Highlights from EHA

Firenze 17.9.2016

GLOBULI ROSSI

Achille Iolascon
M.Domenica Cappellini

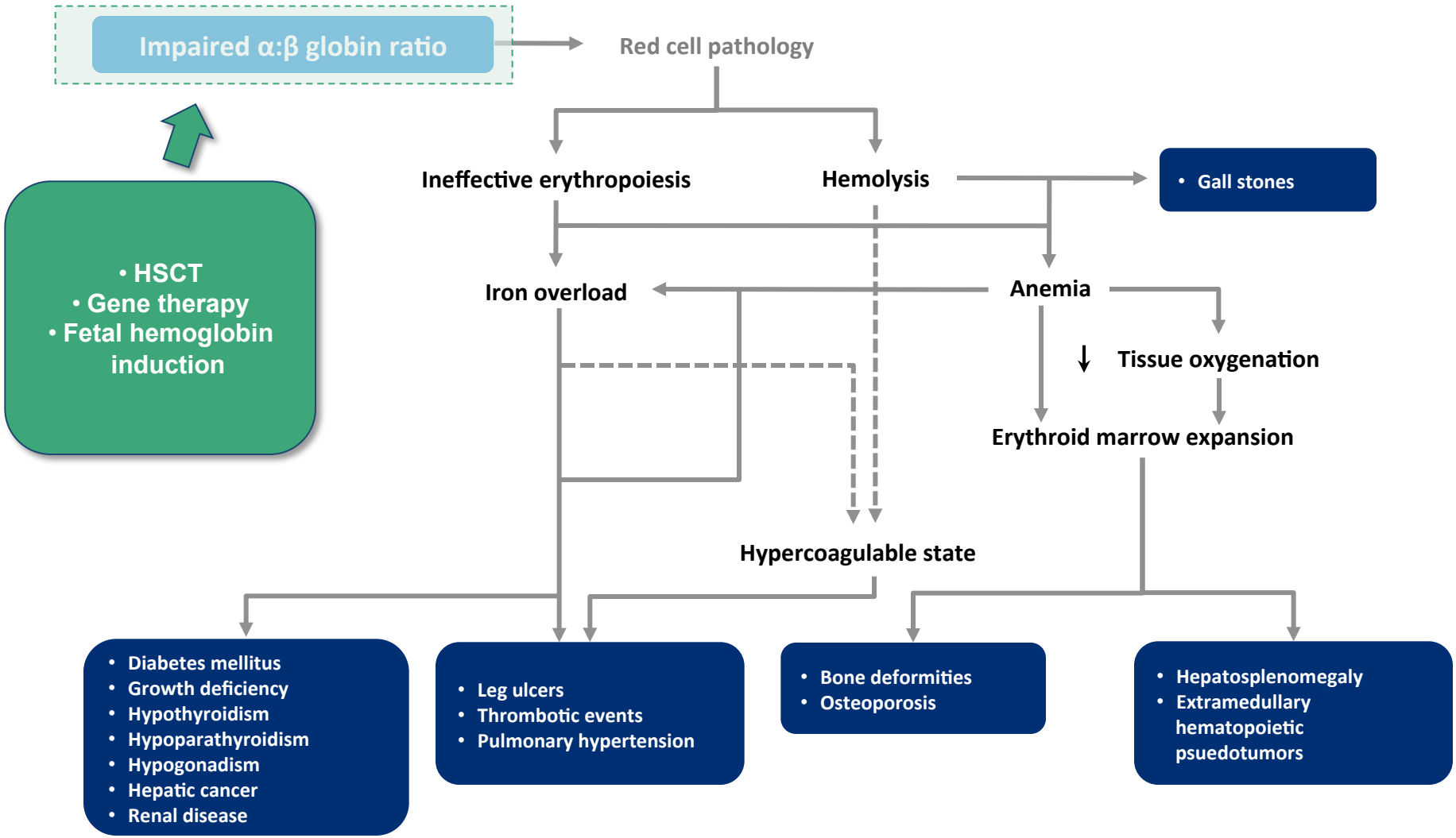
Globuli Rossi

- Nuove tecniche diagnostiche
- Nuove terapie nelle anemie caratterizzate da eritropoiesi
- Nuove terapie della carenza di ferro

What the future treatment for hemoglobinophaties

- Targeting α/β chain imbalance
- Targeting ineffective erythropoiesis
- Targeting iron dysregulation

Targeting α/β chain imbalance



Musallam KM, et al. Haematologica. 2013;98:833-4.

Current BTHAL gene therapy trials

ClinicalTrials.gov
A service of the U.S. National Institutes of Health

A Study Evaluating the Safety and Efficacy of the LentiGlobin® BB305 Drug Product in Beta-Thalassemia Major Subjects

This study is currently recruiting participants. (see Contacts and Locations)

Verified June 2014 by bluebird bio

Sponsor:
bluebird bio

Information provided by (Responsible Party):
bluebird bio

ClinicalTrials.gov Identifier:
NCT01745120

First received: December 6, 2012
Last updated: June 25, 2014
Last verified: June 2014
History of Changes

ß-Thalassemia Major With Autologous CD34+ Hematopoietic Progenitor Cells Transduced With TNS9.3.55 a Lentiviral Vector Encoding the Normal Human ß-Globin Gene

This study is currently recruiting participants. (see Contacts and Locations)

Verified March 2014 by Memorial Sloan-Kettering Cancer Center

Sponsor:
Memorial Sloan-Kettering Cancer Center

Information provided by (Responsible Party):
Memorial Sloan-Kettering Cancer Center

ClinicalTrials.gov Identifier:
NCT01639690

First received: July 11, 2012
Last updated: March 4, 2014
Last verified: March 2014
History of Changes

A phase 1/2 study evaluating safety and efficacy of autologous hematopoietic stem cells genetically modified with GLOBE lentiviral vector encoding for the human globin gene for the treatment of patients affected by transfusion dependent beta thalassemia

