37TH ANNUAL MEETING OF THE HISTIOCYTE SOCIETY

VIRTUAL

OCTOBER 11-12, 2021 @ 11:00AM EASTERN TIME (US)

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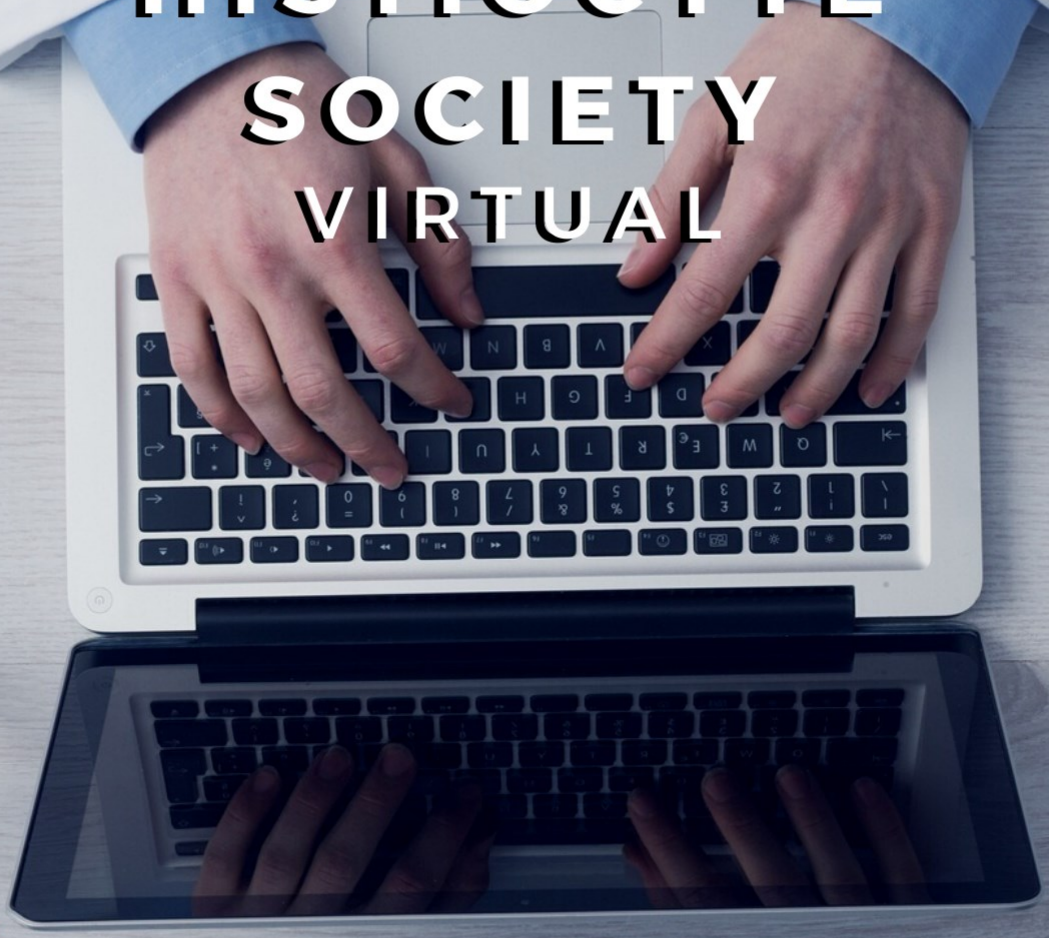

VIRTUAL 2021

37TH ANNUAL MEETING

HISTIOCYTE SOCIETY

ANNUAL MEETING PROGRAM AND ABSTRACTS

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MEETING SPONSORS

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ARTEMIS ASSOCIATION
ON HISTIOCYTOSES

WELCOME TO OUR VIRTUAL ANNUAL MEETING

Dear Colleagues,

I would like to cordially welcome you the 37th Annual Meeting of the Histiocyte Society being held virtually, October 11 and 12, 2021. This is our second year meeting virtually during the COVID19 pandemic. Last year we were due to return to Athens and we had initially hoped that we could simply delay our return to Athens until 2021. Alas, that could not happen (thanks to the annoying 'delta variant' etc.). But things are slowly inching back towards normal! I hope you are all well and looking forward to our meeting this year, which is moving back towards our usual format, though still somewhat 'compact' and virtual.

This year you submitted excellent abstracts- we will hear the most highly rated ones in the Presidential Symposium and we will award the Nezelof and Nesbit prizes to the top scientific and clinical presentations. We will of course have a full poster session where most abstracts will be presented. We will use an innovative and fun 'Spatial Chat' platform, that allows you to move around various rooms to view posters and have direct conversations with presenters. I hope you enjoy it and make good use of the platform. We will also utilize the Spatial Chat platform to enjoy a virtual welcome reception and banquet. This is your chance to catch up with colleagues from around the world in a direct and fun fashion. We will still conduct our regular society business including elections and reports from the executive board and our committees. Members, don't forget to cast your vote in our annual elections (see email notifications for details). Despite the fact that our meeting occurs virtually this year, it still requires work. I would like to thank the Education Committee for scoring abstracts, Kathy for her organizational efforts, and our partner, the Histiocytosis Association, for assisting us in this endeavor.

This year I hope we all appreciate the special worldwide community we have developed and how much we have achieved over the years. This year I hope we stay connected again via technology which is getting easier and more routine. We will host bi-monthly sessions starting later this fall and extending into the Spring, similar to last year. Looking forward, we are planning to host the 38th Annual Meeting of the Histiocyte Society, September 18-20, in person, in Stockholm. We look forward to a full program and welcome a return to an in person meeting next year.

Wishing you all the best during yet another challenging year.

I look forward to seeing you all joyously in 2022, in Stockholm!

Sincerely,
Michael Jordan

President

ABOUT THE HISTIOCYTE SOCIETY

The Histiocyte Society is a professional medical association comprised of more than 200 physicians and scientists from around the world. Members of the organization are considered to be the leaders in understanding and treating histiocytic disorders. The Society is committed to advancing knowledge about histiocytic disorders and improving outcomes for patients through the planning, development, sponsorship and oversight of clinical research.

Building Knowledge

The Histiocyte Society hosts an annual scientific meeting in different locations around the world. Attendance is open to members of the Society as well as other professionals working in the field of histiocytic disorders and related studies. Meetings feature presentations of individual and collective research efforts as well as keynote lectures from guest speakers; special-interest groups convene to discuss and develop new and innovative treatment regimes. This interactive forum allows the best and brightest minds in the histiocytosis community to share the most progressive information and to shape the future of research. Beyond the prolific exchanges that occur during the meeting, presenters work collectively to extend their reach by publishing subsequent articles and manuscripts in scientific journals worldwide.

Advancing Treatment

Through extensive research and collaboration, the Histiocyte Society has made numerous, significant strides in the fight against histiocytic disorders. The Society has established scientific standards for histiocytic disorders that are accepted worldwide; they include: 1) A common language of uniform disease classification, 2) Standardized diagnostic criteria, and 3) Guidelines for patient evaluation and follow-up. The Society remains dedicated to facilitating essential and innovative clinical research – developing critical knowledge and increasingly effective treatments in the pursuit of a cure.

ANNUAL MEETING PROGRAM

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HISTIOCYTE SOCIETY

Photography Consent

Registration for, attendance at, and participation in the 2021 Virtual Histiocyte Society Annual Meeting constitutes an agreement by the participant to allow the Histiocyte Society to use and distribute (both now and in the future) the registrant's or attendee's image and/or voice in photographs, video, electronic reproductions and audio of such events and activities.

HISTIOCYTOSIS ASSOCIATION AND HISTIOCYTE SOCIETY

Separate Organizations, One Goal

The Road to Partnership

Three-month old Bethany Toughill was diagnosed in 1983. Bethany's dad, Jeff, and her mother, Sally, experienced the same fear that today's parents feel when learning that their child has this disease. Alone and frustrated, Sally and Jeff were led to find other families affected by histiocytosis so that they could emotionally support each other. In 1986 they founded the Histiocytosis Association, a community comprised of patients, parents, family members and friends.

At about the same time that the Association was founded, a group of physicians formed the Histiocyte Society – an international group of clinicians and scientists committed to improving the lives of patients with histiocytic disorders by conducting clinical and laboratory research into the causes and treatment of this disease. Recognizing that these two groups had the same goal, Histiocytosis Association Founder Jeffrey Toughill offered the Association's business resources to help manage the Histiocyte Society. Thus began the partnership that still exists today.

A True Partnership

The Histiocytosis Association and the Histiocyte Society remain separate organizations working toward a common goal, with the Histiocytosis Association serving as the Society's administrative center (Secretariat) and primary source of funding. The Histiocytosis Association serves the administrative needs of the Histiocyte Society by:

- Organizing and managing the Society's annual scientific and Executive Board meetings,
- Managing the overall organizational, administrative and financial operations,
- Aiding in the development of organizational guidelines and operating procedures,
- Building, developing and maintaining the organization and annual meeting websites,
- Facilitating communication between Society members and the Executive Board, and
- Building and managing the Society's membership database.

The Histiocytosis Association Mission

The Histiocytosis Association is dedicated to raising awareness about histiocytic disorders, providing educational and emotional support for patients and families and funding research leading to better treatments and a cure.

The Histiocytosis Association Research Program

At the very heart of the Histiocytosis Association is the steadfast commitment to finding a cure for histiocytic disorders. The Histiocytosis Association is committed and focused on a threefold research plan and philosophy:

- Funding basic science research being conducted worldwide identified through a competitive, peer-review process modeled after that of the National Institutes of Health. The Histiocytosis Association requests research proposals on a yearly basis – usually around May. Once research proposals are received the Histiocyte Society Scientific Committee serves as the first-step review. Scores from this review are then submitted to the Histiocytosis Association's Medical & Scientific Advisory Committee (MSAC) for a second review. The SAC then makes a recommendation to the Histiocytosis Association's Board of Trustees concerning grants to be funded.
- Funding clinical studies that result in identifying the best possible treatments for histiocytic disorders.
- Facilitating research by administratively managing the Histiocyte Society.

Since 1992, nearly 200 individual awards have been made to date, representing more than $7 million to support critical research around the world. Grant amounts now average $50,000 per project but have been awarded in amounts up to $100,000 in the past. The Histiocytosis Association of Canada (HAC) has provided $265,000 of that funding.

Two Organizations, One Goal

The Histiocytosis Association and the Histiocyte Society have been committed partners for over 35 years. Though separate organizations, they share a common goal that binds their continued partnership - to find better treatments and, ultimately, a cure leading to a world free of histiocytic disorders.

To learn more about the Histiocytosis Association visit www.histio.org.

ACKNOWLEDGEMENTS AND RECOGNITIONS

HISTIOCYTE SOCIETY EXECUTIVE BOARD

President ..... Michael Jordan
2019-2022
President-Elect ..... Kim Nichols
2020-2022
Treasurer ..... Jennifer Picarsic
2020-2022
Secretary ..... Karin Beutel
2020-2022
Member-at-Large ..... Itziar Astigarraga
2020-2023
Member-at-Large ..... Julien Haroche
2020-2023

HISTIOCYTE SOCIETY EDUCATION COMMITTEE

Michael Henry, Chairperson 2019-2021
Patrick Campbell 2019-2021
Deepak Chellapandian 2020-2022
Paul Hendrie 2020-2022
Melissa Hines 2019-2021
Vassilios Papadakis 2019-2021
Elena Sieni 2019-2021
Julie Talano 2020-2022
Tatiana von Bahr Greenwood 2020-2022

HISTIOCYTE SOCIETY SCIENTIFIC COMMITTEE

Julien Haroche, Chair 2019-2021
Rikhia Chakraborty 2020-2022
Benjamin Durham 2019-2021
Jean-Francois Emile 2019-2021
Michelle Hermiston 2019-2021
Caroline Hutter 2019-2021
Jennifer Picarsic 2019-2021
Yongmin Tang 2020-2022
Astrid van Halteren 2020-2022

HISTIOCYTE SOCIETY STUDY GROUP CHAIRPERSONS

Adult Histiocytosis ..... Michael Girschikofsky
Epidemiology/Late Effects ..... Riccardo Haupt /Vasanta Nanduri
HLH-2004 ..... Jan-Inge Henter
LCH-IV ..... Milen Minkov/Carlos Rodriguez-Galindo
Rare Histiocytic Disorders ..... Oussama Abla

HLH STEERING COMMITTEE

Kim Nichols, Chair 2019-2023
Rebecca Marsh, Vice-Chair 2019-2023
Scott Baker 2018-2022
Ed Behrens 2019-2023
Stephan Ehl 2018-2022
Jan-Inge Henter 2018-2022
Michael Jordan 2017-2021
Kai Lehmberg 2017-2021
Rafal Machowicz 2019-2023
Despina Moshous 2020-2024
Elena Sieni 2019-2023
Zhao Wang 2019-2023
Takahiro Yasumi 2020-2024

LCH STEERING COMMITTEE

Michelle Hermiston, Chair 2017-2021
Matthew Collin, Vice-Chair 2020-2024
Carl Allen 2020-2024
Karin Beutel 2017-2021
Patrick Campbell 2018-2022
Michael Girschikofsky 2019-2023
Rima Jubran 2017-2021
Milen Minkov 2018-2022
Vasanta Nanduri 2020-2024
Barrett Rollins 2017-2021
Kimo Stine 2018-2022

RARE HISTIOCYTIC DISORDERS STEERING COMMITTEE

Eli Diamond, Chair 2019-2023
Jean-Francois Emile, Vice-Chair 2017-2021
Oussama Abla 2020-2024
Jorge Braier 2020-2024
Benjamin Durham 2017-2021
Michael Girschikofsky 2017-2021
Gaurav Goyal 2020-2024
Julien Haroche 2018-2022
Eric Jacobsen 2019-2023
Zdenka Krenova 2019-2023
Akira Morimoto 2017-2021
Jennifer Picarsic 2019-2023

HISTIOCYTE SOCIETY PAST PRESIDENTS

Milen Minkov 2016-2019
Carlos Rodriguez-Galindo 2013-2016
Jim Whitlock 2010-2013
Alexandra Filipovich 2007-2010
Jan-Inge Henter 2004-2007
R. Maarten Egeler 2001-2004
Kenneth McClain 1998-2001
Göran Elinder 1996-1998
Helmut Gadner 1992-1996
Stephan Ladisch 1989-1992
Blaise Favara 1987-1989
Christian Nezelof 1985-1987

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ACKNOWLEDGEMENTS AND RECOGNITIONS

NESBIT PRIZE IN CLINICAL SCIENCE AWARDEES

Paul Kemps....2019
Jennifer Picarsic....2018
Elena Sieni....2017
Francesca Minoia....2016
Alexandra Löfstedt....2015
Vasanta Nanduri....2014
Carl Allen....2013
Stephen Simko....2012
Thomas Lehrnbecher....2011
Rebecca Marsh....2010
Rebecca Marsh....2009
Jorge Braier....2008
Kenneth McClain....2007
Loretta Lau....2006
AnnaCarin Horne....2005
Marie Ouachée-Chardin....2004
Manuel Steiner....2003
Jorge Braier....2002
Wolfgang Holter....2001
Kazuhiro Kogawa....2000

NEZELOF PRIZE IN BASIC SCIENCE AWARDEES

Lauren Meyer....2019
Lauren Meyer....2018
Hirofumi Shibata....2017
Edward Behrens....2016
Benjamin Durham....2015
Samuel Chiang Cern Cher....2014
Gayane Badalian-Very/Kim Nichols....2013
Edward Behrens....2012
Edward Behrens....2011
Michelle Hermiston....2010
Michael Jordan....2009
Matthew Collin....2008
Kejian Zhang....2007
Alessandra Santoro....2006
Udo zur Stadt....2005
Cristiana Costa/Kimberly Risma....2004
Michael B. Jordan....2003
Susan Lee/Joyce Villanueva....2002
Maurizio Aricó....2001
Pieter Leenen....2000

ROBERT J. ARCECI AWARD FOR BEST POSTER

Hirofumi Shibata....2019
Amel Sengal....2018
Caroline Hutter....2017
Sandra Ammann....2016

HISTIOCYTE SOCIETY GOLDEN PIN RECIPIENTS

Jorge Braier....2017
Lisa Filipovich....2017
Gritta Janka....2016
Stephan Ladisch....2016
R. Maarten Egeler....2015
Sheila Weitzman....2014
Shinsaku Imashuku....2010
Helmut Gadner....2008
Jon Pritchard....2006
Giulio D'Angio....2002
Sally Kivilis....2001
Elizabeth Kontoyannis....2000
Paul Kontoyannis....2000
Jeffrey M. Toughill....1998

HISTIOCYTE SOCIETY HONORED MEMBERS

Helmut Gadner....2008
Shinsaku Imashuku....2007
Gritta Janka....2007
Valerie Broadbent....2000
Blaise Favara....1998
Mark Nesbit....1998
Christian Nezelof....1998

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AT-A-GLANCE AGENDA

MONDAY • OCTOBER 11, 2021 - ONLINE

1100 – 1115 **Welcome/Opening Remarks** - Michael Jordan and Paul Kontoyannis

1115 – 1200 **Guest Speaker Presentation** – Florent Ginhoux

SENESCENT LANGERHANS CELL HISTIOCYTOSIS CELLS ARISE FROM BOTH DENDRITIC CELL (DC) AND MONOCYTE/DC3 LINEAGES

1200 – 1230 **Clinical Studies and Registries Update** –

LCH-IV - Milen Minkov
HLH Registry - Stephan Ehl
IRHDR - Oussama Abla

1230 - 1245 **Oral Presentations**

Abstract Presentation #1 - Fabrizio De Benedetti
MACROPHAGE ACTIVATION SYNDROME (MAS) IN SYSTEMIC JUVENILE IDIOPATHIC ARTHRITIS (SJIA): TREATMENT WITH EMAPALUMAB, AN ANTI-INTERFERON GAMMA (IFNγ) MONOCLONAL ANTIBODY

Abstract Presentation #2 - Jan-Inge Henter
RESPONSE TO MAPK INHIBITION OF NEURODEGENERATION IN LCH MONITORED BY CSF NEUROFILAMENT LIGHT AS A BIOMARKER

1245 – 1300 **BREAK**

1300 - 1430 **Poster Session** - via Spatial Chat Platform

1430 - 1530 **Virtual Welcome Reception** - via Spatial Chat Platform

TUESDAY • OCTOBER 12, 2021 - ONLINE

1100 – 1145 **Guest Speaker Presentation -** Jana Pachlopnik Schmid
HEMOPHAGOCYTIC LYMPHOHISTIOCYTOSIS

1145 – 1315 **Presidential Symposium**

Abstracts Nominated for the Nesbit Award in Clinical Science:

THE INTERIM RESULTS OF A MULTICENTER STUDY OF SAFETY AND EFFICACY OF VEMURAFENIB AND CYTARBINE PLUS CLADRIBINE THERAPY IN HIGH-RISK PEDIATRIC LANGERHANS CELL HISTIOCYTOSIS WITH BRAF V600E MUTATION - Dmitry Evseev

PERI-TRANSPLANT ALEMTUZUMAB LEVELS PREDICT RISK OF SECONDARY GRAFT FAILURE AND INVERSELY IMPACT CXCL9 LEVELS AFTER RIC-HCT (A CORRELATIVE BIOLOGY STUDY TO BMT-CTN 1204 RICHI)- Ashley Geerlinks

ALK+ HISTIOCYTOSIS: A NEW CLINICOPATHOLOGIC SPECTRUM HIGHLIGHTING NEUROLOGIC INVOLVEMENT AND RESPONSES TO ALK INHIBITION - Paul Kemps

Abstracts Nominated for the Nezelof Award in Basic Science:
CYTOTOXIC IMPAIRMENT AND EXCESS IL-18 DRIVE SPONTANEOUS, OLIGOCLONAL CD8 T-CELL HYPERACTIVATION AND HYPERINFLAMMATORY DISEASE- Emily Landy

LINEAGE-SWITCH OF CELLS HARBORING BRAF V600E ALLELES IN PATIENTS WITH HIGH RISK LCH TREATED WITH INHIBITORS - Paul Milne

BRAF-V600E EXPRESSION IN DENDRITIC CELLS REPROGRAMS TRANSLATIONAL DYNAMICS TO INCREASE LPS-INDUCED TNFA PRODUCTION - Danielle Minichino

—Continued on Next Page—

AT-A-GLANCE AGENDA (Cont.)

TUESDAY • OCTOBER 12, 2021 - ONLINE (CONTINUED)

1315 – 1330	**BREAK**	
1330 – 1430	**General Assembly Business Meeting***	
	1330 – 1340	President's Report - Michael Jordan
	1340 – 1350	Treasurer's Report - Jennifer Picarsic
	1350 – 1355	Secretary's Report - Karin Beutel
	1355 – 1400	Scientific Committee Chair Report - Julien Haroche
	1400 – 1405	Education Committee Chair Report - Michael Henry
	1405 – 1410	LCH Steering Committee Chair Report - Michelle Hermiston
	1410 – 1415	HLH Steering Committee Chair Report - Kim Nichols
	1415 – 1420	Rare Histiocytoses Committee Chair Report - Eli Diamond
	1420 – 1430	Closing Remarks - Michael Jordan
1430 - 1530	**Virtual Annual Banquet** - via Spatial Chat platform	

GUEST SPEAKER PRESENTATIONS

MONDAY, OCTOBER 11, 2021 | 1115-1200

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SENESCENT LANGERHANS CELL HISTIOCYTOSIS CELLS ARISE FROM BOTH DENDRITIC CELL (DC) AND MONOCYTE/DC3 LINEAGES

Florent Ginhoux

Singapore Immunology Network (SIgN)
*Agency for Science, Technology and Research (A*STAR), Singapore*

Langerhans cell histiocytosis (LCH) is a potentially fatal neoplasm, stemming from the mononuclear phagocyte (MNP) system. Our knowledge of LCH origin and pathogenesis, and thus the development of effective therapies, are hampered by an inability to proficiently discriminate neoplastic cells from normal MNPs at single -cell resolution. Here, we used single-cell RNA-seq and protein analysis to characterize LCH lesions, assessing LCH cell heterogeneity, comparing them to normal MNPs within tumor. We found LCH-discriminatory signatures pointing to senescence and escape from tumor immune surveillance. We also uncovered two major lineages of LCH with DC2- and DC3/Monocyte-like phenotypes, which high-content imaging detected across different pathological tissue sites. Our results support a dual origin model of LCH cell development with an underlying neoplastic "hit" occurring prior to fate commitment to DC2 and DC3/Monocyte lineages. Together these data represent a paradigm expansion in LCH understanding, providing a basis for the development of new therapeutic approaches.

