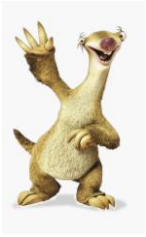

Pee-culiar Blood Groups: The Curious Case of Sid

CLPC SEMINAR: FALL 2025
KRISTIN BUTLER, MPH, MLS (ASCP)CM
LSU HEALTH SHREVEPORT

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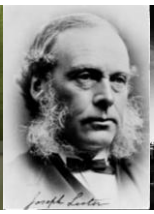
Course Objectives:

1. Define and characterize the Sid blood group system.
2. Explore unique biological features of the Sid antigen.
3. Describe laboratory testing and clinical significance for the Sid antigen and antibodies.

Fun fact: Guinea pig urine is the richest natural source of the Sd^a antigen!

2

Lister Institute of Preventive Medicine



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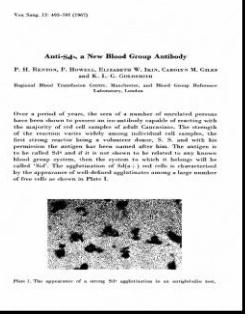
1967, London

- Volunteer donor named Sidney Smith
  - Head of the maintenance department
- System named after him
- Antigen named Sd^a

Renton, P.H., Howell, P., Ikin, E.W., Giles, C.M. and Goldsmith, D.K.L.G. (1967), Anti-Sd^a, a New Blood Group Antibody. Vox Sanguinis, 13: 493-501. https://doi.org/10.1111/j.1423-0410.1967.tb03796.x


Vox Sang. 13: 493–501 (1967)

Anti-Sd^a, a New Blood Group Antibody

P. H. Renton, P. Howell, Elizabeth W. Ikin, Carolyn M. Giles and K. L. G. Goldsmith

Regional Blood Transfusion Centre, Manchester, and Blood Group Reference Laboratory, London

Over a period of years, the sera of a number of unrelated persons have been shown to possess an iso-antibody capable of reacting with the majority of red cell samples of adult Caucasians. The strength of the reaction varies widely among individual cell samples, the first strong reactor being a volunteer donor, S. S., and with his permission the antigen has been named after him. The antigen is to be called Sd^a and if it is not shown to be related to any known blood group system, then the system to which it belongs will be called 'Sd'. The agglutination of Sd^a(+) red cells is characterized by the appearance of well-defined agglutinates among a large number of free cells as shown in Plate I.

Plate I. The appearance of a strong Sd^a agglutination in an antiglobulin test.

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Historical highlights

- Multiple groups describing distinctive agglutination pattern from an elusive antibody (1967)
  - Refractile small agglutinates in a sea of free cells
  - Difficult to distinguish weakly positive from negative RBCs
  - Individuals with positive RBCs varied in agglutination strength with the antibody
  - 1% strong, 80% moderate, 10% weak or neg

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Distinctive agglutination pattern

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Historical highlights

- 1970: group from Radcliffe Infirmary in Oxford found the Sd^a antigen in urine of humans and various animals
  - Over 90% of people secrete Sd^a
  - Expression of the Sd^a antigen varies from barely detectable to extremely strong
  - Found Sd^a "activity" in tissues and fluids, including urine, milk, feces, and saliva

J.A. Morton, M.M. Pickles, A.M. Terry; The Sda Blood Group Antigen in Tissues and Body Fluids. *Vox Sanguinis* 31 December 1970; 19 (5-6): 472–482. https://doi.org/10.1159/000466036

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Historical highlights

- In 1971, anti-Sd^a was associated with the **Cad phenotype**
  - First noted in 1962 by Japanese investigators
  - Later defined as a feature among members of a Mauritian family of India origin (1968)
  - Polyagglutination with anti-Sd^a

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Terminology and nomenclature

- Originally placed in 901 series of high-prevalence antigens
- Confirmation of genetic origin in 2019
  - ISBT acknowledged SID group system as number 038

Red Cell Immunogenetics and Blood Group Terminology | ISBT Working Party | The International Society of Blood Transfusion (ISBT)

Table of blood group systems | The International Society of Blood Transfusion (ISBT)

ISBT International Society of Blood Transfusion

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Antigens and antibodies of SID

