Congress Program & Abstract Book

April 8–11, 2018
Texas Medical Center
Houston, Texas

Chairpersons
Dr. Tarek Elghetany and Dr. Alison Bertuch
<table>
<thead>
<tr>
<th>CONTENTS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Page</td>
</tr>
<tr>
<td>Chairpersons’ Welcome</td>
</tr>
<tr>
<td>Sponsors</td>
</tr>
<tr>
<td>Committees</td>
</tr>
<tr>
<td>Program</td>
</tr>
<tr>
<td>Speaker Biographies</td>
</tr>
<tr>
<td>Speaker Abstracts</td>
</tr>
<tr>
<td>Research Abstracts</td>
</tr>
</tbody>
</table>
Dear Friends and Colleagues,

We are honored and humbled by the opportunity to host the 9th International Congress on Shwachman-Diamond syndrome (SDS) in Houston, Texas. In addition to being known as the fourth largest city in the USA and hub for the energy and space exploration industries, Houston is known for the Texas Medical Center (TMC), which is the largest medical complex in the world internationally recognized for its pioneering medical advances. Thus, we feel it is particularly fitting that, in the collaborative atmosphere of TMC where member institutions strive to deliver the highest standards of patient care and research, the 9th Congress takes place.

The Local and International Organizing Committees have arrived at an ambitious agenda for the 9th Congress, which will cover both research and clinical aspects of SDS. The two years since the last Congress have been noteworthy with respect to advances in the understanding of the genetics of SDS, its underlying molecular basis, and clinical features. We hope that the 9th Congress will provide fertile soil to discuss these new discoveries as well as ongoing and unpublished studies, and nourish new ideas that will help improve the lives of children and adults with SDS.

We would like to thank all Institutions, Associations, Foundations, families, and individuals who have provided critical support to ensure success of the 9th Congress. We are also grateful to all invited speakers, session chairs, and facilitators for their valuable time and expertise.

We welcome you in Houston and hope that you enjoy its warm environment and Southern hospitality during your stay.

M. Tarek Elghetany, MD
Alison Bertuch, MD, PhD

Co-Chairs of the Local Organizing Committee
Texas Children’s Hospital
Generous Sponsors
Generous financial support has been provided by the following Texas Children's Hospital Departments:

- Texas Children’s Cancer and Hematology Centers
- Department of Pathology
- Department of Gastroenterology, Hepatology and Nutrition
- Department of Allergy & Immunology

Generous financial support has also provided by:

- The Department of Molecular and Human Genetics, Baylor College of Medicine
Usua Oyarbide, USA – abstract n. 6
Ayushi Jain, Thailand – abstract n. 7
Amornrat Jensen, Thailand – abstract n. 8
Valentino Bezzerri, Italy – abstract n. 17

Additional travel awards have been generously provided by

the Robbins family to:
Edwardo Tron
Patty Fellin

the SDS Foundation to:
Coleman Lindsley
Amit Grover

the AISS to:
Francesco Pasquali
Cesare Danesino
Antonio Vella
Fabio Cofini
Igor Fanton
Local Organizing Committee

Tarek Elghetany, MD (Co-Chair)
Department of Pathology & Immunology
Baylor College of Medicine
Texas Children's Hospital

Alison Bertuch, MD, PhD (Co-Chair)
Department of Pediatrics
Division of Hematology/Oncology
Baylor College of Medicine
Texas Children's Hospital

Carlos Bacino, MD
Department of Molecular & Human Genetics
Baylor College of Medicine
Texas Children's Hospital

Lisa Forbes, MD
Department of Pediatrics
Division of Allergy & Immunology
Baylor College of Medicine
Texas Children's Hospital

Daniel Leung, MD
Department of Pediatrics
Division of Gastroenterology
Baylor College of Medicine
Texas Children's Hospital

Andrea Marcogliese, MD
Department of Pathology & Immunology
Baylor College of Medicine
Texas Children’s Hospital

Ghadir Sasa, MD
Department of Pediatrics
Division of Hematology/Oncology
Baylor College of Medicine
Texas Children's Hospital
Blanche P Alter, MD, MPH  
Clinical Genetics Branch  
National Cancer Institute  
Rockville, MD  
USA

Marco Cipolli, MD  
Centro Fibrosi Cistica  
Azienda Ospedaliera Universitaria Integrata  
Verona  
Italy

Stella Davies, MBBS, PhD  
Cancer and Blood Diseases Institute  
Cincinnati Children’s Hospital  
Cincinnati, OH  
USA

Inderjeet Dokal, FMedSci  
Barts and The London School of Medicine and Dentistry  
Queen Mary University of London  
London, UK

Jean Donadieu, MD  
Service d’Hémato-Oncologie Pédiatrique  
Hôpital Trousseau  
Paris  
France

Kasiani Myers, MD  
Cancer and Blood Diseases Institute  
Cincinnati Children’s Hospital  
Cincinnati, OH  
USA

Chee Y (Keith) Ooi, MBBS, PhD  
School of Women’s and Children’s Health  
University of New South Wales  
Sydney Children’s Hospital  
Randwick, New South Wales  
Australia

Johanna Rommens, PhD  
Program in Genomics & Genome Biology  
The Hospital for Sick Children  
Peter Gilgan Centre for Research & Learning  
Toronto, ON  
Canada

Akiko Shimamura, MD, PhD  
Dana-Farber/Boston Children’s Cancer and Blood Disorders Center  
Boston, MA  
USA

Alan Warren, MD, PhD  
Department of Haematology  
Cambridge Institute for Medical Research  
Wellcome Trust-Medical Research Council Stem Cell Institute  
University of Cambridge  
Cambridge, UK

Cornelia Zeidler, MD, MPH  
Department of Hematology and Oncology  
Hannover Medical School  
Hannover  
Germany
Program

Sunday, April 8
Houston Marriott Medical Center

Sunday Evening Session

2:00 - 7:00 pm  Registration open

4:00 - 4:30 pm  FAMILY Q&A SESSIONS: English (Akiko Shimamura, USA, Ghadir Sasa, USA, Daniel Leung, USA) and Spanish (Carlos Bacino, USA - Caridad Martinez, USA)

4:30 - 5:30 pm  LECTURE: Review of current diagnostic criteria using the previous two international consensus reports and introduction to the congress agenda (Akiko Shimamura, USA - Alison Bertuch, USA)

5:30 - 7:00 pm  OPENING RECEPTION

Monday, April 9
Rice University, BRC Building

Monday Morning Session  8:15 am – 12:45 pm

SDS DEFINITION AND GENETICS  (Chairs Johanna Rommens, Canada - Carlos Bacino, USA)

8:15 - 8:30 am  Welcome and opening remarks (Local Committee, Dr. Poplack, and Dr. Singh)

8:30 - 9:00 am  SDS as a ribosomopathy (Susan Baserga, USA)

9:00 - 9:30 am  Shwachman-Diamond disease: the canonic definition challenged by the genetic (Jean Donadieu, France)

9:30 - 10:00 am  EFL1 deficiency causes Shwachman-Diamond syndrome (Patrick Revy, France)

10:00 - 10:30 Coffee Break
10:30 - 11:00 am Genetic groups of SDS in Canada, and the SDS underlying hematopoietic phenotype (Yigal Dror, Canada)

11:00 - 12:00 am Selected abstracts:

  - 11:00 - 11:15: Johanna Rommens: Genetic variation in the major shwachman-diamond syndrome gene, SBDS.
  - 11:15 - 11:30: Yves D. Pastore: Is Bone Marrow Failure syndrome (IBMFs) 3, a syndrome due to DNAJC21 mutation part of SDBS or a distinct IBMFs?
  - 11:30 - 11:45: Francesco Pasquali: Chromosome anomalies in bone marrow of patients with Shwachman-Diamond syndrome as successful or unsuccessful attempts to improve ribosome biogenesis.
  - 11:45 - 12:00: Francesco Pasquali: Mild haematological features in patients with deletion of the long arm of chromosome 20 acquired in bone marrow.

12:00 - 12:45 pm SDS genetics and definition roundtable (Johanna Rommens, Canada - Yigal Dror, Canada - Cornelia Zeidler, Germany)

12:45 - 2:00 Lunch and Poster Viewing (odd number posters)

Monday Afternoon Session 2:00 – 6:00 pm

BASIC SCIENCE, HEMATOPOIESIS, AND STEM CELLS (Chairs Tom Vulliamy, UK - Alison Bertuch, USA)

2:00 - 2:30 pm Molecular basis of Shwachman-Diamond syndrome (A. Warren, UK)

2:30 - 3:00 pm Characterization of DNAJC21 as a bone marrow failure gene (Tom Vulliamy, UK)

3:00 - 3:30 pm Decreased Cdc42 Activity Regulates Functional Decline of HSC in SDS (Kasiani Myers, USA)

3:30 - 4:00 Coffee Break

4:00 - 4:30 pm The niche in Schwachman-Diamond Syndrome: inflammation driving evolution? (H.G.P. Raaijmakers, Netherlands)

4:30 - 6:00 pm Selected abstracts:

  - 4:30 - 4:45: Nuria Sánchez-Puig: Energetic basis of the nucleotide-affinity regulation of EFL1 by the SBDS protein.
  - 4:45 - 5:00: Dritan Siliqi: Shwachman-Diamond syndrome: SAXS, inside the structure of the ribosomal GTPASE EFL1, SBDS and their complex.
  - 5:00 - 5:15: Elif Asik: Shwachman-Diamond syndrome cells have reduced homology-directed repair.
  - 5:30 - 5:45: Amornrat Jensen: Decreased accumulation of superoxide dismutase 2 within mitochondria in the yeast model of Shwachman-Diamond syndrome.
  - 5:45 - 6:00: Ayushi Jain: Impaired pre-sequence processing is associated with reduced superoxide dismutase 2 activity in the yeast model of Shwachman-Diamond syndrome.

Monday Evening

Houston Marriott Medical Center

NETWORKING DINNER 6:30 – 8:30 pm
Tuesday, April 10
Rice University, BRC Building

Tuesday Morning Session   8:30 – 11:45 am

CLINICAL ASPECTS I: HEMATOLOGIC MANIFESTATIONS AND CLONAL EVOLUTION
(Chairs Akiko Shimamura, USA – Tarek Elghetany, USA)

8:30 - 9:00 am   Hematologic complications of SDS (Blanche Alter, USA)
9:00 - 9:30 am   Management of hematologic issues in adults and in adolescents transitioning into adulthood (Johnson Liu, USA)
9:30 - 10:00 am  Novel recurrent chromosomal changes and gene expression related to chromosome anomalies (Roberto Valli, Italy)
10:00 - 10:30 Coffee Break
10:30 - 11:00 am Clonal evolution in SDS (Francois Delhommeau, France)
11:00 - 11:45 am Selected abstracts:
   • 11:00 - 11:15: Kenichiro Watanabe: A nationwide cohort for Shwachman-Diamond syndrome in Japan.
   • 11:15 - 11:30: Kasiani Myers: The Shwachman-Diamond syndrome registry: what have we learned and where are we going?
   • 11:30 - 11:45: Kasiani Myers: MDS and AML in Shwachman-Diamond syndrome: clinical features and outcomes.
11:45 - 1:00 Lunch and Poster Viewing (odd number posters)

Tuesday Afternoon Session   1:00 – 4:15 pm

LECTURE: (introduction by Stella Davies, USA)

1:00 - 1:45 pm   Prognostic mutations in Myelodysplastic Syndrome after stem-cell transplantation (Coleman Lindsley, USA)

CLINICAL ASPECTS II: (Chairs Ghadir Sasa, USA, Blanche Alter, USA)

1:45 - 2:15 pm   SDS nutritional status and management ((Keith) Chee Y Ooi, Australia)
2:15 - 2:35 pm   Hepatic abnormalities in SDS (Daniel Leung, USA)
   2:35 - 3:00 Coffee Break
3:00 - 3:20 pm   Pregnancy complications in women with SDS (Neelam Giri, USA)
3:20 - 3:40 pm   Novel myopathy revealed in a newborn with severe hypotonia and thoracic dysplasia (Alexandra Topa, Sweden)
3:40 - 4:00 pm   The brain matters: neurological involvement and functioning in SDS (Elizabeth Kerr, Canada)
4:00 - 4:15 pm   Selected abstracts:
   • 4:00 - 4:15: Sara Loveless: The Patient and Family Perspective: Examining the Impacts of Shwachman-Diamond Syndrome.

Tuesday Evening   6:00 – 10:00 pm

6:00 - 10:00 pm   GALA DINNER   SPACE CENTER
**Wednesday, April 11**  
Rice University, BRC Building  

**Wednesday Morning Session**  
8:30 – 11:45 am  

**TRANSPLANT AND NOVEL THERAPIES**  
(Chairs Kasiani Myers, USA - Jean Donadieu, France)

- **8:30 - 9:00 am**  
  Hematopoietic stem cell transplantation (HSCT) for children with Shwachman-Diamond Syndrome (SDS) (Stella Davies, USA)

- **9:00 - 9:20 am**  
  Novel therapies (Marco Cipolli, Italy)

- **9:20 - 9:40 am**  
  SDS iPS studies of BMF and 7q: To identify novel therapies (Akiko Shimamura, USA)

  **9:40 - 10:00**  
  Coffee Break

- **10:00 - 10:45 am**  
  Selected abstracts:
  - 10:00 - 10:15: *Cornelia Zeidler*: 98 patients with Shwachman-Diamond – Syndrome: An update from the SDS-Registry Europe.
  - 10:15 - 10:30: *Melisa Ruiz-Gutierrez*: induced pluripotent stem cell model of 7q deletion in Shwachman Diamond syndrome identifies a novel therapeutic strategy.
  - 10:30 - 10:45: *Valentino Bezzeri*: Ataluren restores SBDS expression and function in bone marrow cells obtained from SDS patients.

- **10:45 - 11:45 am**  
  **CONFERENCE SUMMARY, FUTURE DIRECTIONS, DATA SHARING, AND ANNOUNCEMENTS**  
  (Akiko Shimamura, USA - Cipolli, Italy, Johanna Rommens, Canada - Cornelia Zeidler, Germany)

  **11:45 - 1:00 Farewell Lunch**
**Blanche P Alter, MD, MPH**

Training:
Radcliffe College (Harvard University): BA in Biochemical Sciences  
Johns Hopkins University School of Medicine: MD  
Johns Hopkins University School of Medicine: Intern and Resident in Pediatrics  
Harvard Medical School: Hematology fellow  
Johns Hopkins School of Public Health: MPH in Epidemiology  

Employment:
Harvard Medical School, Mount Sinai School of Medicine, University of Texas Medical Branch, and currently Senior Clinician, National Cancer Institute, Division of Cancer Epidemiology and Genetics, Clinical Genetics Branch.

I have been involved in the study of rare cancer-prone inherited bone marrow failure syndromes (IBMFS) for my entire career. I joined the NCI in 2000 and established a prospective cohort for families with the most common rare IBMFS, namely Fanconi anemia, dyskeratosis congenita, Diamond-Blackfan anemia, and Shwachman-Diamond syndrome. This cohort is used for time-dependent studies of predictors of cancer types and risks, and genotype/phenotype interactions (close to 150 publications).

---

**Carlos Bacino, MD**

Dr. Carlos Bacino is the Vice Chair of Clinical Affairs at the Department of Molecular Genetics at Baylor College of Medicine. He is also currently the director of the Genetics service at Texas Children’s Hospital and the director of the Genetics Clinic. Dr. Bacino has obtained his medical degree at the University of Buenos Aires in Argentina. He has then completed his Pediatrics residency at Beth Israel Medical Center in New York City and specialized in Clinical Genetics, Clinical Cytogenetics and Molecular Genetics at Cedars-Sinai Medical Center. Dr. Bacino has worked in the field of genetics for over 25 years. Among his many interests he has worked extensively in the field of skeletal dysplasias where he has care and followed many patients with Shwachman-Diamond Syndrome.

---

**Susan Baserga, MD, PhD**

Susan J. Baserga is a Professor at Yale University with a primary appointment in Molecular Biophysics & Biochemistry and joint appointments in the Departments of Genetics and Therapeutic Radiology. Dr. Baserga received a B.S. in Biology from Yale College and an M.D. and Ph.D. (Human Genetics) from Yale. Dr. Baserga was elected to the Connecticut Academy of Science and Engineering in 2012. The focus of her research is on the function of ribonucleoproteins in pre-rRNA processing, pre-ribosome assembly and on the resulting ribosomopathies.

---

**Alison Bertuch, MD, PhD**

Dr. Alison Bertuch received her BS in Biology from Massachusetts Institute of Technology (1981) and MD and PhD in Biology from the University of Rochester (1993). She then ventured to Houston for her residency in Pediatrics at Baylor College of Medicine, where she also received her training in Pediatric Hematology/Oncology. She then joined the faculty and is currently an Associate Professor in the Departments of Pediatrics and Molecular & Human Genetics and Assistant Dean for Curriculum in the Graduate School of Biomedical Sciences. She sees patients at Texas Children’s Hospital where she is director of the Bone Marrow Failure Program. Her research interests are in the areas of telomere biology, DNA repair, and inherited bone marrow failure syndromes.
Stella Davies, MB BS, PhD, MRCP

Cincinnati Children’s Hospital Medical Center

Stella Davies, MD is the Jacob G. Schmidlapp Endowed Chair and Professor of Pediatrics for the Division of Hematology/Oncology, and Director of the Blood and Marrow Transplant Program at Cincinnati Children’s Hospital Medical Center in Cincinnati, Ohio. She received her medical degree and PhD in Philosophy from the University of Newcastle-Upon-Tyne in England. Dr. Davies heads a team in the Division of Experimental Hematology. The team is conducting the most comprehensive research to date to analyze genetic variations and childhood leukemia with DNA from nearly 5,000 children from pediatric oncology centers throughout the United States. As researchers, they hope to identify markers that will indicate the children at greatest risk of developing leukemia as a second malignancy, and to find early, reversible changes in the process. Dr. Davies is internationally recognized as an expert in hematopoietic stem cell transplantation and acute Lymphoblastic leukemia.