GUEST SPEAKER PRESENTATIONS

TUESDAY, OCTOBER 12, 2021 | 1100-1145

VIRTUAL MEETING

HEMOPHAGOCYTIC LYMPHOHISTIOCYTOSIS

Jana Pachlopnik Schmid

University Children's Hospital Zurich, Zurich, Switzerland

Studies of monogenic immune disorders ultimately prove the relevance of an immune system's *component in human biology. Persistent, recurrent, life-threatening or fatal inflammatory syndromes in neonates, infants and toddlers with inborn errors of immunity sadly exemplify the importance of certain gene products in maintaining immune homeostasis. One important group of diseases with immune dysregulation are hemophagocytic lymphohistiocytosis (HLH) and HLH-like diseases. The fact that inherited HLH syndromes are mainly related to genetic impairments in granule-dependent lymphocyte cytotoxicity, emphasizes the perforin/ granzyme-apoptosis pathway's critical role in immune homeostasis. We are applying a joint genomic and transcriptomic approach, combined with thorough functional and phenotypical examinations of leukocytes and fibroblasts from patients with HLH and HLH-like phenotypes. Analyses of these datasets in combination with functional validation in animal models enabled us to identify CD48 and ZNFX1 as important players in human immune homeostasis.*

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***Note: These abstracts can also be found in the Poster Presentations section.**

MACROPHAGE ACTIVATION SYNDROME (MAS) IN SYSTEMIC JUVENILE IDIOPATHIC ARTHRITIS (SJIA): TREATMENT WITH EMAPALUMAB, AN ANTI-INTERFERON GAMMA (IFNγ) MONOCLONAL ANTIBODY

Fabrizio De Benedetti[1], Alexei Grom[2], Paul Brogan[3], Claudia Bracaglia[1], Manuela Pardeo[1], Giulia Marucci[1], Despina Eleftheriou[3], Charalampia Papadopoulou[3], Pierre Quartier[4], Jordi Antón[5], Rikke Frederiksen[6], Veronica Asnaghi[6], Maria Ballabio[6], Cristina de Min[6]

1Division of Rheumatology, Ospedale Pediatrico Bambino Gesù, IRCCS, Rome, Italy; 2Cincinnati Children's Hospital, Division of Rheumatology, Cincinnati, OH, United States; 3UCL Institute of Child Health, and Great Ormond Street Hospital NHS Foundation Trust, London, United Kingdom; 4Université de Paris, IMAGINE Institute, RAISE Reference Centre, Pediatric Immuno-Hematology and Rheumatology Unit, Necker Hospital, Assistance Publique-Hôpitaux de Paris, Paris, France; 5Hospital Sant Joan de Déu, Division of Pediatric Rheumatology, University of Barcelona, Barcelona, Spain; 6Swedish Orphan Biovitrum AG (Sobi), Basel, Switzerland

PURPOSE: MAS is a severe, life-threatening complication of rheumatic diseases that occurs most frequently in patients with sJIA. Preclinical and clinical data suggest that overproduction of IFNγ is a driver of the hyperinflammation observed in MAS. Herein, we report on the efficacy and safety of emapalumab, a fully human anti-IFNγ monoclonal antibody, in patients with MAS associated with sJIA. METHODS: This pilot, open-label, single-arm, phase 2 study (NCT03311854) included patients with MAS (2016 ACR/EULAR criteria) associated with sJIA and with an inadequate response to high dose IV GC and other treatments. Initial emapalumab dose was 6 mg/kg and continued at 3 mg/kg twice weekly for 4 weeks, or less upon achievement of complete response. RESULTS: We report preliminary results from 14 enrolled patients (11 in Europe; 3 in USA) with a median age of 11 (range 2-25) years. All patients had failed high-dose GC or other therapies. Treatment with emapalumab resulted in rapid IFNγ neutralization, as demonstrated by decreased IFNγ-induced chemokine (C-X-C motif) ligand 9 levels, and subsequent deactivation of T cells, as indicated by decreased soluble interleukin-2 receptor levels. Six patients received treatment for 4 weeks, 7 terminated treatment early because of MAS remission (Investigator's assessment), and 1 patient received treatment up to Day 38 to achieve MAS control. Progressive improvement in all clinical and laboratory parameters of MAS was observed. Emapalumab infusions were well tolerated with no discontinuations. A cytomegalovirus reactivation was reported in 1 patient as a serious adverse event related to emapalumab and resolved with antiviral therapy. CONCLUSION: Emapalumab administration led to rapid neutralization of IFNγ and was efficacious in controlling MAS with a favorable safety profile. These RESULTS support the pathogenic role of IFNγ in MAS/sJIA and the therapeutic value of IFNγ neutralization in MAS patients who have failed high-dose GCs and other treatments.

RESPONSE TO MAPK INHIBITION OF NEURODEGENERATION IN LCH MONITORED BY CSF NEUROFILAMENT LIGHT AS A BIOMARKER

Jan-Inge Henter[1,2]; Egle Kvedaraite[1,3,4]; Daniel Martín Muñoz [5,6]; Monica Cheng Munthe-Kaas[7]; Bernward Zeller[7]; Tove Nystad[8]; Caroline Björklund [9]; Isabella Donnér[10]; Magda Lourda[1,3]; Henrik Zetterberg[11,12,13,14]; Kaj Blennow[11,12]; Nikolas Herold[1,2]; Désirée Gavhed[1,2]; and Tatiana von Bahr Greenwood[1,2]

1Childhood Cancer Research Unit, Department of Women's and Children's Health, Karolinska Institutet, Stockholm, Sweden; 2Pediatric Oncology, Astrid Lindgrens Children's Hospital, Karolinska University Hospital, Stockholm, Sweden; 3Center for Infectious Medicine, Department of Medicine Huddinge, Karolinska Institutet, Stockholm, Sweden; 4Department of Clinical Pathology, Karolinska University Laboratory, Stockholm, Sweden; 5Department of Clinical Neuroscience, Karolinska Institutet, Stockholm, Sweden; 6Department of Neuroradiology, Karolinska University Hospital, Stockholm, Sweden; 7Department of Pediatric Hematology and Oncology, Oslo University Hospital, Oslo, Norway; 8Department of Pediatrics, Division of Child and Adolescent Health, University Hospital of North-Norway, Tromsø, Norway; 9Department of Pediatric Hematology and Oncology, Umeå University Hospital, Umeå, Sweden; 10Hallands Hospital, Halmstad, Sweden; 11Clinical Neurochemistry Laboratory, Sahlgrenska University Hospital, Mölndal, Sweden; 12Department of Psychiatry and Neurochemistry, Institute of Neuroscience and Physiology the Sahlgrenska Academy at the University of Gothenburg, Mölndal, Sweden; 13UK Dementia Research Institute at UCL, London, UK; 14Department of Neurodegenerative Disease, UCL Institute of Neurology, London, UK.

PURPOSE: Langerhans cell histiocytosis (LCH) is an inflammatory myeloid neoplasia that can affect multiple organs. CNS involvement (CNS-LCH), which often affects pituitary function, can also cause slowly progressive neurodegeneration (ND-CNS-LCH), which frequently is resistant to conventional LCH-directed therapy. Notably, a population-based study reported that at least 24% of all children with LCH develop signs of ND-CNS-LCH on long-term follow-up with magnetic resonance imaging (MRI). Thus, a strategy for early detection, treatment, and monitoring of ND-CNS-LCH is imperative. In most LCH patients, somatic activating genetic alterations in the mitogen-activated protein kinase (MAPK) pathway can be detected. Targeted MAPK inhibition (MAPKi) has pronounced clinical efficacy in refractory LCH. However, evidence of the therapeutic efficacy of MAPKi in established ND-CNS-LCH is limited. METHODS: In this study, we evaluated the response to MAPKi therapy in five children with CNS-LCH, four of whom with ND-CNS-LCH, by analyzing their cerebrospinal fluid (CSF) for biomarkers of neurodegeneration, including neurofilament light (NFL). CSF-NFL is a sensitive and well-established biomarker of neuroaxonal damage. RESULTS were correlated with clinical and neuroradiological findings. RESULTS: Notably, CSF-NFL levels were initially pathologically elevated in all children with CNS-LCH, but normalized (<380 ng/L) within 6 months in four children and in all five within 9 months after initiation of MAPKi therapy (p=0.041, paired t-test). Notably, in the two patients that MAPKi therapy was discontinued, CSF-NFL levels increased again to abnormal levels within 4 months. MAPKi therapy was associated with perceivable neuroradiological and clinical improvement in three and two children, respectively. For comparison, CSF-NFL levels were normal in all but one (440 ng/L) of 16 samples from 11 additional children with LCH without known CNS-LCH. CONCLUSION: We conclude that CSF-NFL is a relevant surrogate biomarker in ND-CNS-LCH, and that MAPKi therapy appears to effectively reduce this neurodegeneration.

ABSTRACTS NOMINATED FOR THE NEZELOF AWARD IN BASIC SCIENCE:

CYTOTOXIC IMPAIRMENT AND EXCESS IL-18 DRIVE SPONTANEOUS, OLIGOCLONAL CD8 T-CELL HYPERACTIVATION AND HYPERINFLAMMATORY DISEASE

Emily Landy[1], Canna, Scott[2]

1University of Pittsburgh, Pittsburgh PA, USA; 2Children's Hospital of Philadelphia, Philadelphia PA, USA

PURPOSE: Mounting evidence suggests partial defects in cytotoxicity-related genes may promote hyperinflammation in Macrophage Activation Syndrome (MAS) patients, who uniformly have highly-elevated IL-18. In mice, neither excess IL-18 (18tg mice) nor perforin-deficiency individually cause immunopathology without inflammatory challenge. However,18tg mice lacking perforin (18tg;Prf1-/-) develop spontaneous lethal hyperinflammation, while subclinical MAS was found in 18tg;Prf1+/- mice. We sought to understand how these two susceptibility factors, IL-18 and cytotoxic impairment, synergize to drive spontaneous hyperinflammation. METHODS: We tracked the abundance, phenotype, and proliferation of lymphocytes from such mice via flow cytometry, bulk RNAseq, and T Cell Receptor (TCR) sequencing. RESULTS: The expanded CD8 T-cells from 18tg;Prf1-/- mice showed a flow cytometric pattern with low expression of CD44 and CD62L, but high expression of PD-1. Transcriptional analysis in splenic CD8 T-cells from 18tg;Prf1-/- and 18tg;Prf1+/- splenic CD8 T cells demonstrated a hyperactivated, terminal-effector phenotype with increased expression of both inhibitory receptors (e.g. Pdcd1, Cd39, "¦) and effector molecules (Ifng, Gzmk) in comparison to controls and to canonical, publicly-available exhaustion transcriptomes. TCR sequencing showed dramatic clonal hyper-expansion of TCR sequences from 18tg;Prf1-/- CD8 T-cells as compared to controls, but no significant overlap in individual clones between biologic replicate samples. Longitudinal analyses showed an expansion of these hyperactivated CD8 T-cells in peripheral blood as the mice aged. CONCLUSION: These data suggest that IL-18 and cytotoxicity drive progressive and pathologic activation of a stochastic, oligoclonal CD8 T-cell population that underlies life-threatening immunopathology in HLH and MAS. Importantly, all systemic and CD8 T-cell findings observed in 18tg;Prf1-/- mice were present (albeit less dramatically) in 18tg;Prf1+/- mice. This suggests graded defects in perforin-mediated cytotoxicity interact with systemic IL-18 excess to drive CD8 T-cell hyperactivation and cytokine storm susceptibility. Targeting the development and maintenance of this population may be critical for the treatment of MAS and other hyperinflammatory disorders.

LINEAGE-SWITCH OF CELLS HARBORING BRAF V600E ALLELES IN PATIENTS WITH HIGH RISK LCH TREATED WITH INHIBITORS

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Patients with high-risk LCH who progress through front-line therapy have conventionally been treated with high dose salvage chemotherapy. The majority have BRAFV600E mutation, and BRAF inhibitors such as vemurafenib and dabrafenib can also induce dramatic clinical improvement in those how fail salvage. Circulating BRAFV600E alleles are detectable in myeloid mononuclear cells and cell-free DNA prior to and during therapy. However, it is not known how the level of mutation and cellular compartments evolve after long term treatment with conventional chemotherapy or inhibitors. Here we present the profiles of seven high risk patients, two treated with conventional chemotherapy, two treated with inhibitors only and three treated with inhibitors and cytoreductive chemotherapy. In all patients, mutation was detected predominantly in monocytes and myeloid dendritic cells at the start of therapy using a highly sensitive allele-specific quantitative PCR. Patients responding to cytarabine and cladribine on LCHIV completely cleared BRAFV600E within six months. However, patients who were refractory to chemotherapy and went on to receive inhibitors maintained a high level of BRAFV600E in cellular and cell-free DNA for several years. Fractionation of mononuclear cells at later time points revealed a surprising shift of mutated alleles to the T cell compartment, such that this became the major reservoir of mutation in all four patients maintained on inhibitor therapy for more than two years. One of these patients subsequently developed neuro-degeneration whilst on therapy. Involvement of the lymphocyte compartment late in the course of high-risk LCH raises important questions: 1) the presence of mutation in T cells and myeloid cells is consistent with involvement of the hematopoietic stem cell; 2) lymphocytes have access to the CNS suggesting a potential mechanism of neurodegeneration; 3) a transition from myeloid to lymphoid cell reservoirs may mark the potential for withdrawal of targeted therapy without systemic relapse.

BRAF-V600E EXPRESSION IN DENDRITIC CELLS REPROGRAMS TRANSLATIONAL DYNAMICS TO INCREASE LPS-INDUCED TNFA PRODUCTION

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PURPOSE: Langerhans Cell Histiocytosis (LCH) is driven by abnormal mononuclear phagocytes closely related to dendritic cells (DCs) with hyper-MAPK/ERK signaling, most commonly due to the Braf-V600E mutation. Since DCs are poised with environmental sensors such as Toll like Receptors (TLRs) that require MAPK/ERK signaling to incite an immune response, we hypothesized that LCH cells have altered responses to environmental cues. We tested this notion using the model system of LPS-induced TNFa production. METHODS: We utilized the BRAFV600E-Flox: CD11c-Cre mouse model of LCH which expresses Braf-V600E in DCs. The BRAF V600E inhibitor, PLX7904, was used to test for reversibility of phenotypes. We used LPS to stimulate TLR4-induced TNFa production. We measured TNFa by qPCR, ELISA, and intracellular flow cytometry. Translational activity was measured by polysome profiling and qPCR, and activation of translation factors was measured by western blot analysis. RESULTS: LCH mice have increased LPS-induced circulating TNFa compared to WT. V600E-BMDCs and splenic-DCs have a reversible LPS-induced increase in TNFa secretion and intracellular accumulation. However, this is despite a decrease in LPS-induced tnfa transcription compared to WT. Polysome profile data demonstrates increased polysome occupancy of transcripts in V600E-BMDCs, and qPCR confirms an increase in TNFa translation efficiency.

Western blot analysis shows a decrease in levels of phosphorylated-eEF2 consistent with an increase in translational elongation. CONCLUSION: LCH cells demonstrated heighten LPS-induced TNFa production, despite diminished tnfa transcription. Increased intracellular TNFa indicates a cell intrinsic, post-transcriptional mechanism responsible for this disconnect. Polysome profiling suggests increased rates of global translation, including TNFa. Reduced levels of phosphorylated-eEF2 indicate increased elongation rates, potentially contributing to the increased translation. These data indicate novel effects of Braf-V600E on DC translational dynamics that result in abnormal inflammatory behavior and provide a potential novel target for LCH therapies.

ABSTRACTS NOMINATED FOR THE NESBIT AWARD IN CLINICAL SCIENCE:

THE INTERIM RESULTS OF A MULTICENTER STUDY OF SAFETY AND EFFICACY OF VEMURAFENIB AND CYTARBINE PLUS CLADRIBINE THERAPY IN HIGH-RISK PEDIATRIC LANGERHANS CELL HISTIOCYTOSIS WITH BRAF V600E MUTATION

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PURPOSE: A prospective pilot trial to evaluate the safety and efficacy of combined vemurafenib and chemotherapy in high-risk Langerhans' cell histiocytosis (LCH) patients. METHODS: Eighteen patients (8 boys, 10 girls; median age 22 months) with BRAF V600E positive LCH were enrolled. Treatment included a combination of vemurafenib (median dose 21 mg/kg/day) and cytarabine (Ara-C) at 100 mg/m2/12 h, days 1-5) and cladribine (2-CdA, 6 mg/m2/day, days 1-5). Twelve pts with untreated RO+ LCH and 6 pts with RO- multisystem LCH after at least two lines of unsuccessful therapy were included (median DAS 9.5 points, range 1 - 20 points). The induction therapy consisted of 28 days of vemurafenib monotherapy. After that, three courses of Ara-C + 2-CdA were conducted. Vemurafenib treatment was continued between the courses. After three courses of LDAra-C + LD2-CdA, we attempted to cease vemurafenib therapy. Three courses of mono 2-CdA therapy were used as maintenance. MRD was monitored by ddPCR in cell-free DNA (cfDNA) in peripheral blood and flow cytometry sorted bone marrow derived myeloid precursors (CD34+CD19-). RESULTS: All except one patient received the protocol therapy and vemurafenib therapy was stopped. In 13 patients (76%), we did not observe the relapse; their median EFS from vemurafenib cessation is 11.1 months (range 4.1 - 34.2 months). In 4 (24%) patients, we have observed a relapse soon after vemurafenib cessation (median time 2.8 months, range 1.1 - 8 months). In 1 patient, we observed a full-scale relapse with RO lesions, and in the other three patients, local skin and bone lesions were detected. All relapsed patients restarted vemurafenib therapy with full response. No patient died. No vemurafenib induced toxicity with grade 2 or more (according to CTCAE 5.0) was observed. CONCLUSION: Vemurafenib and Ara-C + 2-CdA combination is safe and might allow vemurafenib withdrawal in patients with LCH.

PERI-TRANSPLANT ALEMTUZUMAB LEVELS PREDICT RISK OF SECONDARY GRAFT FAILURE AND INVERSELY IMPACT CXCL9 LEVELS AFTER RIC-HCT (A CORRELATIVE BIOLOGY STUDY TO BMT-CTN 1204 RICHI)

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PURPOSE: Mixed chimerism (MC) leading to secondary graft failure (GF) remains a challenge following alemtuzumab, fludarabine and melphalan reduced-intensity conditioning (RIC) hematopoietic cell transplant (HCT) in hemophagocytic lymphohistiocytosis (HLH) and other primary immunodeficiencies (PID). Mechanisms for GF are not known. Expansion of recipient T-cells and interferon-gamma pathway activation have been proposed as drivers for GF. However, high peri-transplant alemtuzumab levels have been associated with MC and secondary GF, suggesting an inverse relationship between GF and immune activation in RIC-HCT. Our objective was to evaluate cytokine patterns and alemtuzumab levels and their association with durable engraftment. METHODS: Patients enrolled in BMT-CTN 1204 study (NCT01998633) were eligible for this correlative biology study. Secondary GF was defined as donor chimerism <5% after initial engraftment and/or requirement of donor lymphocyte infusion or second HCT. RESULTS: Thirty-three patients were included with HLH (n=25) and other PID (n=8). One patient (3%) developed primary GF and 11 (33%) developed secondary GF. Cytokine analysis revealed patients with secondary GF had lower CXCL9 levels on day +14 and +28. The cumulative incidence (CI) of secondary GF in patients with a day +14 CXCL9 level ≤2394pg/mL (day+14 median) was 73.6% vs 0% in patients with a level >2394pg/mL(p=0.002). Further analysis identified patients with day 0 alemtuzumab levels ≤0.32µg/mL had higher peri-transplant CXCL9 levels compared to patients with levels >0.32µg/mL. Finally, we examined the impact of alemtuzumab levels on secondary GF. The CI of secondary GF was 0% in patients with alemtuzumab levels ≤0.32µg/mL compared to 54.3% in patients with levels >0.32µg/mL(p=0.08). CONCLUSION: This study demonstrates a relationship between alemtuzumab levels and durable engraftment. Further, interferon-gamma activity, as reflected by CXCL9, inversely correlated with peri-transplant alemtuzumab levels. Our findings support that higher alemtuzumab levels drive efficient T-cell depletion of the stem cell product which increases the risk of MC and secondary GF.

ALK+ HISTIOCYTOSIS: A NEW CLINICOPATHOLOGIC SPECTRUM HIGHLIGHTING NEUROLOGIC INVOLVEMENT AND RESPONSES TO ALK INHIBITION

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ALK-positive histiocytosis is a rare subtype of histiocytic neoplasm first described in 2008 in three infants with multisystemic disease involving the liver and hematopoietic system. This entity has subsequently been documented in case reports and series to occupy a wider clinicopathologic spectrum with recurrent KIF5B-ALK fusions. The full clinicopathologic and molecular spectra of ALK-positive histiocytosis remain, however, poorly characterized. Here, we describe the largest study of ALK-positive histiocytosis to date, with detailed clinicopathologic data of 35 cases, including 33 cases with confirmed ALK rearrangements. The clinical spectrum comprised distinct clinical phenotypic groups: infants with multisystemic disease with liver and hematopoietic involvement, as originally described (Group 1A: 6/35), other patients with multisystemic disease (Group 1B: 10/35), and patients with single-system disease (Group 2: 19/35). Seventeen patients of the entire cohort (49%) had neurologic involvement (seven and ten from Groups 1B and 2, respectively). Histology included classic xanthogranuloma features in one quarter of cases, whereas the majority displayed a more densely cellular, monomorphic appearance without lipidized histiocytes but sometimes more spindled or epithelioid morphology. Neoplastic histiocytes were positive for macrophage markers and often conferred strong expression of phosphorylated-ERK, confirming MAPK pathway activation. KIF5B-ALK fusions were detected in 23 patients, while CLTC-ALK, TFG-ALK, EML4-ALK and DCTN1-ALK fusions were identified in single cases. Robust and durable responses were observed in ten patients treated with ALK inhibition, nine with neurologic involvement. This study presents the existing clinicopathologic and molecular landscape of ALK-positive histiocytosis, and provides guidance for the clinical management of this emerging histiocytic entity.

