

Blood Transfusion Symposium 2008: The Future of Blood Transfusion Medicine

**Sticky or Sugar-Free?**  
*The Role of ABO in the World-Wide Scourge of Malaria.*

Christine Cserti-Gazdewich, MD FRCPC  
 Assistant Professor, University of Toronto  
 Departments of Laboratory Hematology (Pathology) & Clinical Hematology (Medicine)  
 Blood Transfusion Laboratory, University Health Network  
 Toronto, Ontario, Canada

Saturday November 8<sup>th</sup> 2008 13:05 – 13:40  
 Four Points Sheraton London Hotel and Suites  
 1150 Wellington Rd S, London, Ontario

## Objectives

- learn how *Plasmodium falciparum* malaria-infected red blood cells adhere to host blood group antigens to cause pathology
- review laboratory and clinical evidence for the importance of ABO in malaria
- understand how this information could be useful for future treatment and prevention strategies

## Support for Research & Clinical Care in Kampala, Uganda

- Grant support:
  - International Society of Blood Transfusion (ISBT) Foundation
  - National Blood Foundation (NBF)
  - Dean's Fund, University of Toronto
  - Luick Fund, Mass General Hospital
  - Division of Global Health and Human Rights, Harvard University
- Corporate medical/humanitarian aid & research support:
  - BD Biosciences
  - Whatman, Streck, Fisher
  - Nonin, Sonosite
  - Heart to Heart Foundation

**Cytoadherence in Pediatric Malaria Study**

## Malaria Today

*"Far from being a problem solved, malaria affects more people on earth than ever before... Today, twice as many children die of malaria than when their parents were children."*  
 - Michael Finkel, National Geographic

Push for New Tactics as War on Malaria Falters

## Where *P falciparum* malaria reigns, so too does the Grim Reaper of children

Child Mortality

<http://www-personal.umich.edu/~mej/cartograms/>

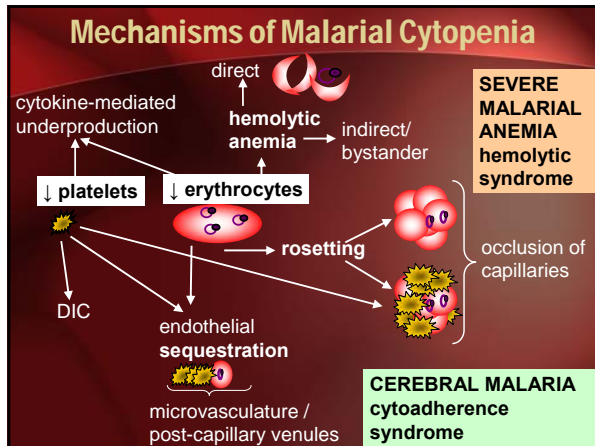
- 1 - 3 million deaths / year
- pediatric case fatality rate: 20x adults
- the death of a child every 15 seconds
- the strongest force of natural selection on the human genome

## Severe Malaria – WHO-defined

**CM SMA LA**

- Depressed CNS, coma, repeated seizures
- Severe anemia (Hct < 15% ; Hgb < 5.0 g/dL)
- Acidosis (lactate > 5 mmol/L)
- Renal failure (Cr > 3 mg/dl)
- Pulmonary edema: ARDS
- Circulatory shock
- DIC; Spontaneous bleeding
- Hemoglobinuria
- Jaundice
- Hypoglycemia (blood glucose < 40 mg/dL or 2.2 mM)
- Parasitemia > 5%

*Even with treatment, mortality of severe malaria: 15-20%*  
*Symptom onset to death: 2.8 d*  
 Greenwood, 1987



### Transfusion for Severe Malarial Anemia (SMA)

- in endemic areas where blood transfusion is available, SMA accounts for >85% of transfusion indications
- although 6,000,000 transfusions are given in sub-Saharan Africa yearly, the need is 18 million
- lack of blood availability = many preventable deaths due to malaria

Lozano M et al. *Global Perspectives in Transfusion Medicine*. aABB Press ©2006.