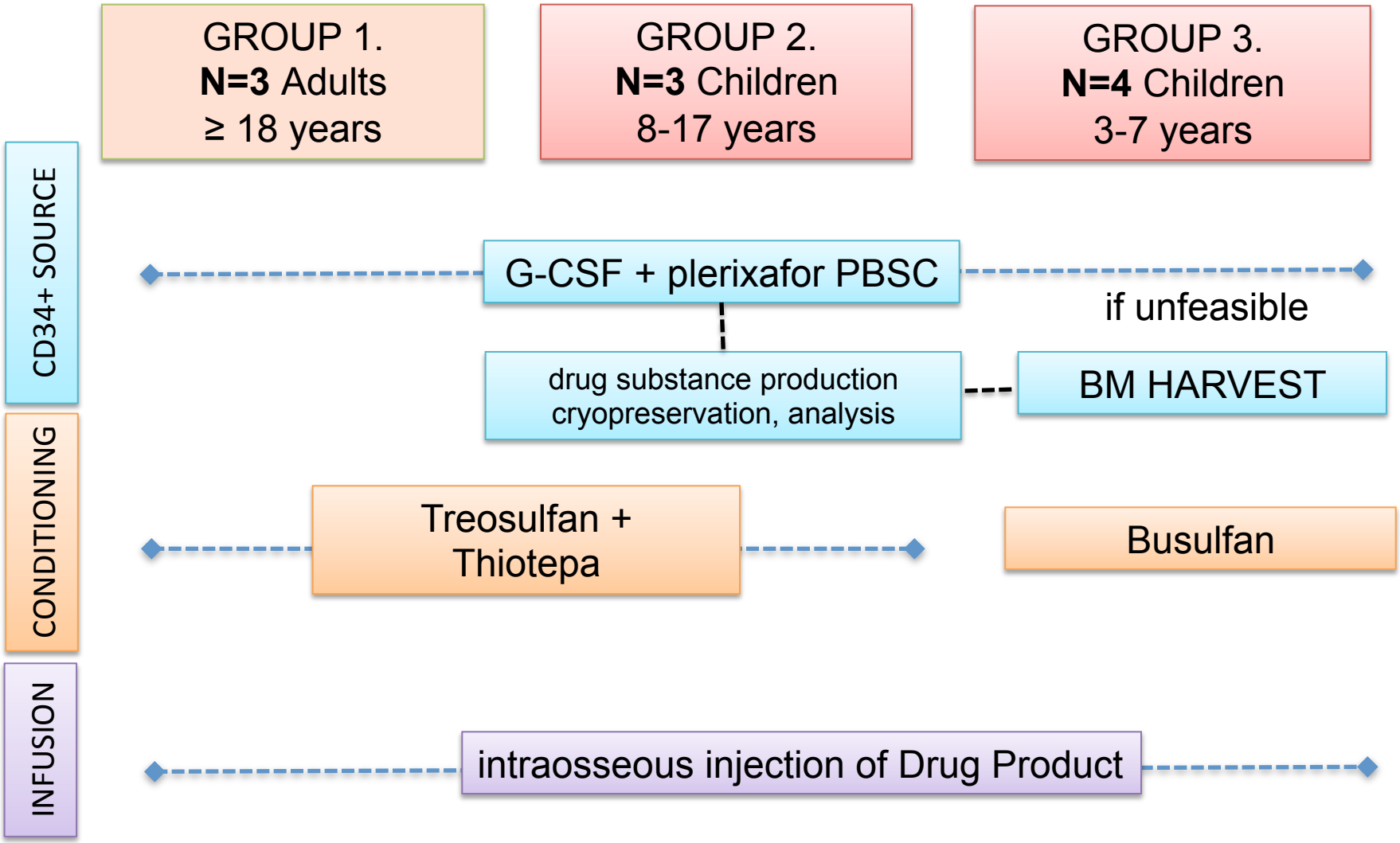
Protocol design

open-label, single center, non randomized, non controlled, prospective, Phase I/II clinical trial

ACRONYM	**TIGET-BTHAL**
PROMOTER	Ospedale San Raffaele (OSR)
FINANCIAL SPONSOR	Fondazione Telethon
BTHAL project leader	Giuliana Ferrari, PhD
PI	Alessandro Aiuti, MD, PhD
Co-PIs	Fabio Ciceri, MD
	Sarah Marktel, MD
	Maria Domenica Cappellini, MD
STATISTICIANS	Stefania Galimberti
	Maria Grazia Valsecchi

TIGET-BTHAL Study Design

N= 10 patients in 3 groups



TIGET-BTHAL
Staggering strategy

Group 1	Group 2	Group 3
ADULTS ≥18 Yrs	CHILDREN 8-17 Yrs	CHILDREN 3-7 Yrs
N=3	**N=3**	**N=4**

The trial will proceed with inclusion of following group if at least 2 patients show all of the following:

1. No SAE related to mobilization
2. No SAE related to intrabone infusion
3. No early SAE related to drug product (within 60 dys)
4. Haematological engraftment achieved by day +60
5. Detection of transduced cells in the bone marrow at +30 or +60

TIGET-BTHAL: *conditioning* RATIONALE

ADULTS & PEDIATRIC > 7 years

Conditioning:
day -5 Thiotepa 6-8 mg
day -4 -2 Treosulfan 14 gr/mq (PK)

Rationale:
- Reduction of organ toxicity
- Effective on reduction of extramedullary haemopoiesis
- Engrafment data from adult allogeneic HSCT protocols in thalassemia (Bernardo, *Blood 2012*);

PEDIATRIC PATIENTS 3-7 years

Conditioning:
day -5 -2 (with PK adjustment)
iv busulfan 120 mg/mq

Rationale:
- Use of conditioning based on results of engraftment obtained from TIGET GT studies (MLD/WAS) and data in pediatric allogeneic transplant (Chiesa, BBMT 2010)

TIGET-BTHAL: *intrabone infusion* RATIONALE

Direct intrabone transplant of unrelated cord-blood cells in acute leukaemia: a phase I/II study

Lancet Oncol. 2008

Francesco Frassoni, Francesca Gualandi, Marina Podestà, Anna Maria Raiola, Adalberto Ibatici, Giovanna Piaggio, Mario Sessarego, Nadia Sessarego, Marco Gobbi, Nicoletta Sacchi, Myriam Labopin, Andrea Bacigalupo

Unrelated Cord Blood Transplantation: Outcomes After Single-Unit Intrabone Injection Compared With Double-Unit Intravenous Injection in Patients With Hematological Malignancies

Vanderson Rocha,[1,2,3,17] Myriam Labopin,[4] Annalisa Ruggeri,[1,2,5] Marina Podestà,[6] Andrea Gallamini,[7] Francesca Bonifazi,[8] Fermin M. Sanchez-Guijo,[9] Montserrat Rovira,[10] Gerard Socie,[11] Ioannis Baltadakis,[12] Mauricette Michallet,[13] Eric Deconinck,[14] Andrea Bacigalupo,[15] Mohamad Mohty,[16] Eliane Gluckman,[1,2] and Francesco Frassoni[6]

- reduces early transplant related adverse events due to the limited number of cells infused
- faster myeloid and PLT recovery
- better engraftment at reduced cell dose?

Thanks to…..

Maria Rosa Lidonnici
Annamaria Aprile
Ylenia Paleari
Claudia Rossi
Giacomo Mandelli
Francesca Tiboni
Emanuela Roselli
Annarita Miccio
Francesco Lotti

Alessandro Aiuti
Luigi Naldini
Eugenio Montini

M. Grazia Roncarolo

OSR BMT UNIT
Fabio Ciceri
Sarah Marktel
Fabio Giglio
Andrea Assanelli
Marta Frittoli
Bernhard Gentner

GLP Test Facility
Patrizia Cristofori
Michela Vezzoli
Paola Albertini
Pamela Rodegher
Vanda Parezanovic