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NOTES

Poster #1

HEMOPHAGOCYTIC LYMPHOHISTIOCYTOSIS: 10 YEARS DATA FROM ARMENIA

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PURPOSE: To analyze the incidence, characteristics, and outcomes of hemophagocytic lymphohistiocytosis (HLH) in Armenia during the last 10 years. METHODS: We conducted a retrospective review of medical records of patients with HLH at the main treatment facilities in Armenia â˜ Hematology Center after Prof. R.Yeolyan and Muratsan Hospital Complex of Yerevan State Medical University over the last 10 years (2011-2021). The HLH-2004 diagnostic criteria were used for the diagnosis. Evaluation of NK-cell activity, soluble CD25 levels, and underlying genetic defects of familial HLH are unavailable in our country. RESULTS: Five patients (3 male and 2 female) with HLH diagnosis were identified with a mean age of 25.5 years, a median age of 12 years, range [5 months-70 years]. In addition to these 5 patients, 1 patient (5-month-old) was suspicious to have HLH (she met 4 out of 8 HLH-2004 criteria, however, she met 11 out of 18 criteria developed by Tamamyan et al, Cancer, 2016), and 1 patient (2.5-year-old) with HLH had unavailable documentation. Two patients with HLH had Hodgkin lymphoma (HL), 1 had acute lymphoblastic leukemia, 1 patient with Niemann-Pick disease, 1 had pneumonia, complicated by sepsis. Four patients (80%) were positive for Epstein-Barr virus (EBV) infection, 1 of them had CMV infection. The more common clinical manifestations include fever, hepatosplenomegaly, pancytopenia, hyperferritinemia, hypoalbuminemia (100%), hypertriglyceridemia, hypofibrinogenemia, elevated hepatic enzymes 60% (⅗) and LDH levels 40% (⅖). Patients with malignancy-associated HLH received treatment with appropriate chemotherapy regimens. Three patients received symptomatic therapy, including corticosteroids. One of the patients was treated with IVIG. Three patients expired during the treatment. Patients with HL (1-year follow-up) and Niemann-Pick disease (11-year follow-up) are alive. CONCLUSION: This is the first report on HLH from Armenia. Although it is a rare disorder, it can be significantly underdiagnosed and needs improvement in diagnostic and treatment approaches.

Poster #2

CONGENITAL SYPHILIS MAY MIMIC HEMOPHAGOCYTIC LYMPHOHISTIOCYTOSIS AND SHOULD BE RULED OUT IN NEWBORNS

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PURPOSE: Hemophagocytic lymphohistiocytosis (HLH) in neonatal period is extremely rare. It is associated with high morbidity and mortality, and early recognition and diagnosis are essential. The differential diagnosis is broad, genetic causes should be investigated and infections that can mimic HLH should be ruled out. METHODS: Review of the initial clinical presentation of one newborn and treatment response. RESULTS: A 12-day-old male with fever and macular rash with lesions in extremities and palmoplantar involvement of 2 days of evolution. First child of non-consanguineous parents, born at week 37. Mother: 22-year-old with normal gestational control and negative syphilis test. Physical examination showed hepatosplenomegaly. Laboratory analysis: normal liver and kidney function, Reactive Protein-C:154.61mg/L, Procalcitonin: 3.30ng/mL, Hemoglobin: 14.2g/dL, Platelets: 37*10^3/µL and Leukocytes: 15.50*10^3/µL (Neutrophils: 5.32*10^3/µL; Lymphocytes: 5.02*10^3/µL) with normal coagulation study. Treatment with broad-spectrum intravenous antibiotic therapy was started. He showed clinical deterioration in the following hours, with respiratory distress requiring respiratory support with non-invasive ventilation, and hemodynamic lability with oliguria and hyperlactacidemia (4mmol/L). When sepsis of viral etiology was suspected, doses of intravenous nonspecific gamma globulin were administered. At 36 hours, he showed an increased hepatosplenomegaly, an elevation of ferritin, hypertriglyceridemia and bicitopenia (anemia and thrombocytopenia). Bone marrow aspirate showed hemophagocytosis. Not elevated sCD25:7,757.11pg/ml. Microbiological studies confirmed syphilis (Rapid Plasma Reagin (RPR) positive (1:8) and Treponema pallidum PCR positive), so specific antibiotic treatment with intravenous penicillin was administered for 10 days. The clinical evolution was favorable with progressive resolution of the infection and HLH. CONCLUSION: In the neonatal period, it is important to identify the clinical pictures of HLH due to their severity and multiorgan involvement. Possible infectious causes should be analyzed, including microbiological studies for syphilis (PCR and RPR). Identification and treatment of the infectious trigger can lead to complete resolution of the clinical picture of HLH, without the need for specific therapies if the response is favorable.

Poster #3

EFFICACY OF ALLOGENEIC HEMATOPOIETIC STEM CELL TRANSPLANTATION IN TREATMENT OF PEDIATRIC CHRONIC ACTIVE EPSTEIN-BARR VIRUS INFECTION COMPLICATED WITH HEMOPHAGOCYTIC LYMPHOHISTIOCYTOSIS: A REPORT OF 18 CASES FROM A SINGLE CENTER

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OBJECTIVE: Hemophagocytic lymphohistiocytosis (HLH) is a serious complication of chronic active EBV infection(CAEBV). To explore the effectiveness of allogeneic hematopoietic stem cell transplantation (allo-HSCT) for CAEBV complicated with HLH, we present our single institutional experience. METHODS: A total of 18 patients diagnosed as CAEBV complicated with HLH were retrospectively reviewed from July 2015 to January 2021. All patients underwent allo-HSCT. RESULTS: There were 12 males and 6 females. The median age at HSCT was 9.1(1.7~17.3) years old. None of the 18 patients got complete remission of HLH, 13 patients got partial remission and 5 patients got HLH progression berore HSCT. EBV-DNA in plasma was over 10^2.5 copies/ml in 8 patients when HSCT. 12, 4 and 2 patients underwent haploid, matched sibling donor and unrelated donor HSCT, respectively.The median mononuclear cell dose in transfusion was 9.0(5.8~15.5)x10^8/kg, and the median CD34+ cell dose was 5.0(3.0~10.8)x10^6/kg. The success rate of engraftment in 18 patients was 83.3% . The occurrence of acute graft-versus-host disease(GVHD) from Ⅱ to Ⅳ degree and chronic GVHD were 53.3% and 30.8%. After 9 (0.3~72) months median follow-up, 7 patients died. Among them, only 3 patients were in haploid HSCT group. The median time from death to transplantation of 7 patients was 1.5(0.3~5) months. The overall survival was 61.1%. CONCLUSIONS: Allo-HSCT seems to be a promising approach for the treatment of CAEBV complicated with HLH, and haploid HSCT could be an suitable option.

Poster #4

MACROPHAGE ACTIVATION SYNDROME (MAS) IN SYSTEMIC JUVENILE IDIOPATHIC ARTHRITIS (SJIA): TREATMENT WITH EMAPALUMAB, AN ANTI-INTERFERON GAMMA (IFNγ) MONOCLONAL ANTIBODY

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PURPOSE: MAS is a severe, life-threatening complication of rheumatic diseases that occurs most frequently in patients with sJIA. Preclinical and clinical data suggest that overproduction of IFNγ is a driver of the hyperinflammation observed in MAS. Herein, we report on the efficacy and safety of emapalumab, a fully human anti-IFNγ monoclonal antibody, in patients with MAS associated with sJIA. METHODS: This pilot, open-label, single-arm, phase 2 study (NCT03311854) included patients with MAS (2016 ACR/EULAR criteria) associated with sJIA and with an inadequate response to high dose IV GC and other treatments. Initial emapalumab dose was 6 mg/kg and continued at 3 mg/kg twice weekly for 4 weeks, or less upon achievement of complete response. RESULTS: We report preliminary results from 14 enrolled patients (11 in Europe; 3 in USA) with a median age of 11 (range 2-25) years. All patients had failed high-dose GC or other therapies. Treatment with emapalumab resulted in rapid IFNγ neutralization, as demonstrated by decreased IFNγ-induced chemokine (C-X-C motif) ligand 9 levels, and subsequent deactivation of T cells, as indicated by decreased soluble interleukin-2 receptor levels. Six patients received treatment for 4 weeks, 7 terminated treatment early because of MAS remission (Investigator's assessment), and 1 patient received treatment up to Day 38 to achieve MAS control. Progressive improvement in all clinical and laboratory parameters of MAS was observed. Emapalumab infusions were well tolerated with no discontinuations. A cytomegalovirus reactivation was reported in 1 patient as a serious adverse event related to emapalumab and resolved with antiviral therapy. CONCLUSION: Emapalumab administration led to rapid neutralization of IFNγ and was efficacious in controlling MAS with a favorable safety profile. These RESULTS support the pathogenic role of IFNγ in MAS/sJIA and the therapeutic value of IFNγ neutralization in MAS patients who have failed high-dose GCs and other treatments.

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Poster #5

FIRST REPORT OF HEMOPHAGOCYTIC LYMPHOHISTIOCYTOSIS (HLH) INDUCED BY CANDIDATUS NEOEHRLICHIA MIKURENSIS. A RETROSPECTIVE FINDING AFTER RESPONSE TO EMPIRICAL DOXYCYCLINE. IS IT TIME FOR UPFRONT MICROBIOME ANALYSIS IN HLH?

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PURPOSE: To report the discovery of a novel HLH-inducing pathogen. METHODS: Case report. RESULTS: A patient with previous non-Hodgkin lymphoma presented in February 2021 with malaise, weight loss, high-spiking fever, and encephalopathy. She was cytopenic and hyperferritinemic. A PET-CT scan showed no signs of lymphoma-relapse apart from splenomegaly. The spinal fluid was normal apart from increased csv-sCD163. Bone marrow examination revealed hemophagocytotic cells. She fulfilled 7/7 HLH-2004 criteria and had an h-score of 266 (99,8% HLH-risk). Evaluation of all common HLH-triggers did not reveal an offending pathogen. As lymphoma relapse remained a suspicion, we deferred from glucocorticoids. Immunoglobulin 1.6 g/kg was without effect. On experimental N-acetylcysteine-treatment, the ferritin level decreased from 7400 to 2000, but the fever persisted. Ruxolitinib 20 mg bid was initiated with swift resolution of fever and symptoms. After six days she was able to be discharged with a walker. Ruxolitinib treatment was tapered over the next 61 days. At ruxolitinib-cessation she was anemic and thrombocytopenic (104 x 109/L). After cessation the hemoglobin improved slightly, but her symptoms started reappearing. We administered empirical doxycycline 100 mg bid for 10 days. After five days, all symptoms had disappeared, and the platelet level was normalized. By day 21 her hematology, sCD163, and sIL2r levels had normalized for the first time. We retrospectively performed 16S PCR on blood and bone marrow, revealing Candidatus Neoehrlichia mikurensis, a tick-borne bacterium of the family Anaplasmataceae, not previously reported to cause HLH, and which has only been described in one single patient in our nation. CONCLUSION: Surprisingly, empirical doxycycline caused swift and complete resolution of hyperinflammatory symptoms in a patient. Microbiome examination revealed a new HLH-inducing pathogen. Given the multitude of rare potentially HLH-causing pathogens and the toxicity of standard HLH therapy, upfront microbiome evaluation of blood and bone-marrow should be considered in HLH.

Poster #6

N-ACETYLCYSTEINE AS A TREATMENT MODALITY IN HYPERINFLAMMATORY DISEASES. PROMISING RESULTS IN HEMOPHAGOCYTIC LYMPHOHISTIOCYTOSIS

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PURPOSE: To describe the rationale behind the use of N-acetylcysteine (NAC) in hyperinflammatory conditions, and to report the response to NAC-treatment in a patient with severe, refractory hemophagocytic lymphohistiocytosis (HLH). METHODS: case report and literature review. RESULTS: A patient with previous T-cell lymphoma developed secondary acute myeloid leukemia. He had an inflammatory phenotype with a ferritin up to >200,000 ug/L and a sCD163 of 55.5 (N:0.69-3.86) mg/L. At debut he experienced strokelike symptoms which rendered him hemiparetic thus limiting the treatment options for the leukemia. He received intravenous cytarabine but progressed to unconsciousness and exhibited a pathological electroencephalography. A spinal tap showed no signs of leukemia or infection but grossly elevated csv-sCD163. Neither intravenous dexamethasone, etoposide, nor intrathecal cytarabine or methotrexate improved his clinical condition. We decided to administer continous NAC 100 mg/kg/24 hours. The day after initiation he started improving, and could eventually be discharged from intensive unit care to rehabilitation. In spite of azacytidine treatment the leukemia relapsed after three months. Again he was hyperinflammed and exhibited neurological symptoms. NAC/dexamethasone was reinitiated for 5 days. The ferritin dropped from 9,770 to 2,250 and he showed neurological improvement. Two days after cessation of NAC the ferritin increased to 5,900 and the clinical condition deterioated on unchanged dexamethasone dose. Ultimately, the patient succombed to the diseases. NAC is an antioxidant, suppress Nuclear-factor-kappa B activity and may modulate macrophage activity. NAC has been shown to reduce sCD163 in healthy subjects. CONCLUSION: We describe marked improvement with temporal relationsship to NAC-treatment in a patient with severe HLH. NAC may have immunomodulatory effects that could benefit patients with HLH. We have some additional experience with NAC in HLH and other inflammatory conditions, however a collaborative effort is needed to unravel the true potential of this readily availiable low-cost treatment modality.

Poster #7

CHARACTERIZATION OF PATHOGENIC VARIANTS CAUSING PRIMARY HEMOPHAGOCYTIC LYMPHOHISTIOCYTOSIS: A CASE REPORT AND GENETIC ANALYSIS

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PURPOSE: Primary hemophagocytic lymphohistiocytosis (HLH) is a rare form of severe immune dysregulation characterized by uncontrolled systemic hyperinflammation leading to life-threatening fevers, cytopenias, organomegaly, hyperferritinemia, and hemophagocytosis. Primary HLH results from genetic deficits in lymphocyte cytotoxicity that are required for negative feedback to prevent hyperinflammation. The genetic variants responsible for primary HLH are still being characterized. METHODS: Next-generation sequencing was used to detect variants in 36 HLH-related genes and PCR was used to detect a large inversion. RNA analysis determined the effect of a start-lost variant. RESULTS: Here we report a 3-month-old with severe primary HLH and characterize two rare pathogenic variants in the PRF1 gene. CONCLUSION: Obtaining next-generation sequencing RESULTS within two days was important for prompt diagnosis and successful treatment.

Poster #8

XIAP DEFICIENCY SUCCESSFULLY MANAGED WITH MONOTHERAPY TADEKINIG ALFA (IL-18BP)

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PURPOSE: X-linked inhibitor of apoptosis (XIAP) deficiency results in dysregulation of inflammasome activity. Patients with XIAP deficiency can have highly elevated levels of total IL-18 and detectable free IL-18 (not bound to IL-18 binding protein). IL-18 is a potent proinflammatory cytokine which enhances IFN-γ production. In this report, we describe a patient with XIAP deficiency with immune dysregulation that was successfully managed with monotherapy tadekinig alfa (recombinant human IL-18BP). METHODS: Retrospective chart review. RESULTS: An 8-year-old male with recurrent fevers, infections and colitis was diagnosed with XIAP deficiency at 8 years of age. Over 2.5 years he developed refractory disease despite multiple therapeutics including anakinra, canakinumab, tocilizumab, colchicine, etanercept, infliximab, mesalamine, rituximab and vedolizumab. His treatment included enrollment into a phase 3, placebo-controlled, randomized trial of tadekinig alfa (ClinicalTrials.gov Identifier NCT03113760). This study remains ongoing and unblinding has yet to occur, thus clinical data from this time period is not included in this report. After completion of this trial, he experienced flares with fevers, pain, and diarrhea despite budesonide, canakinumab and vedolizumab. His inflammatory biomarkers were elevated: total IL-18 was 54,687pg/mL and free IL-18 was 15.12pg/mL. He obtained access to compassionate use of tadekinig alfa 2 mg/kg subcutaneously every 48 hours. Promptly his symptoms resolved, other medications were gradually discontinued, and he has remained symptom free with monotherapy tadekinig alfa for the last 27 months. Free-IL-18 has remained unquantifiable (<10.00 pg/mL) and relevant disease biomarkers have remained low or normal. CONCLUSION: Patients with XIAP deficiency vary in presentation, severity and disease course, and there are no standard treatment recommendations. Our clinical experience suggests patients with XIAP deficiency may benefit from treatment with tadekinig alfa. These promising results highlight the importance of confirming the efficacy of tadekinig alfa in a controlled study, which may lead to broader access to this agent.

Poster #9

A TWO-COHORT, OPEN-LABEL, SINGLE-ARM STUDY OF EMAPALUMAB, AN ANTI-INTERFERON GAMMA (IFNγ) MONOCLONAL ANTIBODY, IN PATIENTS WITH MACROPHAGE ACTIVATION SYNDROME (MAS) IN RHEUMATIC DISEASES

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PURPOSE: MAS is a severe, life-threatening complication of rheumatic diseases such as Still's disease and systemic lupus erythematosus (SLE). Preliminary data from a pilot study including 9 patients with inadequate response to high-dose intravenous glucocorticoids (GCs) (NCT03311854) showed that emapalumab, a fully human anti-IFNγ monoclonal antibody, led to rapid IFNγ neutralization and was efficacious in controlling MAS with a favorable safety profile [De Benedetti et al. Annals Rheumatic Dis 2020;79 (Suppl. 1):194]. Herein, we describe the design and objectives of a Phase 2/3 study to further evaluate the efficacy and safety of emapalumab in patients with MAS in rheumatic diseases. METHODS: This open-label, single arm, multicenter, interventional study is enrolling pediatric and adult patients into two cohorts: 1) MAS in Still's disease (systemic juvenile idiopathic arthritis and adult onset Still's disease; n=25) and 2) MAS in SLE (n=16). Eligible patients must have an inadequate response to high doses of GCs. Each cohort is a single-arm study composed of two phases: an optional run-in phase and an interventional phase. Key eligibility criteria vary by cohort and phase. During the run-in phase, patients are treated with GCs according to the investigators' clinical practice and followed until MAS remission or inadequate response, or for a maximum of 12 weeks. Patients with an inadequate response to GCs (and potentially to other therapies) enroll in the interventional phase to be treated with intravenous emapalumab (initial dose 6mg/kg, subsequent doses 3mg/kg) for 4 weeks or until a complete response (CR) is achieved, and are followed off-drug for 1 year. RESULTS: The primary efficacy endpoint of the study is CR at Week 8 after first emapalumab administration. Other endpoints include GC tapering, survival, pharmacokinetics/pharmacodynamics, and safety. SUMMARY: This ongoing trial is designed to address the unmet need for new efficacious and safe therapies for the treatment of MAS.

Poster #10

SAFETY OF EMAPALUMAB IN PEDIATRIC PATIENTS WITH PRIMARY HEMOPHAGOCYTIC LYMPHOHISTIOCYTOSIS (HLH): RELATIONSHIP TO TREATMENT EXPOSURE

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INTRODUCTION: Primary hemophagocytic lymphohistiocytosis (HLH) is a rare, life-threatening, immune disorder characterized by a hyperinflammatory state in which interferon gamma (IFNγ) is considered a key cytokine. Current conventional therapy for HLH is based on immunochemotherapies, namely etoposide and glucocorticoids; however, this treatment is associated with opportunistic infections and severe myelotoxicity. This study describes prespecified exploratory exposure-safety analyses that were performed on data obtained from the pivotal trial of patients with primary HLH receiving emapalumab, a fully human, anti-IFNγ monoclonal antibody. METHODS: Data from a multicenter, open-label, pivotal phase 2/3 study (NCT01818492) and its long-term follow-up study (NCT02069899) were included in this analysis. Emapalumab safety was assessed in 34 primary HLH patients. Emapalumab was initiated at 1 mg/kg administered intravenously every 3 days, on a background of dexamethasone 5-10 mg/kg/day. Subsequent doses could be increased up to 10 mg/kg, if required, based on predefined laboratory and clinical response parameters. Planned treatment duration was up to 8 weeks. Exploratory graphical and logistic regression analyses were performed to determine the incidence of adverse events (AEs) as a function of emapalumab exposure. AEs emerging after the start of the first infusion until last infusion and prior to initiation of conditioning for transplantation were considered. RESULTS: The exposure-safety analyses did not reveal any apparent relationship between the number of AEs and exposure to emapalumab. In fact, a statistically significant decrease in the incidence of severe AEs and the incidence of AEs related to infusion-related reactions was observed. No clear trend was observed for selected parameters of renal and liver function based on the duration of emapalumab treatment. CONCLUSION: No significant relationships were observed between emapalumab exposure and incidence of AEs. These findings support the primary evidence of a favorable benefit-risk profile of emapalumab across the dose range used in this fragile patient population.