### Lactic Acidosis & Ischemic Vasoocclusion:

**sequestration**

<http://www.yamagiku.co.jp/pathology/case/case206.htm>

**rosetting**  
= intraluminal multicellular aggregates

Pain, PNAS 2001

**rigidification**

3-7 μm diameter  
If rigid...: 7.5 μm diameter

www.nlm.nih.gov/washingtondc/museum  
Glenister, Blood 2002

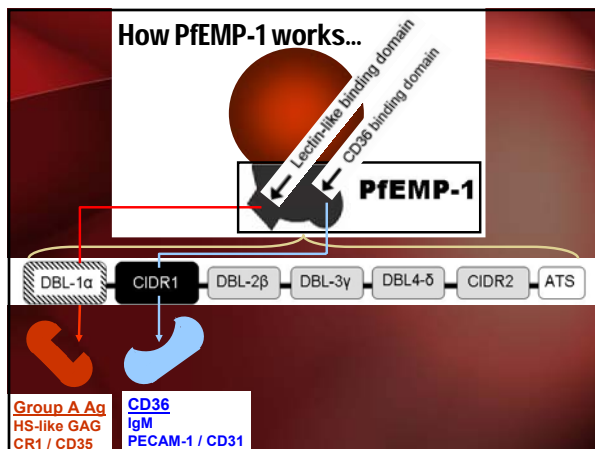
### Cytoadhesion between Hidden Parasite & Host

**Parasite-driven**  
PFEMP-1 = adhesive "knobs" on surface of infected red cell

**Host-driven**  
parasite exploits adhesion ligands

- endothelial (→ sequestration)
  - CD36
  - ICAM-1
- red cell (→ RBC rosettes)
  - group A,B antigens
  - CR1 (Knops) antigens
- platelet (→ platelet rosettes)
  - platelet glycoprotein IV (CD36)

Rug et al, Blood 2006



### ABO Glycosyltransferase Genes... How They Come to Be

Gene	1	2	3	4	5	6	7	
Group A (wild type)						526 (176) Arg	703 (235) Gly	796 (266) Leu, 803 (268) Gly, 1062 (354)
Group B						526 (176) Gly	703 (235) Ser	796 (266) Met, 803 (268) Ala, 1062 (354)
Group O (mutant)						261 del G	351 (117)	

## Explanations for ABO

**BLOOD GROUPS RESEMBLE GERM ANTIGENS**

- **Plague (*Pasteurella pestis*):**  
H(O)-like antigen
- **Smallpox (*Variola v.*):**  
A-like antigen
- **Malaria (*Plasmodium spp.*):**  
A-like antigen

*Mechanistic theories: all for bug-ACQUISITION, not infection-SEVERITY*

- tolerance is bad &
- intolerance = neutralizing anti-A, B IgG & IgM → block germ invasion

## AB(O) Targets Are Expressed Everywhere

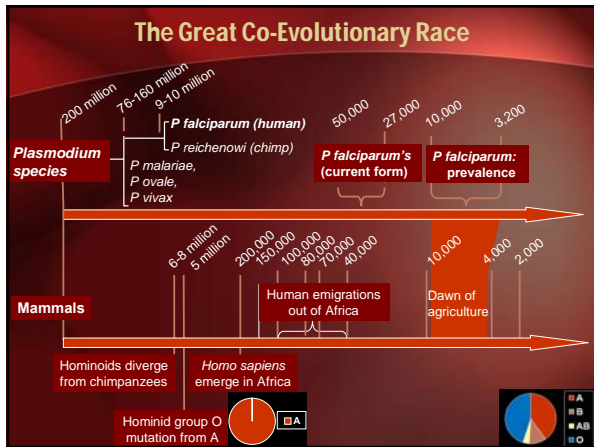
ubiquitous ABO = a more logical SEVERITY factor

