the official badge of the congress is mandatory to access the scientific sessions, exhibits and participated in the networking meetings.

Exhibits

An special area is available for exhibits with the latest information provided by the sponsors of the congress.

Cloakroom

For your convenience a cloakroom service will be available in the technical secretariat area.

Shuttle Bus Services

Shuttle buses depart from the Hotel 30 minutes earlier to start Meeting Session in the morning and return at the end of the last session to Hotels. In addition there is a bus line (C1) every 8 minutes with a stop close to the Boston Hotel and leave to the Auditorium in 20 minutes.

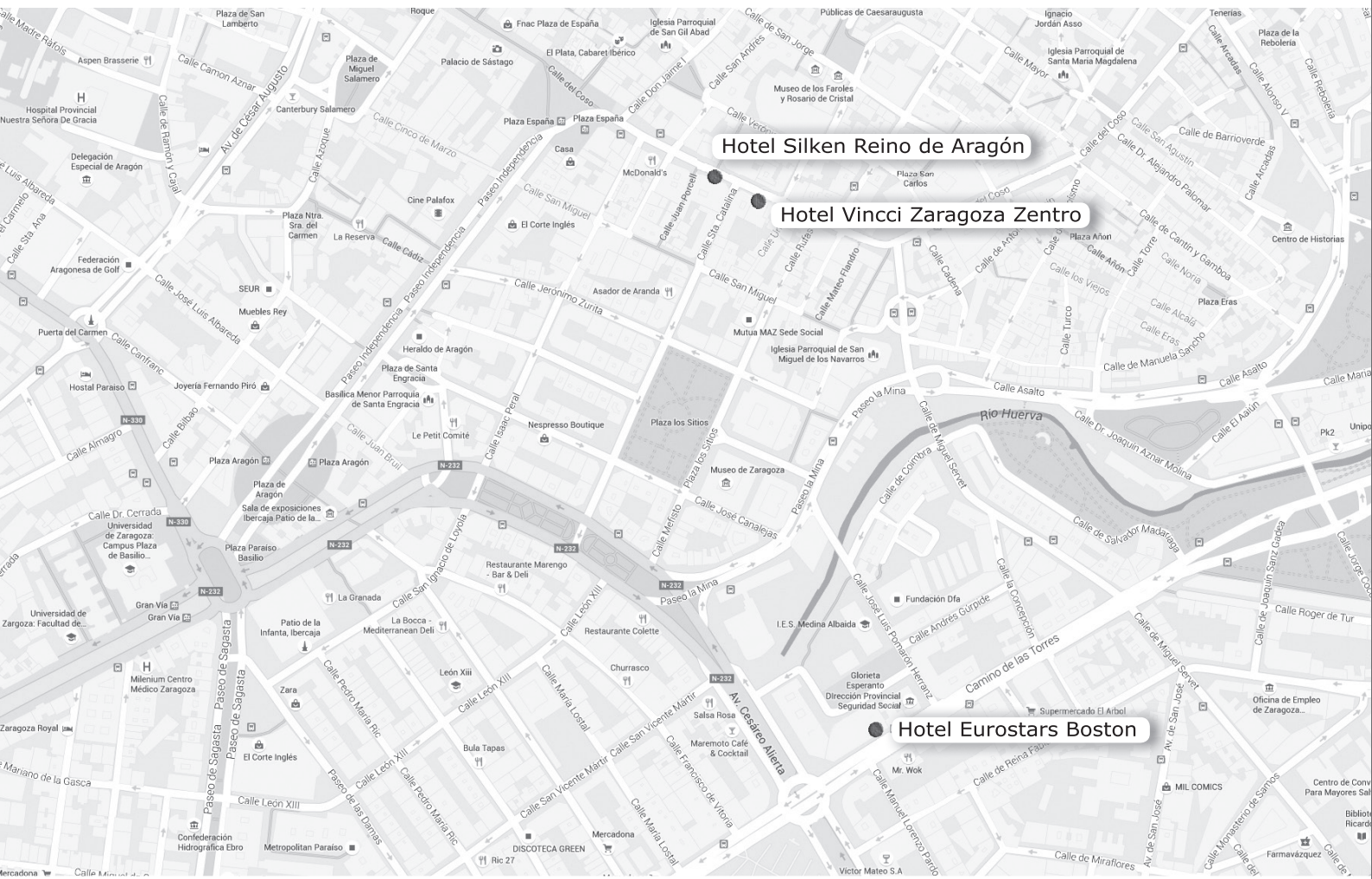
In addition there is a bus line (C1) every 8 minutes with a stop close to the Boston Hotel and leave to the Auditorium in 20 minutes. To return from Auditorium to Boston Eurostar Hotel the bus line (C2) stop is close the Auditorium in C/ Violante de Hungría.

Hotels

Hotel Eurostars Boston ****
Avda las Torres, 28, 50008 Zaragoza
Teléfono: **976 59 91 92**

Hotel Silken Reino de Aragón ****
Calle del Coso, 80, 50001 Zaragoza
Teléfono: **976 46 82 00**

Hotel Vincci Zaragoza Zentro ****
Calle del Coso, 86, 50001 Zaragoza
Teléfono: **976 70 33 00**



EWGGD ZARAGOZA

The European Working Group on Gaucher Disease

Program

Thursday, June 30th

08:00 -09:30	**Session 1: Opening session** *Chairs: Pilar Giraldo, Stephan vom Dahl*n
08:00- 08:30	Welcome
08:30- 09:10	Invited lecture: J. Goldblatt - Genetic aspects of GD
09:10-09:50	**Session 2: Gaucher disease, iron and inflammation** *Chairs: Pilar Giraldo, Stephan vom Dahl* Invited lecture: M. Cappellini - Iron in GD. Italy
09:50 - 10:20	Coffee break
10:20 -11:40	**Session 2: Gaucher disease, iron and inflammation** *Chairs: Pilar Giraldo, Stephan vom Dahl* Oral presentations:
10:20-10:40	Cellular models for monocyte/machrophage studies *Mario de la Mata. Spain*
10:40-11:00	Hyperferritinemia and serum inflammatory cytokines in adult patients with Gaucher disease type 1 *F. Lorenz. Sweden*
11:00-11:20	"Fishing" new molecular targets to gain further insights into the molecular pathogenesis of bone alterations in Type I Gaucher disease (GD1) *Roberto Costa. Italy*
11:20-11:40	Deficiency of monocyte/macrophage KCa3.1-functions in type-1/3 Gaucher disease *Aida Olivan. Spain*
11:40 -12:40	**POSTER Tour I** Chairs: Argiris Symeonidis, Andrea Dardis
12:10 - 13:00	Lunch
13:30 -15:15	**Session 3: Pathophysiology** *Chairs: Lluisa Vilageliu, Hellen Sidransky* Oral Presentations:
13:30-13:50	The fruit fly Drosophila melanogaster as a model system to study Gaucher disease *Or Cabasso. Israel*
13:50-14:15	The contribution of mutant gcase to the aggregation and accumulation of α-synuclein/ The contribution of mutant GBA to the development of Parkinson's disease in Drosophila *Gali Maor. Israel*
14:15-14:35	Patient-derived iPSC provide insights into pathophysiology of Gaucher disease *Ola Awad. USA*

Thursday, June 30th

14:35-14:55	The role of mTor and autophagy in lysosomal storage disease: prospective for therapeutics *Suleiman Igdoura. Canada*
14:55-15:15	Generation of mesenchymal and osteoblastic cellular models for Gaucher disease using human induced pluripotent stem cells *Jenny Serra-Vinardell. Spain*
15:15-15:30	Coffee break
15:30-17:30	**Educational Session** *Chair: Pramod Mistry* Diagnosis and typical/atypical manifestations of GD1. Pramod Mistry Biomarkers. Pilar Giraldo *Neuronopathic GD. Eugen Mengel* Evolving therapeutic landscape in GD. Hanna Rosembaum
17:30-19:00	Business meeting (Members EWGGD)

Friday, July 1st

08:00- 09:45	**Session 4: Diagnosis and monitoring** Chairs: *Ari Zimran, Pramod Mistry*
08:00-08:40	Invited lecture S. Karlsson - Gene therapy. Sweden
08:40 -10:40	Oral Presentations
08:40-09:00	*Proof-of-principle: rapid noninvasive prenatal diagnosis for type 1 Gaucher disease* *Gheona Altarescu. Israel*
09:00-09:25	*Children with type 1 Gaucher disease: changing profiles in the 21st century / Growth parameters in pediatric patients with Gaucher disease: 15-year follow-up from one Israeli clinic* *TBA. Israel*
09:25-09:45	*Benign and malignant hematological complications of Gaucher disease: diagnosis, monitoring and treatment* *Hanna Rosenbaum. Israel*
09:45 - 10:05	Coffee break
10:05 -11:05	**Session 4: Diagnosis and monitoring** *Chairs: Ari Zimran, Pramod Mistry* Oral Presentations
10:05-10:25	Management goals for type 1 Gaucher disease: a consensus document from the European Working Group on Gaucher Disease *Marieke Biegstraaten. The Netherlands.*
10:25-10:45	Revised Belgian guidelines for the diagnosis, treatment and monitoring of Gaucher's disease *François Eyskens. Belgium*
10:45-11:05	Lyso-Gb1 As a Gaucher-specific biomarker: data from clinical trials using velaglucerase alfa *Deborah Elstein. Switzerland*
11:05-12:05	**Session 5: Treatment** *Chairs: G Pastores. Maja di Rocco.* Oral Presentations
11:05-11:25	In Vivo cellular pathways for infused human recombinant glucocerebrosidase *Margarita M Ivanova. USA*
11:25-11:45	Intravenously administered gene therapy for the treatment of neuronopathic Gaucher disease *Giulia Massaro. UK*
11:45-12:10	Long-term safety and efficacy of Taliglucerase Alfa in Pediatric Patients with Gaucher Disease Who Were Treatment-naïve or Previously Treated with Imiglucerase / Long-term efficacy and safety results of Taliglucerase alfa through 5 years in adult treatment-naïve patients with Gaucher disease *Ari Zimran. Israel*
12:10 - 13:00	Lunch
13:00 -13:45	**POSTER Tour II** *Chairs: Maciej Machaczka, Hanna Rosenbaum.*

Friday, July 1st

13:45 -15:05	**Session 5: Treatment** *Chairs: G Pastores, Maja di Rocco*
13:45-14:25	Invited lecture: *C. Hollak. The Netherlands- Treatment options in 2016: clinical view/a proposal for an algorithm* Oral Presentation
14:25-14:45	*CNS-accessible inhibitor of glucosylceramide synthase for substrate reduction therapy of neuronopathic Gaucher disease* *John Marshall. USA*
14:45-15:05	*Four-year follow-up from the encore trial: oral eliglustat in patients with Gaucher disease type 1 stabilized on enzyme therapy* *Ozlem Goker-Alpan. USA*
15:05 - 15:20	Coffee break
15:20 -16:20	**Session 5: Treatment** *Chairs: G Pastores, Maja di Rocco.* Oral Presentation
15:20-15:40	Transformation in pre-treatment presentations of Gaucher disease in first 2 decades of Imiglucerase enzyme replacement therapy: report from the international collaborative Gaucher group Gaucher Registry *Pramod Mistry. USA*
15:40-16:00	Development of a new therapeutic strategy for Gaucher disease (GD) using ex vivo targeted genome editing. *Fabiola Porro. Italy*
16:00-16:20	pH-sensitive glucocerebrosidase ligands as non-inhibitory pharmacological chaperones for Gaucher disease *José M. García Fernández. Spain*
16:20-18:00	**Session 6: Humanitarian Aid Session** *Chair: Pascal Niemeyer*
16:20-16:40	EGA Updates. Pascal Niemeyer
16:40-16:50	The EGA's role in supporting the growing unmet need of Gaucher patients globally that cannot access treatment in their own country *Tanya Collin-Histed (EGA)*
16:50-17:10	*Working within the Governments capabilities to support Gaucher patients in Pakistan; an alternative strategy* *Prof Huma Cheema. Pakistan*
17:10-17:30	Managing the symptoms of Gaucher patients without access to ERT in India *Dr Sujatha Jagadeesh. India*
17:30-17:50	A practical experience of dealing with two siblings from Jordan & the German experience of managing refugees with Gaucher disease in Germany with Gaucher Disease *Dr Jörg Reinke. Germany*
17:50-18:00	Questions & Answers

Saturday, July 2nd

08:00 -10:40	**Session 7: Parkinson disease** *Chairs: Ozlem Goker-Alpan. Marieke Biegstraaten*
08:00-08:40	Invited lecture: M. Horowitz - Molecular mechanisms underlying the association between Gaucher disease and Parkinson disease.
08:40-9:00	Oral Presentations Glucosylceramide synthase inhibition reduces alpha-Synuclein pathology and improves cognition in murine models of synucleinopathy *S. Pablo Sardi. USA*
09:00-09:20	GBA1 haploinsufficiency in a Parkinson mouse model results in an earlier disease onset and more rapid progression of symptoms *Nahid Tayebi. USA*
09:20-09:40	Processing of alpha-Synuclein and Parkin in peripheral blood mononuclear cells in patients with Gaucher disease *Renuka Pudi Limgala. USA*
09:40-10:00	Studying the link between Gaucher and Parkinson's Disease. Cell models and disease-modifying factors *Derek Burke. UK*
10:00-10:20	Glucocerebrosidase variants ARE modifiers for onset and clinical symptoms in Parkinson disease patients – a multinational study of GBA gene sequences using Next Generation Sequencing (NGS) *Rike Bender. Germany*
10:20-10:40	Unbiased approach reveals higher risk for PD in carriers of severe vs mild GBA mutations *David Arkadir. Israel*
10:40 - 11:00	Coffee break
11:00 -13:20	**Session 8: Bone disease** *Chairs: Mercedes Roca. Paula Rozenfeld*
11:00- 11:40	Invited lecture: Nerea Alonso Lopez. Genetic of osteoporosis. Rheumatology and Bone Disease Unit Centre for Genomic and Experimental Medicine, IGMM. University of Edinburgh. UK Oral Presentations
11:40-12:00	Measurement of bone marrow fat fraction with gradient-echo MR imaging in Gaucher disease patients; a practical alternative to Dixon quantitative chemical shift imaging *M. Regenboog. The Netherlands*
12:00-12:20	Osteocyte alterations contribute to bone pathology in Gaucher disease *Juan Marco Mucci. Argentina*

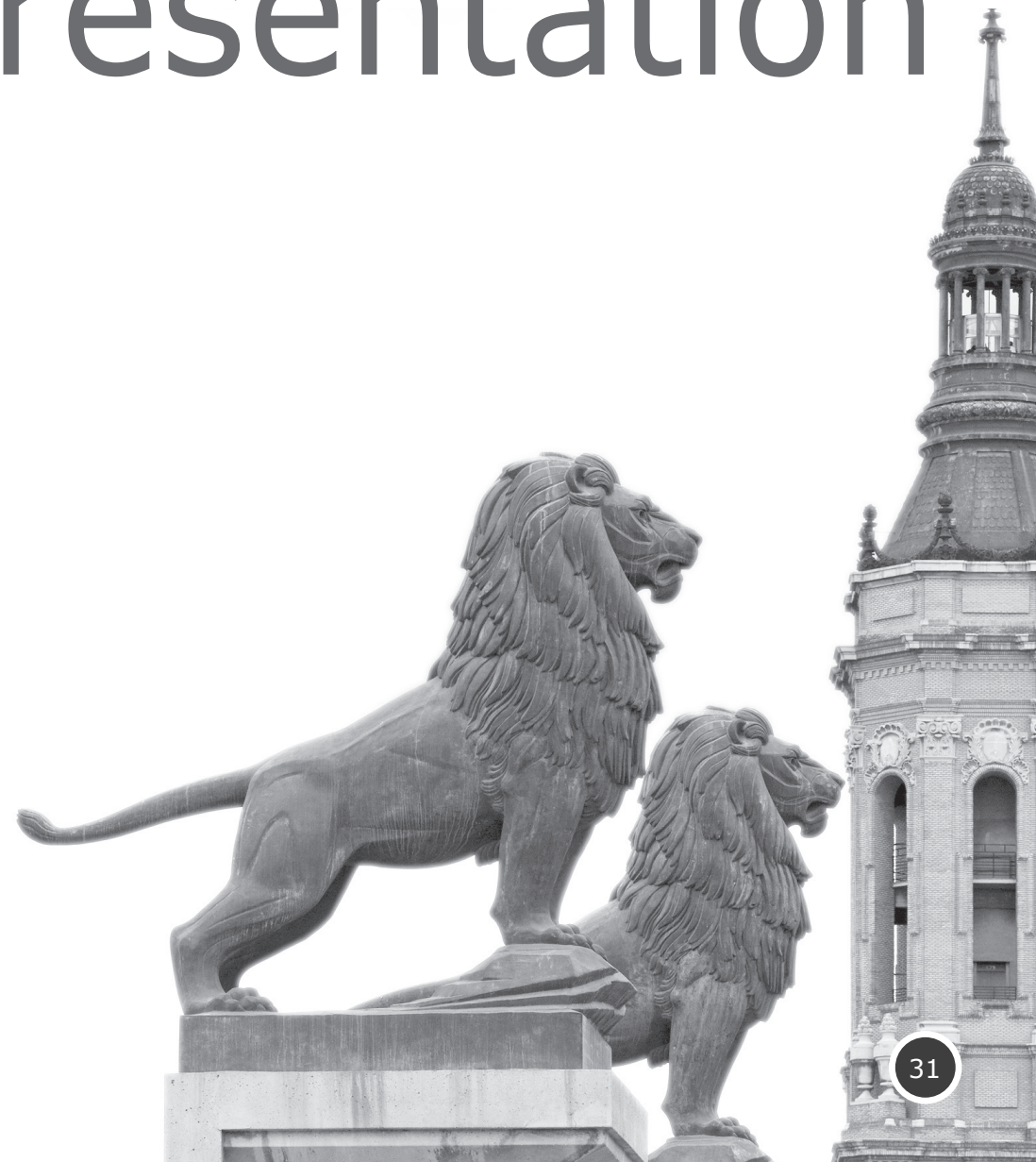
Saturday, July 2nd

12:20-12:40	Effect of two different therapeutic interventions: SRT in comparison to ERT on immune aspects and bone involvement in Gaucher disease *Renuka Pudi Limgala. USA*
12:40-13:00	Risk factors for fractures in Imiglucerase-treated Gaucher disease type 1 patients in the ICGG Gaucher registry *Stephan vom Dahl. Germany*
13:00-13:30	Summary, Poster prices, "EGA young doctor Award" and Invitation to the next EWGGD meeting
13:45	End of the meeting

12th EWGGD ZARAGOZA

The European Working Group on Gaucher Disease

Invited Speakers Presentation



Clinical Professor **JACK GOLDBLATT**

AM, MB ChB, MD, FCP, FRACP (Clin. Geneticist, HGSA)

Director of Genetic Services and
the Familial Cancer Programme of Western Australia.

He has been president of the Human Genetics Society of Australasia and a foundation board member of the International Federation of Human Genetics Societies. He has co-authored 250 papers in international, peer reviewed, journals, including 28 on various aspects of Gaucher disease. He has a doctorate thesis on the biochemical and clinical aspects of Gaucher disease.

He spent two years as a visiting instructor in the medical genetics unit at Mount Sinai Hospital in New York, U.S.A., with a significant involvement in their Jewish genetic disorders metabolic programme. Prof. Goldblatt is a specialist physician and medical geneticist who has worked in the academic field of human genetics since 1975 with research based publications on aspects of clinical, biochemical and molecular genetics. He is particularly interested in the natural course of inherited disorders of metabolism and ran a national Gaucher disease management programme in South Africa over a 13 year period. He was chairman of the Gaucher Disease Advisory Committee which interacted with the Australian Federal Health Department's Life Saving Drugs Program to manage government-subsidised therapy for Gaucher patients. In 2011 he was made a Member of the Order of Australia in the General Division (AM) in the Queen's Birthday Honours List for his service to medicine in the area of human genetics as a clinician and researcher. He has run 70 marathons with a personal best time of 2 hours 55 minutes.

TYPE 1 GAUCHER DISEASE: CLINICAL GENETIC MYTHS AND UNSOLVED ISSUES

Dr Jack Goldblatt, Director Genetic Services and Familial Cancer Program of Western Australia, King Edward Memorial Hospital, Clinical Professor, School of Paediatrics and Child Health, University of Western Australia.

Type 1 Gaucher Disease manifests a considerable clinical variability from severely affected young children to relatively asymptomatic septuagenarians. This variable expression is not explained by beta-glucerebrosidase allelic heterogeneity. This discordance is exemplified by the finding of variable severity in siblings with the same beta-gluc haplotypes. An 11-year old boy had significant haematological and skeletal complications, with a "shepherds crook" femoral deformity, while his 21 year old sister is asymptomatic with minimal splenomegaly and a normal skeletal MRI. Numerous reviews claim that skeletal complications respond sub-optimally to ERT. Our experience on matching ERT dosage to MRI based assessment of skeletal involvement has resulted in no patient experiencing avascular necrosis of femoral or humeral head on therapy. It is particularly important that the MRI is done according to a standardised format and all images are read centrally by a limited number of specifically skilled radiologists.

The MRI reviews should include narrative reports of changes over time as well as using the reproducible semi-quantitative scoring systems. Bone marrow imaging is a vital component of the care of patients and the BMB score is a reliable, semi-quantitative and reproducible technique for evaluating severity and response to therapy and descriptive imaging adds to interpretation. The BMB score has a greater range of signal intensity values assessed than in other scoring systems and is based on the pattern of disease. In terms of skeletal symptoms "bone pain" needs to be differentiated from non-specific limb pain to truly document skeletal complications. Selected patients on long-term ERT who are stable and "debulked" can be considered for altered regimens with prolonged intervals between infusions to suit life-style considerations.

A review of 22 patients, meeting specific criteria, who stopped ERT for up to 6 months due to the drug shortage were shown not to have developed irreversible complications at follow-up at least 2 years after recommencing therapy, and most parameters had returned to "pre-stoppage" levels. Further evidence for this were the data from the placebo group in the Cerdelga ENGAGE trial that showed no significant deterioration over a 9 month period.

Despite more than 20 years of ERT for GD numerous issues remain partially unsolved including: Predicting the natural history of an individual with untreated Gaucher disease (particularly important in the era of Newborn screeningNBS); optimal individualised dosage at different phases of disease; appropriate age of ERT initiation in pre-symptomatic patients; determinants of potential response; effect of ERT on type 3 disease and dosing effect on neurological, pulmonary and skeletal manifestations; application of biomarkers to clinical practice; individualised risk of co-morbidities such as Parkinson's disease; long-term effect of treatment interruptions; role of other/new therapies such as SRT vs ERT; modifying factors on phenotype (epistasis, epigenetic, environmental), why multiple LSDs in the same pathway are all more common in Ashkenazi-Jews.

MARIA DOMENICA CAPPELLINI, MD

Department of Internal Medicine University of Milan, and Policlinico Foundation IRCCS, Milan (Italy)

Professor Cappellini qualified as an MD in 1974 at the University of Milan, Italy. She is Professor of Internal Medicine at the University of Milan and Chief of the Rare Disease Centre at the Policlinico Foundation Hospital, Milan (Italy)

She is a scientific adviser to the Thalassaemia International Federation and a member of the European Gaucher Registry and the Monitoring Committee for Genz-112638. Prof. Cappellini has been an active researcher in the field of rare diseases, including thalassaemia, haemoglobinopathies, porphyrias and lysosomal diseases (mainly Gaucher and Fabry) for over 20 years. She has focused on the phenotypic expression of thalassaemia intermedia and identified the mechanisms underlying the thrombotic risks associated with the disease. She is involved in clinical trials of new iron chelators and has focused recent studies on iron dysregulation in Gaucher disease.

She has published 402 peer-reviewed original articles and is a regular invited speaker national and international meetings. Prof. Cappellini is a member of a number of Societies, including the European Hematology Association (EHA), the Italian Society of Hematology (SIE), the American Society of Hematology (ASH), the Italian Society of Internal Medicine (SIMI), and the European Iron Club. She is Scientific Adviser to the Thalassaemia International Federation (TIF) and contributes to their meetings to establish guidelines for the clinical management of thalassaemia. Professor Cappellini is Past President of the European Federation of Internal Medicine.

She is Honorary Fellow of RCP and ACP.

SESSION: GD IRON AND INFLAMMATION

Maria Domenica Cappellin – Fondazione Ca Granda Policlinico IRCCS, Università di Milano

Approximately 50% of type 1 Gaucher patients have increased levels of ferritin at diagnosis or during the disease course. No clear explanation has been so far provided to explain ferritin increases and particularly it is not proved that Gaucher patients have iron overload. Ferritin is a well known iron storage protein however it is also an acute phase reactant, therefore it remains unclear if increased ferritin levels in Gaucher patients indicate iron overload or an iron homeostasis disregulation or if it is simply an indicator of chronic inflammatory status. The control of iron homeostasis acts at both the cellular and the systemic level and involves a complex system of different cell types, transporters, and signals. In the scenario of iron regulation it is possible that in Gaucher disease, iron absorption and macrophage iron release are modulated by signalling provided by increased levels of IL-6 rather than directly by glucosylceramide loaded macrophages.

Increased ferritin levels in Gaucher disease could simply express a chronic inflammatory status or could be a consequence of hepcidin-related altered iron homeostasis. There have been some observations that enzyme therapy reduces the abnormally high levels of ferritin supporting that ferritin could be a response to inflammation and not an indicator of body iron burden.

In the new scenario of iron homeostasis, the iron status in Gaucher's disease deserves to be explored in order to get further insights in the patophysiology of increased ferritin levels as well as of iron stores in macrophages. Specifically, considering that macrophages are key cells either in Gaucher's disease than in iron trafficking, a blockade in the release of Gaucher-cell iron could be hypothesized. Different potential mechanisms to explain the iron status in Gaucher disease will be discussed.

STEFAN KARLSSON

MD, PhD

Lund University and Lund University Hospital, Lund, Sweden

Stefan Karlsson is Professor of Molecular Medicine and Consultant Physician at Lund University Hospital, Sweden, where he has been based since 1995. Professor Karlsson completed his MD degree at The University of Iceland and received his PhD at University College London. Subsequently, Dr. Karlsson trained as a postdoctoral fellow in Arthur Nienhuis's laboratory at the National Institutes of Health (NIH) and thereafter served as an independent investigator and Chief of the Molecular and Medical Genetics Section in the Developmental and Metabolic Neurology Branch, The National Institute of Neurological Disorders and Stroke, NIH, Bethesda, MD. Dr. Karlsson has a broad interest in hematology, particularly experimental hematology and translational research in hematology. Prof. Karlsson's research is focused on understanding the regulatory pathways involved in hematopoietic stem cell (HSC) self-renewal, survival and expansion and the application of HSCs to development of cell and gene therapies to treat genetic disorders. Professor Karlsson is active on several Scientific Review Boards, and is or has been an Editorial Board member for numerous journals in the field of hematology, including Blood, Stem Cells, European Journal of Haematology, International Journal of Hematology and Experimental Hematology. He is the author of approximately 200 research publications, the majority of which are original research contributions. In 2009, Professor Karlsson was awarded The Tobias Prize by The Royal Swedish Academy of Sciences. Stefan Karlsson served on the Board of The International Society of Experimental Hematology (ISEH) for many years and was the President of ISEH for the year of 2005/2006. He also serves on the Committee for Hematopoiesis, American Society of Hematology and is a member of the Scientific Program Committee of the European Heamatology Association

LENTIVIRAL GENE THERAPY USING CELLULAR PROMOTERS CURES TYPE 1 GAUCHER DISEASE IN MICE

Maria Dahl, Alexander Doyle, Karin Olsson, Jan-Eric Månsson, André R.A. Marques, Mina Mirzaian, Johannes M. Aerts, Mats Ehinger, Michael Rothe, Ute Modlich, Axel Schambach, Stefan Karlsson

Department of Molecular Medicine and Gene Therapy and Department of Pathology, Lund University, Lund, Sweden. Department of Clinical Chemistry, Institute of Biomedicine, the Sahlgrenska Academy at the University of Gothenburg, Gothenburg, Sweden. Department of Medical Biochemistry, University of Amsterdam, Amsterdam, The Netherlands. Institute of Experimental Hematology, Hannover Medical School, Hannover, Germany

Gaucher disease is caused by an inherited deficiency of the enzyme glucosylceramidase. Due to the lack of a fully functional enzyme there is progressive build-up of the lipid component glucosylceramide. Insufficient glucosylceramidase activity results in hepatosplenomegaly, cytopenias and bone disease in patients. Gene therapy represents a future therapeutic option for patients unresponsive to enzyme replacement therapy and lacking a suitable bone marrow donor. By proof-of-principle experiments we have previously demonstrated a reversal of symptoms in a murine disease model of type 1 Gaucher disease, using gammaretroviral vectors harboring strong viral promoters to drive glucosidase beta acid (GBA) gene expression. To investigate whether safer vectors can correct the enzyme deficiency, we utilized self-inactivating lentiviral vectors (SIN LVs) with the GBA gene under the control of human phosphoglycerate kinase (PGK) and myeloid promoters (CD68), respectively. Here we report prevention of, as well as reversal of, manifest disease symptoms after lentiviral gene transfer. Glucosylceramidase activity above levels required for clearance of glucosylceramide from tissues resulted in reversal of splenomegaly, reduced Gaucher cell infiltration and a restoration of hematological parameters. These findings support the use of SIN-LVs with cellular or tissue specific promoters in future clinical gene therapy protocols for type 1 Gaucher disease.

CARLA HOLLAK

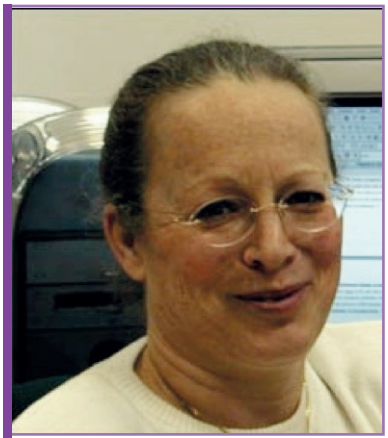
MD

Carla Hollak gained her MD in 1986 from the University of Amsterdam, The Netherlands. Currently, she heads the adult inherited metabolic unit at the Academic Medical Center in Amsterdam, including the centers of excellence for Gaucher and Fabry disease and a rapidly growing service for adults with inborn errors of metabolism. She holds a PhD in Medicine at the same University on studies in Gaucher disease. As co-founder of SPHINX, the Amsterdam Lysosome Center, she is involved in many studies on novel drug treatments for lysosomal storage disorders. Hollak acts repeatedly as expert for regulatory agencies and serves at several boards for rare diseases at a national and international level.

TREATMENT OPTIONS IN 2016: A CLINICAL VIEW

Enzyme replacement therapy (ERT) has revolutionized the care of type 1 Gaucher disease. In 2016, more than twenty-five years later, three recombinant ERT's from different cellular sources and two substrate reduction therapies are available for clinical use. Many more treatments are under development. When initiated timely, severe complications directly related to the build up of storage in spleen, liver and bone can be reversed or prevented. However, several unsolved issues remain. From a clinical point of view, there are still many relevant questions. What is the impact of these treatments on long-term outcomes? Of particular interest is the relationship with Parkinson's disease, metabolic syndrome and rare cancers. Does SRT have advantages over ERT to prevent these complications? A related issue is when we should start these treatments and which one? Currently we tend to decide on the basis of tolerability, convenience or costs. But is that the best way? Or could we perhaps also interrupt therapy for some time? The shortage of enzyme has shown that some patients may deteriorate, while others were stable for months. What are the characteristics of patients that make them susceptible to remain stable or relapse? And what about neuronopathic disease? So far, none of the treatments have been able to ameliorate the central nervous system disease. Last but not least, we should care about those patients that do not have access to these expensive therapies at all. What can we offer them?

The development of an algorithm for treatment of type 1 GD is not difficult if we use the published criteria for initiation of therapy, together with contraindications or obstacles for ERT or SRT. Such algorithms have already been proposed. However, for a truly evidence based algorithm, insufficient data are currently available. Further efforts are needed to investigate which characteristics of Gaucher disease patients predict the response to treatment or the occurrence of complications/ associated conditions in relation to different treatment approaches. This will require both fundamental research to understand the pathophysiological background of complications/associated conditions as well as systematic large cohort studies.



MIA HOROWITZ

Department of Cell Research and Immunology, Tel Aviv University, Israel

Mia Horowitz graduated from the Department of Genetics of The Weizmann Institute of Science, Rehovot Israel, in 1980. Following a post-doctoral training under the supervision of prof. Philip A. Sharp at MIT, Cambridge, MA, USA, she joined the Weizmann institute as an investigator and later joined the Faculty of Life Sciences at Tel Aviv University. In 1985 she initiated a project aiming at understanding the molecular biology underlying Gaucher disease in 1985. Since then she has cloned and sequenced the GBA gene encoding acid–β-glucocerebrosidase (GCase), defective in Gaucher disease (GD), as well as its pseudogene; has characterized their promoters and transcription pattern and identified new mutations in the GBA gene, associated with GD. Mia has shown that mutant GCase variants are recognized as misfolded in the ER and that if they fail to be correctly folded there, they are retrotranslocated from the ER to the cytoplasm, where they undergo polyubiquitination and proteasomal degradation in a process known as the ER Associated Degradation (ERAD). Mia has also shown that ER retention of mutant GCase variants in the ER leads to ER stress and activates the ER stress response, dubbed Unfolded Protein Response (UPR). Lately, using Drosophila as an animal model, she has shown that ER retention and the resulting UPR lead to development of Parkinson disease (PD), underscroring the prominence of mutant GCase in the pathological development of PD in GD patients and carriers of GD mutations.

DROSOPHILA MELANOGASTER AS AN ANIMAL MODEL TO STUDY GAUCHER DISEASE AND ITS ASSOCIATION WITH PARKINSON DISEASE

Mia Horowitz, Department of Cell Research and Immunology, Tel Aviv University, 69978, Israel

In Gaucher Disease (GD) there is accumulation of glucosylceramide that results from mutations in the GBA gene encoding lysosomal acid β-glucocerebrosidase (GCase). Most mutations associated with GD are missense mutations, which lead to synthesis of abnormal enzyme. These mutant variants are recognized as misfolded in the ER and therefore are retained there for refolding attempts. If not successful, the mutant molecules are retrotranslocated from the ER to the cytoplasm, undergo polyubiquitination and proteasomal degradation in a process known as the ER Associated Degradation (ERAD). Chronic ER retention of misfolded molecules induces ER stress and the ER stress response, dubbed unfolded protein response (UPR). We have shown that UPR exists in different cell types that originate from GD patients or carriers of GD mutations, underscoring the importance of mutant GCase in initiating UPR.

GD patients and carriers of GD mutations have a significantly higher propensity to develop Parkinson disease (PD) than the non-GD population, implying dominance which could result from the presence of mutant GCase. It cannot result from substrate accumulation, since it does not occur in carriers of GD mutations nor in brains of Type I GD patients, who develop PD.

To demonstrate that presence of mutant GCase leads to development of PD, we established two different fly models for carriers of GD mutations: flies carrying endogenous mutant GBA alleles and flies transgenic for the normal or mutant human GBA variants. The fruit fly Drosophila melanogaster has two GBA orthologs designated CG31148 and CG31414. There is a transposable element insertion in each of the genes, leading to translation of mutant proteins.

Aging double hetrerozygous flies (CG31148+/-/CG31414+/-) presented UPR and developed motor disabilities. However, flies carrying only one mutant allele (CG31148+/- or CG31414+/-) did not present UPR, nor loss of dopaminergic cells, and did not develop PD like signs. These results suggest that not only presence of mutant GCase but also its level are important factors in the development of PD.

Transgenic flies expressing the human N370S, the L444P or the 84GG mutant alleles also developed PD like disease. The parkinsonian phenotype in the N370S or the L444P transgenic flies, but not in the 84GG expressing flies, could be partially rescued by the addition of ambroxol, a known pharmacological chaperone. These results strongly imply that mutant GCase, which undergoes ER retention and initiates ER stress response (UPR), leads to development of PD.

Alpha-synuclein is an important factor in the development of PD. We generated a fly line which expressed human mutant (A53T) α-synuclein and mutant GCase. An association could be documented between mutant, but not normal, GCase and α-synuclein. These flies presented significantly faster death of dopaminergic cells with reduced life span.

We also generated flies homozygous for each of the two GBA orthologs. The CG31414-/- flies had minimal residual activity, accumulated substrate (in liver-like organ of the fly) and presented signs of neuronopathic GD with a short life span.

To summarize, our results highlight the validity of the fly as an animal model to study Gaucher disease and the association between Gaucher disease and Parkinson disease as well as its use as a valid model to test existing and future therapies, like pharmacological chaperones.

DR NEREA ALONSO

PhD

Dr Nerea Alonso graduated in Biochemistry from the University of Salamanca, Spain in 2003 and then she received her PhD in Cancer Biology and Clinical Studies with Prof Rogelio Gonzalez-Sarmiento at the University of Salamanca. Her PhD research focused on the molecular analysis of patients with Gorlin syndrome, a rare disease involving developmental alterations and basal cell carcinomas. In 2008 she moved to Edinburgh, UK, to do a first postdoc with Prof Stuart Ralston and Dr Omar Albagha, where she characterised the bone phenotype of a murine model for Paget's disease. During her second postdoc, with Prof Ralston, she applied genome-wide technologies to complex traits, such as osteoporosis. During the last five years, she has been part of an international consortium to study osteoporosis (GEFOS), and she is leading a collaborative effort to identify genetic variants predisposing to clinical vertebral fractures in postmenopausal women. Dr Alonso is also interested in pharmacogenomics applied to the treatment of osteoporosis and she has recently been awarded with a grant from the Scottish Chief Scientific Office to perform genetic profiling to predict the response to treatment in patients with severe osteoporosis.

Dr Alonso is a member of the European Calcified Tissue Society (ECTS), the European Human Genetics Society and the British Bone Research Society. She was awarded the ECTS New Investigator Award in 2013, as well as several other prizes at international conferences. Dr Alonso has published articles in top journals in the bone field, such as JBMR, and has co-authored studies published in Nature and Nature Genetics.

Since 2015, Dr Alonso is the vice-director of the Scottish constituency of the Spanish Researchers in the UK Society (SRUK/CERU).

FROM LINKAGE ANALYSIS TO NGS AND BEYOND: UNDERSTANDING THE GENETICS OF OSTEPOROSIS

Osteoporosis is a common metabolic disorder representing a major public health burden. It is characterised by low bone mineral density (BMD) and alterations in the microstructure of the bone, which lead to an increase risk of fracture. It is a clinically silent disease, not diagnosed until a fracture occurs. Fractures related to osteoporosis are associated with significant morbidity in causing pain, disability and impaired quality of life, and increased mortality. Over 9-million osteoporotic fractures occur worldwide, and this figure is predicted to increase by 50% by 2025. Heritability of BMD ranges between 50-80%; fractures show also a heritable component by mechanisms independent of BMD. Genetic factors most likely influence susceptibility to fractures not only involving bone density, but also bone quality, turnover and other non-skeletal phenotypes such as risk of falling.

Osteporosis could also arise as a secondary condition to other disorders, such as Gaucher's disease, where skeletal pathologies result in the most debilitating and potentially irreversible complications of the disease.

A first approach to the genetics of osteoporosis has been done through linkage analysis. Genes responsible for rare inherited diseases associated to changes in BMD have been identified, but their value for osteoporosis was limited. In contrast genome-wide association studies in large collaborative efforts successfully identified more than 60 loci with robust evidence of association with BMD and 14 loci predisposing to clinical fracture. Many of these regions contain genes involved in the RANK/RANKL/OPG and Wnt signalling pathways, whereas others are novel loci for bone metabolism. Currently, exome and whole-genome sequencing are used to identify rare variants of large effect size influencing osteoporosis.

Bisphosphonates are the first line treatment for osteoporosis. They prevent bone loss but are unable to increase bone mass; this requires an anabolic agent, such as teriparatide. However, response to treatment is very variable in each patient. New approaches such as pharmacogenomics are arising as useful tools to identify good and bad responders.