Poster #11

QUERCETIN AMELIORATES XIAP DEFICIENCY ASSOCIATED HYPERINFLAMMATION

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PURPOSE: XIAP deficiency is a rare genetic HLH disorder. XIAP deficiency causes hyperinflammation due to dysregulation of TNF-receptor signaling and NLRP3 inflammasome function. Safe and effective long-term treatments are needed. Here we evaluated the natural flavonoid antioxidant quercetin as a potential therapeutic. METHODS: Bone marrow derived macrophages were derived from XIAP deficient or wild type mice using L929 supernatant. Human monocytes were obtained from normal control or XIAP deficient patient blood samples following informed consent to a study approved by the Institutional Review Board. Murine or human cells were stimulated with TLR-agonists or TNF-α in the presence or absence of quercetin. For in vivo LPS challenge experiments, XIAP deficient or WT mice were fed mouse chow with or without supplemental quercetin (50mg/kg/day exposure) for 7 days followed by

challenge with 10ng/kg LPS. Terminal bleeds were performed 4 hours following LPS challenge. IL-1βand IL-18 were measured by ELISA. RESULTS: In murine studies, quercetin reduced or eliminated IL-1βsecretion by XIAP KO cells following TLR-agonist stimulations or TNF-α stimulation ($p<0.05$). Quercetin strongly reduced constitutive production of IL-18 by both WT and XIAP deficient cells ($p<0.05$). At 4 hours following in vivo LPS challenge, blood levels of IL-1βand IL-18 were significantly decreased in mice that had received quercetin-supplemented chow prior to LPS challenge ($p<0.05$). In experiments using human cells, quercetin greatly reduced IL-1βsecretion by human monocytes from normal control donors and patients with XIAP deficiency following stimulation with TNF-α ($p<0.05$). CONCLUSION: Quercetin may be an effective natural therapeutic for the prevention of hyperinflammation associated with XIAP deficiency. Clinical trials are warranted.

Poster #12

REDUCED INTENSITY CONDITIONING USING BUSULFAN, PENTOSTATIN, AND CYCLOPHOSPHAMIDE WITH POST TRANSPLANT CYCLOPHOSPHAMIDE IN PATIENTS WITH HLH UNDERGOING HEMATOPOIETIC STEM CELL TRANSPLANT

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PURPOSE: Hemophagocytic lymphohistiocytosis (HLH) is a hyperinflammatory condition caused by immune dysregulation. Genetic predisposition, malignancies and infectious etiologies such as the Epstein Barr Virus (EBV) can trigger HLH. Allogeneic hematopoietic stem cell transplantation (HSCT) remains the only curative treatment. Achieving and maintaining donor chimerism with reduced treatment toxicity, while ensuring lack of recurrence of HLH or EBV reactivation remains a challenge with current conditioning regimens. METHODS: We describe two EBV driven HLH patients that underwent HSCT with lymphodepletion-centered reduced intensity conditioning (RIC) as developed at the National Institutes of Health (NIH); consisting of pentostatin, low-dose cyclophosphamide (dose adjusted using absolute lymphocyte count), and 2 days of pharmacokinetically dosed busulfan. Graft-versus-host disease (GVHD) prophylaxis consisted of high-dose post-transplantation cyclophosphamide (PTCy) on days +3 and +4, followed by mycophenolate mofetil (MMF) and sirolimus. RESULTS: First patient - 4-year-old Vietnamese-American male received haploidentical (5/10 alleles) stem cells (bone marrow (BM) source) from mother as the donor; experienced veno-occlusive disease (VOD) and had CMV and late EBV reactivation; all of which were successfully treated and chimerisms maintained 99% donor (1.1 years post-transplant) with low dose sirolimus. Second patient - 19 month old Hispanic male received fully matched (8/8 alleles) stem cells (BM source) from sister; did require three donor lymphocyte infusions (DLI) due to dropping T cell chimerisms, last reported 43% donor (1.8 years post-transplant). Median neutrophil engraftment was 24 days (+/-9.9 days). With a median follow up of 525 days (1.4 years), the 1-year OS for transplanted patients was 100% with minimal GVHD, localized to the skin and Stage 1 Grade 1, requiring topical steroid creams. CONCLUSION: Although the current sample size is small, we describe patients that underwent RIC with PTCy and continue to do well clinically, without HLH or EBV recurrence or significant adverse effects from transplant over 1.4 years post-transplant.

Poster #13

REAL-WORLD TREATMENT PATTERNS AND OUTCOMES IN PATIENTS WITH HEMOPHAGOCYTIC LYMPHOHISTIOCYTOSIS (HLH) AND OTHER CLINICAL CONDITIONS TREATED WITH EMAPALUMAB: THE REAL-HLH STUDY DESIGN

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PURPOSE: Hemophagocytic lymphohistiocytosis (HLH) is a rare, life-threatening hyperinflammatory syndrome characterized by overactivation of the immune system. Emapalumab was approved by the FDA in 2018 for the treatment of adult and pediatric patients with primary HLH with refractory, recurrent or progressive disease or intolerance with conventional HLH therapy. Since approval, no study has evaluated its use in a larger cohort of patients in the real-world clinical setting. METHODS: This retrospective, non-interventional, observational medical chart review study will include US patients treated with emapalumab in a non-clinical trial setting. The study aims to include ≥100 patients treated with ≥1 dose of emaplaumab between November 20, 2018 and December 31, 2020 (patient identification period). The date the patient initiates treatment with emapalumab within the patient identification period is defined as the index date. The post-index date is defined as the period from the index date through to the study end date (June 30, 2021), end of data availability for the patient, or date of death, whichever occurs first. Patients will be classified into three groups (primary HLH, secondary HLH, or non-HLH) based on the information obtained from their charts, the HLH 2004 diagnostic criteria, and adjudication by the Steering Committee. RESULTS: The primary objective of the study is to describe treatment patterns in patients with HLH treated with emapalumab in a real-world clinical setting, including emapalumab dose, treatment duration, and reasons for initiating or discontinuing treatment. The secondary objectives are to describe demographic and clinical characteristics of patients with HLH treated with emapalumab, and their outcomes. The exploratory objectives include detailing the demographic, clinical characteristics, treatment patterns and outcomes of patients with non-HLH clinical conditions treated with emapalumab. CONCLUSION: The study aims to assess treatment patterns and outcomes, clinical and demographic characteristics among patients treated with emapalumab in real-world clinical settings.

Poster #14

ANTI-PD-1 ANTIBODY AND LENALIDOMIDE IN CHRONIC ACTIVE EB VIRUS INFECTION

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PURPOSE: Chronic active Epstein-Barr virus infection (CAEBV) is a disease with poor outcome. Except for allogeneic hematopoietic stem cell transplantation, almost all current therapies are ineffective. Long-term elimination of EBV cannot be achieved. New treatment strategy is urgently needed. METHODS: We investigated the efficacy of anti-PD-1 antibody, combined with lenalidomide, in an open-label, single-center, prospective study involving CAEBV patients. Patients received regimens every 2 weeks for at least 6 cycles, until they reached indications for discontinuation. RESULTS: As of Nov 15, 2020, 32 patients had received anti PD-1+lenalidomide regimen. As of the Feb 1, 2021 analysis cutoff date, 24 cases of them completed at least 3 courses of treatment and were included in analysis. The overall response rate is 54.2% (13/24, 45.8% complete response; 8.3% partial response). EBV-DNA copies in PBMC decreased significantly (p=0.002) and the proportion of CD8+T cells in lymphocytes increased (p=0.007) with the course of treatment. With a median follow-up time of 17.8 months, 22 of 24 patients were alive with an estimated probability of survival of 91.3% at 1 year. All CR patients didn't suffer any relapse. The comparative analysis between response group and non-response group showed the proportion of EM CD8+ T cells in the Response-group increased significantly after treatment, while the NR group had a significantly higher copy number variation load before treatment. CONCLUSION: Anti-PD-1 antibody combined with lenalidomide was an effective and safe therapy for CAEBV patients. The significant therapeutic effect and the different characteristics between response and non-response group, provides a new conception for the possible pathogenesis of CAEBV.

Poster #15

EARLY CHANGE IN SOLUBLE INTERLEUKIN-2 RECEPTOR LEVEL DURING STANDARD ETOPOSIDE/DEXAMETHASONE HLH TREATMENT IS PREDICTIVE OF MORTALITY

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PURPOSE: Standardized treatment with etoposide and dexamethasone in patients with hemophagocytic lymphohistiocytosis (HLH) has improved survival, but approximately 15% of patients die in the first months after diagnosis and salvage therapy is often needed for patients not adequately responding. Identifying these patients promptly is likely to improve outcomes. This study aims to define early treatment response markers which are predictive of outcomes to better identify refractory HLH. METHODS: We conducted a multi-institutional, retrospective study of patients treated for HLH from 2008-2019. Patients treated with protocols other than HLH-1994 or HLH-2004, HLH secondary to an undiagnosed malignancy, or with insufficient data were excluded. Biweekly data of laboratory markers were obtained during the first 100 days of treatment. The endpoint was survival to bone marrow transplant (BMT) or to approximately 1-year if no BMT was pursued. Classification and regression tree analysis and receiver operating curves were used to identify optimal prognostic indicators. RESULTS: Eighty-one patients from three institutions met criteria for inclusion. Median age was 21 months (range 0-282). Pre-BMT mortality was 14.8% (n=12) and overall mortality (including up to day 180 post BMT) was 24.7% (n=20). Improvement in soluble interleukin-2 receptor (sIL2r; sCD25) was the earliest and most consistent predictor of outcome. A ratio of sIL2r from the peak pre-treatment level >0.83 at day 7 (sensitivity 80%, specificity 100%, AUC 0.90) and >0.51 at day 14 (sensitivity 80%, specificity 89%, AUC 0.81) were associated with death prior to BMT. Early change in ferritin was not indicative of prognosis (day 7 ferritin ratio >1.03, sensitivity 33%, specificity 80%, AUC 0.53), but had better performance than most other parameters at later time points. CONCLUSION: Lack of improvement in sIL2r during the first two weeks of standard HLH therapy is associated with pre-BMT mortality and may indicate the need for earlier escalation in therapy.

Poster #16

ROLE OF SERUM CYTOKINES IN THE DIAGNOSIS AND TREATMENT OF PEDIATRIC HEMOPHAGOCYTIC LYMPHOHISTIOCYTOSIS

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PURPOSE: Our previous studies indicated that IFN-γ and IL-10 are significantly elevated in pediatric hemophagocytic lymphohistiocytosis (HLH) and is helpful for the early and accurate diagnosis of HLH. The PURPOSE of this retrospective study is to investigate the role of the cytokines in differentiating the subtypes of HLH and stratifying the patients into different risks. METHODS: From 2010 through 2020, 256 patients with newly diagnosed HLH were enrolled. IL-6, IL-10 and IFN-γ were quantitatively measured at admission. HLH-related variants were determined in 158 patients by sanger sequencing or whole exome sequencing. Patients were divided into four groups according to different treatment regimens. RESULTS: Thirty-three patients were diagnosed with primary HLH (pHLH), 178 with EBV-HLH and 47 with other causes. UNC13D, LYST, STXBP2 and PRF1were the most frequently affected genes. IL-6 and IL-10 levels were comparable between primary and secondary HLH while IFN-γ was much lower in primary HLH. Compared with patients with familial HLH (FHL), those with X-linked lymphoproliferative (XLP) presented only slightly elevated IL-10 and IFN-γ. The IFN-γ level and the ratio of IL-10 to IFN-γ could be a helpful marker to differentiate FHL/XLP from EBV-HLH. The probability of 5-year overall survival (OS) of the whole cohort was 70.1±2.9%, which were 77.2±3.2% and 41.5±10.2% for patients with EBV-HLH and pHLH. The 5-year OS were 80.9±4.8%, 69.6±3.8% for patients treated with dexamethasone (DXM) and HLH-94/04, with much lower IL-10 and IFN-γ levels in DXM group. Patients with low IL-10 (<190pg/mL) presented superior outcome, while those with

IFN-γ≥1150pg/mL had the worst survival rate. CONCLUSION: EBV-HLH, FHL and XLP presented different cytokine patterns. Patients with low IL-10 and IFN-γ levels presented superior outcome. DXM could be a first line choice for EBV-HLH patients with low IL-10 and IFN-γ levels.

Poster #17

HLA B 15:01 AND HLA DQB1 06:02 IS ASSOCIATED WITH EBV STATUS IN CHINESE

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PURPOSE: Although most adults are infected Epstein-Barr virus (EBV), human leukocyte antigen (HLA) complex, may influence the pathogenic infection pathway and therefore may affect EBV-associated disease. Several studies have reported HLA class I alleles with protective or susceptibility associations. Several studies have reported an association of HLA class I alleles with protective or susceptibility, however, the RESULTS are conflicting and such studies are still not available in China. METHODS: In this study, 269 patients with EBV associated disease and 213 EBV seronegative hematopoietic stem cell donors were analyzed, among whom the HLA alleles were sequenced using a high-resolution genotyping method. RESULTS: Our results showed that individuals carrying the HLA-DQB1 06:02 allele had a reduced risk of EBV-related disease, OR (Odds ratio) =0.5699, 95% confidence interval (95% CI) (0.3486-0.9317) (p<0.05), while individuals carrying the HLA-B 15:01 allele individual risk was increased (OR=1.763,95% CI) (0.3486-0.9317) (p<0.05). The B15:01 group was also found to be at much higher risk in patients with T-cell, NK-cell, and multicellular EBV infection than in patients with other genotypic subgroups, suggesting a worse prognosis. CONCLUSIONS: The rate of EBV infection was much higher in the B 15:01 group than in other genotypic subgroups. These findings emphasis the importance of a single locus HLA influencing EBV infectivity. B 15:01 is more prone to non-B-cell infections and should be examined for the dominant type of EBV-infected lymphocytes.

Poster #18

PLASMA D-DIMER AS A POTENTIAL BIOMARKER FOR THE PROGRESSION OF HLH

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PURPOSE: Although hypofibrinogenemia is a feature of hemophagocytic lymph histiocytosis, there are no data about D-dimer as a biomarker to identify prognosis of hemophagocytic lymphohistiocytosis. We hypothesized that increase levels of D-dimer are associated with poor outcomes. METHODS: The study population consisted of a consecutive cohort of 500 patients with HLH diagnosed by Beijing Friendship Hospital from January 2015 to December 2017 were analyzed retrospectively. The survival time was counted until1 December 2020. RESULTS: 500 patients were diagnosed with HLH, the median age was 32years (range, 1-79 years), and 264 (52.8%) were male. The underlying cause of HLH was infectious (n = 248, 49.6%), followed by malignancies (n = 92, 18.4%), unidentified causes (n = 79, 15.8%), and autoimmune disorders (n = 47, 9.4%). After a median follow-up of 26 months, 290 patients (58%) had died. The median overall survival of the entire cohort was 17 months. Patients with increased levels of D-dimer (D-dimer >1.5 μg/mL) were more likely to have a poor prognosis (10 months vs 37 months; p < 0.001), mortality (65.22% vs 48.17%, p < 0.001). Multivariate analysis of COX model showed increased levels of D-dimer was independent factors for OS (Overall Survival). CONCLUSION: HLH is a disease of poor prognosis and high mortality. In this large series of patients' diagnosis with HLH from a single center, patients with increased levels of D-dimer had a markedly worse survival. Increased levels of D-dimer had prognostic value in HLH and was an independent prognostic factor for HLH.

Poster #19

THE DIAGNOSTIC SIGNIFICANCE OF MULTIPLE CYTOKINES IN ADULT HAEMOPHAGOCYTIC SYNDROME

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PURPOSE: Hemophagocytic lymphohistiocytosis (HLH) is a group of immune disorders resulting in uncontrolled activation of T lymphocytes and macrophages with life-threatening cytokine storms. The gold standard for the diagnosis of hemophagocytic syndrome is the HLH-2004 diagnostic criteria, but there are still some diagnostic challenges and therefore there is a need to explore the diagnostic value of multiple cytokines for the diagnosis of hemophagocytic syndrome. Study design: In this retrospective study patients attending Beijing Friendship Hospital of Capital Medical University from January 2016 to December 2020 were included, of which 166 patients with confirmed HLH and 142 febrile patients requiring differential diagnosis completed the sum of multiplex cytokine assays, applying multifactor liquid phase microarray technology- The Luminex analytical platform system detects 30 cytokines (MIP-1α, SDF-1α, IL-27, IL-1β, IL-2, IL-4, IP-10, IL-6, IL-7, IL-8, IL-10, Eotaxin, IL-12p70, IL-13, IL-17A, IL-31, IL-1 RA (RANTES, IFNγ, GM-CSF, TNFα, MIP-1β, IFNα, MCP-1, GRO-α, IL-1 α, IL-15, IL-18, IL-21, IL-22). RESULTS: IL-1 RA, IL-18, IFNγ, IP-10, RANTES, Eotaxin, GRO-α and MIP-1α were higher in the HLH group than in the non-HLH group, and the differences were statistically significant. Among them, the area under the curve of cytokine IL-18 for HLH diagnosis was reported for the first time as 82.69%, sensitivity 76.32% and specificity 79.61%, the AUC of IL-1 RA was 72.34%, sensitivity 62.71% and specificity 75.97%, the AUC of IP-10 was 71.73%, sensitivity 60.14% and specificity 75.15%, while the AUC of the combined diagnostic test for IL-1 RA, IL-18, IFNγ, IP-10 and RANTES was 99.6%, with a sensitivity of 95.8% and a specificity of 98.6%.

Poster #20

SUCCESSFUL TREATMENT OF CNS-HLH REACTIVATION AFTER BMT

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PURPOSE: to report a case of successful treatment with donor lymphocyte infusions (DLI) and etoposide/tocilizumab/dexamethasone combination of patient with hemophagocytic lymphohistiocytosis and central nervous system involvement (CNS-HLH) reactivation in the early post bone marrow transplantation (BMT) period. METHODS and RESULTS: A 13-year-old girl presented with febrile fever, vomiting, depressed level of consciousness and drowsiness. MRI and the examination of cerebrospinal fluid revealed encephalitis without evidence of infection. Therefore, she received therapy with 25 mg/kg/d methylprednisone (4 days) with a rapid and complete resolution of neurological symptoms. Later the patient fulfilled the HLH diagnostic criteria. According to the data of genetic analysis, compound heterozygous mutations in the 14th and 24th exons of the UNC13D gene have been identified. The patient received treatment according to the HLH-94 protocol with subsequent BMT from the matched sibling donor. The early post-BMT period proceeded without significant infectious and immunological complications. Basic immunosuppression included Cyclosporin A and Mycophenolate mofetil. The engraftment was registered on day +18 after BMT. Donor Chimerism (DC) on day +30 after BMT was mixed (92% of donor cells). On day +62 the patient was presented with fever, vomiting, neurological symptoms and full HLH criteria without any evidence of infection. The DC was only 49.5% of donor cells. We performed treatment with tocilizumab 8 mg/kg, etoposide 150 mg/m^2 twice and dexamethasone 10 mg/m^2/d for 14 days. Afterwards she received 4 DLI with CD3+ cell dose escalated from 0.5 to 4x10^5/kg. 100% donor chimerism has been achieved by 100 days after BMT. No signs of disease have been observed for 280 days after BMT. CONCLUSIONS: DLI can be a curative and safe treatment option in patients with reactivation of the CNS-HLH in the early post-BMT period with the background of mixed donor chimerism.

Poster #21

NOVEL MUTATION IN SLC29A3 GENE AND CONCURRENT MUTATION IN BRCA1 GENE IN A CHILD WITH ROSAI-DORFMAN DISEASE

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PURPOSE: to describe Rosai-Dorman disease (RDD) patient with novel mutation in SLC29A gene with concurrent mutation in BRCA1 gene. METHODS: The 5-year old child from Azerbaijani consanguineous family was initially admitted to the hospital in 2019 with complaints of ear's pain and maxillary lymph node (LN) enlargement. Familial history included multiple cases of lymphadenopathies, ovarian and breast cancers. Antibacterial therapy was without response. Further LN enlargement was observed. A biopsy of LN was performed. Morphological signs of sinus histiocytosis with massive lymphadenopathy (RDD) were found in the samples. Initial treatment was performed in Azerbaijan. Course of prednisone was without significant response. Interferon-alpha was administered for 4 weeks with partial response. The further course of disease was recurrent. Patient consistently was treated with interferon-alpha, 6-mercaptopurine, methotrexate and 2-chlordeoxyadenosine with partial result in all episodes of LN enlargement. Next generation sequencing (NGS) examination of the patient revealed two mutations: SLC29A3 and BRCA1. RESULTS: SLC29A3 is typical for Faisalabad histiocytosis the hereditary form of RDD, associated with presence H-symptoms (hepatomegaly, cutaneous hyperpigmentation and hypertrichosis, hearing loss, heart anomalies and hypogonadism). A homozygous mutation in SLC29A3 (c.303_320dupCTACTTT GAGAGCTACCT) identified in our case has not been described in literature before. In our case only RDD signs were present without H-symptoms. Heterozygous BRCA1 mutation (c.1059G>A) is known to be pathogenic and described in numerous cases of patients with ovarian and breast cancers. CONCLUSION: Combination of these two mutations, recurrent character of the disease, lack of significant response in all previous lines of therapy, makes this case challenging in terms of treatment approaches. The possible impact of pathogenic BRCA1 mutation to the course of disease also remains unknown. The patient requires continued follow-up.

Poster #22

18F-FDG PET/CT FOR IDENTIFYING THE POTENTIAL CAUSES AND PROGNOSIS OF SECONDARY HEMOPHAGOCYTIC LYMPHOHISTIO-CYTOSIS IN CHILDREN

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OBJECTIVE: To investigate the value of 18F-FDG PET/CT metabolic parameters in etiological differentiation and prognosis prediction of secondary hemophagocytic lymphohistiocytosis (sHLH) in children. METHODS: Sixty-six newly diagnosed sHLH children, who underwent 18F-FDG PET/CT examination from January 2018 to December 2020 were retrospectively analysed. Except for 4 cases with unknown etiology, patients were divided into malignant disease group (n = 13) and non-malignant disease group (n = 49). Data relating to the clinical manifestations, metabolic parameters of liver, spleen, bone marrow and lymph nodes and their ratios to hepatic blood pool and mediastinal blood pool were extracted from medical records. RESULTS: The SUVmax of lymph node, spleen, and liver and SUVmean of spleen in malignant disease group were significantly higher than those in non-malignant disease group (P=0.031, 0.035, 0.016 and 0.032). Malignant disease should be considered when lymph node SUVmax was higher than 4.41 (sensitivity 61.5%, specificity 81.6%). Focal involvement of extranodal organs was more likely to occur in malignant disease group than in non-malignant disease group (P=0.011). IFN- γ was positively correlated with SUVmax and SUVmean in bone marrow, liver, and spleen. Ferritin, sCD25, IL-6 and IL-10 were positively correlated with spleen SUVmax and SUVmean. In EBV-HLH, high level of

bone marrow SUVmax, SUVmean and the ratio to mediastinal blood pool and hepatic blood pool were correlated with poor 2-week treatment response, overall survival rate and event-free survival rate. CONCLUSIONS: Some metabolic parameters of 18F-FDG PET/CT are helpful to identify the etiology of sHLH in children. Malignant disease should be considered when the SUVmax of lymph nodes is higher than 4.41 and focal involvement of extranodal organs occurs. In EBV-HLH, the high SUV value of bone marrow is associated with poor prognosis.