Marco Cipolli, MD

Specialisation in Gastroenterology and Digestive Endoscopy – 1994.
Scholarship Doctor at Veneto’s Regional Centre for Cystic Fibrosis from 1988 to 1992.
Assistant Doctor at Veneto’s Regional Centre for Cystic Fibrosis from 1993 to 1994.
Head of the Gastroenterological Clinic Service of the Cystic Fibrosis Centre of the Ospedale Civile Maggiore since 2/5/1997.
Head of the Clinical Research Centre of the Cystic Fibrosis Centre, Ospedale Civile Maggiore Verona, from 2008 to 2016
Director of the Cystic Fibrosis Centre Marche Region, University Hospital Ancona, from 01 June 2016 to present.

Stella Davies, MB BS, PhD, MRCP

Cincinnati Children’s Hospital Medical Center

Stella Davies, MD is the Jacob G. Schmidlapp Endowed Chair and Professor of Pediatrics for the Division of Hematology/Oncology, and Director of the Blood and Marrow Transplant Program at Cincinnati Children’s Hospital Medical Center in Cincinnati, Ohio. She received her medical degree and PhD in Philosophy from the University of Newcastle-Upon-Tyne in England. Dr. Davies heads a team in the Division of Experimental Hematology. The team is conducting the most comprehensive research to date to analyze genetic variations and childhood leukemia with DNA from nearly 5,000 children from pediatric oncology centers throughout the United States. As researchers, they hope to identify markers that will indicate the children at greatest risk of developing leukemia as a second malignancy, and to find early, reversible changes in the process. Dr. Davies is internationally recognized as an expert in hematopoietic stem cell transplantation and acute Lymphoblastic leukemia.

François Delhommeau, PharmD, PhD

François Delhommeau is PharmD, PhD, and Full Professor of Hematology at the Medical School of Sorbonne University, Paris, France. He is Head of the Hematology Laboratory at Saint-Antoine Hospital (Assistance Publique-Hôpitaux de Paris). He spent 10 years in William Vainchenker’s Lab at Institut Gustave Roussy, Villejuif, where he worked on myeloproliferative neoplasms and co-discovered TET2 mutations in these syndromes. He currently drives a research group dedicated to the understanding of initiating events in adult and children myeloid cancers at the Saint-Antoine Research Centre.

Main publications:

Jean Donadieu, MD

Hemato oncologist and epidemiologist, Jean DONADIEU, 60, is working in the Trousseau Hospital, Paris.
He founded the french severe chronic registry in 1993, initially to evaluate the safety of GCSF in chronic neutropenia, latter extended to describe the natural history of any type of syndrome associated with neutropenia, including shwachman diamond syndrome. The french SCN registry is currently working with all french hematological centers, with the genetic laboratory of Pitie salpêtrière and with many other specialists involved in the care or in the evaluation or research in this field. A biobank is associated to this project in order to allow further research. This year, his team has been appointed as a reference center for this family of disorders in France.
Dr. Giri joined the National Institutes of Health, NIH in 2004 as a Senior Staff member and holds a position of Staff Clinician at the Clinical Genetics Branch, DCEG. She is an associate investigator on Dr. Blanche Alter’s clinical research program conducting Etiologic Investigation of Cancer Susceptibility in Inherited Bone Marrow Failure (http://marrowfailure.cancer.gov/).

Dr. Giri received her MBBS and MD in Pediatrics from Grant Medical College in Mumbai, India. She completed her US residency training in Pediatrics through New York Medical College, and a fellowship in Pediatric Hematology/Oncology at the Pediatric Oncology Branch of the National Cancer Institute. Dr. Giri is board certified in Pediatrics and in Pediatric Hematology/Oncology.

To better understand the natural history and consequences of various phenotypes on genotypes, Dr. Giri’s research focuses on clinical, genetic and epidemiological characterization major inherited bone marrow failure syndromes including Fanconi anemia, dyskeratosis congenita, Diamond Blackfan anemia and Shwachman Diamond syndrome.

Dr. Yigal Dror is Director of the Marrow Failure and Myelodysplasia Program, senior scientist at the Genetics and Genome Biology Program at The Hospital for Sick Children, Toronto, and a member of the Institute of Medical Sciences at the University of Toronto.

After graduating from Medical School and completing pediatric residency in the Hebrew University in Jerusalem, Dr. Dror completed clinical fellowship in pediatric hematology/oncology and a research fellowship in the field of hematopoiesis and marrow failure syndromes/myelodysplasia at SickKids hospital, Toronto. In 2000 Dr. Dror assumed his current position as a clinician scientist at SickKids.

His main clinical interests are in the area of bone marrow failure and myelodysplastic syndrome. His research focuses on characterization of stem cells and blood cells in these conditions, genetic etiologies and clinical outcome. He heads two multicenter pan-Canadian studies and networks: the Canadian Inherited Marrow Failure Registry and the Canadian Aplastic Anemia and Myelodysplastic Syndrome Study.

Dr. Dror lab study the landscape of mutations and affected genes in inherited bone marrow failure syndromes. The lab found that mutations in PARN cause developmental delay and bone marrow failure and are associated with defects in ribosomes and telomeres. The lab unravel that PARN-deficiency disrupts the polyadenylated state of H/ACA box RNA molecules that in turn influences ribosome profile and telomere length. The lab also identified DNAJC21 as the second gene associated with Shwachman-Diamond syndrome (SDS).

Data from the Canadian Inherited Marrow Failure Registry have contributed to understanding the clinical features and outcome of various IBMFS, risk of clones/myelodysplastic syndrome/leukemia, and phenotype-genotype correlations.

A main focus of the lab has been on dissecting the hematopoietic defects in SDS, and have provided insight into cell growth, cell death, oxidative stress, telomere length and gene expression in marrow progenitor cells; as well as, stromal function and angiogenesis.

Dr. Elghetany is Professor of Pathology & Immunology and Pediatrics at Baylor College of Medicine / Texas Children’s Hospital, Houston, Texas, USA. He is also Director of the Departmental Quality and Safety Programs. He graduated from Ain Shams University in Cairo, Egypt with highest honors among top 2% of the class. After completing residency in internal medicine in Egypt, he started residency in pathology at Baylor College of Medicine followed by hematopathology fellowship at State University of New York at Syracuse. He was awarded “Best Clinical Pathology Faculty” several times by the residents at the University of Texas Medical Branch at Galveston and Baylor College of Medicine. He has more than 70 publications and book chapters. He has a broad area of interest in anemias with special interest in inherited bone marrow failure syndromes and myelodysplastic syndromes.
**Elisabeth Kerr, PhD**

Elizabeth N Kerr, PhD, CPsych. has been a clinical neuropsychologist and health psychologist at the Hospital for Sick Children (SickKids) in Toronto, Canada since 1995. She is the program director of the Epilepsy Classroom, an active scientific staff with the Department of Paediatrics, Division of Neurology, and project investigator with the Research Institute at SickKids. She holds a cross appointment as an adjunct professor with the Department of Paediatrics at the University of Toronto. She is currently a member of the Board of Directors for SDS Canada and was previously a member of an advisory sounding board for the CIHR Institute of Genetics GE3LS and Health Service and Policy Research in Genetics/Genomics. Her research activities focus on neurocognitive and social-emotional outcomes in medically intractable epilepsy and genetic conditions.

**Daniel Leung, MD**

Dr. Leung is an Associate Professor of Pediatrics at Baylor College of Medicine and Director of Clinical Research within the section of Pediatric Gastroenterology, Hepatology, and Nutrition. He is a transplant hepatologist within the Texas Children’s Liver Center where he also directs the Viral Hepatitis Program. Dr. Leung’s clinical interests and research spans disciplines within hepatology and gastroenterology such as CF liver disease and the intestinal microbiome, SDS, liver fibrosis biomarkers, and liver transplantation. He serves as site PI of the CF Foundation/NIH-funded Prediction by Ultrasound of the Risk of Hepatic Cirrhosis in Cystic Fibrosis (PUSH) study and is leading research efforts studying biomarkers and novel imaging modalities in children with cholestatic liver diseases within the NIDK sponsored Childhood Liver Disease Research Network (ChiLDReN).

**Coleman Lindsley, MD, PhD**

Dr. Lindsley is an Assistant Professor of Medicine at Dana-Farber Cancer Institute and Harvard Medical School. He received his BA in music from Swarthmore College, and his M.D. and Ph.D. in Immunology from Washington University School of Medicine, then completed a residency in internal medicine at Brigham and Women’s Hospital and a fellowship in oncology at the Dana-Farber Cancer Institute. The primary focus of his laboratory is the biology and treatment of myeloid malignancies. His genetic studies have led to new genomic models of leukemia classification and MDS outcome after stem cell transplantation.

**Johnson Liu, MD**

The Feinstein Institute for Medical Research; Cohen Children’s Medical Center of NY; Monter Cancer Center

Dr. Johnson Liu holds the Les Nelkin Memorial Endowed Chair in Pediatric Oncology and is a Professor at the Donald and Barbara Zucker School of Medicine at Hofstra/Northwell. Dr. Liu received BS and MD degrees from the University of Michigan and the University of Michigan Medical School. He was previously awarded a Commendation Medal from the United States Public Health Service for his research at the National Institutes of Health. To date, Dr. Liu has authored or co-authored 111 papers, reviews, monographs and book chapters in the scientific and medical literature.
Dr. Kasiani Myers is an Assistant Professor in the Division of Bone Marrow Transplantation and Immune Deficiency at Cincinnati Children’s Hospital Medical Center within the University of Cincinnati Department of Pediatrics. Clinically she is focused in the diagnosis and treatment of bone marrow failure including Shwachman Diamond syndrome and hematopoietic stem cell transplant for non-malignant diseases. She is currently the co-Director of the Shwachman Diamond Syndrome Registry and has ongoing clinical and translational research in the fields of bone marrow failure and late effects of hematopoietic stem cell transplant.

Dr. Caridad Martinez is a member of the Center for Cell and Gene Therapy (CAGT) translational clinical research and the pediatric bone marrow transplant clinical team, which is part of the Bone Marrow Transplant Program. Her clinical interests include bone marrow transplantation (BMT), umbilical cord blood transplantation, stem cell transplantation for pediatric patients with congenital childhood diseases, and cell and gene therapy.

Clinical Research
Survival and Immunoreconstitution after Umbilical Cord Blood Transplant

Developing strategies to improve overall survival and immunoreconstitution after an umbilical cord blood transplant in pediatric patients with malignant or non-malignant diseases. Currently, she is the principal investigator of several clinical protocols using umbilical cord blood transplant for non-malignant and malignant diseases.

Treatment of Patients with Immunodeficiency Disorders
Dr. Martinez is also very involved with the Department of Allergy and Immunology at Texas Children’s Hospital, where she is engaged in evaluating, enrolling and taking care of patients with primary immunodeficiency disorders that will require a stem cell transplant as a curative option of their disease.

Education
M.D. University of Puerto Rico School of Medicine
Residency, Pediatrics, University of Puerto Rico Children’s Hospital
Fellowship, Pediatric Hematology-Oncology, St. Jude Children’s Research Hospital
Fellowship, Pediatric Bone Marrow Transplant, University of Minnesota Children’s Hospital
Board Certifications
American Board of Pediatrics

(Keith) Chee Y Ooi, MBBS, Dip Paeds, PhD, FRACP
Dr (Keith) Chee Y. Ooi, is a Clinical Academic at the University of New South Wales (UNSW) and Consultant Pediatric Gastroenterologist at Sydney Children’s Hospital Randwick, Australia. He graduated from the University of Melbourne with the Clara Myers Prize, and trained at The Hospital for Sick Children, Toronto, Canada.
Dr Ooi is an internationally and nationally recognized clinician and translational researcher in cystic fibrosis (CF), CF gastroenterology and childhood pancreatic diseases. He established and leads the CF gut research program at UNSW and Sydney Children’s Hospital Randwick. He has been awarded over USD3.5 million as a principal investigator, regularly presents at prestigious conferences, and has won multiple research awards, including the Dean’s Rising Star Award (UNSW Medicine) in 2015. He has published in high impact journals in the fields of gastroenterology (e.g. Gastroenterology), cystic fibrosis (e.g. Thorax, Chest) and pediatrics (e.g. Pediatrics, JAMA Peds).
**Marc H.G.P. Raaijmakers, MD, PhD**

Prof. dr. Marc H.G.P. Raaijmakers is a haematologist in the Department of Hematology at the Erasmus MC Cancer Institute, Rotterdam, the Netherlands. He received his MD from the University Utrecht and completed training in Internal Medicine and Hematology at the Radboud University Hospital in Nijmegen, the Netherlands. He completed postdoctoral research at the Department of Stem Cell and Regenerative Biology at Harvard University and Stem Cell Institute, revealing a concept of niche-induced oncogenesis in the hematopoietic system. He co-authored papers and comments in leading journals including Nature, Cell, Cell Stem Cell, J. Exp. Med, Blood and Leukemia, served in the editorial boards of leading journals in the field of Hematology and provided numerous invited lectures at international meetings. His laboratory studies micro-environmental contributions to the pathogenesis of hematopoietic disease with an emphasis on the initiation and evolution of preleukemic disorders. He received awards from the Dutch Cancer Society, the Dutch Society of Hematology, the Dutch Ministry of Science and Innovation and the Leukemia & Lymphoma Society U.S.A. His clinical focus is in bone marrow failure syndromes and acute myeloid leukemia.

**Patrick Revy, PhD**

Patrick Revy qualified in Immunology at the University Pierre et Marie Curie of Paris. He gained his PhD in Immunology in the laboratory of Alain Fischer in Necker Hospital (Paris, France) where he discovered that the Activation-induced cytidine deaminase AID is essential for class switch recombination and somatic hypermutation processes in humans. Patrick Revy is a researcher from CNRS and co-heads, with Jean-Pierre de Villartay, an INSERM Team « Dynamique du génome et système immunitaire » within the Imagine Institute (Paris, France). The primary goal of the Team is to identify the molecular mechanisms of genetic disorder of the immune system accompanied by DNA repair defect and/or telomere dysfunction in humans. This serves as the basis for both basic studies on the function and the regulation of the immune system and for medical applications in the diagnosis and treatment of diseases. Patrick Revy has contributed to the description of several molecular defects associating DNA repair/modification or telomere dysfunction with immunodeficiencies/bone marrow failure syndromes (e.g. AID, UNG, Cernunnos, ATR, Apollo, MYSM1, RTEL1).

**Johanna Rommens, PhD**

Scientist Emeritus, Program in Genetics & Genome Biology, The Hospital for Sick Children. Professor, Department of Molecular Genetics, University of Toronto. SickKids Research Institute, Room 12.9716, PGCRL, 686 Bay Street, Toronto, Canada M5G 0A4. The major research interests of Dr. Rommens relate to mechanisms that underlie inherited diseases and their presentations. Recent endeavours include i) studies of ribosomal deficiency as applied to Shwachman-Diamond syndrome and ii) modification of mendelian disease expression as applied to cystic fibrosis.

Dr. Rommens has extensive experience in disease gene identification, genome analysis, and gene expression. Her laboratory has developed mouse models for Shwachman-Diamond syndrome that are being used to understand how SDS-associated alleles and loss of Sbds manifest at cellular and tissue levels, and lead to disease phenotypes.

**Ghadir Sasa, MD**

Dr. Ghadir Sasa is interested in inherited and acquired bone marrow failure disorders. During her fellowship under the mentorship of Dr. Alison Bertuch, Dr. Sasa’s research focused on dyskeratosis congenita, an inherited bone marrow failure that results from abnormal telomere shortening. She is also clinically interested in myelodysplastic & myeloproliferative syndromes, paroxysmal nocturnal hemoglobinuria, and general hematology.

**Education:**

M.D., Jordan University of Science and Technology - Internship/Residency, Miami Children’s Hospital - Fellowship, Baylor College of Medicine

**Board Certifications:**

American Board of Pediatrics - American Board of Pediatric Hematology/ Oncology

**Clinical Interests:**

General Hematology, Bone marrow failure disorders, Dyskeratosis congenital, Myelodysplastic & myeloproliferative syndromes, Paroxysmal nocturnal hemoglobinuria
Akiko Shimamura, MD, PhD
Dr. Akiko Shimamura is an Associate Professor of Pediatrics at Harvard Medical School, serves as Director of the Bone Marrow Failure and Myelodysplastic Syndrome Program of the Dana Farber/Boston Children’s Cancer and Blood Disorders Center, and holds the Samuel E. Lux IV Chair in Pediatric Hematology/Oncology. Her research focuses on translational studies spanning clinical through basic science investigations to understand the genetic and molecular basis of bone marrow failure, MDS, and leukemia predisposition with the goal of developing improving diagnosis and therapy. Dr. Shimamura, in collaboration with Dr. Kasiani Myers, directs the North American Shwachman Diamond Syndrome Registry.

Alexandra Topa, MD, MSc
Graduate of the Faculty of Medicine "Carol Davila" in Bucharest (Romania) in 2002, I obtained the specialist degree in Pediatrics in 2008 in France followed by the Master degree in Genetics in 2009. From this year on, I have worked as full-time clinician at the Department of Clinical Genetics at the Sahlgrenska University Hospital in Gothenburg, Sweden. I have obtained the specialist degree in Clinical Genetics in 2012 and I currently work as a senior consultant. My main activity is focused on the diagnostics and genetic counselling of patients with developmental abnormalities including foetopathology and prenatal diagnostics. I have a teaching activity with lectures and seminars in clinical genetics for medical students at the Faculty of Medicine “Sahlgrenska Academy”, University of Gothenburg. My main area of research is the etiology and mechanisms of occurrence of craniofacial developmental abnormalities with focus on craniosynostosis.

Roberto Valli, PhD
Department of Medicine and Surgery, University of Insubria – Varese – Italy.
Born in Varese (Italy) in 1973. In 2001 he graduates in Biological Sciences. In December 2001 he starts the PhD program at the University of Insubria and in 2005 he obtained the PhD degree. During the PhD studies, he works for a period in the Biology and Genetics labs of the D.A.P.E.G. Department of the University of Bari (Italy).
From 2005 to 2008 he started the Post-doc research program at the University of Insubria. Since 2008 he is researcher in Medical Genetics of the University of Insubria and Aggregate Professor of General and Applied Biology and Medical Genetics at the School of Medicine. Since 2002 is member of the Italian’s Human Genetics Society (S.I.G.U.)
Since 2015 is member of the European Cytogenetic Association (E.C.A.)