Despite the major advances in identifying genetic variants predisposing to osteoporosis, many more genetic markers remain to be discovered. Besides, little is known about using genetics to predict response to therapy. Larger studies and combination of genetic analysis and functional studies are crucial to understand the pathophysiology of the disease, as well as to set the basis for precision medicine

12th EWGGD ZARAGOZA

The European Working Group on Gaucher Disease

Oral Presentations

No	LIST OF ORAL PRESENTATIONS	Session
01	**CELLULAR MODELS FOR MONOCYTE/MACHROPHAGE STUDIES** *Centro Andaluz De Biología del Desarrollo-CSIC, Universidad Pablo de Olavide. Sevilla. Spain*	**2**
02	**HYPERFERRITINEMIA AND SERUM INFLAMMATORY CYTOKINES IN ADULT PATIENTS WITH GAUCHER DISEASE TYPE 1** *Lorenz F[1], Pawłowicz E[2], Bulanda Brustad A[3], Machaczka M[2,3*]* *[1] Department of Radiation Sciences, Section of Hematology, Umeå University, Umeå, Sweden* *[2] Hematology Center, Karolinska University Hospital Huddinge, Stockholm, Sweden* *[3] Medical Faculty, University of Rzeszow, Rzeszow, Poland*	**2**
03	**DEFICIENCY OF MONOCYTE/MACROPHAGE KCA3.1-FUNCTIONS IN TYPE-1/3 GAUCHER DISEASE** *Aida Oliván-Viguera[1], Pilar Alfonso[2], Pilar Giraldo[2], Ralf Köhler[1,31]* *[1.] Aragon Institute of Health Sciences (IACS) and University of Zaragoza, Spain* *[2.] Grupo de Enfermedades Hematológicas y Metabólicas, Instituto Investigación Sanitaria Aragón (IIS Aragón) and CIBERER, Zaragoza, Spain* *[3.] Aragon Agency for Research and Development (ARAID), Zaragoza, Spain*	**2**
04	**"FISHING" NEW MOLECULAR TARGETS TO GAIN FURTHER INSIGHTS INTO THE MOLECULAR PATHOGENESIS OF BONE ALTERATIONS IN TYPE I GAUCHER DISEASE (GD1)** *Roberto Costa[1], Stefania Bellesso[2], Marina Stroppiano[3], Mirella Filocamo[3], Enrico Moro[1*].* *[1] Department of Molecular Medicine, University of Padova, Italy.* *[2] Department of Women's and Child's Health, University of Padova, Italy.* *[3] Lab. Diagnosi Pre-Postnatale Malattie Metaboliche - IRCCS G. Gaslini, Genova, Italy.*	**2**
05	**THE FRUIT FLY DROSOPHILA MELANOGASTER AS A MODEL SYSTEM TO STUDY GAUCHER DISEASE** *Or Cabasso1 and Mia Horowitz[1]* *[1]Department of Cell Research and Immunology, Tel Aviv University, Ramat Aviv, Israel*	**3**
06a	**THE CONTRIBUTION OF MUTANT GCASE TO THE AGGREGATION AND ACCUMULATION OF A-SYNUCLEIN** *Gali Maor[1], Orly Sela[1], Daniel Segal[2], [3] and Mia Horowitz[1]* *[1]Dept of Cell Research and Immunology, [2]Department of Molecular Microbiology and Biotechnology and [3]Sagol Interdisciplinary School of Neurosciences, Tel Aviv University, Tel Aviv, Israel*	**3**
06b	**THE CONTRIBUTION OF MUTANT GBA TO THE DEVELOPMENT OF PARKINSON'S DISEASE IN DROSOPHILA** *Gali Maor[1], Or Cabasso[1], Olga Krivoruk[1], Joe Rodriguez[2], Hermann Steller[2], Daniel Segal[3, 4] and Mia Horowitz[1],* *[1]Dept of Cell Research and Immunology, [2]Strang Laboratory of Cancer Research, The Rockefeller University, 1230 York Avenue, New York, NY 10021, USA, [3]Department of Molecular Microbiology and Biotechnology and [4]Sagol Interdisciplinary School of Neurosciences, Tel Aviv University, Israel*	**3**
07	**PATIENT-DERIVED iPSC PROVIDE INSIGHTS INTO PATHOPHYSIOLOGY OF GAUCHER DISEASE** *Ola Awad, Leelamma M. Panicker, Judi A. Sgambato, Manasa Srikanth, and Ricardo A. Feldman.* *Department of Microbiology and Immunology, University of Maryland School of Medicine, Baltimore. USA*	**3**
08	**THE ROLE OF MTOR AND AUTOPHAGY IN LYSOSOMAL STORAGE DISEASE: PROSPECTIVE FOR THERAPEUTICS** *Suleiman Igdoura, PhD;* *Depts. of Biology and Pathology, McMaster University, Hamilton, ON Canada*	**3**
09	**GENERATION OF MESENCHYMAL AND OSTEOBLASTIC CELLULAR MODELS FOR GAUCHER DISEASE USING HUMAN INDUCED PLURIPOTENT STEM CELLS** *Jenny Serra-Vinardell1*, Laura de-Ugarte2, Ricardo A. Feldman3 Gustavo Tiscornia4, Natàlia García-Giralt2, Daniel Grinberg1, Lluïsa Vilageliu1* *1 Departament de Genètica, Universitat de Barcelona; IBUB; CIBER de Enfermedades Raras (CIBERER), Barcelona, Spain*	**3**
10	**PROOF-OF-PRINCIPLE:RAPID NONINVASIVE PRENATAL DIAGNOSIS FOR TYPE I GAUCHER DISEASE** *Altarescu_A *1,2, Zeevi_D1, Weinberg-Shukron_A,1,2 Zahdeh_F,3,4 Dinur_T,5 Chicco_G,5 Herskovitz_Y,1Renbaum_P,1 Elstein_D,5 Rolfs_A,6,7 Zimran_A 2,5* *1.Medical Genetics Institute, Shaare Zedek Medical Center, Jerusalem, Israel*	**4**
11 a	**CHILDREN WITH TYPE 1 GAUCHER DISEASE: CHANGING PROFILES IN THE 21ST CENTURY** *Altarescu_G[1], Abrahamov_A[1], Elstein_D[1], Zimran_A[1],* *[1] Gaucher Clinic, Shaare Zedek Medical Center, the Hebrew University and Hadassah School of Medicine, Jerusalem Israel*	**4**
11 b	**GROWTH PARAMETERS IN PEDIATRIC PATIENTS WITH GAUCHER DISEASE: 15-YEAR FOLLOW-UP FROM ONE ISRAELI CLINIC** *Espen Mendelson_MD1*, Floris Levy-Khademi_MD2, Amos Meir_MD3, Aya Abrahamov_MD3, Deborah Elstein_PhD3, Ari Zimran_MD3* *1Department of Pediatrics, 2Division of Pediatric Endocrinology and the 3Gaucher Clinic, Shaare Zedek Medical Center, affiliated with the Hebrew University Hadassah School of Medicine, Jerusalem Israel*	**4**
12	**BENIGN AND MALIGNANT HEMATOLOGICAL COMPLICATIONS OF GAUCHER DISEASE: DIAGNOSIS, MONITORING AND TREATMENT** *Hanna Rosenbaum* *Hematology day care unit, the center for consultant medicine, Clalit services, Nazareth, Israel.*	**4**
13	**MANAGEMENT GOALS FOR TYPE 1 GAUCHER DISEASE: A CONSENSUS DOCUMENT FROM THE EUROPEAN WORKING GROUP ON GAUCHER DISEASE** *M. Biegstraaten, T.M. Cox, N. Belmatoug, M.G. Berger, T. Collin-Histed, S. Vom Dahl, M. Di Rocco, C. Fraga, F. Giona, P. Giraldo, M.Hasanhodzic, D.A. Hughes, P.O. Iversen, A.I. Kiewiet, E. Lukina, M. Machaczka, T. Marinakis, E. Mengel, G.M. Pastores, U. Plöckinger, H. Rosenbaum, C. Serratrice, A. Symeonidis, J. Szer, J. Timmerman, A. Tylki-Szymańska, M. Weisz Hubshman, D.I. Zafeiriou, A. Zimran, C.E.M. Hollak*	**4**
14	**REVISED BELGIAN GUIDELINES FOR THE DIAGNOSIS, TREATMENT AND MONITORING OF GAUCHER'S DISEASE** *Eyskens François1*, Cassimans David2, Roland Dominique3* *For the Belgian Working Group on Gaucher's Disease.* *1Department of Pediatrics - Metabolic diseases, UZA, AZ Middelheim, Antwerp; 2Department of Hepatology & Metabolic center, UZ-Leuven, Leuven; 3Institut de Pathologie et de Génétique-Metabolic diseases, Gosselies (Charleroi), Belgium*	**4**

15	**LYSO-GB1 AS A GAUCHER-SPECIFIC BIOMARKER: DATA FROM CLINICAL TRIALS USING VELAGLUCERASE ALFA** *Qiu3, Yulin Qin3, Claudia Cozma4, Sabrina Eichler4, Arndt Rolfs4,5* *1 Shire Global Medical Affairs, Shire, Zug, Switzerland* *2 Gaucher Clinic, Shaare Zedek Medical Center, affiliated with the Hebrew University-Hadassah Medical School, Jerusalem, Israel* *3 Biostatistics and Statistical Programming Department, Shire, Lexington, MA, USA* *4 Centogene AG, Rostock, Germany* *5 Albrecht-Kossel-Institute for Neuroregeneration, University of Rostock, Rostock, Germany*	**4**
16	**IN VIVO CELLULAR PATHWAYS FOR INFUSED HUMAN RECOMBINANT GLUCOCEREBROSIDASE** *Margarita M Ivanova*1, Erk Changsila1, Chadima Iaonou1.2 and Ozlem Goker-Alpan1,2* *1Lysosomal and Rare Disorders Research and Treatment Center, Fairfax, VA, USA 2O&O Alpan LCC, Fairfax, VA, USA*	**5**
17	**INTRAVENOUSLY ADMINISTERED GENE THERAPY FOR THE TREATMENT OF NEURONOPATHIC GAUCHER DISEASE** *Massaro Giulia1*, Perocheau Dany2, Baker Rob3, Karlsson Stefan4, Cheng Seng H.5, Waddington Simon N.2, Rahim Ahad A.1* *1Department of Pharmacology, UCL School of Pharmacy, London, UK. 2. Gene Transfer Technology Group, UCL Institute for Women's Health, London, UK. 3.* *Molecular Diagnostic Lab, Haematology – Royal Free Site, HSL LLP, London, UK. 4. Division of Molecular Medicine and Gene Therapy, Lund University, Lund, Sweden. 5. Genzyme, A Sanofi Company, USA.*	**5**
18 a	**LONG-TERM SAFETY AND EFFICACY OF TALIGLUCERASE ALFA IN PEDIATRIC PATIENTS WITH GAUCHER DISEASE WHO WERE TREATMENT-NAÏVE OR PREVIOUSLY TREATED WITH IMIGLUCERASE** *Ari Zimran, MD1, Derlis Emilio Gonzalez-Rodriguez, MD2, Aya Abrahamov, MD1, Peter A. Cooper, PhD3, Sheeba Varughese, PhD3, Einat Brill-Almon, PhD4, Raul Chertkoff, MD4, 1Gaucher Clinic, Shaare Zedek Medical Center, Jerusalem, Israel, and the Hadassah Medical School - Hebrew University, Jerusalem, Israel; 2Instituto Privado de Hematologia e Investigacion Clinica, Asuncion, Paraguay; 3Department of Paediatrics, University of the Witwatersrand, Johannesburg, South Africa; 4Protalix BioTherapeutics, Carmiel, Israel*	5
18 b	**LONG-TERM EFFICACY AND SAFETY RESULTS OF TALIGLUCERASE ALFA THROUGH 5 YEARS IN ADULT TREATMENT-NAÏVE PATIENTS WITH GAUCHER DISEASE** *Ari Zimran, MD,1 Gloria Durán, MD,2 Pilar Giraldo, MD, PhD,3 Hanna Rosenbaum, MD,4 Fiorina Giona, MD,5 Milan Petakov, MD,6 Sergio Eduardo Solorio-Meza, MD,7 Peter A. Cooper, FC Paed (SA), PhD,8 Sari Alon,9 and Raul Chertkoff, MD9, 1Gaucher Clinic, Shaare Zedek Medical Center, Hebrew University and Hadassah Medical School, Jerusalem, Israel; 2Pontificia Universidad Catolica de Chile, Santiago, Chile; 3CIBERER, Hospital Universitario Miguel Servet, Zaragoza, Spain; 4Rambam Medical Center, Haifa, Israel; 5Sapienza University, Rome, Italy; 6Clinical Center of Serbia, Clinic of Endocrinology, Diabetes and Metabolic Disease, Belgrade University Medical School, Belgrade, Serbia; 7Hospital de Especialidades No1, Leon, México; 8Faculty of Health Sciences, University of the Witwatersrand & Charlotte Maxeke Johannesburg Academic Hospital, Johannesburg, South Africa; 9Protalix BioTherapeutics, Carmiel, Israel*	**5**
19	**CNS-ACCESSIBLE INHIBITOR OF GLUCOSYLCERAMIDE SYNTHASE FOR SUBSTRATE REDUCTION THERAPY OF NEURONOPATHIC GAUCHER DISEASE** *John Marshall1, Dinesh Bangari1, Jennifer B. Nietupski1, Mary A. Cromwell1, Gregory A. Grabowski2, John P. Leonard1, Seng H. Cheng1, 1Sanofi Genzyme, Framingham, MA, USA 2Cincinnati Children's Hospital Medical Center, Cincinnati, OH, USA*	**5**
20	**FOUR-YEAR FOLLOW-UP FROM THE ENCORE TRIAL: ORAL ELIGLUSTAT IN PATIENTS WITH GAUCHER DISEASE TYPE 1 STABILIZED ON ENZYME THERAPY** *Ozlem Goker-Alpan1*, Timothy M Cox2, Guillermo Drelichman3, Renata Cravo4,Manisha Balwani5, Thomas Andrew Burrow6, Ana Maria Martins7, Elena Lukina8, Barry Rosenbloom9, Nora Watman10, Amal el Beshlawy11, Priya Kishnani12, Maria Lucia Pedroso13, Sebastiaan JM Gaemers14, Regina Tayag15, M Judith Peterschmitt14 1Lysosomal Disorders Unit, O&O Alpan, LLC, Fairfax, VA, USA; 2University of Cambridge, Addenbrooke's Hospital, Cambridge, UK; 3Hospital de Niños Ricardo Gutiérrez, Buenos Aires, Argentina; 4State Institute of Haematology 'Arthur de Siqueira Cavalcanti', Rio de Janeiro, Brazil; 5Mount Sinai Hospital, New York, NY, USA; 6Cincinnati Children's Hospital Medical Center, Cincinnati, OH, USA, 7Universidade Federal de São Paulo, São Paulo, SP, Brazil; 8National Research Center for Hematology, Moscow, Russia; 9Cedars-Sinai/Tower Hematology , Beverly Hills, CA, USA; 10Hospital Ramos Mejia, Buenos Aires, Argentina; 11Pediatric Hematology, Cairo University, Egypt; 12Duke University School of Medicine, Department of Pediatrics, Durham, NC, USA; 13Hospital de Clinicas, Universidade Federal do Paraná, Curitiba, PR, Brazil; 14Sanofi Genzyme, Cambridge, MA, USA; 15Prometrika, Cambridge, MA, USA*	**5**
21	**TRANSFORMATION IN PRE-TREATMENT PRESENTATIONS OF GAUCHER DISEASE IN FIRST 2 DECADES OF IMIGLUCERASE ENZYME REPLACEMENT THERAPY: REPORT FROM THE INTERNATIONAL COLLABORATIVE GAUCHER GROUP GAUCHER REGISTRY** Pramod Mistry1*, Hans C. Andersson2, Manisha Balwani3, Julie L. Batista4, T. Andrew Burrow5, Joel Charrow6, Paige Kaplan7, Neal Khan8, Priya S. Kishnani9, Edwin H. Kolodny10, Barry Rosenbloom11, C. Ronald Scott12, Neal Weinreb13 1 Department of Internal Medicine, Yale University School of Medicine, New Haven, CT, USA 2 Tulane University Medical School, New Orleans, LA, USA 3 Icahn School of Medicine at Mount Sinai, New York City, NY, USA. 4 Sanofi Genzyme, Cambridge, MA, USA 5 Cincinnati Children's Hospital Medical Center, Cincinnati, OH, USA 6 Northwestern University Feinberg School of Medicine, Chicago, IL, USA 7 Children's Hospital of Philadelphia, Philadelphia, PA, USA 8 The University of Calgary, Calgary, Alberta, Canada 9 Duke University School of Medicine, Durham, NC, USA 10 New York University School of Medicine, New York, NY, USA 11 Cedars-Sinai/Tower Hematology Oncology, Beverly Hills, CA, USA 12 University of Washington, Seattle, WA, USA 13 University Research Foundation for Lysosomal Storage Diseases, Coral Springs, FL, USA	**5**
22	**DEVELOPMENT OF A NEW THERAPEUTIC STRATEGY FOR GAUCHER DISEASE (GD) USING EX VIVO TARGETED GENOME EDITING** *Fabiola Porro2, Erika Malini1, Bruno Bembi1, Andres Muro2, Andrea Dardis1** *1 Regional Coordinator Centre for Rare Diseases, University Hospital Udine, Italy* *2 International Centre for Genetic Engineering and Biotechnology (ICGEB), Trieste, Italy*	**5**
23	**pH-SENSITIVE GLUCOCEREBROSIDASE LIGANDS AS NON-INHIBITORY PHARMACOLOGICAL CHAPERONES FOR GAUCHER DISEASE** *José M. Garcia-Fernández1*, Carmen Ortiz-Mellet2, Teresa Mena-Barragán2, Katsumi Higaki3, Eiji Nanba3, Gustavo Tiscornia4, Aya Narita5, Kousaku Ohno5,6, Yoshiyuki Suzuki7* *1 Instituto de Investigaciones Químicas (IIQ), CSIC-Universidad de Sevilla, Sevilla, Spain* *2 Department of Organic Chemistry, Faculty of Chemistry, University of Sevilla 3 Division of Functional Genomics, Research Center for Bioscience and Technology, Tottori University, Tottori, Japan 4 Center for Biomedical Research, Depertamento de Ciencias, Biomedicas y Medicina/Programa de Medicina Regenerativa, Universidad do Algarve, Faro, Portugal 5 Division of Child Neurology, Faculty of Medicine, Tottori University, Tottori, Japan 6 Sanin Rosai Hospital, Tottori, Japan 7 Tokyo Metropolotan Institute of Medical Science*	**5**

24	**GLUCOSYLCERAMIDE SYNTHASE INHIBITION REDUCES a-SYNUCLEIN PATHOLOGY AND IMPROVES COGNITION IN MURINE MODELS OF SYNUCLEINOPATHY** *S. Pablo Sardi, Catherine Viel, Jennifer Clarke, James C. Dodge, John Marshall, Bing Wang, Lamya S. Shihabuddin, Seng H. Cheng, Sanofi Genzyme, Framingham, MA, USA*	**7**
25	**GBA1 HAPLOINSUFFICIENCY IN A PARKINSON MOUSE MODEL RESULTS IN AN EARLIER DISEASE ONSET AND MORE RAPID PROGRESSION OF SYMPTOMS** *Nahid Tayebi1, Loukia Parisiadou2, Ashley N Gonzalez1, Mieu Brooks3, Chrissy Makariou-Pikis2, Bahafta Berhe1, Niraj Trivedi4, Abdel Elkahloun5, Richard Grey1, Benoit I Giasson3 and Ellen Sidransky1* *1 MGB, 4CSGB and 5CGDGB, NHGRI, NIH, Bethesda MD,2 Department of Radiation Oncology, Feinberg School of Medicine, Chicago, IL and 3Department of Neuroscience, University of Florida College of Medicine, Gainesville, FL USA*	**7**
26	**PROCESSING OF ALPHA-SYNUCLEIN AND PARKIN IN PERIPHERAL BLOOD MONONUCLEAR CELLS IN PATIENTS WITH GAUCHER DISEASE** Renuka Pudi Limgala*, Ariel Badger, Chandni Sejpal and Ozlem Goker-Alpan Lysosomal and Rare Disorders Research and Treatment Center, Fairfax, VA, USA O & O Alpan, LLC, Fairfax, VA, USA	**7**
27	**STUDYING THE LINK BETWEEN GAUCHER AND PARKINSON'S DISEASE. CELL MODELS AND DISEASE-MODIFYING FACTORS** Derek Burke1,2*, Carmen de la Fuente-Barrigon2, Maria Garcia-Gomez2, and Simon Heales1,2. 1 Chemical Pathology, Great Ormond Street Hospital, London, UK. 2 Genetics and Genomic Medicine, UCL Institute of Child Health, University College London, UK	**7**
28	**GLUCOCEREBROSIDASE VARIANTS ARE MODIFIERS FOR ONSET AND CLINICAL SYMPTOMS IN PARKINSON DISEASE PATIENTS – A MULTINATIONAL STUDY OF GBA GENE SEQUENCES USING NEXT GENERATION SEQUENCING (NGS)** Rike Bender1*, Anne-Katrin Giese1/2, Eva-Juliane Vollstedt3/4, Ulrike Grittner5, Maximilian Weiss6, Ilona Csoti7, Roongroj Bhidayasiri8, Gregor Kuhlenbäumer9, Matthias Wittstock10, Tobias Böttcher10, Matthias Hahne11, Wolfgang Jost12, Birgit Herting13, Meike Kasten3/4, Christine Klein3, Arndt Rolfs1/6 1 Albrecht-Kossel-Institute for Neuroregeneration, University of Rostock, Rostock, Germany 2 Department of Neurology, Massachusetts General Hospital, Boston, USA 3Institute of Neurogenetics, University of Lübeck, Lübeck, Germany 4 Department of Psychiatry and Psychotherapy, University of Lübeck, Lübeck, Germany 5 Department for Biostatistics and Clinical Epidemiology, Charité-University Medical Centre, Berlin, Germany 6 Centogene AG, The Rare Disease Company, Rostock, Germany 7 Department of Neurology, Gertrudis Hospital, Leun-Biskirchen, Germany 8 Chulalongkorn Centre of Excellence for Parkinson's Disease and Related Disorders, Chulalongkorn University Hospital Bangkok 10330, Thailand 9 Department of Neurology, Christian-Albrechts-University of Kiel and University Medical Center, Kiel,Germany 10 Department of Neurology, University of Rostock, Rostock, Germany 11 Clinic of Neurology, University of Marburg, Bad Neustadt, Germany 12 Department of Neurology, Deutsche Klinik für Diagnostik Wiesbaden, Wiesbaden, Germany 13 Diakonie-Klinikum Schwäbisch Hall GmbH, Schwäbisch Hall, Germany	**7**
29	**UNBIASED APPROACH REVEALS HIGHER RISK FOR PD IN CARRIERS OF SEVERE VS. MILD GBA MUTATIONS** David Arkadir*1, Tama Dinur2, Strphen Mullin3, Atul Mehta4, Hagit Baris-Feldman5 Ari Zimran2. 1Department of Neurology, Hadassah Medical Center and the Hebrew University, Jerusalem Israel 2Gaucher Clinic, Shaare Zedek Medical Center nad Hebrew University, Jerusalem, Israel 3Department of clinical Neuroscience, UCL Institute on Neurology, London, UK 4LSD Unit Royal Free Hospital and UCL School of Medicine, London, UK. 5The Genetics Institute, Rambam Health Care Campus, Haifa, Israel, and Rappaport School of Medicine, Technion, Haifa, Israel	**7**
30	**MEASUREMENT OF BONE MARROW FAT FRACTION WITH GRADIENT-ECHO MR IMAGING IN GAUCHER DISEASE PATIENTS; A PRACTICAL ALTERNATIVE TO DIXON QUANTITATIVE CHEMICAL SHIFT IMAGING** M. Regenboog1-2*, K.M. Beekman1, C.E.M. Hollak2, M. Maas1 E.M. Akkerman1, 1 Department of Radiology, 2 Department of Internal Medicine, division of Endocrinology & Metabolism, Academic Medical Center Amsterdam, The Netherlands.	**8**
31	**OSTEOCYTE ALTERATIONS CONTRIBUTE TO BONE PATHOLOGY IN GAUCHER DISEASE** Mucci Juan Marcos1*, Ferreyra Malena1, Crivaro Andrea1, Bondar Constanza1, Ormazabal Maximiliano1, Delpino Victoria2, Rozenfeld Paula1 1 Instituto de Estudios Inmunológicos y Fisiopatológicos, CONICET-UNLP, La Plata, Argentina 2 Instituto de Inmunología, Genética y Metabolismo, CONICET, CABA, Argentina	**8**
32	**EFFECT OF TWO DIFFERENT THERAPEUTIC INTERVENTIONS: SRT IN COMPARISON TO ERT ON IMMUNE ASPECTS AND BONE INVOLVEMENT IN GAUCHER DISEASE** Renuka Pudi Limgala*, Miriam Arnone & Ozlem Goker-Alpan Lysosomal and Rare Disorders Research and Treatment Center, Fairfax, VA 22030 USA	**8**
33	RISK FACTORS FOR FRACTURES IN IMIGLUCERASE-TREATED GAUCHER DISEASE TYPE 1 PATIENTS IN THE ICGG GAUCHER REGISTRY Patrick Deegan1, Julie L. Batista2, Stephan Vom Dahl3* 1Lysosomal Disorders Unit, Department of Medicine, Addenbrooke's Hospital, Cambridge, UK 2Sanofi Genzyme, Cambridge, MA, USA 3Department of Gastroenterology, Hepatology and Infectious Diseases, University of Duesseldorf	**8**

Session 2

(1)

CELLULAR MODELS FOR MONOCYTE/MACHROPHAGE STUDIES

Mario de la Mata, Marina Villanueva Paz, Mónica Álvarez Córdoba, David Cotán, Manuel Oropesa-Ávila, Ana Delgado Pavón, Isabel de Lavera, José A. Sánchez-Alcázar.

Centro Andaluz De Biología del Desarrollo-CSIC, Universidad Pablo de Olavide

41013 Sevilla, Spain.

*Corresponding author : José A Sánchez Alcázar. jasanalc@upo.es

Gaucher disease (GD), the lysosomal storage disease with the highest prevalence, is caused by mutations in the GBA1 gene that results in defective and insufficient activity of the enzyme β-glucocerebrosidase (GCase). Decreased catalytic activity and/or instability of GCase leads to accumulation of glucosylceramide (GlcCer) in the lysosomes of macrophage cells. The accumulation of substances affects the function of lysosomes and other organelles such as mitochondria. In order to mimic the pathological phenotype of the disease, an in vitro cellular model of Gaucher disease was developed by treating the THP-1, a human monocytic cell line differentiated into macrophage, with a specific inhibitor of GCase, conduritol beta epoxide, and/or supplementation with exogenous GlcCer. In the cellular chemical model, GlcCer accumulation led to impaired autophagy and mitochondrial dysfunctionin TPH-1 macrophages.

In turn, dysfunctional mitochondria induced excessive species reactive oxygen that promoted autophagy and inflammasome activation. In addition, the elimination of dysfunctional mitochondria played a crucial role in protecting cells from the damage caused by perturbed mitochondrial function. Treatment of Gaucher TPH-1 macrophages with coenzyme Q_{10} (CoQ), an electron and proton carrier with antioxidant properties, improved mitochondrial/lysosomal function and increased autophagic flux and the degradation of damaged mitochondria. Altogether, our results point out that cell alterations in cell models of GD are attenuated by supplementation with CoQ.

Session 2

(2)

HYPERFERRITINEMIA AND SERUM INFLAMMATORY CYTOKINES IN ADULT PATIENTS WITH GAUCHER DISEASE TYPE 1

Lorenz F[1], Pawłowicz E[2], Bulanda Brustad A[3], Machaczka M[2,3]*

[1] Department of Radiation Sciences, Section of Hematology, Umeå University, Umeå, Sweden

[2] Hematology Center, Karolinska University Hospital Huddinge, Stockholm, Sweden

[3] Medical Faculty, University of Rzeszow, Rzeszow, Poland

*Corresponding author: maciej.machaczka@ki.se

The storage of glucosylceramide in macrophages in Gaucher disease produces an inflammatory response with iron recycling dysregulation and a release of cytokines. This study was undertaken to evaluate ferritinemia, iron metabolism profiles and inflammatory cytokine concentrations in Swedish patients with Gaucher disease type 1 (GD1). Patients and methods: The study included 16 adults with GD1 aged 20–86 years. All but one patient (94%) carried at least one allele with c.1226A>G (N370S) mutation in the GBA1 gene. Zimran's severity score index (SSI) was calculated for all patients at the time of their inclusion in the study. Iron profile (s-ferritin, p-Fe, p-TSAT); HFE gene mutations; s-hepcidin; LFTs; CRP; serum IL-1β, IL-6, IL-8, IL-10, TNF-α, and sIL-2Rα were collected from fresh blood for analysis. Assessment of these variables was performed at baseline and at follow-up (every 6–12 months). Results: Hyperferritinemia >500 μg/L was present in 13/16 pts (81%). Transferrin, Fe, and TSAT were within normal limits for 15/16 pts. There was no correlation between hyperferritinemia and sex, splenectomy, and SSI. HFE gene mutations were analyzed in 11 pts: 4 pts were heterozygous for His63Asp, one pt was heterozygous for both His63Asp and Cys282Tyr, and 6 pts had no mutation in the HFE gene. No obvious correlation between ferritinemia and HFE genotype was detected. Hepcidin levels were normal in 7/10 pts; one pt with a normal ferritin had a low hepcidin concentration at 4 μg/L and 2 pts with a hyperferritinemia had elevated hepcidin. The serum levels of IL-1β, IL-8, IL-10 were normal in all pts. The concentrations of TNF-α, IL-6, sIL-2Rα, and CRP were within normal limits in all pts. In 5/11 pts TNF-α was moderately increased. Two patients with the highest TNF-α levels (28.4, and 30.4 ng/L) showed mildly elevated IL-6 (5.9 and 10.7 ng/L). IL-6 was within normal limits in the remaining 14 pts (87%). sIL-2Rα was within normal range in 62% of pts (10/16). Conclusions: Hyperferritinemia is common in Swedish GD1 patients. Unlike HLH, hyperferritinemia in GD1 is not associated with high serum levels of sIL-2Rα. In some, but not all GD1 patients TNF-α and IL-6 levels could be mildly elevated.

Session 2

(3)

DEFICIENCY OF MONOCYTE/MACROPHAGE KCA3.1-FUNCTIONS IN TYPE-1/3 GAUCHER DISEASE

Aida Oliván-Viguera[1], Pilar Alfonso[2], Pilar Giraldo[2], Ralf Köhler[1,31]

[1.] Aragon Institute of Health Sciences (IACS) and University of Zaragoza, Spain

[2.] Grupo de Enfermedades Hematológicas y Metabólicas, Instituto Investigación Sanitaria Aragón (IIS Aragón) and CIBERER, Zaragoza, Spain

[3.] Aragon Agency for Research and Development (ARAID), Zaragoza, Spain

*Corresponding author: rkoehler43@gmx.de

Background: The calcium-calmodulin-gated KCa3.1 channel in macrophages regulates secretion of pro-inflammatory cytokines and macrophage differentiation in tissues. In Gaucher Disease (GD) lysosomal accumulation of glucosylceramide, in macrophages (Gaucher cells), results in progressive splenomegaly and bone lesions. Here we hypothesized that macrophage KCa3.1 functions are impaired in a GD-related fashion.

Methods: We measured KCa3.1-currents by whole-cell patch-clamp in monocytes and macrophages activated by 4-day exposure to erythrocyte lysates from GD-1/3 patients and healthy volunteers (Ctrl).

Results: Comparison of maximal KCa3.1-current amounts revealed similar K^+ current densities in monocytes in GD-1 and Ctrl, while currents densities in GD-3 monocytes were very low ($P<0.05$). In differentiated macrophages, maximal current densities were three times larger than in monocyte precursors. However, GD-1 macrophages showed significantly smaller increase of current densities (% 40 of Ctrls) while in GD-3 macrophages KCa3.1 currents were still hardly detectable ($P<0.01$). Concerning the cellular mechanisms, we found that glucosylceramide inhibited KCa3.1-membrane function with an IC50 of ≈5μM.

Conclusions: We demonstrate for the first time impaired macrophage/monocyte KCa3.1 functions in GD, which is more pronounced in GD-3. Accumulation of glucosylceramide may cause channel inhibition or defective lysosomal KCa3.1 recycling. Considering KCa3.1 important in macrophage differentiation and cytokine production, impaired KCa3.1-activities could be of pathomechanistic relevance in Gaucher cell malfunction by promoting a pro-inflammatory environment. Pharmacological targeting KCa3.1 may represent a novel strategy to achieve neuroprotection in GD-3 and to reduce bone lesions in GD-1.

This work has been partially granted by FEETEG and FIS PS15/00616

Session 2

(4)

"FISHING" NEW MOLECULAR TARGETS TO GAIN FURTHER INSIGHTS INTO THE MOLECULAR PATHOGENESIS OF BONE ALTERATIONS IN TYPE I GAUCHER DISEASE (GD1)

Roberto Costa[1], Stefania Bellesso[2], Marina Stroppiano[3], Mirella Filocamo[3], Enrico Moro[1*].

[1] Department of Molecular Medicine, University of Padova, Italy. (moroe@bio.unipd.it)

[2] Department of Women's and Child's Health, University of Padova, Italy.

[3] Lab. Diagnosi Pre-Postnatale Malattie Metaboliche - IRCCS G. Gaslini, Genova, Italy.

Corresponding Author: moroe@bio.unipd.it

In the past few years much effort has been spent to address the question as to whether osteoblasts or osteoclasts defects may account for the type I Gaucher disease (GD1)-related bone pathogenesis. While a proinflammatory state and increased osteoclast activity in GD1 patients has been deeply described, limited research has been performed to understand the impairment in the osteoblast lineage. Using a novel approach, based upon the use of different transgenic fish lines and a glucocerebrosidase mutant fish, we have recently demonstrated that defective canonical Wnt signaling is involved in a disrupted differentiation program of Gba1-depleted osteoblast precursors. The Wnt signaling impairment was further demonstrated in fibroblasts from GD1 affected patients, suggesting a direct link between glucocerebrosidase activity and Wnt pathway transduction. We have started to explore individual Wnt pathway related molecules using alternative assays and live imaging, in order to identify target cell signaling molecules, which are directly modulated by glucocerebrosidase function.

Our preliminary results strengthen the potential role of the canonical Wnt pathway in the onset of type I Gaucher disease-related bone defects and emphasize the potential application of Wnt pathway modulators as adjuvant treatment of GD-related osteopenia.

*This work has been fully supported by Genzyme Generation Program 2013 to E.Moro

Session 3

(5)

THE FRUIT FLY DROSOPHILA MELANOGASTER AS A MODEL SYSTEM TO STUDY GAUCHER DISEASE

Or Cabasso[1] and Mia Horowitz[1]

[1]Department of Cell Research and Immunology, Tel Aviv University, Ramat Aviv, Israel

*Corresponding author horwitzm@post.tau.ac.il

Gaucher disease (GD) results from mutations in the GBA1 gene, which lead to decreased activity of the lysosomal enzyme acid-β-glucocerebrosidase (GCase) and as a result to accumulation of glucosylceramide (GlcCer). Following its synthesis on ER bound polyribosomes, GCase undergoes four N-linked glycosylations and folding. Correctly folded enzyme shuttles to the lysosomes while mutant misfolded enzyme is retained in the ER, which leads to ER stress and activation of the ER stress response (Unfolded protein response-UPR).

In order to study the consequences of UPR in an animal model, we decided to establish the fly Drosophila melanogaster as a model for GD.

In Drosophila, there are two GBA1 orthologs, GBA1a (CG31148) and GBA1b (CG31414). They were both manipulated to have a transposable element insertion (Minos element insertion), which leads to synthesis of shorter, mutant GCase proteins. The GBA1a mutant protein produces a truncated GCase, missing 34 C-terminal amino acids, while the GBA1b mutant protein has a 129 amino acid, C-terminal deletion, which includes one amino acid of the active site. As a result, The GBA1a-/- flies are mildly affected while the GBA1b-/- flies present a severe neuronopathic disease.

We could show decreased GCase activity, substrate accumulation, neurodegeneration and earlier death in the fly models. These results highlight the resemblance between the fly models and GD forms and underlie their importance in studying the disease as well as possible therapies.

Session 3

(6a)

THE CONTRIBUTION OF MUTANT GCASE TO THE AGGREGATION AND ACCUMULATION OF A-SYNUCLEIN

Gali Maor[1], Orly Sela[1], Daniel Segal[2, 3] and Mia Horowitz[1],

[1]Dept of Cell Research and Immunology, [2]Department of Molecular Microbiology and Biotechnology and [3]Sagol Interdisciplinary School of Neurosciences, Tel Aviv University, Tel Aviv, 69978, Israel.

A widely studied association has been found between GD and Parkinson disease (PD). GD patients and carriers of GD mutations have a significantly higher propensity to develop PD in comparison to the non-GD population.

A key factor contributing to PD is α-synuclein. One of the major characteristics of PD is the presence of insoluble oligomeric and fibrillar α-synuclein-positive inclusions known as Lewy bodies and Lewy neurites in neurons in the substantia nigra pars compacta. In brain autopsies from PD patients or carriers of GBA mutations, accumulation of α-synuclein aggregates was noted in the substantia nigra. Furthermore, in α-synuclein-KO mice, transgenic for the human A53T mutant α-synuclein and heterozygous for the L444P mutant GBA allele there was a significant stabilization of α-synuclein, compared to mice expressing the human A53T mutant α-synuclein only, highlighting the effect mutant GCase on α-synuclein [1]. We could recapitulate the significant stabilization of α-synuclein in the presence of mutant GCase in dopaminergic cells expressing WT, N370S or L444P human GCase variants. We also documented association between mutant, but not WT GCase and α-synuclein.

We used Drosophila melanogaster as an animal model to further study the association between mutant GCase and α-synuclein. In transgenic flies, expressing the human A53T α-synuclein and human mutant GCase in dopaminergic cells there was an earlier death of the cells and shorter life span in comparison to flies expressing only the mutant α-synuclein or in the presence of WT GCase.

Our results strongly indicate that association between mutant GCase and α-synuclein leads to their stabilization and to a phenotype reminiscent of Parkinson disease.

1. Fishbein, I., et al., *Augmentation of phenotype in a transgenic Parkinson mouse heterozygous for a Gaucher mutation. Brain*, 2014. 137(Pt 12): p. 3235-47.

Session 3

(6b)

THE CONTRIBUTION OF MUTANT GBA TO THE DEVELOPMENT OF PARKINSON'S DISEASE IN DROSOPHILA

Gali Maor[1], Or Cabasso[1], Olga Krivoruk[1], Joe Rodriguez[2], Hermann Steller[2], Daniel Segal[3, 4] and Mia Horowitz[1],

[1]Dept of Cell Research and Immunology, 2Strang Laboratory of Cancer Research, The Rockefeller University, 1230 York Avenue, New York, NY 10021, USA, [3]Department of Molecular Microbiology and Biotechnology and [4]Sagol Interdisciplinary School of Neurosciences, Tel Aviv University, Tel Aviv, 69978, Israel.

Gaucher disease (GD) results from mutations in the acid β-glucocerebrosidase (GCase) encoding gene, GBA, which leads to accumulation of glucosylceramides. GD patients and carriers of GD mutations have a significantly higher propensity to develop Parkinson's disease (PD) in comparison to the non-GD population.

In the present study we used the fruit fly Drosophila melanogaster to show that development of PD in carriers of GD mutations results from the presence of mutant GBA alleles. *Drosophila* has two GBA orthologs (CG31148 and CG31414), each of which has a minos insertion, which creates C-terminal deletion in the encoded GCase. Flies double heterozygous for the endogenous mutant *GBA* orthologs presented Unfolded Protein Response (UPR) and developed parkinsonian signs, manifested by death of dopaminergic cells, defective locomotion and a shorter life span. We also established transgenic flies carrying the mutant human N370S, L444P and the 84GG variants. UPR activation and development of parkinsonian signs could be recapitulated in flies expressing these three mutant variants.

UPR and parkinsonian signs could be partially rescued by growing the double heterozygous flies, or flies expressing the N370S or the L444P human mutant GCase variants, in the presence of the pharmacological chaperone ambroxol, which binds and removes mutant GCase from the ER. However flies expressing the 84GG mutant, that does not express mature GCase, did not exhibit rescue by ambroxol.

Our results strongly suggest that the presence of a mutant *GBA* allele in dopaminergic cells leads to ER stress and to their death, and contributes to development of Parkinson's disease.

Session 3

(7)

PATIENT-DERIVED iPSC PROVIDE INSIGHTS INTO PATHOPHYSIOLOGY OF GAUCHER DISEASE

Ola Awad, Leelamma M. Panicker, Judi A. Sgambato, Manasa Srikanth, and Ricardo A. Feldman.