Poster #23

HAPLOIDENTICAL HEMATOPOIETIC STEM CELL TRANSPLANTATION FOR PEDIATRIC PATIENTS WITH CHRONIC ACTIVE EPSTEIN-BARR VIRUS INFECTION: A RETROSPECTIVE ANALYSIS OF A SINGLE CENTER

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OBJECTIVE: To evaluate the feasibility and clinical effect of haplo-HSCT for the treatment of pediatric patients with CAEBV. METHODS: Children with CAEBV who did not have matched donors and underwent haplo-HSCT in Beijing Children's Hospital, Capital Medical University, from October 2016 to June 2020 were retrospectively analyzed. RESULTS: Twenty-five patients, including 16 males and 9 females, with an onset age of 5.0±2.6 years and a transplantation age of 6.9±2.9 years, were enrolled in this study. The mean observation time was 19.0±12.0 months. Three patients received the Reduced Intensity Conditioning regimen, and the remaining patients all received the modified Myeloablative Conditioning regimen. By the end of follow-up, 23 patients were characterized by disease-free survival (DFS), 22 patients were characterized by event-free survival (EFS), and 2 patients died. One of the patients died of TMA, and another died of GVHD; this patient discontinued the treatment for economic reasons. The 3-year overall survival (OS) rate was estimated to be 92.0% ±5.4%, and the 3-year EFS rate was estimated to be 87.4%±6.8%. All active patients survived after HSCT event-free. Acute GVHD degrees 1-3 were observed in 10 patients (40.0%), and degree IV was observed in 6 patients (24.0%), who were all cured except for one patient. Chronic GVHD was observed in 9 patients (36.0%), and most of the cases were mild. The incidence of TMA was 28.0%, and the incidence of VOD was 4.0%. CONCLUSION: Haploidentical hematopoietic stem cell transplantation is safe and effective in the treatment of pediatric CAEBV and can be used as an alternative therapy without matched donors or emergency transplantation. Patients with active disease before HSCT also benefited from haplo-HSCT. Haplo-HSCT requires careful monitoring for complications such as GVHD and TMA. Early detection of TMA and timely treatment can reduce mortality and improve the survival rate.

Poster #24

A GRANULOMA-LIKE PROLIFERATION OF S100P-NEGATIVE INDETERMINATE CELLS.

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PURPOSE: To describe an exceptional case of indeterminate cells proliferation. METHODS: A 70-year-old man presented with erythematous papules and plaques, slowly progressing over his trunk. Clinical record includes a history of colorectal cancer and bilinear cytopenia with persistent monocytosis, awaiting hematologic diagnosis. RESULTS: Skin biopsy revealed a dermal, non epidermotropic, multinodular proliferation of CD1a+, CD207/langerin-, S100p-, CD163-, medium-sized mononuclear, cells, with abundant, slightly eosinophilic cytoplasm and large, occasionally irregular nuclei, with prominent nucleoli. These cells were organized in large granuloma -like structures with a necrotic center (coagulative type). A polymorphous, inflammatory infiltrate surrounds the proliferation, including hyperplastic lymphoid follicle snd reactive phagocytes. Collectively, the morpho-phenotypic features were indicative of an indeterminate cell histiocytosis/tumor (iCH). However, the unusual, multinodular architecture, and with the low proliferative index (Ki67: 5%), lead us to consider also a reactive proliferation of indeterminate cells. CONCLUSION: With around 100 reported cases, iCH is one of the rarest histiocytosis. Due to its morphological and phenotypical similarities with Langerhans cell histiocytosis (LCH), it is currently included in the L-group of histiocytoses. Despite 90% of cases involve the skin, any organ may be involved. Up to 20% of iCH cases associates with hematological neoplasms. Moreover, Vitte et.al. described a specific type of leukemia cutis with iCH features. Most iCH cases feature a CD1a+/S100p+ phenotype, but roughly 10% of them lacks S100p. As with all other histiocytoses, typical iCH features diffuse infiltrates. However, our case, displayed a well-defined, multinodular architecture with central necrosis, resembling granulomas. Tardio et.al. described the only other report of cutaneous S100p- iCH showing a multinodular architecture and central necrosis, in a patient with unremarkable clinical and laboratory findings. This case experienced a self-regressing course while our patient's records, seems to point toward an emerging hematological dyscrasia/neoplasia. Further follow-up is needed due to reveal the clinical significance of such proliferation.

Poster #25

JUVENILE XANTHOGRANULOMATOSES - EXPERIENCES IN THE GERMAN RARE HISTIOCYTOSES REGISTRY AND CONSULTATION CASES

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BACKGROUND: Rare or Non-Langerhans cell histiocytoses (non-LCH) include a number of different diseases. In pediatrics, besides Rosai-Dorfman disease (RDD) (juvenile) xanthogranulomatosis (JXG) is most frequent. METHODS: In 2012, the German registry and consultation study for non-LCH - part of the International Rare Histiocytic Disease Registry - was initiated. RESULTS: 28 JXG patients were reported, all are survivors (3 months to 5 years follow-up); 10/18 female/male; 7 were neonates, 4 older infants, 7 were 1y old, 7 were 2-10 y old, and 3 >10y). 9 patients had localized skin or skin plus subcutaneous involvement which could be resected completely, with no relapse (3 neonates); 4 patients had multiple skin lesions (two were still completely resected, two observed, with one regredient, one slowly progressive). 5 neonatal as well as older patients had inoperable local organ involvement (eye, mediastinal masses, intracerebral); 9 neonatal as well as older pts. had multiple organ involvement (combinations of skin, peritoneum, liver, lung, intracerebral). In two, a mixed or overlap histiocytosis was discussed. Genetic analysis was performed in 6 cases; showing 1 KRAS, 1 BRAF, and 1 ALK mutation (all systemic cases). Those patients who had multiple organ involvement, received nonsurgical therapy if progressive (all patients > 3 years), consisting of steroid therapy in one case, and LCH-type polychemotherapy in 4. In 3 of these, there was insufficient response, and a molecular alteration was identified, so they received targeted therapy, and all three responded. CONCLUSION: JXG represents a very heterogeneous disease. In localized cases, resection and observation are appropriate, in particular in neonates. Older children occasionally have multisystem disease, which may show insufficient response to steroids or LCH-type therapy. Then, molecular analyses are crucial in order to enable targeted therapies - which may induce response. Appropriate consultation depends on international prospective data collection, which should be further propagated.

Poster #26

SHAPIRO'S JUVENILE XANTHOGRANULOMA PRESENTING AS CLUSTERED PAPULONODULAR LESIONS IN AN INFANT

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Non-Langerhans cell histiocytosis (n-LCH) is a concerted term for a class of benign proliferative disorders of histiocytes, macrophages and dendritic cells which do not fulfil the criteria for Langerhans cell histiocytosis. Juvenile xanthogranuloma (JXG) is the most frequent form of n-LCH and is characterised by solitary or a few papulonodular lesions in infants and children with preponderance in males. It customarily manifests in a generalised cutaneous form while the eyes are the commonest extra cutaneous involved organ. When multiple it may be associated with neurofibromatosis-1, Neimann -Pick disease and juvenile myelomonocytic leukaemia. keratotic, lichenoid, flat plaques, giant, agminated, granuloma annulare like and subcutaneous or destructive forms are rarer morphological variants. Herein we describe a case of a 6-month-old girl child born to a nonconsanguineous couple who presented with four months' duration of multiple, discrete, soft to firm, non-tender, yellowish red, papulonodules clustered over the anterolateral aspect of the chest with a few of them having superficial ulceration and crusting. She had no systemic symptoms and her general physical examination and rest of the mucocutaneous examination were unremarkable with no palpable lymph nodes or organomegaly. Dermoscopy revealed yellowish structureless areas surrounded by a rim of erythema and telangiectasia. On histopathological examination diffuse dermal infiltrate extending into subcutaneous tissue was seen. On higher magnification infiltrate was composed of elongated to epithelioid cells with vesicular chromatin and eosinophilic cytoplasm arranged in sheets and fascicles with occasional cells showing cytoplasmic vacuolization suggestive of Shapiro's JXG. Occasional Touton giant cells were identified. Immunohistochemistry revealed CD68, CD163 positivity of cells while CD1a, S-100, Langerin and ALK-1 stain were negative. The unusual clustered distribution of lesions of Shapiro JXG adds to the peculiarity of our case which clinicians should be well aware of to differentiate it from the surfeit of dermatoses which could have similar manifestations.

Poster #27

KIDNEY INVOLVEMENT IN ERDHEIM-CHESTER DISEASE: A MULTICENTER COHORT STUDY

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PURPOSE: Little is known about kidney function and prognosis in Erdheim-Chester disease (ECD). We investigated kidney involvement and outcome in a large ECD cohort. METHODS: Consecutive patients with biopsy-proven ECD followed at referral centers in Italy, France, and Israel between 2005-2020 were included. Data on kidney function, imaging studies, and treatment history were collected at diagnosis, at one, two, five years, and last visit. RESULTS: We included 195 patients, 72% of whom men (mean age at diagnosis 56.5+/-14.3y). Perirenal involvement was found in 142 patients (73%). Of them, 38% had hydronephrosis, 8% kidney atrophy, 42% ureteral, 31% vascular peduncle, and 19% adrenal gland involvement. Perirenal involvement was associated with older age (p=0.001), male sex (p=0.004), hypertension (p=0.001), large vessel (p<0.001) and heart involvement (p<0.001), and BRAFV600E mutation (p=0.008). Baseline kidney function was worse in patients with perirenal infiltration than in those without (median eGFR 75 vs 98mL/min/1.73m2, p<0.001). At follow-up median eGFR remained lower in patients with perirenal involvement (p=0.001). The eGFR variation at last visit was similar between the two groups (p=0.8). Patients receiving BRAF-/mTOR-inhibitors had more frequently radiologic (p=0.02) and metabolic (p=0.001) improvement of perirenal infiltration, compared with patients receiving conventional therapies (e.g., interferon-alpha). There was no difference in mortality or end-stage kidney disease (ESKD) between patients with and without perirenal involvement (log-rank test, p=0.177). Unadjusted predictors of both chronic kidney disease (CKD) 4-5/eGFR decrease >25% and ESKD/death at last visit were age at onset >50y, hypertension, BRAFV600E

mutation, and low eGFR at baseline. At multivariate analysis, cardiovascular risk factors were associated with CKD 4-5 or eGFR decrease>25% (p=0.005); age>50y was associated to ESKD/death (p=0.005). Conventional therapies had a protective effect on ESKD/death (p=0.029). CONCLUSIONS: Perirenal infiltration in ECD is associated with worse renal function at diagnosis. Cardiovascular risk factors and age are independent predictors of kidney outcome.

Poster #28

MULTIPLE EXTRANODAL ROSAI DORFMAN DISEASE IN AN ADOLESCENT WITH A CUTANEOUS LEOPARD PHENOTYPE AND GERMLINE MAP2K2 MUTATION

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PURPOSE: To present an atypical case of Rosai Dorman Disease (RDD) in an adolescent with a constitutional defect in RAS-MAPK pathway; and to analyse its possible prognostic and therapeutic implications. METHODS: We analyse the case of a multiple extranodal RDD in an adolescent with a cutaneous LEOPARD phenotype due to MAP2K2 germline mutation. RESULTS: A 16 years old male, with history of moderate mental retardation, intestinal invagination in infancy, and multiple lentigins, presented with exophthalmos and diplopia. Magnetic Resonance of the brain and orbit showed an orbital mass of 20 x 7 mm in the left orbit roof, ocular proptosis, congestion of paranasal mucosa, and an intracranial, extra-axial frontal/ temporal brain tumour of 20 mm, compatible with meningioma. An almost complete resection of the orbital tumour was performed in two times and histology was compatible with RDD. Because of the multiple lentigins associated with this rare tumour, we performed a genetic sequencing panel for rasopathies and a MAP2K2 de novo heterocygous mutation was identified c.692G>T (Arg231Leu). The patient has stable disease and received no further treatment to date. In case of progression a MEK inhibitor should be considered. CONCLUSION: Rosai Dorfman disease is a rare inflammatory/ oncologic condition. The therapeutic decisions depend on the extent of the nodal or extranodal disease and symptoms. Recent studies show an important role of the RAS MAPK pathway in the physiology of this disease which allows a therapeutic option with MEK inhibitors and may confer a prognostic significance like in other disease associated with alterations is this pathway, Juvenile Mielo-Monocitic Leukemia (JMML). A better understanding of RDD pathophysiology and RAS MAPK alterations certainly will allow better therapeutic approaches in patients affected by this disease.

Poster #29

ROSAI-DORFMAN DISEASE - A SINGLE TERTIARY CARE CENTRE EXPERIENCE OF A UNIFORM PROTOCOL BASED APPROACH

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PURPOSE: Rosai- Dorfman Disease (RDD), is a rare histiocytic disorder, with natural course ranging from spontaneously resolving lymphadenopathy to life threatening emergencies. This heterogenous nature poses many difficulties in diagnosis and management. Our objective was to report the clinical features and treatment outcomes of children with RDD presented to our centre. METHOD: In this retrospective study, the clinical details were collected from files of children diagnosed with RDD and on follow up with Pediatric Oncology Clinic, All India Institute of Medical Sciences, New Delhi from January 2012 to December 2020. RESULTS: Seven children were diagnosed with RDD during the study period with median follow up of 54 months. Median age of the cohort was 5 years, with male preponderance. All seven children had cervical lymph nodal involvement. Extranodal involvement was seen in one child who had cutaneous and osseous disease in addition to hepatomegaly in two children and hepatosplenomegaly in one child. Diagnosis could be established by fine needle aspiration cytology alone in 6 children. Positron Emission Tomography (PET-CT) was done at baseline and for response assessment. One child with uncomplicated lymphadenopathy was treated with observation alone. Remaining six children were started on corticosteroids with median duration of 5 months. Two children attained remission with corticosteroids alone, of which one child relapsed after 3 years. Due to toxicity and poor response, 4 out of 6 children were shifted to second line therapy with 6-Mercaptopurine (6MP) and Methotrexate, which they received for a median of 13 months. None had relapse during the follow up. CONCLUSION: RDD is one of the rare diseases to be considered in the differential diagnosis of significant cervical lymphadenopathy. Diagnosis could be established by FNAC alone. PET CT emerged to be useful for response assessment. 6MP and Methotrexate combination is safe and effective alternative option to corticosteroids.

Poster #30

CLOFARABINE AS MONOTHERAPY AND SALVAGE THERAPY IN HISTIOCYTIC SARCOMA

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PURPOSE: Describe two pediatric patients treated with Clofarabine for Histiocytic sarcoma (HS). METHOD: Descriptive retrospective case series. RESULTS: A 13 y/o female presented with an enlarging, painless lump on the posterior right thigh for three weeks. MRI of the right leg showed a 2.8 cm mass arising from the semimembranous muscle. A CT chest, PET-CT, bone marrow biopsy, MRI brain/spine and LP were negative. Biopsy showed discohesive sheets of large atypical epithelioid cells with abundant eosinophilic cytoplasm negative for pan-keratin, EMA, CD34 and desmin with normal INI-1

with positivity for CD163, CD68, and LCH and CD4. Sequencing identified DOC8-BRAF and AXON1-P314L fusions. Following complete resection, she was started on Clofarabine monotherapy. Patient is in remission with no clinical or radiographic evidence of disease. Our second patient is a 15 y/o male who presented with supraclavicular and axillary adenopathy. Biopsy showed large atypical cells with spindled and epithelioid morphology positive for CD4, CD56, lysozyme, CD163, CD43 and vimentin with variable CD45, S100 and CD68 consistent with metastatic histiocytic sarcoma. Marrow and CSF studies were negative. He was treated with a modified version of the T-cell leukemia protocol AALL0434 with complete clinical resolution at the end of induction. Four weeks into consolidation, PET-CT confirmed avidity at the primary site with new lesions consistent with refractory disease. Sequencing revealed mutations in NF-1, FBXW7, and CBL. While on salvage therapy of Clofarabine and Tremitinib, he had an aggressive recurrence. A third outside opinion favored a myeloid sarcoma (MS) over HS and patient was switched to an AML-based regimen. Unfortunately, patient died of his disease. Although diagnosis remains unclear, patient did show initial response to Clofarabine. CONCLUSION: HS is a rare, highly aggressive cancer. Clofarabine has been used as a monotherapy and in combination with MAPK pathway inhibition and salvage therapy to treat HS and MS.

Poster #31

BENIGN CEPHALIC HISTIOCYTOSIS - DERMOSCOPY

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BACKGROUND: Benign cephalic histiocytosis is a rare self-limiting, non-Langerhans cell histiocytosis of infants and young children. The dermoscopic features of benign cephalic histiocytosis have hitherto been undescribed in the literature. AIM: To describe the dermoscopic features of benign cephalic histiocytosis. METHOD: An 8-month infant presented with a two-month history of an asymptomatic protuberant nodule over the right cheek. On examination, a well-defined, 2x2x3 cm solitary skin-coloured, non-tender, firm, nodule was present over the right mandibular region with areas of yellowish discoloration and telangiectasia present over the summit of the nodule, and overlying scaling. Histopathological examination revealed acanthotic epidermis and collection of histiocytes in reticular dermis in sheets and nodules, admixed with few scattered foamy macrophages and multinucleated giant cells, neutrophils and numerous eosinophils. The histiocytes showed round to oval nuclei with bland chromatin, small conspicuous nucleoli and moderate amount of cytoplasm. On immunohistochemistry, these cells were positive for CD68, S100 and CD163; and were negative for CD1a, Langerin and BRAF. Dermoscopy of the lesion showed yellowish-brown orange areas, linear and curved telangiectatic vessels, a peripheral rim of scales with an upturned inner edge, and red structureless areas. Dermatoscopic evaluation showing yellowish orange areas represent dermal accumulation of histiocytes, red structureless areas were due to epidermal hemorrhage and superficial telangiectatic vessels represent dilated blood vessels on histopathology. CONCLUSION: Benign cephalic histiocytosis has overlapping clinical and histological features with Juvenile xanthogranuloma and dermoscopy may be helpful in differentiating these two entities.

Poster #32

DERMOSCOPY OF VARIOUS HISTIOCYTOSIS

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PURPOSE: Histiocytosis are rare disorders characterized by the accumulation of macrophages, dendritic cells and monocyte-derived cells in various tissues and organs. Cutaneous presentation in different histiocytosis varies, however, lesions may have overlapping clinical features making differentiation from other dermatosis difficult. Dermoscopy may serve as an non-invasive auxiliary tool in diagnosis of such lesions. METHODS: We report dermoscopic features of eight histopathologically and immunohistochemistry proven cases of histiocytosis, papular xanthoma [n=3], xanthoma disseminatum (XD) [n=2], and Langerhans cell histiocytosis (LCH), benign cephalic histiocytosis and juvenile xanthogranuloma (JXG) [n=1 each]. RESULTS: Dermoscopy of papular xanthoma showed central yellowish orange area with preserved pigment network and surrounding rim of erythema. Dermoscopy of early XD revealed yellowish-orange areas surrounded by a rim of erythema and linear vessels while late stages of XD showed white streaks and structureless areas with minimal background erythema. Dermoscopy of LCH showed central white structureless areas and scaling, linear and dotted vessels, surrounded by erythematous ring and telangiectasias, and peripheral brownish hyperpigmentation. Dermoscopy of benign cephalic histiocytosis showed yellowish-brown orange areas, linear and curved telangiectatic vessels, a peripheral rim of scales and red structureless areas, while JXG showed a characteristic "setting sun" appearance with linear and dotted vessels. CONCLUSION: Central orange-yellow structureless area surrounded by an erythematous rim (‘setting sun’ appearance) is a common dermoscopic feature of all xanthogranulomatous disorders. Additional features such as reddish-purple areas and vascular blotches and scaling seen in LCH may suggest specific diagnosis.

Poster #33

NOVEL GENE FUSIONS AND SUSTAINED RESPONSE TO TARGETED THERAPY IN CHILDREN WITH SYSTEMIC JUVENILE XANTHOGRANULOMA

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Juvenile xanthogranuloma (JXG) is a non-Langerhans cell histiocytic disease, most commonly affecting children, manifesting as self-resolving skin lesions. Rarely, JXG can affect vital organs, resulting in significant morbidity. We describe two children with systemic JXG, in whom novel gene-fusions were identified, and targeted therapy led to prompt and sustained response. Patient 1 is a 6-year-old boy who developed widespread rash and diabetes insipidus. Biopsy of the rash revealed features consistent with JXG. Brain MRI demonstrated enhancing lesions in the brainstem, left temporal lobe, and neurohypophysis. Since increased MAP-kinase signaling is the most frequent driver of histiocytoses, we treated him with the MEK-inhibitor trametinib, with

prompt improvement in rash and his overall condition. Targeted sequencing of the biopsy demonstrated a novel GAB2-BRAF fusion; elevated MAP-kinase signaling was confirmed by increased p-ERK staining. Experimental expression of GAB2-BRAF in HEK293 cells demonstrated increased p-MEK and p-ERK expression by western blotting, while expression in BAF3 cells induced cytokine-independent growth. Three years later, he remains well on desmopressin and trametinib with resolution of brain lesions on MRI. Patient 2 is an infant who had thrombocytopenia at birth, which progressed to pancytopenia, and hepatosplenomegaly. Bone marrow biopsy showed histiocytic hyperplasia, but was overall nonspecific. He remained transfusion dependent. Extensive diagnostic investigations failed to identify a diagnosis. By 4 months of age, he developed skin lesions; biopsy revealed features consistent with JXG. Trametinib was initiated, with prompt resolution of pancytopenia, organomegaly, and flattening of the skin lesions. Sequencing of biopsy tissue revealed a novel TFG-RET fusion. Experimental expression of TFG-RET in HEK293 cells demonstrated increased p-ERK expression. At three months since initiation of therapy, the child is asymptomatic and thriving. Novel sequencing methodologies allow for identification of driver mutations and application of targeted therapies that result in rapid and sustained disease control in patients with complex histiocytoses.