**rosetting**

- red cells: > 2 million ABO sites via:
- platelets

**sequestration**

- endothelial cells
- macromolecules
  - von Willebrand factor
  - cellular adhesion molecules (selectins, PECAM [CD31])
  - platelet glycoprotein IV [CD36]



## Prevalence of O:A by Malaria Endemicity

Location	O-to-A ratio	Malaria	Location	O-to-A ratio	Malaria
Norway/Sweden	0.79	No	Sudan	3.86	Yes
Finland	0.83	No	Kenya	3.16	Yes
Czech Republic	0.68	No	S America	> 9	Yes
Austria/Hungary	0.82	No	Vietnam	1.91	Yes
Portugal	0.66	No	Philippines	2.04	Yes
Switzerland	0.80	No	P N Guinea	1.52	Yes

## Clinical Studies on ABO in Malaria...

**23** studies dating back to 1979

Serious shortcomings:  
sample size small  
inadequate control  
inappropriate cases

**6** higher merit studies

larger sample size  
test ABO association to severity  
last 10 years  
performed throughout Africa

## In-Vitro Rosettes are Strongest for Group A

RBC rosette capacity from 52 donors of various ABO types (using O infected RBC)

Carlson J et al. J Exp Med 1992; 176: 1311-7

## In-Vitro Rosetting and A-Binding: Severity-correlated

111 fresh Kenyan pediatric parasitized RBC isolates

	Mild Malaria	Severe Malaria	p - value
Rosetting (%)	12.9	21.4	0.010
Giant rosetting (%)	2.3	9.8	< 0.001
Soluble Group A binding (%)	1.8	5.4	0.031

Heddini A et al. Infect & Immun 2001; 58:49

## A vs O: Severity and In-Vitro Rosetting

### Severity: A vs O

n = 567, Mali, 2003  
n = 45, Kenya, 1993  
n = 39, Kenya, 2003

### Rosette Frequency: A vs O

Rowe JA et al. PNAS 2007; 104: 17471

## Soluble Group A Inhibits Rosetting In-Vitro

Enzymatic conversion of A → O  
abolished rosettes.

Barrağan et al. Infect & Immun 2000; 68: 2971

## Implications:

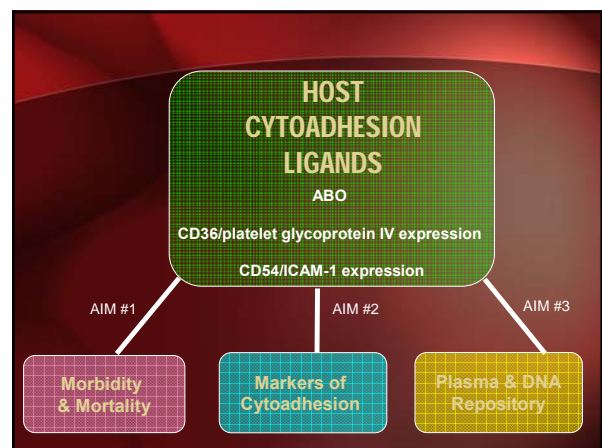
- ABO status: patient risk assessment
- group O RBCs (less sticky) may be preferred for transfusion
- malarial selection pressure may explain the higher prevalence of the Le(a-b-)/ Se+ status among Africans
- knowledge suggests a potential new strategy for treatment, using blockers of cytoadhesion.

### Caveats:

- malaria is complex and depends on more than ABO
  - CD36, ICAM-1
  - Knops (CR-1)
  - immune response
  - platelet - vascular interactions

## The Cytoadherence in Pediatric Malaria Study

[www.clinicaltrials.gov](http://www.clinicaltrials.gov) : NCT 00707200  
 HS 356 (UNCST)  
 TAHSN REB 07-0548-AE





## Summary

- Malaria is a disease of global importance.
- Pathogenesis from cytoadhesion via PfEMP-1 depends, in part, on the ABO status of host.
- Clinical severity & mortality is related to ABO.
- Evidence suggests that *P falciparum* has been a principal shaper of the distribution of ABO groups in humans.



Blood, Oct 2007