Department of Microbiology and Immunology, University of Maryland School of Medicine, Baltimore MD, 21201, USA. Corresponding author

E-Mail: rfeldman@som.umaryland.edu

To model Gaucher disease (GD) we reprogrammed fibroblasts from patients with types 1, 2 and 3 GD, into induced pluripotent stem cells (iPSC). Directed differentiation of GD iPSC revealed phenotypic abnormalities in hematopoietic, osteogenic and neuronal cells, which recapitulated characteristic hallmarks of the disease. GD iPSC macrophages had a striking defect in their ability to clear phagocytosed RBC and produced elevated levels of inflammatory cytokines. All of the phenotypic abnormalities we observed were reversed by incubation with recombinant glucocerebrosidase. Treatment with chaperones, in particular isofagomine, was also able to reverse the abnormal phenotype of GD cells. Differentiation of GD iPSC-derived hematopoietic progenitors showed that myeloid, erythroid and megakaryocytic differentiation was compromised. Similarly, GD osteoblasts had defective differentiation, exhibited abnormal lysosomal exocytosis, and a decreased ability to carry out protein and mineral bone matrix deposition, suggesting that bone manifestations in GD patients may be caused in part by dysfunctional osteoblasts. This system also provided insights into the mechanisms by which mutant glucocerebrosidase may cause neurodegeneration. GD iPSC-derived neurons with severe GBA1 mutations had widespread depletion of lysosomes and autophagy defects, in part due to downregulation of TFEB, the master regulator of lysosomal biogenesis and autophagy. Overexpression of TFEB partially reversed lysosomal depletion in GD neurons. In sum, the use of GD iPSC enabled us to uncover developmental and lysosomal defects in a number of cell types affected by the disease, and to begin elucidating the mechanisms involved. Patient-derived iPSC also provide a new and relevant platform for therapeutic development.

Session 3

(8)

THE ROLE OF MTOR AND AUTOPHAGY IN LYSOSOMAL STORAGE DISEASE: PROSPECTIVE FOR THERAPEUTICS

Suleiman Igdoura, PhD;

Depts. of Biology and Pathology, McMaster University, Hamilton, ON Canada L8S 4K1

EMail: igdoura@mcmaster.ca

Defective lysosomal enzymes can result in disease marked by suppression of lysosomal degradation capacity. Accumulation of undegraded lysosomal substrates as well as cytosolic substrates due to impairment of autophagy describe the state of lysosomal storage diseases (LSDs). Recently, the transcription factor TFEB was found to sense and regulate lysosomal homeostasis in a mammalian target of rapamycin (mTOR)-dependent manner. mTOR is part of a protein complex (mTORC1) that is active when localized to the lysosomal surface; therefore lysosomes can regulate their own homeostasis. In this study, we utilized fibroblasts from Sialidosis and Tay Sachs patients to assess the cross-talk between mTORC1 and mTORC2 activities during the modulation of autophagy in LSDs. Our results indicate accumulations of autophagy markers LC3 and p62 in LSD fibroblasts that are partially or completely rescued by mTOR with rapamycin or torin-1. We also found increased phosphorylation of the mTORC1-target 4E-BP1, and cytoplasmic sequestration of TFEB in the presence of rapamycin but not torin-1. We have also utilized specific siRNAs targeting human raptor and rictor to knockdown mTORC1 and mTORC2 respectively and found that mTORC2 substrate specificity is broader in LSD fibroblasts to include mTORC1 targets. These results points to mTORC2 as a more effective modulator of TFEB phosphorylation and phagolysosome formation in LSDs. These findings place further importance on the role of mTORC2 in autophagy and lysosomal biogenesis and point to mTOR as a potential therapeutic target for the treatment of LSDs.

Session 3

(9)

GENERATION OF MESENCHYMAL AND OSTEOBLASTIC CELLULAR MODELS FOR GAUCHER DISEASE USING HUMAN INDUCED PLURIPOTENT STEM CELLS

Jenny Serra-Vinardell[1*], Laura de-Ugarte[2], Ricardo A. Feldman[3] Gustavo Tiscornia[4], Natàlia García-Giralt[2], Daniel Grinberg[1], Lluïsa Vilageliu[1]

[1] Departament de Genètica, Universitat de Barcelona; IBUB; CIBER de Enfermedades Raras (CIBERER), Barcelona, Spain. [2] Musculoskeletal research group, IMIM (Hospital del Mar Medical Research Institute), Red Temática de Investigación Cooperativa en Envejecimiento y Fragilidad (RETICEF), ISCIII, Barcelona, Spain. [3] Department of Microbiology and Immunology, University of Maryland School of Medicine, Baltimore, Maryland 21201, USA. [4] Department of Biomedical Sciences and Medicine, University of Algarve, 8005-139 Faro, Portugal. *Corresponding author: jny.serra@gmail.com

Skeletal alterations in Gaucher disease (GD) include Erlenmeyer flask deformity, osteoporotic fractures, osteoesclerosis and osteonecrosis. Approximately 90% of GD type 1 patients exhibit bone involvement and is the major cause of morbidity. Most of these manifestations can be explained by the disruption of the balance between bone formation (by osteoblasts) and bone resorption (by osteoclasts). However, the pathological mechanisms causing this disruption are still poorly understood.

The role of Mesenchymal Stem Cells (MSCs) and osteoblasts in bone formation is well documented. We hypothesize that bone manifestations in GD are a result of either (1) down-regulation of the bone formation process, or (2) an increased osteoclastogenesis due to the effect of MSCs and/or osteoblasts on monocyte differentiation into osteoclasts.

In the present study, we generated cellular models of MSCs and osteoblasts using GD and wild type (WT) human induced pluripotent stem cells (hiPSC). We are performing an extensive characterization in terms of gene expression and osteoblast function to compare the differentiation process of MSCs into osteoblasts between GD and WT models. In order to evaluate the potential of MSC and osteoblast in stimulating osteoclastogenesis, we have set up a co-culture experiment for our models. Preliminary results demonstrated the ability of MSCs derived from both GD and WT hiPSC to differentiate into osteoblasts and produce mineralized extracellular matrix.

We will use these cellular models to ascertain the role of bone cells in bone pathology in GD and to assay new drugs such as pharmacological chaperones.

Session 4

(10)

PROOF-OF-PRINCIPLE: RAPID NONINVASIVE PRENATAL DIAGNOSIS FOR TYPE I GAUCHER DISEASE

Altarescu_A *[1,2], Zeevi_D[1], Weinberg-Shukron_A,[1,2] Zahdeh_F,[3,4] Dinur_T,[5] Chicco_G,[5] Herskovitz_Y,[1]Renbaum_P,[1] Elstein_D,[5] Rolfs_A,[6,7] Zimran_A [2,5]

1.Medical Genetics Institute, Shaare Zedek Medical Center, Jerusalem, Israel. 2.The Hebrew University of Jerusalem, Faculty of Medicine, Jerusalem, Israel. 3. Department of Genetics, The Alexander Silberman Institute of Life Sciences, Faculty of Science, The Hebrew University of Jerusalem, Edmond J. Safra Campus, Givat Ram, Jerusalem, Israel. 4. Hereditary Research Lab, Life Sciences Department, Bethlehem University, Bethlehem, Palestinian Authority. 5. Gaucher Clinic, Shaare Zedek Medical Center, Jerusalem, Israel. 6. Centogene AG, Rostock, Germany.

7. Albrecht Kossel Institute for Neuroregeneration, University of Rostock, Medical Faculty, Rostock, Germany.

*Corresponding author: Gheona Altarescu, MD

Altarescu_A and Zeevi _D equally contributed to this paper

BACKGROUND. Noninvasive prenatal testing can be used to accurately detect chromosomal aneuploidies in circulating fetal DNA. For noninvasive prenatal diagnosis (NIPD) of monogenic disease, the necessity of parental haplotype construction is a primary drawback. Family-specific haplotype assembly is essential for accurate diagnosis of minuscule amounts of circulating cell-free fetal DNA. Current haplotyping techniques are time-consuming , laborious to be carried out within the limited time constraints of prenatal testing, hampering practical application of NIPD in the clinic. We have devised a universal strategy for rapid NIPD for Gaucher Disease type I, common in the Ashkenazi Jewish (AJ) population. METHODS. Pregnant AJ couples, carrying mutation(s) in GBA gene (causing Gaucher disease), were recruited. Targeted next-generation sequencing of GBA-flanking SNPs was performed on peripheral blood samples from each couple, relevant mutation carrier family members, and unrelated individuals who are homozygotes for an AJ founder mutation. Allele-specific haplotypes were constructed based on linkage, and a consensus Gaucher disease–associated founder mutation–flanking haplotype was fine mapped. Together, these haplotypes were used for NIPD. All test results were validated by conventional prenatal or postnatal diagnostic methods. RESULTS. All eight unrelated fetuses (ten parental alleles) (100%) were correctly diagnosed based on the noninvasive method developed, some as early as 14 weeks of pregnancy. The consensus mutation–flanking haplotype aided diagnosis for 6 of 9 founder mutation alleles. CONCLUSIONS. The founder NIPD method developed and described is rapid, economical, and readily adaptable for prenatal testing of prevalent autosomal recessive disease-causing mutations in an assortment of worldwide populations.

Session 4

(11a)

CHILDREN WITH TYPE 1 GAUCHER DISEASE: CHANGING PROFILES IN THE 21ST CENTURY

Altarescu_G[1], Abrahamov_A[1], Elstein_D[1], Zimran_A[1],

[1] Gaucher Clinic, Shaare Zedek Medical Center, the Hebrew University and Hadassah School of Medicine, Jerusalem Israel

Corresponding author: Ari Zimran. azimran@gmail.com

Gaucher disease (GD) has a broad clinical spectrum but only partial correlation with genotypes. Increased awareness for GD especially among at-risk populations like Ashkenazi Jews (AJ) and availability of non-invasive diagnosis induced a trend to prenatal screening that led to increasing numbers of diagnosed patients. We assessed pediatric (<16 years) Israeli AJ patients with GD to ascertain demographics and clinical features at presentation and over time because many are products of large-scale screening.

55/67 of our pediatric patients born since 01/01/2000 are AJ with non-neuronopathic GD: 28 are N370S/N370S; 24 are N370S/other; 3 are other/other. 30 were diagnosed by screening; 10 others have a sibling previously diagnosed by screening. Of 19 receiving enzyme replacement therapy (ERT), four were by screening, (N370S/N370S; N370S/L444P, N370S/84GG, N370S/IVS2+1); others were diagnosed because of GD symptoms and/or a sibling with symptomatic GD. Four began ERT at <2 years of age; nine at 3-5 years; six at 6-12 years. 49% presented with height/weight growth percentiles ≤25%, but group means were rather comparable up to 12 years follow-up including in 10 receiving ERT (8 for >5 years). 22% presented with anemia and 20% with thrombocytopenia; at last follow-up 4% and 6%, respectively, remained cytopenic.

We present a new demographic profile for pediatric GD because so many are products of screening with few GD signs/symptoms at presentation and at follow-up. Nonetheless, early diagnosis is good, especially for non-N370S/mild genotypes. However, the potential advantage of screening would be genotype-specific prognostication and appropriate genetic counseling.

Session 4

(11b)

GROWTH PARAMETERS IN PEDIATRIC PATIENTS WITH GAUCHER DISEASE: 15-YEAR FOLLOW-UP FROM ONE ISRAELI CLINIC

Espen Mendelson_MD[1]*, Floris Levy-Khademi_MD[2], Amos Meir_MD[3], Aya Abrahamov_MD[3], Deborah Elstein_PhD[3], Ari Zimran_MD[3],

*In partial fulfillment of requirements for an MD degree at the Hebrew University Hadassah School of Medicine

[1]Department of Pediatrics, [2]Division of Pediatric Endocrinology and the [3]Gaucher Clinic, Shaare Zedek Medical Center, affiliated with the Hebrew University Hadassah School of Medicine, Jerusalem Israel

Corresponding author: Ari Zimran. azimran@gmail.com

Enzyme Replacement Therapy (ERT) putatively accelerates height growth in Gaucher disease so patients achieve mid-parental height. This study presents growth parameters in adult patients followed for 15 years. 41 non-neuronopathic patients at least 21 years old were assessed. Parental height, mid-parental sex-adjusted height, and siblings' heights and age of sexual maturation were compared to that of the patient. Student's t-test was employed.

Mean final height was below expected (relative to population norms) but this did not mean short stature because patients achieved mid-parental height. Mean final height standard deviation score (SDS) was significantly lower in males [$p=0.036$] but not in females. There was no effect of ERT, gender, or genotype. Expected age of menarche was not achieved [$p<0.0001$]. Mean age of menarche among patients was significantly later than in their [obligate carrier] mothers [$p=0.0005$]; mean age of start shaving in boys was as in healthy boys.

This study confirms mean height significantly lower in Gaucher disease than healthy populations, but patients were not of short stature. ERT-treatment did (but not significantly) impact mean final height SDS. That height SDS was significantly decreased in boys but not girls is new.

Session 4

(12)

BENIGN AND MALIGNANT HEMATOLOGICAL COMPLICATIONS OF GAUCHER DISEASE: DIAGNOSIS, MONITORING AND TREATMENT

Hanna Rosenbaum

Hematology day care unit, the center for consultant medicine, Clalit services, Nazareth, Israel.

Corresponding author:roseerlich@gmail.com

Type I Gaucher disease (GD) the autosomal recessively inherited lysosomal storage disorder caused by glucocerebrosidase deficiency, leads to the accumulation of glucocerebroside in macrophages, namely Gaucher cells (GCs) which infiltrate the reticuloendothelial system, bone marrow, spleen, liver, resulting in organ damage.

Hematological complications namely anemia thrombocytopenia and bleeling phenomena are considered the hallmark and frequently are the presenting signs and symptoms of Type I GD.

Not rarely, the course of GD is complicated by autoimmune phenomena and hematological malignancies which are attributed in recent studies to inflammatory processes and immune dysregulation induced by Gaucher macrophages.

Guidelines and a logarithm for the evaluation of early and late hematological complications will be delineated.

The frequency and causes of the association with malignant hematological neoplasias will be discussed focusing on the diagnosis and new therapeutic modalities.

Guideliness for the evaluation of anemia thrombocytopenia and bleeding tendency will be presented including iron, vitamin B12 profiles and inflammatory status. New aspects for estimation of chronic disease parameters in GD will be discussed.

Regarding "late" hematological complications namely hematological malignancies, the discussion will include special attention to the immune and lymphoproliferative disorders: gammopathies, multiple myeloma and malignant lymphomas.

Early diagnosis of "benign" and malignant hematological disorders in GD patients will improve outcome and prognosis. Increased awareness for the hematological manifestations and the high frequency of hematological malignancies will enable to analyze the risk factors for these co morbidities in GD.This approach might enable prevention of malignant hematological disorders during the course of GD.

Session 4

(13)

MANAGEMENT GOALS FOR TYPE 1 GAUCHER DISEASE: A CONSENSUS DOCUMENT FROM THE EUROPEAN WORKING GROUP ON GAUCHER DISEASE

M. Biegstraaten, T.M. Cox, N. Belmatoug, M.G. Berger, T. Collin-Histed, S. Vom Dahl, M. Di Rocco, C. Fraga, F. Giona, P. Giraldo, M.Hasanhodzic, D.A. Hughes, P.O. Iversen, A.I. Kiewiet, E. Lukina, M. Machaczka, T. Marinakis, E. Mengel, G.M. Pastores, U. Plöckinger, H. Rosenbaum, C. Serratrice, A. Symeonidis, J. Szer, J. Timmerman, A. Tylki-Szymańska, M. Weisz Hubshman, D.I. Zafeiriou, A. Zimran, C.E.M. Hollak

Presenting author:

Marieke Biegstraaten, MD, PhD Department of Internal Medicine, Division Endocrinology and Metabolism, Room F5-166 Academic Medical Center PO Box 22660 1100 DD Amsterdam, The Netherlands E m.biegstraaten@amc.uva.nl

Introduction: Gaucher Disease type 1 (GD1) is a lysosomal disorder that affects many systems. Enzyme therapy improves the principal visceral and haematological manifestations of the condition and, as a consequence, many patients show a modified phenotype which reflects manifestations of their disease that are refractory to treatment. More generally, it is increasingly recognised that information as to how a patient feels and functions [obtained by patient- reported outcome measurements (PROMs)] is critical to any comprehensive evaluation of treatment in the long term. Both trends should thus be reflected in management goals for GD1.

Methods: A modified Delphi procedure among GD1 experts was performed to reach consensus on management goals for GD1. Based on a literature review and with input from GD1 patients, 65 potential goals were formulated as statements. Participants were asked to indicate whether or not they agreed to include that specific statement in the management goals. Consensus was considered to be reached when ≥75% of the participants agreed.

Results: Twenty-five experts reached consensus on 42 management goals. In addition to the traditional goals concerning haematological, visceral and bone manifestations, improvement in quality of life, fatigue and participation in social activities, as well as early detection of long-term complications or associated diseases were included.

Conclusion: This new set of management goals for GD1 gives greater weighting than former treatment goals to long-term complications, associated conditions and PROMs. When applying this set of goals in medical practice, the clinical status of the individual patient should be taken into account.

Session 4

(14)

REVISED BELGIAN GUIDELINES FOR THE DIAGNOSIS, TREATMENT AND MONITORING OF GAUCHER'S DISEASE

Eyskens François[1]*, Cassimans David[2], Roland Dominique[3]

For the Belgian Working Group on Gaucher's Disease.

[1]Department of Pediatrics - Metabolic diseases, UZA, AZ Middelheim, Antwerp; [2]Department of Hepatology & Metabolic center, UZ-Leuven, Leuven; [3]Institut de Pathologie et de Génétique-Metabolic diseases, Gosselies (Charleroi), Belgium

*Corresponding author: francois.eyskens@provincieantwerpen.be

Background: In 2014 the idea came up to revise the Belgian Gaucher Guidelines that already dated from 2004. These guidelines aim the treating physician with respect to the necessary assessment(s) at diagnosis, the treatment of Gaucher's Disease (GD) as well as the monitoring of the patient during the follow up. It reflects not only international recommendations and literature but also the knowledge and expertise of Belgian experts. Methods: A review of the literature, based on a search of peer-reviewed literature published between2004-2015, was performed from May till November 2015. The quality of the body of evidence was determined by the different experts involved, and statements were formulated. Whenever possible statements were evidence based, if not they were based on expert opinion. Consensus was determined by a consensus meeting on November 26th 2015, followed by consensus e-form rounds and a final consensus meeting. The available data of treatment were presented by the invited medical directors of Genzyme/Sanofi and Shire in November 2015. Results: The current guidelines were developed by the working group focusing on a profound revising of the former Guidelines according to the recent knowledge, and adding new chapters on Genetics, Liver and Pulmonary involvement, the link with Parkinson's disease and some rare disease manifestations. Conclusion: With these Guidelines we aim to create broad awareness in order to promote the inclusion of GD in the differential diagnosis and to allow- and facilitate a prompt and correct diagnosis. Classification according to severity allows for an individual fine-tuned therapeutic approach.

Session 4

(15)

LYSO-GB1 AS A GAUCHER-SPECIFIC BIOMARKER: DATA FROM CLINICAL TRIALS USING VELAGLUCERASE ALFA

Qiu[3], Yulin Qin[3], Claudia Cozma[4], Sabrina Eichler[4], Arndt Rolfs[4,5]

[1] Shire Global Medical Affairs, Shire, Zug, Switzerland
[2] Gaucher Clinic, Shaare Zedek Medical Center, affiliated with the Hebrew University-Hadassah Medical School, Jerusalem, Israel
[3] Biostatistics and Statistical Programming Department, Shire, Lexington, MA, USA
[4] Centogene AG, Rostock, Germany
[5] Albrecht-Kossel-Institute for Neuroregeneration, University of Rostock, Rostock, Germany

*Corresponding author: delstein-c@shire.com

Background: Gaucher disease (GD) management may include evaluation of biomarkers but, to date, none are GD-specific. Glucosylsphingosine (lyso-Gb1) is a promising GD-specific biomarker, investigated previously in heterogeneous populations.

Methods: This was an exploratory analysis of lyso-Gb1 in blood samples from velaglucerase alfa clinical trials of ERT-naïve patients (164 samples) and those switched from imiglucerase (141). Lyso-Gb1, chitotriosidase and CCL18 were quantified and correlations with clinical GD markers investigated.

Results: Mean baseline lyso-Gb1 concentration was higher in ERT-naïve (n=22) than in switch patients (n=22): 323.2±140.5 ng/mL vs 91.8±84.8 ng/mL ($p<0.0001$). During velaglucerase alfa therapy, lyso-Gb1 decreased significantly from baseline in all patients: by 41.0% ($p<0.0001$) in ERT-naïve vs 14.1% ($p=0.0241$) in switch patients at week 25; and by 83.2% ($p<0.001$) vs 51.9% ($p<0.0001$) at week 161. In ERT-naïve patients, lyso-Gb1 reduction rate was fastest within the first 24 weeks and, as expected, was faster than in switch patients throughout the first 100 weeks ($p<0.0001$), although reduction in switch patients continuously accelerated. Percentage reductions in lyso-Gb1 correlated with chitotriosidase/CCL18 in ERT-naïve ($r\geq0.60$ at weeks 25, 53, 101) and switch patients ($r\geq0.19$). Lyso-Gb1 concentrations correlated with platelet counts and normalized spleen volumes over time in ERT-naïve ($p<0.0001$ and $p<0.0001$) and switch ($p=0.0109$ and $p=0.0008$) patients, despite interpatient variation in these clinical markers.

Conclusions: These data support earlier studies and demonstrate the utility of lyso-Gb1 as a GD-specific biomarker that also correlates with GD-specific parameters. Treatment with velaglucerase alfa resulted in significant decreases in lyso-Gb1 in ERT-naïve patients and in those switched from imiglucerase.

Session 5

(16)

IN VIVO CELLULAR PATHWAYS FOR INFUSED HUMAN RECOMBINANT GLUCOCEREBROSIDASE

Margarita M Ivanova [*1], Erk Changsila[1], Chadima Iaonou[1,2] and Ozlem Goker-Alpan[1,2]

[1]Lysosomal and Rare Disorders Research and Treatment Center, Fairfax, VA, USA

[2]O&O Alpan LCC, Fairfax, VA, USA

[*] Corresponding author: mivanova@ldrtc.org

The major cellular pathway for organelle turnover and clearance of unwanted proteins is the autophagy-lysosome pathway (ALP). Lysosomes not only house proteolytic enzymes, but also traffic organelles, sense nutrients, repair mitochondria. ALP dysfunction is expected to be the initial step for the cascade of events leading to Gaucher disease (GD). In vitro studies in different cell models demonstrate lysosomal dysfunction in GD, little is known about the in vivo effects of enzyme replacement therapy (ERT) on different cellular functions, including ALP and mitochondrial function. We investigated the effects of the infused ERT on different dynamic cellular events in PBMCs obtained before and 3h after the ERT infusion. Autophagosome and lysosome staining, ATP assay, RNA and protein expression analysis of ALP and mitochondrial genes were performed. Western blot analysis of GBA demonstrated robust uptake of recombinant GBA by PBMCs during the ERT infusion. The rate of autophagosome and lysosome formation in PBMCs did not change after infusion. However, LC3-II accumulation and decreased LC3-I/LC3-II ratios were observed after ERT, indicating autophagy initiation. Levels of lysosomal, autophagy, and mitochondrial markers - LAMP1, Beclin1, and Tfam - were declined to normal after ERT infusion. In vitro treatment with rhGCase improved autophagosome vesicle formation level in control and GD PBMC. 3h rhGCase treatment induced lysosome levels in control PBMCs but not in GD samples. Enzyme replacement therapy normalizes ALP and energy metabolism. While the infused hrGCase is known to be cleared immediately from the circulation, in vivo effects continue to much longer after the administration.

Session 5

(17)

INTRAVENOUSLY ADMINISTERED GENE THERAPY FOR THE TREATMENT OF NEURONOPATHIC GAUCHER DISEASE

Massaro Giulia[1]*, Perocheau Dany[2], Baker Rob[3], Karlsson Stefan[4], Cheng Seng H.[5], Waddington Simon N.[2], Rahim Ahad A.[1]

[1]Department of Pharmacology, UCL School of Pharmacy, London, UK. [2]. Gene Transfer Technology Group, UCL Institute for Women's Health, London, UK. [3].

Molecular Diagnostic Lab, Haematology – Royal Free Site, HSL LLP, London, UK. 4. Division of Molecular Medicine and Gene Therapy, Lund University, Lund, Sweden. 5. Genzyme, A Sanofi Company, USA.

*Corresponding author: giulia.massaro.13@ucl.ac.uk

Gaucher Disease is caused by mutations in the GBA gene encoding the enzyme glucocerebrosidase. Acute neuronopathic Gaucher Disease (nGD) is characterised by neuronal loss, astrocytosis and microglial proliferation. nGD is untreatable since enzyme replacement therapy cannot cross the blood-brain barrier.

AAV9 has been demonstrated to be able to efficiently transduce the Central Nervous System and visceral organs following intravenous administration to mice and non-human primates.

In this study, I tested the hypothesis that neonatal intravenous injection of adeno-associated viral vector serotype 9 (AAV9), carrying functional GBA gene, would improve lifespan, behavior, brain and visceral pathology in a mouse model of nGD.

Untreated KO mice die 12-14 days after birth. Treated mice showed a >10-fold increase in their lifespan. Neuropathological markers such as microglia- mediated inflammation, astrogliosis and lysosomal accumulation were ameliorated and some of the most affected areas of the brain, like thalamus, brain stem and cerebellum were partially rescued. Histologic analysis, enzymatic assay and blood test revealed improvement in the visceral pathology; in the lung, spleen and liver the presence of Gaucher cells is significantly reduced and tissue architecture is preserved.

Session 5

(18a)

LONG-TERM SAFETY AND EFFICACY OF TALIGLUCERASE ALFA IN PEDIATRIC PATIENTS WITH GAUCHER DISEASE WHO WERE TREATMENT-NAÏVE OR PREVIOUSLY TREATED WITH IMIGLUCERASE

Ari Zimran, MD[1], Derlis Emilio Gonzalez-Rodriguez, MD[2], Aya Abrahamov, MD[1], Peter A. Cooper, PhD[3], Sheeba Varughese, PhD[3], Einat Brill-Almon, PhD[4], Raul Chertkoff, MD[4]

[1]Gaucher Clinic, Shaare Zedek Medical Center, Jerusalem, Israel, and the Hadassah Medical School - Hebrew University, Jerusalem, Israel; [2]Instituto Privado de Hematologia e Investigacion Clinica, Asuncion, Paraguay; [3]Department of Paediatrics, University of the Witwatersrand, Johannesburg, South Africa; [4]Protalix BioTherapeutics, Carmiel, Israel

Taliglucerase alfa is an enzyme replacement therapy approved for treatment of patients with Type 1 Gaucher disease (GD) and is the first approved plant cell–expressed recombinant therapeutic protein. This extension study of taliglucerase alfa in pediatric patients included those who were either treatment-naïve (n=10) or who were previously switched from imiglucerase (n=5). Patients received taliglucerase alfa 30 U/kg or 60 U/kg (treatment-naïve patients) or at the same dose as previously treated with imiglucerase (switch patients). In treatment-naïve patients, taliglucerase alfa 30 and 60 U/kg, respectively, increased mean hemoglobin concentration (+19.7% and +23.3%) and mean platelet count (+23.9% and +156.6%) while also reducing mean spleen volume (-67.8% and -68.9%), liver volume (-37.0% and -34.3%), and chitotriosidase activity (-72.7% and -84.4%) from baseline through 36 total months of treatment. In patients previously treated with imiglucerase, these disease parameters remained stable through 33 total months of treatment with taliglucerase alfa. In both studies, most adverse events were mild/moderate and treatment was well tolerated. These long-term results of taliglucerase alfa in pediatric patients with GD extend the taliglucerase alfa clinical safety and efficacy data set.

Disclosures: This study was sponsored by Protalix BioTherapeutics. Pfizer and Protalix entered into an agreement in November 2009 to develop and commercialize taliglucerase alfa.

Keywords: Gaucher disease; enzyme replacement therapy; taliglucerase alfa.

Session 5

(18b)

LONG-TERM EFFICACY AND SAFETY RESULTS OF TALIGLUCERASE ALFA THROUGH 5 YEARS IN ADULT TREATMENT-NAÏVE PATIENTS WITH GAUCHER DISEASE

Ari Zimran, MD,[1] Gloria Durán, MD,[2] Pilar Giraldo, MD, PhD,[3] Hanna Rosenbaum, MD,[4] Fiorina Giona, MD,[5] Milan Petakov, MD,[6] Sergio Eduardo Solorio-Meza, MD,[7] Peter A. Cooper, FC Paed (SA), PhD,[8] Sari Alon,[9] and Raul Chertkoff, MD[9]

[1]Gaucher Clinic, Shaare Zedek Medical Center, Hebrew University and Hadassah Medical School, Jerusalem, Israel; [2]Pontificia Universidad Catolica de Chile, Santiago, Chile; [3]CIBERER, Hospital Universitario Miguel Servet, Zaragoza, Spain; [4]Rambam Medical Center, Haifa, Israel; [5]Sapienza University, Rome, Italy; [6]Clinical Center of Serbia, Clinic of Endocrinology, Diabetes and Metabolic Disease, Belgrade University Medical School, Belgrade, Serbia; [7]Hospital de Especialidades No1, Leon, México; [8]Faculty of Health Sciences, University of the Witwatersrand & Charlotte Maxeke Johannesburg Academic Hospital, Johannesburg, South Africa; [9]Protalix BioTherapeutics, Carmiel, Israel;

Taliglucerase alfa is an enzyme replacement therapy approved for treatment of type 1 Gaucher disease (GD). This extension study of the long-term efficacy and safety of taliglucerase alfa in adult treatment-naïve patients enrolled 19 patients with GD (8 patients received 30 U/kg; 9 patients received 60 U/kg; 2 patients initially received 30 U/kg but had their doses adjusted to 45 and 60 U/kg after 27 total months and who were analyzed separately). Seventeen patients completed the study and received taliglucerase alfa for 60 months; 2 discontinued. Throughout the study, spleen and liver volumes decreased continuously while hemoglobin concentrations and platelet counts increased. Percent changes from baseline to 60 months were: spleen volume, −56.7% (30 U/kg; n=7), −67.9% (60 U/kg; n=7), and −61.0% (adjusted dose; n=2); liver volume, −32.5% (30 U/kg; n=7), −23.3% (60 U/kg; n=7), and −30.4% (adjusted dose; n=2); hemoglobin concentration, +18.8% (30 U/kg; n=7), +20.5% (60 U/kg; n=8), and +14.6% (adjusted dose; n=2); platelet counts, +44.8% (30 U/kg; n=7), +168.0% (60 U/kg; n=8), and +52.8% (adjusted dose; n=2). The biomarkers of chitotriosidase activity and CCL18 concentration decreased through 60 months. Taliglucerase alfa was well tolerated, with no treatment-related adverse events (AEs) observed. All AEs were mild to moderate in intensity, except for 2 serious AEs which were not considered treatment-related; both patients completed the study. Treatment-naïve adult patients with GD demonstrated continued improvement in disease parameters with taliglucerase alfa, with no new safety concerns through 60 months, extending the clinical efficacy and safety dataset for taliglucerase alfa.

Session 5

(19)

CNS-ACCESSIBLE INHIBITOR OF GLUCOSYLCERAMIDE SYNTHASE FOR SUBSTRATE REDUCTION THERAPY OF NEURONOPATHIC GAUCHER DISEASE

John Marshall[1], Dinesh Bangari[1], Jennifer B. Nietupski[1], Mary A. Cromwell[1], Gregory A. Grabowski[2], John P. Leonard[1], Seng H. Cheng[1]

[1]Sanofi Genzyme, Framingham, MA, USA

[2]Cincinnati Children's Hospital Medical Center, Cincinnati, OH, USA.

*Corresponding author: seng.cheng@sanofi.com

Gaucher disease (GD) is caused by a deficiency of glucocerebrosidase and the consequent lysosomal accumulation of unmetabolized glycolipid substrates. Enzyme-replacement therapy adequately manages the visceral manifestations of non-neuronopathic type 1 Gaucher patients, but not the brain disease in neuronopathic types 2 and 3 GD. Substrate reduction therapy through inhibition of glucosylceramide synthase has also been shown to effectively treat the visceral disease. Here, we evaluated the efficacy of a novel small molecule inhibitor of glucosylceramide synthase with CNS access (Genz-682452) to treat the brain disease. Treatment of the conduritol B epoxide-induced mouse model of neuronopathic GD with Genz-682452 reduced the accumulation of liver and brain glycolipids (>70% and >20% respectively), extent of gliosis, and severity of ataxia. In the genetic 4L;C* mouse model, Genz-682452 reduced the levels of substrate in the brain by >40%, the extent of gliosis, and paresis. Importantly, Genz-682452-treated 4L; C* mice also exhibited an ~30% increase in lifespan. Together, these data indicate that an orally available antagonist of glucosylceramide synthase that has CNS access is effective at attenuating several of the neuropathologic and behavioral manifestations associated with mouse models of neuronopathic GD. Therefore, Genz-682452 holds promise as a potential therapeutic approach for patients with type 3 GD.

Session 5

(20)

FOUR-YEAR FOLLOW-UP FROM THE ENCORE TRIAL: ORAL ELIGLUSTAT IN PATIENTS WITH GAUCHER DISEASE TYPE 1 STABILIZED ON ENZYME THERAPY

Ozlem Goker-Alpan[1*], Timothy M Cox[2], Guillermo Drelichman[3], Renata Cravo[4],Manisha Balwani[5], Thomas Andrew Burrow[6], Ana Maria Martins[7], Elena Lukina[8], Barry Rosenbloom[9], Nora Watman[10], Amal el Beshlawy[11], Priya Kishnani[12], Maria Lucia Pedroso[13], Sebastiaan JM Gaemers[14], Regina Tayag[15], M Judith Peterschmitt[14]

[1]Lysosomal Disorders Unit, O&O Alpan, LLC, Fairfax, VA, USA; [2]University of Cambridge, Addenbrooke's Hospital, Cambridge, UK; [3]Hospital de Niños Ricardo Gutiérrez, Buenos Aires, Argentina; [4]State Institute of Haematology 'Arthur de Siqueira Cavalcanti', Rio de Janeiro, Brazil; [5]Mount Sinai Hospital, New York, NY, USA; [6]Cincinnati Children's Hospital Medical Center, Cincinnati, OH, USA, [7]Universidade Federal de São Paulo, São Paulo, SP, Brazil; [8]National Research Center for Hematology, Moscow, Russia; [9]Cedars-Sinai/Tower Hematology Oncology, Beverly Hills, CA, USA; [10]Hospital Ramos Mejia, Buenos Aires, Argentina; [11]Pediatric Hematology, Cairo University, Egypt; [12]Duke University School of Medicine, Department of Pediatrics, Durham, NC, USA; [13]Hospital de Clinicas, Universidade Federal do Paraná, Curitiba, PR, Brazil; [14]Sanofi Genzyme, Cambridge, MA, USA; [15]Prometrika, Cambridge, MA, USA

*Corresponding author: ogokeralpan@oandoalpan.com

Eliglustat is an oral substrate-reduction therapy approved in the US and Europe for adults with Gaucher disease type 12 months (Cox; Lancet. 2015;385:2355). During the trial extension, patients (all eliglustat-treated) could accrue 2.5-4 years of eliglustat treatment, depending on when they enrolled, how they had been randomized, and where they lived (51 US patients were switched to commercial drug when it became available). We report long-term safety and efficacy with respect to years on eliglustat for all 157 eliglustat-treated patients in ENCORE, 46 of whom have 4-year data. Throughout the trial, mean values for hemoglobin concentration, platelet count, spleen volume, and liver volume remained stable. Year to year, all four measures remained stable collectively (composite endpoint relative to baseline values) in ≥85% of patients and individually in ≥91% of patients. All four therapeutic goals established for patients on ERT for hemoglobin, platelet, spleen, and liver were maintained in ≥92% of patients and each individual goal was maintained by ≥94%. Mean bone mineral density Z-scores (lumbar spine and femur) remained normal throughout the trial. Eliglustat was well-tolerated; 4 patients (2.5%) withdrew for adverse events considered related to eliglustat. No long-term safety concerns were identified. In summary, clinical stability by composite and individual measures was maintained in eliglustat-treated patients with GD1 who remained in the ENCORE trial long-term.

Session 5

(21)

TRANSFORMATION IN PRE-TREATMENT PRESENTATIONS OF GAUCHER DISEASE IN FIRST 2 DECADES OF IMIGLUCERASE ENZYME REPLACEMENT THERAPY: REPORT FROM THE INTERNATIONAL COLLABORATIVE GAUCHER GROUP GAUCHER REGISTRY

Pramod Mistry[1]*, Hans C. Andersson[2], Manisha Balwani[3], Julie L. Batista[4], T. Andrew Burrow[5], Joel Charrow[6], Paige Kaplan[7], Neal Khan[8], Priya S. Kishnani[9], Edwin H. Kolodny[10], Barry Rosenbloom[11], C. Ronald Scott[12], Neal Weinreb[13]

[1] Department of Internal Medicine, Yale University School of Medicine, New Haven, CT, USA [2] Tulane University Medical School, New Orleans, LA, USA [3] Icahn School of Medicine at Mount Sinai, New York City, NY, USA. [4] Sanofi Genzyme, Cambridge, MA, USA [5] Cincinnati Children's Hospital Medical Center, Cincinnati, OH, USA [6] Northwestern University Feinberg School of Medicine, Chicago, IL, USA [7] Children's Hospital of Philadelphia, Philadelphia, PA, USA [8] The University of Calgary, Calgary, Alberta, Canada [9] Duke University School of Medicine, Durham, NC, USA [10] New York University School of Medicine, New York, NY, USA [11] Cedars-Sinai/Tower Hematology Oncology, Beverly Hills, CA, USA [12] University of Washington, Seattle, WA, USA [13] University Research Foundation for Lysosomal Storage Diseases, Coral Springs, FL, USA

*Corresponding author: pramod.mistry@yale.edu

We hypothesized that the prevalence of Gaucher disease type 1(GD1) clinical manifestations at the time of treatment initiation has changed since alglucerase/imiglucerase enzyme replacement therapy (ERT) was approved in the United States in 1991. US imiglucerase-treated GD1 patients (N=1,516) from the International Collaborative Gaucher Group Gaucher Registry (ClinicalTrials.gov NCT00358943, sponsored by Sanofi Genzyme) were stratified by age at ERT initiation: n=434 pediatric patients(<18 y); n=724 young adults (18 to <50 y); n=358 older adults (≥50 y). We analyzed pre-treatment demographics, splenectomy status, hematologic parameters, and bone manifestations for each cohort by year of ERT initiation using the chi-square test or Fisher's exact test, when appropriate. The most striking differences between early and later time periods (Table below) included reduced prevalence of splenectomy, avascular necrosis (AVN), bone crises in the two younger age groups, and fractures. However, the results for fracture prevalence in all age groups and bone crisis prevalence among older adults did not reach statistical significance (defined as $p<0.05$), likely due to sparse data. Changes were not observed in hematologic parameters. Compared with patients who started treatment when ERT was first introduced, patients now tend to have a less severe GD1 phenotype and fewer irreversible manifestations of GD1 when starting treatment.