Poster #34

DABRAFENIB OR TRAMETINIB ARE EFFECTIVE FIRST-LINE THERAPIES IN PATIENTS WITH LCH AND OTHER HISTIOCYTOSES

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Conventional chemotherapy treatments for Langerhans Cell Histiocytosis (LCH) are fraught with the risk of recurrence after therapy or of progression while on therapy. Since most LCH are driven by activating mutations in the BRAF-MEK-ERK pathway, BRAF- and MEK-inhibitors have been used successfully in some patients suffering relapse. There is limited knowledge on the efficacy of these agents as first-line therapies for LCH or other histiocytic disorders. We present data on 34 patients (26 LCH, 2 Juvenile Xanthogranuloma, 2 Rosai-Dorfman Disease, 4 presumed CNS histiocytosis) treated at a single center with the BRAF inhibitor dabrafenib and/or the MEK inhibitor trametinib. In 28 patients with available tissue specimens, 26 had activating mutations in the BRAF-MEK-ERK pathway. Sixteen patients had received therapy before receiving the inhibitor, the majority of whom had multisystem LCH with risk-organ involvement and progressed on therapy or relapsed. In this group (ages 1.3-31 years), 7 patients received dabrafenib, 7 trametinib, and 2 were treated with both. With a median treatment duration of 3 years, 15 (94%) have sustained favorable responses {12 No Active Disease (NAD) or NAD with residual diabetes insipidus (DI) and/or sclerosing cholangitis, 1 Active Disease Better (ADB)}. One patient with isolated CNS disease had stabilization. Eighteen patients (ages 0.2-45 years), received the inhibitor as first-line treatment, 4 dabrafenib, 14 trametinib. With a median treatment duration of 1.75 years, all have had sustained favorable responses (13 NAD, 2 ADB). Three patients with presumed isolated CNS/pituitary-stalk histiocytosis demonstrated stabilization or improvement of disease. The inhibitors were tolerated well. The most common side effect was skin rash with trametinib that resolved with dose reduction. Two patients with single system LCH discontinued therapy and remain off therapy without recurrence. Our experience demonstrates that patients with histiocytoses can be treated safely, and effectively with dabrafenib or trametinib as first-line therapy.

Poster #35

DRAMATIC EFFICACY OF VEMURAFENIB ON PSYCHIATRIC SYMPTOMS REVEALING BRAF-V600E ERDHEIM-CHESTER DISEASE

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PURPOSE: Psychiatric manifestations in Erdheim-Chester Disease (ECD) are not described in the spectrum of neurological manifestations, particularly as the initial onset of the disease, making the diagnostic challenging. METHODS: We present the first case of neuro-histiocytosis, presenting as a psychiatric delirium with hallucinations, recovering upon targeted therapy with BRAFV600E inhibitor. RESULTS: An 81-year-old Caucasian woman suffered from delirium with hallucination. Her medical history included Grave's disease, deep venous thrombosis, retinal vein occlusion, and macular degeneration. She had auditory hallucinations for 7 months for the current condition, followed by a 2 months history of visual hallucinations and logorrhea. She also had a persecutory delusion and hallucinations without underlying psychiatric pathology. The neurological examination showed a cerebellar syndrome (Scale for the assessment and rating of ataxia (SARA) score: 13/40). The patient also had acute heart failure. The echography showed a pericardial effusion requiring drainage. Brain magnetic resonance imaging (MRI) showed confluent FLAIR hyperintensity lesions in the pons and superior cerebellum peduncles. Lumbar puncture was unremarkable except for elevated neopterin. Body computed tomography showed a "hairy kidney" aspect and an adventitia vessels thickening. The 18-fluorodeoxyglucose PET showed bilateral symmetric osteosclerosis of long bone suggestive of ECD. The diagnostic was confirmed with a perirenal biopsy revealing diffuse infiltration of CD68+, CD1a- histiocytes with BRAFV600E gene mutation on pyrosequencing. The patient received interferon-alpha (180 micrograms/week) twice then specific BRAF inhibitor Vemurafenib. Rapid regression of the psychiatric symptoms and neurological improvement (SARA Score: 12/40) occurred within days of treatment. After 8 weeks, the patient had recovered clinically. The brain MRI showed partial regression of the lesions. 18FDG-PET shows a reduction in radiotracer uptake on all ECD sites compatible with a partial metabolic response. CONCLUSION : This case highlights the first description of delirium as a manifestation of neuro-ECD with dramatic improvement with targeted therapy.

Poster #36

DISTURBANCE OF MONOCYTE HOMEOSTASIS IN HISTIOCYTOSIS IS CLOSE TO CHRONIC MYELOMONOCYTIC LEUKEMIA AND IS CORRELATED WITH PHENOTYPE AND DISEASE ACTIVITY

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PURPOSE: Monocytes have a significant role in histiocytosis pathogenesis. Little is known about their phenotype in histiocytosis and the difference with other myeloproliferative or inflammatory conditions. METHODS: The phenotype of monocytes from patients with histiocytosis was compared to the one of patients with chronic myelocytic monocytic leukemia (CMML), essential thrombocythemia (ET), giant cell arteritis (GCA), and healthy donors (HD). Monocytes were defined as classical (CD14++CD16-), intermediate (CD14+CD16+) and non-classical (CD14+CD16++) by flow cytometry analysis. RESULTS: Seventy-two patients were included (16 histiocytosis, 7 ET, 7 CMML, 21 GCA, and 21 HD). Among histiocytosis, 8 patients had Erdheim-Chester disease, 4 had Langerhans-cell histiocytosis, and 4 had Rosai-Dorfman disease. The frequency of classical monocytes was higher in histiocytosis compared to ET (92% [70-98] vs 76% [54-90]). The frequency of intermediate monocytes was lower in histiocytosis compared to ET (5% [1.5-24] vs 13% [5-42.8]) and GCA(5% [1.5-24] vs7.91% [2.38-69]). The frequencies of classical (87.50% [71-94] vs 97% [96-99]), transitional (5% [4-25] vs 2.5% [0.5-3.5]), and non-classical monocytes (4% [1-11] vs 0.5% [0.25-1.5]) differed between CMML patients and patients with both histiocytosis and clonal hematopoiesis. Monocyte subsets were similar between CMML patients and patients with histiocytosis harboring MAP-kinase pathway gene mutation. Monocyte phenotype did not differ depending on the type of histiocytosis, molecular status, association with myeloid neoplasms/clonal hematopoiesis, or targeted therapy treatment. The frequency of non-classical monocytes was lower in patients with vascular involvement (1% [0.1-2] vs 4% [0.5-11]). Intermediate monocytes were less frequent in responder patients (3% [0.1-8] vs 8% [4-25]). CONCLUSION: The distribution of monocyte subsets is homogenous between the different types of histiocytosis. It's close to CMML, a myeloproliferative disorder involving the MAP-kinase pathway, but different from ET and GCA. Monocyte subset distribution analysis in histiocytosis could be helpful for the diagnosis and be a surrogate marker of disease activity.

Poster #37

MONOCYTE POLARIZATION AS A MARKER OF DISEASE ACTIVITY IN A SEVERE VASCULAR FORM OF ERDHEIM-CHESTER DISEASE

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PURPOSE: Disease activity in Erdheim-Chester Disease (ECD) is assessed with 18fluorodeoxyglucose PET-CT, but no biological marker is validated to evaluate disease activity. METHODS: We have correlated monocyte phenotype and disease activity in a patient with an aggressive vascular form of ECD requiring transplantation. RESULTS: A twenty-five-year-old woman was hospitalized for progressive abdominal pain. She had a medical history of Budd-Chiari syndrome at 14 years old, requiring long-term vitamin-K antagonist therapy. For the current condition, computed tomography showed mesenteric ischemia secondary to the sheathing of the superior mesenteric artery and coeliac trunk. She underwent thrombectomy of superior mesentery artery associated with stenting with successful reimplantation on abdominal aorta associated with heparin-anticoagulation therapy. Despite anticoagulation therapy, she had digestive ischemia requiring partial jejunum surgical removal. Search for hereditary or acquired thrombophilia was negative. Mesentery artery biopsy showed fibrosis in the adventitia with lymphocyte and histiocyte infiltration. Immunostaining showed CD68+, CD1a- histiocytes with strong p-ERK expression. Bone scintigraphy showed radiotracer uptake in the long bones characteristic of ECD. Patient received interleukin-1 receptor antagonist followed by MEK-inhibitor inducing remission on PET-CT. At that time, monocyte phenotype analyses showed 92.9% of "classical" (CD14+CD16-) monocytes, 5.7% of "intermediate" (CD14+ CD16+) and 1.2% of "non-classical" (CD14+lowCD16+) monocytes. Unfortunately, the patient presented a new coeliac trunk thrombosis causing an acute liver failure which required liver transplantation in emergency. MEK-inhibitor was stopped after transplantation. Despite immunosuppressive treatment, patient had a new ECD's flare and died. During this flare, monocyte phenotype analyses showed an increase in "classical" monocytes (97.5%), and a decrease in "non-classical monocytes (1%) with 0.7% of "intermediate" monocytes. CONCLUSION: An increase in "classical" monocytes and a decrease in "non-classical" monocytes have been noticed in patients with active ECD. This case favors the hypothesis that monocyte immunophenotype analysis can be a simple tool to assess disease activity.

Poster #38

A RARE RAB27A VARIANT ASSOCIATED WITH A CASE OF HEMOPHAGOCYTIC LYMPHOHISTIOCYTOSIS ALTERS EFFECTOR PROTEIN BINDING AFFINITIES

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PURPOSE: Here we describe an adult-onset hemophagocytic lymphohistiocytosis (HLH) with a novel homozygous RAB27A c.551G>A (p.R184Q) variant. We sought to assess the contribution of this novel RAB27A variant to melanocyte and lymphocyte function and determine whether it could be disease-causing. METHODS: Hair pigmentation and melanocyte distribution in skin from patient biopsies were studied. In addition, lymphocyte responses were evaluated. The patient-derived Rab27a variant was also examined using reconstitution systems, including both Rab27a-deficient mouse and human cells. In addition, Rab27a interactions were studied by immunoprecipitation. RESULTS: A 35-year old male with consanguineous parents initially presented with recurrent fever and was diagnosed with Epstein-Barr virus-driven chronic lymphoproliferation and eventually fulfilled all eight HLH criteria. The patient displayed normal pigmentation as well as Rab27a expression in blood-derived cells. However, patient NK and CD8+ T cell exocytosis was low. Ectopic expression of the Rab27a p.R184Q variant rescued melanosome distribution in mouse Rab27a-deficient melanocytes, but did not increase exocytosis upon reconstitution of human Rab27a-deficient CD8+ T cells. Mechanistically, the Rab27a p.R184Q variant displayed reduced binding to Slp2a but augmented binding to Munc13-4, two key Rab27a effector proteins in immune cells. Munc13-4 binding was particularly strong to a Rab27a p.T23N/p.R184Q double mutant, mimicking inactive Rab27a. CONCLUSIONS: Rab27a p.R184Q was stably expressed and could facilitate melanosome trafficking, yet did not support lymphocyte exocytosis by mechanisms of altered effector protein binding. The HLH-associated Rab27a variant increased Munc13-4 binding, representing a novel mode of selectively impairing hematopoietic cell Rab27a-mediated exocytosis and potentially contributing to development of late-onset HLH.

NOTES

Poster #39

ASSESSMENT OF BRAF V600E MUTATIONAL STATUS IN BIOPSIES AND PLASMA SAMPLES OF LANGERHANS CELL HISTIOCYTOSIS (LCH) PATIENTS: A PRELIMINARY REPORT FROM THE LCH ITALIAN REGISTRY

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PURPOSE: to assess lesional and circulating BRAFV600E in LCH patients referred to the Italian Registry. METHODS: Lesional BRAFV600E was analyzed by immunohistochemistry (IHC), Sanger sequencing or digital droplet PCR (ddPCR). Plasmatic cell-free DNA was extracted by QiAmp Circulating Nucleic Acid Kit (Qiagen) and subjected to ddPCR BRAFV600E FAM/HEX assay (BioRad) within 24h. Clinical data were collected and correlated to the mutational status. RESULTS: BRAFV600E was detected in the biopsy of 89/179 (49.7%) patients by IHC or Sanger sequencing. ddPCR unveiled the mutation in additional 10/32 (31.3%) wild-type cases. Plasmatic BRAFV600E at diagnosis/reactivation (baseline) was evaluated in 62 patients resulting positive in 5/5 (100%) multisystem risk organ positive (MS RO+), 14/25 (56%) multisystem risk organ negative (MS RO-) and 13/32 (40.6%) single system (SS) patients. Plasma from 17 BRAFV600E positive patients at baseline was longitudinally analyzed for a median time of 30 (6-36) months. Out of the 8 SS patients analyzed, negativization of plasmatic BRAFV600E within 6 months correlated with clinical remission in 5/5 SS unifocal bone patients and 1/3 SS multifocal bone patient treated with Vemurafenib. Differently, 2/3 SS multifocal bone patients exhibited persistent negative BRAFV600E levels despite recurrent active disease over a one-year follow-up. In 3/6 MS RO- and 3/3 MS RO+ patients, plasmatic BRAFV600E varied accordingly to treatment response, while no clear correlation was observed in the remaining 3/6 MS RO- patients. CONCLUSION: the use of ddPCR improved diagnostic accuracy of lesional BRAFV600E in LCH patients. Plasmatic BRAFV600E confirms as a potential biomarker for disease activity. However, discrepancies between clinical course and circulating BRAFV600E in about 30% of observed cases warrant investigation of BRAFV600E levels in blood cellular subsets and the role of BRAFV600E-indipendent pathogenetic patwhays. A larger prospective multi-center study is ongoing within the European consortium of Histiocytosis in order to extend actual findings.

Poster #40

EFFICACY OF SINGLE AGENT ORAL INDOMETHACIN IN CHILDREN WITH NEWLY DIAGNOSED AND RELAPSED LANGERHANS CELL HISTIOCYTOSIS

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PURPOSE: Indomethacin is a potent inhibitor of prostaglandin E2 synthesis; limited data has shown promising efficacy in children with Langerhans cell histiocytosis (LCH). METHODS: We conducted a single center retrospective study to assess the efficacy of single agent oral indomethacin on the overall survival, reactivation rates and late sequelae of children with newly diagnosed and relapsed LCH treated at the Hospital for Sick Children from Jan 1995 - Dec 2020. RESULTS: Twenty-one patients were treated with indomethacin monotherapy during their course of treatment. Twelve patients had single system (SS) disease while 9 had multisystem (MS) disease, of whom five (55%) were risk organ positive (RO+) and four (44%) were RO-. Nine patients (43%) were treated as upfront therapy, all had SS disease, nine (43%) had salvage/reactivated disease and 3 (14%) as maintenance therapy. Seven patients were BRAFV600E positive (33%), seven were BRAFV600E negative (33%) and seven patients had unknown BRAFV600E status (33%). Following indomethacin therapy twelve (57%) patients did not have disease reactivation during the study timeframe; of these, the majority had unifocal bony disease (n=9, 75%), SS (n=9, 75%), BRAFV600E negative (n=5, 42%) and were RO- (n=11, 92%). Nine (43%) patients reactivated with a median time of 8 months from the start of indomethacin therapy. The majority had MS disease (n=6, 67%) and 44% were RO+. These RO+ patients all reactivated in a low-risk organ (skin/bone). Five (55%) were BRAFV600E positive who reactivated. Five re-activated while on indomethacin and four re-activated after stopping indomethacin. The main toxicities were gastrointestinal side effects, nausea and GERD (n=4). Six patients developed long term LCH sequelae, including ND-LCH (n=2). All patients are alive with an overall survival of 100%. CONCLUSIONS: Indomethacin is a safe and effective drug especially for unifocal bone, SS, BRAFV600E negative, RO- LCH.

Poster #41

THREE DIFFERENT STRATEGIES OF TARGET THERAPY (VEMURAFENIB) IN PATIENTS WITH HIGH-RISK REFRACTORY LANGERHANS CELL HISTIOCYTOSIS TREATED IN ARGENTINA

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PURPOSE: To report the response of three patients with risk organ multisystem Langerhans cell Histiocytosis (RO-MS-LCH) to vemurafenib. Material and METHODS-RESULTS.Patient 1: One-year-old male with LCH-MS RO+ at diagnosis (skin, multifocal bone with CNS risk+RO+). Patient had progressive disease after treatments with prednisone(PDN)-vinblastine (VBL); cytarabine(ARAC)-PDN-Indomethacin(IMT) and cladribine (2CDA)-ARAC. BRAFV600E mutation was detected at a skin biopsy. After

vemurafenib 20 mg/kg administration, the patient showed complete response, although dermatological toxicity grade 3 was also observed. After 8 weeks, maintenance with methotrexate-mercaptopurine replaced the targeted therapy without evidence of reactivation.Patient 2: Male 3-years old, one-months old at diagnosis. LCH MS RO + with severe systemic compromise. After treatment with PDN-VBL and PDN-Vincristine-ARAC his disease progressed with bone lesions. 2CDA-ARAC therapy did not show any response. Treatment with Clofarabine showed partial response (severe transfusion requirements and severe hypoalbuminemia). BRAFV600E mutation was detected in a bone marrow biopsy, thus treatment with Vemurafenib was tailored at 40 mg/kg. Afterwards,the patient's condition improved. Transfusional requirement diminished and albumin levels returned to normal. His disease showed complete response of non-risk organs, but severe hepatosplenomegaly, moderated anemia and thrombocytopenia persisted. After 28 months, he is still on targeted therapy.Patient 3: Female full-term newborn with LCH MS RO+ (skin, liver, spleen, hematopoietic system and lungs) and with RO dysfunction and progressive cholestasis parameters. Even though BRAFV600E mutation was not detected, empirical treatment with vemurafenib 20 mg/kg/day was initiated at 20 days of life, with complete response. After 22 months, she is still on treatment with no evidence of active disease and no toxicity. CONCLUSION: Targeted therapy seems to be a viable option in high risk refractory patients with LCH. Its empirical use in these patients could be considered. More evidence, nevertheless longer follow-up and guidelines are necessary regarding targeted therapy.

Poster #42

LCH INVOLVING THE HEAD AND NECK: A SINGLE PEDIATRIC INSTITUTION EXPERIENCE

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PURPOSE: Langerhans cell histiocytosis (LCH) is a rare disorder caused by clonal expansion of cells of the mononuclear phagocyte system. The presentation is highly variable, ranging from a single lesion to multi-focal multisystemic disease. LCH frequently arises in the head and neck, but may be mistaken for more common pathologies resulting in delays in diagnosis. Skull-base and vertebral lesions present a higher risk due to increased likelihood of anterior pituitary dysfunction. METHODS: This single institution retrospective study included all pediatric patients with LCH of the head and neck presenting to our center between 2009 and 2019. Patients were classified by extent of disease. Patients with single-system disease were further classified by involvement of CNS-risk site(s). RESULTS: Sixty-one patients were included. Forty-eight had single-system disease, 28 of whom had disease affecting a CNS-risk site. Thirteen patients presented with multisystem disease. Most patients were diagnosed within 1-3 months of presentation; however, there were patients in all groups whose disease remained undiagnosed for 7-9 months. Of 20 patients with single-system non-CNS-risk LCH, 14 were managed with surgery alone and 6 with chemotherapy. In this group, 5 reactivations occurred in 3 patients. All patients with a CNS-risk site (n=28) and/or multisystem disease (n=13) received chemotherapy. In the CNS-risk group, 2 reactivations occurred in 1 patient. In the multisystem group, 9 reactivations occurred in 6 patients. One patient with multisystem disease died within 4 months of presentation due to liver failure. Long-term sequelae occurred most frequently in the CNS-risk and multisystem groups, and included headaches, developmental delay, anterior pituitary dysfunction, and hearing loss. CONCLUSION: Head and neck LCH presents a diagnostic challenge. The outcome is varied, depending on site(s) of disease, therapy, and genetics. Although many low-risk patients experience good outcomes with limited intervention, high-risk patients can require extensive therapy and may suffer long-term consequences.

Poster #43

MULTISYSTEMIC LANGERHANS CELL HISTIOCYTOSIS IN A YOUNG ADULT MALE WITH PLEOMORPHIC CUTANEOUS PRESENTATION

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PURPOSE: Langerhans cell histiocytosis (LCH) is an inflammatory myeloid neoplasia characterized by clonal proliferation of CD1a+ dendritic cells with infiltration of one or more organs. Although multisystem involvement in adults is rare, pulmonary involvement is the common feature seen as a part of multisystem involvement or as isolated presentation. Here, we describe a rare case of multisystemic LCH in an adult male with pleomorphic cutaneous manifestations. METHODS: A 29-year-old non-smoker male presented with multiple erythematous to pigmented papulonodular lesions over face involving forehead, nasolabial folds, perioral, bilateral concha, right axilla, anterior chest and perineal region associated with foul smelling purulent discharge at inter-triginous areas for the duration of 2 years. Patient also had history of dry cough which was intermittent in onset, associated with chest pain from 1½ month. There was no history of bony tenderness, symptoms suggestive of other systemic involvement. RESULTS: His baseline laboratory evaluation did not reveal any abnormality. Histopathological examination of skin lesions revealed dense collection of histiocytes with irregular nuclear contours with nuclear grooves admixed with eosinophils at dermo-epidermal junction and superficial dermis. At places these histiocytes show epidermotropism and extend up to deep dermis. No well-formed epitheloid granuloma, adnexal structure involved. Immunohistochemistry showed CD1a and langerin positivity. High-resolution computed tomography (HRCT) chest revealed right gross hydropneumothorax with multiple cysts in bilateral lung and few lytic lesions in right 10th and left 3rd rib. A total body PET scan evaluation did not reveal LCH at other sites. Diagnosis of multisytemic LCH was made and started on induction chemotherapy with oral prednisolone and vinblastine. CONCLUSIONS: LCH is considered as a great imitator mimicking wide range of dermatoses. Our patient had lesions masquerading seborrheic dermatoses, hidradenitis suppurativa and facial lesions resembling angiofibroma which could be a reason for delay in diagnosis or misdiagnosis and management.