Francesca Sanvito
Claudio Doglioni
Gabriella Passerini
Ferruccio Ceriotti

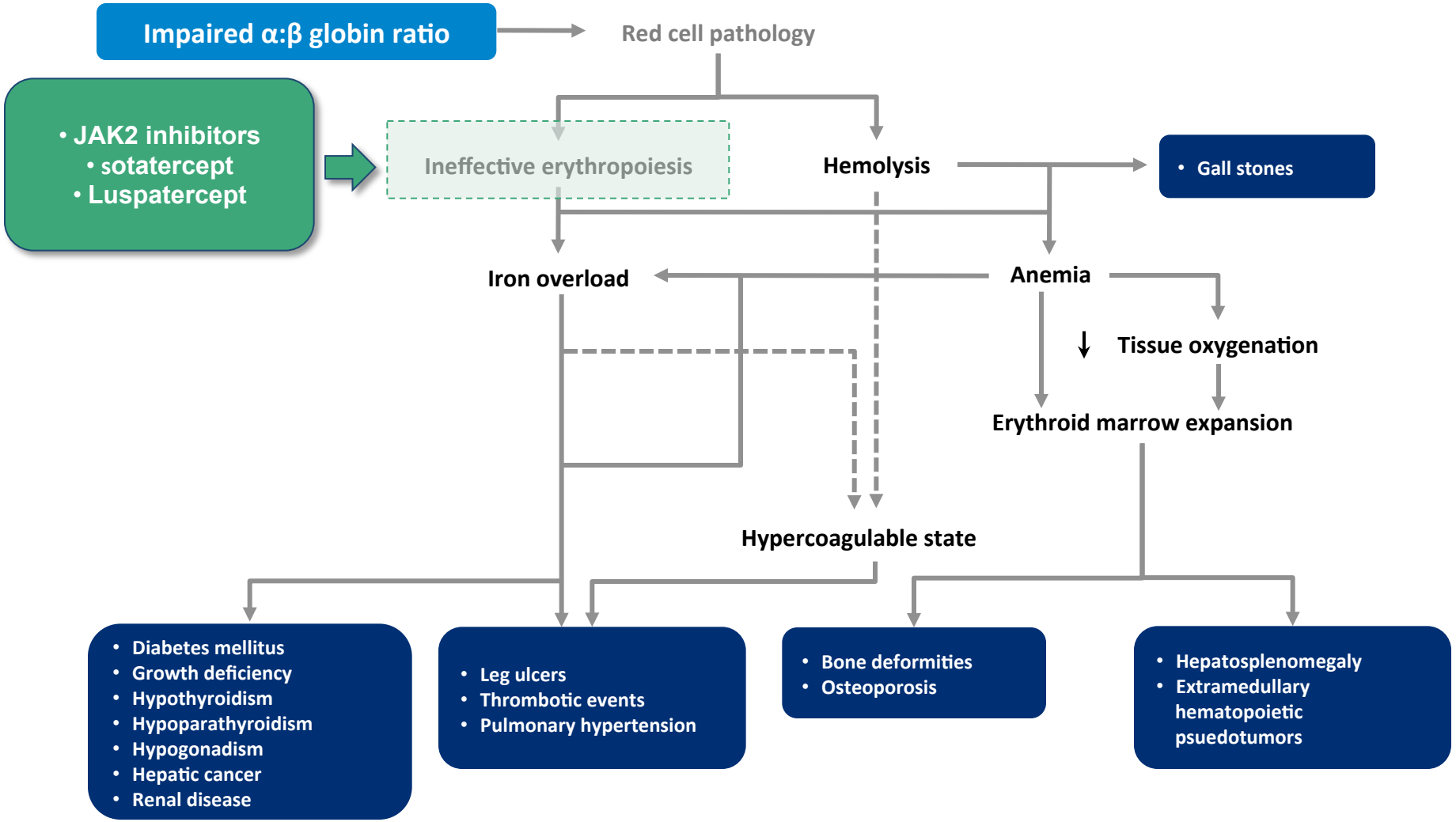
Michela Gabaldo
GSK Alliance Manager

TIGET Clinical Trial Office
M. Bonopane
M. Facchini
E. Mrak

MolMed S.p.A

Nica Cappellini — ***Policlinico, UniMi,Milan***
Chiara Refaldi
Elena Cassinerio
Laura Zanaboni
Marco Andreani — ***IME, Rome***
Guido Lucarelli
Mike Antoniou
King'sCollege,London
Antonio Amato — ***ANMI, Rome***

Targeting ineffective erythropoiesis

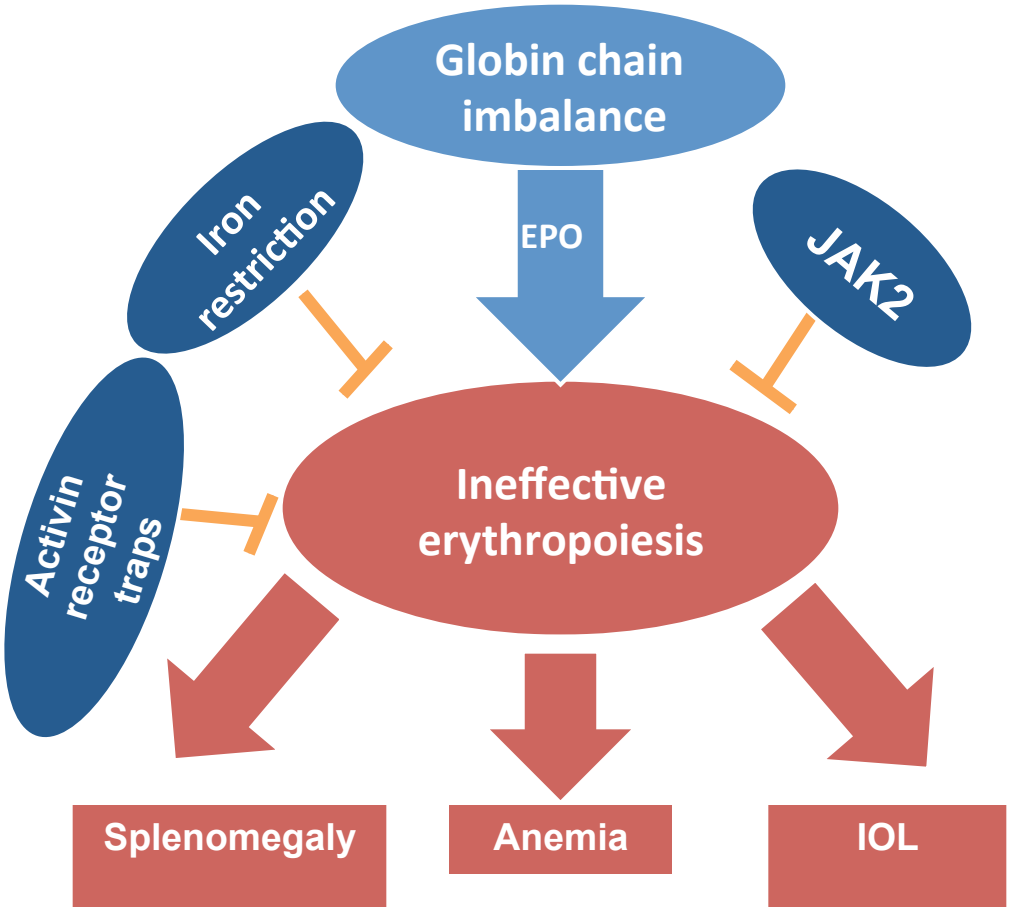


Musallam KM, et al. Haematologica. 2013;98:833-4.