TREATMENT INITIATION TIME PERIOD

	Age Group	1991-1995 n/N (%)*	1996-2000 n/N (%)*	2001-2005 n/N (%)*	2006-2009 n/N (%)*	p-value
Splenectomy	*Pediatric*	*44/189 (23.3)*	*4/120 (3.3)*	*2/86 (2.3)*	*2/39 (5.1)*	*<0.001*
	Young Adult	*182/389 (46.8)*	*34/161 (21.1)*	*22/126 (17.5)*	*5/46 (10.9)*	*<0.001*
	Older Adult	*73/158 (46.2)*	*26/88 (29.5)*	*15/71 (21.1)*	*10/41 (24.4)*	*<0.001*
AVN	*Pediatric*	*19/31 (61.3)*	*4/40 (10.0)*	*3/18 (16.7)*	*0/10 (0.0)*	*<0.001*
	Young Adult	*41/65 (63.1)*	*14/67 (20.9)*	*7/50 (14.0)*	*4/18 (22.2)*	*<0.001*
	Older Adult	*21/32 (65.6)*	*11/45 (24.4)*	*7/27 (25.9)*	*3/13 (23.1)*	*<0.001*
Bone Crises	*Pediatric*	*26/81 (32.1)*	*10/82 (12.2)*	*4/58 (6.9)*	*1/26 (3.8)*	*<0.001*
	Young Adult	*44/133 (33.1)*	*9/74 (12.2)*	*2/66 (3.0)*	*2/23 (8.7)*	*<0.001*
	Older Adult	*14/67 (20.9)*	*5/55 (9.1)*	*2/40 (5.0)*	*2/25 (8.0)*	*0.077*
Fractures	*Pediatric*	*3/9 (33.3)*	*3/41 (7.3)*	*2/24 (8.3)*	*0/7 (0.0)*	*0.141*
	Young Adult	*4/24 (16.7)*	*3/46 (6.5)*	*2/32 (6.3)*	*0/8 (0.0)*	*0.425*
	Older Adult	*2/14 (14.3)*	*8/34 (23.5)*	*1/17 (5.9)*	*1/6 (16.7)*	*0.437*

** 'n' indicates an occurrence; 'N' indicates the total number of patients evaluated in each stratum.*

Session 5

(22)

DEVELOPMENT OF A NEW THERAPEUTIC STRATEGY FOR GAUCHER DISEASE (GD) USING EX VIVO TARGETED GENOME EDITING

Fabiola Porro[2], Erika Malini[1], Bruno Bembi[1], Andres Muro[2], Andrea Dardis[1*]

[1] Regional Coordinator Centre for Rare Diseases, University Hospital Udine, Italy

[2] International Centre for Genetic Engineering and Biotechnology (ICGEB), Trieste, Italy

[*] Corresponding author: dardis.andrea@aoud.sanita.fvg.it

Ex-vivo gene therapy using lentivirus to transduce autologous hematopoietic stem cell (HSCs) has been used in mouse models and presymptomatic patients with neurological lysosomal disorders. This approach holds the concern of insertional mutagenesis, which could be overcome by using site-specific genome targeting techniques. The aim of this work was to develop an ex-vivo gene therapy for Gaucher disease (GD) based on targeted genome editing.

Our approach was based on the site-specific insertion of the wild type GBA cDNA into the AAVS1 safe harbour locus. Therefore, engineering nucleases (TALENs and CRISPR-Cas9) were used to specifically create double strand breaks within the AAVS1 locus to trigger the insertion of the therapeutic cDNA by homologous direct repair.

The donor vector AAVS1-EGFP-PGKpromGBA was obtained by cloning the EGFP cDNA (devoid of promoter), in frame with the 2A peptide, and the wild type GBA cDNA (under the control of the PGK promoter) into the AAVS1-SA-2A-puro-pA-donor vector. This plasmid, which contains the AAVS1 homology sequences, was co-transfected with TALENs or CRISPR-Cas9 into Hela cells. After integration, EGFP expression is driven by the endogenous AVVS1 promoter; thus, only edited cells express EGFP. Editing efficiency (% of EGFP expressing cells) was 5-10 % depending on the used nuclease. GBA overexpression, after sorting edited cells, was assessed.

Using this strategy we were able to efficiently and specifically target the insertion of the GBA cDNA within the AVVS1 safe harbour locus. This approach could be used for the in vitro correction of autologous HSCs for the treatment of GD.

Session 5

(23)

pH-SENSITIVE GLUCOCEREBROSIDASE LIGANDS AS NON-INHIBITORY PHARMACOLOGICAL CHAPERONES FOR GAUCHER DISEASE

José M. Garcia-Fernández[1]*, Carmen Ortiz-Mellet[2], Teresa Mena-Barragán[2], Katsumi Higaki[3], Eiji Nanba[3], Gustavo Tiscornia[4], Aya Narita[5], Kousaku Ohno[5,6], Yoshiyuki Suzuki[7]

[1] Instituto de Investigaciones Químicas (IIQ), CSIC-Universidad de Sevilla, Sevilla, Spain

[2] Department of Organic Chemistry, Faculty of Chemistry, University of Sevilla

[3] Division of Functional Genomics, Research Center for Bioscience and Technology, Tottori University, Tottori, Japan

[4] Center for Biomedical Research, Depertamento de Ciencias, Biomedicas y Medicina/

Programa de Medicina Regenerativa, Universidad do Algarve, Faro, Portugal

[5] Division of Child Neurology, Faculty of Medicine, Tottori University, Tottori, Japan

[6] Sanin Rosai Hospital, Tottori, Japan

[7] Tokyo Metropolotan Institute of Medical Science

*Corresponding author: jogarcia@iiq.csic.es

Objective: Pharmacological chaperone therapy (PCT) represents an interesting small-drug option for protein folding diseases in general and lysosomal storage disorders in particular. Most of the PCs under investigation, either developed as de novo candidates or discovered from drug repositioning programs, were identified as active site-directed competitive inhibitors for the targeted enzyme. Their mechanism of action implies binding at the active site, stabilizing the misfolded mutant protein in the endoplasmic reticulum (ER) and restoring trafficking to the lysosome, resulting in function recovery. However, there is a risk that progressive accumulation of the PC in the lysosome will prevent substrate processing even if enzyme rescuing was successful. To circumvent this problem, we have now designed a new generation of pH-responsive PCs that undergo fast self-inactivation at the acidic environment of the lysosome, thereby maximizing chaperone versus inhibitor behavior. Methods: To impart acid vulnerability to PCs, a prototype was develeoped that combines an iminosugar moiety and a hydrophobic tail connected through an orthoester group. Both segments are necessary for enzyme binding. At neutral pH (ER) the orthoester functionality remains stable and the compound display its PC potential, but at acidic pH (lysosome) it undergoes fast hydrolysis and the resulting moieties bocome no ligands for the enzyme, which remains free for substrate processing. Results: New highly pH-responsive glycomimetics targeting human glucocerebrosidase have been designed and a proof of concept of their potential as PCs for the treatment of Gaucher disease has been provided by immunodetection and enzyme activity enhancement determinations.

Session 7

(24)

GLUCOSYLCERAMIDE SYNTHASE INHIBITION REDUCES a-SYNUCLEIN PATHOLOGY AND IMPROVES COGNITION IN MURINE MODELS OF SYNUCLEINOPATHY

S. Pablo Sardi, Catherine Viel, Jennifer Clarke, James C. Dodge, John Marshall, Bing Wang, Lamya S. Shihabuddin, Seng H. Cheng

Sanofi Genzyme, Framingham, MA, USA

*Corresponding author: seng.cheng@sanofi.com

Mutations in *GBA1*, the gene encoding glucocerebrosidase, confer a heightened risk of developing synucleinopathies such as Parkinson's disease (PD). Recent studies have also demonstrated that genetic variation in *GBA1* can impact the progression of PD. Patients harboring mutations in *GBA1* present higher prevalence and severity of motor and non-motor symptoms. However, the precise mechanisms by which mutations in *GBA1* increase PD risk and exacerbate its progression remain unclear. Here, we investigated the merits of glucosylceramide synthase (GCS) inhibition as a potential treatment for synucleinopathies. A Gaucher-related synucleinopathy mouse model ($Gba^{D409V/D409V}$) was treated with an orally available and brain-penetrant GCS inhibitor, Genz-667161 for 8 months. This intervention prevented CNS substrate lipid accumulation. Most notably, treatment with the GCS inhibitor slowed the accumulation of hippocampal aggregates of α-synuclein, ubiquitin and tau, and improved the associated memory deficits. The effects of the GCS inhibitor were also studied in a mouse model overexpressing α-synuclein, *PrP-A53T-SNCA,* and harboring wild type alleles of *GBA1*. Treatment of *PrP-A53T-SNCA* mice with Genz-667161 for 6.5 months reduced membrane-associated α-synuclein in the CNS and ameliorated cognitive deficits. Collectively, the data indicate that inhibition of GCS can modulate processing of α-synuclein and reduce various α-synuclein entities, thereby reducing the progression of synucleinopathies in mice with and without mutations in *GBA1*. The present study provides supporting evidence for the clinical development of brain-penetrant GCS inhibitors in PD and other synucleinopathies.

Session 7

(25)

GBA1 HAPLOINSUFFICIENCY IN A PARKINSON MOUSE MODEL RESULTS IN AN EARLIER DISEASE ONSET AND MORE RAPID PROGRESSION OF SYMPTOMS

Nahid Tayebi[1], Loukia Parisiadou[2], Ashley N Gonzalez[1], Mieu Brooks[3], Chrissy Makariou-Pikis[2], Bahafta Berhe[1], Niraj Trivedi[4], Abdel Elkahloun[5], Richard Grey[1], Benoit I Giasson[3] and Ellen Sidransky[1]

[1] MGB, [4]CSGB and [5]CGDGB, NHGRI, NIH, Bethesda MD,[2] Department of Radiation Oncology, Feinberg School of Medicine, Chicago, IL and [3]Department of Neuroscience, University of Florida College of Medicine, Gainesville, FL.

Background: Mutations in *GBA1* are a leading genetic risk factor for PD and, overall, patients with *GBA1* mutations experience an earlier PD onset and more severe symptoms. Severe or null *GBA1* mutations are associated with a higher risk for PD and a more progressive course.

Methods: Transgenic mice hemizygous or homozygous for human A53 α-synuclein (*SNCA*^{A53T}) were mated with C57Bl/6J *Gba*$^{+/-}$ mice, yielding offspring that overexpressed SNCA^{A53T} and carried a null *Gba* allele. More than 200 mice were followed for up to 2 years. SNCA copy number was estimated using TaqMan assays. Weight loss was the initial sign of illness, followed by arching, and limb paralysis. Symptomatic mice were euthanized together with their age-matched controls (wt/wt, wt/SNCA^{A53T} and *Gba*$^{+/-}$). A whole transcriptome array was performed on mice with each genotype. Immunohistochemistry (IHC) and protein fractionation were used to quantify the monomeric and aggregate forms of SNCA ^{A53T} and related proteins.

Results: Survival analysis of 84 mice showed that *gba*$^{+/-}$//SNCA^{A53T} hemizygous and homozygous mice exhibit symptoms on average 9.76 weeks and 4.04 weeks earlier (p-values 0.023, 0.0030). Further, disease progression was more rapid in the *Gba*$^{+/-}$//SNCA^{A53T} mice (p-value <0.0001). Quantification of SNCA aggregates showed no significant effect of the *Gba*$^{+/-}$ allele. The expression arrays revealed several significantly over expressed genes. *Gba*$^{+/-}$// SNCA^{A53T} mice had less high molecular weight SNCA^{A53T} and more Glial fibrillary acidic protein than SNCA^{A53T} mice.

Conclusion: Glucocerebrosidase haploinsufficiency enhances the severity and progression of neurological symptoms, providing a valuable mouse model to elucidate the basis of this association.

Session 7

(26)

PROCESSING OF ALPHA-SYNUCLEIN AND PARKIN IN PERIPHERAL BLOOD MONONUCLEAR CELLS IN PATIENTS WITH GAUCHER DISEASE

Renuka Pudi Limgala*, Ariel Badger, Chandni Sejpal and Ozlem Goker-Alpan
Lysosomal and Rare Disorders Research and Treatment Center, Fairfax, VA, USA
O & O Alpan, LLC, Fairfax, VA, USA
*Corresponding author: rlimgala@ldrtc.org

Gaucher disease (GD) patients display a wide range of clinical symptoms and present with serious comorbidities. It has been shown that patients with GD, even carriers with one mutated *GBA* gene are at a higher risk for developing Parkinson's disease (PD), and at an earlier age. Mutations within *SNCA (*a-synuclein)*, LRRK2, PRKN* (Parkin) are implicated in other genetic forms of parkinsonism. a-Synuclein is known to form aggregates in cases of PD, and shown to decrease solubility of Parkin. While the role of LRRK2 is largely unclear, the neuropathology may share similarities to *GBA*-associated PD cases. As an attempt to explore the relationship between these molecules and *GBA*, we investigated the expression at both molecular and protein level in the peripheral blood mononuclear cells (PBMCs). We compared patients with GD manifesting PD symptoms (GD-PD) to GD patients without PD symptoms (GD-nonPD) and controls. At the mRNA level, there was increased expression of LRRK2 and SNCA, in both GD-PD and GD-nonPD compared to controls. However, there was a significant increase in expression of LRRK2 and a-synuclein in subjects with GD-PD only, at protein level. Since a-synuclein aggregation is known to affect solubility and subsequent accumulation of Parkin, we studied Parkin in PBMCs and found a correlation with a-synuclein levels. Flow cytometry also revealed a fraction of PBMCs from GD-PD with overexpression of both Parkin and a-synuclein indicating possible aggregation of these proteins. As this finding was unique to GD-PD, the mechanisms underlying this observation may shed light to development of future biomarkers.

Session 7

(27)

STUDYING THE LINK BETWEEN GAUCHER AND PARKINSON'S DISEASE. CELL MODELS AND DISEASE-MODIFYING FACTORS

Derek Burke[1,2*], Carmen de la Fuente-Barrigon[2], Maria Garcia-Gomez[2], and Simon Heales[1,2].

[1] Chemical Pathology, Great Ormond Street Hospital, London, UK.

[2] Genetics and Genomic Medicine, UCL Institute of Child Health, University College London, UK;

*Corresponding author :Derek.burke@gosh.nhs.uk

Research funded by [1]Great Ormond Street Hospital Children's Charity and [2] TINTIN European Project and Marie Curie Initial Training Network.

GBA1 mutations, oxidative stress and loss of mitochondrial function are associated with Parkinson's Disease (PD). GBA1 activity is decreased in PD patients without GBA1 mutations. In view of this, we have studied the effects of;

- Oxidative stress on cell viability, GBA1and other lysosomal enzyme activities
- GBA1 inhibition on cell viability +/- oxidative stress
- Complex I deficiency on GBA1 activity
- GBA1 deficiency upon mitochondrial content and dopamine metabolism

SHSY5Y cells were exposed to hydrogen peroxide (H_2O_2). Cell viability and the activities of GBA1, beta-galactosidase (b-gal), total beta-hexosaminidase (b-Hex), acid alpha-glucosidase (GAA) and citrate synthase (mitochondrial content) were assessed. Complex I deficiency was created by rotenone exposure and GBA1 inhibition. Dopamine metabolism was assessed, following a L-dopa challenge, in the extracellular medium. H_2O_2 caused a dose dependent loss of cell viability, which was not altered by GBA1 inhibition. Oxidative stress did not affect GBA1 or b-gal activity but b-Hex activity was significantly increased. Complex I inhibition had no effect on GBA1 activity. GBA1 inhibition was associated with increased generation of the dopamine metabolite, DOPAC. Loss of GBA1 activity in idiopathic PD may not be explained by oxidative stress or loss of mitochondrial complex I. However, the oxidative stress mediated increase in b-Hex may provide a mechanism for the recently reported increased activity of b-hex in PD. GBA1 deficiency may lead to increased catabolism of dopamine, raising the possibility of an involvement of monoamine oxidase. The latter is required for DOPAC formation and is reported to be upregulated in PD.

Session 7

(28)

GLUCOCEREBROSIDASE VARIANTS ARE MODIFIERS FOR ONSET AND CLINICAL SYMPTOMS IN PARKINSON DISEASE PATIENTS – A MULTINATIONAL STUDY OF GBA GENE SEQUENCES USING NEXT GENERATION SEQUENCING (NGS)

Rike Bender1*, Anne-Katrin Giese[1/2], Eva-Juliane Vollstedt[3/4], Ulrike Grittner[5], Maximilian Weiss[6], Ilona Csoti[7], Roongroj Bhidayasiri[8], Gregor Kuhlenbäumer[9], Matthias Wittstock[10], Tobias Böttcher[10], Matthias Hahne[11], Wolfgang Jost[12], Birgit Herting[13], Meike Kasten3[/4], Christine Klein[3], Arndt Rolfs[1/6]

[1] Albrecht-Kossel-Institute for Neuroregeneration, University of Rostock, Rostock, Germany 2 Department of Neurology, Massachusetts General Hospital, Boston, USA [3]Institute of Neurogenetics, University of Lübeck, Lübeck, Germany 4 Department of Psychiatry and Psychotherapy, University of Lübeck, Lübeck, Germany [5] Department for Biostatistics and Clinical Epidemiology, Charité-University Medical Centre, Berlin, Germany [6] Centogene AG, The Rare Disease Company, Rostock, Germany [7] Department of Neurology, Gertrudis Hospital, Leun-Biskirchen, Germany [8] Chulalongkorn Centre of Excellence for Parkinson's Disease and Related Disorders, Chulalongkorn University Hospital Bangkok 10330, Thailand [9] Department of Neurology, Christian-Albrechts-University of Kiel and University Medical Center, Kiel,Germany [10] Department of Neurology, University of Rostock, Rostock, Germany [11] Clinic of Neurology, University of Marburg, Bad Neustadt, Germany [12] Department of Neurology, Deutsche Klinik für Diagnostik Wiesbaden, Wiesbaden, Germany [13] Diakonie-Klinikum Schwäbisch Hall GmbH, Schwäbisch Hall, Germany

* Corresponding author: rike.bender@uni-rostock.de

Gaucher disease (GD) is caused by mutations in the glucocerebrosidase (GBA) gene and is associated with a 5-10-fold increased risk for development of Parkinson disease (PD). To examine this link further, we carried out a multinational study screening for GD variants in extensively phenotyped PD patients. In total, 1,391 patients with idiopathic PD from 19 participating centers were enrolled (IRB approval by University of Rostock, HV-2009-0013; NCT01272687). Demographic and clinical data was obtained for all patients including age of onset, family history, standardized clinical PD rating, and neuropsychological testing. The complete GBA gene was sequenced using NGS. Overall, 215 subjects (15.5%) with GBA-sequence variations were detected in the PD cohort. Of those, 61 (4.4%) had GD-associated mutations. The remaining 154 subjects (11.1%) carried genetic variants, which are most likely modifiers influencing PD risk. PD subjects with GD-associated mutations had a lower age of onset (mean: 51, SD: 11) than subjects without variants (mean: 56, SD: 13; p=0.001). Family history for PD (25.5% vs. 19.8%, p=0.33) and depressive symptoms (25.0% vs. 19.4%, p=0.45) were more frequent in PD patients with GD-associated mutations, albeit not statistically significant. In conclusion, genetic variants in the GBA gene have a high frequency in PD patients and are significantly associated with early age of onset. Positive family history for PD and worse neuropsychological symptoms were more frequent in the presence of GBA variants. Elucidating the link between GD and PD may yield new insights into the pathophysiology of PD and identify new targets for treatment.

Session 7

(29)

UNBIASED APPROACH REVEALS HIGHER RISK FOR PD IN CARRIERS OF SEVERE VS. MILD GBA MUTATIONS

David Arkadir*[1], Tama Dinur[2], Strphen Mullin[3], Atul Mehta[4], Hagit Baris-Feldman[5] Ari Zimran[2].

[1]Department of Neurology, Hadassah Medical Center and the Hebrew University, Jerusalem Israel

[2]Gaucher Clinic, Shaare Zedek Medical Center nad Hebrew University, Jerusalem, Israel

[3]Department of clinical Neuroscience, UCL Institute on Neurology, London, UK

[4]LSD Unit Royal Free Hospital and UCL School of Medicine, London, UK.

[5]The Genetics Institute, Rambam Health Care Campus, Haifa, Israel, and Rappaport School of Medicine, Technion, Haifa, Israel

* Corresponding author: arkadir@hadassah.org.il

Increased incidence of Parkinson's disease (PD) exists among carriers of mutations in the Acid beta-glucocerebrosidase (GBA) gene. Large studies detected correlation between different GBA mutations and the risk for PD. These studies concluded that severe GBA mutations impose a higher risk for PD compared to mild mutations but were based on the measured frequencies of the different GBA mutations in the healthy, ethnically-matched, population. This methodology is prone to biases. To test for the validity of this finding, while avoiding these biases, we tested the genetic status of patients with PD with *a priori* equal chances of carrying mild (N370S) or severe GBA mutation.

Participants were traced among parents of compound heterozygous Gaucher patients. To confirm the diagnosis of PD, a movement disorders specialist examined all living individuals (N=98, males 76, age at onset 56-71 years, Hoehn & Yahr scale 2-5) or reviewed the medical record of already deceased ones (N=4, 4 males, age at onset 67-69 years). Genotyping the GBA gene of PD patients was directed by the known GBA mutations of their descendents. The genetic status of deceased patients was extrapolated based on the combined GBA mutation status of their living spouse and descendents.

In agreement with our hypothesis, 9/12 of the obligatory carriers with PD had a severe mutation (P=0.07, binomial test). This finding is in agreement with previous reports who used the measured mutation frequencies in a population of healthy controls to calculate the correlation between mutation status and the risk for PD.

Session 8

(30)

MEASUREMENT OF BONE MARROW FAT FRACTION WITH GRADIENT-ECHO MR IMAGING IN GAUCHER DISEASE PATIENTS; A PRACTICAL ALTERNATIVE TO DIXON QUANTITATIVE CHEMICAL SHIFT IMAGING

M. Regenboog[1-2*], K.M. Beekman[1], C.E.M. Hollak[2], M. Maas[1] E.M. Akkerman[1],

[1] Department of Radiology, [2] Department of Internal Medicine, division of Endocrinology & Metabolism, Academic Medical Center Amsterdam, The Netherlands.

[*] presenting author: m.regenboog@amc.uva.nl

Introduction. Quantification of bone marrow involvement in Gaucher disease (GD) is of importance for initial assessment and follow-up of patients. Dixon quantitative chemical shift imaging (QCSI) to measure bone marrow fat fractions (Ff) has been applied at the AMC and validated as a surrogate marker for bone marrow involvement with Gaucher cells. A disadvantage of this technique is the limited availability of the requisites to perform these scans in other centers. One of the alternatives to measure bone marrow Ff is the use of gradient-echo (GRE) MR imaging. We examined this technique in a subset of GD patients and report our initial experiences.

Methods. In 11 adult, treated, GD1 patients, Ff measurements were performed using two imaging protocols, GRE and Dixon QCSI (spin-echo sequence), on the same day and same 1.5 Tesla MRI-scanner. Regions-of-interest (ROIs) were drawn in the lumbar vertebrae 3-5. In this ROIs the fat fraction as percentage of total signal could be obtained after averaging the pixels from the image. The average of the Ff in L3-L5 was reported for both methods. Comparison of GRE to Dixon QCSI was made by creating a Bland-Altman plot and calculate the intraclass correlation coefficient (ICC).

Results. Ff was mostly in the normal range, as expected for well treated patients. Values ranged from 30.9 to 65.3 (mean 46.4) using QCSI, and 25.7 to 63.0 (mean 42.0) by GRE imaging. The mean difference in Ff between GRE and QCSI was 4.5 (SD 6.0, limits of agreement -7.3-16.3). Intraclass correlation coefficient for absolute agreement between the two methods was 0.80 (95% CI 0.32-0.95, $p<0.001$).

Conclusion. Bone marrow Ffs measured with GRE MRI show a high absolute agreement when compared to Dixon QCSI Ffs. This indicates that GRE might serve as a practical alternative to Dixon QCSI. Further research is needed to confirm this observation, specifically in the lower range of Ff. In addition, the relationship to disease parameters and other semi-quantitative bone marrow imaging including the bone marrow burden score should be studied.

Session 8

(31)

OSTEOCYTE ALTERATIONS CONTRIBUTE TO BONE PATHOLOGY IN GAUCHER DISEASE

Mucci Juan Marcos[1*], Ferreyra Malena[1], Crivaro Andrea[1], Bondar Constanza[1], Ormazabal Maximiliano[1], Delpino Victoria[2], Rozenfeld Paula[1]

1 Instituto de Estudios Inmunológicos y Fisiopatológicos, CONICET-UNLP, La Plata, Argentina
2 Instituto de Inmunología, Genética y Metabolismo, CONICET, CABA, Argentina

Corresponding author: juanmarcosmucci@gmail.com

The aim of our work was to evaluate the involvement of osteocytes in the bone pathology of Gaucher disease (GD). The study was performed with an *in vitro* model using the MLO-Y4 osteocyte cell line treated with CBE at different time points. Osteoclastogenic potential of conditioned media (CM) from CBE treated ostecytes was evaluated by osteoclast differentiation assays, and RANKL levels were evaluated by immunofluorescence. Connexin-43 expression was assessed by qPCR and apoptosis was studied by Annexin-V and TUNEL stainings. To study the mechanism on osteoclast differentiation, the apoptotic body fraction of the CM and its supernatant were obtained by centrifugation. Both fractions were used in osteoclastogenesis assays with or without OPG. The CM from CBE treated osteocytes induced higher levels of osteoclast differentiation ($p<0,01$) compared to control CM and higher surface RANKL levels were observed in treated cells ($p<0,05$). Connexin-43 expression diminished with CBE treatment ($p<0,01$) and osteocyte apoptosis was increased ($p<0,05$). The induction of osteoclast differentiation by apoptotic bodies and supernatant was higher in CBE treated CM in both fractions ($p<0,01$), and treatment with OPG reduced osteoclast levels in both cases ($p<0,001$). In conclusion, we showed a potential role of osteocytes in bone pathology of GD by induction of osteoclast differentiation. This effect could be related to a higher apoptotic state in osteocytes as well as the RANKL induction pathway.

Session 8

(32)

EFFECT OF TWO DIFFERENT THERAPEUTIC INTERVENTIONS: SRT IN COMPARISON TO ERT ON IMMUNE ASPECTS AND BONE INVOLVEMENT IN GAUCHER DISEASE

Renuka Pudi Limgala*, Miriam Arnone & Ozlem Goker-Alpan
Lysosomal and Rare Disorders Research and Treatment Center, Fairfax, VA 22030 USA
O & O Alpan, LLC, Fairfax, VA 22030 USA

*Corresponding author: rlimgala@ldrtc.org

Macrophage-directed enzyme replacement therapy (ERT) has been the most accepted form of treatment for Gaucher disease (GD), however there are still unmet needs in treating all aspects of the disease. As an alternative to ERT, substrate reduction therapy (SRT) was developed using glucosylceramide synthase inhibitors. In current IRB approved study (NCT02605603), we closely monitor and compare the effects of ERT vs SRT particularly the immunological aspects as well as bone remodeling. For this, we performed in-depth immunophenotyping of patients being administered ERT (n=8) or Eliglustat (n=10, 8 patients earlier treated with ERT and 2 patients untreated before starting SRT). To investigate if the extent of bone remodeling could be predicted or explained using markers from peripheral blood, expression of RANK/RANKL pathway components were assessed on relevant immune cell types. In subjects continuing ERT, no significant changes were observed in surface expression of RANK or RANKL on the immune subsets over 3-6 months. However, in 4 out of the10 subjects treated with SRT, there is a significant increase ($p<0.05$) in the expression of RANK on T cells and monocytes and increase in RANKL on Tcell subsets. The results from individual GD patients were compared in parallel to the secreted forms of RANKL and OPG as well as their disease severity and bone density findings. Insights from the study highlight personalized differences between subjects and possible use of components of RANK pathway as markers for bone disease progression. This study is funded by a research grant from Sanofi Genzyme (GZ-2014-11295).

Session 8

(33)

RISK FACTORS FOR FRACTURES IN IMIGLUCERASE-TREATED GAUCHER DISEASE TYPE 1 PATIENTS IN THE ICGG GAUCHER REGISTRY

Patrick Deegan[1], Julie L. Batista[2], Stephan Vom Dahl[3*]

[1]*Lysosomal Disorders Unit, Department of Medicine, Addenbrooke's Hospital, Cambridge, United Kingdom*

[2]*Sanofi Genzyme, Cambridge, MA, USA*

[3]Department of Gastroenterology, Hepatology and Infectious Diseases, University of Duesseldorf

*Corresponding author: stephan.vomdahl@med.uni-duesseldorf.de

Background: Skeletal complications, particularly fractures, can be disabling consequences of Gaucher disease type 1 (GD1) and successful management of patients requires understanding the associated clinical risk factors. Aims and methods: Demographics and risk factors for first fracture were evaluated in imiglucerase-treated GD1 patients of all ages with data sets from the International Collaborative Gaucher Group (ICGG) Gaucher Registry. Patients were excluded if fractures occurred prior to treatment initiation or after imiglucerase discontinuation, or if splenectomy status, date of diagnosis and of treatment initiation were missing. Selected candidate predictors (sex, year of diagnosis, age at treatment initiation, time between diagnosis and treatment initiation, bone pain, bone crises, splenectomy status, chitotriosidase level) were analyzed using multivariate logistic regression with forward selection. Results: Patients (N=3554) were 47% male; 26% had splenectomy; median age of diagnosis was 15.3 years; 293 patients (8%) experienced a fracture during treatment. Significant predictors of total fracture risk were: age at treatment initiation (per 5yr increase; OR=1.045; 95% CI: 1.008,1.083; $p<0.05$); time between diagnosis and treatment initiation (per 5-yr increase; OR=1.092; 95% CI:1.033,1.154; $p<0.01$); bone pain (yes vs. no; OR=1.902; 95% CI: 1.476, 2.452; $p<0.0001$); splenectomy (yes vs. no; OR=1.628; 95% CI: 1.221, 2.171; $p<0.001$); chitotriosidase level (>75th vs. ≤75th percentile; OR=1.510; 95% CI: 1.009, 2.261; $p<0.05$). These key predictors were similar for risk of spinal fracture (n=103 events). Conclusion: Shorter duration between diagnosis and treatment, earlier age at treatment initiation, no splenectomy, no bone pain, and lower chitotriosidase levels may predict a lower fracture risk in imiglucerase-treated GD1 patients. (NCT00358150, sponsored by Sanofi Genzyme)

12th EWGGD ZARAGOZA

The European Working Group on Gaucher Disease

Poster Presentations

No	LIST OF POSTER PRESENTATIONS
P1	***UPR ACTIVATION AND CHOP INDUCTION OF GBA TRANSCRIPTION IN GD DERIVED CELLS*** *Hila Braunstein1*, Gali Maor1*, Mirella Filocamo2 Ari Zimran3, and Mia Horowitz1#,* *1Department of Cell Research and Immunology, Tel Aviv University, 69978, Israel, 2Centro di Diagnostica Genetica e Biochimica delle Malattie Metaboliche, IRCCS G. Gaslini, Genova, Italy, 3Department of Internal Medicine, Shaare Zedek Medical Center and Hebrew University-Hadassah Medical School, Jerusalem, Israel.*
P2	***ANALYSIS OF HUMAN GLUCOCEREBROSIDASE EXPRESSED IN Pichia pastoris*** *Marcela de Oliveira Vitarelli1,2*, Eliane Namie Miyaji2 , Paulo Lee Ho2* *1 Chemistry Institute, University of São Paulo, São Paulo, Brazil 2 Biotechnology Center, Butantan Institute, São Paulo, Brazil*
P3	***DECREASES IN GLUCOSYLSPHINGOSINE (LYSO-GL-1) CORRELATE WITH CLINICAL RESPONSE IN ADULTS WITH GAUCHER DISEASE TYPE 1 TREATED WITH ORAL ELIGLUSTAT*** *M. Judith Peterschmitt1*, Kate Zhang1, Lin Liu1, Ames Macdougall2, Gerald F. Cox1* *1Sanofi Genzyme, Cambridge, MA, USA; 2Prometrika, LLC, Cambridge, MA, USA*
P4	***CROSS-SECTIONAL STUDY OF GLUCOSYLSPHINGOSINE (LYSO GB1) IN AN UNSELECTED COHORT OF ADULT PATIENTS WITH GAUCHER DISEASE REVEALS LOWEST LEVELS IN PATIENTS TREATED WITH VELAGLUCERASE ALFA*** *Dinur_T1, Chicco_G1, Elstein_D1, Bdolach-Avram_T1, Cozma_C3, Eichler_S3, Rolfs_A2/3, Zimran_A1*.* *1 Gaucher Clinic, Shaare Zedek Medical Center, Jerusalem, Israel* *2 Albrecht-Kossel-Institute for Neuroregeneration, University of Rostock, and CentoGene, Rostock, Germany* *3 Centogene AG, Institute for Rare Diseases, Rostock, Germany*
P5	***THE UTILITY OF GLUCOSYLSPHINGOSINE AS A PATHOPHYSIOLOGICALLY RELEVANT BIOMARKER OF GAUCHER DISEASE*** *Vagishwari Murugesan1*, Wei-Lien Chuang2, Jun Liu1, Andrew Lischuk3, Katherine Kacena4, Haiqun Lin5, Gregory M Pastores6, Ruhua Yang1, Joan Keutzer2, Kate Zhang2, Pramod K Mistry7* *1 Department of Internal Medicine, Yale University School of Medicine, New Haven, Connecticut, USA 2Genzyme Corporation, Framingham, Massachusetts, USA 3 Department of Radiology, Yale University School of Medicine, New Haven, Connecticut, USA 4 Kacena Consulting, Natick, Massachusetts, USA 5 Department of Biostatistics, Yale School of Public Health, USA 6 Department of Neurology, New York University School of Medicine, New York, USA 7 Department of Internal Medicine & Pediatrics, Yale University School of Medicine, New Haven, Connecticut, USA*
P6	***GLUCOSYLSPHINGOSINE (LYSO-GB1) CONCENTRATION IN THE BLOOD OF GAUCHER PATIENTS REFLECTS THE SEVERITY OF GBA MUTATIONS*** *Claudia Cozma1, Gabriela Oprea1, Guido Kramp1, Sabrina Eichler 1, Anne Giese2, Jan Lukas2, Tobias Bötcher2, Arndt Rolfs 1,2* *1 Centogene AG; Rostock, Germany. 2 Albrecht-Kossel-Institute, University of Rostock, Rostock, Germany*
P7	***QUANTIFICATION OF GLUCOSYLSHINGOSINE (LYSO-GB1) FOR THE DIAGNOSIS AND MONITORING OF GAUCHER DISEASE -METHODOLOGY AND VALIDATION*** *Claudia Cozma1, Guido Kramp1, Sabrina Eichler 1, Arndt Rolfs 1,2* *1 Centogene AG; Rostock, Germany. 2 Albrecht-Kossel-Institute, University of Rostock, Rostock, Germany*
P8	***ROLE OF RED BLOOD CELL ON THE PATHOPHYSIOLOGY OF GAUCHER DISEASE AND EFFECT OF THE ENZYME REPLACEMENT THERAPY*** *Franco M1-4, Reihani N1-4, Marin M1-4, Collec E1-4, Billette de Villemeur T5,6, Rose C6,7, Mignot C5,6, Colin Y1-4, Belmatoug N6,8 & Le Van Kim C1-4.* *1Inserm U1134, Paris, France; 2Université Paris Diderot, Sorbonne Paris Cité, UMR_S 1134, Paris, France; 3Institut National de la Transfusion Sanguine, Paris, France; 4Laboratoire d'excellence GR-Ex, Paris, France; 5Service de Neuropédiatrie, Hôpital Trousseau, Assistance Publique–Hôpitaux de Paris and Université Pierre et Marie Curie, Paris, France; 6Centre de Référence des Maladies Lysosomales, Clichy, France; 7Service d'Hématologie et de cardiologie Hôpital Saint Vincent de Paul, Université Catholique de Lille, Lille; 8Service de Médecine Interne, Hôpital Beaujon, Clichy, France;*
P9	***IRON IMAGING AND PARAMETERS OF IRON METABOLISM IN ADULT TYPE 1 GAUCHER DISEASE*** *Regenboog M1-2*, Bohte AE1, Akkerman EM1, Stoker J1, Hollak CEM2* *1Department of Radiology, 2Department of Internal Medicine, division of Endocrinology & Metabolism. Academic Medical Center Amsterdam, The Netherlands.*
P10	***SERUM FERRITIN AND LIVER FEATURES IN A COHORT OF 15 PATIENTS AFFECTED BY GAUCHER DISEASE*** Miriam Rigoldi1, Lorenza Borin2, Raffaella Mariani1, Irene Pelloni1, Federico Greni3, Cristina Arosio4, Alb*erto Piperrno1,2,4* *1Center for rare disease, Internal Medicine 2, ASST- Monza S.Gerardo Hospital, Italy 2Hematology Unit, ASST-Monza S.Gerardo Hospital, Italy . 3University of Milano-Bicocca, School of Medicine and Surgery – Monza, Italy* *4Consortium fo*r Human Molecular Genetics – Monza, Italy
P11	***GAUCHER DISEASE AND DEFICIENCY OF INTESTINAL DISACHARIDASES*** *Medrano-Engay B1, 2, Irún MP1, 2, 3, Andrade-Campos M1, 2, 3, Pocoví M2, 4, Giraldo P1, 2, 3, 5* *1Unidad de Investigación Traslacional. Hospital Universitario Miguel Servet. Zaragoza. España. 2Instituto Investigación Sanitaria Aragón. Zaragoza. España. 3Centro de Investigación Biomédica en Red de Enfermedades Raras (CIBERER), ISCIII. Zaragoza. España. 4Departmento de Bioquímica y Biología Molecular y Celular. Universidad de Zaragoza. España. 5Fundación para el Estudio y Terapéutica de la Enfermedad de Gaucher. Zaragoza, España.*

P12 ***PERIPHERAL BLOOD LYMPHOCYTE IMMUNOPHENOTYPE ABNORMALITIES IN PATIENTS WITH TYPE I GAUCHER DISEASE***

*Eugenia Verigou1, Christos Sotiropoulos1, Kalliopi Zafeiropoulou1, Theodore Marinakis2, Constantina Repa3, Panagiotis Tsaftaridis4, Vasileios Lazaris1, Alexandra Kourakli1, Argiris Symeonidis1**
1Hematology Division, Dept of Int.Medicine, University of Patras Medical School, Patras, Greece. 2Dept of Hematology, "G.Gennimatas" General Hospital, Athens, Greece. 3Thalassemia Center, Laikon General Hospital, Athens, Greece. 4Dept of Haematology, Laikon General Hospital, National and Kapodistrian University of Athens, Greece.

P13 ***SCREENING FOR DELETIONS/DUPLICATIONS IN THE GBA1 GENE USING MULTIPLEX LIGATION-DEPENDENT PROBE AMPLIFICATION METHOD IN GAUCHER PATIENTS***

Marina Siebert1,2,3, Suelen P. Basgalupp2,4, Filippo P. Vairo2,5, Ida V. D. Schwartz2,4,5,6.
1Postgraduate Program in Sciences of Gastroenterology and Hepatology, Faculty of Medical Sciences, Universidade Federal do Rio Grande do Sul, Porto Alegre, RS, 90035-003, Brazil; 2Basic Research and Advanced Investigations in Neurosciences (BRAIN) Laboratory, Experimental Research Center. Hospital de Clínicas de Porto Alegre (HCPA), Porto Alegre, RS, 90035-903 Brazil; 3Unit of Molecular and Protein Analyses, Experimental Research Center, Hospital de Clínicas de Porto Alegre (HCPA), Porto Alegre, RS, 90035-903 Brazil; 4Postgraduate Program in Medical Sciences, Faculty of Medical Sciences, Universidade Federal do Rio Grande do Sul, Porto Alegre, RS, 90035-003, Brazil; 5Medical Genetics Service, Hospital de Clínicas de Porto Alegre (HCPA), Porto Alegre, RS, 90035-903 Brazil; 6Department of Genetics, Universidade Federal do Rio Grande do Sul, Porto Alegre, Brazil

P14 ***SCREENING FOR LYSOSOMAL STORAGE DISEASES IN FLANDERS – BELGIUM: PILOT STUDY AND DIAGNOSTIC OPPORTUNITIES***

Eyskens François, Devos Sylvie*
PCMA vzw-Metabolic Lab, University of Antwerp, Antwerp, Belgium

P15 ***ANALYSIS OF 8 MARKERS ASSOCIATED TO N370S AND L444P ALLELES ON MEXICAN PATIENTS WITH GAUCHER DISEASE***

Garcia-Ortiz, Jose Elias1, Magaña-Torres María Teresa, da Silva-Jose Thiago Donizete2.*
1División de Genética Centro de Investigación Biomédica de Occidente, IMSS. Guadalajara, México.
2Doctorado en Genética Humana, CUCS-Universidad de Guadalajara. Guadalajara, México.