Poster #44

RESPONSE TO MAPK INHIBITION OF NEURODEGENERATION IN LCH MONITORED BY CSF NEUROFILAMENT LIGHT AS A BIOMARKER

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PURPOSE: Langerhans cell histiocytosis (LCH) is an inflammatory myeloid neoplasia that can affect multiple organs. CNS involvement (CNS-LCH), which often affects pituitary function, can also cause slowly progressive neurodegeneration (ND-CNS-LCH), which frequently is resistant to conventional LCH-directed therapy. Notably, a population-based study reported that at least 24% of all children with LCH develop signs of ND-CNS-LCH on long-term follow-up with magnetic resonance imaging (MRI). Thus, a strategy for early detection, treatment, and monitoring of ND-CNS-LCH is imperative. In most LCH patients, somatic activating genetic alterations in the mitogen-activated protein kinase (MAPK) pathway can be detected. Targeted MAPK inhibition (MAPKi) has pronounced clinical efficacy in refractory LCH. However, evidence of the therapeutic efficacy of MAPKi in established ND-CNS-LCH is limited. METHODS: In this study, we evaluated the response to MAPKi therapy in five children with CNS-LCH, four of whom with ND-CNS-LCH, by analyzing their cerebrospinal fluid (CSF) for biomarkers of neurodegeneration, including neurofilament light (NFL). CSF-NFL is a sensitive and well-established biomarker of neuroaxonal damage. results were correlated with clinical and neuroradiological findings. RESULTS: Notably, CSF-NFL levels were initially pathologically elevated in all children with CNS-LCH, but normalized (<380 ng/L) within 6 months in four children and in all five within 9 months after initiation of MAPKi therapy (p=0.041, paired t-test). Notably, in the two patients that MAPKi therapy was discontinued, CSF-NFL levels increased again to abnormal levels within 4 months. MAPKi therapy was associated with perceivable neuroradiological and clinical improvement in three and two children, respectively. For comparison, CSF-NFL levels were normal in all but one (440 ng/L) of 16 samples from 11 additional children with LCH without known CNS-LCH. CONCLUSION: We conclude that CSF-NFL is a relevant surrogate biomarker in ND-CNS-LCH, and that MAPKi therapy appears to effectively reduce this neurodegeneration.

Poster #45

BRAFV600E-POSITIVE CELLS AS MOLECULAR MARKERS OF BONE MARROW DISEASE IN PEDIATRIC LANGERHANS CELL HISTIOCYTOSIS

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Risk organ involvement in Langerhans cell histiocytosis (LCH), including the hematopoietic system, liver, and spleen, is associated with relapse, leading to permanent complications. The BRAFV600E mutation in LCH is associated with an inadequate early response to chemotherapy and reactivation. However, the BRAF mutation status and clinical impact at the molecular level of bone marrow disease (BMD) are not fully understood. We retrospectively performed mutational analyses of 59 LCH tumors by targeted amplicon sequencing using custom-designed primers and subsequently analyzed somatic mutations in 41 paired bone marrow (BM) samples by allele-specific droplet digital PCR. We examined BMD at the molecular level in 38 paired BM samples, excluding three patients for whom quantitative mutational analysis was not applicable. We detected BMD before treatment at various allele frequencies (0.03-7.0%, median 0.83%) in 21 of the 38 (55%) cases, consisting of 12 MSRO+, 7 MSRO-, 2 MFB, and 0 SS cases. The

BRAFV600E mutation was detected in 21 of 25 (84%) BM samples from BRAFV600E-positive LCH, whereas no other variants were identified. The rates of BM involvement in the MSRO+, MSRO-, MFB, and SS cases were 100%, 54%, 25%, and 0%, respectively. In the 25 cases of BRAFV600E-positive LCH, the median variant allele frequencies (VAFs) of BRAFV600E in the BM of the MSRO+, MSRO-, and MFB cases were 1.0% (0.20-7.0%), 0.030% (0-2.2%), and 0% (0¬-0.51%), respectively. However, no somatic mutations were detected in the 13 cases with other types of mutations. Mutational burden varied between the four different clinical phenotypes (Kruskal-Wallis, $p < 0.001$). Notably, MSRO+ disease showed the highest mutational burden (Mann-Whitney test, $p < 0.001$). A high mutational burden in the BM defines a distinct clinical phenotype of high-risk, young-age patients with multisystem LCH and potential alternative risk factors.

Poster #46

CLONAL EVOLUTION OF T-LYMPHOBLASTIC LEUKEMIA/LYMPHOMA TO LANGERHANS CELL HISTIOCYTOSIS WITH MALIGNANT FEATURES IN AN 8-YEAR-OLD BOY

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PURPOSE: Malignant histiocytosis (MH) is an uncommon neoplastic disorder of Langerhans cells with malignant cytologic features. MH may arise de novo, as progression from Langerhans cell histiocytosis (LCH), or rarely following lymphoid malignancy. Therapies remain limited for disseminated disease, with hematopoietic stem cell transplant (HSCT) reported to be curative. METHODS: We present a pediatric patient with disseminated LCS transdifferentiated from T-lymphoblastic leukemia/lymphoma (T-ALL). RESULTS: An 8-year-old boy with a history of T-ALL (NOTCH1 and NRAS gene mutations) in remission developed hyperpigmented papules 1 month after completing therapy. Skin biopsy was diagnostic for LCH (S100, Langerin, CD1a+). Molecular studies showed T-gamma gene rearrangement and NOTCH1 and NRAS mutations identical to his original T-ALL, in addition to KRAS and GATA3 gene mutations. No BRAF-V600E mutation or evidence of T-ALL was detected. Patient had skin and lymph node involvement. He received standard LCH therapy per LCH III with initial response. However, skin lesions progressed. Therapy was switched to cytarabine/vincristine and prednisone with minimal response. Two lesions became large and ulcerated and biopsy showed LCH with malignant histologic features (aggressive infiltrative growth pattern, cytologic pleomorphism), identical NOTCH1 and NRAS mutations, with a novel BRAF non-V600E mutation. He underwent surgical resection of the largest lesions with clear margins and was initiated on MEK and BRAF inhibitors with rapid resolution of the remaining ulcerated papules. Two months after therapy, a new MH lesion developed. Therapy was switched to clofarabine with partial response. He currently awaits HSCT. CONCLUSION: Due to the paucity of cases of MH in pediatrics, there is no standard of care. MH following lymphoid malignancy, in this case T-ALL, may show different behavior than de novo MH. Multimodality systemic therapy plus surgical excision with clear margins for local disease control appear to be successful in achieving partial response to date in this pediatric patient.

Poster #47

SPECIFIC CLINICAL INDICATORS AND TREATMENT REGIMEN STUDY OF FEBRILE CHILDREN WITH LANGERHANS CELL HISTIOCYTOSIS

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Langerhans cell histiocytosis (LCH) is a myeloproliferative disorder characterized by infiltration and accumulation of CD207+CD1a+ langerhans cells, which resulted in lesions in different tissues or organs. Some patients have recurrent or persistent fever with a body temperature over 38â„ƒ on their first visit. We firstly figured out the clinical indicators significantly correlated with initial fever of LCH children, including age, skin involvement, risk organ involvement, HLH phenomenon, BRAFV600E mutation, ferritin level, procalcitonin etc. Levels of many clinical indicators also had significant differences between patients with fever and patients without fever. Then we compared the progression-free survival rates (PFS) of current LCH regimens, including prognosis of 6 weeks after first-line treatment, first-line treatment, second-line treatment and target therapy. Only the PFS for first-line treatment therapy of these two groups (patients with or without fever) showed significantly different. Furthermore, we analyzed the clinical indicators correlated with prognosis of first-line treatment, such as fever, age, HLH phenomena, ferritin, skin involvement etc. Further multivariate liner regress analyzation showed that HLH phenomena (P=0.002) and BRAFV600E mutation in tissue (P=0.026) were the critical affecting factor for first-line treatment. Our research also found that IL-10 level could predict initial fever and the prognosis of 6 weeks after first-line treatment, with a cut-off value of 4.1950 pg/mL and 4.2150pg/mL separately.

Poster #48

HISTIOCYTOSIS LANGERHANS IN CHILDREN AND SOLID TUMORS: ON THE SAME MECHANISM ? A CASE REPORT.

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PURPOSE: Aim of our study was to investigate the coincidence of Langerhans cell histiocytosis (LCH) with other primary tumors. The association between LCH and other malignancies has been described, with frequencies varying from 2.6% in children who received chemotherapy to 32% in adults. Commonly occurs during treatment, or later after treatment. There are no reports for children with asymptomatic LCH without treatment, who developed solid tumors. METHODS: We present a girl 17-years old, who was diagnosed at the age of 8 with low-risk LCH, on the parietal bone. Nine years later, while she was in remission for LCH, she presented with a second primary "benign" brain tumor. RESULTS: A previously healthy girl, at the age of 8 had an accidental fallen. On the routine check-up with a cranial x-ray after fallen, three lytic lesions on the right parietal bone were diagnosed. The CT and MRI imaging revealed a normal brain. The biopsy of the bone lytic lesions revealed Langerhans cell histiocytosis. The laboratory investigation of this case, classified as a local lesions-single system and according to LCH III protocol, no treatment was performed. The bone lesions progressively resolved. Nine years later, while the girl had a systematic follow-up, she presented with progressively worsening polydipsia and polyuria. Î‘ urine test was performed while the brain CT-scan and the MRI imaging revealed a mass in the supra cellar mainly cystic with a small solid component. No evidence for reactivation of LCH was documented. Total surgical resection of the brain mass was performed and the histology revealed a craniopharyngioma. CONCLUSIONS: The coincidence of LCH with a second primary "benign" brain tumor, is remarkable with an interval of 9 years after the first diagnosis of LCH, even in childhood. Awareness in the follow-up of patients with LCH for a second primary tumor is needed.

Poster #49

CHILDREN WITH LANGERHANS CELL HISTIOCYTOSIS (LCH): GREEK NATIONAL REGISTRY RESULTS OVER THE LAST TWO DECADES

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PURPOSE: To describe the characteristics, treatment and outcome of pediatric patients with LCH in Greece. METHODS: Between 2000-2018, 7 Hematology-Oncology Centers retrospectively registered data of 169 patients (62.72% males). Mean age at diagnosis was 5.59 years (range: 0.01-20.51), 52<2 years. RESULTS : One-hundred-thirty-three (78.7%) patients presented with SS-disease, bones 110 (82.71%), skin 11 (8.27%), hypothalamus/ pituitary 3 (2.26%), other 3 (2.26%). Forty-six (34.6%) patients had CNS-Risk+ lesions, no neurodegenerative disease recorded, 4 patients had diabetes insipidus (DI). Initially 66 (49,6%) were observed only, 5(7.5%) relapsed. All patients are alive in CR (OS:100%, EFS:92.5%, median-time of follow-up (MTFup) 4.2 years (range, 0.1-19.7). Systemic chemotherapy received 71 (53.4%) SS-disease children, all Prednisolone/Vinblastine. Thirty (22.6%) patients had CNS-Risk+ lesions and 4 developed DI. Two patients died (pneumothorax, lymphoma), OS:98.5%. Thirteen patients relapsed, EFS:85.7%, MTFup 6.51 years (range 0.3-19.7). Thirty-six patients (21.3%) presented with MS-disease. Median age 1.7 years (range: 0.35-10.33 / 21 <2 years, 58.3%), with 2-5 system involvement and hematopoietic/liver/spleen involvement in 6/9/6 patients, respectively. Twenty-seven patients had bone localization (CNS-Risk+21). Risk-Organ+ had 22 patients (59.5%). Ten patients (27.0%) developed DI. The majority received Prednisolone/ Vinblastine, 3 Prednisolone/Vinblastine/Etoposide and 1 Prednisolone-only, for median treatment duration of 12 months (range 1-43). First relapse or resistant disease was observed in 9 patients (24.3%): EFS:75.0%, second/ third relapse in 6/3 patients. One patient following Cladribine treatment succumbed (CMV-infection, multi-organ system failure), 1 patient is alive (CR) following stem-cell-transplant, OS 97.3%. Overall, 27/51 (52.94%) evaluated patients were BRAF-V600E+, 21/39 SS and 6/12 MS. Five SS BRAF-V600E+ patients relapsed (all 5 with upfront chemotherapy), 6/6 MS BRAF-V600E- patients are in CR1 and 3/6 BRAF-V600E+ have relapse/resistant disease. No targeted therapy given. CONCLUSIONS: These retrospective results of Greek children with LCH are comparable with international and are the basis for our participation to the LCH-IV Protocol, supported by the Artemis Association.

Poster #50

RELAPSES IN PEDIATRIC PATIENTS WITH MULTI-SYSTEM OR MULTI-FOCAL BONE LANGERHANS CELL HISTIOCYTOSIS - DATA FROM THE JLSG 96/02 STUDY

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PURPOSE: Relapses of LCH are common. We assessed pediatric patients with relapsed LCH who had been treated with the JLSG-96/02 protocol. METHODS: 317 patients with LCH (111 with multifocal bone [MFB] and 206 with multisystem [MS]) treated from 1996 to 2009 were studied. Median follow -up was 10.6 years. Relapse of LCH was defined as exacerbation after attaining non-active disease. RESULTS: 101/317 (31.9%) patients (MFB: 31/111 [27.9%], MS: 70/206 [34.5%], p=0.33) had first relapse at median 1.5

(range, 0.2 -5.0) years after the initiation of therapy. Cases with subsequent (2 -6) relapses were also similar (14/31 vs. 37/70; p=0.52) between MFB and MS patients. Incidence of more than 2 relapse episodes was 22/51 vs. 64/132 (p=0.63) in MFB and MS diseases, respectively. Of total 187 relapse episodes, relapse at single - system accounted for 85%; in which bone relapse was the most common (55%) followed by central diabetes insipidus (CDI; N=23;12 %) in both MFB and MS diseases. Relapse at multi - system with risk organ involvement was extremely rare (6 relapses [3%] in total, 5 relapses at 1st and one relapse at 2nd) and was significantly higher in MS than MFB diseases (p<0.05). Two MS patients died of relapses in risk organs at first and 2nd relapse, respectively. Regarding treatment, patients received chemotherapy in 122/187 (65%) at relapses; of which 101/122[82%] were treated with the initial regimen. Of 164 relapses, excluding 23 CDI relapses, 142 (87%) were well controlled. Some patients with multiple relapses up to 6 showed permanent sequelae. CONCLUSIONS: In JLSG cohorts, incidence of relapses did not differ between MFB and MS disease; however, rare relapses at multi - system with risk organ involvement occurred more in MS diseases. The most relapses were well controlled by chemotherapy including initial regimen. Further improvement is needed for patients with relapsed LCH.

Poster #51

HYDROXYUREA THERAPY FOR PEDIATRIC RELAPSED LANGERHANS CELL HISTIOCYTOSIS

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BACKGROUND: Despite a significant improvement of the overall survival in pediatric Langerhans cell histiocytosis (LCH) patients, relapse is still common, and fundamental treatment for relapsed LCH has not yet been established. Hydroxyurea (HU) therapy for relapsed LCH was proposed by Zinn DJ et al. (Blood 2016). In their report, only 2/15 (13%) patients are children. Therefore, the efficacy and toxicity profile of HU therapy for pediatric LCH remains unknown. Herein, we report two pediatric cases of HU therapy for relapsed LCH. METHODS: We treated two relapsed pediatric LCH patients with HU following a standard frontline chemotherapy during 2019 to 2021. RESULT: Case 1: A 13.4 years old boy with the 7th multi focal bone (MFB) recurrence. Due to the vertebral lesions with intraspinal soft tissue extension, vinblastine and prednisolone were administered for 6 weeks. After partial response (PR) has achieved, he received HU (500mg/day) with MTX therapy as maintenance therapy. He maintained non-active disease (NAD) without recurrence for 2.5 years. Case 2: A 7.7 years old girl with the second MFB recurrence at parietal bone and mandible. HU (500mg/day) monotherapy was effective for all lesions and she is currently on treatment after 5 months. CONCLUSIONS: In these two cases, the therapeutic response of HU and/or MTX therapy is comparable to that of conventional treatment for relapsed pediatric LCH patients. In both cases, partial response was achieved at early period of treatment and the disease status of NAD has been confirmed at their last follow-up. No serious adverse event was observed and dose reduction was unnecessary. Such an oral regimen reduces the physical and mental stress of painful injections for children, in addition to the number of clinic visits and medical cost. Thus, it gives a great benefit to pediatric LCH patients who are likely to repeat recurrences.

Poster #52

CLINICAL STUDY OF MAP2K1-MUTATED LANGERHANS CELL HISTIOCYTOSIS IN CHILDREN

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PURPOSE: To analyze the genetic and clinical features of children with MAP2K1-mutated Langerhans cell histiocytosis (LCH). METHODS: The clinical data of newly treated patients with MAP2K1-mutated LCH from July 2017 to October 2020 were collected. At the same time, two control groups were compared, consisting of all newly treated patients who had either the BRAFV600E mutation (n=133) or no known mutation of the MAPK pathway (n=59). RESULTS: We found 13 mutations of the MAP2K1 gene, which were mainly concentrated at p.53-62 and p.98-103. The most common mutation site was c.172_186del (12/37). Compared with the BRAFV600E mutation group, the patients with MAP2K1 mutations were mainly characterized by single system multiple bone involvement (P=0.022), with later disease onset (P=0.029) as well as less involvement of risk organs, especially liver (P=0.024). There was no significant difference in clinical features compared with the no known mutation group. The 2-year progression-free survival rate of first-line treatment in MAP2K1-mutated patients was 65.6% ± 9.5%, without a significant difference among the three groups. The prognosis of patients with lung involvement was poor [hazard ratio (95% confidence interval) = 6.312 (1.769-22.526), P = 0.005]. More progression or relapses could be found in patients with bony thorax involvement (8/17 vs. 2/20, P=0.023), yet involvements in other sites of bones were not correlated to disease progression or relapse. CONCLUSION: MAP2K1 mutations in LCH are concentrated at amino acids 53-62 and 98-103, and c.172_186del is the most common mutation site. Compared with patients with the BRAFV600E mutation, patients with the MAP2K1 mutation were mainly characterized by more SS-multiple bone involvement, less risk organ and skin involvement and later disease onset, requiring clinical stratification and precise treatment. The prognosis of patients with lung involvement, frequently associated with bony thorax involvement, is poor and should be given more attention in further studies.

Poster #53

GENETIC LANDSCAPE AND ITS PROGNOSTIC SIGNIFICANCE IN CHILDREN WITH LANGERHANS CELL HISTIOCYTOSIS

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PURPOSE: The aim of this study was to interrogated the LCH molecular landscape in childhood LCH biopsy using next-generation sequencing, and explored the prognostic significance of different gene mutations. METHODS: There were 223 patients with available formalin-fixed paraffin-embedded (FFPE) biopsy samples for targeted sequencing of a panel of genes at diagnosis were included in this study. RESULTS: Overall, 32 genomic alterations in MAPK pathway were observed in 186 out of 223 cases (83.4%). 21 (65.6%) mutations have been reported in children with LCH and are significantly pathogenic. A novel fusion gene FNBP1-BRAF and two mutations of MAP3K10 were identified and verified to resulted in activation of MAPK pathway. FNBP1-BRAF fusion gene, MAP3K10 p. A17T and MAP3K10 p. R823C were confirmed sensitize to Trametinib but not to Dabrafenib. Among different genetic groups of patients, those with ARAF mutation or more than one mutation had higher progression/reactivation rates (75% and 50% respectively). And, there was significant difference in 2-year PFS among different genetic groups of patients, with the lowest PFS in those with ARAF mutation or more than one mutation (BRAFV600E : 70.9% ± 5.3%, BRAF other mutations : 88.2% ± 6.4%, MAP2K1 mutations : 57.9% ± 17.7%, ARAF mutations : 25.0% ± 21.7%, more than one mutation : 31.3% ± 23.7% and others: 69.5% ± 8.3%, respectively, P = 0.027). We also found independent prognostic significance of ARAF mutation and more than one mutation for PFS in childhood LCH (ARAF mutation, Hazard Ratio, 4.250; 95%CI, 1.264-14.296; P = 0.019; more than one mutation, Hazard Ratio, 3.531; 95%CI, 1.258-9.915; P = 0.017). CONCLUSION: In summary, the majority of LCH patients carries somatic mutations involving MAPK pathway. The children with ARAF mutations and two mutations had poor prognosis. Different genotypes contribute to disease risk stratification and guide of medical treatments.

Poster #54

THE ROLE OF BRAF-MUTATED HEMATOPOIETIC CELLS IN PATIENTS WITH LANGERHANS CELL HISTIOCYTOSIS

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PURPOSE: Langerhans cell histiocytosis (LCH) is a neoplasm marked by the accumulation of CD1A+CD207+ cells. It is most commonly driven by a somatic, activating mutation in the BRAF serine-threonine kinase (BRAF-V600E) which can also be found in other cells of the hematopoietic system in patients with LCH. Here we ask which role these BRAF-mutated CD1A-CD207 - cells might have in patients with LCH and how RAF-inhibition affects the function of these cells. METHODS: We performed single-cell transcriptome analyses of peripheral blood and bone marrow cells using the 10x Genomics Chromium Single Cell Controller. The downstream analysis was performed using Seurat package for R. BRAFV600E content in different cell types at time of diagnosis and during treatment was determined using ddPCR. Serum cytokine levels at different timepoints were measured using the Inflammation 20-Plex Human ProcartaPlexâ„¢ Panel. Fluorescence-activating cell sorting was performed using a FACScan flow cytometer (BD FACSAria). RESULTS: Using a single patient as an example, we provide evidence that peripheral blood BRAFV600E levels do not correlate with clinical presentation during treatment with a RAF-inhibitor, but that RAF inhibtion leads to a fast but reversible clinical remission. We also show that serum inflammatory cytokines exactly mirror RAF inhibition. Genotyping analysis identified the BRAFV600E mutation in hematopoietic stem cells and both myeloid and lymphoid lineages. Finally, the single-cell transcriptome analyses of peripheral blood and bone marrow cells at time of diagnosis and during treatment RAF-inhibition identifies pathways involved in LCH pathogenesis. CONCLUSION: BRAF-mutated cell populations other than the pathognomonic CD1A+CD207+ cells may directly contribute to the clinical picture of LCH.