Alan Warren, FRCP, FRCPath, PhD, FMedSci
University of Cambridge, England, UK
Alan Warren graduated in Biochemistry (1983) and Medicine (1986) from the University of Glasgow and trained in Haematology at the Hammersmith Hospital in London and Addenbrooke’s Hospital in Cambridge. He was appointed Professor of Haematology in 2003. He gained his PhD in molecular biology at the MRC Laboratory of Molecular Biology in Cambridge under the supervision of Terry Rabbitts FRS, where he discovered an essential role for the leukaemia-associated LIM-only protein Lmo2 in haematopoeis. The Warren laboratory aims to elucidate basic mechanisms of eukaryotic ribosome assembly to better understand how corruption of this process causes stem cell subversion and leukaemia predisposition. He was elected Fellow of the Academy of Medical Sciences in 2005 and received a FEBS Award from the Austrian Society for Molecular Biology and Biotechnology in September 2016.
Cornelia Zeidler, MD

Pediatric Hematologist and Oncologist
Training: Medicine 1981-1987, Medical School Hannover, MD degree 1987, doctoral thesis 1993
Medical School Hannover, 2013 Master of Public Health (MPH)

Academic Appointments:

1986-1987 Internship, Children Hospital, Medical School, Hannover
1988- 1989 Laboratory of the Department Pediatric Hematology and Oncology, Medical School Hannover
1989- 1996 Residency, Fellowship, Children Hospital, Medical School, Hannover
1995-present Clinical Consultant and Coordinator of the Severe Chronic Neutropenia International Registry in Europe
1997-present Attending, Children Hospital, Medical School, Hannover
2011-present Deputy speaker of the centre for rare diseases at Hannover Medical School

Membership: EHA, German Society of Pediatrics, German Society of Pediatric Hematology/ Oncology

2000-present Member of the SCNIR Advisory Board
2003-2013 Member of the RDTF, now EUCERD of the EU

Major Fields of Interest:

• Epidemiology and Pathophysiology of Neutropenia
• Congenital Bone Marrow Failure Syndromes
• Secondary Leukemias

Research Activities (all supported by grant funding):

• Coordinating Investigator of the SCNIR in Europe (1999-present)
• Coordination of the German Network on Congenital BMFS (2003-2012)
• Incidence and Natural Course of Congenital Neutropenia with Respect to the Different Subtypes (project in the bmfs-network)
• Coordinating Investigator of the GPOH Long-term observational trial for severe chronic neutropenia (2011-present)
Review of current diagnostic criteria using the previous two international consensus reports and introduction to the Congress agenda

Akiko Shimamura, MD PhD\textsuperscript{1}, Alison Bertuch, MD, PhD\textsuperscript{2}

\textsuperscript{1} Dana-Farber/Boston Children’s Cancer and Blood Disorders Center
300 Longwood Avenue, Boston, MA 02115

\textsuperscript{2} Texas Children’s Cancer and Hematology Centers/Baylor College of Medicine
1102 Bates Street, Houston, TX 77030

Abstract

In the first half of the Introductory Lecture, Dr. Shimamura will discuss the recent evolution of our understanding of Shwachman-Diamond syndrome, highlighting areas of progress in our understanding of the genetics, underlying biology, and clinical features of SDS since the last consensus guidelines were published. In the second half of the Introductory Lecture, Dr. Bertuch will review how advances made since the 8th International Congress on Shwachman-Diamond Syndrome have shaped the agenda of the current Congress, which will be reviewed.
Shwachman-Diamond Syndrome as a ribosomopathy
Susan J. Baserga, MD, PhD
Departments of Molecular Biophysics & Biochemistry, Genetics, and Therapeutic Radiology,
Yale School of Medicine, 333 Cedar St., New Haven, CT 06520

Abstract
Ribosome biogenesis, beyond being a critical requirement for growth in all eukaryotic cells, has a broad impact on the etiology of numerous human disorders. Human genetic diseases caused by mutations in structural components of the ribosome and in factors required to make ribosomes are called ribosomopathies. The number of defined ribosomopathies has been steadily increasing in the last 15 years paralleling our increased understanding of the complexity of how ribosomes are made at the molecular level. A central conundrum is the apparent tissue-specificity of a particular ribosomopathy. For example, mutation or deletion of more than 10 different ribosomal proteins causes bone marrow failure (Diamond-Blackfan anemia, DBA) yet mutation in the ribosomal protein Rps23 causes short stature and intellectual disability without the bone marrow failure characteristic of DBA. Similarly, mutation in the ribosomal protein RpsA causes Isolated Congenital Asplenia, with no evidence of bone marrow failure. Like these ribosomopathies, Shwachman-Diamond syndrome (SDS) is caused by mutation in proteins required for making functional ribosomes: SBDS, DNAC21 and perhaps EFL1. SDS is characterized by bone marrow failure, exocrine pancreatic insufficiency, skeletal abnormalities and cancer predisposition. I will discuss how, to understand the molecular basis of ribosomopathies, we must now superimpose the physiology onto the biochemistry, especially during embryonic development. I will further highlight two models put forth to provide a mechanistic basis for the conundrum of their tissue specificity.
Shwachman-Diamond disease: the canonic definition challenged by the genetic

Jean Donadieu, MD, PhD

Service d’Hémato-Oncologie Pédiatrique APHP Hopital Trousseau Université Paris sorbonne Registre des neutropénies - Centre de référence des neutropénies chroniques www.neutropenie.fr 26 avenue du Dr Netter F 75012 Paris E mail: jean.donadieu@aphp.fr

Abstract

The canonic phenotypic definition of Shwachman-Diamond Syndrome (SDS) is based on the association of 2 main criteria: exocrine pancreas deficiency and chronic neutropenia. Additional features may participate to the definition of the syndrome: bone dysplasia, developmental delay and several less frequent features like heart abnormalities, liver cytolysis, while the natural history is frequently complicated by pancytopenia, MDS and leukemia...Such criteria have been published in a consensus guidelines(1). And, since its discovery in 2003, germline recessive mutations of the SBDS gene have been considered sufficient to define universally this syndrome whatever the spectrum of phenotype manifestations. It results that patients who have no SBDS mutations, but some features belonging to SDS should be considered as ‘SD-like syndrome’. In our experience, 10 genes (table 1) can be found in such SD like syndrome and this list is incomplete as the molecular background is still not determined in some patients. But in the literature, the criteria used to call the disease ‘SDS like’ are larger than those commonly found in SDS. As example, because natural history of SDS is known to be complicated by pancytopenia, anemia, thrombocytopenia, MDS, any of these complications are considered sufficient in SDS like syndrome, even without chronic neutropenia. The second example is the definition of pancreatic exocrine insufficiency, which may be considered in case of a large group of manifestations: pancreas lipomatosis, abnormal signal or shape on MRI or CT scan, low fat vitamin levels, chronic diarrhea with steatorrhea, low elastase level, low secretion level of pancreas enzymes...

The question of the definition of SD like appears to be much broader than a purist debate. If any wants to describe the epidemiology, the natural history of such entities or to understand their pathophysiology, or even to find new genes, such issues should be clarified, otherwise, a lot effort will be wasted.

Table 1: an (incomplete) list of diseases with ‘bone marrow failure’ and exocrine pancreas insufficiency

<table>
<thead>
<tr>
<th>Syndrome or gene</th>
<th>Specific features</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pearson syndrome. Mitoc. DNA deletion</td>
<td>Acidemia / Kearns-Sayre syndrome</td>
<td>(2)</td>
</tr>
<tr>
<td>DNAJC21</td>
<td>Pancytopenia mostly thrombocytopenia</td>
<td>(3)</td>
</tr>
<tr>
<td>NDUFS2</td>
<td>dystonia</td>
<td>(4)</td>
</tr>
<tr>
<td>EFL1</td>
<td>Mainly red cell deficiency</td>
<td>(5)</td>
</tr>
<tr>
<td>SRP54</td>
<td>Myeloid maturation arrest</td>
<td>(6)</td>
</tr>
<tr>
<td>GLUD1</td>
<td>Hypoglycemia Hyper amonemia</td>
<td>(7)</td>
</tr>
<tr>
<td>G6PC3</td>
<td>Skin heart crohn disease</td>
<td>(8)</td>
</tr>
<tr>
<td>JAGN1</td>
<td>Myeloid maturation arrest</td>
<td>(9)</td>
</tr>
<tr>
<td>COX412</td>
<td>Mainly red cell deficiency</td>
<td>(10)</td>
</tr>
<tr>
<td>EIF2AK3</td>
<td>Neonatal diabetes</td>
<td>(11)</td>
</tr>
</tbody>
</table>

Reference List

**EFL1 deficiency causes Shwachman-Diamond Syndrome**

*Patrick Revy, PhD*

INSERM UMR 1163, Laboratory of Genome Dynamics in the Immune System, Equipe Labellisée Ligue contre le cancer, Paris, France

Paris Descartes–Sorbonne Paris Cité University, Imagine Institute, Paris, France

**Abstract**

Entry of the large ribosomal subunit into active translation is licensed by removal of the anti-association factor eIF6, a key step in the translational activation of ribosomes that is catalysed by elongation factor-like GTPase 1 (EFL1) and its allosteric regulator SBDS. Mutations in the SBDS gene cause autosomal recessive Shwachman-Diamond syndrome (SDS) that is typified by progressive bone marrow failure and leukaemia predisposition. Here, we report biallelic loss of function mutations in the EFL1 gene in three unrelated individuals with clinical features akin to SDS. The mutations cause defective ribosome assembly and reduced global translation due to impaired eIF6 release. Additionally, we show that mice carrying a homozygous missense Efl1 mutation recapitulate the SDS phenotype. Our findings strongly support the hypothesis that impaired terminal 60S subunit maturation and attenuated translation is the primary molecular defect in SDS.
Genetic groups on Shwachman-Diamond syndrome and underlying hematopoietic phenotype

Yigal Dror, MD, FRCP(C)

Genetics and Genome Biology Program, Research Institute, The Hospital for Sick Children, and Institute of Medical Sciences, University of Toronto, and Marrow Failure and Myelodysplasia Program, Division of Haematology/Oncology, Department of Paediatrics, University of Toronto.

Abstract

Shwachman-Diamond syndrome is an inherited multi-system disorder with bone marrow failure, exocrine pancreatic dysfunction, metaphyseal dysplasia, among other manifestations. Several genetic groups have thus far been described.

In the first part of the talk the genetic basis of the disease in Canada will be discussed. The most common SDS group that comprises 85% of the genetically tested cases with this disease on the Canadian Inherited marrow failure registry (CIMFR) is of patients with SBDS mutations. Patients with DNAJC21 comprises 10% of the cases, Thus far, only one case with SRP54 have been identified in our registry. Patients with SBDS mutations tend to have more severe pancreatic insufficiency. Patients with DNAJC21 mutations tend to have more severe hematological phenotype. MDS was observed in both genetic groups. The patient with SRP54 had clinical phenotype that more resembled severe congenital neutropenia that SDS.

In the second part of the talk the hematopoietic phenotype of SDS will be discussed.
Abstract

The synthesis of new ribosomes is a fundamental conserved process in all cells. The large (60S) ribosomal subunit is pre-assembled in the nucleus and exported to the cytoplasm where it completes a final series of maturation steps to gain translational competence. These events include formation of the catalytic center, recruitment of the last remaining ribosomal proteins and the removal of inhibitory assembly factors. However, the mechanisms underlying this process remain poorly understood due to limited structural information. Using the latest advances in single-particle cryo-electron microscopy, we have determined the structures of multiple sequential cytoplasmic ribosomal subunit maturation intermediates that provide fundamental new insights into the process of 60S maturation and the molecular basis of the ribosomopathy Shwachman-Diamond syndrome. I will discuss how these data may lead to improved therapies for patients with SDS.
Characterization of DNAJC21 as a bone marrow failure gene

Tom Vulliamy, PhD

Blizard Institute, Barts and The London School of Medicine and Dentistry, Queen Mary University of London, London, UK

Abstract

Many of the patients referred to our bone marrow failure (BMF) registry have been hard to classify. While a significant proportion are clearly idiopathic, in more than 700 of them there is a suggestion of a constitutional basis to the disease but without sufficient features to diagnose a recognized syndrome. The indicative signs may either be a subtle extra-hematopoietic abnormality, a family history of disease or presentation at a very young age.

We have been carrying out exome sequencing in these patients in an attempt to uncover novel genetic causes of this apparently constitutional BMF. One quite dramatic allelic series that we identified recently were homozygous rare/novel loss of function variants in DNAJC21 in four unrelated families. None of these cases had been diagnosed with Shwachman Diamond syndrome.

Little was known about the human DNAJC21 protein at the time, but it’s yeast orthologue (Jjj1) was known to be involved in ribosome biogenesis, working just upstream of the SBDS orthologue in the maturation of the 60S subunit. We were able to confirm a role for the human protein in this process. Of interest, we also found an association with rRNA as well as an unexplained response to the transcription inhibitor, actinomycin D. We have also shown a surprising interaction with dyskerin, the protein responsible for X-linked dyskeratosis congenita.

Finally, through CRISPR Cas9 engineering, we have developed a model of the disease by introducing loss of function variants into exon 3 of the zebrafish gene. Preliminary analysis shows that these fish are viable and do not appear to have significant neutropenia. We believe it will be interesting to study how these animals respond to hematopoietic stress.
Decreased Cdc42 Activity Regulates FunctionalDecline of HSC in Shwachman-Diamond syndrome

Sachin Kumar, PhD 1,2, Kalpana J Nattamai 1, Rebbekah Karns 4, Akiko Shimamura, MD, PhD 5, Stella M Davies, MBBS, PhD 3, Hartmut Geiger, PhD 6,7 Kasiani C Myers, MD 2

1 Division of Experimental Hematology and Cancer Biology, Cincinnati Children's Research Foundation, Cincinnati, OH 45229, USA.
2 Pharmacology Division, CSIR-Central Drug Research Institute, Lucknow-226031, India.
3 Division of Bone Marrow Transplantation and Immune Deficiency, Cincinnati Children's Hospital Medical Center, Cincinnati, OH;
4 Division of Gastroenterology, Hepatology and Nutrition, Cincinnati Children's Hospital Medical Center and University of Cincinnati, Cincinnati, OH, USA.
5 Boston Children’s Hospital, Dana Farber Cancer Institute, Boston, MA
6 Institute of Molecular Medicine, Ulm University, Ulm, Germany.
7 Aging research center, Ulm University, Ulm, Germany.

Abstract

Shwachman-Diamond syndrome (SDS) is an inherited bone marrow failure syndrome associated with an increased risk of myelodysplasia and acute myeloid leukemia. Detailed cellular and molecular mechanisms of bone marrow failure remain unclear. Here we report drastically reduced frequencies of hematopoietic stem cells (HSCs) (CD34+, CD38- cells) in bone marrow of SDS patients, and significant functional defects of HSC from SDS individuals confirmed ex vivo. Primitive hematopoietic cells from SDS patients presented with a strongly reduced activity of the small RhoGTPase Cdc42. Changes in the activity of Cdc42 in HSCs result in changes of polarity in HSCs, and thus SDS HSCs presented with a reduced frequency of HSCs polar for both cytoplasmic polarity proteins as well as epigenetic markers like H4K16ac. The level of apolarity of SDS HSCs was tightly linked to the magnitude of HSC depletion in SDS patients, implying a causative role for this phenotype to bone marrow failure. Importantly, raising Cdc42 activity ex vivo in primitive hematopoietic cells from SDS patients to the level seen in normal HSCs via exogenous Wnt5a correlated strongly with elevated numbers of functional SDS HSCs. Our data identify a decreased activity of the small RhoGTPase Cdc42 in SDS HSCs as a novel target to improve HSCs number and function in SDS and imply that bone marrow failure in SDS is a Cdc42 activity driven and thus reversible pathology.
The niche in Shwachman-Diamond Syndrome: inflammation driving evolution?
Marc H.G.P. Raaijmakers, prof. dr.