Targeting ineffective erythropoiesis in thalassemia

- **Benefits of reducing IE include:**[1]
  - Improved anemia
  - Reduced spleen and extramedullary expansion
  - Indirect increase in serum hepcidin
  - Improved QoL

- **Approaches:**
  - Hb F promotion
  - Stimulation of proliferation (EPO)
  - JAK2 inhibitors[1]
  - Activin receptor traps
    - Sotatercept and luspatercept[3,4]
  - Iron restriction[1,2]

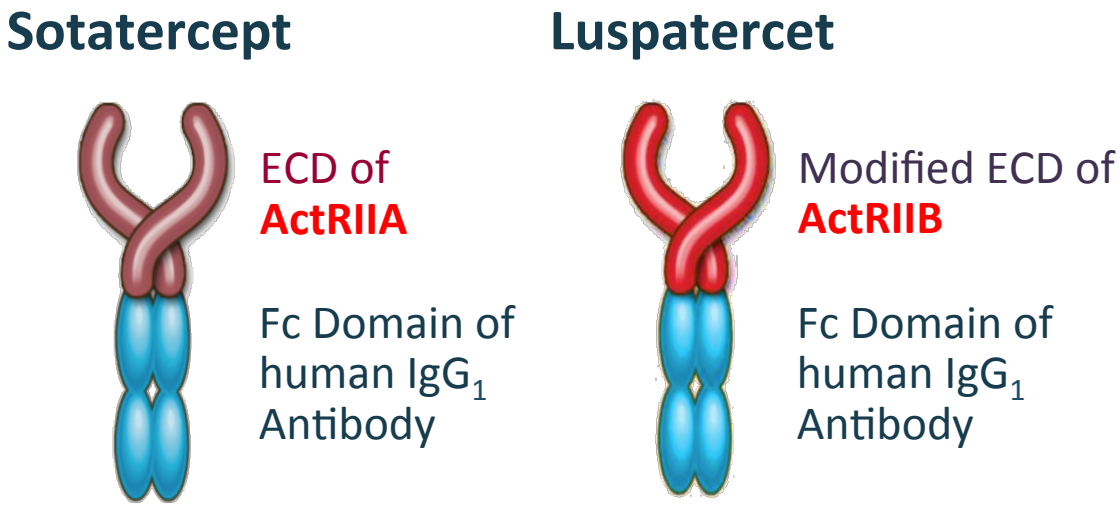


IE, ineffective erythropoiesis;
EPO, erythropoietin;
IOL, iron overload

1. Adapted from Melchiori L *et al. Adv Hematol* 2010;2010:938640; 2. Gardenghi S *et al. J Clin Invest* 2010;120:4466–4477; 3. Suragani R *et al. Blood* 2014;123:3864–3872; 4. Iancu-Rubin C *et al. Exp Hematol* 2013;41:155–166

ACE 011- ACE-536

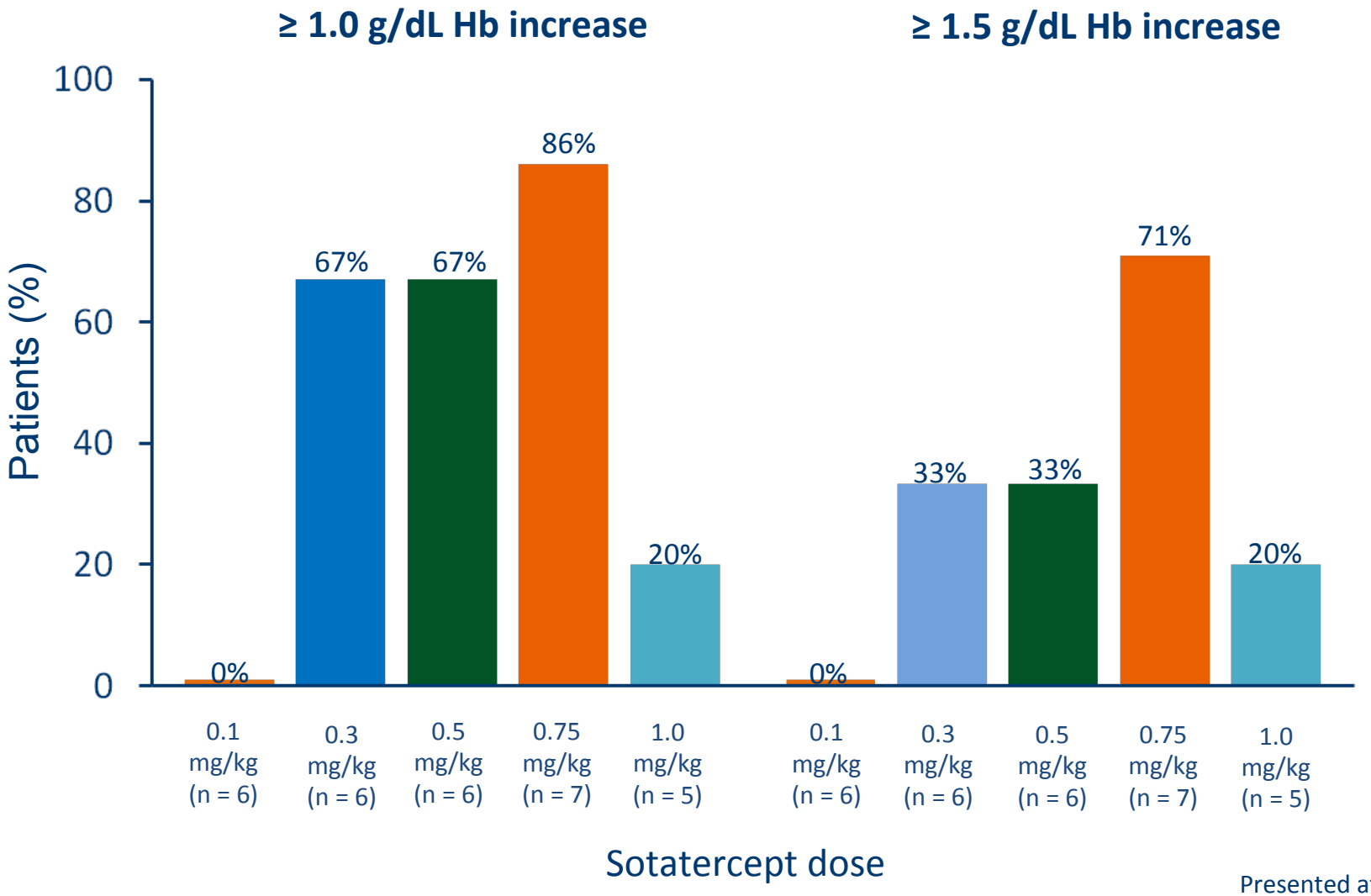
- ACE-011, ACE-536 promote late-stage erythroid differentiation via a mechanism distinct from ESAs *(Suragani R et al., Nature Med 2014)*



- Luspatercept and sotatercept bind to various ligands in the TGF-β superfamily with differing affinities
  - Both bind to GDF11 and inhibit Smad 2,3 signaling