P16 ***GAUCHER DISEASE IN GREECE.LABORATORY INVESTIGATIONS***

E.Dimitriou1, M.Moraitou1, L.Vilageliu2, D.Grinberg2, J. Serra-Vinardell2, M. Cozar2, I.Mavridou1 and H.Michelakakis1*
1. Dept Enzymology and Cellular Function, Institute of Child Health, Athens ,Greece 2. Dept. of Genetics, University of Barcelona, CIBERER, IBUB, Barcelona, Spain

P17 ***CYP2D6 GENOTYPE AND METABOLIZER STATUS IN SPANISH PATIENTS WITH TYPE 1 GAUCHER DISEASE***

Alfonso P1,2,3, López de Frutos L1,3, Andrade-Campos M1,2,3, Giraldo P1,2,3. 1Instituto de Investigación Sanitaria Aragón (IIS Aragón), Zaragoza, Spain, 2Centro de Investigación Biomédica en Red de Enfermedades Raras (CIBERER), Zaragoza, Spain, 3Fundación Española para el Estudio y la Terapéutica de la Enfermedad de Gaucher y otras Lisosomales (FEETEG), Zaragoza, Spain

P18 ***A COMPARISON OF THE CHINESE AND GLOBAL POPULATIONS FROM THE INTERNATIONAL COLLABORATIVE GAUCHER GROUP (ICGG) GAUCHER REGISTRY***

*Duan, Yanlong1; Taylor, John S.2**; Luo, Xuequn3; Gu, Longjun4; Gu, Xufan5; Meng, Yan6; Zhang, Huiwen5; Huang, He7; Zheng, Weiyan7; Zhang, Yonghong1**
1Beijing Children's Hospital, Beijing, China; 2 Sanofi Genzyme, Cambridge, MA, USA; 3 First Affiliated Hospital of Sun Yat-Sen University, Guangzhou, China; 4 Shanghai Children's Medical Center, Shanghai, China; 5 Shanghai Xinhua Hospital, Shanghai, China; 6 Chínese PLA General Hospital, Beijing, China; 7 Zheng Zhejiang University Affiliated 1st Hospital, Hangzhou, China

P19 ***TYPE 1 GAUCHER DISEASE: HOMOZYGOSITY FOR p.R496H MUTATION***

P. Irún1,2 , M Andrade-Campos1,2 , E Mingot3 , P. Giraldo1,2,4
1Centro de Investigación Biomédica en Red de Enfermedades Raras (CIBERER), Instituto de Salud Carlos III, Zaragoza. Spain. 2 Instituto Investigación Sanitaria Aragon (IIS Aragón). Zaragoza. Spain. 3Haematology Department. Carlos Haya University Hospital. Malaga. 4Spanish Foundation of Gaucher Disease and other Lysosomal Disorders (FEETEG). Zaragoza. Spain.

P20 ***WANDERING SPLEEN IN 2 GAUCHER PATIENTS ON ENZYME REPLACEMENT THERAPY: DIAGNOSIS AND MANAGEMENT***

Lau Heather1, Liang Howard2, Elstein Deborah3, Alberton Joseph4, Zimran Ari3*
1 Department of Neurology, NYU Langone Medical Center, New York, NY USA 2 Department of Surgery, NYU Langone Medical Center, New York, NY USA 3 Gaucher Clinic, Shaare Zedek Medical Center, Jerusalem Israel
4 Department of Surgery, Shaare Zedek Medical Center, Jerusalem Israel BioTherapeutics, Carmiel, Israel

P21 ***MUTATIONS IN THE GBA1 GENE IN SPANISH POPULATION WITH PARKINSON DISEASE AND PLASMA miRNAs***

Gervas-Arruga J1,2,3, Alfonso P1,2,3, Garcia-Rodriguez B4 , Capablo JL5, Pocovi M6, Giraldo P1,2,3*
1 Centro de Investigación Biomédica en Red de Enfermedades Raras (CIBERER), Zaragoza, Spain 2 Instituto de Investigación Sanitaria Aragón (IISAragón), Zaragoza, Spain 3 Fundación Española para el Estudio y la Terapéutica de la Enfermedad de Gaucher y otras Lisosomales (FEETEG), Zaragoza, Spain 4 Biochemistry department, Miguel Servet University Hospital, Zaragoza, Spain 5 Neurology department, Miguel Servet University Hospital, Zaragoza, Spain 6 Biochemistry; Cellular and Molecular Biology department, Zaragoza University, Zaragoza, Spain

P22 ***GENETIC STUDIES ON A PORTUGUESE PARKINSON DISEASE PATIENT COHORT***

Gabriel Miltenberger-Miltenyi, Leonor Guedes, Tiago Soares, Marcos Gomes, Joaquim Ferreira, Tiago Outeiro.
Instituto de Medicina Molecular Faculdade de Medicina Universidade de Lisboa

P23 ***EXAMINING THE RISK OF PARKINSONISM IN SIX SIBLING PAIRS WITH GAUCHER DISEASE***

Grisel Lopez1, MD, Gianina Monestime1, BS, MPH, Catherine Groden1, RN, NP, Edythe Wiggs1, PhD, Nahid Tayebi1, PhD, Ari Zimran2, MD, and Ellen Sidransky1, MD*
1Medical Genetics Branch, NHGRI, NIH, Bethesda MD, USA 2Gaucher Clinic Jerusalem, Israel

P24 ***GAUCHER DIEASE TYPE 3 IN SPAIN. OUTCOMES AND CHARACTERIZATION***

Marcio M Andrade-Camposa, Javier Gervasb, I. Garciac, O. Salamerod, P. Martinez-Odriozolae, JA Mendezf, F. Garcia-Bragadog, C. Calvoh, E. Luñoi, I. Sancho-Valk, H. Canol, J, JM Hernandez-Rivasm, D. Lorenzon, M. Lopez-Duplao, V. Callaop, P. Giraldoa
aIIS Aragon. CIBER enfermedades Raras, Zaragoza, Spain; bIACS, Zaragoza; cMiguel Servet University Hospital, Zaragoza; dVall d'Hebron University Hospital, Barcelona; eBasurto University Hospital, Bilbao;fOurense Hospital, Ourense; gJosep Trueta Hospital, Girona;hSan Jorge Hospital, Huesca; iCabueñes Hospital, Asturias;kAlcañiz Hospital, Alcañiz; lArcos del Mar Menor University Hospital, Murcia; mSalamanca University Hospital, Salamanca; nComplejo Hospitalario, Burgos; oJoan XXIII Hospital, Tarragona; pArnau de Vilanova Hospital, Lleida, Spain

P25 ***NEUROLOGICAL MANIFESTATIONS AND COURSE OF TYPE 1 AND TYPE 3 (NORRBOTTNIAN) GAUCHER DISEASE IN SWEDEN***

Machaczka M1,2, Kämpe Björkvall C3, Paucar Arce M4, Myhr-Eriksson K3, Forsgren L5, Svenningsson P4*
1 Hematology Center, Karolinska University Hospital Huddinge, Stockholm, Sweden 2 Medical Faculty, University of Rzeszow, Rzeszow, Poland 3 Department of Medicine, Sunderby Regional Hospital of Norrbotten County, Luleå, Sweden 4 Department of Clinical Neuroscience, Center for Molecular Medicine, Karolinska Institutet, Stockholm, Sweden 5 Department of Pharmacology and Clinical Neuroscience, Umeå University, Umeå, Sweden.

P26 ***A SEVERITY SCORING TOOL FOR NON NEUROLOGICAL MULTISYSTEM INVOLVEMENT OF TYPE 3 GAUCHER DISEASE: AN IMPORTANT STEP TO MOVE FORWARD***

Magy Abdelwahab, Neal Weinreb
1 Department of Pediatric Hematology, Cairo University Pediatric Hospital, Cairo, Egypt 2 University Research Foundation for Lysosomal Storage Diseases, USA

P27 ***PHENOTYPIC VARIABILITY IN GAUCHER TYPE 3***

Eugen Mengel1, Jörg Reinke1, Tatiana Bremova3, Juliane Arndt1, Silvia Naumann1, Susanne Pitz2, Gundula Staatz2 and Julia Hennermann.1
1 JGU Mainz UMC, Villa metabolica Mainz, Germany 2 JGU Mainz UMC, Neuroophtalmology and Radiology department Mainz, Germany 3 German Center for Vertigo and Balance Disorders, Grosshadern Campus, University Hospital Munich, Germany

P28 ***LIVER CIRRHOSIS AND MESENTERIC LYMPHADENOPATHY: AN ASSOCIATION OR AN INDICATION OF DISEASE SEVERITY IN 3 GAUCHER DISEASE EGYPTIAN CHILDREN***

Magy Abdelwahab1, Neal Weinreb2
1 Department of Pediatric Hematology,Cairo University Pediatric Hospital, Cairo, Egypt 2 University Research Foundation for Lysosomal Storage Diseases, USA

P29 ***INCREASED RISK OF HEPATOCELLULAR CARCINOMA (HCC) IN GAUCHER DISEASE: THREE CASE REPORTS: BIOLOGICAL COURSE AND IMPACT ON MONITORING***

*David Santosa1, Eugen Mengel2, Martin Merkel3, Ute Gebert4, Dieter Häussinger1, Stephan vom Dahl1**
1Department of Gastroenterology, Hepatology and Infectious Diseases, Heinrich-Heine-University, Düsseldorf, Germany 2Villa Metabolica, University Children's Hospital, Mainz, Germany 3Endocrinologicum, Amedes Integrated Diagnostics, Hannover, Germany 4Department of Internal Medicine, St. Franziskus-Hospital, Cologne, Germany

P30 ***SPLENIC ARTERY ANEURYSMS, A RARE COMPLICATION OF GAUCHER DISEASE: REPORT OF FOUR CASES***

C Serratrice1, T Cox2, 3, E Morris3, K Benyousfi4, O Monnet5, A Sibert6, W Allaham6, N Belmatoug4*
1. Department of Internal Medicine and Rehabilitation, University Hospital of Geneva, Geneva, Switzerland 2. Department of Medicine, University of Cambridge, UK 3. Lysosomal Disorders Unit, Addenbrooke's Hospital, Cambridge, UK 4. Reference Centre of Lysosomal Diseases, University Hospital Beaujon, Clichy, France 5. Department of Radiology, Saint Joseph Foundation, Marseille, France 6. Department of Radiology, University Hospital Beaujon, Clichy, France

P31 ***LYMPADENOPATHY IN GAUCHER DISEASE IS ASSOCIATED WITH TINGIBLE BODY MACROPHAGES OF M2 PHENOTYPE***

*Ozlem Goker-Alpan, MD1, 2 *, Renuka Pudi Limgala, PhD1,2, Erk Changsila,BS1, Chadima Iaonou, BS1,2, Ariel Badger, MS1, and Margarita Ivanova,PhD1,2,*
1Department of Translational Medicine, Lysosomal and Rare Disorders Research and Treatment Center, 11212 Waples Mill Rd. #103, Fairfax, VA, 22030 USA
2 O&O Alpan, LLC, 11212 Waples Mill Rd. #100, Fairfax, VA, 22030 USA

P32 ***OPTIMAL RESPONSE IN GAUCHER DISEASE IN REPLACEMENT THERAPY AND CONCOMITANT CHRONIC MYELOID LEUKEMIA.***

MS Noya2, P Irun1, M Andrade1, MF Lopez-Fernandez2, P Giraldo1
1.Traslational Research Unit IIS Aragon CIBER enfermedades raras CIBERER, Zaragoza, Spain. 2. Department Hematology, CHUAC A Coruña, Spain.

P33 ***MYELODYSPLASTIC SYNDROMES IN TYPE 1 GAUCHER DISEASE :DIAGNOSTIC AND TREATMENT CHALLENGES***

Hanna Rosenbaum
Hematology day care unit, the center for consultant medicine, Clalit services, Nazareth, Israel.

P34 ***OCCURENCE OF NEOPLASTIC DISEASES AND OF PRENEOPLASTIC CONDITIONS, AMONG GREEK PATIENTS WITH GAUCHER DISEASE***

Constantina Repa, Theodore Marinakis, Alexandra Kourakli, Dimitrios Zafeiriou, Panagiotis Tsaftaridis, Helen Michelakakis,, Veroniki Konminaka, Vassiliki Pappa, Paraskevi Roussou, Efthimia Vlachaki, Maria Kotsopoulou, Alexandros Makis, Evdokia Mandala, Ersi Voskaridou, Nikolaos Anagnostopoulos, Argiris Symeonidis for the Hellenic (Greek) National Gaucher Disease Study Group.*

P35 ***STUDY OF COMORBIDITIES IN A SPANISH COHORT OF GAUCHER DISEASE TYPE 1 PATIENTS***

J. Pérez-López1, V. Giner Galvañ2, MA. Torralba Cabeza3, RJ Núñez4, M. Moltó Abad1, Inmaculada Roig5, L. Vicente6, A. Luaña Galan7, E. Cabezudo8, X. Solanich9, B. Roig Espert10, E. Patera11, ML. Lozano Almela12, R. Hurtado García13, M. Reyes14, N. Revilla12, S. Novelli15, J. Esteve16.
1 Inborn Errors of Metabolism in Adults. Unit of Rare Diseases, H. Vall d'Hebron (Barcelona) 2 Service of Internal Medicine. H. Virgen de los Lirios (Alcoy)3 Service of Internal Medicine. H. Lozano Blesa (Zaragoza) 4 Service of Hematology. H. Virgen del Rocío (Sevilla) 5 Service of Hematology. Consorci Hospitalari Parc Taulí (Sabadell) 6 Service of Hematology. H. de Terrassa (Terrassa) 7 Service of Hematology. H. Arnau de Vilanova (Lleida)8 Service of Hematology. H. Sant Joan de Deu (Manresa) 9 Service of Internal Medicine. H. de Bellvitge (Hospitalet de Llobregat) 10 Service of Internal Medicine. H. de Manises (Valencia) 11 Service of Internal Medicine. H. Sant Jaume (Calella)12 Service of Hematology. H. Morales Meseguer (Murcia) 13 Service of Internal Medicine. H. Vega Baja (Orihuela) 14 Service of Hematology. H. Lozano Blesa (Zaragoza)15 Service of Hematology. H. Santa Creu i Sant Pau Pau (Barcelona) 16 Service of Hematology. H. Clínic (Barcelona)

P36 ***THE APPEARANCE OF INTRAOCULAR LESIONS IN GAUCHER DISEASE TYPE 3 DESPITE LONG-TERM GLUCOCEREBROSIDASE REPLACEMENT THERAPY***

Sawicka-Gutaj N1, Machaczka M2,3, Kulinska-Niedziela I4, Bernardczyk-Meller J5, Gutaj P1, Sowinski J1, Ruchala M1*
1 Department of Endocrinology, Metabolism and Internal Medicine, Poznan University of Medical Sciences, Poznan, Poland 2 Hematology Center, Karolinska University Hospital Huddinge, Stockholm, Sweden 3 Medical Faculty, University of Rzeszow, Rzeszow, Poland 4 Heliodor Swiecicki Clinical Hospital, Ophthalmology Outpatient Clinic, Poznan University of Medical Sciences, Poznan, Poland 5 Department of Ophthalmology, NZOZ "OCU SERVICE", Poznan, Poland

P37 ***IMIGLUCERASE TREATMENT ASSOCIATED WITH REDUCTION OF BONE CLAIMS IN GAUCHER PATIENTS: ANALYSIS OF U.S. CLAIMS DATA***

Luba Nalysnyk1, Martin Selzer1, Alaa Hamed1, Neal Weinreb2 1Sanofi Genzyme, Cambridge, MA*
2University Research Foundation for Lysosomal Storage Diseases, Coral Spring, FL

P38 ***THREE-DIMENSIONAL PRINTING OF BONE LESSIONS IN GAUCHER DISEASE***

A Lacleriga1, Luis Sarria2, Alfaro-Santafé JV3, Mercedes Roca4, Pilar Giraldo5
1Traumatology Department. Miguel Servet University Hospital. Zaragoza 2 Radiodiagnostic Department. Miguel Servet University Hospital. Zaragoza 3 Podoactiva. Zaragoza 4 Radiological Centre. Zaragoza 5 Traslational Research Unit. IIS Aragón. CIBER enfermedades raras CIBERER. Zaragoza

P39 ***THE IMPACT OF CHITOTRIOSIDASE ON THE OCCURRENCE OF BONE EVENTS IN GAUCHER DISEASE TREATED USING ENZYME REPLACEMENT THERAPY: THE CONTRIBUTION OF JOINT MODELS***

Marie Vigan1,2, Nadia Belmatoug3, Marc G Berger4,5,6, France Mentré1,2, Jérôme Stirnemann7 and all members of the Gaucher Disease Treatment Evaluation Committee
1INSERM, IAME, UMR 1137, F-75018 Paris, France 2 Université Paris Diderot, IAME, UMR 1137, Sorbonne Paris Cité, F-75018 Paris, France 3Médecine Interne et CRML, Hôpital Beaujon, AP-HP, 100 Bd Général Leclerc, 92110 Clichy, France 4Hématologie Biologique, CHU Estaing, 1 place Lucie Aubrac, 63003 Clermont-Ferrand Cedex 1, France 5CRB Auvergne, CHU Estaing, 1 place Lucie Aubrac, 63003 Clermont-Ferrand Cedex 1, France 6Hématologie Clinique, CHU Estaing, 1 place Lucie Aubrac, 63003 Clermont-Ferrand Cedex 1, France 7Service de Médecine Interne Générale, Hôpitaux Universitaires de Genève, Rue Gabrielle-Perret-Gentil 4, CH-1211 Genève 14, Suisse

P40 ***EVALUTAION OF BONE QUALITY IN PATIENTS WITH TYPE 1 GAUCHER DISEASE WITH MICROINDENTATION. PRELIMINARY RESULTS***

Jordi Pérez-López1, Sabina Herrera2, Marc Moltó1, Roberto Güerri-Fernández2, Elena Cabezudo3, Silvana Novelli4, Jordi Esteve5, Albert Hernández6, Inmaculada Roig7, Xavier Solanich8, Daniel Prieto-Alhambra9, Xavier Nogués2, Adolfo Díez-Pérez2.
1 Inborn Errors of Metabolism in Adults. Unit of Rare Diseases. Hospital Vall d'Hebron, Barcelona. 2 Internal Medicine, Hospital del Mar, Barcelona. 3 Hematology Department, Hospital de Sant Joan de Déu, Manresa, Barcelona. 4 Hematology Department, Hospital de Sant Pau, Barcelona. 5 Hematology Department, Hospital Clínic, Barcelona. 6 Internal Medicine Department, Hospital Comarcal de Sant Jaume, Calella, Barcelona. 7 Hematology Department, Consorci Hospitalari Parc Taulí, Sabadell, Barcelona. 8 Internal Medicine Department, Hospital de Bellvitge, Barcelona. 9 Oxford NIHR Musculoskeletal Biomedical Research Unit, Nuffield Department of Orthopaedics, Rheumatology, and Musculoskeletal Sciences, University of Oxford, Oxford, United Kingdom

P41 ***OCCURRENCE OF ADVERSE BONE EVENTS IN ADULTS AND CHILDREN WITH GAUCHER DISEASE TREATED WITH TALIGLUCERASE ALFA***

Michael Wajnrajch1, Betina Hernandez1, Li-Jung Tseng1, Sukirti Bagal1, Roy Mansfield2, Mireille Fernet3
1 Pfizer Inc, New York, USA, 2 Pfizer Inc. Kent, United Kingdom 3 Pfizer Canada, Kirkland, Canada

P42 ***BONE MINERAL DENSITY AND LEAN MUSCLE MASS CHARACTERISTICS IN CHILDREN WITH GAUCHER DISEASE TREATED WITH ENZYME REPLACEMENT THERAPY OR UNTREATED***

Liron Dar MD, Deborah Elstein PhD, Ari Zimran MD, Ehud Lebel MD***
**In partial fulfillment of requirements for an MD degree at the Hebrew University Hadassah School of Medicine*
***Gaucher Clinic and the Department of Orthopedic Surgery, Shaare Zedek Medical Center, affiliated with the Hebrew University Hadassah School of Medicine, Jerusalem Israel*

P43 ***BONE CRISIS IN SMALL'S FOOT BONES IN PEDIATRIC GD PATIENTS UNDER LONG-TERM ENZYME REPLACEMENT THERAPY***

Andrade-Campos Marcio1,2, Sancho-Val Ignacio3, García Inmaculada4, Roca Mercedes5, Giraldo Pilar1,2,6
1Traslational Research Unit. IIS Aragón. 2CIBER enfermedades raras CIBERER, Zaragoza. 3Haematology Department. Alcañiz Hospital. 4Pediatric Hospital Miguel Servet. 5MRI Center, 6FEETEG. Zaragoza. Spain

P44 ***ESTIMATING THE UTILITY ASSOCIATED WITH MODE OF TREATMENT ADMINISTRATION IN GAUCHER DISEASE***

Luba Nalysnyk1, Paul Swinburn2, Monica Hadi2, Alaa Hamed1 1 Sanofi Genzyme, Cambridge, MA, USA*
2Mapi Group, London, UK

P45 ***THERAPEUTIC GOALS AND NORMAL CLINICAL VALUES ACHIEVED WITHIN 4 YEARS OF INITIATING VELAGLUCERASE ALFA IN TREATMENT-NAÏVE PATIENTS WITH GAUCHER DISEASE IN PHASE 3 STUDIES***

Ari Zimran1, Deborah Elstein2, Derlis E Gonzalez3, Elena A Lukina4, Yulin Qin5, Quinn Dinh6, Hadhami Ben Turkia7*
1 Gaucher Clinic, Shaare Zedek Medical Center, affiliated with the Hebrew University-Hadassah Medical School, Jerusalem, Israel 2 Shire Global Medical Affairs, Shire, Zug, Switzerland 3 Instituto Privado de Hematología e Investigación Clínica (IPHIC), Sanatorio Español, Asunción, Paraguay 4 Department of Orphan Diseases, National Research Center for Hematology, Moscow, Russia 5 Biostatistics and Statistical Programming Department, Shire, Lexington, MA, USA 6 Shire Global Medical Affairs, Shire, Waltham, MA, USA 7 Paediatric Department, La Rabta Hospital, Tunis, Tunisia

P46 ***FREQUENCY, DURATION, RELATEDNESS, SEVERITY, AND SERIOUSNESS OF ADVERSE REACTIONS AMONG A POOLED ELIGLUSTAT DATASET OF 393 GAUCHER PATIENTS FROM FOUR CLINICAL TRIALS***

M Judith Peterschmitt1, Gerald F Cox1, Jennifer Ibrahim1, James Macdougall2, Lisa H Underhill1, Sebastiaan JM Gaemers1*
1Sanofi Genzyme, Cambridge, MA, USA; 2Prometrika, LLC, Cambridge, MA,

P47 ***ARIMOCLOMOL AS A CLINICAL CANDIDATE FOR TREATMENT OF TYPE 2 AND TYPE 3 GAUCHER DISEASE***

Cathrine K. Fog-Tonnesen1, Mingshu Ming1, Nikolaj H. T. Petersen 1, Claus Bornaes1, Anders Hinsby1 and Thomas Kirkegaard1*
1 Orphazyme ApS, Copenhagen, Denmark

P48 ***SIGNIFICANT AND CONTINUOUS IMPROVEMENT IN QUANTITATIVE CHEMICAL SHIFT IMAGING (QCSI) IN PATIENTS WITH GAUCHER DISEASE TREATED WITH TALIGLUCERASE ALFA DURING THE EARLY ACCESS PROGRAM***

Ari Zimran, MD1; Tama Dinur, PhD1; Erik Akkerman, PhD2; Carla E. M. Hollak, MD, PhD2; Raul Chertkoff, MD3; Mario Maas, MD2*
1 Gaucher Clinic and Genetic Unit, Shaare Zedek Medical Center, Jerusalem, Israel 2University of Amsterdam, Academic Medical Centre, Amsterdam, the Netherlands 3 Protalix BioTherapeutics, Carmiel, Israel

P49 ***TALIGLUCERASE ALFA DURING PREGNANCY FOR PATIENTS WITH TYPE 1 GAUCHER DISEASE***

Ari Zimran1; Hanna Rosenbaum2; Raul Chertkoff3*
1 Gaucher Clinic and Genetic Unit, Shaare Zedek Medical Center, Jerusalem Israel. 2Rambam Medical Center, Haifa, Israel 3 Protalix BioTherapeutics, Carmiel, Israel

P50 ***PHARMACOKINETICS, SAFETY, AND EFFICACY OF RAPID INFUSIONS OF VELAGLUCERASE-ALFA IN ADULT PATIENTS WITH GAUCHER DISEASE: PRELIMINARY REPORT OF AN INVESTIGATOR-INITIATED PILOT STUDY***

Zimran_A1, Elstein_D1, Arbel_N2, Chicco_G1.
1Gaucher Clinic, Shaare Zedek Medical Center, the Hebrew University Hadassah School of Medicine, Jerusalem Israel, 2Medison Pharma, Petach Tikva, Israel

P51 ***INTRA-MONOCYTIC GLUCOCEREBROSIDASE ACTIVITY: A KEY PARAMETER TO PERSONALIZE IMIGLUCERASE DOSING?***

Juliette Berger1,2, Marie Vigan3,4, Jérôme Stirnemann5, Roseline Froissart6, Nadia Belmatoug7, Florence Dalbies8, Christian Rose9, Agathe Masseau10, Christine Serratrice11, Yves-Marie Pers12, Ivan Bertchansky13, Fabrice Camou14, Monia Bengherbia7, Céline Bourgne1, Alexandre Janel1, Sandrine Saugues1,2, Thu Thuy Nguyen3,4, Bruno Pereira15, France Mentré3,4 and Marc G. Berger1,2,16
1Hématologie Biologique, CHU Estaing, 1 place Lucie Aubrac, 63003 Clermont-Ferrand Cedex 1, France 2 CRB Auvergne, CHU Estaing, 1 place Lucie Aubrac, 63003 Clermont-Ferrand Cedex 1, France 3INSERM, IAME, UMR 1137, F-75018 Paris, France 4 Université Paris Diderot, IAME, UMR 1137, Sorbonne Paris Cité, F-75018 Paris, France 5 Service de Médecine Interne Générale, Hôpitaux Universitaires de Genève, Rue Gabrielle-Perret-Gentil 4, CH-1211 Genève 14, Suisse 6Laboratoire Maladies Héréditaires du Métabolisme, Centre de Biologie Est Groupement Hospitalier Est, 69677 Bron Cedex, France 7Médecine Interne et CRML, Hôpital Beaujon, AP-HP, 100 Bd Général Leclerc, 92110 Clichy, France 8Institut de Cancérologie et Hématologie, Hopital morvan, Brest, France 9 Onco-Hématologie, Hôpital Saint-Vincent de Paul, Boulevard de Belfort, 59000 Lille, France 10 Service de Médecine Interne, CHU, Nantes, France 11University hospital of Geneva , chemin du pont bochet 3, 1226 thonex, Switzerland. 12CHU Montpellier, Hopital St Eloy, Montpellier, France 13CHU de Montpellier, Hopital La Peyronnie, Montpellier 14Réanimation Médicale, Hôpital Saint André, CHU de Bordeaux, 33000 Bordeaux, France. 15DRCI, CHU G. Montpied, 58 rue Montalembert, 63003 Clermont-Ferrand Cedex 1, France 16Hématologie Clinique, CHU Estaing, 1 place Lucie Aubrac, 63003 Clermont-Ferrand Cedex 1, France

Poster 1.

(1)

UPR ACTIVATION AND CHOP INDUCTION OF GBA TRANSCRIPTION IN GD DERIVED CELLS

Hila Braunstein[1*], Gali Maor[1*], Mirella Filocamo[2] Ari Zimran[3], and Mia Horowitz[1#],

[1]Department of Cell Research and Immunology, Tel Aviv University, 69978, Israel, [2]Centro di Diagnostica Genetica e Biochimica delle Malattie Metaboliche, IRCCS G. Gaslini, Genova, Italy, [3]Department of Internal Medicine, Shaare Zedek Medical Center and Hebrew University-Hadassah Medical School, Jerusalem, Israel.

Chronic presence of mutant, misfolded proteins in the endoplasmic reticulum (ER) initiates ER stress and induces the ER Associated Degradation (ERAD) of misfolded proteins and the unfolded protein response (UPR).

In Gaucher disease (GD), resulting from mutations in the GBA gene, encoding lysosomal acid β-glucocerebrosidase, a certain fraction of the mutant variants is retained in the ER. This retention activates the UPR. We have shown in the past UPR activation in GD derived fibroblasts, in fibroblasts that derived from carriers of GD mutations and in Drosophila models of carriers of GD mutations.

In the present work we extended our studies to include a large collection of GD derived fibroblasts, GD derived-EBV-transformed B-cells and white blood cells derived directly from patients. Our results show that UPR is activated in all the tested cells. They also indicate that transcription of the GBA gene is upregulated through activation of the UPR-induced CHOP transcription factor. Transcription of MAN2B gene, encoding alpha-mannosidase and ACP gene, encoding acid phosphatase was also elevated presumably through CHOP activation.

Our results highlight the existence of chronic stress in GD derived cells due to the presence of ER-retained, mutant GCase.

(2)

ANALYSIS OF HUMAN GLUCOCEREBROSIDASE EXPRESSED IN PICHIA PASTORIS

Marcela de Oliveira Vitarelli[1,2]*, Eliane Namie Miyaji[2] , Paulo Lee Ho[2]

[1] *Chemistry Institute, University of São Paulo, São Paulo, Brazil*
[2] *Biotechnology Center, Butantan Institute, São Paulo, Brazil*

*Corresponding author: vitarelli.marcela@gmail.com

Introduction: Gaucher disease (GD) is a rare autosomal recessive disorder, affecting approximately 600 individuals in Brazil. Therapies consist of human glucocerebrosidases (GCR), produced in human or vegetable cells, containing exposed mannose residues. Even with a relatively low number of patients, government expenses in purchasing GCR are considerably high. Pichia pastoris has a high-mannose glycosylation pattern that we seek to explore in order to produce a national low-cost biosimilar. Objectives: Evaluate the expression of GCR and its glycosylation pattern in P.pastoris aiming to produce a biosimilar for enzyme replacement therapy for GD. Methods: Human GCR genes were amplified by PCR and two secretion signal sequences were used. pPIC3.5/spalbGCR and pHIL-S1/GCR plasmids were constructed and used to transform P.pastoris GS115. GCR expression was induced with methanol. Western blot with anti-GCR serum was performed to confirm expression in both cell lysate and culture supernatant. Results: GCR bands with the expected size (1,5Kb) were detected by PCR. Constructed vectors were confirmed by restriction enzyme digestion and sequencing. GCR expression in P.pastoris was detected in both cell lysate and supernatant, presenting a low secretion level. Discussion: GCR expressed intracellularly presents a molecular mass of approximately 66kDa, suggesting the presence of the Man8GlcNAc2 intermediate, which may be clinically functional. The enzyme is also secreted (molecular mass > 97kDa) and low levels of the protein in the supernatant may be due to the non-yeast codon optimized sequence. Purification steps will follow for further analysis of the GCR glycosylation pattern and activity.

Support: Fapesp, CAPES and Fundação Butantan

(3)

DECREASES IN GLUCOSYLSPHINGOSINE (LYSO-GL-1) CORRELATE WITH CLINICAL RESPONSE IN ADULTS WITH GAUCHER DISEASE TYPE 1 TREATED WITH ORAL ELIGLUSTAT

M. Judith Peterschmitt[1]*, Kate Zhang[1], Lin Liu[1], Ames Macdougall[2], Gerald F. Cox[1]

1Sanofi Genzyme, Cambridge, MA, USA;
2Prometrika, LLC, Cambridge, MA, USA

*Corresponding author: Judith.Peterschmitt@Genzyme.com

Glucosylsphingosine (lyso-GL-1), a minor substrate of acid β-glucosidase, is increasingly recognized as a reliable and highly specific biomarker of Gaucher disease that is associated with disease pathogenesis. In this post-hoc analysis, lyso-GL-1 levels and correlations with changes in spleen and liver volumes, hemoglobin, platelet count, chitotriosidase, CCL18, and GL-1 were evaluated in 26 treatment-naïve patients who participated in the eliglustat Phase 2 trial (NCT00358150, Sanofi Genzyme) for up to 4 years. Lyso-GL-1 was extracted from dried blood spots using methanol: acetonitrile:water (80:15:5) and analyzed by HPLC tandem-mass spectrometry with dimethyl psychosine as the internal standard. The coefficient of variation was 8.8%. Baseline median lyso-GL-1 levels (942 ng/mL, range: 248–2418) were more elevated than other biomarkers tested (165-fold), with no overlap with normal values (5.7 ng/mL, range: 3.0–14.8). After 1 year, median lyso-GL-1 decreased by 61% (SD: 11.0%) and continued to decrease in parallel with chitotriosidase to a final median reduction of 83% (SD: 11.8%) at year 4. Absolute decreases in lyso-GL-1 correlated with absolute improvements in spleen volume (r=0.690), hemoglobin (r=0.515), platelet count (r=-0.279), and chitotriosidase (r=0.386). Correlations between lyso-GL-1 and liver volume and GL-1 were small and clinically insignificant. The association between lyso-GL-1 levels and clinical parameters using a Repeated Measures Mixed Model were statistically significant ($P<0.05$ for all associations). Because it is highly specific to Gaucher disease, significantly elevated in all patients, and in the causal pathway of the disease, lyso-GL-1 may prove to be a more useful clinical marker of treatment response than chitotriosidase.

(4)

CROSS-SECTIONAL STUDY OF GLUCOSYLSPHINGOSINE (LYSO GB1) IN AN UNSELECTED COHORT OF ADULT PATIENTS WITH GAUCHER DISEASE REVEALS LOWEST LEVELS IN PATIENTS TREATED WITH VELAGLUCERASE ALFA

Dinur_T1, Chicco_G1, Elstein_D1, Bdolach-Avram_T1, Cozma_C3, Eichler_S3, Rolfs_A2/3, Zimran_A1*.

1 Gaucher Clinic, Shaare Zedek Medical Center, Jerusalem, Israel
2 Albrecht-Kossel-Institute for Neuroregeneration, University of Rostock, and CentoGene, Rostock, Germany
3 Centogene AG, Institute for Rare Diseases, Rostock, Germany

* Corresponding author: azimran@gmail.com

Glucosylsphingosine (LysoGb1) has recently emerged as a specific and sensitive biomarker for Gaucher disease. Accurate and reproducible measures are performed on dry blood spots (DBS); the results are correlated with disease severity, and the reduction with effective treatment tends to occur faster than other biomarkers. Therefore we have added LysoGb1 to the routine follow-up of our patients, carried by DBS and performed at Centogene AG/Germany.

Herein we report the results of 205 consecutive patients who have attended our clinic for a first-time assessment or for a follow-up visit from July 2014 to February 2015. We have analyzed the results according to the following 7 sub-groups: Untreated (n=61) and 6 ERT-treated groups: Imiglucerase (n=24), velaglucerase (n=29), taliglucerase (n=21), imiglucerase to velaglucerase (n=52), imiglucerase to taliglucerase (n=12) and taliglucerase to velaglucerase (n=6). Statistical analysis using Kruskal-Wallis Test, Mann-Whiteny test and the Bonferroni correction revealed that the only group showing a statistically significant lower value of LysoGB1 ($p<0.001$) compared to all others was the velaglucerase-alfa only.

The lack of statistical significant difference between each of the ERT groups except for velaglucerase and untreated patients, reflects the beneficial impact of ERT on disease severity. The second noteworthy finding is the emergence of velaglucerase-alfa as a potentially more effective ERT with regard to reduction of LysoGb1 levels ($p<0.0013$). While this result is compatible with our previous report of a booster-effect in patients switched from imiglucerase to velaglucerase-alfa, it should be considered preliminary, and awaits further studies, preferably via prospective, double-blind clinical trials.

(5)

THE UTILITY OF GLUCOSYLSPHINGOSINE AS A PATHOPHYSIOLOGICALLY RELEVANT BIOMARKER OF GAUCHER DISEASE

Vagishwari Murugesan1*, Wei-Lien Chuang2, Jun Liu1, Andrew Lischuk3, Katherine Kacena4, Haiqun Lin5, Gregory M Pastores6, Ruhua Yang1, Joan Keutzer2, Kate Zhang2, Pramod K Mistry7

1 Department of Internal Medicine, Yale University School of Medicine, New Haven, Connecticut, USA 2Genzyme Corporation, Framingham, Massachusetts, USA 3 Department of Radiology, Yale University School of Medicine, New Haven, Connecticut, USA 4 Kacena Consulting, Natick, Massachusetts, USA 5 Department of Biostatistics, Yale School of Public Health, USA 6 Department of Neurology, New York University School of Medicine, New York, USA 7 Department of Internal Medicine & Pediatrics, Yale University School of Medicine, New Haven, Connecticut, USA

*Presenting Author: Vagishwari Murugesan: Vagishwari.murugesan@gmail.com

Introduction: Gaucher disease (GD) leads to accumulation of glucosylceramide (GL1) and its lysolipid, glucosylsphingosine (lyso-GL1). Lyso-GL1 has been implicated in mediating immune dysregulation and skeletal disease in GD. Methods: Serum levels of lyso-GL1 were measured in 169 patients with GD by LC/MS. Significant predictors of Lyso-GL1 levels were assessed by Pearson's correlation co-efficient, Wilcoxon Mann Whitney test and multiple linear regression. Propensity score were used to match patients on treatment mode- those receiving Enzyme Replacement Therapy (ERT) and Eliglustat Tartrate SRT (ET-SRT). Results: Of the 169 patients, 41 patients were followed from prior to initiation of ERT. In healthy controls, serum lyso-GL1 was barely detectable: 1.5 ng/ml (1.3 – 1.7; 95% CI). In untreated GD patients, plasma lyso-GL1 level was dramatically elevated (180.9 ng/ml: 95% CI, 145.4 – 216.5) and imiglucerase ERT resulted in marked reduction of plasma lyso-GL1 levels (89 ng/ml: 95% CI, 69.2 – 129.4) ($p<0.001$). Lyso-GL1 levels correlated with chitotriosidase ($r=0.59$ $p<0.0001$), CCL18 ($r= 0.62$ $p <0.0001$), hepatomegaly ($r=0.28$ $p<0.0006$), splenomegaly ($r=0.27$ $p=0.003$), splenectomy ($p=0.01$) and treatment mode ($p<0.001$). By multiple linear regression, the strongest predictors of Lyso-GL1 were age ($p<0.001$), splenectomy ($p=0.02$), Chitotriosidase ($p<0.001$) and CCL18 levels ($p=0.001$). After propensity score matching to obtain comparable groups of patients on ERT vs ET-SRT, Lyso-GL1 levels were significantly lower among patients receiving ET-SRT by 113 ng/ml (95% CI : 136 – 90.3 ng/ml $p<0.001$). Conclusion: Circulating lyso-GL1 is a pathophysiologically relevant biomarker to assess disease severity in GD. Compared to ERT, E-SRT was associated with significantly greater reduction of this bioactive lipid.

(6)

GLUCOSYLSPHINGOSINE (LYSO-GB1) CONCENTRATION IN THE BLOOD OF GAUCHER PATIENTS REFLECTS THE SEVERITY OF GBA MUTATIONS

Claudia Cozma1, Gabriela Oprea1, Guido Kramp1, Sabrina Eichler 1, Anne Giese2, Jan Lukas2, Tobias Bötcher2, Arndt Rolfs 1,2

1 Centogene AG; Rostock, Germany
2 Albrecht-Kossel-Institute, University of Rostock, Rostock, Germany

* claudia.cozma@centogene.com

Background and aims: We report here the identification of 113 unique GBA genetic variants from 637 genetically confirmed Gaucher patients and carriers.

Method: In our laboratory diagnosis of Gaucher disease is performed in a high-throughput stepwise manner: enzymatic activity, lyso-Gb1 quantification in DBS or plasma followed by GBA gene sequencing. Determination of lyso-Gb1 is performed by LC/MRM-MS, an analytical method proven to be reliable and reproducible in plasma and DBS.

Results: Lyso-Gb1 was investigated in three GBA cohorts: (A) comprising 103 homozygotes GBA cases; (B) 286 compound heterozygotes GBA cases and (C) 149 heterozygotes GBA carriers. The lyso-Gb1 levels found in blood from GBA carriers and Gaucher patients were divided in: very mild (<15.0 ng/mL), mild (15.1-50 ng/mL), moderate (50.1-200 ng/mL) and severe (>200 ng/mL).

Cohort A was characterized by 52.5% of cases with severe lysoGb1 levels, followed by 35.9% moderate and 11.6% mild biomarker levels. B included 40.9% severe, 38.9% moderate, 14.6% mild and 5.6% very mild lyso-Gb1 levels. All GBA carriers were characterized by Lyso-Gb1 levels < 12 ng/mL.

The most severe mutations were c.1060G>A; p.D354N (783+/-192 ng/ml), 408_412del; p.P137Cfs*7 (769+/-573), followed closely by c.463T>C; p.Y155H (717.5+/-65.7). Statistically, concentrations >300 ng/mL lyso-Gb1 are correlated with mutations in GBA exons 2, 3, 4, 8 and 10 and with severe clinical symptoms.