Erasmus MC cancer Institute, Department of Hematology, Rotterdam, the Netherlands

Abstract

Congenital bone marrow failure syndromes with leukemia predisposition, in which mutations in a single gene, present in both HSPC and niche cells, drives tissue failure, clonal evolution and malignant transformation, provide an unprecedented model system to study the molecular mechanisms and human relevance of niche contributions to bone marrow failure and leukemogenesis. The lecture will address the findings of studies in Shwachman-Diamond Syndrome (SDS), caused by constitutive, bi-allelic, loss-of-function, mutations in the ribosome biogenesis gene SBDS, and characterized by bone abnormalities, neutropenia and a striking propensity to develop MDS/AML. Commonalities will be discussed between findings in different murine models of niche induced/facilitated leukemogenesis and biological observations in SDS patients. The findings complement other emerging data from mouse models of niche-facilitated oncogenesis and human disease, pointing at inflammatory signaling in HSPC niches as a biologic commonality in preleukemia and myeloid neoplasm. A hypothetical model can be proposed of ‘mesenchymal niche inflammation’ promoting bone marrow failure, genotoxic stress, genetic instability and clonal evolution, of broader relevance to congenital bone marrow failure and leukemia predisposition syndromes. Recent data as well as controversies and uncertainties concerning this evolving new paradigm will be discussed.
Hematologic Complications of SDS

Blanche P Alter, MD, MPH 1, Neelam Giri, MD 2, Irina Maric 2, and M Tarek Elghetany 3

1 Clinical Genetics Branch, Division of Cancer Epidemiology and Genetics, National Cancer Institute, National Institutes of Health, Bethesda MD; 2 Department of Laboratory Medicine, National Institutes of Health, Bethesda, MD, and 3 Texas Children’s Hospital, Baylor College of Medicine, Houston, TX

Abstract

Patients with Shwachman Diamond syndrome (SDS) usually present with exocrine pancreatic insufficiency; neutropenia is the hallmark hematologic finding. Most patients have macrocytosis and may have or develop anemia and thrombocytopenia (pancytopenia). They may progress to significant bone marrow failure (BMF) and are at risk of myelodysplastic syndrome (MDS), acute myeloid leukemia (AML), or syndrome-specific solid tumors. The NCI Inherited Bone Marrow Failure Syndromes (IBMFS) program opened in 2002 and has enrolled 35 patients with SDS among more than 600 patients with an IBMFS. This multis Syndrome cohort provided an opportunity for cross-syndrome comparisons of many parameters. We evaluated blood and marrow features, including the specificity of certain diagnostic tests for IBMFS. Data revealed that patients with SDS do not have increased chromosome breakage following culture with DNA-crosslinking agents (diagnostic of Fanconi anemia, FA), elevated red cell adenosine deaminase (diagnostic of Diamond Blackfan anemia, DBA), or very short leukocyte telomeres (as in dyskeratosis congenita, DC). The diagnosis of MDS in the context of an IBMFS is challenging, since many bone marrows tend to have varying degrees of dyspoiesis. We examined bone marrow findings in 82 NIH patients with IBMFS, and classified the types and levels of dyspoiesis as well as the cytogenetic findings. There were 27 DBA, 26 DC, 20 FA, 6 SDS, and 3 patients with thrombocytopenia absent radii (TAR). All 6 patients with SDS had multilineage dyspoieses, and 2 had del(20q). None of the patients with SDS has evolved to clinical MDS or AML to date. We suggest that dyspoietic bone marrow morphology per se may not be a predictor of an adverse hematologic prognosis in these patients; larger studies are needed with longer follow-up.
Management of Hematologic Issues in Adults and in Adolescents
Transitioning into Adulthood
Johnson M. Liu, MD

The Feinstein Institute for Medical Research, Manhasset, NY; Cohen Children’s Medical Center of NY, New Hyde Park, NY; Monter Cancer Center, Lake Success, NY; Zucker School of Medicine at Hofstra/Northwell, Hempstead, NY

Abstract

Patients with Shwachman-Diamond Syndrome (SDS) demonstrate varying degrees of cytopenia, ranging from isolated neutropenia, mild to moderate thrombocytopenia, anemia, and/or pancytopenia. Neutropenia is the most common hematologic finding, whereas anemia and thrombocytopenia are reported less frequently. Granulocyte colony-stimulating factor is used to treat severe neutropenia and does not appear to be associated with CSF3R mutations as can be seen in severe congenital neutropenia (Blood 2018;131:408). Defined hematologic complications include aplastic anemia, clonal hematopoiesis, myelodysplastic syndrome (MDS), and acute myeloid leukemia (AML). Bone marrow failure in SDS is poorly understood mechanistically but develops in ~40% of patients by age 50. MDS is a clonal stem cell disorder characterized by ineffective hematopoiesis, cytopenia, morphologic dysplasia, and a tendency for leukemic transformation. MDS is uncommon in children and young adults, often arising secondary to inherited predisposition or to acquired bone marrow failure, and it may have a different genetic basis from adult MDS. The cumulative risk of MDS/leukemia was 36% by age 30 in a large French cohort (Haematologica 2012; 97:1312), but lower frequencies for AML have been reported in other smaller cohorts. Stem cell transplantation is typically considered for SDS patients who develop severe cytopenia, MDS, or leukemia. The pathogenesis of MDS in patients is an active area of investigation. Clonal cytogenetic abnormalities in the bone marrow can occur, including i(7q), del(20q), del(7q), monosomy 7, and others. Furthermore, recent molecular studies have suggested clonal hematopoiesis due to TP53 mutations in nearly half of a small cohort of 27 patients (Blood 2018;131:408). SDS patients with MDS and TP53 mutations also appear to have a poor prognosis after stem cell transplantation (NEJM 2017;376:536). This presentation will review the hematologic complications of SDS, particularly focusing on the propensity of SDS patients to develop clonal hematopoiesis and MDS and on management issues in adolescents and adults.
Novel recurrent chromosomal changes and gene expression related to chromosome anomalies
Roberto Valli, PhD

Human and Medical Genetics, Department of Medicine and Surgery, University of Insubria, Varese, Italy

Abstract

Starting in 1999, we have monitored the cytogenetic picture in the bone marrow (BM) of a cohort of 96 Italian patients with SDS by all suitable cytogenetic methods and with molecular investigations related to the chromosome changes.

During this follow-up, we have found also clonal chromosome anomalies recurrent, but different from the most frequent ones, the well characterized isochromosome i(7)(q10), and the interstitial deletion del(20)(q).

These newly recognized clonal recurrent anomalies were: unbalanced structural anomalies of chromosome 7, a further complex rearrangement of the del(20)(q) with duplicated and deleted portions, and an unbalanced translocation t(3;6), with partial trisomy of the long arm of chromosome 3 and partial monosomy of the long arm of chromosome 6.

As to the del(20)(q), we have identified a common region of deletion that includes the EIF6 gene in 12/12 patients analysed. The EIF6 loss was shown to be consistent with the possible improvement of ribosome biogenesis, and with a better hematological prognosis. Through a collaboration with the Centro Ricerca Tettamanti, University of Bicocca (Milan, Italy), by SNPs-arrays we have recently found an intriguing recurrent region of copy-neutral loss of heterozygosis (CN-LOH) in 3 patients, regarding the chromosome 20 and, very interestingly, encompassing the EIF6 gene.

The del(20)(q) is considered a good prognostic sign, associated with a lower risk of developing myelodysplasia (MDS) and/or acute myeloid leukemia, but also with a significantly milder hematological basic condition. These features are consistent with the EIF6 haploinsufficiency. To better investigate this assumption, we performed whole transcriptome expression studies on RNA from BM of 7 patients carrying the del(20)(q) at various clonal percentages. This study demonstrated that the patients with the del(20)(q) show an expression pattern more similar to normal subjects than SDS patients without the del(20)(q).
Abstract

Pancytopenia, myelodysplastic syndrome (MDS), or acute myeloid leukemia (AML) are life-threatening complications of Shwachman Diamond syndrome (SDS). To determine which lesions underlie clonal development and leukemic onset in SDS, we screened bone marrow samples from 42 patients with SDS using cytogenetics and targeted next generation sequencing (NGS) of 41 genes.

We found somatic TP53 mutations in 18/42 SDS cases with variant allele frequencies ranging from 0.5 to 83%. In one patient, a co-occurring mutation in FLT3 was detected, and in four patients, isolated somatic mutations in IDH1, SF3B1, and PHF6 were observed. Strikingly, TP53 mutations were seldom observed in non-SDS cases of the French Severe Chronic Neutropenia Registry. SDS patients without any detectable mutations (VAF<0.5%) had no severe hematological expression at the time of sampling. By contrast, among the 21 SDS patients with somatic mutations, AML or MDS were observed in 4 cases. Severe cytopenias without MDS or AML were found in two other cases, one with an isolated TP53 mutation at 24%, and one with both TP53 and FLT3 mutations around 45%. In the 12 remaining patients, allele frequencies of TP53 variants ranged from 0.5 to 37%. In several patients, concomitant cytogenetic and NGS analyses of either sequential or MDS/AML samples led us to build putative clonal evolutions. Our data suggest that TP53 mutations frequently initiate the clonal evolution process in SDS, with subsequent accumulation of chromosomal aberrations which lead to MDS and AMLs with complex karyotype.
Abstract

Myelodysplastic syndrome (MDS) is a clinically heterogeneous disease characterized by functional impairment of hematopoiesis and abnormal bone marrow morphology. The type and severity of hematopoietic dysfunction in MDS is highly variable and the kinetics of disease progression are difficult to predict. Genomic studies have shown that MDS is typically driven by a multi-step somatic genetic process affecting a core set of genes. Recurrent, somatic MDS driver mutations all drive clonal dominance, although they can have stereotyped positions in the clonal hierarchy or patterns of co-mutation association and exclusivity. Furthermore, environmental context, such as exposures to cytotoxic chemotherapy or the presence of germline predisposition, can influence disease pathogenesis and clinical outcomes.

To determine whether genetic mutations may predict clinical outcomes after allogeneic hematopoietic stem-cell transplantation, we performed targeted mutational analysis on samples obtained before transplantation from 1514 patients with MDS who were enrolled in the Center for International Blood and Marrow Transplant Research Repository. TP53 mutations were present in 19% of the patients and were associated with shorter survival and a shorter time to relapse. Among patients 40 years of age or older who did not have TP53 mutations, the presence of RAS pathway mutations was associated with shorter survival, owing to a high risk of relapse, and the presence of JAK2 mutations was associated with shorter survival, owing to a high risk of death without relapse. Notably, in young adults, 4% of the patients had compound heterozygous mutations in the Shwachman–Diamond syndrome–associated SBDS gene, despite the fact that most patients had no clinical diagnosis of SDS. Cryptic, biallelic SBDS mutations were associated with short stature, concurrent somatic TP53 mutations, and a poor prognosis compared to other young MDS patients. In follow-up studies, we are evaluating the spectrum of somatic drivers of clonal hematopoiesis and leukemia in SDS patients.
Abstract
Shwachman-Diamond Syndrome (SDS) is a rare autosomal recessive condition, associated with pancreatic and haematological abnormalities. Patients with SDS typically present with exocrine pancreatic dysfunction, poor growth / failure-to-thrive, weight loss and/or nutritional deficiency. An enteropathy affecting the small intestine of children with SDS has also been described. Identification, evaluation and management of nutritional issues in SDS will be discussed. A practical guide to management of exocrine pancreatic insufficiency will also be covered.

Finally, the results of an international survey on gastrointestinal and nutritional knowledge and issues affecting patients with SDS by gastroenterologists and dietitians will be presented.
Hepatic abnormalities in SDS
Daniel H. Leung, MD

Baylor College of Medicine1, One Baylor Plaza, Houston, TX 77030 and Texas Children’s Hospital2, 6621 Fannin St. MWT 1010, Houston, TX 77030

Abstract

Liver abnormalities, ranging from elevated liver biochemistries to acute liver failure have been reported in patients with Shwachman-Diamond Syndrome (SDS). Biochemical perturbances are notably more common in early childhood but spontaneously normalize with age. The etiology of hepatic inflammation during these early years in unclear and our current understanding of these hepatic abnormalities suggest minimal to no impact on morbidity or mortality. This may change as life span increases in people with SDS. It is important for SDS providers to understand the relevance of diagnostic laboratories and imaging from a hepatology perspective.
Pregnancy Complications in Women with Shwachman Diamond Syndrome

Neelam Giri, MD, MBBS and Blanche P Alter, MD, MPH

Clinical Genetics Branch, Division of Cancer Epidemiology and Genetics, National Cancer Institute, National Institutes of Health, Bethesda, MD

Abstract

Pregnancy in women with Shwachman Diamond syndrome (SDS) has not been well studied, with only 5 cases reported between 1991 to 2018. These women had more complications than expected, similar to other inherited bone marrow failure syndromes. Two women experienced miscarriages; 4 had 6 full-term pregnancies resulting in 7 live births (1 set of twins). Three women developed hematological complications including mild anemia, neutropenia and thrombocytopenia; one received platelet transfusions for thrombocytopenia. Poor maternal weight gain and fetal growth restriction were noted in one, which improved with resumption of pancreatic enzyme supplementation. One woman was required monitoring for hypoglycemia. Labor was induced in 4 at 36-40 weeks gestation; 3 underwent cesarean delivery for cephalopelvic disproportion/contracted pelvis, failure to progress or fetal distress due to chorioamnionitis. Post-partum course was complicated by excessive bleeding in 2 due to uterine atony and thrombocytopenia that required red blood cell transfusions. Six babies (1 set of twins) were healthy, 1 baby had hypoplastic left heart and intrauterine growth restriction. One woman subsequently died from ovarian adenocarcinoma.

Personal communication with SDS Foundation indicated an additional 6 women who had at least one pregnancy. These pregnancies were associated with cytopenia, particularly thrombocytopenia with excessive hemorrhage during labor. Poor fetal growth was also noted. Two women subsequently died from complications related to stem cell transplantation.

Pregnancy in patients with SDS is high-risk due to frequent multi-organ complications that require a multidisciplinary healthcare team including maternal-fetal medicine specialists, gastroenterologists, endocrinologists and hematologists with expertise in bone marrow failure syndromes. Involvement of multidisciplinary team should be at the earliest opportunity.
Novel myopathy revealed in a newborn with severe hypotonia and thoracic dysplasia
Alexandra Țopa, MD, MSc

Department of Clinical Pathology and Genetics, Sahlgrenska University Hospital, Gothenburg, Sweden
Department of Pathology and Genetics, University of Gothenburg, The Sahlgrenska Academy, Gothenburg, Sweden

Abstract

This is the case of a boy of Scandinavian origin with molecularly confirmed Shwachman-Diamond syndrome (SDS) and a rare neonatal presentation with pronounced hypotonia and severe thoracic dysplasia causing respiratory distress. The clinical picture initially suggested a neuromuscular disease and a muscle biopsy showed unspecific myopathic changes with prominent variability in muscle fiber size and abnormal expression of developmental isoforms of myosin. However, the skeletal presentation and other phenotypic features as pancreatic insufficiency and hematological abnormalities that occurred later on were suggestive for SDS. The clinical diagnosis was confirmed by the detection of compound heterozygous mutations in SBDS using whole-exome-sequencing. This method has allowed to screen for potentially co-occurring disease causing variants in neuromuscular genes; no such variants have been detected. Despite his severe condition, the boy survived with a long period of total dependency on assisted ventilation. The follow-up showed severe motor delay, but also a progressive amelioration of the respiratory status with need of assisted ventilation only during nighttime. This case illustrates the challenges in differential diagnosis of pronounced neonatal hypotonia with asphyxia and highlights the muscular involvement in SDS with, to our knowledge, the first documented myopathy in a patient with clinically and molecularly confirmed SDS.
The brain matters: neurological involvement and functioning in SDS

Elizabeth Kerr, BASc, MA., PhD., CPsych.

The Hospital for Sick Children and Department of Pediatrics, The University of Toronto.
Toronto, Ontario, Canada.

Abstract

SDS impacts all organs including the brain. This presentation addresses typical brain functioning, describes the little we know about the impact of SDS on brain matter, and summaries the consequences of alterations of matter on functioning. Specifically, the cognitive, behavioural, and adaptive functioning, that is, neuropsychological functioning, will be reviewed. Broad recommendations for supporting patients’ development are included for clinician involved in care. The cause of neurological involvement is not understood. A defect in ribosome biogenesis is raised as the potential antagonist of neurological functioning and consequently neurodevelopment in SDS. Elucidation of the mechanisms whereby abnormalities in ribosome biogenesis cause specific brain alterations in SDS remained to be studied.
Hematopoietic stem cell transplantation (HSCT) for children with Shwachman-Diamond Syndrome (SDS)

Stella M Davies

Cincinnati Children’s Hospital Medical Center, Cincinnati, OH

Abstract

Children with SDS have a wide range of clinical phenotypes, and only a minority will have sufficiently severe hematological abnormalities to warrant consideration of HSCT. Recent registry data have expanded SDS from a largely pediatric disorder to an important disorder of adults, with recognition of newly diagnosed SDS in adults, often presenting with myeloid malignancy, and it appears the diagnosis of SDS may be commonly overlooked or only recognized in the setting of excessive transplant toxicity. Moreover, children diagnosed with SDS in childhood are now reaching adulthood and further expanding the described phenotype. The published literature regarding transplant for SDS focuses largely on pediatric experience. The indication for transplant was generally cytopenias, and few transplants were performed for leukemia. While data for leukemia are few, it appears that tolerance of chemotherapy for AML in SDS is impaired and outcomes of treatment with or without transplant are disappointing. Identification of biomarkers that might signal later evolution to leukemia might improve survival if HSCT could be performed early, and support the use of annual screening marrow examinations for surveillance. Cytogenetic abnormalities have been challenging as markers indicating need for transplant because stable abnormalities such as iso-chromosome 7 or 20delq can be present for years without evolution.

Early pediatric studies, involving small numbers of children, described cardiac necrosis in SDS after exposure to transplant doses of cyclophosphamide. These data lead to the evolution of reduced intensity chemotherapy regimens, in general using alternative alkylating agent and avoiding cyclophosphamide. These data suggest reasonable outcomes with a majority of children with SDS transplanted prior to evolution to AML surviving. HSCT can be necessary and life-saving in children with critical cytopenias. It is important to recognize that HSCT is associated with important late effects, and families should be aware of this, and careful follow-up should be instituted to reduce late complications. Growth and impaired fertility are particular issues. Pre-transplant intervention with fertility preservation and serial monitoring after transplant to review fertility potential can reduce the impact of such effects.
**Abstract**

Shwachman-Diamond syndrome is a rare inherited disease mainly caused by mutations in SBDS gene and characterized by multiple organ involvement. Bone marrow failure and subsequent hematological issues are the main causes of morbidity and mortality in SDS. In fact, the Italian SDS registry reveals that almost 15% of SDS patients develop myelodysplasia or acute myeloid leukemia over the years. SDS patients currently undergo enzyme supplementation to restore normal pancreatic enzyme levels or to bone surgery to correct severe malformations. Unfortunately, no therapy able to counteract bone marrow failure has been developed so far. Since almost 50% of SDS patients present a nonsense mutation in one SBDS allele, we are currently developing a new therapeutic option based on a small nonsense suppressor molecule, namely ataluren. Ataluren is an orally available drug that has been already approved by EMA for the treatment of Duchenne muscular dystrophy. Our preliminary in vitro results show that ataluren can restore both SBDS protein expression and function in bone marrow hematopoietic progenitors and mesenchymal stromal cells, thus fostering a first explorative clinical trial. However, ataluren showed also some pitfalls due to its randomness in correcting the genetic defect by read-through mechanism. Thus, we are also testing other nonsense suppressor molecules with improved efficacy. But the next challenge would be the development of other therapeutic options aimed to correct other mutations that exist in SBDS gene as well as in other SDS-related genes such as DNAJC21, SRP54 and EFL1. In order to address this issue, we are designing novel strategies that can correct splicing mutations, including the pre-mRNA manipulation by U1 small nuclear RNA.
SDS iPS studies of BMF and 7q: 
To identify novel therapies

Akiko Shimamura, MD PhD

Dana-Farber/Boston Children’s Cancer and Blood Disorders Center
300 Longwood Avenue Boston, MA 02115

Abstract

Monosomy 7 or deletion of 7q (del(7q)) frequently arise in patients with Shwachman Diamond syndrome (SDS) and are associated with high grade MDS and leukemia which are major causes of mortality. Non-transplant treatments for marrow failure are complicated by potential stimulation of clonal outgrowth. We utilized induced pluripotent stem cells to model bone marrow failure and del(7q) MDS. This isogenic SDS+/- del(7q) model did not demonstrate a relative advantage of proliferation or hematopoiesis with 7q deletion in the context of marrow failure. RNA sequencing analysis revealed that the TGFβ pathway is hyperactivated in SDS and reduced in SDS del(7q). Inhibition of the TGFβ pathway improved hematopoietic colony formation and myeloid differentiation in the SDS iPSC without promoting outgrowth of the del7q line. These studies utilizing an iPSC model of BMF and MDS identified a potential therapeutic target for treatment of SDS.
Research Abstracts
Genetic Variation In The Major Shwachman-Diamond Syndrome Gene, Sbds
Sophie Rossini, Liliana Vertel Morales, Johanna M. Rommens

Program in Genetics & Genome Biology, Research Institute, The Hospital for Sick Children and Department of Molecular Genetics, University of Toronto, Toronto, ON Canada

Abstract

Shwachman-Diamond syndrome (SDS; OMIM#260400; also known as Shwachman syndrome, Shwachman-Bodian-Diamond syndrome, Pancreatic insufficiency and bone marrow dysfunction) is an autosomal recessive disease characterized by exocrine pancreatic dysfunction and bone marrow failure. Skeletal anomalies and neurodevelopmental and behavioral issues are also reported. Individuals with SDS present with malabsorption, malnutrition, growth failure and hematologic abnormalities including single- or multi-lineage cytopenias. Susceptibility to severe infections and acute myelogenous leukemia are concerning morbidities.