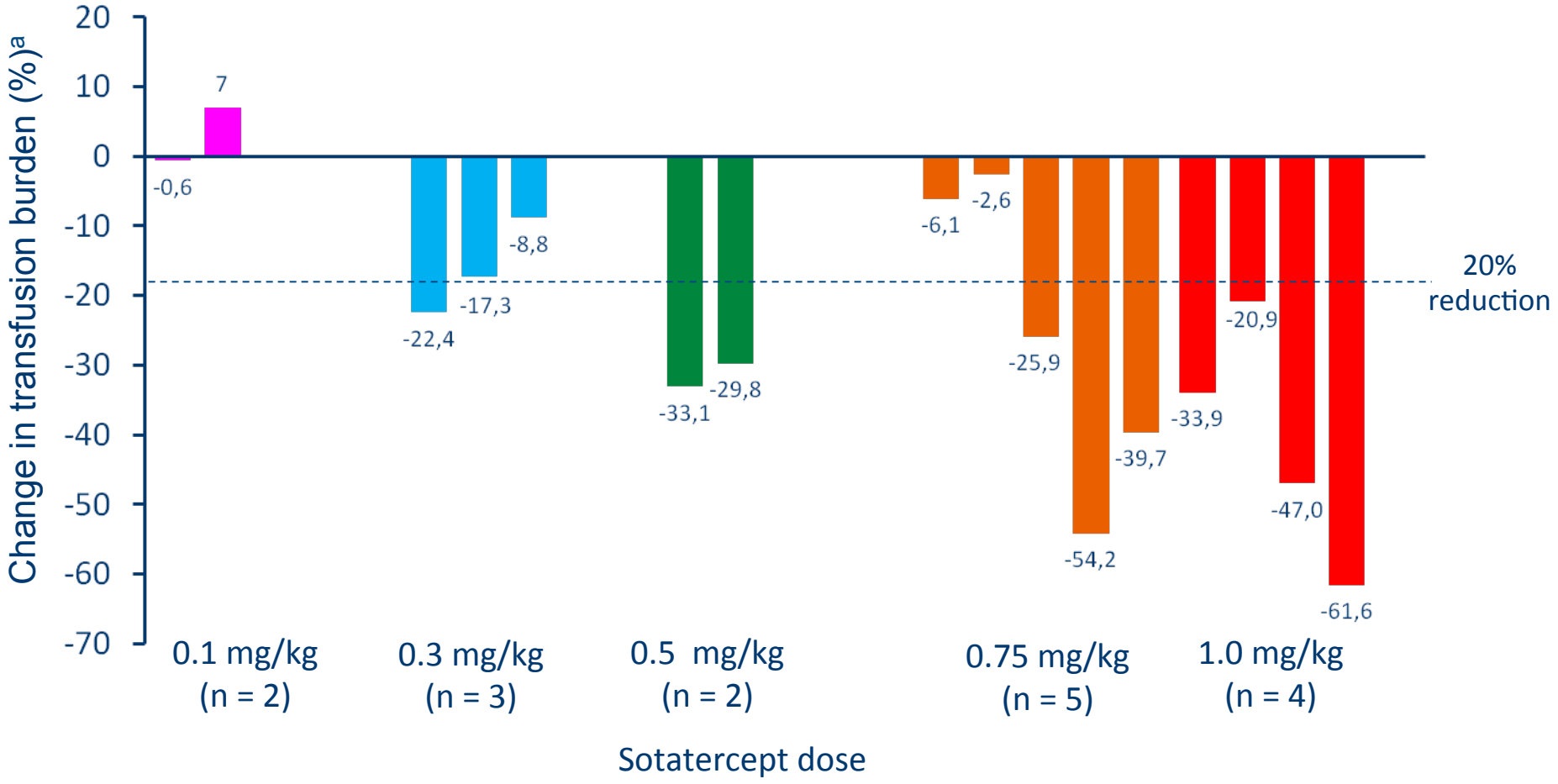
ESA, erythroid stimulating agent; ECD, extracellular domain;
TGF-β, transforming growth factor β; GDF, growth differentiation factor

Results: NTD Thalassemia Patients With Hb Increase Sustained for ≥ 12 Weeks



Presented at EHA 2016.

Results: Reduction in Transfusion Burden for TD β-Thalassemia Patients



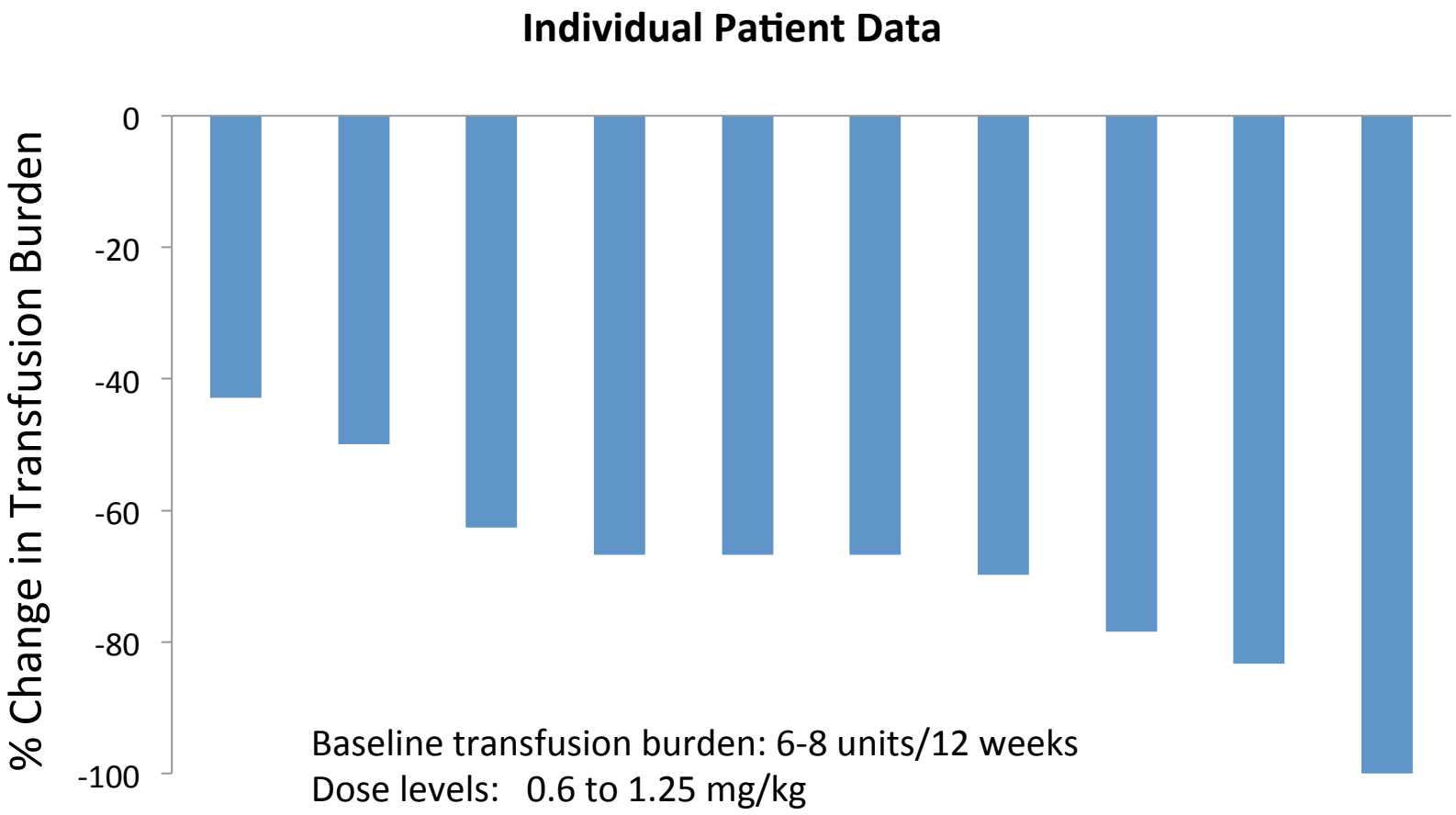
- **Mean transfusion burden reduction among patients treated with sotatercept ≥ 0.5 mg/kg was 32.25%**

[a] Percentage change in transfusion burden (units/168 days) from baseline to on-treatment.