Conclusion: The levels of lyso-Gb1 in blood can be used for the easy diagnosis of Gaucher patients and for the monitoring of the disease progression.

(7)

QUANTIFICATION OF GLUCOSYLSHINGOSINE (LYSO-GB1) FOR THE DIAGNOSIS AND MONITORING OF GAUCHER DISEASE -METHODOLOGY AND VALIDATION

Claudia Cozma1, Guido Kramp1, Sabrina Eichler 1, Arndt Rolfs 1,2

1 Centogene AG; Rostock, Germany
2 Albrecht-Kossel-Institute, University of Rostock, Rostock, Germany

* claudia.cozma@centogene.com

Gaucher disease (GD) is an autosomal recessive disease caused and characterized by a deficient beta-glucocerebrosidase activity, resulting in accumulation of glycolipids in cells and tissues of the affected patients. Standard diagnostic procedures include measurement of enzyme activity, genetic testing as well as analysis of biomarkers. We present here the characteristics of the lyso-Gb1 (glucosylsphingosine) determination in clinical set-up as a clinically relevant and important alternative to less specific biomarkers, such as chitotriosidase and CCL18/PARC.

The method is based on the extraction of biomarker lyso-Gb1 from dried blood spots (DBS) or plasma and specific determination and quantification by multiple reaction monitoring mass spectrometry (MRM-MS).

The method validation process revealed the following characteristics for lyso-Gb1 determination: (i.) Intra- and inter assay precision were found to have CV% < 10% for both DBS and plasma for all tested concentrations. (ii.) For accuracy of the MRM-MS method the CV% was < 8% and linearity was demonstrated for the analytical range. (iii.) Reference values were determined on normal controls (average +2*STD) at 1.2 ng/mL (plasma) and 4.8 ng/mL (DBS). (iv.) Pathological range 23.2 to 226.0 ng/mL (plasma) and 25.4 to 771.0 ng/mL (DBS). (v.) LOD and LOQ was determined on blanks, (vi.) specificity and sensitivity was determined to be 100 % (vii.) lyso-Gb1 concentration is stable for years in DBS stored at room temperature (viii.) a correlation factor was found between plasma concentration and DBS.

Lyso-Gb1 was demonstrated to have the highest sensitivity and specificity to date for diagnosis and monitoring of GD.

(8)

ROLE OF RED BLOOD CELL ON THE PATHOPHYSIOLOGY OF GAUCHER DISEASE AND EFFECT OF THE ENZYME REPLACEMENT THERAPY

Franco M1-4, Reihani N1-4, Marin M1-4, Collec E1-4, Billette de Villemeur T5,6, Rose C6,7, Mignot C5,6, Colin Y1-4, Belmatoug N6,8 & Le Van Kim C1-4.

1Inserm U1134, Paris, France; 2Université Paris Diderot, Sorbonne Paris Cité, UMR_S 1134, Paris, France; 3Institut National de la Transfusion Sanguine, Paris, France; 4Laboratoire d'excellence GR-Ex, Paris, France; 5Service de Neuropédiatrie, Hôpital Trousseau, Assistance Publique–Hôpitaux de Paris and Université Pierre et Marie Curie, Paris, France; 6Centre de Référence des Maladies Lysosomales, Clichy, France; 7Service d'Hématologie et de cardiologie Hôpital Saint Vincent de Paul, Université Catholique de Lille, Lille; 8Service de Médecine Interne, Hôpital Beaujon, Clichy, France;

melanie.franco@inserm.fr

Gaucher disease (GD) is a rare genetic disease caused by lysosomal glucocerebrosidase deficiency, impairing glucosylceramide catabolism. Macrophages are considered as responsible for the major GD symptoms. However, other symptoms such as anemia, vascular occlusion and spleen and bone infarcts might involve red blood cells (RBCs). We have previously demonstrated that RBCs from GD patients exhibit abnormalities indicating new role in GD pathophysiology.

To determine whether enzyme replacement therapy (ERT) normalizes RBC parameters from GD patients, we compared RBCs properties between healthy volunteers (n=19), untreated naïve GD patients (n=16) and Velaglucerase treated-patients group (n=15). 7 GD patients were also followed propsectively before and after starting ERT with at least 1 year of follow-up. RBC morphology, hemorheologic measurements and RBC adhesion to laminin were examined.

Although RBCs from untreated-patients exhibit higher aggregation properties and adhesion to laminin compared to controls, we do not observe any amelioration of these parameters in Velaglucerase-treated patients. However, RBC from untreated-patient exhibits abnormal morphology and lower deformability index compared to controls and these parameters are normalised by ERT. Moreover these parameters correlates with marker of disease severity such as low hemoglobin and high CCL18 levels.

Our data indicate that some GD RBCs properties might be target of ERT and new marker of treatment efficiency. This study highlight also that other RBC abnormalities does not seem to be affected by ERT and suggest that other additive therapeutic options might be useful in order to minimize RBC abnormalities.

This work was supported in part by grants from Shire.

(9)

IRON IMAGING AND PARAMETERS OF IRON METABOLISM IN ADULT TYPE 1 GAUCHER DISEASE

Regenboog M1-2*, Bohte AE1, Akkerman EM1, Stoker J1, Hollak CEM2

1Department of Radiology, 2Department of Internal Medicine, division of Endocrinology & Metabolism. Academic Medical Center Amsterdam, The Netherlands.

*presenting author: m.regenboog@amc.uva.nl

Introduction. Gaucher Disease (GD) is characterized by large amounts of lipid-storing macrophages and is associated with accumulation of iron. High levels of ferritin are a hallmark of the disease. We hypothesized that magnetic resonance imaging R2* iron quantification can serve as a marker of (residual) disease burden. In addition, liver iron could play a role in development of hepatic fibrosis and hepatocellular carcinoma (HCC). In the past, patients developing HCC all had high levels of ferritin and evidence of fibrosis. Methods. 40 GD1 patients and 40 matched healthy controls were examined using a whole-body MRI protocol. Regions-of-interest (ROI) were defined for abdominal viscera and bone marrow compartments. R2* values of a ROI were calculated as marker for the amount of iron. Differences in median R2* values were analyzed per organ. Results. Iron distribution in GD shows a heterogeneous picture, with signs of iron detected in typical GD localizations (spleen, liver and bone marrow), not in other sites. Liver R2* values were significantly higher in GD patients as compared to healthy controls, median 41 Hz (range 28-165) versus 38 Hz (range 28-53) (p=0.0064). Splenectomized patients (n=10) did not show a significant difference in liver R2* values compared to their matched controls. Splenic R2* values in GD patients showed a median of 32.26 Hz (range 13.12-114.5) versus 28.95 Hz (range 15.23-48.9) in controls (p=0.08). Liver R2* correlated with spleen R2* (R2 0.338, p=0.001). Ferritin levels were elevated in 58% of patients (median 654 µg/L, range 296-1520). Hepcidin levels did not differ between patients and controls (median 4.3 nM (<0.5-20.3) versus 5.0 nM (<0.5-15.3) respectively). In a multivariate linear regression analysis, liver R2* correlated well with ferritin levels (standardized β = 0.448, p=0.017), but not with hepcidin levels. In the six patients with the highest liver iron, three were heterozygous for the C282Y mutation, one had iron overload in the past after frequent transfusions and two were on miglustat or eliglustat. Splenic iron was specifically high in three patients with extensive Gaucheroma in the spleen. In several patients, despite elevated chitotriosidase levels indicative of residual Gaucher cells, normal iron imaging was established. Conclusion. Iron imaging with MRI shows accumulation of iron in liver and spleen in a subset of patients. This modality does not detect all residual disease sites. We hypothesize that measurement of iron status and liver iron content is of importance for early detection of hepatic injury including fibrosis and hepatocellular carcinoma.

(10)

SERUM FERRITIN AND LIVER FEATURES IN A COHORT OF 15 PATIENTS AFFECTED BY GAUCHER DISEASE

Miriam Rigoldi[1], Lorenza Borin[2], Raffaella Mariani[1], Irene Pelloni[1], Federico Greni[3], Cristina Arosio[4], Alberto Piperrno[1,2,4]

[1]Center for rare disease, Internal Medicine 2, ASST- Monza S.Gerardo Hospital, Italy
[2]Hematology Unit, ASST-Monza S.Gerardo Hospital, Italy
[3]University of Milano-Bicocca, School of Medicine and Surgery – Monza, Italy
[4]Consortium for Human Molecular Genetics – Monza, Italy

*Corresponding author: rigoldimiriam@gmail.com

Aim: To evaluate serum iron indices and liver features before and after ERT in a cohort of Italian patients with Gaucher disease.

Patients. Fifteen patients (7F, 8M), affected by type I (N=14) and III (N=1), aged 1 to 72 years at diagnosis, with a median follow up of 9 years (1st-3rd quartils: 2-16.5).

Results: At diagnosis, 14/15 patients showed increased serum ferritin (SF) levels that was over 1000 ng/ml in six. Transferrin saturation (TS) was 23.5+9.2% (mean+SD). In the six patients with the highest SF levels no causal mutations in HFE and ferroportin genes were found. At liver ultrasound, 3 patients showed steatosis, 3 multiple nodules, and 1 a single nodule. Three patients underwent to liver biopsy. The first showed hepatocellular carcinoma in noncirrhotic liver (splenectomised) with severe iron overload (IOD) (Scheuer IV); the second marked steatosis without fibrosis and moderate IOD (Scheuer II-III) the third normal liver structure with mild-moderate IOD (Scheuer I-II). Iron deposits involved not only reticulo-endothelial cells but also hepatocytes suggesting the existence of different patterns of liver iron accumulation.

Thirteen patients underwent to ERT showing improvement of hemoglobin ($p<0.01$), splenomegaly ($p=0.015$), chitotriosidase ($p=0.045$), platelet count ($p=0.05$), and SF ($p=0.08$).

Conclussion: In our cohort of Gaucher patients increased SF with normal TS was common, being the first manifestation in four. It was not related to hemochromatosis genes mutations, but associated with other indicators of disease and improved by ERT.

(11)

GAUCHER DISEASE AND DEFICIENCY OF INTESTINAL DISACHARIDASES

Medrano-Engay B[1, 2], Irún MP[1, 2, 3], Andrade-Campos M[1, 2, 3], Pocoví M[2, 4], Giraldo P[1, 2, 3, 5]

Corresponding author: mbmedrano@iisaragon.es

[1]Unidad de Investigación Traslacional. Hospital Universitario Miguel Servet. Zaragoza. España. [2]Instituto Investigación Sanitaria Aragón. Zaragoza. España. [3]Centro de Investigación Biomédica en Red de Enfermedades Raras (CIBERER), ISCIII. Zaragoza. España. [4]Departmento de Bioquímica y Biología Molecular y Celular. Universidad de Zaragoza. España. [5]Fundación para el Estudio y Terapéutica de la Enfermedad de Gaucher. Zaragoza, España.

Patients under miglustat therapy experience a high incidence of Gastrointestinal Disturbances (GaD). We have tried to deep in the origin of GaD in Gaucher Disease (GD) patients and its relationship to the deficient activity of intestinal disacharidases. Miglustat induce GaD like bad digestion carbohydrate symptoms, in vitro studies have demonstrate that miglustat is a potent inhibitor of intestinal disaccharidases, such as maltase and sucrase and lactase somewhat weaker. We have designed an analytical study to determine the activity profile of these three disaccharidases enzymes in 40 GD patients and 20 healthy controls and their relationship with the occurrence of GaD. GD patients were classified in two groups: exposed to miglustat (21) and never treated with miglustat (19). All subjects were surveyed for the presence and intensity of gastrointestinal symptoms, eating habits, intestinal rhythm and intake another drugs or concomitant diseases. For the study of disacharidase activities a breath test were performed for every sugar sucrase, maltase and lactase. Molecular study of the polymorphism SNP -13910C>T in the MCM6 gene was determined. GD patients have a higher proportion of positive breath test results to at least one of the three sugars tested (50%) than the control group (35%). A total of 14/21 (67%) GD patients exposed to miglustat showed a malabsorption phenotype vs 35% in control group and 32% in GD patients unexposed to miglustat ($p = 0.028$). High correlation between the CC genotype in the MCM6 gene and the lactase activity was observed.

This work has been partially granted by FEETEG

(12)

PERIPHERAL BLOOD LYMPHOCYTE IMMUNOPHENOTYPE ABNORMALITIES IN PATIENTS WITH TYPE I GAUCHER DISEASE

Eugenia Verigou[1], Christos Sotiropoulos[1], Kalliopi Zafeiropoulou[1], Theodore Marinakis[2], Constantina Repa[3], Panagiotis Tsaftaridis[4], Vasileios Lazaris[1], Alexandra Kourakli[1], Argiris Symeonidis[1]*

[1]Hematology Division, Dept of Int.Medicine, University of Patras Medical School, Patras, Greece. [2]Dept of Hematology, "G.Gennimatas" General Hospital, Athens, Greece. [3]Thalassemia Center, Laikon General Hospital, Athens, Greece. [4]Dept of Haematology, Laikon General Hospital, National and Kapodistrian University of Athens, Greece.

*Corresponding author: argiris.symeonidis@yahoo.gr

Patients – Methods: We performed peripheral blood flow cytometry for evaluation of 28 lymphocyte subsets in 19 patients with type-I Gaucher disease (GD) and 10 controls. We used 11 surface markers, combined in three 5-color protocols. Measurements were performed on FC500 cytometer (Beckman Coulter), listmode files were analyzed with Kaluza 1.2 and statistics was performed with SPSS-21. Results are displayed in parentheses, with median and interquartile range. Results: GD patients exhibited absolute lymphopenia (31.85%, 8.45 vs 41%, 16.02, p=0.031), reflecting significant decrease of all major T-cell subsets (CD4+, CD8+, HLA-DR+ T-cell, NK-T, NK-Tγδ). Different activation status of early activated lymphocytes was not confirmed. However, different activation pattern was presented, and late activated- or HLA-DRdim cells were slightly decreased (3.92%, 2.2 vs 5.63%, 4.78), resulting in significantly reduced absolute HLA-DR+ T-lymphocytes (66.79/μL, 42.55 vs 93.87/μL, 70.23, p=0.035), whereas HLA-DRbright lymphocytes were elevated (11.7%, 6.93 vs 9.87%, 4.91). Interestingly, NK-T subsets were decreased in percentage and/or absolute counts (NK-T: 2.19%, 4.86 vs 3.19%, 6.08, and 39.38/μL, 64.22 vs 70.22/μL, 144.04, p=0.04, NK-Tγδ: 0.92%, 2.85 vs 1.46%, 1.75 and 22.83/μL, 23.32 vs 33.07/μL, 34.38, p=0.024) whereas NK cell percentage was not significantly different (10%, 10.65 vs 9.29%, 10.02). Conversely, patients had increased memory T-cells (CD4+CD45RO+: 21.9%, 5.20 vs 16.22%, 7.61, p=0.11, and CD8+CD45RO+: 4.92%, 3.24 vs 3.27%, 1.40, p=0.21) which resulted in similar absolute numbers between the two groups. B-lymphocytes were slightly increased in patients, but differences were not significant.

Conclusions: Patients with GD exhibit various immunoregulatory abnormalities, impairing also immune surveillance T/NK-cell subsets

(13)

SCREENING FOR DELETIONS/DUPLICATIONS IN THE GBA1 GENE USING MULTIPLEX LIGATION-DEPENDENT PROBE AMPLIFICATION METHOD IN GAUCHER PATIENTS

Marina Siebert[1,2,3], Suelen P. Basgalupp[2,4], Filippo P. Vairo[2,5], Ida V. D. Schwartz[2,4,5,6*].

*Corresponding author: idadschwartz@gmail.com

[1]Postgraduate Program in Sciences of Gastroenterology and Hepatology, Faculty of Medical Sciences, Universidade Federal do Rio Grande do Sul, Porto Alegre, RS, 90035-003, Brazil; [2]Basic Research and Advanced Investigations in Neurosciences (BRAIN) Laboratory, Experimental Research Center. Hospital de Clínicas de Porto Alegre (HCPA), Porto Alegre, RS, 90035-903 Brazil; [3]Unit of Molecular and Protein Analyses, Experimental Research Center, Hospital de Clínicas de Porto Alegre (HCPA), Porto Alegre, RS, 90035-903 Brazil; [4]Postgraduate Program in Medical Sciences, Faculty of Medical Sciences, Universidade Federal do Rio Grande do Sul, Porto Alegre, RS, 90035-003, Brazil; [5]Medical Genetics Service, Hospital de Clínicas de Porto Alegre (HCPA), Porto Alegre, RS, 90035-903 Brazil; [6]Department of Genetics, Universidade Federal do Rio Grande do Sul, Porto Alegre, RS, 91501-970, Brazil.

Gaucher disease (GD) is caused by a deficiency of glucocerebrosidase due to mutations in the GBA1 gene. Our aims were to validate the P338-X1 GBA kit (MRC-Holland) for Multiplex Ligation-dependent Probe Amplification (MLPA), and to detect large deletions and/or duplications present in GBA1 in GD patients from Southern Brazil. The sample group included 36 unrelated GD patients in whom the GBA1 gene was previously analysed by Sanger sequencing, being 31 with both mutations identified, 4 with only one mutation and one with none identified mutation. The MLPA kit contains one probe for each of the following regions of GBA1: 5'UTR, exons 3, 4, 6, 8, 9, 10, and intron 7. The exon 10 probe generates a normal signal at L444, but reduced signal when L444P or L444R is present. Amplified products were analyzed with the ABI3500 equipment and Coffalyser software. Out of 72 alleles, 35 (48.6%) were found to have L444P and 1 (1.4%), L444R, confirming our sequencing results; however, we were not able to distinguish if the reduced signal was due to the presence of RecNciI, L444P+A456P or L444P+E326K. Only one patient presented a heterozygous deletion in intron 7; according to the sequencing, this patient is also heterozygous for RecNciI. No other evidence for deletion/duplication was found. After MLPA analysis, it is still remaining 5 uncharacterized alleles from 4 GD patients. Additional approaches are necessary to evaluate uncovered regions of GBA1 (exons 1, 2, 5, 11, promoter and 3'UTR).

(14)

SCREENING FOR LYSOSOMAL STORAGE DISEASES IN FLANDERS – BELGIUM: PILOT STUDY AND DIAGNOSTIC OPPORTUNITIES

Eyskens François*, Devos Sylvie

PCMA vzw-Metabolic Lab, University of Antwerp, Antwerp, Belgium

*Corresponding author : francois.eyskens@provincieantwerpen.be

Background Lysosomal storage diseases (LSD) are a group of metabolic disorders with various clinical presentations, which complicates diagnosis. Recent studies highlight the advantages of early diagnosis. A pilot study was performed to test the appropriateness and effectiveness of the method for UHPLC-MS/MS newborn and diagnostic screening for Gaucher, Fabry, Pompe, MPS I, MPS II, MPS IVA and MPS VI in dried blood spots.

Methods Around 20,000 newborn samples, collected in addition to the routine Flemish screening program, were analyzed with UHPLC-MS/MS for 7 lysosomal enzyme activities: α-glucosidase (GAA, Pompe's disease), α-galactosidase (GLA, Fabry's disease) and α-iduronidase (IDUA, MPS I), β-glucocerebrosidase (ABG, Gaucher's disease), iduronate-2-sulfatase (ID2S, MPS II), N-acetyl-galactosamine-6-sulfatase (GALN, MPS IVA) and N-acetyl-galactosamine-4-sulfatase (ASB, MPS VI).

Results The method was validated in our laboratory. Employing the technique, it was noticed that 2 siblings suffering from Pompe's disease had GAA enzyme activity levels below cut-off just before receiving ERT, whereas 2 siblings suffering from Gaucher's disease type 3 had very high ABG enzyme activity levels just before ERT when receiving 120 U/kg/2 weeks. Moreover, an undiagnosed 6 month old boy and a 7 year old boy tested positive for Pompe and MPS II, respectively.

Conclusion We report for the first time results of a 7-plex enzymatic activity UHPLC-MS/MS assay for the lysosomal storage disorders Gaucher, Fabry, Pompe, MPS I, MPS II, MPS IVA and MPS VI in Europe. Moreover, this technique can be employed for diagnostic testing and the possibility for ERT pharmacokinetics on individual patient level should be further explored.

(15)

ANALYSIS OF 8 MARKERS ASSOCIATED TO N370S AND L444P ALLELES ON MEXICAN PATIENTS WITH GAUCHER DISEASE

Garcia-Ortiz, Jose Elias[1*], Magaña-Torres María Teresa, da Silva-Jose Thiago Donizete[2].

1División de Genética Centro de Investigación Biomédica de Occidente, IMSS. Guadalajara, México.
[2]Doctorado en Genética Humana, CUCS-Universidad de Guadalajara. Guadalajara, México.

*Corresponding autor: jose.elias.garcia@gmail.com

Gaucher disease is caused by mutations in the GBA gene, more than 350 variants have been described but N370S and L444P are the most common worldwide. In some populations, the ancestral origin of these mutations has been established. Here we describe the distribution of five STR (D1S305, D1S2777, D1S2715, D1S2624, D1S1653) and three SNP (rs1800438, rs3754485, rs1800442) in 16 individuals with Gaucher Disease with either N370S or L444P alleles and 70 healthy Mexican Mestizos. Allele and genotype frequencies were obtained by direct counting, Hardy-Weimberg equilibrium was determined for all the polymorphism and haplotypes were constructed with a high heterogeneity Bayesian algorithm using the Arlequin 3.5 program. The distributions of the alleles and haplotypes were reduced in patients with Gaucher Disease compared with the healthy individuals and interestingly we were able to associate a partial region of the haplotype with either N370S or L444P in the 3' region of GBA, in 5' the distributions of combinations were less evident. Even though a direct comparation with the haplotypes described in Askhenazi Jews, Spanish and Colombians was not possible due to the use of different markers and just inference of the alleles due to ambiguous nomenclature, it might be possible to infer an Spanish origin of these mutations.

(16)

GAUCHER DISEASE IN GREECE. LABORATORY INVESTIGATIONS

E.Dimitriou[1*], M.Moraitou[1], L.Vilageliu[2], D.Grinberg[2], J. Serra-Vinardell[2], M. Cozar[2], I.Mavridou[1] and H.Michelakakis[1]

1. Dept Enzymology and Cellular Function, Institute of Child Health, Athens ,Greece

2. Dept. of Genetics, University of Barcelona, CIBERER, IBUB, Barcelona, Spain

*Corresponding author: ecfdept@ich.gr

Gaucher Disease (GD) is a rare lysosomal storage disease. Three main subtypes are recognized depending on the presence and rate of progression of neurological symptoms. In most cases the disease results from mutations in the GBA gene encoding the lysosomal enzyme b-glucocerebrosidase (GCase). We present our findings regarding the laboratory diagnosis of GD in Greece. Overall 120 cases, including 27 siblings, were diagnosed during a period of 30 years. They included 101 Type I, 11Type II, 6 Type III and two cases with non -defined subtype. For laboratory diagnosis assaying of plasma chitotriosidase activity is used as an initial screening test. Final diagnosis is established through GCase assaying in either white blood cell or cultured fibroblast homogenates. The results are confirmed further through mutation analysis. Increased plasma chitotriosidase activity (4x-239x the upper normal limit was found in 106 tested cases, including 4 presymptomatic cases, whereas 2 had zero activity. In all GCase activity was diagnostic. Mutation analysis was carried out in 103 unrelated cases and identified 23 different mutations in 199 alleles in GBA (17 previously described and 6 novel mutations: IVS6-2A>G, T231I, N462Y, L175P, F81L, Y135S) and 31 different genotypes. In all but one type 1 cases the N370S mutation was detected either in hetero- or homozygosity. The latter was confirmed through parental mutation analysis. Patients originated from all parts of Greece, one mutation was seen in Greek patients from Asia Minor. Our cohort includes 10 patients of Albanian origin, 2 from Egypt, 1 from Italy and 1 from Ethiopia.

(17)

CYP2D6 GENOTYPE AND METABOLIZER STATUS IN SPANISH PATIENTS WITH TYPE 1 GAUCHER DISEASE

Alfonso P[1,2,3], López de Frutos L[1,3], Andrade-Campos M[1,2,3], Giraldo P[1,2,3]. [1]Instituto de Investigación Sanitaria Aragón (IIS Aragón), Zaragoza, Spain, [2]Centro de Investigación Biomédica en Red de Enfermedades Raras (CIBERER), Zaragoza, Spain, [3]Fundación Española para el Estudio y la Terapéutica de la Enfermedad de Gaucher y otras Lisosomales (FEETEG), Zaragoza, Spain

*Corresponding autor: giraldocastellano@gmail.com

Type 1 Gaucher disease (GD1), the most prevalent lysosomal disease, has two therapy approaches: enzymatic replacement therapy and substrate reduction therapy (SRT). Recently a new SRT has been approval for adult GD1 patients. The indication and dose are individualized and dependant of the metabolized status of CYP2D6, a polymorphic cytochrome P450 isoenzyme. CYP2D6 gene, of autosomal recessive inheritance, has multiple allelic variants, resulting in four phenotypic types: poor metabolizers (PM) with gene inactivation of both alleles; intermediate metabolizers (IM); extensive metabolizers (EM); and ultra-rapid metabolizers (UM) with excess enzymatic activity. In order to know the CYP2D6 profile in Spanish GD1 patients we have designed a prospective study in 30 selected GD1 patients included in the Spanish GD Registry. Genomic DNA was isolated from whole blood and the CYP2D6 genotype was determined using xTAG™ CYP2D6 kit v.3 (Luminex) based on known CYP2D6 variants. This Kit analyzes the most common alleles within CYP2D6 gene. Each sample was amplified in two multiplex PCR reactions, these were pooled and purified. Multiplex allele specific Primer Extension was then performed followed by bead hybridization, wash and analysis on xMAP™ instrument. After analysis, 87,6% patients were IMs and EMs and 12,4% patients were PMs. No UM was observed in our series. Genomic test used to determine the CYP2D6 status is a simple and useful procedure with a minimal variability. CYP2D6 is the way used by numerous drugs to be metabolized; nevertheless other factors can influence drug interactions via affecting expression of the CYP proteins.

This work has been partially granted by FEETEG

(18)

0A COMPARISON OF THE CHINESE AND GLOBAL POPULATIONS FROM THE INTERNATIONAL COLLABORATIVE GAUCHER GROUP (ICGG) GAUCHER REGISTRY

Duan, Yanlong1; Taylor, John S.2**; Luo, Xuequn3; Gu, Longjun4; Gu, Xufan5; Meng, Yan6; Zhang, Huiwen5; Huang, He7; Zheng, Weiyan7; Zhang, Yonghong1*

Beijing Children's Hospital, Beijing, China;
Sanofi Genzyme, Cambridge, MA, USA;
First Affiliated Hospital of Sun Yat-Sen University, Guangzhou, China;
Shanghai Children's Medical Center, Shanghai, China;
Shanghai Xinhua Hospital, Shanghai, China;
Chínese PLA General Hospital, Beijing, China;
Zheng Zhejiang University Affiliated 1st Hospital, Hangzhou, China

*yhzhang58@hotmail.com

**JST is currently employed at Johnson & Johnson Vision Care, Inc.

Background: Gaucher disease (GD) is best characterized in patients of Ashkenazi-Jewish ancestry; however, interesting differences have been described among more numerically dominant patient populations of other ethnic backgrounds. Methods: This analysis compares the pre-treatment demographic, genotype, and phenotypic disease characteristics of GD patients in China enrolled in the ICGG Gaucher Registry (NCT00358943, sponsored by Sanofi Genzyme) (N=134) with the Worldwide ICGG Gaucher Registry population (N=5980) as of 14 January 2013. Results: Most Chinese patients enrolled in the Registry had at least one Rare (77%) or L444P allele (53%); Worldwide the primary mutation reported for GD patients in the Registry was N370S (79%). At Baseline, more patients in China reported anemia (52%), moderate to severe thrombocytopenia (non-splenectomized, 90%), severe splenomegaly (93%), and moderate to severe hepatomegaly (85%) relative to Worldwide patients (43%, 76%, 48%, and 73%, respectively). Fewer patients in China reported bone pain (26%) and bone crisis (4%) relative to Worldwide patients (49% and 13%, respectively). Conclusion: This analysis describes the largest cohort of patients with GD in China evaluated to date. Compared with Worldwide patients, the subset of patients from China had a higher prevalence of Rare and L444P alleles; were qualitatively more likely to have severe anemia, thrombocytopenia, splenomegaly and moderate to severe hepatomegaly at baseline; and are qualitatively less likely to have bone pain or bone crises. Greater participation in the Registry will enable even more comprehensive charaterization studies of GD patients in China and throughout Asia relative to other ethnic populations, which might impact the management of Gaucher Disease in Chinese and Asian populations.

Disclosures:

YLD, LJG, HH receive honoraria and expense reimbursement for serving on the ICGG Gaucher Registry board of advisors. XQL, YM, YHZ receive honoraria and expense reimbursement for serving on the Registry board of advisors, and fees from Sanofi Genzyme for speaking engagements. XFG receives fees from Sanofi Genzyme for speaking engagements, and research support from Genzyme. HZ receives fees from Sanofi Genzyme for speaking engagements.

(19)

TYPE 1 GAUCHER DISEASE: HOMOZYGOSITY FOR P.R496H MUTATION

P. Irún1,2 , M Andrade-Campos1,2 , E Mingot3 , P. Giraldo1,2,4

1Centro de Investigación Biomédica en Red de Enfermedades Raras (CIBERER), Instituto de Salud Carlos III, Zaragoza. Spain. 2 Instituto Investigación Sanitaria Aragon (IIS Aragón). Zaragoza. Spain. 3Haematology Department. Carlos Haya University Hospital. Malaga. 4Spanish Foundation of Gaucher Disease and other Lysosomal Disorders (FEETEG). Zaragoza. Spain.

*Corresponding author: mpirun.uit@gmail.com

The p.R496H mutation has been reported previously in type 1 Gaucher disease (GD) only in combination with a different mutation on the other allele. It has been classified as a very mild mutation, having discordance between studies that conclude that in combination with N370S, symptoms will most likely not develop and others indicating disease manifestations including bone deformations. No neuronopathic disease has been reported even when it is in combination with a severe or null allele. Carriers are frequent in Ashkenazi population. In the Spanish GD Registry, we have identified two families whose GD index cases are homozygous for p.R496H. Family studies have allowed us to identify one more case in homozygosity and three carriers. The purpose of this study is to describe clinical and biological characteristics of these patients. Two women from two unrelated families were diagnosed of type 1 GD after 40 years old. Index cases showed a GC activity below 26% of a healthy control, mean ChT activity (5,479±790 nmol/mL/h) and mean CCL18/PARC (622±337 ng/mL). The third case detected in familiar studies had normal biomarkers values and GC activity of 22% of a healthy control. Only one patient has bone disease with bone crisis and osteonecrosis in right femoral head, is under ERT and need joint replacement. The other two are asymptomatic and out of therapy. In the two families there are relatives with different neurological disorders (Parkinson disease and dementia). In conclusion the wide clinical variability of GD is also present in patients homozygous for p.R496H.

This work is partially granted by FEETEG

(20)

WANDERING SPLEEN IN 2 GAUCHER PATIENTS ON ENZYME REPLACEMENT THERAPY: DIAGNOSIS AND MANAGEMENT

Lau Heather1*, Liang Howard2, Elstein Deborah3, Alberton Joseph4, Zimran Ari3

1 Department of Neurology, NYU Langone Medical Center, New York, NY USA
2 Department of Surgery, NYU Langone Medical Center, New York, NY USA
3 Gaucher Clinic, Shaare Zedek Medical Center, Jerusalem Israel
4 Department of Surgery, Shaare Zedek Medical Center, Jerusalem Israel

*Corresponding author: Heather.lau@nyumc.org

Wandering spleen is a rare clinical entity characterized by splenic migration into the lower abdomen and/or pelvis, either congenital or acquired, and may be exacerbated by conditions that cause splenomegaly. Serious complications include torsion of the splenic pedicle and subsequent splenic infarction, requiring emergent splenectomy. Clinical presentation is variable. There is one prior report of wandering spleen in a newly diagnosed Gaucher patient (Dweck et al 2001). Here we report 2 more cases of wandering spleen in known Gaucher patients who have been on long standing enzyme replacement therapy (ERT). The first case occurred in a 32 year old Ashkenazi G4P4 woman with Gaucher Type 1 disease (N370S/IVS2+1) on ERT for 15 years. She presented with acute onset of severe generalized abdominal pain. Urgent imaging revealed a displaced and enlarged spleen. The patient underwent successful laparoscopic splenectomy without complication. The second case is a 32 year old Ashkenazi G1P1 woman with Gaucher Type 1 disease (N370S/IVS2+1) on ERT for the past 24 years. She presented with a six-week history of intermittent generalized abdominal pain, exacerbated by eating and prolonged periods of standing. Abdominal imaging confirmed a displaced and enlarged spleen with torsion and parenchymal infarction. Laparoscopic splenectomy was converted to an open and successful splenectomy. In conclusion, this rare condition occurred in 2 Gaucher patients who had been stable on ERT. This report expands the differential diagnosis for onset of new abdominal pain in otherwise stable Gaucher patients and discusses the appropriate evaluation and management of wandering spleen.

(21)

MUTATIONS IN THE GBA1 GENE IN SPANISH POPULATION WITH PARKINSON DISEASE AND PLASMA MIRNAS

Gervas-Arruga J1,2,3*, Alfonso P1,2,3, Garcia-Rodriguez B4 , Capablo JL5, Pocovi M6, Giraldo P1,2,3

1 Centro de Investigación Biomédica en Red de Enfermedades Raras (CIBERER), Zaragoza, Spain
2 Instituto de Investigación Sanitaria Aragón (IISAragón), Zaragoza, Spain
3 Fundación Española para el Estudio y la Terapéutica de la Enfermedad de Gaucher y otras Lisosomales (FEETEG), Zaragoza, Spain
4 Biochemistry department, Miguel Servet University Hospital, Zaragoza, Spain
5 Neurology department, Miguel Servet University Hospital, Zaragoza, Spain
6 Biochemistry; Cellular and Molecular Biology department, Zaragoza University, Zaragoza, Spain

*Corresponding author :gervas.j@gmail.com

The link between Parkinson disease (PD) and Gaucher disease (GD) related to mutations in GBA1 is established. Recently information about additional roles of miRNAs in neurodegenerative processes had been described. The objective of this study is to present the incidence of GBA1 mutations in a cohort of PD patients in our region and compare the profile of some miRNAs in patient plasma samples. The study included 112 PD and 109 unrelated controls. Complete sequencing of GBA1 was performed and plasma miRNAs were quantified using TaqMan miRNA RT-PCR and digital droplet PCR, in 10 controls, 10 PD patients and 10 PD + GBA1 variant carrier patients. In seven PD (6.3%) and four controls (3.7%) GBA1 variants were detected in heterozigosity: in PD group, c.(203)A>G, E326K, T369M (2) and L444P(3) and in controls, N370S (2), c.(203)A>G and E326K. PD patients with GBA1 variants were younger than non-carriers (age at onset: 52.6 ± 12.77 years vs 60.2 ± 10.20 years; P=0.065), specifically, the carriers of L444P mutation (40.5 ± 4.95 years; P= 0.031). We found significant differences in miRNA-365-1 expression between PD + GBA1 variant carriers and PD patients (p=0.038) and between control and PD groups (p=0.006). This study reinforces the role of the GBA1 gene as a susceptibility gene for PD, and that GBA1 gene variants are associated with earlier age at PD onset. The miRNA expression changes in plasma are increasingly being recognized not only as regulators of developmental processes but also contributors.

This work is partially granted by FEETEG

(22)

GENETIC STUDIES ON A PORTUGUESE PARKINSON DISEASE PATIENT COHORT

Gabriel Miltenberger-Miltenyi, Leonor Guedes, Tiago Soares, Marcos Gomes, Joaquim Ferreira, Tiago Outeiro.

Instituto de Medicina Molecular
Faculdade de Medicina
Universidade de Lisboa

Corresponding author: gmiltenyi@medicina.ulisboa.pt

Introduction: Genetic studies of Parkinson's disease (PD) have revealed a number of loci conferring high or lower risk for PD. However, the relationships between the identified genes and the relevant pathways are still not fully understood. Mutations in GBA1 were recognized as one of the most prevalent risk factors for sporadic and familial parkinsonism. Deficiency in GBA1, which encodes the lysosomal enzyme glucocerebrosidase, gives rise to Gaucher disease, the most common lysosomal storage disorder which manifests with variable phenotypic abnormalities. Methods: 415 PD patients of our center were clinically and genetically characterized. Mutation screening was carried out for the LRRK2 gene and for the GBA1 gene. Subsequently, we analyzed 66 patients divided in three groups (19 LRRK2 positive, 37 GBA1 positive and 10 non-mutated) for the Chitotriosidase enzyme activity in serum. 19 of the 415 patients carried mutation in the LRRK2 gene. We found three different variants, of which the most common mutation p.Gly2019Ser was found in 16 patients. 37/415 patients showed exonic variants in the GBA1 gene (8.9%). Results: We found 16 different GBA1 mutations and describe a novel mutation. One patient had mutations in homozygous state and two patients carried compound heterozygous mutations. No patients shared mutations in both LRRK2 and GBA1 genes. The chitotriosidase enzyme activity did not show any significant changes in the LRRK2 positive or the GBA1 positive group when compared with the non-mutated patients. The clinical characteristics, e.g. age-at-onset of the symptoms, were comparable in the different groups. Conclusion: We analyzed the frequency of LRRK2 and GBA1 mutation carriers in a large number of Portuguese PD patients. We described 39 pathogenic GBA1 mutations, including a novel mutation, as potential risk factors for PD.

(23)

EXAMINING THE RISK OF PARKINSONISM IN SIX SIBLING PAIRS WITH GAUCHER DISEASE

Grisel Lopez[1], MD, Gianina Monestime[1], BS, MPH, Catherine Groden[1], RN, NP, Edythe Wiggs[1], PhD, Nahid Tayebi[1], PhD, Ari Zimran[2], MD, and Ellen Sidransky[1]*, MD

[1]*Medical Genetics Branch, NHGRI, NIH, Bethesda MD, USA*

[2]*Gaucher Clinic Jerusalem, Israel*

* sidranse@mail.nih.gov

Background: While the association between mutations in the gene encoding glucocerebrosidase (GBA1) and parkinsonism is well established, most GBA1 mutation carriers do not develop parkinsonian features. In Parkinson disease (PD), by the time patients present with symptoms, more than 50% of dopaminergic neurons in their substantia nigra have been lost. Identifying patients at highest risk for developing PD is critical to impact patients' prognoses and quality of life.

Methods: We studied six sibling pairs with Gaucher disease longitudinally at the National Institutes of Health Clinical Center. In each discordant sibling pair (DSP), both siblings had the same genotype in GBA1, but only one sibling was clinically diagnosed with PD. In all 12, neurological evaluations, family pedigrees, neuropsychological testing and olfactory testing were performed and motor and non-motor symptoms of PD were assessed. Medical information such as age at onset, genotype, clinical presentation, family history, and current medications were also collected. DNA and RNA samples were banked.

Results: Five genotypes were detected; N370S/N370S (in two pairs), N370S/L444P, N370S/V394L, c.84insG/N370S and N370S/IVS2+1. General severity of Gaucher disease was determined based on the degree of skeletal involvement, hematological abnormalities, and organomegaly. In four of the six pairs, the sibling with PD was younger and had milder Gaucher symptoms.

Conclusions: Continued longitudinal evaluations and recruitment of additional DSPs will help to power this study and elucidate factors contributing to parkinsonism in these individuals. Genetic studies will explore potential genetic modifiers or environmental factors that may diminish or augment risk.

(24)

GAUCHER DIEASE TYPE 3 IN SPAIN. OUTCOMES AND CHARACTERIZATION

Marcio M Andrade-Campos[a], JL Capablo[b], I. Garcia[c], O. Salamero[d], P. Martinez-Odriozola[e], JA Mendez[f], F. Garcia-Bragado[g], C. Calvo[h], E. Luño[i], I. Sancho-Val[k], H. Cano[l], J, JM Hernandez-Rivas[m], D. Lorenzo[n], M. Lopez-Dupla[o], V. Callao[p], P. Giraldo[a]

[a]IIS Aragon. CIBER enfermedades Raras, Zaragoza, Spain; [b]Neurology Department. Miguel Servet University Hospital, [c]Children Miguel Servet University Hospital, Zaragoza; [d]Vall d'Hebron University Hospital, Barcelona; [e]Basurto University Hospital, Bilbao;[f]Ourense Hospital, Ourense; [g]Josep Trueta Hospital, Girona;[h]San Jorge Hospital, Huesca; [i]Cabueñes Hospital, Asturias;[k]Alcañiz Hospital, Alcañiz; [l]Arcos del Mar Menor University Hospital, Murcia; [m]Salamanca University Hospital, Salamanca;nComplejo Hospitalario, Burgos; [o]Joan XXIII Hospital, Tarragona; [p]Arnau de Vilanova Hospital, Lleida, Spain

Corresponding author: marcioandrade_hn@yahoo.com

Type 3 Gaucher disease (GD3) is the less common presentation of the disease and a continue challenge for physicians. The onset age usually occurs in the first 10 years of age, with wide interindividual variations, making diagnosis and therapy unmet needs. Here we summarize the experience in 19 GD3 patients, the largest Spanish cohort.