Loss of function variants in SBDS lead to SDS (Boocock et al, 2003 Nat Genet. 33: 97-101). SBDS maps to human chromosome 7, is comprised of five exons and encodes a highly conserved polypeptide of 250 amino acids. A dysfunctional and unprocessed pseudogene, SBDSP with 97% sequence identity, also resides on chromosome 7. Two common recurring variants, c.183_184delinsCT and c.258+2T>C that arise by gene conversion with SBDSP have been seen in all populations reporting cases of SDS. Rare missense, nonsense, deletion, insertion, splice site and genome rearrangement disease-associated variants have also been identified. Based on literature and public database surveys, at least 80 distinct SBDS disease-causing variants have been described. No individuals have been reported with two null alleles, highlighting that SBDS is an essential gene.

Recently, additional genes have been reported with loss of function variants that lead to clinical phenocopy or resemble SDS including DNAJC21 (Tummala et al, 2016 Am J Hum Genet. 99: 115-124; Dhanraj et al, 2017 Blood 129: 1557-1562; Bluteau et al, 2017 Blood Nov 16, Epub ahead of print), EFL1 (Stepensky et al, 2017 J Med Genet. 54: 558-566) and SRP54 (Carapito et al, 2017 J Clin Invest. 127: 4090-4103). The shared theme of disturbed ribosome biogenesis and function appear to underlie the consequent phenotypes. The number of SDS individuals without causal variants in SBDS remains unknown, but is less than 10%, with adherence to strict clinical diagnostic criteria as described in the most recent Consensus Guidelines (Dror et al, 2011 Ann NY Acad Sci. 1242: 40-55).

Presenting author name and contact information:
Johanna M Rommens, PhD
Program in Genetics & Genome Biology
Research Institute, The Hospital for Sick Children
PGCRL, Room 12.9716
686 Bay Street, Toronto, ON M5G 0A4 Canada
Phone: +1 416 813 7095  email: jrommens@sickkids.ca
Is Bone Marrow Failure syndrome (IBMFs) 3, a syndrome due to DNAJC21 mutation part of SDBS or a distinct IBMFs?

Guylaine D’Amours 1,2, Fátima D. Lopes 3, Julie Gauthier 4, Virginie Saillour 5, Christina Nassif 3, Nathalie Alos 6,8, Sonia Nizard 1,8, Emmanuelle Lemuye 1,8, Jacques L. Michaud 1,3,5,8, Véronique-Anne Pelletier 8, Jean-François Soucy 1,4,8, Yves D. Pastore 2,7,9

1 Service de Génétique Médicale, CHU Sainte-Justine, Montréal, Canada
2 Faculté de Médecine, Université de Montréal, Montréal, Canada
3 Centre de Recherche, CHU Sainte-Justine, Montréal, Canada
4 Laboratoire de Diagnostic Moléculaire, CHU Sainte-Justine, Montréal, Canada
5 Centre Intégré de Génomique Clinique Pédiatrique, Montréal, Canada
6 Service d’Endocrinologie, CHU Sainte-Justine, Montréal, Canada
7 Département de Pédiatrie, CHU Sainte-Justine, Montréal, Canada
8 Département de Pédiatrie, Université de Montréal, Montréal, Canada
9 Service d’Hématologie-Oncologie, CHU Sainte-Justine, Montréal, Canada.

Abstract

Introduction: DNAJC21 mutation has been first reported in 4 individuals as a new cause of IBMFs (IBMFs 3, OMIM #617052). A more recent report of additional patients presenting phenotypic similarities with patients suffering Shwachman-Diamond syndrome (SDBS) questions if DNAJC21 is a distinct IBMFs or part of the SDBS spectrum of disease. We present five additional patients, two siblings and three cousins from a First Nation community who presented multi-organ anomalies associated with bone marrow failure. We report their molecular data, phenotype, and clinical history, and compare with all individuals reported in the literature.

Case series: All of our patients were referred to the hematology and genetic clinic for evaluation of mild pancytopenia and multi-organ anomalies. This included congenital hip dysplasia, joint hypermobility, severe atypical eczema, neuro-developmental delay, postnatal growth retardation, and teeth anomalies; in addition, three had cerebral atrophy. None had evidence of pancreatic insufficiency. All had a homozygous DNAJC21 100A>G mutation (exome sequencing, confirmed by Sanger sequencing). Reduced telomere length <1st percentile was observed in all tested patients, predominantly in granulocytes and NK cells. Two patients required blood during the first months of life, but had gradual improvement in the pancytopenia after the age of one. Repeat bone marrow revealed mild aplastic anemia. Acquired loss of p53 mutation in bone marrow cells was observed in one patient with progressive mild macrocytosis but no other feature for myelodysplasia. To our knowledge, only one of the 10 reported patients to date had developed acute myeloid leukemia M7.

Discussion: Our case series may suggest that DNAJC21 is a distinct IBMFs. The improvement of the pancytopenia during the first year of life questions the role of DNAJC21 in the hematopoiesis. As other ribosomopathies, long term risk for MDS/LMA or SAA requires strict hematological and multidisciplinary follow-up, including pediatricians, geneticists and hematologists.

Presenting author name and contact information:

Yves D Pastore, MD
Hematology Service and Charles Bruneau Cancer Center, Pediatric Dpt, CHU Ste-Justine.
3175 Cote Ste-Catherine, Montreal (QC), Canada.
Phone: +1 5149 284 203 email: yves.pastore@umontreal.ca
Chromosome anomalies in bone marrow of patients with Shwachman-Diamond syndrome as successful or unsuccessful attempts to improve ribosome biogenesis

Pasquali F1*, Valli R1, Minelli A2, D'Amico G3, Frattini A1-4, Montalbano G1, Galbiati M2, Cazzaniga G3, Danesino C2, Maserati E1

1 Genetica Umana e Medica, Dipartimento di Medicina e Chirurgia, Università dell’Insubria, Varese, Italy
2 Genetica Medica, IRCCS Policlinico S. Matteo and University of Pavia, Pavia, Italy
3 Immunology and Cell Therapy, Centro Ricerca Tettamanti, Pediatric Clinic, University of Milan Bicocca/MBBM, Monza, Italy
4 IRGB, National Council of Research, Milano, Italy

Abstract

Biallelic mutations of the SBDS gene cause Shwachman-Diamond syndrome in most cases. In these patients, the cells of bone marrow seem to plan programmatically an improvement of their function, impaired by the defective SBDS protein, through the acquisition of chromosome anomalies. The attempts may be successful or unsuccessful, but a number of observations witness them. The success is reached when the two most frequent clonal anomalies arise: the i(7)(q10) (isochromosome of the long arm of chromosome 7) and the del(20)(q) (interstitial deletion of the long arm of chromosome 20), which imply consequently a benign prognosis. Two are the strategies followed.

1 - Attempt to duplicate the region of chromosome 7 with the gene SBDS: the i(7)(q10) may thus arise, and it includes in all cases analysed (11/11) a duplication of the mutation c.258+2T>C, milder because this mutation leads to a small amount of functional SBDS protein (success). An i(7)(q10) with the more severe mutation c.183_184TA>CT duplicated was never observed. We found and reported other different anomalies of chromosome 7 not clonal, but in single cells: they witness the attempts to rearrange the chromosome.

2 - Attempt to eliminate the region of chromosome 20 where the gene EIF6 is located, as it interacts with SBDS in ribosome biogenesis and the less the EIF6 protein is, the better for ribosome formation. This attempt leads to the del(20)(q) with loss of the EIF6 (success). Microsatellite analysis showed in three patients out of 11 with del(20)(q) results compatible with mitotic recombination in the region of EIF6, or near it. In six other patients out of 17, with del(20)(q) or i(7)(q10) or other anomalies, SNP array results showed copy-neutral loss of heterozygosity in the region of EIF6, or near it. These data witness the attempts to rearrange the chromosome to produce the del(20)(q).

Presenting author name and contact information:

Dr. Francesco Pasquali, professor emeritus
Genetica Umana e Medica, Dipartimento di Medicina e Chirurgia, Università dell’Insubria, Varese, Italy
Via J. H. Dunant, 5
21100 Varese, Italy
Phone: +39 0332 217180 email: francesco.pasquali@uninsubria.it
Mild haematological features in patients with deletion of the long arm of chromosome 20 acquired in bone marrow

Valli R1, Frattini A1,2, Montalbano G1, Minelli A3, Danesino C3, Pasquali F1*, Maserati E1

1 Genetica Umana e Medica, Dipartimento di Medicina e Chirurgia, Università dell’Insubria, Varese, Italy
2 IRGB, National Council of Research, Milano, Italy
3 Genetica Medica, IRCCS Policlinico S. Matteo and University of Pavia, Pavia, Italy

Abstract

In our cohort of 95 Italian patients with Shwachman-Diamond syndrome (SDS) followed-up since 1999, 17 bear an interstitial deletion of the long arm of chromosome 20, del(20)(q), in bone marrow (BM). In six of them, we already reported the results of array-comparative genomic hybridization (a-CGH), and we demonstrated the consistent loss of the gene EIF6: we postulated that ribosome biogenesis is consequently more efficient, leading to a lower risk of developing Myelodysplastic Syndrome (MDS) and Acute Myeloid Leukaemia (AML). We update here our results leading the total number of patients with del(20)(q) to twelve. We confirm that all deletions are interstitial, that their proximal breakpoints are clustered in a rather small region of about 2,600 Kb, while the distal breakpoints are more variable. Excluding two patients, who present a very small deletion, with loss of 4,150 and 4,700 Kb respectively, the distal breakpoints in the other ten patients cluster in a segment of 11,227 Kb, and the material lost is in the range 14,008 – 26,863 Kb. The gene EIF6 is lost in all the 12 patients.

The loss of EIF6 due to the del(20)(q) is confirmed as a good prognostic sign: none of these twelve patients encountered a transformation into MDS/AML, nor an evolution into severe BM aplasia.

We considered some basic information on the haematological condition of these patients at the time of a-CGH analysis, together with their age and with the proportion of BM cells bearing the del(20)(q). We concluded that the benign prognostic significance of del(20)(q) and loss of EIF6 may be expanded to the basic haematological features of these patients: BM picture and peripheral blood counts give evidence of a very mild condition in all our patients, both as to BM morphology and to peripheral blood cytopenias, which are absent or mild.

Presenting author name and contact information:

Dr. Francesco Pasquali, professor emeritus
Genetica Umana e Medica, Dipartimento di Medicina e Chirurgia, Università dell’Insubria, Varese, Italy
Via J. H. Dunant, 5
21100 Varese, Italy
Phone: +39 0332 217180 email: francesco.pasquali@uninsubria.it
Whole Exome Sequencing Discloses Heterozygous Variants In Dnajc21 (2 Cases) And Efl1 (5 Cases) But Not In Srp54 In 16 Patients With Shwachman Diamond Syndrome Carrying Biallelic Sbds Mutations

Jacopo Morini 1*, Lucia Nacci 2*, Gabriele Babini 1*, Simone Cesaro 3, Roberto Valli 4, Andrea Ottolenghi 1, Elena Nicolis 5, Emily Pintani 5, Emanuela Maserati 4, Claudia Scotti 2, Marco Cipolli 6, Cesare Danesino 2, Antonella Minelli 2.

*equally contributed

1: JM; GB; AO; CS; Department of Physics, University of Pavia, Pavia, Italy; 2: LN;CD; AM; Department of Molecular Medicine, University of Pavia, Pavia, Italy; 3: SC; Oncoematologia Pediatrica, Azienda Ospedaliera Universitaria Integrata, Verona, Italy; 4 RV; EM; Department of Clinical and Experimental Medicine, University of Insubria, Varese, Italy; 5: EN; EP Cystic Fibrosis Centre, Azienda Ospedaliera Universitaria Integrata, Verona, Italy; 6:Cystic Fibrosis Regional Centre, Ospedali Riuniti, Ancona, Italy

Abstract

Purpose: to investigate if variants in the newly identified genes are present in SDS patients carrying SBDS mutations.

Shwachman Diamond Syndrome (SDS) is a rare autosomic recessive disorder (OMIM 260400) first described in 1964 by Shwachman and coworkers. Boocock et al. identified the disease causing gene, SBDS (OMIM 607444), localized on chromosome 7q11.21.

No genetic heterogeneity was known until 2017 when Dhanraj et al. reported biallelic mutations in DNACJ21; shortly after, Stepensky et al. reported biallelic mutations in EFL1 in four cases and Carapito et al. reported de novo dominant mutations in the SRP54 gene, in three independent patients.

Methods: Exome sequencing and modelling of the variations in identified DNACJ21 and EFL1.

Results: We analysed our data about whole exome sequencing (WES) in 16 patients with biallelic mutations in SBDS (9 cases [183-184TA>CT / 258+2T>C]; 6 cases [258+2T>C / rare mutation]; 1 case homozygous for 258+2T>C). Among them, we found two cases carrying rare heterozygous SNPs in DNAJC21 and five cases carrying three heterozygous SNPs in EFL1. All SNPs were predicted as pathogenetic in at least two among Polyphen-2; SIFT; Provean; Mutation Taster; SNAP2. SRP54 did not contain any SNP.

We confirmed all variants by Sanger sequencing, and demonstrated that all of them are inherited from one of the parents. DDG (Delta Delta G, relative free energy) assessment demonstrated that the variants may affect the stability of the protein.

Conclusions: the genetic heterogeneity recently demonstrated in SDS indicates how genetic analysis in patients with a phenotype suggestive of SDS, and no mutation in SBDS, should be extended at least in the newly recognized disease causing genes. Moreover, mutations additional to the classic ones can be found and that their clinical significance, if any, should also be investigated.

Presenting author name and contact information:

Prof. Cesare Danesino
Molecular Medicine Department – University of Pavia
Via Forlanini 14,
27100 Pavia , Italy
Phone: +39 0382 987737 email: cidi@unipv.it
TP53 mutations in Shwachman-Diamond Syndrome

Alyssa L. Kennedy 1, R. Coleman Lindsley 2, Kasiani C. Myers 3, Stella Davies 4, Akiko Shimamura 5

1, 5 Dana Farber/Boston Children’s Hospital Cancer and Blood Disorders Center, 450 Brookline Avenue, Boston, MA, USA. 2 Department of Medical Oncology, Division of Hematological Malignancies, Dana Farber Cancer Institute 450 Brookline Avenue, Boston, MA, USA 3, 4 Cincinnati Children’s Hospital Medical Center, 3333 Burnet Ave, Cincinnati, OH, USA.

Abstract

Patients with Shwachman-Diamond syndrome (SDS) have a high risk of developing myelodysplastic syndrome (MDS) and acute myeloid leukemia (AML). A collaborative study coordinated by the SDS Registry found that progression to leukemia carries an especially poor prognosis for SDS patients. Enhanced surveillance strategies that can identify patients at particularly high risk for progression could improve overall outcomes by highlighting patients that would benefit from early transplantation. Although genomic analyses of MDS and AML in the general population have identified somatic mutations that inform risk stratification, little is known about the somatic mutational landscape in SDS. Recent studies have shown that somatic TP53 mutations are very common in SDS patients with MDS, but TP53 mutations are also detectable in patients without documented clinical progression, suggesting that other clinical or genetic factors contribute to transformation. To investigate the molecular and biological characteristics of TP53 mutations in SDS and define the prognostic implications of somatic TP53 mutations in patients with SDS, we first performed whole exome sequencing of bone marrow samples from twenty-three patients from the SDS registry. We identified somatic TP53 alterations in 8 of the 23 patients (34%). To investigate the effects of TP53 on hematopoiesis in SDS, we introduced p53 mutations into SDS patient-derived induced pluripotent stem cells (iPSCs). We extended these studies to hematopoietic stem/progenitor cells using shRNAs to knockdown both SBDS and TP53. We are currently assaying the effect of TP53 mutations on proliferation, survival, checkpoint controls including those dependent on the DNA damage response, and hematopoietic differentiation of these SBDS-deficient iPSC and CD34 cells. Next, we plan to assess TP53 somatic mutations in serial samples from 128 SDS patients in the SDS Registry and BMF Registry to determine whether TP53 mutations are associated with risk of clonal progression and adverse clinical outcomes in patients with SDS.

Presenting author name and contact information:

Dr. Alyssa L. Kennedy
Dana Farber/Boston Children’s Hospital Cancer and Blood Disorders Center
450 Brookline Avenue, Boston, MA, USA
Phone: +1 617 919 1456 +1 617 632 4410 email: Alyssa_Kennedy@dfci.harvard.edu
Energetic basis of the nucleotide-affinity regulation of EFL1 by the SBDS protein
Axel Luviano 1, Roberto Castañeda-Cruz 1, Nuria Sánchez-Puig 1 and Enrique García-Hernández 1

1 Instituto de Química Universidad Nacional Autónoma de México, Circuito Exterior, Ciudad Universitaria, México 04630, D.F., México

Abstract

The binding energetics of yeast Efl1 to guanosine nucleotides and Sdo1 (the yeast ortholog of SBDS) were characterized by means of high-sensitive isothermal titration calorimetry. The binding of both nucleotides to isolated Efl1 are enthalpy and entropy driven and exhibited significant dependence on temperature.