Presented at EHA 2016.

Luspatercept Reduced Transfusion Burden

- 10/14 patients were treated for ≥12 weeks and were evaluable for change in transfusion burden
- All 10 evaluable patients had >40% reduction in transfusion burden over 12 weeks



Data presented at EHA June 2016

The BELIEVE Study

Phase 3 study of luspatercept in beta-thalassemia

BELIEVE

PATIENT POPULATION	Adult beta-thalassemia patients who are regularly transfused
STUDY DESIGN	Randomized, double-blind, placebo-controlled 300 patients, randomized 2:1 (200 luspatercept, 100 placebo)
KEY INCLUSION CRITERIA	Patients who receive 6-20 units of RBCs over past 24 weeks and no transfusion-free period ≥ 35 days (regularly transfused patients)

The BELIEVE Study

Phase 3 study of luspatercept in beta-thalassemia

BELIEVE

- Multicenter (110 sites), multinational (20 countries) study
- 12-week prospective pre-treatment period to calculate baseline transfusion burden

Luspatercept Treatment Leads to Long Term Increases in Hemoglobin and Reductions in Transfusion Burden in Patients with Low or Intermediate-1 Risk Myelodysplastic Syndromes (MDS): Preliminary Results from the Phase 2 PACE-MDS Extension Study

Uwe Platzbecker, MD[1], Aristoteles Giagounidis, MD, PhD[2], Ulrich Germing, MD[3], Katharina Götze, MD[4], Philipp Kiewe, MD[5], Karin Mayer, MD[6], Oliver Ottmann, MD[7], Markus Radsak, MD[8], Thomas Wolff, MD[9], Detlef Haase, MD[10], Monty Hankin[11], Dawn Wilson[11], Xiaosha Zhang[11], Adberrahmane Laadem, MD[12], Matthew L. Sherman, MD[11] and Kenneth M. Attie, MD[11]

[1]Universitätsklinikum Carl Gustav Carus, Dresden, [2]Marien Hospital Düsseldorf, [3]Universitätsklinikum Düsseldorf, [4]Technical University of Munich, [5]Onkologischer Schwerpunkt am Oskar-Helene-Heim, Berlin, [6]University Hospital Bonn, [7]Universitätsklinikum Frankfurt, Goethe Universitaet, Frankfurt/Main, [8]Johannes Gutenberg-Universität, Mainz, [9]OncoResearch Lerchenfeld UG, Hamburg, [10]Universitätsmedizin Göttingen, Germany; [11]Acceleron Pharma, Cambridge, MA, [12]Celgene Corporation, Summit, NJ, USA

Ineffective Erythropoiesis in MDS

- Anemia, a hallmark of MDS, is a significant clinical challenge to treat, particularly after failure of ESAs[1]
- Defects in maturation of erythroid precursors (ineffective erythropoiesis) lead to erythroid hyperplasia and anemia
- Ineffective erythropoiesis is driven by excessive Smad2/3 signaling[2]

ESA: erythropoiesis stimulating agent; EPO: erythropoietin; GDF: growth and differentiating factor; RBC: red blood cell

1. Fenaux P, et al. Blood. 2013;121:4280

2. Zhou L, et al. Blood 2008;112:3434

Luspatercept Lower-Risk MDS Phase 2 Study

- A phase 2, multicenter, open-label, 3-month dose escalation study in adults with lower-risk MDS, followed by a 5-yr extension study
- **Eligibility criteria:**
  - Age ≥ 18 years
  - IPSS Low or Int-1-risk MDS
  - Anemia, defined as high transfusion burden (**HTB**; ≥ 4 RBC units/8 weeks) or low transfusion burden (**LTB**; < 4 RBC units/8 weeks with Hb < 10.0 g/dL)
  - Nonresponsive or refractory to ESA or EPO > 500 U/L
  - No prior AZA or DAC and no current LEN, ESA, G-CSF, GM-CSF
- **Study endpoints include:**
  - Hb increase in LTB patients (≥ 1.5 g/dL over 8 weeks)
  - Reduction in RBC transfusion burden in HTB patients (4 U or 50% reduction over 8 weeks)
  - Achievement of transfusion independence ≥ 8 weeks
  - Time to/duration of HI-E response
  - Safety, PK
  - HI-N, HI-P, HR-QoL (FACT-An), PD and iron biomarkers

Luspatercept MDS Studies A536-03/05

A536-03 Dose Escalation Phase
(3 Months Treatment)

Completed

- **Cohort 1** 0.125 mg/kg (N=3)
- **Cohort 2** 0.25 mg/kg (N=3)
- **Cohort 3** 0.50 mg/kg (N=3)5
- **Cohort 4** 0.75 mg/kg (N=6)
- **Cohort 5** 1.0 mg/kg (N=3)
- **Cohort 6** 1.33 mg/kg (N=6)
- **Cohort 7** 1.75 mg/kg (N=3)

A536-03 Expansion Phase
(3 Months Treatment)

Completed

Expansion Cohort 1
Starting dose 1.0 mg/kg
Individually titrated dose
(N=31)

Active

Expansion Cohort 2
Starting dose 1.0 mg/kg
Individually titrated dose
(N=~50, ongoing)

Active

A536-05 Extension Study (5 years Treatment) (N-32+EC2 Patients)
Starting dose 1.0 mg/kg (treatment interruption patients); Individually titrated dose

Safety Summary – AEs by Dose Level (3 Months)

- Majority of adverse events (AEs) were grade 1 or 2
- One possibly related grade 3 AE: blast cell count increase
- Two possibly related serious AEs: gr 3 myalgia; gr 3 worsening of general condition

Adverse Events (All Grade Regardless of Causality) in ≥ 4 Patients

Preferred Term n (%)	0.125-0.5 mg/kg N=9	0.75-1.75 mg/kg N=49	Overall N=58
Diarrhoea	2 (22%)	5 (10%)	7 (12%)
Myalgia	2 (22%)	5 (10%)	7 (12%)
Nasopharyngitis	1 (11%)	6 (12%)	7 (12%)
Fatigue	0	6 (12%)	6 (10%)
Abdominal pain upper	1 (11%)	4 (8%)	5 (9%)
Bone pain	1 (11%)	4 (8%)	5 (9%)
Bronchitis	0	5 (10%)	5 (9%)
Headache	0	5 (10%)	5 (9%)
Hypertension	0	5 (10%)	5 (9%)
Anaemia	0	4 (8%)	4 (7%)
Muscle spasms	2 (22%)	2 (4%)	4 (7%)

Safety Summary - Patients in Long-Term Extension Study

- No serious or grade 3 or 4 adverse events related to study drug reported during the extension study
- 7/32 (22%) patients discontinued early: patient request (n=3), lack of effect (n=2), progression (n=1), death (n=1)

Adverse events at least possibly related to study drug during the extension study (N=32)

Preferred Term	No. Patients (%)
At least 1 related AE	3 (9.4)
Bone pain	1 (3.1)
Headache	1 (3.1)
Hypotonia	1 (3.1)
Myalgia	1 (3.1)
Nausea	1 (3.1)