Median follow-up was 15.7 years (.035-41.4 years), males (15, 78.9%); mean age at diagnosis 2.7 years (0.1-10), 13(84.2%) patients were diagnosed bellow 5 years old. Genotype distribution: L444P/L444P: 7(36.8%), L444P/D409H: 2(10.5%), D409H/D409H: 1(5.3%), L444P/other: 3(15.8%), Other/other: 6(31.6%). One patient was diagnosed during familiar study; splenomegaly was part of the primary GD manifestation in 16 (84.2%) cases, mean 9.25 (0.0-22.0) cm bellow left rib costal. Twelve (63.2%) cases presented anemia (mean 10.8 g/dL, range: 6.9-17.0) and 42.3% thrombocytopenia (mean 116 x109/mL; 21-254), 3 cases with <50 x106. Mean chitotriosidase: 8258.2 (871-22251). GD3 manifestations appear at mean 7.9 y.o. (0.9-44), initial manifestation was: oculomotor apraxia (10), myoclonic epilepsy (3), hypotonic and spasms in extremities (2), tremor and gait problems (2), one patients developed kyphosis (L444P homozygous), one cardiac valve calcification (D409H homozygous) and one patient (L444P homozygous) are asymptomatic. All, except 2 patients started ERT at a mean 3.5 years after diagnosis, alglucerase and imiglucerase were the most common enzyme used (15 cases, 88.2%) and 6 (35.3%)patients underwent combined therapy with miglustat. Response to therapy was variable, 3 patients had die; but improvements were recorded during therapy.

This work has been partially granted by FEETEG

(25)

NEUROLOGICAL MANIFESTATIONS AND COURSE OF TYPE 1 AND TYPE 3 (NORRBOTTNIAN) GAUCHER DISEASE IN SWEDEN

Machaczka M[1,2]*, Kämpe Björkvall C[3], Paucar Arce M[4], Myhr-Eriksson K[3], Forsgren L[5], Svenningsson P[4]

[1] *Hematology Center, Karolinska University Hospital Huddinge, Stockholm, Sweden*

[2] *Medical Faculty, University of Rzeszow, Rzeszow, Poland*

[3] *Department of Medicine, Sunderby Regional Hospital of Norrbotten County, Luleå, Sweden*

[4] *Department of Clinical Neuroscience, Center for Molecular Medicine, Karolinska Institutet, Stockholm, Sweden*

[5] *Department of Pharmacology and Clinical Neuroscience, Umeå University, Umeå, Sweden*

*Corresponding author: maciej.machaczka@ki.se

Neuronopathic Gaucher disease (GD) type 3 (GD3) is frequent in northern Sweden, whereas patients with GD type 1 (GD1) are found throughout the country. We present neurological manifestations and clinical course in 13 patients with GD1 and 12 patients with Norrbottnian GD3. Methods The patients were evaluated by standardized assessments of neurological symptoms and signs. Every sixth month, the GD3 patients were rated with the modified Severity Scoring Tool. At baseline and at the 3 years follow-up, patients underwent University of Pennsylvania Smell Identification Test, Montreal Cognitive Assessment and Hospital Anxiety and Depression Scale. When clinical signs were present, additional examinations were undertaken. Results Marked clinical heterogeneity was evident in both GD1 and GD3 groups. Most patients with GD3 have abnormalities of horizontal gaze, ataxia and focal epilepsy, some also had mild cognitive impairment, anxiety, hyposmia and a hitherto unreported, tic-like hyperkinetic myokymia. Six GD3 patients (all of whom were homoallelic for the L444P mutation in the GBA1 gene), have lived beyond 40 years of age; and none has developed Parkinsonism. In contrast, two of the GD1 patients suffer from Parkinsonism. Mild to complete hyposmia was present in 6 GD3 patients and 5 GD1 patients. Neither the group of GD1 nor GD3 patients had detectable progression of their neurological manifestations. Conclusions This is the first comprehensive follow-up investigation of neurological manifestations of middle-aged and older Swedish GD1 or Norrbottnian GD3 patients. While the patients are clinically stable over time, we have identified unusual clinical features, discordant phenotypes, and tic-like hyperkinetic myokymia which appears to unique for this disease variant (Norrbottnian) in Sweden.

(26)

A SEVERITY SCORING TOOL FOR NON NEUROLOGICAL MULTISYSTEM INVOLVEMENT OF TYPE 3 GAUCHER DISEASE: AN IMPORTANT STEP TO MOVE FORWARD

Magy Abdelwahab, Neal Weinreb

1 Department of Pediatric Hematology, Cairo University Pediatric Hospital, Cairo, Egypt
2 University Research Foundation for Lysosomal Storage Diseases, USA

Corresponding author: magywahab@yahoo.com

Gaucher disease includes non-neuronopathic Gaucher disease type 1 (GD1) and neuronopathic (GD2 and GD3). Severity scores systems were published by Zimran (1992: all GD) Davies et al (2007: GD3 neurological involvement), Kallish and Kaplan (2013: pediatric GD1). In GD3, non-CNS involvement, more extensive than usually found in pediatric GD1, causes significant morbidity and mortality. Sites of non-neurological GD3 involvement include liver, spleen, bone marrow, lymph nodes, eye, teeth, bone and chest. There is no GD3-specific tool for grading severity of these manifestations. Egyptian GD3 patients typically have hepatosplenomegaly, focal lesions of liver and spleen, blood cytopenias and mesenteric/mediastinal lymphadenopathy, bone involvement (pain, fractures, avascular necrosis, infarcts, osteopenia and Erlenmeyer's flask deformity), pulmonary complications (recurrent infections, obstructive symptoms, bronchiectasis, interstitial lung disease, miliary infiltrates, consolidation, and pulmonary hypertension). Dental complications include abnormalities in dentin structure with cell entrapment, loss of interdigitation in dentinoenamel junctions and fibrosed pulps infiltrated with Gaucher cells. Eye involvement include infiltration of the cornea, conjunctiva, vitreous and retina sometimes with visual impairment. Persistence of these manifestations despite generally adequate ERT indicates severe, refractory disease. We will review the Egyptian GD3 cohort data with the intention of formulating a severity scoring tool for clinical and investigational use that encompasses domains for all multi-systemic, non-CNS manifestations of GD3 patients. Each domain will be scored as mild, moderate or severe (1, 2, 3). The average of the summed domain scores comprises the composite severity score that will be correlated with a Clinical Global Severity Impression Scale and with prognostic outcomes.

(27)

PHENOTYPIC VARIABILITY IN GAUCHER TYPE 3

Eugen Mengel1, Jörg Reinke1, Tatiana Bremova3, Juliane Arndt1, Silvia Naumann1, Susanne Pitz2, Gundula Staatz2 and Julia Hennermann.1

[1] JGU Mainz UMC, Villa metabolica Mainz, Germany
[2] JGU Mainz UMC, Neuroophtalmology and Radiology department Mainz, Germany
[3] German Center for Vertigo and Balance Disorders, Grosshadern Campus, University Hospital Munich, Germany

*Corresponding author: eugen.mengel@unimedizin-mainz.de

Objective: To determine phenotypic variability regarding characteristic disease manifestations and mortality in the multiethnic cohort of Mainz. Methods: 28 patients with Gaucher disease type 3 were followed with a standardizied follow up protocol up to 23 years. The protocol includes detailed neurological examination including epilepsy diagnostics and cognitive testing, assessment of scoliosis, lung function, mSST of Elin Davis, testing of abnormal saccades and motor abnormalities. All patients were treated with ERT, usually with a dose 60U/kg/biweekly. Results: 7 patients deceased during follow up in the age range between 1 and 19 years. 2/7 because of visceral complications, 2/7 caused by laryngospasm because of progressive bulbar signs and 3 because of progressive myoclonic epilepsy. Only the patients with death caused by visceral complications were homocygous for L444P. Noticeablely patients homocygous for L444P were highly variable regarding neurological symptoms like ataxia and dystonia. IQ varies from 59 – 131. Turkish and Palestinian patients had worser IQ and scoliosis than German and middle east patients. Those patients from German and middle east origin had also a later disease manifestation up to the age of 25 years. Restrictive lung function abnormalities were found in only 3/28 patients. Abnormal saccades were seen in all patients. 3 patients with adult onset had maximal saccades velocity near normal, but gain and voluntary saccades were clearly abnormal. Mesenterial lymphadenopathy was diagnosed in 3 patients, 2 of them had also protein loosing enteropathy. Conclusion: IQ, neurological functions and degree of scoliosis were highly variable. Patients homocygous for L444P with German or middle east back ground had better IQ, less scoliosis and less movement restrictions than patients with Turkish or Palestinian background. Causes of death reflect the whole spectrum of important disease manifestation, in particular severe visceral manifestation, bulbar dysfunction and progressive myoclonic epilepsy.

(28)

LIVER CIRRHOSIS AND MESENTERIC LYMPHADENOPATHY: AN ASSOCIATION OR AN INDICATION OF DISEASE SEVERITY IN 3 GAUCHER DISEASE EGYPTIAN CHILDREN

Magy Abdelwahab[1], Neal Weinreb[2]

[1] *Department of Pediatric Hematology,Cairo University Pediatric Hospital, Cairo, Egypt*
[2] *University Research Foundation for Lysosomal Storage Diseases, USA*

Corresponding author: magywahab@yahoo.com

Untreated adults with type 1 Gaucher disease (GD1) may develop hepatic fibrosis, cirrhosis and portal hypertension. Cirrhosis is rare in children (a few reports in GD1; none in GD3). We describe 3 non-splenectomized children (2-GD3, 1-GD1; 2 males; current age 7-10y) with cirrhosis and mesenteric and mediastinal lymphadenopathy. All had proven glucocerebrosidase deficiency. Patient 1 (P1) was born with jaundice, hepatosplenomegaly and umbilical bleeding. His initial diagnosis was progressive familial intrahepatic cholestasis due to bile synthesis defect. At 1.5y, P1 developed cirrhosis with esophageal and gastric varices. By age 5, he had supranuclear gaze palsy and behavioral problems and was diagnosed with GD3. A younger sister was diagnosed with GD3 at 16 months with splenomegaly, supranuclear gaze palsy and bulbar dysfunction. P1 and his sister receive imiglucerse 30 U/kg/2W despite which, both have mesenteric lymphadenopathy.

Patient 2 (P2) exhibited delayed motor development and hepatosplenomegaly at 1.5y. She was diagnosed as GD1 and started imiglucerase (60 units/kg/2W) with regression of organomegaly .After 2.5y, hepatosplenomegaly recurred with cirrhosis, ascites, mesenteric and mediastinal lymphadenopathy.

Patient 3(P3, L444P/L444P) presented at 14 months with hepatosplenomegaly, growth delay, anemia, thrombocytopenia and stridor. He received imiglucerase (60U/kg/2W) with improvement in growth, CBC and organomegaly. However, after 3y, he deteriorated with supranuclear gaze palsy, behavioral problems, kyphoscoliosis, lung infections and focal seizures. Hepatosplenomegaly worsened accompanied by abdominal pain, cirrhosis, varices and mesenteric lymphadenopathy.

These severely affected GD patients illustrate that besides CNS manifestations, organs such as liver and lymph nodes may sometimes also be refractory to ERT.

(29)

INCREASED RISK OF HEPATOCELLULAR CARCINOMA (HCC) IN GAUCHER DISEASE: THREE CASE REPORTS: BIOLOGICAL COURSE AND IMPACT ON MONITORING

David Santosa[1], Eugen Mengel[2], Martin Merkel[3], Ute Gebert[4], Dieter Häussinger1, Stephan vom Dahl1*

[1]*Department of Gastroenterology, Hepatology and Infectious Diseases, Heinrich-Heine-University, Düsseldorf, Germany*

[2]*Villa Metabolica, University Children's Hospital, Mainz, Germany*

[3]*Endocrinologicum, Amedes Integrated Diagnostics, Hannover, Germany*

[4]Department of Internal Medicine, St. Franziskus-Hospital, Cologne, Germany

*Corresponding author: David.Santosa@med.uni-duesseldorf.de

Introduction: Besides hematooncologic tumours, GD poses an increased risk for solid tumors, especially in elder patients. In normal individuals, in absence of chronic alcohol exposition, viral hepatitis or HFE-dependent iron storage, the major established risk factor for HCC today is NAFLD (non-alcoholic liver disease). Here, pathophysiological mechanisms like PNPLA3 polymorphisms have been discussed, Aims: We report on the incidence of HCC in a cohort of approx. 250 patients from 4 centers in Germany. Case #1: The diagnosis of GD was made in a non-splenectomized male patient (*1952) at age 50 and ERT with imiglucerase was commenced. Liver cirrhosis was histologically diagnosed in 2007. Multifocal HCC was diagnosed in 2013 and TACE (transarterial chemoembolization) was performed. Fibroscan showed increased liver stiffness (19 kPa-27.7 kPa). The patient deceased in 2015. Case #2: A female patient, *1942, was diagnosed with GD in 1971 after splenectomy. ERT with imiglucerase, later velaglucerase, started in 1993. In 2010, ultrasound revealed a progredient liver mass with elevated AFP (26.1 ng/ml, reference < 8.1 ng/ml) and liver stiffness (Fibroscan: 10.3 kPa). After hemihepatectomy of poorly differentiated multifocal HCC in 2011, tumour progress on the contralateral side occurred, with subsequent death in 2013. Case #3: A male patient, *1984, was diagnosed with GD in 1992 after splenectomy and ERT with imiglucerase was started. In 2012, HCC was diagnosed (AFP 41 ng/ml) and successful liver resection was carried out. Conclusion: GD is a risk factor for the development of HCC, also in non-cirrhotic liver. Splenectomy might contribute to this development. Intense screening (abdominal ultrasound, AFP, Fibroscan) is recommended.

(30)

SPLENIC ARTERY ANEURYSMS, A RARE COMPLICATION OF GAUCHER DISEASE: REPORT OF FOUR CASES

C Serratrice[1]*, T Cox[2, 3], E Morris[3], K Benyousfi[4], O Monnet[5], A Sibert6, W Allaham[6], N Belmatoug[4]

[1.] Department of Internal Medicine and Rehabilitation, University Hospital of Geneva, Geneva, Switzerland [2.] Department of Medicine, University of Cambridge, UK [3.] Lysosomal Disorders Unit, Addenbrooke's Hospital, Cambridge, UK [4.] Reference Centre of Lysosomal Diseases, University Hospital Beaujon, Clichy, France [5.] Department of Radiology, Saint Joseph Foundation, Marseille, France [6.] Department of Radiology, University Hospital Beaujon, Clichy, France

*Corresponding author: Christine.Serratrice@hcuge.ch

Splenic artery aneurysms (SAAs) account for more than half of all visceral aneurysms and are the third most frequent intra-abdominal aneurysms. Small SAAs (≤ 2cm) are usually asymptomatic, and giant SAAs (≥5cm) more frequently symptomatic, can result in life-threatening complications. Hypertension, atherosclerosis, cirrhosis, portal hypertension, liver transplantation, female sex (with pregnancy and multiparity) are known etiological factors. Although splenomegaly in Gaucher disease (GD) has been associated with SAAs, bibliographic analysis identified only a single report of this complication1. Here we report four patients with giant or life-threatening SAAs in GD: 2 males and 2 females, aged 10, 19, 30 and 33 years at diagnosis of GD. With GBA1 genotypes: N370S/N370S, N370S/84GG, N370S/c1098_1099insA, N370S/D218A, patients had been classified as GD1. The diagnosis of SAA was made between 12 and 36 years after diagnosis of GD1 (fatal rupture was diagnosed at post-mortem in one). Two patients were already treated by enzyme replacement therapy (ERT) when SAA was diagnosed and the fatal case had also received ERT, but intermittently. One patient has been treated after diagnosis. Follow-up: One patient died with catastrophic aneurysmal rupture; portal hypertension with varices was present. Another patient refused intervention despite a 3.3 cm diameter, symptomatic aneurysm which has been monitored for 8 years uneventfully. One patient had successful splenic artery embolization; another was splenectomized electively. Conclusion: SAAs is a rare complication of GD but can lead to fatal rupture. In GD, the aneurysms can occur independently of portal hypertension as illustrated by three of the four cases reported.

1. Colovic R. Splenic artery aneurysm in a patient with Gaucher disease. Srp Arh Celok Lek, 1989; 117(1-2):107-113

(31)

LYMPADENOPATHY IN GAUCHER DISEASE IS ASSOCIATED WITH TINGIBLE BODY MACROPHAGES OF M2 PHENOTYPE

Ozlem Goker-Alpan, MD[1, 2] *, Renuka Pudi Limgala, PhD[1,2], Erk Changsila,BS1, Chadima Iaonou, BS1,2, Ariel Badger, MS1, and Margarita Ivanova,PhD[1,2,]

[1]*Department of Translational Medicine, Lysosomal and Rare Disorders Research and Treatment Center, 11212 Waples Mill Rd. #103, Fairfax, VA, 22030 USA*
[2] *O&O Alpan, LLC, 11212 Waples Mill Rd. #100, Fairfax, VA, 22030 USA*

*corresponding author: ogokeralpan@oandoalpan.com

In Gaucher disease (GD), the deficiency of the lysosomal enzyme acid b-glucosidase (GCase) and the accumulation of substrate in macrophages lead to immune activation. There are multiple comorbidities in GD that may be related to immune dysregulation: development of lymphadenopathy, multiple myeloma (MM) and lymphomas that are observed in increased frequency in GD. Giant mesenteric or mediastinal lymphadenopathy mimicking lymphoma has been reported in GD pediatric cases. The mechanisms leading to lymph node enlargement and development of tumors are not known.

We investigated the role of the immune activation in the development of lymphadenopathy in GD and the contribution of the activated macrophages to the tumor progression.

While on enzyme replacement therapy, a patient with GD (N370S/RecNciI) and monoclonal gammopathy of undetermined significance, presented with a large submandibular lymphadenopathy. Reactive follicular hyperplasia with intrafollicular monotypic plasma cells (IgG kappa), was demonstrated on biopsy with variable sized follicles in polarized germinal centers with macrophage infiltration. Staining with macrophage (M2) marker and Ki67, a marker of proliferative activity, indicated that the lesion was composed of active proangiogenic macrophages. MM malignancy of plasma cells was ruled out, the immunophenotyping of peripheral blood showed clonal B-cell proliferation, and a reduced number of circulating marginal zone memory B-cells.

M2 macrophages promote tumor formation through cell-to-cell interaction by differentiating into tumor-associated macrophages and promote vascularization, formation of lymphatic vessels. This data highlights the mechanisms of lymphadenopathy in GD, but also may bridge the gap between the cell types interplaying in the development of B cell malignancies.

(32)

OPTIMAL RESPONSE IN GAUCHER DISEASE IN REPLACEMENT THERAPY AND CONCOMITANT CHRONIC MYELOID LEUKEMIA

MS Noya[2], P Irun[1], M Andrade[1], MF Lopez-Fernandez[2], P Giraldo[1]

1.Traslational Research Unit IIS Aragon CIBER enfermedades raras CIBERER, Zaragoza, Spain.
2. Department Hematology, CHUAC A Coruña, Spain.

Corresponding author: giraldocastellano@gmail.com

Association of Gaucher disease (GD) with haematological malignancies has been reported. Only 2 cases of chronic myeloid leukemia(CML) in GD have been published and both previous to imatinib therapy. A 55 years old woman diagnosed in 2005 as type-1 GD (N370S/L444P) following pathologic fracture, pancytopenia and hepatosplenomegaly with spleen nodes. MRI: Diffuse infiltration in the axial skeleton. β-glucocerebrosidase activity: 0.93 nM/mgprot.h (12%). Chitotriosidase: 16608 nM/m. CCL18 / PARC:1969 ng/ml. She started ERT(Imiglucerase) in 2006 (mean dose 40 U/Kg) and reached complete response. In May 2015 she continues asymptomatic, but presents leukocytosis (23.43x109/L) with myeloid precursors and numerous basophiles, without anemia or thrombocytopenia and increase of ALT, AST (<2N), GGT (9N) and LDH:(4,5N). Bone marrow aspirate: granulocytic hiperplasia and Gaucher cells. A Ph'+ t(9; 22), BCR-ABL p210+ was detected according diagnosis CML in chronic phase, Sokal 0.7, and started therapy with imatinib 400 mg together with ERT. She developed generalized itchy rash and increase of liver enzymes (>2N) and need to reduce the dose of Imatinib the first 3 months. Later reintroduce dose of Imatinib 400 mg/d and at 6th month, she was in CML optimal response: BCR ABL1 0.3% and CCyR was confirmed. At 9th month a major molecular response (0.18%) is reached, without toxicity. This is the first case reported of Gaucher disease and CML.treated with Imiglucerase and Imatinib. Some studies implicate glucosylceramide synthase in the resistance of imatinib-induced apoptosis in CML cells via ceramide accumulation. In this case, the patient achieved optimal response for both entities at this moment.

(33)

MYELODYSPLASTIC SYNDROMES IN TYPE 1 GAUCHER DISEASE: DIAGNOSTIC AND TREATMENT CHALLENGES

Hanna Rosenbaum

Hematology day care unit, the center for consultant medicine, Clalit services, Nazareth, Israel.

Corresponding author:roseerlich@gmail.com

Pancytopenia in Gaucher disease(GD)is attributed to the infiltration of bone marrow with Gaucher cells, fibrosis and hypersplenism. The concurrence of GD and myelodysplastic syndrome (MDS) was reported in anecdotal cases. We report four GD1 patients with cytopenias due to myelodysplastic syndromes.

Patient 1: diagnosed at age 67 with GD1.Bone marrow aspirate (BMA) and biopsy (BMB) performed because of persistent cytopenia showed aggregates of Gaucher cells ,myelodysplasia and dyserythropoesis with a normal karyotype.

Patient 2: a 42 y.o. female presented with refractory anemia. A BMA and BMB revealed a massive Gaucher cells infiltrate with ring sideroblasts.The patient responded to recombinant erythropoietin.

Patient 3 a 69 y. o. female diagnosed in childhood with GD1 and.ERT was instituted because of bone complications .Following 22 years of ERT with trilineage dysplasia in the BMA and 15% CD 117 positive cells. Only few Gaucher cells were seen. in. These findings are compatible with refractory anemia with excess of blasts.The patient died following transformation to AML.

Patient 4 diagnosed at age 59 with GD.Six years later Waldenstrom macroglobulinemia was diagnosed & hyper viscosity.BMB revealed massive GD infiltration with areas of small IGM K positive lymphocytes. Complete remission was achieved foolowing CHOP+ Rituxumab. Four years later a nephrectomy was performed due to renal cell carcinoma. MDSwas diagnosed, requiring frequent red cells infusions. He died from metastatic renal cell carcinoma.

Concurrence of myelodysplastic syndrome and Gaucher is not frequent it should be considered in GD patients with persistent cytopenias.

(34)

OCCURENCE OF NEOPLASTIC DISEASES AND OF PRENEOPLASTIC CONDITIONS, AMONG GREEK PATIENTS WITH GAUCHER DISEASE

Constantina Repa, Theodore Marinakis, Alexandra Kourakli, Dimitrios Zafeiriou, Panagiotis Tsaftaridis, Helen Michelakakis, Veroniki Konminaka, Vassiliki Pappa, Paraskevi Roussou, Efthimia Vlachaki, Maria Kotsopoulou, Alexandros Makis, Evdokia Mandala, Ersi Voskaridou, Nikolaos Anagnostopoulos, Argiris Symeonidis* for the ***Hellenic (Greek) National Gaucher Disease Study Group***.

*Corresponding author: argiris.symeonidis@yahoo.gr

We aimed to investigate the incidence, type and course of the neoplastic diseases, occuring in the Greek Gaucher Disease (GD) patient population. Patient-files and databases including 79 patients with GD, followed-up in 12 Hematology Departments, reporting data to the Hellenic (Greek) National Gaucher Disease Study Group, were reviewed and patients with neoplastic diseases were retrieved and analyzed.

There were 37 male and 42 female patients, with a median age of 27 years at diagnosis of GD, including 12 pediatric patients. Seventy-seven patients had type I GD and the remaining 3 had type III GD. A definite diagnosis of a neoplastic disease was done in 8 patients, and two additional patients have been diagnosed with a Monoclonal Gammopathy of Undetermined Significance (MGUS). The diagnosed neoplastic diseases were colon cancer in 3 patients, lung cancer, pancreatic cancer, multiple myeloma, Hodgkin's lymphoma and high-grade B-cell non-Hodgkin's lymphoma of the Central Nervous System in one patient each. Two of the three patients, who presented a lymphoproliferative disease, but none of the 5 with a solid tumor had been previously splenectomized. Of interest two of the 3 patients with colon cancer exhibited the N370S/D409H disease genotype, which was not found in any other of the remaining patients. Five of the 8 patients with a neoplastic disease and the 2 with the MGUS are alive and routinely followed-up.

In view of the high incidence of neoplastic diseases, occuring in patients with GD, the significance of high level of alertness and clinical suspicion is stressed

(35)

STUDY OF COMORBIDITIES IN A SPANISH COHORT OF GAUCHER DISEASE TYPE 1 PATIENTS

J. Pérez-López1*, V. Giner Galvañ2, MA. Torralba Cabeza[3], RJ Núñez[4], M. Moltó Abad1, Inmaculada Roig[5], L. Vicente[6], A. Luaña Galan[7], E. Cabezudo[8], X. Solanich[9], B. Roig Espert[10], E. Patera[11], ML. Lozano Almela[12], R. Hurtado García[13], M. Reyes[14], N. Revilla[12], S. Novelli[15], J. Esteve[16].

[1] Inborn Errors of Metabolism in Adults. Unit of Rare Diseases, H. Vall d'Hebron (Barcelona) [2] Service of Internal Medicine. H. Virgen de los Lirios (Alcoy)[3] Service of Internal Medicine. H. Lozano Blesa (Zaragoza) [4] Service of Hematology. H. Virgen del Rocío (Sevilla) [5] Service of Hematology. Consorci Hospitalari Parc Taulí (Sabadell) [6] Service of Hematology. H. de Terrassa (Terrassa) [7] Service of Hematology. H. Arnau de Vilanova (Lleida)[8] Service of Hematology. H. Sant Joan de Deu (Manresa) [9] Service of Internal Medicine. H. de Bellvitge (Hospitalet de Llobregat) 1[0] Service of Internal Medicine. H. de Manises (Valencia) [11] Service of Internal Medicine. H. Sant Jaume (Calella)[12] Service of Hematology. H. Morales Meseguer (Murcia) [13] Service of Internal Medicine. H. Vega Baja (Orihuela) [14] Service of Hematology. H. Lozano Blesa (Zaragoza)[15] Service of Hematology. H. Santa Creu i Sant Pau Pau (Barcelona) [16] Service of Hematology. H. Clínic (Barcelona)

*Corresponding author: jordpere@vhebron.net

OBJECTIVES: Specific treatment in Gaucher type 1 (GD1) has allowed a lifespan similar to general population. Aging process results in an increase frequency of comorbidities and poly-pharmacy, so that it may affect the outcomes of these patients. We studied the comorbidities and poly-pharmacy in a Spanish cohort of GD1 patients. METHODS: 37 GD1 were enrolled. Disease complications (including Severity Score Index [SSI]), simultaneous comorbidities (including Charlson Index [ChS]), as well as concomitant drugs were evaluated. Mean and standard deviation were measured in quantitative variables. RESULTS: 20 women and 17 men were studied, with mean of age 46.8(17.4) years. SSI was 7.4(3.7). All patients were under treatment for GD, whose length of treatment was 11.1(6) years. Seventeen (46%) patients presented bone complications (5 osteonecrosis, 5 osteoporosis, 4 hip replacement and 3 bone fractures), 8(20.5%) were splenectomized, 7(17.9%) monoclonal gammapathy, 1(2.7%) Parkison disease and none with cancer. ChS was 4(1.4), including dyslipemia in 6(16.2%) patients, diabetes mellitus in 2(5.4%). Fourteen (37.8) patients showed poly-pharmacy with a mean of 4.5(2.8) drug per patient, including psychotropic drugs in 11(30%), calcium and D vitamin supplements in 10(27%), pain kills in 7(17.9%), statins in 6(16.2%), antiarrhytmics in 2(5.4%), and biphosphonates, antiagregants and antiparkinsonians in 1(2.7%) respectively. CONCLUSIONS: Our data suggests that bone involvement leads to the most important disease complications in GD1 (46% of patients), and that poly-pharmacy is very frequent among these patients (37.8%). Psychotropic drugs, calcium/vitamin D supplements and statins are the most common concomitant medication.

(36)

THE APPEARANCE OF INTRAOCULAR LESIONS IN GAUCHER DISEASE TYPE 3 DESPITE LONG-TERM GLUCOCEREBROSIDASE REPLACEMENT THERAPY

Sawicka-Gutaj N[1], Machaczka M[2,3]*, Kulinska-Niedziela I[4], Bernardczyk-Meller J[5], Gutaj P[1], Sowinski J[1], Ruchala M[1]

[1] Department of Endocrinology, Metabolism and Internal Medicine, Poznan University of Medical Sciences, Poznan, Poland
[2] Hematology Center, Karolinska University Hospital Huddinge, Stockholm, Sweden
[3] Medical Faculty, University of Rzeszow, Rzeszow, Poland
[4] Heliodor Swiecicki Clinical Hospital, Ophthalmology Outpatient Clinic, Poznan University of Medical Sciences, Poznan, Poland
[5] Department of Ophthalmology, NZOZ "OCU SERVICE", Poznan, Poland

*Corresponding author: maciej.machaczka@ki.se

Gaucher disease (GD) is an autosomal recessive lipid storage disorder caused by deficient lysosomal enzyme glucocerebrosidase activity. The presence of central nervous system disease is a hallmark of the neuronopathic forms of GD (types 2 and 3). Intraocular lesions (e.g., corneal clouding, retinal lesions, and vitreous opacities) have been infrequently reported in GD type 3 (GD3). Moreover, there are virtually no published data on the occurrence and natural course of intraocular lesions in GD3 patients treated with enzyme replacement therapy (ERT).

We present the case of a 26-year-old Polish male with L444P homozygous GD3 (mutation c.1448T > C in the GBA1 gene) who developed fundus lesions despite 10 years of ERT. At the age of 23 years, a spectral domain optical coherence tomography (OCT) examination was performed which disclosed the presence of discrete lesions located preretinally, intraretinally in the nerve fiber layer, and in the vitreous body. A 3-year follow-up OCT examination has not shown any significant progression of the fundus lesions.

To the best of our knowledge, this is the first published report describing the occurrence of newly identified retinal and preretinal lesions occurring during long-term ERT in GD3. We recommend that a careful ophthalmic assessment, including a dilated fundus examination, should be included as part of annual follow-up in patients with GD3. Further studies are needed to understand the nature and clinical course of these changes and whether or not these intraocular findings have any predictive value in the context of neurologic and skeletal progression in GD3.

(37)

IMIGLUCERASE TREATMENT ASSOCIATED WITH REDUCTION OF BONE CLAIMS IN GAUCHER PATIENTS: ANALYSIS OF U.S. CLAIMS DATA

Luba Nalysnyk[1]*, Martin Selzer[1], Alaa Hamed[1], Neal Weinreb[2]

[1]Sanofi Genzyme, Cambridge, MA

[2]*University Research Foundation for Lysosomal Storage Diseases, Coral Spring, FL*

**Corresponding* author: lubomyra.nalysnyk@genzyme.com

Background: Bone involvement is a frequent and debilitating complication of Gaucher disease type 1 (GD1) and can significantly decrease patients' quality of life due to pain, disability, and need for orthopedic intervention. OBJECTIVE: To examine the impact of treatment with imiglucerase on bone claims in Gaucher patients before and after initiation of treatment using US administrative claims data. Methods: A retrospective study of medical claims data was conducted using Optum's Clinformatics Datamart database. Gaucher patients treated with imiglucerase between January 1, 1997 and June 30, 2013 were identified. Patients with ≥3 months continuous insurance coverage prior to their first treatment with imiglucerase (index date) and ≥3 months follow-up post-index date were eligible. The percentage of patients with bone claims and the average bone claims per patient were compared pre- and post-treatment. Results: 107 GD patients were evaluated; 59 (55%) were male, 85 (79%) were ≥18 years. Overall, 31 (29%) patients reported bone claims within 3 months of beginning imiglucerase. In all 3-month intervals during follow-up, the number of patients with bone claims was significantly reduced when compared to the pre-treatment period. When data were analyzed separately for adults and children, similar trends were observed with more pronounced results among children. Conclusion: A reduction in the number of bone claims and number of patients with bone claims was observed after initiation of treatment with imiglucerase in Gaucher patients in the US. These results suggest that imiglucerase treatment is associated with a reduction of bone complaints in both adult and pediatric patients.

Key words: Gaucher disease, bone disease, claims analysis Study sponsored by Sanofi Genzyme.

(38)

THREE-DIMENSIONAL PRINTING OF BONE LESSIONS IN GAUCHER DISEASE

A Lacleriga[1], Luis Sarria[2], Alfaro-Santafé JV[3], Mercedes Roca[4], Pilar Giraldo[5]

Traumatology Department. Miguel Servet University Hospital. Zaragoza
Radiodiagnostic Department. Miguel Servet University Hospital. Zaragoza
Podoactiva. Zaragoza
Radiological Centre. Zaragoza
Traslational Research Unit. IIS Aragón. CIBER enfermedades raras CIBERER. Zaragoza

Corresponding author : laclerigaa@me.com

The 3-dimensional(3D) printed images have been recently introduced into the aid plan surgery process. Biotechnology research based on 3D printing principles is being applied even to biological material with the goal of creating functional tissues and organs. One of the early adopters of 3D printing technology was the orthopedic field. Applying this technique to diagnosis of bone lesions permits us to obtain more accurate information about the rendered-volume to prepare their surgical reconstruction. Nowadays in patients with complex fractures or deformities axial3D is an effective tool for providing an anatomic scale piece with advantage on conventional CT/MRI.

The advanced bone complications in Gaucher disease lead to severe and complex joint problems and frequently need joint replacements. The application of computed tomography (CT) with 3-dimensional CT could improve the quality of bone assessment and custom prosthesis.

We have applied this technique to study bone deformities in some Gaucher disease patients with severe bone lesions in the shoulder, hip and knee. Plain X-ray records were reviewed, and CT scan data retrieved. CT scans were included if they were performed before any surgical treatment and were of sufficient quality to enable fine resolution 3-dimensional reconstruction. The images obtained could be used to model appropriate prosthesis in individual subjects.

In our experience 3D printed models offer an innovative approach to studying bone lesions in GD and also for preoperative planning of complex surgeries, the creation of custom prosthesis, and in the education and training of physicians.

This work has been partially granted by FEETEG.

(39)

THE IMPACT OF CHITOTRIOSIDASE ON THE OCCURRENCE OF BONE EVENTS IN GAUCHER DISEASE TREATED USING ENZYME REPLACEMENT THERAPY: THE CONTRIBUTION OF JOINT MODELS.

Marie Vigan[1,2,] Nadia Belmatoug[3], Marc G Berger[4,5,6,] France Mentré[1,2,] Jérôme Stirnemann[7] and all members of the Gaucher Disease Treatment Evaluation Committee

[1]INSERM, IAME, UMR 1137, F-75018 Paris, France 2 Université Paris Diderot, IAME, UMR 1137, Sorbonne Paris Cité, F-75018 Paris, France 3Médecine Interne et CRML, Hôpital Beaujon, AP-HP, 100 Bd Général Leclerc, 92110 Clichy, France 4Hématologie Biologique, CHU Estaing, 1 place Lucie Aubrac, 63003 Clermont-Ferrand Cedex 1, France 5CRB Auvergne, CHU Estaing, 1 place Lucie Aubrac, 63003 Clermont-Ferrand Cedex 1, France 6Hématologie Clinique, CHU Estaing, 1 place Lucie Aubrac, 63003 Clermont-Ferrand Cedex 1, France 7Service de Médecine Interne Générale, Hôpitaux Universitaires de Genève, Rue Gabrielle-Perret-Gentil 4, CH-1211 Genève 14, Suisse

Corresponding author: Marie Vigan
e-mail: marie.vigan@inserm.fr

Object: Bone events (BE: Avascular necrosis, bone infarct or pathological fracture) are important complications of Type 1 Gaucher disease. We aim to analyse the link between longitudinal biomarker data (especially chitotriosidase) and repeated BEs occurring over time.

Methods: We analysed data of 241 patients of the French Gaucher registry, treated with imiglucerase. We used a non-linear mixed effects model to analyse longitudinal repeated chitotriosidase dosages and a frailty model to analyse the occurrence of repeated BEs in Gaucher patients after treatment initiation. We then propose to develop a joint model, capable of analysing together the link between chitotriosidase and the occurrence of repeated BEs occurring over time.

Results: Thirty-seven % of patients had BE during the follow-up. Total number of BEs under ERT was 65, among 45 patients. The occurrence of a BE before treatment increased the risk of another occurrence during treatment. A BE before treatment is associated with double of the concentration of chitotriosidase at the start of treatment and a tripling of the risk of a BE during treatment. A link between chitotriosidase and occurrence of BE has been demonstrated: a five-fold increase in the concentration of chitotriosidase multiplied the risk of a BE by 1.2.

Conclusion: Chitotriosidase will be able to be used more effectively for estimating the risk of a bone event and identifying which patients require more intensive treatment.

(40)

EVALUTAION OF BONE QUALITY IN PATIENTS WITH TYPE 1 GAUCHER DISEASE WITH MICROINDENTATION. PRELIMINARY RESULTS.

Jordi Pérez-López[1], Sabina Herrera[2], Marc Moltó[1], Roberto Güerri-Fernández[2], Elena Cabezudo[3], Silvana Novelli[4], Jordi Esteve[5], Albert Hernández[6], Inmaculada Roig[7], Xavier Solanich[8], Daniel Prieto-Alhambra[9], Xavier Nogués[2], Adolfo Díez-Pérez[2].

[1] Inborn Errors of Metabolism in Adults. Unit of Rare Diseases. Hospital Vall d'Hebron, Barcelona. [2] Internal Medicine, Hospital del Mar, Barcelona. [3] Hematology Department, Hospital de Sant Joan de Déu, Manresa, Barcelona. [4] Hematology Department, Hospital de Sant Pau, Barcelona. [5] Hematology Department, Hospital Clínic, Barcelona. [6] Internal Medicine Department, Hospital Comarcal de Sant Jaume, Calella, Barcelona. [7] Hematology Department, Consorci Hospitalari Parc Taulí, Sabadell, Barcelona. [8] Internal Medicine Department, Hospital de Bellvitge, Barcelona. [9] Oxford NIHR Musculoskeletal Biomedical Research Unit, Nuffield Department of Orthopaedics, Rheumatology, and Musculoskeletal Sciences, University of Oxford, Oxford, United Kingdom

Corresponding author: jordpere@vhebron.net

OBJECTIVES: Over 80% of patients with type 1 Gaucher disease (GD1) shows bone affectation, causing predisposition for bone fractures. Although with significant limitations, the most used techniques for bone assessment are dual energy X-ray absorptiometry (DXA) and magnetic resonance imaging (MRI). However, microindentation has showed a good correlation with bone fractures in patients with osteoporosis. Our goal was to study the bone quality in patients with GD1 using the microindentation and its new measurement parameter called Bone Mineral Strength index (BMSi), as a new and accurate tool for this assessment. METHODS: 14 GD1 patients and 29 controls were enrolled. Clinical and laboratory parameters of all individuals were collected, followed by Bone Mineral Density (BMD) in their column and hip and BMSi in tibia. Impact Bone Microindentation measurements were carried out (Osteoprobe, Active Life Scientific, Santa Barbara, CA, USA) in areas not affected by bone infarcts or necrosis. RESULTS: Comparing GD1 patients with controls, there were no significant differences among patients with GD1 and controls regarding age, gender, height and weight. We found a significant difference in BMSi (81.7(7.78) $p<0.05$) and BMD in column (0.99(0.14) $p<0.05$), but not in BMD in hip (0.76(0.10) $p=0.14$). No differences were found between GD1 patients regarding Severity Score Index (SSI) or treatment.