Poster #55

PLASMA SIGNALLING FACTORS IN PATIENTS WITH LANGERHANS CELL HISTIOCYTOSIS (LCH) CORRELATE WITH RELATIVE FREQUENCIES OF LCH CELLS AND T CELLS WITHIN LESIONS

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PURPOSE: Langerhans cell histiocytosis (LCH) lesions contain an inflammatory infiltrate of immune cells including myeloid-derived LCH cells that often harbour mutations. Cell-signalling proteins within the lesion environment suggest that LCH cells and T cells contribute majorly to the inflammation. Foxp3+ regulatory T cells (Tregs) are enriched in lesions and blood from patients with LCH and are likely involved in LCH pathogenesis. In contrast, mucosal associated invariant T (MAIT) cells are reduced in blood from these patients and the consequence of this is unknown. Serum/plasma levels of cytokines have been associated with LCH disease extent and may play a role in the recruitment of cells to lesions. This study aimed at understanding whether plasma cell-signalling factors corresponded with LCH cells and/or LCH-associated T cell subsets in patients with LCH. METHODS: Cell-signalling factors including cytokines, chemokines and soluble immune checkpoint molecules (48 analytes total), were measured in patient plasma using LEGENDplex panels (BioLegend). Matched lesion-derived single-cell suspensions and/or peripheral blood mononuclear cells were analysed using flow cytometry. We tested for correlations between plasma factors and the relative frequency of (a) LCH cells, (b) Foxp3+ Tregs and (c) MAIT cells. RESULTS: The relative frequency of LCH cells in lesions (n=7) correlated with plasma levels of IL-11 (r=0.808, p=0.038), soluble CD27 (r=-0.857, p=0.024) and CCL2 (r=-0.893, p=0.012). In addition, plasma CCL2 and Tim-3 levels correlated with lesion (n=7, r=0.893, p=0.012) and blood (n=10, r=0.830, p=0.005) Treg proportions respectively. Furthermore, lesion MAIT cells (n=6) correlated with plasma CCL17 (r=0.886, p=0.033) and CCL5 (r=0.893, p=0.012) levels. CONCLUSION: This study highlights plasma cell-signalling factors that are associated with LCH cells, MAIT cells or Tregs in patients, thus they are potentially important in LCH pathogenesis. Further study into these associations is needed to determine whether these factors may become suitable prognostic indicators or therapeutic targets to benefit patients.

HISTIOCYTE SOCIETY GOVERNING BY-LAWS

The Society shall be governed and guided by the principles set forth in these By-Laws.

ARTICLE I
OFFICES, REGISTERED OFFICE, AND REGISTERED AGENT

Section 1. Offices. The principal office of Histiocyte Society, Inc. (the "Corporation") shall be located within or without the State of New Jersey, at such place as the Board (as defined below), in its sole discretion, shall from time to time designate. The Corporation may also maintain additional offices at such other places as the Board may from time to time designate.

Section 2. Registered Office and Registered Agent.
The Corporation shall have and continuously maintain a registered office and a registered agent in the State of New Jersey, as required by the New Jersey Nonprofit Corporation Act (the "Act"). The registered agent shall be either an individual resident of the State of New Jersey or a corporation authorized to transact business in the State of New Jersey, in accordance with the Act.

ARTICLE II
PURPOSES AND MISSION

Section 1. Purposes. The purposes for which the Corporation is formed are as set forth in the Corporation's Certificate of Incorporation (the "Certificate of Incorporation").

Section 2. Mission. The mission of the Corporation is to: (i) improve the state of knowledge of the histiocytic disorders and improve the welfare of patients with these disorders; (ii) promote, facilitate, and carry out research in histiocytic disorders; (iii) facilitate and provide a forum for health care professionals for effective communication concerning these aims; (iv) promote education and to educate physicians, scientists, and others in matters related to the histiocytic disorders; (v) advise lay organizations in educational and other matters concerning the histiocytic disorders; and (vi) collaborate with organizations that have common goals.

ARTICLE III
MEMBERSHIP

Section 1. Classes. The Corporation shall have three (3) classes of members: (i) ordinary members (the "Ordinary Members"); (ii) honored members (the "Honored Members"); and (iii) emeritus members (the "Emeritus Members").

A. Ordinary Members. Ordinary Members shall be health care professionals who are active in patient care, education or research in the histiocytic disorders. Ordinary Members pay dues, may participate in all activities of the Corporation, and hold office.

B. Honored Members. Honored Members are distinguished individuals, who, in the view of the Board, have made extraordinary contributions to the Corporation. Honored Members enjoy all the rights and privileges of Ordinary Members, but do not pay dues and may not hold office.

C. Emeritus Members. Emeritus Members are Ordinary Members who have retired from active involvement in the field. Emeritus Members enjoy all rights and privileges of Ordinary Members, but do not pay dues and may not hold office.

Section 2. Qualifications. The Board shall determine, in its sole discretion, the qualifications, dues, terms, and other conditions of each class of member.

Section 3. Voting Rights. All members shall have the right to vote on the following matters: (i) election of the Board and officers; (ii) election of members of the Education and Scientific Committees and other committees as deemed appropriate by the Board; (iii) approval of the annual budget proposed by the Board; (iv) approval of any amendments to these Amended and Restated Bylaws (these "Bylaws"); and (v) other issues as the Board may choose to bring before the members. Voting on all other matters is expressly reserved for the Board.

Section 4. Member Meetings. There shall be an annual meeting of the members upon such date, time, and place as the Board shall determine. Special meetings of the members may be called by the President or upon the request of a majority of the voting members.

Section 5. Notice. Members shall receive not less than thirty (30) nor more than sixty (60) calendar days prior written notice of all member meetings. Notice shall be given in the manner specified in Article VIII of these Bylaws. The purpose for which a special meeting is called shall be stated in the notice. Any member may waive notice of any meeting by a written (including electronic) statement, either before or after the meeting. Notwithstanding the foregoing, attendance and participation at a meeting without objection to notice shall constitute a waiver of notice.

Section 6. Quorum and Voting. Each voting member shall have one vote on each voting matter. A quorum shall consist of at least ten percent (10%) of the total voting members. A majority of the votes cast on each voting matter at which a quorum exists shall constitute a valid action of the members.

Section 7. Removal. Any member may be removed from membership by a majority vote of the Board only: (i) for cause, which is defined as failure to pay dues for three (3) consecutive years; or (ii) other causes as determined by the Board in its sole discretion. The Board shall be the sole judge of moral, ethical, and professional qualifications required for election to or termination of membership.

Section 8. Action by Written Consent. Except as otherwise expressly prohibited by the Act, applicable law, the Certificate of Incorporation or these Bylaws, any action required or permitted to be taken at a meeting of the members (other than the biennial election of Board members), may be taken without a meeting upon the written consent of members who would have been entitled to cast the minimum number of votes which would be necessary to authorize the action at a meeting at which all members entitled to vote thereon were present and voting; provided, that: (i) the Corporation provides to all other members advance notification setting forth the proposed action consented to; (ii) the proposed action is not consummated before the expiration of ten (10) days from the giving of the notice (and twenty (20) days from the giving of the notice in the case of any action taken pursuant to Chapter 10 of the Act); and (iii) the notice sets forth the existence of such ten (10) day period; provided further, that the writings are filed with the minutes of the members.

ARTICLE IV
BOARD OF TRUSTEES

Section 1. Powers. There shall be a Board of Trustees of the Corporation (the "Board"), which shall supervise and control the business, property, and affairs of the Corporation, except as otherwise expressly provided by the Act, applicable law, the Certificate of Incorporation, or these Bylaws. All members of the Board shall serve without financial compensation.

Section 2. Number and Qualifications. The Board of the Corporation shall be composed of no less than five (5) and no more than nine (9) individuals. The number of Board members may be decreased (but in no event to fewer than three (3) members), however, no decrease shall have the effect of shortening the term of any incumbent member of the Board.

Section 3. Composition. The Board shall consist of those individuals then serving as the President, the President-Elect, the Past President, the Secretary, the Treasurer, and two Members-at-Large.

Section 4. Election and Term of Office. The members of the Board shall be elected by the voting members as set forth in Article V, and shall serve until their successors are elected and qualified, or their earlier removal, resignation or death.

Section 5. Resignation and Removal. Any Board member may resign at any time by giving written notice to the President. Such resignation shall take effect at the time specified therein, or, if no time is specified, at the time of acceptance thereof as determined by the President. A Board member may be removed with cause by vote of a two-thirds majority of the Board at a duly held meeting with a quorum present. The remaining Board members of the Corporation shall be the sole judge of moral, ethical, and professional qualifications required for removal from the Board.

Section 6. Vacancies. Vacancies on the Board, whether caused by resignation, removal, death, an increase in the authorized number of Board members or otherwise, may be filled by the affirmative vote of a majority of the remaining Board members, although less than a quorum, or by a sole remaining Board member. A Board member elected to fill a vacancy shall serve for the unexpired portion of such term.

Section 7. Meetings. A regular annual meeting of the Board of the Corporation shall be held each year, at such time, day, and place as shall be designated by the Board. Special meetings of the Board may be called at the direction of the President or by a majority of the Board members then in office, to be held at such time, day, and place as shall be designated in the notice of the meeting.

Section 8. Telephone Meetings. Any one or more Board members may participate in a meeting of the Board by means of a conference telephone or similar telecommunications device that allows all persons participating in the meeting to hear each other. Participation by telephone or other telecommunications devices shall be equivalent to presence in person at the meeting for purposes of determining if a quorum is present.

Section 9. Notice. Notice of the time, day, and place of any meeting of the Board shall be given not less than twenty-four (24) hours prior to such meeting, in the manner set forth in Article VIII. The purpose for which a special meeting is called shall be stated in the notice. Any Board member may waive notice of any meeting by a written (including electronic) statement, either before or after the meeting. Notwithstanding the foregoing, attendance and participation at a meeting without objection to notice shall constitute a waiver of notice.

Section 10. Quorum. A majority of the Board members then in office shall constitute a quorum for the transaction of business at any meeting of the Board.

Section 11. Manner of Acting. Except as otherwise expressly required by the Act, applicable law, the Certificate of Incorporation or these Bylaws, the affirmative vote of a majority of the Board members present at any meeting at which a quorum exists shall be the act of the Board. Each Board member shall have one vote.

Section 12. Action by Written Consent. Except as otherwise expressly prohibited by the Act, applicable law, the Certificate of Incorporation or these Bylaws, any action required or permitted to be taken at any meeting of the Board, or any committee thereof, may be taken without a meeting if all the members of the Board or of such committee consent thereto in writing (including by electronic transmission), and the writings are filed with the minutes of the Board or committee.

ARTICLE V
OFFICERS

Section 1. Officers. The officers of the Corporation shall consist of: (i) president (the "President"); (ii) president-elect (the "President-Elect"), whenever this office is occupied in accordance with Section 1.B of this Article V below; (iii) immediate past-president (the "Past-President"), when this office is occupied in accordance with Section 1.C. of this Article V below; (iv) secretary (the "Secretary"); (v) treasurer (the "Treasurer"); and (vi) two (2) members-at-large (each, a "Member-at-Large" and together, the "Members-at-Large"). The Corporation shall have such other assistant officers as the Board may deem necessary in its sole discretion, and such officers shall have such authority as prescribed by the Board. One person may hold more than one office.

A. President. The President shall give active direction and have control of the business and affairs of the Corporation for a 3-year term. The President may be elected for no more than two terms, provided, however, that such terms shall not be consecutive. The President may sign contracts and other instruments, which the Board has authorized to be executed, and shall perform all duties incident to the office of President, as may be prescribed by the Board.

B. President-Elect. The President-Elect is an officer of the Corporation and assumes the office of President two (2) years following such individual's appointment as President-Elect. If for any reason, as determined by the Board, the President is unable to carry out the duties of such office, the President-Elect shall assume the office of President for the remainder of the President's term. The President-Elect shall be elected by the voting members of the Corporation at the time of the annual meeting of the members that occurs one year following the annual meeting of the members that elected the President. For the avoidance of doubt, the President-Elect shall remain vacant during the term that the Past-President serves in office.

C. Past-President. After serving one full term as President, such individual becomes the Past-President and remains an officer of the Board for one year immediately thereafter.

D. Secretary. The Secretary shall keep or cause to be kept the minutes of all meetings of the Board and shall perform such other duties and possess such other powers as are incident to the office of Secretary or as shall be assigned to such individual by the President or the Board. The Secretary serves a two year term with two additional terms permitted by re-election.

E. Treasurer. The Treasurer shall, subject to oversight by the Board, maintain general supervision over the financial affairs of the Corporation and shall cause to be kept accurate books of account. The Treasurer shall oversee the disbursement of funds of the Corporation and shall from time to time, or upon request from the Board, account for all the transactions undertaken as Treasurer, and of the financial condition of the Corporation. The Treasurer serves a two year term with two additional terms permitted by re-election.

F. Members-at-Large. Each Member-at-Large shall assist the other Board members in the conduct of their duties as directed by the President or by consensus of the Board. Candidates for a
Member-at-Large position shall be ordinary members who have not served on the Board for at least two years prior to assuming a term as a Member-at-Large. The Members-at-Large shall each serve a three year term with one additional term permitted by re-election.

Section 2. Election of Officers. The President-Elect, Secretary, Treasurer, and Members-at-Large shall be elected, as the case may be, by the voting members of the Corporation at an annual meeting of the members in

accordance with the applicable term structures set forth in Section 1 of this Article V.

Section 3. Term of Office. Each officer of the Corporation shall be installed at the annual meeting of members at which they are elected, and shall hold office for terms as set forth in Section 1 of this Article V, or until their respective successors shall have been duly elected and qualified, or their earlier removal, resignation or death.

Section 4. Resignation and Removal. Any officer may resign at any time by giving written notice to the President. Such resignation shall take effect at the time specified therein, or, if no time is specified, at the time of acceptance thereof as determined by the President. An officer may be removed with cause by vote of a two-thirds majority of the Board at a duly held meeting with a quorum present.

Section 5. Vacancies. Vacancies shall be filled by a majority vote of the Board.

ARTICLE VI
COMMITTEES

Section 1. Standing Committees. Standing Committees include the: (i) nominating committee (the "Nominating Committee"); (ii) program committee (the "Program Committee"); (iii) scientific committee (the "Scientific Committee"); (iv) education committee (the "Education Committee"); and (v) disease steering committee (the "Disease Steering Committee"). The Board in its sole discretion may create other committees on an ad-hoc basis.

A. Nominating Committee. The Nominating Committee shall be composed of the President, President-Elect, Past-President, Secretary, and Treasurer, and shall be responsible for providing the Board with candidates for office, membership, and standing committees, as requested by the Board from time to time.

B. Program Committee. The Program Committee shall be composed of the President, Secretary, Chairperson of the Education Committee, Chairperson of the Scientific Committee, the Secretariat, and additional members chosen from among the members of the Corporation (as determined by the Board, in its sole discretion). The President shall act as Chairperson of the Program Committee. The Program Committee shall be responsible for planning, organizing, and executing the annual meeting of members and for presenting the program materials to the Board prior thereto for Board approval. The Program Committee may, in its sole discretion, solicit assistance from others, who may or may not be members of the Corporation.

C. Scientific Committee. The Scientific Committee shall be composed of no less than five (5) and no more than nine (9) Ordinary Members. The Scientific Committee shall review proposals for research and related activities according to guidelines developed by the Board, make recommendations to the Board, and present the Board with annual reports and plans concerning the Corporation's research activities. Members of the Scientific Committee will be elected by voting members of the Corporation at the time of the annual meeting. Members of the Scientific Committee will serve two (2) year terms and may serve up to six (6) consecutive years if re-elected. The Scientific Committee will select its own chairperson within ten (10) days of the close of the annual meeting.

D. Education Committee. The Education Committee shall be composed of no less than five (5) and no more than none (9) Ordinary Members. The Education Committee will oversee the educational activities of the Corporation, and review and score the abstracts to be presented at the annual meeting of members. The Education Committee will also present the Board with annual reports and plans concerning the Corporation's educational activities. Members of the Education Committee will be elected by voting members of the Corporation at the time of the annual meeting of the member. Members will serve two (2) year terms and may serve up to six (6) consecutive years if re-elected. The Education Committee will select its own chairperson within ten (10) days of the close of the annual meeting.

E. Disease Steering Committees. The Disease Steering Committees shall oversee the scientific agenda for their respective diseases and will present the Board with annual reports and plans concerning the research and educational activities for those diseases. Members of the Disease Steering Committees will be appointed by the Board, per standard operating procedures as defined by the Board.

Section 2. Committees and Task Forces. The Board may create and appoint members to such other committees and task forces, as it shall deem appropriate in its sole discretion. Such committees and task forces shall have the power and duties designated by the Board, and shall give advice and make recommendations to the Board.

Section 3. Vacancies. Temporary vacancies in the membership of committees may be filled by the Board until the time of an annual meeting and election as specified above.

Section 4. Rules. Each committee and task force may adopt rules for its meetings not inconsistent with the Act, applicable law, the Certificate of Incorporation, these Bylaws or any rules adopted by the Board.

ARTICLE VII
AGENTS

Section 1. Agents. The Board may appoint agents, such as a secretariat (the "Secretariat"), with such powers and to perform such acts and duties on behalf of the Corporation, as the Board may determine from time to time, in its sole discretion.

ARTICLE VIII
MISCELLANEOUS PROVISIONS

Section 1. Fiscal Year. The fiscal year of the Corporation shall be the calendar year.

Section 2. Notice Procedures. Whenever under the provisions of these Bylaws notice is required to be given to a Board member, officer, committee member or member, such notice shall be given in writing by first-class mail or overnight delivery service with postage prepaid to such individual at such individual's address as it appears on the records of the Corporation. Such notice shall be deemed to have been given when deposited in the mail or the delivery service. Alternatively, notice may also be given by facsimile, electronic mail, or hand delivery, and will be deemed given when received.

ARTICLE IX
INDEMNIFICATION

Section 1. Indemnification Generally. Unless otherwise prohibited by the Act or applicable law, the Corporation may indemnify any current or former Board member or officer, and may by resolution of the Board indemnify any agent, against any and all expenses and liabilities incurred by such individual in connection with any claim, action, suit or proceeding to which such individual is made a party by reason of being a Board member, officer or agent. However, there shall be no indemnification in relation to matters as to which such individual shall be adjudged to be guilty of a criminal offense or liable to the Corporation for damages arising out of such individual's own gross negligence in the performance of a duty to the Corporation. Amounts paid in indemnification of expenses and liabilities may include, but shall not be limited to, counsel fees and other fees, costs and disbursements, and judgments, fines, and penalties against, and amounts paid in settlement by, such Board member, officer or agent. The Corporation may advance

HISTIOCYTE SOCIETY GOVERNING BY-LAWS

expenses or, where appropriate, may itself undertake the defense of any officer or agent. However, such officer or agent shall repay such expenses if it should be ultimately determined that such individual is not entitled to indemnification under this Article IX.

Section 2. Insurance. The Board may also authorize the purchase of insurance on behalf of any Board member, officer or other agent, against any liability incurred by such individual which arises out of such individual's status as a Board member, officer or agent, whether or not the Corporation would have the power to indemnify the person against that liability under the law.

ARTICLE X
DISTRIBUTION OF ASSETS UPON DISSOLUTION

Section 1. Distribution of Assets Upon Liquidation. In the event of the liquidation or dissolution of the Corporation, after payment of all debts, all remaining assets shall be distributed only as permitted by the Act, applicable law, and the Certificate of Incorporation.

ARTICLE XI
AMENDMENTS TO BYLAWS

Section 1. Amendments to Bylaws. These Bylaws may be amended (or new bylaws adopted) upon the affirmative vote of a majority of the voting members; provided, that the proposed changes have been approved by the Board, and circulated to the voting members not less than thirty (30) nor more than sixty (60) calendar days prior to such vote to approve same.

NOTES

HISTIOCYTE SOCIETY CONSTITUTION

Article I: Name
The name of the society shall be the "Histiocyte Society". This is a non-profit organization duly registered in the United States of America.

Article II: Aims
1. To improve the state of knowledge of the histiocytic disorders and the welfare of patients with these disorders and their families.
2. To promote, facilitate and carry out research in histiocytic disorders.
3. To facilitate and provide a forum for health professionals for effective communication concerning these aims.
4. To promote education and to educate physicians, nurses, other health professionals, scientists, legislators, and other lay persons in matters related to the histiocytic disorders.
5. To advise lay organizations in educational and other matters concerning histiocytic disorders.
6. To collaborate with other organizations with common aims.

Article III: Amendments and Revisions
1. Changes may be made by an affirmative vote of two-thirds (2/3) of a quorum at a general meeting of the Society, provided proposed changes have been submitted to and approved by the Board and circulated among the members at least one (1) month prior to the general meeting.
2. Proposed changes may originate with any ordinary member of the Society. They should be submitted to the Secretary at least two (2) months prior to the general meeting.
3. Changes properly proposed to the Board will be presented at the next general meeting with the recommendation of the Board.

Article IV: Dissolution
1. Dissolution shall be proposed, processed, and voted on as for amendments and revisions, Article III.
2. In a case of dissolution of the Society, any funds remaining after payment of all outstanding debts will be donated to one or more organizations, with aims and objectives consonant with those of the Society, to be selected by the Board.

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STOCKHOLM 2022
38TH ANNUAL MEETING
HISTIOCYTE SOCIETY
Save the Dates
for the 38th Annual Meeting of the Histiocyte Society
September 18-20, 2022
Stockholm, Sweden
Registration opens in April 2022