Data as of 31 Aug 2015

Conclusions

- Lower risk MDS patients treated with therapeutic doses of luspatercept demonstrated a robust increase in hemoglobin levels and reduced transfusion burden
- Patients with ring sideroblasts (RS+) and SF3B1 mutation were most likely to respond to luspatercept treatment
- Luspatercept was generally safe and well-tolerated
- Increased Hb and transfusion independence up to 1 year (ongoing) were observed in responders
- Responders included patients who were refractory to prior ESA or had serum EPO >200 U/L
- These results supported the initiation of a Phase 3 study of luspatercept in patients with lower-risk MDS (MEDALIST)

Data as of 31 Aug 2015

The MEDALIST Study

Phase 3 Study of Luspatercept in MDS

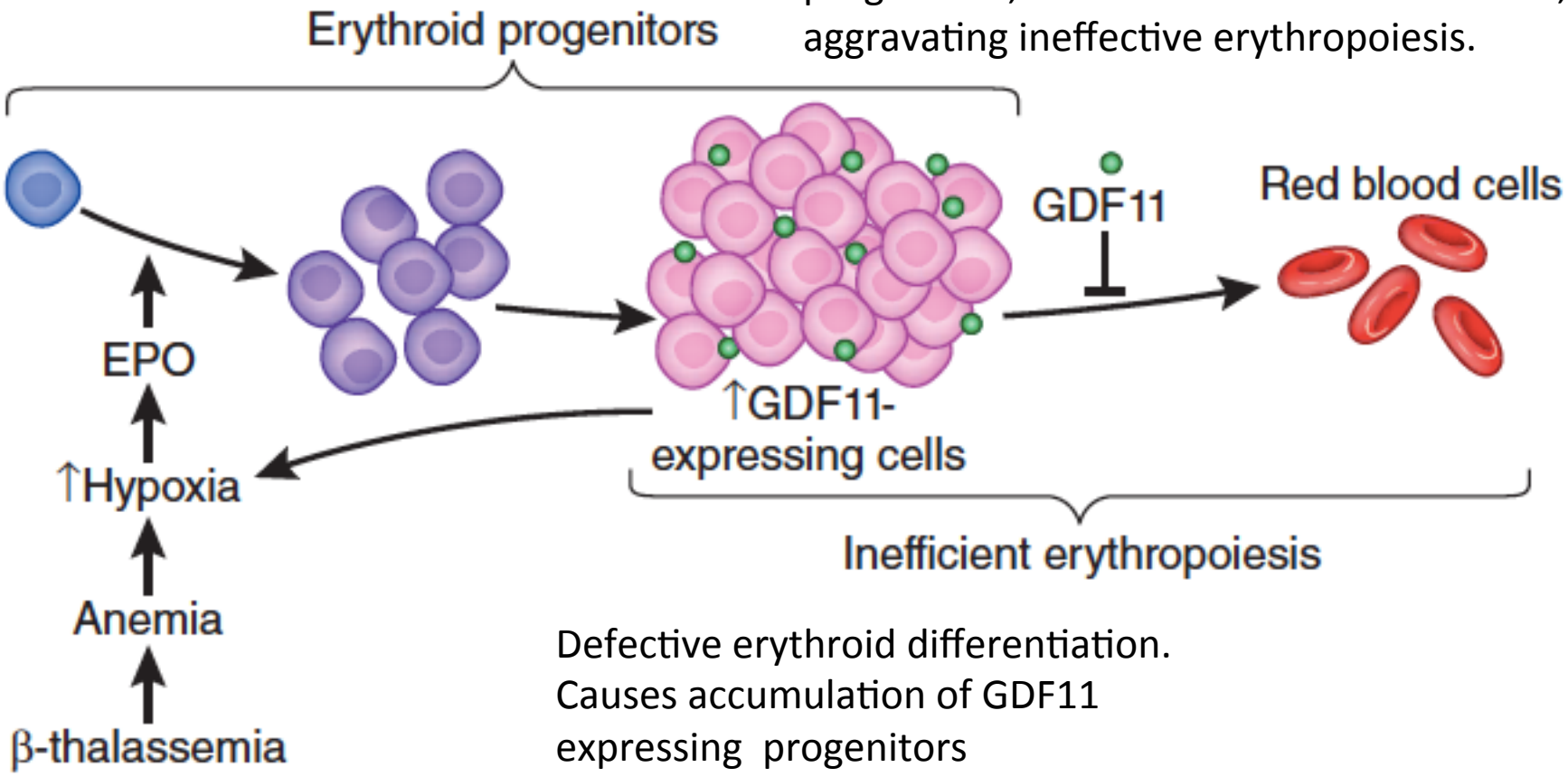
MEDALIST

Patient Population / Study Design	Randomized, double-blind, placebo-controlled study in very low, low or intermediate risk (IPSS-R) MDS patients with ring sideroblasts (RS+) who require RBC transfusion 210 patients randomized 2:1; luspatercept 1 mg/kg SC every 3 weeks, titration up to 1.75 mg/kg possible
Key Inclusion Criteria	Refractory / intolerant to prior ESA *or* EPO > 200 U/L RS+; <5% blasts; no prior HMA or lenalidomide ≥ 2 units RBCs transfused / 8 weeks Excluded: del(5q), secondary MDS
Primary Efficacy Endpoint	Proportion of patients that become RBC-transfusion independent (≥ 8 weeks) during the first 24 weeks

Sponsored by Celgene

Role of GDF11 in IE

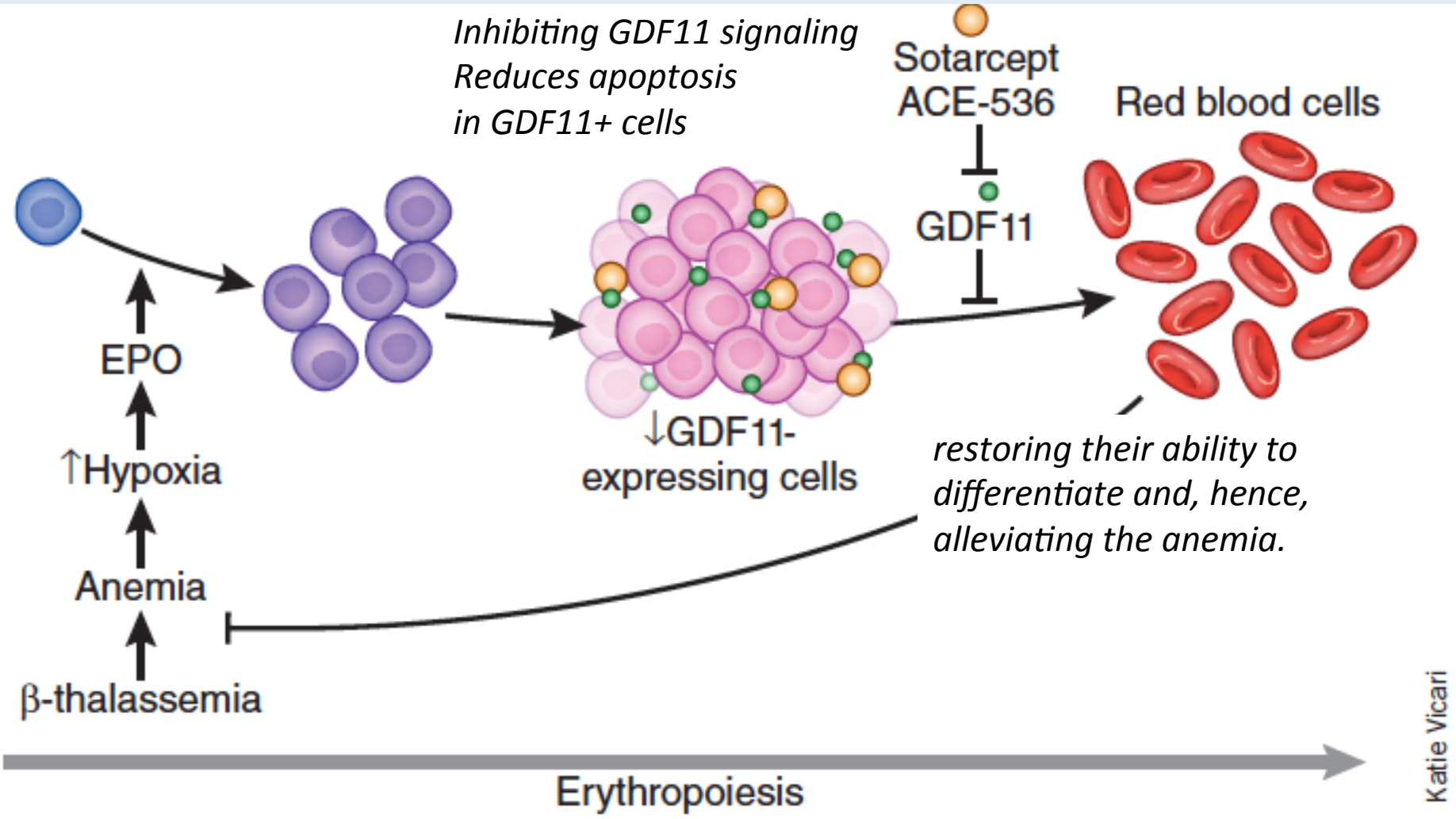
GDF11 maintains the survival of these progenitors, inhibits further differentiation, aggravating ineffective erythropoiesis.



