Sickle cell disease and hyperhemolysis: epidemiology, risk factors and new therapeutic approaches in France

WORLD APHERESIS ASSOCIATION AND FRENCH SOCIETY FOR HEMAPHERESIS
April 27-29, 2016, PARIS-FRANCE

France Pirenne, EFS, INSERM, Henri Mondor Hospital, Créteil, France
Sickle Cell Disease

- Sickle Cell Disease (SCD): most frequent inherited disorder in France (20,000 patients): mutation of the globin $\beta$ gene inducing an abnormal Hb, the HbS

- Polymerisation of HbS:
  - Fragility of the membrane: chronic hemolysis
  - Adhesion and low deformability of RBCs: vaso occlusion crisis (VOC)
  - Infections because of asplenia

- Transfusion remains the main treatment of the disease
  - For treatment: VOC, Acute Chest Syndrom, priapism...
  - For prevention
    - Pregnancy, surgery, high symptomatology...
    - Vasculopathy in children +++
Red cell exchange

30% ↑ of HbA

Simple transfusion
Hemolytic Transfusion Reaction in SCD: the most harmful effect of transfusion and alloimmunization

- Frequently delayed: 5 to 10 days following transfusion
  - Delayed hemolytic transfusion reaction (DHTR)
- Destruction of both transfused and autologous RBCs
  - Hyperhemolysis
- Profound reticulopenia: worsen the anemia
- Clinical presentation resembling a vaso occlusive crisis
  - Explains why DHTR is under recognized
- Additional transfusions exacerbate the anemia
- Specific Immuno hematological characteristics
  - Main cause: allo immunization
  - In some cases: no detectable antibodies...
Diagnosis of DHTR

• Retrospective analysis of cases at the Reference Center of Sickle Cell Disease in the Henri Mondor Hospital, during 11 years

  – 99 cases in 69 patients
  – 6 deaths

Habibi, Pirenne, in preparation
Transfusion

DHTR diagnosis

11 Days +/-6.5 after transfusion

D0

D30

Dark urines: 94%
Pain: 89% (symptoms of VOC)
Fever: 63%
Symptoms related to anemia: 44%
# DHTR : Biological parameters

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<td><strong>Biology at Emergency room (n=66)</strong></td>
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<td>Delay between TF and readmission, days</td>
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<td>Day of LDH max</td>
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<td>Lowest reticulocyt count, /mm³</td>
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<td>Highest leucocytes</td>
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### Characteristics of dead patients

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<tr>
<th>Patient</th>
<th>TF indication</th>
<th>Previous DHTR</th>
<th>DHTTR Antibody</th>
<th>Hb post TF</th>
<th>ΔHb</th>
<th>Nadir Hb</th>
<th>Day of nadir Hb</th>
<th>LDH max</th>
<th>Day of LDH max</th>
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<tr>
<td>N 1</td>
<td>VOC</td>
<td>No</td>
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<td>8.1</td>
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<td>Yes</td>
<td>antiHI</td>
<td>8.2</td>
<td>3.2</td>
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<td>N 3</td>
<td>Delivery</td>
<td>Yes</td>
<td>none</td>
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<td>2.2</td>
<td>6</td>
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<td>Leg ulcer</td>
<td>Yes</td>
<td>S, Lea</td>
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<td>Surgery</td>
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<td>6.7</td>
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Mean ± SD  

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<tr>
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<th>Mean ± SD</th>
<th>Median</th>
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<tbody>
<tr>
<td>Hb post TF</td>
<td>9.98 ± 1.64</td>
<td>10.9</td>
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<tr>
<td>ΔHb</td>
<td>5.1 ± 2.3</td>
<td>5.2</td>
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<tr>
<td>Nadir Hb</td>
<td>4.4 ± 1</td>
<td>4.5</td>
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<tr>
<td>Day of nadir Hb</td>
<td>10.5 ± 3.3</td>
<td>9.5</td>
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<tr>
<td>LDH max</td>
<td>4275 ± 2002</td>
<td>5118</td>
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<tr>
<td>Day of LDH max</td>
<td>10 ± 2</td>
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</tbody>
</table>

EFS
Diagnostic nomogram of DHTR

\[
\text{(PT}\ Hb \text{ g/dl x %HbA)} - \text{(TFT}\ Hb \text{ g/dl x %HbA)} \\
\text{(TFT Hb g/dl x %HbA)}
\]

* Post transfusion: Between 0 to 48H
** Time from Transfusion: any time when DHTR is suspected

Mekontso Dessap et al, Medicine, 2016, in revision
Immuno-hematological characteristics of the 99 cases