Dissection of the ΔCpb values revealed that upon GDP/MgGDP binding, Efl1 undergoes a conformational change accompanied by the burying of ~1200 Å2 of surface area (~27/25 residues). In contrast, GTP/MgGTP binding elicits an even larger conformational change, involving the burying of ~3100/2400 Å2 of surface area (~66/51 residues). This is in agreement with the number of rotational bonds changing calculated from deconvolution of the conformational entropy where the GTP complexes are twofold larger than those involved in the formation of GDP complexes. SBDS has been identified to function as a GEF for EFL1, i.e., it strengthens the interaction with GTP, and weakens that with GDP. 1-2 This result was confirmed using calorimetry and provided information on the thermodynamics of the process. Calculation of cooperative heterotropic parameters indicate that the Sdo1-induced affinity loss for GDP is due to a somewhat unfavorable entropic effect, while the affinity gain for GTP is entropically driven, although counterbalanced by a significant endothermic enthalpy change. Additionally, SBDS pre-induces a conformational change in Efl1 that makes GTP binding an event in which the net changes in ∆ASA and the number of free rotatable bonds are significantly reduced in relation to the changes seen with isolated Efl1, while the opposite behavior is observed for the interaction with GDP.


Presenting author name and contact information:

Dr. Nuria Sánchez-Puig
Instituto de Química Universidad Nacional Autónoma de México,
Circuito Exterior, Ciudad Universitaria,
México 04630,
D.F., México
Phone: +52 1 55 269 67605  email: nuriasp@unam.mx
**Shwachman-Diamond Syndrome: Saxs, Inside The Structure Of The Ribosomal Gtpase Efl1, Sbds And Their Complex**

Dritan Siliqi *, Davide Altamura 1, Abril Gijsbers 2, Alfonso Méndez-Godoy 3, Cinzia Giannini 1, Teresa Sibillano 1, Michele Saviano 1, Nuria Sanchez-Puig 3

1 Istituto di Cristallografia, CNR, Via G. Amendola 122/O, 70126 Bari, Italy;  
2 Maastricht MultiModal Molecular Imaging Institute (M4I), Universiteitssingel 50, 6229 ER Maastricht, Netherlands  
3 Instituto de Química, Departamento de Química de Biomacromoléculas, Universidad Nacional Autónoma de México, Ciudad Universitaria, 04510 México, DF, México

**Abstract**

Ribosome biogenesis is closely linked to the cell growth and proliferation. Dysregulation of this process causes several diseases collectively known as ribosomopathies. One of them is the Shwachman-Diamond Syndrome, and the SBDS protein mutated in this disease participates with EFL1 in the cytoplasmic maturation of the 60S subunit. We have shown that the interaction of EFL1 with SBDS resulted in a decrease of the Michaelis-Menten constant (KM) and increase the dissociation constant for GDP and thus SBDS acts as a GEF for EFL1 (1). The interaction of EFL1 alone or in complex with SBDS to guanine nucleotides is followed by a conformational rearrangement. Mutations in the SBDS proteins are present in 90% of patients, however, the etiology of the remaining patients was unknown. We have shown that surface missense mutations disrupt the binding to EFL1 with the consequent loss in guanine exchange regulation. Recently together with two other clinician groups, we described two mutations in EFL1 in patients with SDS clinical symptoms [2]. In this study, we aim to show the conformational changes resulting from the interactions between EFL1 and its binding partners using SAXS technique (3). As well, some screening of small organic molecules capable of modulating the function of EFL1 will be shown.


The authors acknowledge financial support PGR2015-2017 Italy-Mexico, MAECI and the CNCCS Consortium Project “Collezione di Composti Chimici ed attività di screening”

**Presenting author name and contact information:**

Dr. Dritan Siliqi  
Istituto di Cristallografia, Consiglio Nazionale delle Ricerche (IC-CNR)  
Via G. Amendola, 122/O  
70216 Bari, Italy  
Phone: +39 080 59 29 164  email: dritan.siliqi@ic.cnr.it
Shwachman-Diamond Syndrome Cells Have Reduced Homology-Directed Repair

Elif Asik 1, Nimrat Chatterjee1,2, and Alison A. Bertuch1
1Department of Pediatrics, Hematology/Oncology, Baylor College of Medicine, Houston, TX
2Present address: Department of Biology, Massachusetts Institute of Technology, Cambridge, MA

Abstract

Shwachman-Diamond syndrome (SDS) is known principally as a ribosomopathy, with impaired ribosomal joining and decreased 60S subunits. Although ribosome biogenesis and DNA repair are considered distinct biological pathways, accumulating evidence suggests that ribosomal defects, such as in SDS, may be associated with decreased DNA damage repair. The mechanism by which SBDS deficiency impacts the repair of DNA damage warrants further study as it may contribute to bone marrow failure and malignant transformation in patients with SDS. We found lymphoblastoid cell lines (LCLs) derived from patients with SDS were hypersensitive to γ- or ionizing radiation (IR) with decreased colony survival and increased apoptosis. This hypersensitivity was associated with an increase in γ-H2AX, p53, phospho-ATM, and DNA-PKcs protein levels compared to controls. We also found that SDS LCLs had a delay in resolution of γ-H2AX foci and protein levels at 1 and 24 hrs after 2 and 10 Gy IR as compared to control LCLs. We hypothesized that the decreased survival, and increased and sustained DNA damage response was due to a defect in one or more pathways of DNA double strand break (DSB) repair. To investigate this, we utilized U2OS and HCT 116 (nonhematopoietic) cells containing different integrated GFP reporter transgenes to monitor the repair of an I-SCE1-induced DSB via two types of homology-directed repair (HDR), gene conversion (GC) and single-strand annealing (SSA). Following induction of the DSB in SBDS-depleted and control cells, proficiency of DSB repair was assessed by measurement of GFP+ cells by flow cytometry. Compared to the scrambled control, SBDS depletion significantly reduced DSB repair by GC and SSA. Collectively, these findings indicate for the first time deficiency of SBDS impairs HDR. Ongoing studies are examining the mechanism underlying this reduction in HDR and assessing whether the nonhomologous end-joining pathway of DSB repair is similarly affected.

Presenting author name and contact information:
Elif Asik, PhD
Department of Pediatrics, Hematology/Oncology, Baylor College of Medicine
TX Childs Feign Center, Room C1240.09, MS: BCM320
Houston, TX 77030
Phone: +1 832 824 4539 email: Elif.Asik@bcm.edu
Sbds-Deficient Zebrafish Phenocopies Human Shwachman-Diamond Syndrome And Shows P53 Pathway Activation

U Oyarbide 1, W Amaya-Mejia 1, Stillman A 1, J Topczewski 2,3, SJ Corey 1.

1Departments of Pediatrics, Microbiology/Immunology, Human and Molecular Genetics, VCU Massey Cancer Center and Children's Hospital of Richmond; 2Program in Developmental Biology, Stanley Manne Children's Research Institute, 3Department of Pediatrics, Northwestern University's Feinberg School of Medicine.

Abstract

Shwachman-Diamond Syndrome (SDS) is rare inherited recessive disorder characterized by exocrine pancreatic insufficiency, bone marrow dysfunction, and an increase risk of acute myeloid leukemia. Patients with SDS may exhibit abnormalities in other organs, including bone, liver and cognitive disorders. Most cases of SDS result from biallelic mutations in the SBDS gene. SBDS is involved in ribosomal biogenesis and stabilization of mitotic spindle microtubules and cell division. The precise mechanisms for its pathophysiology remain to be determined.

Sbds-knockout mice result in early embryonic lethality. We created a zebrafish line null for Sbds that phenocopies the human SDS. Immunoblotting showed a progressive decrease of Sbds during the first 8 days post fertilization (dpf), with absence at 10 dpf. These results suggest that larval viability is due to maternal deposition of sbds transcript and corresponding protein, which was confirmed by RT-qPCR and immunoblot at earliest developmental stages. Homozygous mutant fish live up to 4 weeks. They display less cell proliferation in digestive epithelia at 10 dpf and growth retardation from 15 dpf. Neutropenia was observed as early as 5 dpf. Pancreas, liver, digestive tract, and eye showed histologic evidence of atrophy at 21 dpf.

RNA-seq analysis showed that Sbds-deficiency activated the p53 pathway (upregulation of p53, p21, bax and puma). In addition, we found an increase of expression of genes involved in autophagy (atg5, atg7 and mTOR) and lipid metabolism (pparg, ppara, and srbp1). To further study dysregulation of p53 and function of Sbds to stress, we exposed 5 and 10 dpf fish to UV irradiation. We observed an abnormal response after irradiation in mutants comparing to control by 10 dpf.

We conclude Sbds deficiency activated the p53 tumor suppressor pathway in zebrafish, that can cause aberrant proliferation, organ malformation and starvation that leads to dead. Our results make it a highly relevant model to investigate its pathophysiology and develop new small molecule therapies.

Presenting author name and contact information:

Usua Oyarbide, PhD
Virginia Commonwealth University
401 College st GRL 278
Richmond, Virginia 23298
Phone: +1 312 927 7137 email: Usua.oyarbide@vcuhealth.org
Impaired pre-sequence processing is associated with reduced superoxide dismutase 2 activity in the yeast model of Shwachman-Diamond syndrome

Ayushi Jain 1*, The Phyu 1, Laran T. Jensen 2, Amornrat Naranuntarat Jensen 1

1Department of Pathobiology, Faculty of Science, Mahidol University, Bangkok 10400 Thailand; 2Department of Biochemistry, Faculty of Science, Mahidol University, Bangkok 10400 Thailand

Abstract

Yeast lacking Sdo1p, the S. cerevisiae homologue of human SBDS, display increased oxidative damage to mitochondrial proteins. Both the abundance and activity of superoxide dismutase 2 (Sod2p), a key mitochondrial antioxidant enzyme, are significantly reduced in sdo1Δ cells. The reduction of Sod2p protein and its activity do not appear to result from impaired mitochondrial protein import as a YFP fusion targeted to the mitochondrial matrix using the Cox4p pre-sequence shows normal mitochondrial localization in sdo1Δ cells. Active Sod2p can be formed in sdo1Δ cells when SOD2 is over-expressed using a strong promoter indicating that the pathways required to generate functional Sod2p are largely intact. However, immature forms of Sod2p are present in the sdo1Δ strain, suggesting that processing of the Sod2p pre-sequence is impaired. Cells lacking pre-sequence protease Cym1p are known to accumulate immature forms of Sod2p and cym1Δ yeast exhibit a reduction in both Sod2p activity and protein levels similar to that observed in sdo1Δ cells. Overall, our results implicate impaired processing of the Sod2p pre-sequence in sdo1Δ cells on reduced activity of this enzyme. Loss of Sod2p activity in mammalian cells is known to impair the function of several critical cellular pathway and a complete deletion of SOD2 leads to neonatal lethality. We propose that a reduction of Sod2p activity in SDS patients may have an impact on disease progression.

Presenting author name and contact information:

Miss Ayushi Jain
Department of Pathobiology, Faculty of Science, Mahidol University
272 Rama VI Rd., Ratchathewi District
Bangkok 10400, Thailand
Phone: +66 851282731  email: ayushi225@hotmail.com
Decreased accumulation of superoxide dismutase 2 within mitochondria in the yeast model of Shwachman-Diamond syndrome

Laran T. Jensen 1, The Phyut 2, Ayushi Jain 2, Chonlada Kaewwanna 2, Amornrat Naranuntarat Jensen 2*

1Department of Biochemistry, Faculty of Science, Mahidol University, Bangkok 10400 Thailand;
2Department of Pathobiology, Faculty of Science, Mahidol University, Bangkok 10400 Thailand

Abstract

The SBDS protein participates in ribosome biogenesis; however, effects beyond reduced translation efficiency are thought to be involved in SDS progression. Impaired mitochondrial function and mitochondrial DNA (mtDNA) loss have been reported for cells lacking either SBDS or Sdo1p, the Saccharomyces cerevisiae SBDS ortholog. To better understand how loss of SBDS/Sdo1p leads to mitochondria damage, we utilized the S. cerevisiae model of SDS. Yeast deleted for SDO1 show increased oxidative damage to mitochondrial proteins and a marked decrease in protein levels and activity of mitochondrial superoxide dismutase 2 (Sod2p), a key enzyme involved in defense against oxidants. A rho0 strain, lacking mtDNA, exhibited Sod2p activity similar to WT, suggesting that reduced Sod2p activity in sdo1Δ cells is not due to mtDNA loss. Reduced activity of Sod2p does not appear to be associated with impaired ribosome biogenesis as the expression of TIF6 gain-of-function mutant which partially bypass the reduced translation in sdo1Δ did not significantly alter either Sod2p protein levels or activity. In addition, chemical inhibition of protein translation does not result in loss of Sod2p activity or protein levels, in support of the Sod2p defect being independent of impaired translation in sdo1Δ cells. Sod2p protein levels and activity are largely restored in a por1Δ sdo1Δ strain, lacking the major mitochondrial voltage dependent anion channel (VDAC) found overexpressed in cells lacking Sdo1p. Overall, it appears that over-abundance of Por1p contributes to loss of the key antioxidant enzyme Sod2p and this may contribute to mitochondrial insufficiency in sdo1Δ cells.

Presenting author name and contact information:

Dr. Amornrat Naranuntarat Jensen
Department of Pathobiology, Faculty of Science, Mahidol University
272 Rama VI Rd., Ratchathewi District
Bangkok 10400, Thailand
Phone: +66 841236118  +66 23547158  email: amornrat.nar@mahidol.ac.th
Osteoblast functionality in patients with Shwachman-Diamond syndrome

Annalisa Frattini1,2, Isabella Villa3, Roberto Valli4*, Michela Signo5, Maria Rita Pinto4, Marco Zecca5, Maria Rita Frau6, Alessandro Rubinacci3, Emanuela Maserati1, Francesco Pasquali2

1 Human and Medical Genetics, Department of Medicine and Surgery, University of Insubria, Varese, Italy
2 IRGB, National Council of Research, Milan, Italy
3 Bone Metabolism Unit, San Raffaele Scientific Institute, Milan, Italy
4 Department of Pediatric Hematology/Oncology and Transfusion Medicine, Bambino Gesù Hospital IRCCS, Rome, Italy
5 Department of Pediatric Oncohematology, IRCCS Policlinico San Matteo, Pavia, Italy
6 Pediatric and Intensive Care Unit, San Francesco Hospital, Nuoro, Italy

Abstract

Skeletal anomalies in Shwachman-Diamond syndrome (SDS) are present in about half of the patients, and include various features, as metaphyseal dysostosis, abnormal development of growth plates, delay of secondary center of ossification, thoracic dystrophies, early-onset low turnover osteoporosis, delayed bone age. Poor growth manifested by both short stature and/or abnormal weight gain is also common, and may present even at birth.

Although the defect in growth plate may suggest altered function of chondrocytes, we analyzed if also the function of osteoblasts (OBs) may be compromised in SDS patients.

We obtained bone biopsies from eight SDS patients (5 males and 3 females, age range 9-43 years, all carrying bi-allelic mutations of the SBDS gene) and from five healthy donors (2 males and 3 females, age range 5-24 years).

Western blot analysis showed a reduced/undetectable SBDS protein amount in all the patients.

The expression of SBDS and of other two genes involved in ribosome biogenesis (EIF6 and EFL1), analysed by qRTP−CR, showed a decreasing trend, and it was significant (p<0.01) for SBDS.

The expression of genes involved in the differentiation and maturation of OBs (RUNX2, OSX, OPN and Col1A) was evaluated by qRTPCR in OBs primary culture. In basal culture condition, SDS-OBs showed a similar behaviour compared to healthy OBs.

When cultured in osteogenic medium, SDS-OBs showed a reduced ability to mineralize compared to control cells. At variance, osteogenes (RUNX2, OSX, OPN, ALP, BSP) mRNA were upregulated, suggesting a positive feedback due to reduced protein synthesis.

In conclusion, according to the function of SBDS in ribosome biogenesis, we may hypothesize that in unstressed condition SDS-OBs are able to fulfill cellular needs, while in stressed condition they may not, result consistent with the skeletal condition of SDS.

Presenting author name and contact information:

Dr. Roberto Valli
Human and Medical Genetics, Department of Medicine and Surgery – University of Insubria
Via J. Dunant, 5
21100 Varese - Italy
Phone: +39 0332 217112  email: roberto.valli@uninsubria.it
A nationwide cohort for Shwachman-Diamond syndrome in Japan
Kenichiro Watanabe1, Hirokazu Kanegane2, Takayuki Hamabata3, Kagehiro Kozuki3, Katsutsugu Umeda4, Asahito Hama4 Yusuke Okuno4, Hideki Muramatsu4, Yoshiyuki Takahashi5, Daisuke Hasegawa5, Atsushi Manabe5, Akira Ohara6, Masafumi Ito7, Seiji Kojima4, Etsuro Ito8

1 Department of Hematology and Oncology, Shizuoka Children’s Hospital
2 Department of Pediatrics and Developmental Biology, Graduate School of Medical and Dental Sciences, Tokyo Medical and Dental University
3 Department of Pediatrics, Graduate School of Medicine, Kyoto University
4 Department of Pediatrics, Nagoya University Graduate School of Medicine
5 Department of Pediatrics, St. Luke's International Hospital
6 Department of Transfusion Medicine, Toho University School of Medicine
7 Department of Pathology, Japanese Red Cross Nagoya First Hospital
8 Department of Pediatrics, Hirosaki University Graduate School of Medicine

Abstract

Background: Shwachman-Diamond syndrome (SDS) is an inherited bone marrow failure syndrome (IBMFS) characterized by bone marrow failure and pancreatic insufficiency. Although it is the third most common IBMFS following Fanconi anemia and Diamond-Blackfan anemia in western countries, the incidence of SDS has been considered to be much lower in Japan. Patients and methods: In addition to the cases of SDS diagnosed by SBDS gene mutation analysis at Kyoto University, we captured those from the central review system of the Japanese Society of Pediatric Hematology and Oncology and the targeted sequencing system for IBMFS at Nagoya University as well as the previous Japanese nationwide survey for SDS that was conducted in 2010. Results: A total of 45 cases with biallelic SBDS gene mutation were captured. After the previous nationwide survey, 19 cases were accumulated in the past 7 years: 2.7 cases per year in average. Male-female ratio of the patients was 2.2:1. Most common mutation was 183-184TA>CT/258+2T>C (73%) followed by 258+2T>C /258+2T>C (6.6%). Clinical features at initial visit were various kinds of cytopenia, failure to thrive, steatorrhea, liver dysfunction, short stature, and skeletal abnormalities. Pancreatic insufficiency and/or pancreatic abnormality in imaging studies were found in almost all patients. Neutropenia was documented in one-third of the cases at initial visit; however, eventually 89% of the patients had neutropenia. Other hematological abnormalities were anemia (64%), thrombocytopenia (69%), and pancytopenia (40%). Six percent of the cohort was documented to develop overt leukemia at present. Conclusion: Along with growing recognition of the disease among physicians and establishment of mutation analysis system, newly diagnosed cases of SDS were accumulated constantly. The SDS cohort in Japan may be useful to clarify clinical and genetic features of the patients with relatively uniform genetic background that is distinct from European ethnicity.