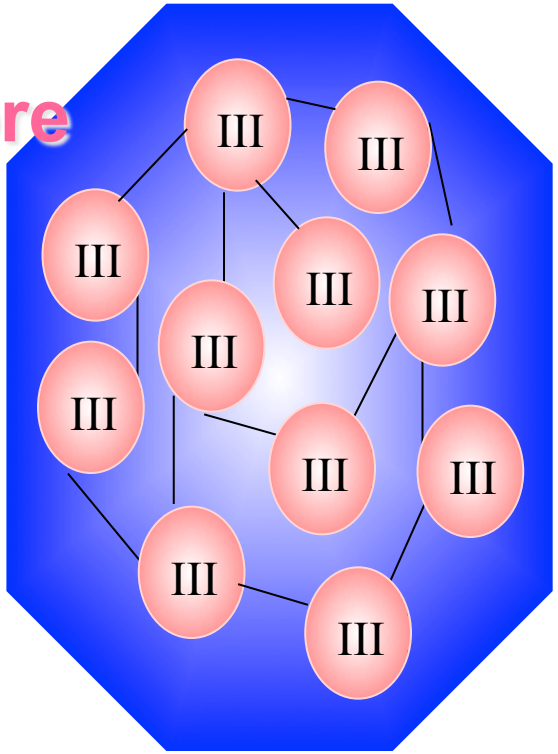
Defective erythroid differentiation.
Causes accumulation of GDF11 expressing progenitors

Paulson. Nature Medicine Editorial , 20 , 4 , 2014
After: Dussiot *et al*. Nat Med, 4, 398-407, 2014
Suragani *et al*. Nat Med, 4, 408-14 2014

Role of ACE-11 and ACE-536 GDF11 in IE



Advances on i.v. iron



1947: Fe-Saccharide

↓

1954: Fe-Dextran (HMW)

↓

1999: Fe-Gluconate

↓

2000: Fe-Sucrose

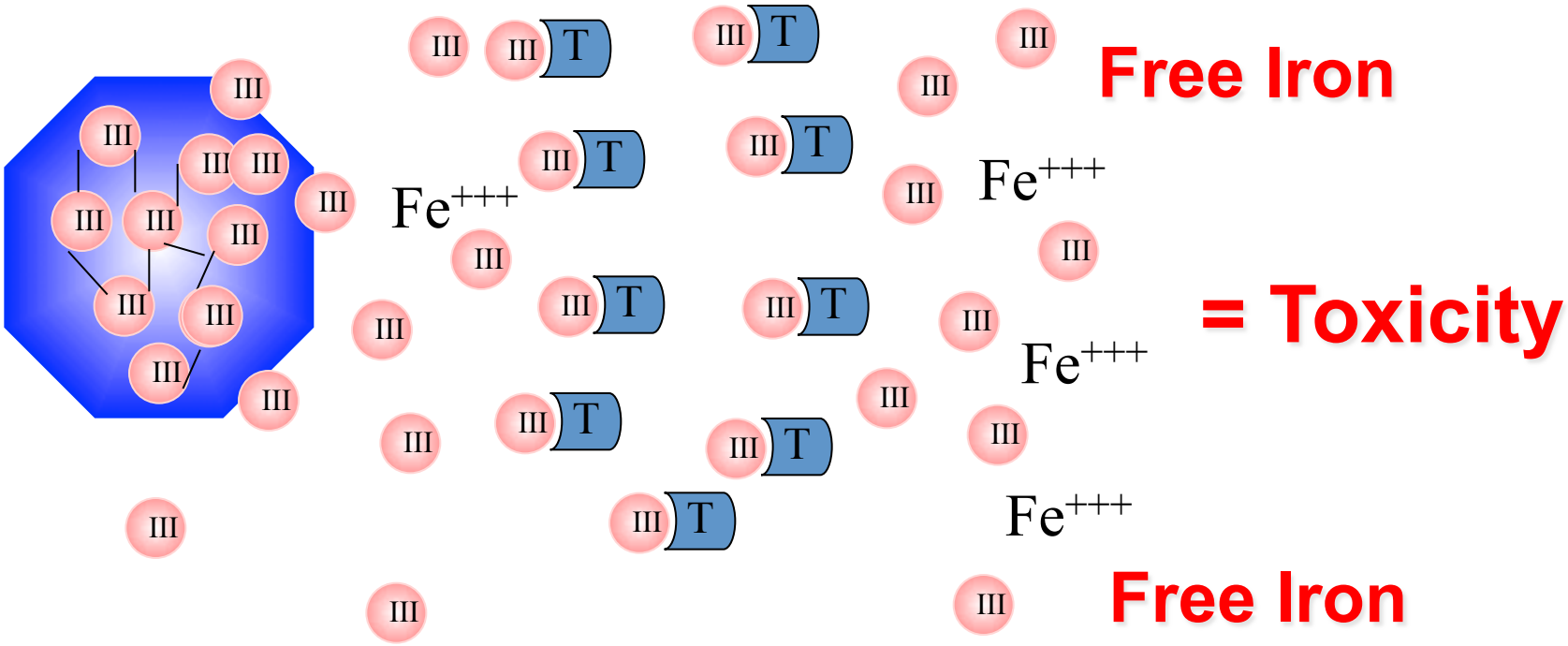
↓

2009: Ferumoxytol

↓

2011: Fe-Carboxymaltose

Iron toxicity (limiting dose) depends on stability of the iron/carbohydrate complex



Features of new Fe(III)-hydroxide carbohydrate complexes

- highly stable complexes, do not release large amounts of ionic iron
- "Similar" to ferritin
- Macrophage uptake