- **group 1**: DHTR in non-immunized patients who developed antibodies;
- **group 2**: DHTR in previously immunized patients who developed newly formed antibodies;
- **group 3**: DHTR in non-previously immunized patients who did not develop antibodies, and
- **group 4**: DHTR in previously immunized patients who did not develop newly formed antibodies.
Immuno-hematological characteristics of the 99 cases

Antibodies appeared in 62% of the cases

Group 1: DHTR in non-immunized patients who developed antibodies;
Group 2: DHTR in previously immunized patients who developed newly formed antibodies;
Group 3: DHTR in non-previously immunized patients who did not develop antibodies, and
Group 4: DHTR in previously immunized patients who did not develop newly formed antibodies.
Immuno-hematological characteristics of the 99 cases

Antibodies appeared in 62% of the cases

group 1: DHTR in non-immunized patients who developed antibodies;
group 2: DHTR in previously immunized patients who developed newly formed antibodies;
group 3: DHTR in non-previously immunized patients who did not develop antibodies, and
group 4: DHTR in previously immunized patients who did not develop newly formed antibodies.

- antibodies known to be pathogenic (RH, FY, JK, MNS, high frequency..)
- antibodies not known as pathogenic (AUS, auto)
Immuno-hematological characteristics of the 99 cases

In 37% of the cases, newly formed antibodies were not detected.

- group 1: DHTR in non-immunized patients who developed antibodies;
- group 2: DHTR in previously immunized patients who developed newly formed antibodies;
- group 3: DHTR in non-previously immunized patients who did not develop antibodies, and
- group 4: DHTR in previously immunized patients who did not develop newly formed antibodies.
A severity score was calculated based on the number of severe complications.

There is no statistical correlation between the severity score and:
- The immuno-hematological profile
- The type of antibodies that appear in groups 1 and 2
Prospective monocenter observational study: between November 2011 and June 2014 in the Henri Mondor hospital, Creteil

Patients included:
- Age 18 and over with an intention to transfuse followed by effective transfusion in the hospital
- DHTR confirmed by: rapid disappearance of HbA associated with 2 to 3 of the following criteria a few days to 3 weeks after transfusion
  - Symptoms of VOC
  - Dark urines
  - Worsening of anemia
  - Increase in LDH level

Incidence of DHTR

Narbey, Habibi, Pirenne, in preparation
Incidence of DHTR

722 transfusion episodes (TE)

Punctual transfusion
n=266 (195 patients)

15 DHTR (for 15 TE and 15 patients)

Chronic transfusion program
N=456 (157 patients)

No DHTR
Incidence and risks

• There is a significant higher risk to develop DHTR when patients are transfused punctually as compared to patients transfused on a transfusion program (p<0.001)

• Incidence of DHTR when patients are transfused punctually:
  – DHTR developed after 5.6% of the TE
  – 7.6% of the patients developed DHTR during the studied period

• Risk factors
  – punctual transfusion
  AND
  – history of DHTR, history of immunization, patients with few transfusions, pregnancy
Prevention and treatment of DHTR in SCD

• In order to be efficient: action at 3 levels

1- The trigger of the reaction
2- The mechanism of destruction
3- The consequences of the hemolysis
Prevention and treatment of DHTR in SCD

• In order to be efficient: action at 3 levels

  1- The trigger of the reaction

    • Allo immunization

      Prevention
      – RH/K matched RBCs in non immunized patients
      – Extended matched RBCs in immunized patients

    • But not only ...
      – many cases with poorly significant antibodies (auto, AUS) and 30% of cases without detectable antibodies
      – Trigger unknown ....
8 patients highly immunized with history of DHTR were preventively treated with RITUXIMAB before a new transfusion

For all patients: No appearance of new formed antibodies,
5 patients: non eventful clinical course
3 patients: mild DHTR
Rituximab can at least prevent recurrence of newly formed antibodies in high responders patients and potentially minimizes the risk of severe DHTR.

Confirmation that DHTR is complex in SCD and does not only rely on antibody mediated hemolysis.

Rituximab should be considered when a new transfusion seems inevitable in SCD patients with a previous history of severe DHTR linked to immunization.