When cultured in osteogenic medium, SDS-OBs showed a reduced ability to mineralize compared to control cells. At variance, osteogenes (RUNX2, OSX, OPN, ALP, BSP) mRNA were upregulated, suggesting a positive feedback due to reduced protein synthesis.

In conclusion, according to the function of SBDS in ribosome biogenesis, we may hypothesize that in unstressed condition SDS-OBs are able to fulfill cellular needs, while in stressed condition they may not, result consistent with the skeletal condition of SDS.

Presenting author name and contact information:

Dr. Kenichiro Watanabe
Department of Hematology and Oncology, Shizuoka Children’s Hospital
860 Urushiyama, Shizuoka
420-8660 JAPAN
Phone: +81 54 247 6251   email: wataken@kuhp.kyoto-u.ac.jp
The Shwachman-Diamond Syndrome Registry: What Have We Learned And Where Are We Going?

Kasiani C. Myers, MD, Elissa Furtani, MD, Sara Loveless, RN, Maggie Malsch, RN, Ashley Galvin, Ellie Fratt, Jordan Larson, Jacob Cotton, Leah Cheng, Kelly Mcintosh, RN, Joan Moore, RN, Leann Mount, RN, Stella Davies, MBBS, PhD, MRCP, Akiko Shimamura MD, PhD

1 Shwachman-Diamond Syndrome Registry (SDSR), Dana-Farber/Boston Children’s Cancer and Blood Disorders Center and Cincinnati Children’s Hospital Medical Center, Cancer and Blood Diseases Institute

Abstract

The Shwachman-Diamond Syndrome Registry (SDSR) was established in December 2008 with the goal of understanding the natural history and biology of SDS to improve the lives of people with SDS. The SDSR has enrolled 208 patients with biallelic SBDS mutations or SDS-Like features. Longitudinal data has been collected for 160 patients with a median duration of follow-up of 11.3 (0.3-52.8) years. 110 individuals carry biallelic SBDS mutations, one with DNAJC21 mutations, none with EFL1 or SRP54 mutations and 97 individuals remain genetically undefined. Ongoing characterization of SBDS mutation-negative individuals has identified subgroups of clinically defined SDS individuals, as well as a more heterogenous subgroup with features of SDS but do not meet classic diagnostic criteria. In those with SBDS mutations overall survival is 88% with deaths mainly caused by MDS and AML. A detailed analysis of hematologic complications is ongoing with the goal of improving surveillance and treatment strategies. The range of non-hematologic phenotypic severity differs significantly from that reported in the literature. The outcomes of adults with SDS are a major focus of the SDSR. Outreach by the SDSR to the lay community has disseminated clinical updates, cutting edge research advances, and clinical trials for SDS patients. Outreach by the SDSR to the medical community has resulted in collaborative studies to improve outcomes for SDS. The SDSR has also organized collaborative biological studies of the causes of bone marrow failure and MDS/AML and has identified potential novel therapeutic strategies. The SDSR also provides a platform for future clinical trials. The SDSR is a critically important resource for clinical and biological research, education and outreach and will continue to engage families and physicians to advance our understanding of genetic etiology, disease pathophysiology, and to develop effective treatment strategies.

Presenting author name and contact information:

Dr. Kasiani C. Myers
Cincinnati Children’s Hospital Medical Center
3333 Burnet Ave. MLC 11027
Cincinnati, OH 45229
Phone: +1 513 803-3218  +1 513-803-1969  email:Kasiani.myers@cchmc.org
Abstract

Shwachman-Diamond syndrome (SDS) is associated with myelodysplasia (MDS) and acute myeloid leukemia (AML). This multi-institutional retrospective study investigated clinical features, treatment, and outcomes of 38 SDS patients who presented with AML (n=9), MDS-EB1/2 (n=5) or MDS (n=23). One patient had an isolated somatic p53 mutation. Blinded central pathology review was performed.
Median ages in years (range) at diagnosis were 16 (0.5-30) for MDS, 9 (0.7-20) for MDS-EB/1.2, and 28.8 (5.5-47) for AML. One MDS-EB1 and 1 MDS patient progressed to AML. The leukemias had complex cytogenetic abnormalities with the exception of one case with normal cytogenetics. Follow-up was available for 10 AML patients; 9 are deceased. Nine had received cytoreductive chemotherapy in preparation for hematopoietic stem cell transplant (HSCT). Four failed to achieve remission and died with disease prior to transplant. One proceeded to HSCT without chemotherapy. Four of six transplanted subjects died with relapsed disease. Treatment-related mortality was largely infectious or GVHD. The sole surviving AML patient had normal cytogenetics, achieved remission, then underwent HSCTs with 3 separate stem-cell infusions for primary graft failures.

Four of the 5 MDS-EB1/2 patients underwent RIC HSCT, 3 are alive and 1 died of infection. The fifth patient has stable disease on decitabine monotherapy for 4.75 years.

Of the nineteen MDS patients with treatment data, 13 had upfront HSCT therapy, 2 had upfront chemotherapy, and 4 had no therapy. 3 required ≥2 HSCTs due to graft failure. Follow-up is available for 18, 11 of whom are deceased, 6 with relapsed disease. Treatment-related mortality was largely infectious or graft failure. 7 MDS patients are alive in remission.

In summary, prognosis is poor for SDS patients who develop AML due to resistant disease and treatment-related complications. Better markers for risk stratification to inform early transplant and novel therapeutic strategies are urgently needed to improve outcomes.

Presenting author name and contact information:

Dr. Kasiani C. Myers
Cincinnati Children’s Hospital Medical Center
3333 Burnet Ave. MLC 11027
Cincinnati, OH 45229
Phone: +1 513 803 3218   +1 513 803 1969   email: Kasiani.myers@cchmc.org
Immunophenotypic analysis of peripheral blood obtained from a cohort of patients affected by Shwachman-Diamond syndrome

Antonio Vella¹*, Valentino Bezzerrì², Gianfranco Di Gennaro³, Riccardo Ortolani¹, Elena Nicolis⁴, Vincenzo Bronte¹ and Marco Cipolli⁵

¹Unit of Immunology, Azienda Ospedaliera Universitaria Integrata di Verona, Italy; ²Department of Medicine, University of Verona, Italy; ³Department of Pathology and Diagnostics, Azienda Ospedaliera Universitaria Integrata di Verona, Italy; ⁴Unit of Transfusion Medicine, Azienda Ospedaliera Universitaria Integrata di Verona, Italy; ⁵Cystic Fibrosis Center, Azienda Ospedaliero Universitaria Ospedali Riuniti, Ancona, Italy.

Abstract

SDS is characterized by bone marrow failure syndrome, which in turn reflects in impaired hematopoiesis, including thrombocytopenia, neutropenia and aplasia. However, little is known about the immunophenotype of peripheral blood. SDS patients reported lower percentage of bone marrow (BM) CD34+ cells, with increased number of monocytes and decreased number of granulocytes. Peripheral blood immunophenotype has been solely studied in a cohort of 6 Canadian patients so far, showing decreased number of circulating NK cells and abnormalities of B- and T-lymphocyte cell number in some cases. The aim of this study is gain further insights into the complete SDS peripheral blood immunophenotype. Here we report a comprehensive analysis of 54 SDS patients with ages range between 0.5 and 38 years (mean 13.3 ± 11.1). We compared SDS patients with an enlarged cohort of 617 healthy control subjects with the same ages range (mean 7.3 ± 6.3). Besides the already described severe neutropenia, we observed a very significant decreased number of circulating eosinophils (-87.6%) and of CD4/CD8 double negative (DN) T cells (-81.1%) in SDS patients. On the contrary, we reported a significant increase of monocytes (127.3%). No statistical difference was observed for B cells, NK cells, NKT nor other T lymphocyte subsets. Our results indicate that myeloid differentiation is affected in the opposite way between granulocyte progenitors, which show a strong maturation impairment, and monocyte progenitors which seem even more urged to differentiation. Interestingly, here we show for the first time a dysregulation in DN T cell maturation. Since DN T cells can act as suppressors (DN Tregs) or effectors (DN T cells) of the immune and inflammatory response, the impairment of DN T cell maturation might at least partially explain the susceptibility to recurrent infections observed in the early stages of life of SDS patients.

Presenting author name and contact information:

Dr. Antonio Vella
Unit of Immunology, Azienda Ospedaliera Universitaria Integrata di Verona
Piazzale A. Scuro 10
37134 Verona, Italy
Phone: +39 045 812 6445 fax: +39 802 7468 email: antonio.vella@univr.it
The Patient and Family Perspective: Examining the Impacts of Shwachman-Diamond Syndrome

Sara Loveless\textsuperscript{1}, Akiko Shimamura\textsuperscript{1}, Kelly McIntosh\textsuperscript{1}, Maggie Malsch\textsuperscript{1}, Joan Moore\textsuperscript{1}, Elissa Furtani\textsuperscript{1}, Leann Mount\textsuperscript{1}, Stella Davies\textsuperscript{1}, Kasiani Myers\textsuperscript{1}

\textsuperscript{1} Shwachman-Diamond Syndrome Registry (SDSR), Dana-Farber/Boston Children’s Cancer and Blood Disorders Center and Cincinnati Children’s Hospital Medical Center, Cancer and Blood Diseases Institute

Abstract

The SDSR (Shwachman-Diamond Syndrome Registry) conducted a survey of SDSR participants in order to broaden the understanding of issues important to SDS patients and families. An anonymous, self-report survey of 20 questions was distributed to enrolled patients and families with available email addresses (123 families). Forty-two responses (34\%) were received on 49 patients. Median patient age was 13 (0.25-67 years). Responders often reported concerns with feeding (75\%), height (63\%), and elevated liver enzymes (74\%). Many respondents indicated that they has issues related to their bones including skeletal abnormalities and osteoporosis (52\%).

While many (66\%), felt that they had adequate access to providers with knowledge of SDS, they felt that other healthcare team members did not understand the disease as well and coordinating care across disciplines was challenging. Ninety-five percent described significant concerns with emotional, social and/or educational issues including mood, depression and learning difficulties. Many parents described social difficulties in children with SDS due to short stature and often missing school. While SDS patients face significant challenges, their families also face numerous challenges. Family members (80\%), identified several issues including financial hardships from healthcare costs, and concerns regarding future insurance and care needs, career opportunities and choice of job location. Parents (73\%) also mentioned that SDS has impacted family planning as well as their own relationships including increased stress with extended family members and friends. Despite these challenges, responders (86\%) described inspiring accomplishments and successes of children and adults with SDS. Self-identified issues important to SDS families revolved around concerns about prognosis (risk of leukemia or need for transplant) as well as transition to adulthood. These issues should be prioritized in the research and care of SDS patients.

Presenting author name and contact information:

Sara Katherine Loveless, RN
Shwachman-Diamond Registry
3333 Burnet Avenue
Cincinnati, Ohio 45229
Phone: +1 513 803 7656 +1 513 636 6927 email: Sara.loveless@cchmc.org
98 patients with Shwachman-Diamond –Syndrome: An update from the SDS-Registry Europe

Mellor-Heineke, S.¹, Froemling, F.¹, Welte, K. ², Zeidler, C.¹ for the SDS-Registry

¹ SCNIR, Hannover Medical School, Carl-Neuberg-Str.1, 30625 Hannover, Germany
² University Children’s Hospital, Department of General Pediatrics and Pediatric Hematology and Oncology, Hoppe-Seyler-Str.1, 72076 Tübingen, Germany

Abstract

We report on 98 patients with a clinical diagnosis of Shwachman-Diamond-Syndrome (SDS) collected by the SDS-Registry Europe. In 81 of 82 patients tested (98.8 %) compound heterozygous SBDS mutations were detectable.

Patient characteristics: 98 SDS patients (48 female, 50 male), including 9 families with 2 affected siblings. At least contact 90 patients are alive, 8 patients expired. Median age at clinical SDS diagnosis is 1,51 years (range 0 -18.22 years). Median age at last follow up is 11,37 years (range 1,68- 39,49 years). 25 patients have reached adulthood (> 18 years old). The median time at deaths is 14,12 years (range 0,95- 45,57 years).

Besides anemia, neutropenia, recurrent infection or epistaxis, failure to thrive was the most common reason for diagnostic evaluation.

G-CSF therapy was given short-term (<1year) in 4 patients, long-term (>1 year) in 18 patients and interventionally in 2 patients. Median G-CSF dose was 2.8 μg/kg/day.

8 of 98 patients developed myelodysplastic syndrome (MDS) or leukemia (AML). 3/8 patients were pre-treated with G-CSF; 5/8 did not receive G-CSF at all. Between 01/1999 and 08/2015 stem cell transplantation (SCT) was performed in a total of 14 patients (2 patients received 2.SCT due to primary or secondary graft failure). Reason for SCT was MDS/AML (6 patients) or pancytopenia (6 patients) or other reason (2 patients). 2/6 patients transplanted for MDS/AML are alive at 6.33 and 12.18 years after SCT. 6/8 patients with PC/other are alive at 2.96 – 14.98 years after SCT.

In summary, the registry has current information on SDS patients including 25 adults up to 45 years of age. Besides leukemia and transplant related mortality only 1 patient died due to primary infection. Further studies are necessary to improve the outcome of leukemia and SCT.

Presenting author name and contact information:

Dr. Cornelia Zeidler
Severe Chronic Neutropenia International Registry at the Hannover Medical School
Carl-Neuberg-Str. 1
Hannover, 30625 Germany
Phone: +49 511 557105   fax: +49 511 557106   email: Zeidler.Cornelia@mh-hannover.de
Induced Pluripotent Stem Cell Model Of 7q Deletion In Shwachman Diamond Syndrome Identifies A Novel Therapeutic Strategy

Melisa Ruiz-Gutierrez, MD, PhD1*, Cailin Joyce, PhD2, Ozge Vargel Bolukbasi, PhD1, Adriana Kotini, PhD3, Gabriela Alexe, PhD2, Jennifer Whangbo, MD, PhD1, David Russell, MD, PhD4, Kimberly Stegmaier, MD3, Carl Novina, PhD2, Eirini Papapetrou, MD, PhD3 and Akiko Shimamura, MD, PhD1

1Division of Hematology/Oncology, Boston Children’s Hospital and Dana Farber Cancer Institute, Boston, MA. 2Dana Farber Cancer Institute, Boston, MA. 3Department of Oncological Sciences, Tisch Cancer Institute, Icahn School of Medicine at Mount Sinai, New York, NY. 4Division of Hematology, University of Washington, Seattle, WA

Abstract

Monosomy 7 or deletion of 7q (del(7q)) are common cytogenetic abnormalities in pediatric myelodysplastic syndrome (MDS) and frequently arise in patients with Shwachman Diamond Syndrome (SDS). Monosomy 7/del(7q) is associated with high grade MDS and propensity to progress to acute myelogenous leukemia, a major cause of morbidity and mortality. The aim of this study is to investigate the molecular consequences of del(7q) in the context of SDS with the goal of developing more effective treatments.

To study the biological and molecular consequences of monosomy/del(7q) in SDS, induced pluripotent stem cells were generated from SDS patients (SDS-iPSC). A deletion of the MDS-associated region of the long arm of chromosome 7 was then introduced using a previously published modified Cre-Lox approach. We next explored whether deletion of 7q conferred a relative fitness advantage within the context of bone marrow failure. Proliferation and hematopoietic differentiation of the SDS-del(7q) iPSCs was reduced below that of both the isogenic SDS-iPSCs and normal controls. These data demonstrate that deletion of 7q fails to confer a relative growth advantage relative to isogenic SDS-iPSCs and results in further impairment of hematopoiesis.

To gain insight into the mechanisms of del7q-associated clonal evolution in SDS, we performed RNA sequencing (RNA seq) of SDS+/-del(7q) iPSC. Expression of TGFβ pathways and downstream targets were reduced in SDS-del(7q) iPSCs compared to isogenic SDS iPSC. Single cell RNAseq analysis of primary SDS bone marrow cells confirmed that the TGFβ pathway is hyperactivated in SDS. Pharmacological targeting of TFGβ with small molecule inhibitors resulted in selective improvement of SDS hematopoietic colony formation and myeloid differentiation without stimulating outgrowth of the isogenic SDS-del(7q) cells or normal controls. Thus, TGFβ inhibition selectively rescues hematopoiesis in SDS but not in isogenic del7q cells, suggesting a potential strategy to treat SDS without stimulating del7q clonal outgrowth.