CONCLUSIONS: Our preliminary results suggest that could become microindentation as a novel and accurate technique for the study of the bone quality in GD1 patients. However, more data is needed to find out its role in clinical practice.

(41)

OCCURRENCE OF ADVERSE BONE EVENTS IN ADULTS AND CHILDREN WITH GAUCHER DISEASE TREATED WITH TALIGLUCERASE ALFA

Michael Wajnrajch[1], Betina Hernandez[1], Li-Jung Tseng[1], Sukirti Bagal[1], Roy Mansfield[2], Mireille Fernet[3]

[1] *Pfizer Inc, New York, USA,*

[2] *Pfizer Inc. Kent, United Kingdom*

[3] *Pfizer Canada, Kirkland, Canada*

Bone manifestations among Gaucher Disease patients are common with acute bone crises occurring in up to 20% of the worldwide population of patients with GD type 1. The objective of this abstract is to summarize the bone events (avascular necrosis, pathological fractures and bone crises) from the taliglucerase alfa clinical studies. A total of 132 subjects, adults and children, were exposed to taliglucerase alfa in phase III clinical trials. In the adult population 57 subjects received at least 24 months of treatment of taliglucerase alfa and 33 subjects received more than 36 months. In the pediatric population 16 children received more than 12 months of exposure to taliglucerase alfa. Twelve bone events were documented (avascular necrosis/osteonecrosis, pathological fractures or bone crises) involving 9 adult patients treated with taliglucerase alfa across the 7 studies of our clinical program. None of the adverse events were considered related to treatment by the investigators. The frequency of adverse clinical bone events during the phase III clinical development of taliglucerase alfa suggests a relatively low occurrence, consistent with the previously documented findings during ERT. This report adds to the understanding of the overall safety profile of taliglucerase alfa in adult and pediatric patients with GD.

Disclosures: This study was sponsored by Pfizer.

(42)

BONE MINERAL DENSITY AND LEAN MUSCLE MASS CHARACTERISTICS IN CHILDREN WITH GAUCHER DISEASE TREATED WITH ENZYME REPLACEMENT THERAPY OR UNTREATED

Liron Dar MD*, Deborah Elstein PhD, Ari Zimran MD, Ehud Lebel MD**

**In partial fulfillment of requirements for an MD degree at the Hebrew University Hadassah School of Medicine*

***Gaucher Clinic and the Department of Orthopedic Surgery, Shaare Zedek Medical Center, affiliated with the Hebrew University Hadassah School of Medicine, Jerusalem Israel*

Presenting & Corresponding author: Ehud Lebel. 972-50-868-5068, lebel@szmc.org.il

Bone Mineral Density (BMD) characteristics are standard indices of risk of osteoporosis in adults with Gaucher disease. Data is limited regarding changes in BMD in pediatric patients. This study evaluated BMD, Lean Muscle Mass (LMM) and Fat Fraction (FF) in treated and untreated children (>3 years) with enzymatically-proven (type 1) Gaucher disease. All underwent Dual-energy X-ray absorptiometry (DXA) as a total-body scan and results for BMD, FF and LMM analyzed. Demographics included age, gender, genetic diagnosis, body dimensions, and treatment status.

Forty-eight patients (24 males; aged 3-16 years) were included: 13 had the genotype N370S/N370S and were not receiving Enzyme Replacement therapy (ERT); the remaining 35 were N370S/other of which 26 were receiving ERT. Treated patient had significantly higher average total-body BMD values ($p=0.005$) and there was a strong positive linear correlation between age and bone and soft tissue values ($R=0.5$-0.7) representing steady growth. Their findings were closely aligned with those of the untreated N370S/N370S patients. The nine untreated N370S/other patients showed slower accumulation of BMD and LMM. Age-adjusted values of BMD were low among all patients, representing lower than normal skeletal dimensions and/or delayed bone-age.

This is a first study to consider components of body composition in children with Gaucher disease. Administration of ERT invariably improved bone-mass and musculature. BMD showed steady accumulation, but was below expected values for age/gender. Recording of BMD and LMM should become part of routine evaluation of children with Gaucher disease: such data would supply substantial support for decision-making regarding ERT and other interventions.

(43)

BONE CRISIS IN SMALL'S FOOT BONES IN PEDIATRIC GD PATIENTS UNDER LONG-TERM ENZYME REPLACEMENT THERAPY

Andrade-Campos Marcio[1,2,] Sancho-Val Ignacio[3], García Inmaculada[4], Roca Mercedes[5], Giraldo Pilar[1,2,6]

[1]Traslational Research Unit. IIS Aragón. [2]CIBER enfermedades raras CIBERER, Zaragoza. [3]Haematology Department. Alcañiz Hospital. [4]Pediatric Hospital Miguel Servet. 5MRI Center, 6FEETEG. Zaragoza. Spain.

Corresponding author: marcioandrade_hn@yahoo.com.

Long-term effect of enzyme replacement therapy (ERT) on skeletal involvement are relate to baseline burden disease and therapy regularity and adherence.

About 30% of child patients had bone crisis before the start of ERT. Only ~2.5% of early treated children without previous bone crisis (BC) present a BC during ERT. These are reported usually in long bones. Here we report 3 cases of BC in small bones in GD patients diagnosed during childhood and under long-term ERT in Spain.

Case 1. Male, diagnosed at 18 months with spleen enlargement, anaemia and thrombocytopenia, genotype [R463C]+[G377S], he started ERT (60 U/Kg) with good outcomes. After 10 years on ERT he developed a bone crisis in right astragalus and calcaneus, the episode was resolved after ERT dose increase.

Case 2. Female, diagnosed at 4 years of GD1, genotype [N370S]+[84GG] due to epistaxis, splenomegaly and thrombocytopenia, She started ERT after diagnosis developing infusion reaction controlled after switch enzyme, and after 13 years under ERT, developed bone crisis in her right calcaneus.

Case 3. Male diagnosed at 3.5 years old, genotype [N370S]+[R130W], due to splenomegaly and started ERT at 60 UI/2 weeks, after 10 years on ERT he developed a bone crisis in rigth tarso. Apparently without a trigger, but during growth period.

Bone crisis usually occurs on long bones epiphysis predominantly in patients without therapy or esplenctomyzed. In small and distant bones as our cases is an uncommon event and would be related to the intensity of physical activity in loading areas.

This work has been partially granted by FEETEG

(44)

ESTIMATING THE UTILITY ASSOCIATED WITH MODE OF TREATMENT ADMINISTRATION IN GAUCHER DISEASE

Luba Nalysnyk[1]*, Paul Swinburn[2], Monica Hadi[2], Alaa Hamed[1]

[1] *Sanofi Genzyme, Cambridge, MA, USA*
[2]*Mapi Group, London, UK*

*Corresponding author: lubomyra.nalysnyk@genzyme.com

Background: As both intravenously-administered imiglucerase and orally-administered eliglustat treatments have demonstrated comparable efficacy in patients with Gaucher disease type 1, questions arise in considering the value of oral versus intravenous treatment in relation to patient's benefit, compliance, preference and quality of life.

Objective: To capture UK societal-based utility values for health states related to treatment mode of administration for Gaucher disease

Methods: Health states were developed on the basis of a review of relevant literature and expert clinical input. Health states were piloted with members of the UK general public to ensure their suitability for use in a valuation exercise. Valuation study population: 100 members of the general public in the UK. Participants were asked to complete a brief socio-demographic form, the EQ-5D and then participate in a visual analog scale (VAS) ranking exercise and Time Trade-Off (TTO) interview.

Results: Mean age of the study participants (n=100) was 35 years, 66% were women. Participants reported high VAS (86.13) and EQ-5D index scores (0.95) indicating very good health status. Data revealed that varying mode of administration had a significant impact on health-related quality of life (HRQoL) with the "oral treatment" state (VAS 70.9; utility 0.85) valued more positively than the "intravenous treatment" state (VAS 49.9; utility 0.73) suggesting reduced burden for patients.

Conclusion: The data demonstrates the HRQL impact of changes in treatment mode of administration. The data suggests that these differences are significant and could potentially have important consequences for patients.

Key words: Gaucher disease, health states, utility Study sponsored by Sanofi Genzyme

(45)

THERAPEUTIC GOALS AND NORMAL CLINICAL VALUES ACHIEVED WITHIN 4 YEARS OF INITIATING VELAGLUCERASE ALFA IN TREATMENT-NAÏVE PATIENTS WITH GAUCHER DISEASE IN PHASE 3 STUDIES

Ari Zimran[1*], Deborah Elstein[2], Derlis E Gonzalez[3], Elena A Lukina[4], Yulin Qin[5], Quinn Dinh[6], Hadhami Ben Turkia[7]

1 Gaucher Clinic, Shaare Zedek Medical Center, affiliated with the Hebrew University-Hadassah Medical School, Jerusalem, Israel 2 Shire Global Medical Affairs, Shire, Zug, Switzerland 3 Instituto Privado de Hematología e Investigación Clínica (IPHIC), Sanatorio Español, Asunción, Paraguay 4 Department of Orphan Diseases, National Research Center for Hematology, Moscow, Russia 5 Biostatistics and Statistical Programming Department, Shire, Lexington, MA, USA 6 Shire Global Medical Affairs, Shire, Waltham, MA, USA 7 Paediatric Department, La Rabta Hospital, Tunis, Tunisia

*Corresponding author: azimran@gmail.com

In a phase 3 extension study (ClinicalTrials.gov number, NCT00635427), velaglucerase alfa was efficacious for long-term treatment in patients with type 1 Gaucher disease (GD). A post hoc analysis was conducted to evaluate efficacy using published therapeutic goals for GD treatment. Thirty-nine patients were treatment-naïve before initiating velaglucerase alfa treatment (ages at baseline, 6-62 years). Patients received every-other-week intravenous infusions (mean dose 56.6 U/kg) for up to 5.8 years. At year 4, of patients whose baseline clinical values were outside the therapeutic goal ranges, all 20 patients had achieved the goal for platelet count (≥100% increase or ≥100-120×109/L depending on baseline value and splenectomy status), all 16 patients achieved the goal for spleen volume (≥50% decrease or ≤8.0 multiples of normal [MN]), 19/20 patients achieved the hemoglobin concentration goal (≥11-12 g/dL depending on age and sex), and 15/16 patients achieved the liver volume goal (≥30% decrease or ≤1.5 MN). In line with skeletal goals, the lumbar spine bone density Z-score increased in 20/23 adult patients (aged ≥18 years). In patients who had abnormal baseline values (compared with healthy individuals, regardless of whether the therapeutic goal was met), normal values were achieved in 24/28 patients for hemoglobin and 12/20 patients for platelets (based on testing laboratory's reference values), in 12/18 patients for spleen volume (normalization considered ≤5.0 MN), in 11/19 non-splenectomized patients for liver volume (≤1.0 MN), and in 10/19 adults for lumbar spine bone density (Z-score ≥-1). Normalization of GD parameters, beyond achieving therapeutic goals, may be achievable with velaglucerase alfa.

(46)

FREQUENCY, DURATION, RELATEDNESS, SEVERITY, AND SERIOUSNESS OF ADVERSE REACTIONS AMONG A POOLED ELIGLUSTAT DATASET OF 393 GAUCHER PATIENTS FROM FOUR CLINICAL TRIALS

M Judith Peterschmitt[1]*, Gerald F Cox[1], Jennifer Ibrahim[1], James Macdougall[2], Lisa H Underhill[1], Sebastiaan JM Gaemers[1]

[1]*Sanofi Genzyme, Cambridge, MA, USA;* [2]*Prometrika, LLC, Cambridge, MA, USA*

Corresponding author: *Judith.Peterschmitt@Genzyme.com

Eliglustat is a first-line oral substrate reduction therapy approved for adults with Gaucher disease type 1 (GD1) who have compatible CYP2D6 metabolizer phenotypes (>90% of patients). To better inform patients about what to expect on eliglustat, clinicians have requested more information about adverse reactions (AEs) associated with eliglustat. We therefore examined AE data from 393 eliglustat-treated GD1 patients in four Sanofi-Genzyme-sponsored clinical trials (NCT00358150; NCT00891202; NCT00943111; NCT01074944) representing 535 patient-years of eliglustat exposure with a mean treatment duration of 1.4 years.

We evaluated the frequency, duration, relatedness, severity, and seriousness of the following AEs listed in the EU Summary of Product Characteristics, shown with the overall incidence by AE and percent of patients in whom the investigator assessed the AE as related to eliglustat: headache (17% overall and 5% related), arthralgia (14% overall and 2% related), diarrhea (10% overall and 4% related), nausea (8% overall and 3% related), fatigue (7% overall and 2% related), abdominal pain (6% overall and 3% related), and flatulence (2% overall and 2% related). Overall, 5% of these AEs were severe and none was serious. By type of AE, 64-88% of patients reported the AE just once. Symptoms associated with GD1 (arthralgia and fatigue) usually lasted >2 weeks. The majority (67%) of all other AEs lasted ≤2 weeks. In summary, the majority of these AEs in eliglustat-treated patients were occasional, non-severe, and (except for AEs related to GD1) short-term.

(47)

ARIMOCLOMOL AS A CLINICAL CANDIDATE FOR TREATMENT OF TYPE 2 AND TYPE 3 GAUCHER DISEASE

Cathrine K. Fog-Tonnesen[1]*, Mingshu Ming[1], Nikolaj H. T. Petersen [1], Claus Bornaes[1], Anders Hinsby[1] and Thomas Kirkegaard[1]

1 Orphazyme ApS, Copenhagen, Denmark

*Corresponding author: cft@orphazyme.com

Gaucher disease (GD) is the most frequent lysosomal storage disorder (LSD) (incidence of 1:40,000 newborns). It is caused by the deficient activity of the lysosomal enzyme acid beta-glucosidase (GBA), caused by autosomal recessive mutations in the GBA gene. Although GD presents as a continuum of disease states, it is clinically divided in visceral (GD1), acute neuronopathic (GD2) and sub-acute neuronopathic (GD3) forms.

So far, there is no effective treatment for the neurological forms of the disease. The aim of this study is therefore to investigate the potential of arimoclomol as a new therapeutic agent for the treatment of GD2/3.

Arimoclomol is a hydroxylamine derivative, belonging to a group of compounds known to have unique properties as co-inducers of the heat shock response (HSR). The cytoprotective actions of the HSR involve central cellular compartments such as the endoplasmic reticulum (ER) and endo-lysosomal system and has been shown, among others, to facilitate protein folding, restore activity to misfolded proteins and protect against lysosomal membrane permeabilization. Arimoclomol may therefore promote mutant GBA folding/trafficking and restore lysosomal function in GD cells. Since arimoclomol has been extensively clinically tested and readily crosses the blood-brain-barrier (BBB), it may present a new therapeutic opportunity for the treatment of neurological Gaucher disease.

(48)

SIGNIFICANT AND CONTINUOUS IMPROVEMENT IN QUANTITATIVE CHEMICAL SHIFT IMAGING (QCSI) IN PATIENTS WITH GAUCHER DISEASE TREATED WITH TALIGLUCERASE ALFA DURING THE EARLY ACCESS PROGRAM

Ari Zimran, MD[1*]; Tama Dinur, PhD[1]; Erik Akkerman, PhD[2]; Carla E. M. Hollak, MD, PhD[2]; Raul Chertkoff, MD[3]; Mario Maas, MD[2]

1 Gaucher Clinic and Genetic Unit, Shaare Zedek Medical Center, Jerusalem, Israel 2University of Amsterdam, Academic Medical Centre, Amsterdam, the Netherlands 3 Protalix BioTherapeutics, Carmiel, Israel

*Corresponding author: azimran@gmail.com

Taliglucerase alfa, the first plant-cell-expressed recombinant therapeutic protein FDA-approved for human use, is the third enzyme replacement therapy (ERT) approved for treating Type 1 Gaucher disease (GD). In trials, efficacy endpoints were organomegaly reduction and hematological parameter improvement; impact on bone was exploratory because of slower skeletal response to other ERTs. Using more sensitive MRI, improvement in bone-marrow infiltration was demonstrable ≤12 months. We report the impact of taliglucerase alfa on bone marrow, assessed by QCSI, in 26 patients treated in the Early Access Program (EAP) after the 2009 imiglucerase shortage. The investigator-initiated study was designed for patients with fat fraction (FF)≤0.30 and predictive of bony complications when FF<0.23. Of 15 treatment-naive patients (10 female; mean age 50.6 years), 9 had baseline FF≤0.30, 3 discontinued, and 4 with baseline FF≤0.23 and follow-up measurements improved to FF>0.23. Among 11 treatment-switch patients (6 female; mean age 40.5 years; mean imiglucerase exposure 9.5 [range 1-17] years), 8 had baseline FF<0.26, 5 of whom had FF<0.23; 3 improved to FF≥0.29 (1 year) and 1 increased to FF=0.26 (2 years). EAP protocol mandated increasing dosage to 30U/kg EOW; 2 patients at this dosage (17 years' imiglucerase exposure) with baseline FF=0.13 and FF=0.19 increased to FF=0.26 at 1 year, implying a booster effect (possibly due to 100% mannose residues resulting in greater bone uptake vs 40-60% mannose residues for imiglucerase). Booster effect requires further studies. This report adds to the overall taliglucerase alfa efficacy profile in adult patients with Type 1 GD, emphasizing bone benefit.

Disclosures: This study was sponsored by Pfizer. Pfizer and Protalix BioTherapeutics entered into an agreement in November 2009 to develop and commercialize taliglucerase alfa.

Keywords: Gaucher disease; enzyme replacement therapy; taliglucerase alfa.

(49)

TALIGLUCERASE ALFA DURING PREGNANCY FOR PATIENTS WITH TYPE 1 GAUCHER DISEASE

Ari Zimran[1*]; Hanna Rosenbaum[2]; Raul Chertkoff[3]

1 Gaucher Clinic and Genetic Unit, Shaare Zedek Medical Center, Jerusalem Israel. 2Rambam Medical Center, Haifa, Israel 3 Protalix BioTherapeutics, Carmiel, Israel

*Corresponding author: azimran@gmail.com

Bleeding tendency, organomegaly, and skeletal complications such as osteonecrosis of the hip joints in Gaucher disease (GD) may complicate pregnancy and delivery. Because pregnancy itself may worsen GD by inducing thrombocytopenia, anemia, and at times even bone crises, enzyme replacement therapy (ERT) may have an important role during this major life event. Label warnings with regard to pregnancy, even for the new ERTs with reproductive toxicology included in the drug development program, have led to concerns about the safety of ERT during pregnancy. Our group was the first to administer ERT in pregnancy and recommend that our female patients conceive with and continue ERT during their pregnancy. We reviewed patient records starting in 2009 for pregnant women receiving taliglucerase alfa from conception through the post-partum period. In all, 9 patients had 15 pregnancies: 1 patient had 3 pregnancies (1 missed abortion, 2 full-term, healthy infants; similar events were reported with imiglucerase before she switched to taliglucerase alfa); 1 patient had 3 pregnancies (1 missed abortion, 1 therapeutic abortion, 1 full-term, healthy infant); 1 patient had 2 pregnancies (2 full-term, healthy infants); and the remaining 6 patients had 6 viable pregnancies (full-term, healthy infants). Follow-up was up to 6 years. The overall 86% live birth rate with taliglucerase alfa (excluding the therapeutic abortion) is similar to that for patients with GD treated with imiglucerase or velaglucerase alfa. In fact, the maternal and neonatal outcomes were similar to the general population. These data add to the safety profile for taliglucerase alfa.

Disclosures: This study was sponsored by Pfizer. Pfizer and Protalix BioTherapeutics entered into an agreement in November 2009 to develop and commercialize taliglucerase alfa.

Keywords: Gaucher disease; enzyme replacement therapy; taliglucerase alfa

(50)

PHARMACOKINETICS, SAFETY, AND EFFICACY OF RAPID INFUSIONS OF VELAGLUCERASE-ALFA IN ADULT PATIENTS WITH GAUCHER DISEASE: PRELIMINARY REPORT OF AN INVESTIGATOR-INITIATED PILOT STUDY

Zimran_A[1], Elstein_D[1], Arbel_N[2], Chicco_G[1].

1Gaucher Clinic, Shaare Zedek Medical Center, the Hebrew University Hadassah School of Medicine, Jerusalem Israel, 2Medison Pharma, Petach Tikva, Israel

Corresponding author: Ari Zimran. azimran@gmail.com

Following anecdotal reports of patients with Gaucher disease who have been treated by enzyme replacement at accelerated rates, estimated to be between 5'-10', we designed an investigators-initiated pilot trial to assess the safety and efficacy of rapid infusions of velaglucerase alfa (VPRIV) in adult patients with Gaucher disease without significant co-morbidities, who have been treated for a minimum of 3 months. We chose velaglucerase because its safety profile includes fewer allergic reactions and anti-drug antibodies. Our "change in practice" protocol received IRB approval and all participating patients signed an informed consent. We recruited 15 patients age 20-43 (8 female, 7 male) with minimally 3 months of VPRIV exposure at 15-60 units/kg body-weight EOW. The study design includes 3 months at the standard time of 60 minutes (to document adverse effects), then acceleration phase of 3 months (30 minutes, 20 minutes and 4 infusions of 10 minutes – all in hospital) and a final stage of 3 months with 5 rapid infusions at home and final infusion with end-of-study (EOS) tests at the hospital. Safety is the primary end-point` efficacy parameters include spleen and liver volume assessment, hemoglobin and platelet counts and LysoGb1 as the most sensitive biomarker. In addition, pharmacokinetic study is done at baseline (60 minutes infusion) and at EOS (10 minutes). The study is on-going; so far we have not encountered any clinically significant adverse, and by the end of June 11/15 patients will be infused for 10 minutes. Preliminary safety results and patients' satisfaction will be presented.

Inclusion criteria: Aged 18 years and older, non-pregnant and non-lactating, non-splenectomized. Enzymatic diagnosis & molecular analysis indicative of type 1 Gaucher disease. Receiving VPRIV for at least 6 infusions (3 months) prior to Screening/Baseline at a constant dose and frequency.

Exclusion criteria: Existence of a clinically significant co-morbidity

(51)

INTRA-MONOCYTIC GLUCOCEREBROSIDASE ACTIVITY: A KEY PARAMETER TO PERSONALIZE IMIGLUCERASE DOSING?

Juliette Berger[1,2], Marie Vigan[3,4], Jérôme Stirnemann[5], Roseline Froissart[6], Nadia Belmatoug[7], Florence Dalbies[8], Christian Rose[9], Agathe Masseau[10], Christine Serratrice[11], Yves-Marie Pers[12], Ivan Bertchansky[13], Fabrice Camou[14], Monia Bengherbia[7], Céline Bourgne[1], Alexandre Janel[1], Sandrine Saugues[1,2], Thu Thuy Nguyen[3,4], Bruno Pereira[15], France Mentré[3,4] and Marc G. Berger[1,2,16]

[1]Hématologie Biologique, CHU Estaing, 1 place Lucie Aubrac, 63003 Clermont-Ferrand Cedex 1, France

2 CRB Auvergne, CHU Estaing, 1 place Lucie Aubrac, 63003 Clermont-Ferrand Cedex 1, France 3INSERM, IAME, UMR 1137, F-75018 Paris, France 4 Université Paris Diderot, IAME, UMR 1137, Sorbonne Paris Cité, F-75018 Paris, France 5 Service de Médecine Interne Générale, Hôpitaux Universitaires de Genève, Rue Gabrielle-Perret-Gentil 4, CH-1211 Genève 14, Suisse 6Laboratoire Maladies Héréditaires du Métabolisme, Centre de Biologie Est Groupement Hospitalier Est, 69677 Bron Cedex, France 7Médecine Interne et CRML, Hôpital Beaujon, AP-HP, 100 Bd Général Leclerc, 92110 Clichy, France 8Institut de Cancérologie et Hématologie, Hopital morvan, Brest, France 9 Onco-Hématologie, Hôpital Saint-Vincent de Paul, Boulevard de Belfort, 59000 Lille, France 10 Service de Médecine Interne, CHU, Nantes, France 11University hospital of Geneva , chemin du pont bochet 3, 1226 thonex, Switzerland. 12CHU Montpellier, Hopital St Eloy, Montpellier, France 13CHU de Montpellier, Hopital La Peyronnie, Montpellier 14Réanimation Médicale, Hôpital Saint André, CHU de Bordeaux, 33000 Bordeaux, France. 15DRCI, CHU G. Montpied, 58 rue Montalembert, 63003 Clermont-Ferrand Cedex 1, France 16Hématologie Clinique, CHU Estaing, 1 place Lucie Aubrac, 63003 Clermont-Ferrand Cedex 1, France

Corresponding author: Prof Marc G. Berger. e-mail: mberger@chu-clermontferrand.fr

Rationale: Enzyme replacement therapy (ERT) improves many type 1 Gaucher disease (GD1) symptoms but with unexplained variability. The standard therapeutic regimen is at odds with the reported few minutes of plasma half-life and we hypothesized that intra-monocytic pharmacokinetics could be different. Patients and methods: We assessed intra-monocytic glucocerebrosidase activity (IMoGlc) using an adapted flow cytometry technic (FAU: Fluorescence Arbitrary Units). We included 71 healthy subjects (HS) and 42 GD1 patients: i) 11 untreated patients, ii) 16 patients restarting imiglucerase in whom we followed residual IMoGlc and iii) 3 patients among these and 15 patients treated with a stable dose in whom IMoGlc was assessed at different times between infusions (ClinicalTrials #NCT01951989). IMoGlc kinetics of treated patients (n=31) were analysed by population pharmacokinetic approach using a one compartment model. Results: Endogeneous IMoGlc (27 patients) accounts for about 10% of IMoGlc in HS. Median of endogeneous IMoGlc before treatment was 159x103 FAU (min-max: 74-509x103FAU). It increased to a residual level of 233x103FAU (155-599x103) under treatment with a half-life of 35 days. Intra-monocytic pharmacokinetics appeared dramatically different from plasma, with a transient peak estimated at 2603x103FAU (1841-3211x103), and a half-life of 0.5 day (0.2-1.4). Interestingly, the individually adapted low-dose imiglucerase allowed a similar level of residual IMoGlc as standard dose. Conclusion: This proves for the first time that the intra-cellular pharmacokinetics of imiglucerase could explain the efficacy of typical imiglucerase doses and that the cases where decreased doses are required due to less aggressive disease may be explained by individual pharmacokinetic characteristics.

This study was sponsored by Sanofi Genzyme

12th EWGGD ZARAGOZA

The European Working Group on Gaucher Disease

Participants List

Abdelwahab	Magy	Egypt
Abkulut	Ilknur	Turkey
Abrahamov	Ayala	Israel
Adunsky	Nitzan	Israel
Aleksovska	Vesna	Macedonia
Ali Ibrahim Antaki	Mohammad	Jordan
Almeida Conde	Alberto	Spain
Alonso Lopez	Nerea	UK
Altarescu	Gheona	Israel
Amato	Dominick	Canada
Andrade	Marcio	Spain
Arkadir	David	Israel
Arshad Cheema	Huma	Pakistan
Aules Leonardo	Anastasia	Spain
Azevedo	Simone	USA
Bagal	SUKirti	USA
Becker	Michal	Israel
Belogurova	Margarita	Russia
Beresford	Paul	Switzerland
Berger	Marc	France
Berger	Juliette	France
Biegstraaten	Marieke	The Netherlands
Borin	Lorenza	Italy
Bosch	Aimee- Kate	South Africa
Braustein	Hila	Israel
Burke	Derek	UK
Cabasso	Or	Israel
Camou	Fabrice	France
Cano	Horacio	Spain
Capablo	Jose Luis	Spain
Cappellini	Maria Domenica	Italy
Carubbi	Francesca	Italy
Cavalcanti	David	Brasil
Cebolla Sanz	Jorge	Spain
Ceberio Hualde	Leticia	Spain
Cheng	Seng	USA
Chertkoff	Raul	Israel
Chicco	Gaya	Israel
Chis	Bogdan Augustin	Romania
Chronopoulou	Effie	UK
Collin-Histed	Tanya	UK
Cozma	Claudia	Germany
Croal	Laura	USA
Da Silva	Gerson	Brasil
Dar	Liron	Israel
Dardis	Andrea	Italy
Dayan	Galit	Israel
De Castro	Isabel	Spain
De La Serna	Javier	Spain
De Oliveira Vitarelli	Marcela	Brazil
De Pascale	Francesca	Italy
De Piccioto Shamia	Naam	Israel
Degulys	Andrius	Lithuania
Deng	Yifang	China
Di Rocco	Maja	Italy
Dimitriou	Evangelia	Greece
Dimoula	Ersi	Greece
Dinur	Tama	Israel
Djurik	Borislav	Bosnia Herzegovina
Domb	Yael	Israel
Dragoshi	Fation	Albania
Drewiacki	Tatiana	Brasil
Duan	Yanlong	China
Duboka	Davor	Serbia
Elstein	Deborah	Israel
Escuder Azuara	Beatriz	Spain
Eyskens	Francois	Belgium
Feder Krengel	Neta	Israel
Feldman	Ricardo	USA
Fernet	Mireille	Canada
Fog Tonnesen	Catherine	Denmark
Foronda Herrera	Hector	Spain
Franco	Melanie	France
Freisens	Selena	Germany
Fumic	Ksenija	Croatia
Gabriel	Jocelyne	Switzerland
Gaona	Manuel	Spain
Garcia	Salvador	Spain
Garcia	Jose Manuel	Spain
Garcia Jimenez	Inmaculada	Spain
Gervas	Javier	Spain
Gil	Faran	Israel
Giona	Fiorina	Italy
Giraldo	Pilar	Spain

Giuliani	Margaret	France
Goker-Alpan	Ozlem	USA
Goldblatt	Jack	Australia
Gomez Garate	Igor	Spain
Gomez Notario	Carlos	Spain
Gonzalez	Diana	Paraguay
Gonzalez	Derlis Emilio	Paraguay
Grauholt	Anne Marie	
Grey	Richard	USA
Gundobina	Olga	Russia
Hampson	Rasa	Lithuania
Hermida	Alvaro	Spain
Hernandez	Betina	USA
Higgs	Joanne	UK
Hollak	Carolina	The Netherlands
Horowitz	Mia	Israel
Igdoura	Suleiman	Canada
Ion	Luca	Republic Of Moldova
Irun	Pilar	Spain
Ivanov Vassilev	Yanko	Bulgaria
Ivanova	Margarita	USA
Johnson	Malcolm	UK
Jovanovic	Biljana	Serbia
Jung	Olive	USA
Karasev	Yuriy	Russia
Karlsson	Stefan	Sweden
Kiec Wilk	Beata	Poland
Klein	Shirley	Netherlands
Köhler	Ralf	
Komninaka	Veroniki	Greece
Kotsopoulou	Maria	Greece
Kyriazi	Maria	Greece
Lakocevic	Milan	Serbia
Latre	Paz	Spain
Lauridsen	Anne-Grethe	Denmark
Le Van Kim	Caroline	France
Leonard	Fintan	Switzerland
Lidon	Fernando	Spain
Limgala	RenUKa	USA
Lopez De Frutos	Laura	Spain
Lopez Velasco	Marta	Spain
Lucki	Patricia Noemi	Guatemala
Machaczka	Maciej	Sweden
Mackrell	Margaret	Canada
Malini	Erika	Italy
Manuel	Jeremy	UK
Maor	Gali	Israel
Marinakis	Theodoros	Greece
Martins	Ana Maria	Brasil
Massaro	Giulia	UK
Medina	Astrid	Colombia
Medrano Engay	Blanca	Spain
Mengel	Karl Eugen	Germany
Michelakakis	Helen	Greece
Michieletto Chiara	Maria	Italy
Mistry	Pramod	USA
Morales	Montserrat	Spain
Moro	Enrico	Italy
Morris	Liz	UK
Mosso	Luca	Italy
Mucci	Juan	Argentina
Murugesan	Vagishwari	USA
Nalysnyk	Luba	Nalysnyk
Neirinck	Anne	Belgium
Niemeyer	Pascal	Germany
Nogueira	Mariana	Switzerland
Ohana	Deddi	Israel
Olivan Viguera	Aida	Spain
Oswiecinski	Wojciech	Polonia
Ouzren	Nawal	Switzerland
Oz	Anat	Israel
Panahloo	Zoya	Switzerland
Parkkinen	Johanna	Finland
Pastores	Gregory	Ireland
Patterson	Mary Anne	Canada
Paz	Antonio	Spain
Peña Aragon	Paulina	Mexico
Perez Lopez	Jordi	Spain
Perez Valero	Teresa	Spain
Pérez Valero	Conchita	Spain
Petakov	Milan	Serbia
Peterschmitt	Judith	USA
Piperno	Alberto	Italy
Plamenov Dimitrov	Kristiyan	Bulgaria

Plaza	Sylvia	Spain
Plotkin	Horacio	USA
Podushkina	Ekaterina	Rusia
Popovici	Mariana	Romania
Prieto	Soledad	Spain
Prosekar	Barbara	Austria
Qureshi	Atif	Pakistan
Ramon	Enrique	Spain
Raviv	Miriam Milca	Israel
Reinholtsen	Frank	Norway
Reinke	Jörg	Sweden
Rengifo	Lyda	Colombia
Repa	Konstantina	Greece
Rigoldi	Miriam	Italy
Rintell	David	USA
Robertson	Gregory	Switzerland
Roca Espigau	Mercedes	Spain
Roig	Inma	Spain
Rolando Franco Franco	Emiliano	Paraguay
Rosenbaum	Hana	Israel
Rotner	Mira	Israel
Roy Sánchez	Ana	Spain
Royo	Pilar	Spain
Rozenfeld	Paula	Argentina
Rubio	Yolanda	Spain
Ryu	Eun Ju	USA
Sanchez	Maria Teresa	Spain
Santosa	David	Germany
Sanz	Javier	Spain
Sathe	Suyog	India
Saulite	Ieva	Latvia
Scheele	Willem	USA
Sekulic	Davorka	Croatia
Serra	Jenny	Spain
Serratrice	Christine	Switzerland
Sevitz	Hylton	South Africa
Sidransky	Ellen	USA
Siebert	Marina	Brazil
Sierra Monzon	Jose Luis	Spain
Sinca	George	Romania
Singh	Manjit	India
Siqueira	Ana Cecilia	Brasil
Smockova	Marketa	Rep Checa
Solanich	Xavier	Spain
Sorensen	Sven	Canada
Spartali	Argyro	Greece
Sulic	Darinka	Serbia
Surrel	Ghislaine	France
Symeonidis	Anargyros	Greece
Szer	Jeffrey	Australia
Taurisano	Roberta	Italy
Terekhova	Marina	Russia
Theochari	Aikaterini	Greece
Tiomkin	Maayan	Israel
Torquati	Fernanda	Italy
Trindade	Saide	Brasil
Tsaftaridis	Panagiotis	Greece
Tummolo	Albina	Italy
Tyagi	Shashank	India
Underhill	Lisa	USA
Vaino	Mariliis	Estonia
Van Dongen	Patricia	Netherlands
Vasilevsky	Alexey	Rusia
Veerhuis	Lydia	Netherlands
Verigou	Evgenia	Greece
Vigan	Marie	France
Viggiano	Emanuela	Italy
Vilageliu	Lluisa	Spain
Villarrubia	Jesus	Spain
Vlachaki	Efthymia	Greece
Vollstedt	Eva Juliane	Germany
Vom Dahl	Stephan	Germany
Wagner	Jasenka	Croatia
Wajnrajch	Michel	USA
Warwick	Alison	UK
Wellhoefer	Hartmann	Switzerland
White	Christine	Canada
White	Katherine	Canada
Yew Tan	Chong	UK
Zaken	Hagit	Israel
Zarina	Sandra	Latvia
Zimran	Ari	Israel
Znidar	Irena	Slovenia

Acknowledgments

Partner

EGA
European Gaucher Alliance
Driven by passion for patients

EWGGD ZARAGOZA
The European Working Group on Gaucher Disease

Collaborators

IIS Aragón
Instituto de Investigación Sanitaria Aragón

IACS Instituto Aragonés de Ciencias de la Salud

ciberer isciii

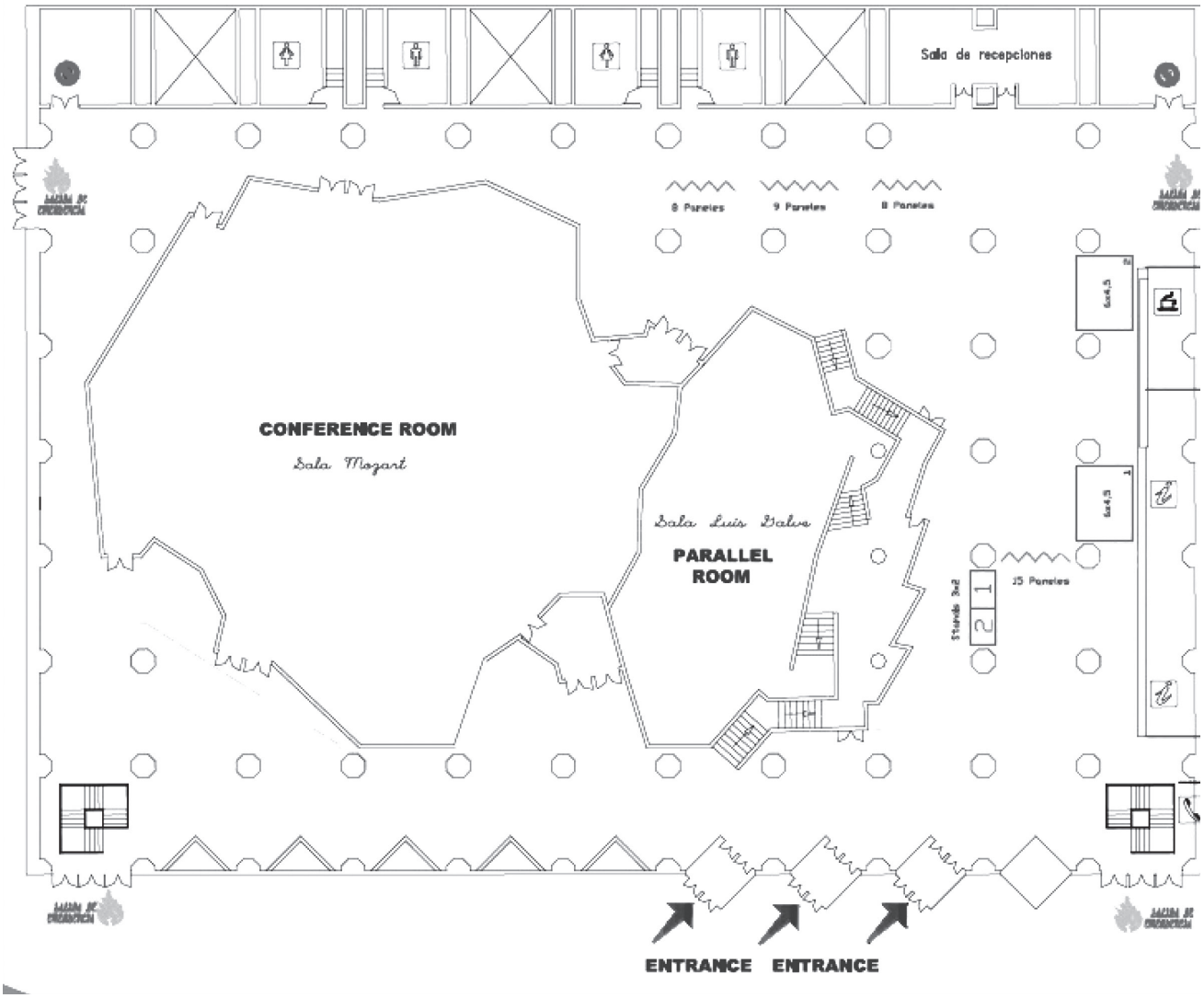
quirónsalud
La salud persona a persona

FEETEG

entidad de utilidad pública
feder
FEDERACIÓN ESPAÑOLA DE ENFERMEDADES RARAS

ASPHER

Plans of Auditorio and Stands



Map of the hotels

Hotel Eurostars Boston ****
Avda las Torres, 28, 50008 Zaragoza
T.: **976 59 91 92**

Hotel Silken Reino de Aragón ****
Calle del Coso, 80, 50001 Zaragoza
T.: **976 46 82 00**

Hotel Vincci Zaragoza Zentro ****
Calle del Coso, 86, 50001 Zaragoza
T.: **976 70 33 00**

ACKNOWLEDGMENTS