Caution is absolutely necessary in SCD because of the higher risk of infectious diseases.
Prevention and treatment of DHTR in SCD

- In order to be efficient: action at 3 levels

2- The mechanism of destruction

Antibody mediated hemolysis

- action on effector cells (macrophages, NK) and complement

Complement and effector cells involved without detectable antibodies?
Steroids

+  
-  

- Frequently used
- Synergistic with IgIV
- Low cost
- VOC and ACS recurrence
Immunoglobulins

+  
  -

• Frequently used
• Good benefit/risk ratio
• Animal models

• Viscosity (but low Hb)
• Kidney

• Not immunosuppressing

*Delayed hemolytic transfusion reaction in children with sickle cell disease*

*Haematologica 2011*
Other treatments to act on destruction

• Action of RITUXIMAB on destruction?
  – Bachmeyer, 2009; Delmonte, 2013: Rituximab was given to treat DHTR associated with other treatments

• Eculizumab
  – To stop complement activation and the cascade of events produced by the release of RBC content
Prevention and treatment of DHTR in SCD

• In order to be efficient: action at 3 levels

3- The consequences of the hemolysis
In some cases, the evolution of DHTR can be disastrous:

- Frequently linked with intra vascular hemolysis
- Within a few hours: multi organ failure

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Extracellular hemin crisis triggers acute chest syndrome in sickle mice

Samit Ghosh, Olufolake Adetoro Adisa, Prasanthi Chappa, Fang Tan, Kesmic Ann Jackson, David Robert Archer, and Solomon Fifi Ofori-Acquah

JCI 2014
Intra vascular hemolysis

Free hemoglobin → Hemolysis → Haptoglobin, Hemopexin

Heme oxygenase

NADPH reductase

Biliverdin

CO + Fe^{2+}

NADP^{+} + H_{2}O

Anti inflammatory
Anti oxidant
Intra vascular hemolysis

Hemolysis

Free hemoglobin → heme

Haptoglobin
Hemopexin

NADPH

NADPH reductase

Heme oxygenase

Oxygen

Biliverdin

CO + Fe^{2+}

NADP^+ + H_2O

Anti inflammatory
Antioxidant
Plasma Exchange

+ free-hemoglobin

- Not easy to perform
- Low Hb level
- Faisability

Successful treatment of recurrent hyperhemolysis syndrome with immunosuppression and plasma-to-red blood cell exchange transfusion

Transfusion 2014

Erik J. Uhlmann, Shalini Shenoy, and Laurence T. Goodnough
Other potential treatments to inhibit toxicity of RBC content

- Haptoglobin
- Hemopexin
Rationale to use Eculizumab

In the most severe cases of DHTR, there is a need of a treatment that stop hemolysis and the disastrous cascade of events that are induced by free heme and free hemoglobin as in SCD patients, haptoglobin and hemopexin are frequently overwhelmed because of chronic hemolysis.
Rationale to use Eculizumab

Complement activation likely involved in post-transfusion hyperhemolysis in SCD

First hit: yes when antibodies are produced
First hit: ?? low titer of antibodies against RBCs

Second hit: hypothesis: free heme
The use of Eculizumab in severe DHTR

Life-threatening delayed hyperhemolytic transfusion reaction in a patient with sickle cell disease: effective treatment with eculizumab followed by rituximab

Boonyasampant et al. Volume 00, April 2015 Transfusion

Eculizumab salvage therapy for delayed hemolysis transfusion reaction in sickle cell disease patients

Guillaume Dumas,1 Anoosha Habibi,1-3 Thierry Onimus,4 Jean-Claude Merle,5 Keyvan Razazi,6 Armand Mekontso Dessap,4 Frederic Galactéros,2 Marc Michel,1 Veronique Frémeaux Bacchi,7 France Noizat Pirenne,3,8 and Pablo Bartolucci1,3

Blood, 25 February 2016 • Volume 127, Number 8
TREATMENT Of DHTR
Primum non nocere

AVOID
Retransfusion
only life-threatening complications should authorize a new transfusion
All known antibodies in every patient’s history must be considered transfusion matching should be extended to the main immunogenic blood groups: FY, JK and MNS

Erythropoietin

![Graph showing reticulocytes G/L before and after EPO treatment]
Management of DHTR and outcome (n = 99)

Supportive Treatment of VOC
- Hydration
- Oxygenation
- Analgesia
N = 53 (53.5%)

No Additional Transfusion
N = 34

Additional transfusion
N = 19
- 3 Deaths

Supportive Treatment of VOC
+ EPO
+/- IVIG, Eculizumab, Corticosteroids, Rituximab
N = 46 (46.5%)

No Additional Transfusion
N = 30
- 1 Death

Additional Transfusion
N = 16
- 2 Deaths
Conclusions

- DHTR can be life threatening and under recognized
- Incidence is high when patients are punctually transfused
- Mechanism remains enigmatic in some cases
- New transfusion should be avoided
- Treatment has to be decided early in case of severe hyperhemolysis, but DHTR can resolve without specific treatment
- Prevention of immunization is necessary but not sufficient