Presenting author name and contact information:

Dr. Melisa Ruiz-Gutierrez, MD PhD
Boston Children’s Hospital
300 Longwood Ave, Karp 08215
Boston MA 02115
Phone: +1 617 919 2009  fax +1 617 730 0934  email: melisa.ruiz-gutierrez@childrens.harvard.edu
Ataluren restores SBDS expression and function in bone marrow cells obtained from SDS patients

Valentino Bezzerri1*, Donatella Bardelli2, Jacopo Morini3, Antonio Vella4, Simone Cesaro5, Claudio Sorio1, Andrea Biondi6, Cesare Danesino7, Piero Farruggia8, Baroukh Maurice Assael9, Giovanna D’Amico2 and Marco Cipolli10

1Department of Medicine, University of Verona, Italy; 2Unit of Immunology and Immunotherapy, Centro Ricerca Tettamanti, Pediatric Department, University of Milano Bicocca, Fondazione MBBM, Italy; 3Department of Physics, University of Pavia, Italy; 4Unit of Immunology, Azienda Ospedaliera Universitaria Integrata di Verona, Italy; 5Unit of Pediatric Hematology Oncology, Azienda Ospedaliera Universitaria Integrata di Verona, Italy; 6School of Medicine and Surgery, University of Milano-Bicocca, Italy; 7Department of Molecular Medicine, University of Pavia, Italy; 8Department of Oncology, ARNAS Ospedale Civico Palermo, Italy; 9Department of Pulmonology, Adult CF center, IRCCS Fondazione Cà Granda, Policlinico Milano, Italy; 10Cystic Fibrosis Center, Azienda Ospedaliero Universitaria Ospedali Riuniti, Ancona, Italy

Abstract

Shwachman-Diamond syndrome (SDS) affects several organs, causing bone marrow failure, exocrine pancreatic insufficiency, skeletal malformations, and cognitive disorders. About 15% of SDS patients develop myelodysplastic syndromes (MDS) and are at higher risk of developing acute myeloid leukemia (AML). Deficiency in SBDS expression has been associated with increased mTOR phosphorylation in leukocytes and excessive apoptotic rate and lack of myeloid differentiation in bone marrow hematopoietic progenitors. Importantly, most SDS patients carry the c.183-184TA>CT nonsense mutation in SBDS. Ataluren is an orally bio-available compound approved by the European Medicines Agency (EMA) for the treatment of Duchenne muscular dystrophy (DMD). Ataluren promotes the insertion of near-cognate tRNAs at the nonsense codon site resulting in amino acid replacements similar to those observed in endogenous read-through. Here we have assessed the efficacy of this drug in restoring SBDS expression in hematopoietic cells obtained from a cohort of 13 SDS patients. We show that ataluren treatment readily restores SBDS protein expression in different cell types, including bone marrow stem cells. Ataluren promotes myeloid differentiation in hematopoietic progenitors, reduces apoptotic rate in primary PBMCs, and reduces mTOR phosphorylation levels both in lymphoblasts and in bone marrow mesenchymal stromal cells. Since a specific therapy against SDS is currently lacking, these results provide the rationale for ataluren repurposing clinical trials. Moreover, this study opens new therapeutic perspectives also for other genetic bone marrow failure syndromes caused by nonsense mutations.

Presenting author name and contact information:

Dr. Valentino Bezzerri, Ph.D.
Department of Medicine, University of Verona
Strada Le Grazie 8
37134 Verona, Italy
Phone: +39 045 802 7274   fax: +39 045 802 7127   email: valentino.bezzerri@univr.it
Androgen Therapy In Patients With Shwachman-Diamond Syndrome

Albert Catala1, Salah Ali2, Lawrence Jardine3, Supanun Lauhasurayotin1,4, Bozana Zlateska4, Michaela Cada1, Yigal Dror1,4,5

#1The Marrow Failure and Myelodysplasia Program, Haematology Section, Division of Haematology/Oncology, Department of Paediatrics, The Hospital for Sick Children, Toronto, Canada, #2Division of Haematology/Oncology, Department of Paediatrics, The Hospital for Sick Children, Toronto, Canada, #3Division of Haematology/Oncology, Children's Hospital of Western Ontario, London, Canada, #4Program in Genetics and Genome Biology, Research Institute, The Hospital for Sick Children, Toronto, Canada, #5Institute of Medical Sciences, University of Toronto, Toronto, Canada

Abstract

The majority of data on androgen therapy for inherited bone marrow failure syndromes comes from the Fanconi Anemia literature, where a 50-70% hematological response rate is reported. The use of androgens among Shwachman-Diamond syndrome (SDS) patients is anecdotal and not well documented.

Purpose: To report the experience of androgen use in patients with SDS from the Canadian Inherited Bone Marrow Failure Registry (CIMFR).

Methods: Clinical, laboratory and liver imaging data were reviewed retrospectively from CIMFR records from 2001 to 2017.

Results: Three patients with SDS received androgens. Patient 1 had SBDS gene mutation and severe pancytopenia, and was transfusion dependent. She received oxymetholone (0.5mg/Kg/day) and was switched to danazol (1.5mg/Kg/day) at 34 months due to funding concerns. She achieved sustained response at 12 months and became transfusion independent by 3 months. She remains well on danazol at 63 months. Patient 2 had DNAJC1 gene mutation and severe bicytopenia. He received danazol (1.5-3mg/Kg/day) and achieved sustained response by 5 months. He continues on danazol at 36 months. Patient 3 had DNAJC1 gene mutation and severe pancytopenia, and was transfusion dependent. He received only 2 months of oxymetholone (0.5mg/kg) before proceeding to stem cell transplant (SCT). He had no response. All patients had mild transaminitis prior to androgens, and in one patient this increased to above baseline. Patients 1 and 2 had mild behavioural changes and virilisation.

Implications and applications:
The only curative treatment for bone marrow failure (BMF) in SDS patients is SCT. The present report suggest that androgen therapy may be useful and has limited toxicities in those with progressive BMF, but these results must be confirmed in larger series. Androgen impact on prevention of BMF or development of myelodysplastic syndrome or acute myeloid leukemia is unknown and requires further investigation.

Presenting author name and contact information:
Dr. Albert Catala
The Marrow Failure and Myelodysplasia Program, Haematology Section, Division of Haematology/Oncology, Department of Paediatrics, The Hospital for Sick Children, Toronto, Canada
SickKids, 555 University Avenue
Toronto, Ontario, Canada, M5G 1X8
Phone: +1 416 460 5511  email: albert.catala@sickkids.ca
<table>
<thead>
<tr>
<th>First Name</th>
<th>Last Name</th>
<th>Institute</th>
<th>City</th>
<th>Country</th>
<th>Email Address</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blanche</td>
<td>Alter</td>
<td>National Cancer Institute</td>
<td>Rockville</td>
<td>USA</td>
<td><a href="mailto:alterb@mail.nih.gov">alterb@mail.nih.gov</a></td>
</tr>
<tr>
<td>Elf</td>
<td>Arik</td>
<td>Baylor College of Medicine</td>
<td>Houston</td>
<td>USA</td>
<td><a href="mailto:ElfArik@bcm.edu">ElfArik@bcm.edu</a></td>
</tr>
<tr>
<td>Carlos</td>
<td>Bacino</td>
<td>Texas Children’s Hospital, Baylor College of Medicine</td>
<td>Houston</td>
<td>USA</td>
<td><a href="mailto:cbacino@bcm.edu">cbacino@bcm.edu</a></td>
</tr>
<tr>
<td>Susan</td>
<td>Baserga</td>
<td>Yale School of Medicine</td>
<td>New Haven</td>
<td>USA</td>
<td><a href="mailto:susan.baserga@yale.edu">susan.baserga@yale.edu</a></td>
</tr>
<tr>
<td>Allison</td>
<td>Bertuch</td>
<td>Texas Children’s Hospital, Baylor College of Medicine</td>
<td>Houston</td>
<td>USA</td>
<td><a href="mailto:aberuch@bcm.edu">aberuch@bcm.edu</a></td>
</tr>
<tr>
<td>Valentina</td>
<td>Bezzerri</td>
<td>University of Verona</td>
<td>Verona</td>
<td>Italy</td>
<td><a href="mailto:valentino.bezzerri@univr.it">valentino.bezzerri@univr.it</a></td>
</tr>
<tr>
<td>Marco</td>
<td>Cipolli</td>
<td>Università degli Studi di Verona</td>
<td>Pavia</td>
<td>Italy</td>
<td><a href="mailto:marco.cipolli@ospedaliriuniti.marche.it">marco.cipolli@ospedaliriuniti.marche.it</a></td>
</tr>
<tr>
<td>Cesare</td>
<td>Danesino</td>
<td>University of Pavia</td>
<td>Pavia</td>
<td>Italy</td>
<td><a href="mailto:cdi@unipv.it">cdi@unipv.it</a></td>
</tr>
<tr>
<td>Stella</td>
<td>Davies</td>
<td>Cincinnati Children’s Hospital</td>
<td>Cincinnati</td>
<td>USA</td>
<td><a href="mailto:Stella.Davies@cchmc.org">Stella.Davies@cchmc.org</a></td>
</tr>
<tr>
<td>Francois</td>
<td>Delhommeau</td>
<td>Saint-Antoine Hospital</td>
<td>Paris</td>
<td>France</td>
<td><a href="mailto:francois.delhommeau@aphp.fr">francois.delhommeau@aphp.fr</a></td>
</tr>
<tr>
<td>Jean</td>
<td>Donadieu</td>
<td>APHP</td>
<td>France</td>
<td>France</td>
<td><a href="mailto:jean.donadieu@aphp.fr">jean.donadieu@aphp.fr</a></td>
</tr>
<tr>
<td>Vidal</td>
<td>Dror</td>
<td>The Hospital for Sick Children</td>
<td>Tokyo</td>
<td>Japan</td>
<td><a href="mailto:vgal.dror@sickkids.org">vgal.dror@sickkids.org</a></td>
</tr>
<tr>
<td>Tank</td>
<td>Eighttanay</td>
<td>Texas Children’s Hospital, Baylor College of Medicine</td>
<td>Houston</td>
<td>USA</td>
<td>tank <a href="mailto:Eighttanay@texaschildrens.org">Eighttanay@texaschildrens.org</a></td>
</tr>
<tr>
<td>Ayushi</td>
<td>Jain</td>
<td>Mahidol University</td>
<td>Bangkok</td>
<td>Thailand</td>
<td><a href="mailto:ayushi225@hotmail.com">ayushi225@hotmail.com</a></td>
</tr>
<tr>
<td>Amornrat</td>
<td>Jensen</td>
<td>Mahidol University</td>
<td>Bangkok</td>
<td>Thailand</td>
<td>amornrat.harvard.edu</td>
</tr>
<tr>
<td>Alyssa</td>
<td>Kennedy</td>
<td>Dana Farber and Boston Children’s Cancer and Blood Disorders Center</td>
<td>Boston</td>
<td>USA</td>
<td><a href="mailto:alyssa.kennedy@childrens.harvard.edu">alyssa.kennedy@childrens.harvard.edu</a></td>
</tr>
<tr>
<td>Elizabeth</td>
<td>Kerr</td>
<td>The Hospital for Sick Children</td>
<td>Toronto</td>
<td>Canada</td>
<td><a href="mailto:elizabeth.kerr@sickkids.ca">elizabeth.kerr@sickkids.ca</a></td>
</tr>
<tr>
<td>Coleman</td>
<td>Lindley</td>
<td>Dana-Farber Cancer Institute</td>
<td>Boston</td>
<td>USA</td>
<td><a href="mailto:coleman.lindley@dfci.harvard.edu">coleman.lindley@dfci.harvard.edu</a></td>
</tr>
<tr>
<td>Sara</td>
<td>Loveless</td>
<td>Cincinnati Children’s Hospital</td>
<td>Cincinnati</td>
<td>USA</td>
<td><a href="mailto:sara.loveless@ccmc.org">sara.loveless@ccmc.org</a></td>
</tr>
<tr>
<td>Caridad</td>
<td>Martinez</td>
<td>Texas Children’s Hospital, Baylor College of Medicine</td>
<td>Houston</td>
<td>USA</td>
<td><a href="mailto:caridad.martinez@texaschildrens.org">caridad.martinez@texaschildrens.org</a></td>
</tr>
<tr>
<td>First Name</td>
<td>Last Name</td>
<td>Institute</td>
<td>City</td>
<td>Country</td>
<td>Email Address</td>
</tr>
<tr>
<td>-----------</td>
<td>-----------------</td>
<td>----------------------------------------------------------------------------</td>
<td>---------------</td>
<td>---------</td>
<td>--------------------------------------</td>
</tr>
<tr>
<td>Kasiiani</td>
<td>Myers</td>
<td>Cincinnati Children's Hospital Medical Center</td>
<td>Cincinnati</td>
<td>USA</td>
<td><a href="mailto:kasiani.myers@cchmc.org">kasiani.myers@cchmc.org</a></td>
</tr>
<tr>
<td>(Keith)</td>
<td>Chee Ooi</td>
<td>University of New South Wales and Sydney Children's Hospital</td>
<td>Sydney</td>
<td>Australia</td>
<td><a href="mailto:keith.ooi@unsw.edu.au">keith.ooi@unsw.edu.au</a></td>
</tr>
<tr>
<td>Usua</td>
<td>Oyarbide</td>
<td>Virginia Commonwealth University</td>
<td>Richmond</td>
<td>USA</td>
<td><a href="mailto:usua.oyarbide@vcuhealth.org">usua.oyarbide@vcuhealth.org</a></td>
</tr>
<tr>
<td>Francesco</td>
<td>Pasquali</td>
<td>University of Insubria</td>
<td>Varese</td>
<td>Italy</td>
<td><a href="mailto:francesco.pasquali@uninsubria.it">francesco.pasquali@uninsubria.it</a></td>
</tr>
<tr>
<td>Yves</td>
<td>Pastore</td>
<td>CHUSainte-Justine / University of Montreal</td>
<td>Montreal</td>
<td>Canada</td>
<td><a href="mailto:yves.pastore@umontreal.ca">yves.pastore@umontreal.ca</a></td>
</tr>
<tr>
<td>Marc</td>
<td>Raaijmakers</td>
<td>Erasmus MC Cancer Institute</td>
<td>Rotterdam</td>
<td>NL</td>
<td><a href="mailto:m.h.g.raaijmakers@erasusmc.nl">m.h.g.raaijmakers@erasusmc.nl</a></td>
</tr>
<tr>
<td>Patrick</td>
<td>Revy</td>
<td>Imagine Institute</td>
<td>Paris</td>
<td>France</td>
<td><a href="mailto:patrick.revy@insERM.fr">patrick.revy@insERM.fr</a></td>
</tr>
<tr>
<td>Johanna</td>
<td>Rommens</td>
<td>The Hospital for Sick Children</td>
<td>Toronto</td>
<td>Canada</td>
<td><a href="mailto:jrommens@sickkids.ca">jrommens@sickkids.ca</a></td>
</tr>
<tr>
<td>Melisa</td>
<td>Ruiz-Gutierrez</td>
<td>Boston Children's Hospital/Dana Farber Cancer Institute</td>
<td>Boston</td>
<td>USA</td>
<td><a href="mailto:melisa.ruiz-gutierrez@childrens.harvard.edu">melisa.ruiz-gutierrez@childrens.harvard.edu</a></td>
</tr>
<tr>
<td>Nuria</td>
<td>Sanchez</td>
<td>Universidad Nacional Autónoma de México</td>
<td>México D.F.</td>
<td>Mexico</td>
<td><a href="mailto:nuriasp@unam.mx">nuriasp@unam.mx</a></td>
</tr>
<tr>
<td>Ghadir</td>
<td>Sasa</td>
<td>Texas Children’s Hospital, Baylor College of Medicine</td>
<td>Houston</td>
<td>USA</td>
<td><a href="mailto:gssasa@texaschildrens.org">gssasa@texaschildrens.org</a></td>
</tr>
<tr>
<td>Akiko</td>
<td>Shimamura</td>
<td>Dana Farber/Boston Children’s Cancer and Blood Disorders Center</td>
<td>Boston</td>
<td>USA</td>
<td><a href="mailto:akiko.shimamura@childrens.harvard.edu">akiko.shimamura@childrens.harvard.edu</a></td>
</tr>
<tr>
<td>Dritan</td>
<td>Siliqi</td>
<td>Istituto di Cristallografia - CNR</td>
<td>Bari</td>
<td>Italy</td>
<td><a href="mailto:dritan.siliqi@ic.cnr.it">dritan.siliqi@ic.cnr.it</a></td>
</tr>
<tr>
<td>Alexandra</td>
<td>Topa</td>
<td>Sahlgrenksa University Hospital, University of Gothenburg, Sweden</td>
<td>Gothenburg</td>
<td>Sweden</td>
<td><a href="mailto:alexandra.topa@vgregion.se">alexandra.topa@vgregion.se</a></td>
</tr>
<tr>
<td>Roberto</td>
<td>Valli</td>
<td>University of Insubria - Varese - Italy</td>
<td>Varese</td>
<td>Italy</td>
<td><a href="mailto:roberto.valli@uninsubria.it">roberto.valli@uninsubria.it</a></td>
</tr>
<tr>
<td>Antonio</td>
<td>Vella</td>
<td>University Hospital of Verona</td>
<td>Verona</td>
<td>Italy</td>
<td><a href="mailto:antonio.vella@univr.it">antonio.vella@univr.it</a></td>
</tr>
<tr>
<td>Tom</td>
<td>Vulliamy</td>
<td>Rats adn The London, Queen Mary University of London</td>
<td>London</td>
<td>UK</td>
<td><a href="mailto:t.vulliamy@qmul.ac.uk">t.vulliamy@qmul.ac.uk</a></td>
</tr>
<tr>
<td>Alan</td>
<td>Warren</td>
<td>University of Cambridge</td>
<td>Cambridge</td>
<td>UK</td>
<td><a href="mailto:ajw1.000@cam.ac.uk">ajw1.000@cam.ac.uk</a></td>
</tr>
<tr>
<td>Kenichiro</td>
<td>Watanabe</td>
<td>Shizuoka Children’s Hospital</td>
<td>Shizuoka</td>
<td>Japan</td>
<td><a href="mailto:wataken@kuhp.kyoto-u.ac.jp">wataken@kuhp.kyoto-u.ac.jp</a></td>
</tr>
<tr>
<td>Cornelia</td>
<td>Zeidler</td>
<td>Medical School Hannover and SDS Europe</td>
<td>Hannover</td>
<td>Germany</td>
<td><a href="mailto:Zeidler.Cornelia@mh-hannover.de">Zeidler.Cornelia@mh-hannover.de</a></td>
</tr>
</tbody>
</table>