- Only one antigen, Sd^a
- Anti-Sd^a is naturally occurring IgM
  - Generally considered insignificant
  - IgG and rise in titer have been described
  - Hemolytic transfusion reactions have been described when donor RBC had strong expression of Sd^a

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Antigens and antibodies of SID

- Sd^a antigen is lacking on RBC of fetus and newborn
- Becomes evident around 7 – 10 months of age
- Shares similarities with Lewis system antigens
  - Sd^a appears to be passively absorbed onto RBC
  - Pregnant women may transiently appear Sd(a=)
  - Antibodies can be neutralized by various body fluids

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Related Cad phenotype

"Super Sid" Sd(a+++)

- Extremely low-prevalence phenotype
- RBCs highly reactive to anti-Sd^a
- 1962: Japanese group O and B donors
- 1968: Family of group B donors
  - Found RBC reacted with *Dolichos biflorus* (anti-A1 lectin)
  - Sanger et al. (1971) established link with Sd^a
  - Additional studies followed to confirm
  - Higher prevalence in Asian population

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Prevalence of SID phenotypes

RBC phenotype	Prevalence (%)	Population	Size of study/ies
Sd(a−)	9 (2–4 in other tissue)	British	290 and 144
Sd(a+)	91	British	
Cad	0–0.07	French	250,000* and 78,526*
	0.03	Japanese	51,429* and 3183†
	0.14	Canadian	1425*
	0.26	Thai	14,261*

Stenfelt, L., Hellberg, Å.. & Olsson, M.L.. (2023). SID: a new carbohydrate blood group system based on a well-characterized but still mysterious antigen of great pathophysiologic interest. Immunohematology, 39(1), 2023. 1-10. https://doi.org/10.21307/immunohematology-2023-002

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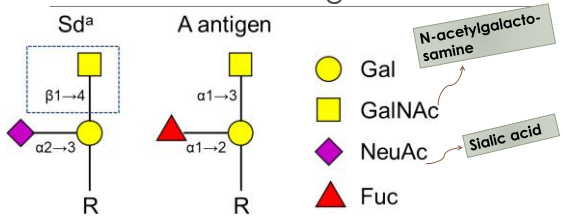
Structure of Sd^a antigen



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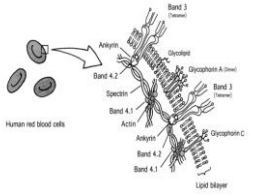
Biochemistry of SID on RBC



- Antigen structures of Sda and Cad decorate Glycophorin A and Band 3
- Has the same terminal sugar as A antigen, N-acetylgalactosamine (GalNAc)

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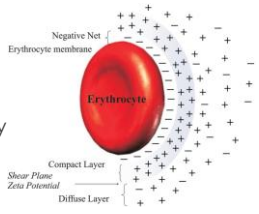
Remember the RBC zeta potential?

➢**Sialic acid (NeuAc)**

- Difference in electrical potential around cells
- Found on the surface of many vertebrate cells as part of the "sugar coating" of the cell membrane



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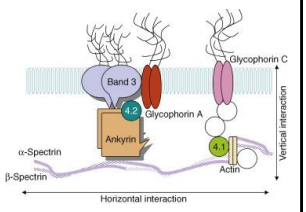
Glycophorin A on RBC membrane

- Major intrinsic membrane protein of RBC
- Required for high activity of Band 3 anion exchange
- Highly decorated with sialic acid (NeuAc)



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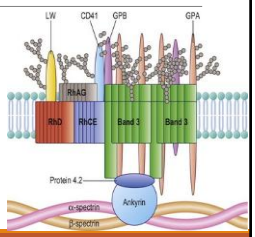
Band 3 on the RBC membrane

- Most abundant membrane protein in human RBC
- Facilitates exchange of CO_2 and HCO_3
- Associated with Glycophorin A
- ABO, H/h, I/i antigens



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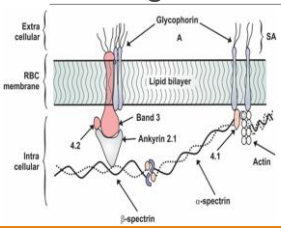
Genetics and molecular basis of Sd^a antigen formation



- The gene, ***B4GALNT2***, codes for a transferase enzyme that adds the terminal sugar N-acetylgalactosamine (GalNAc) onto the sialic acid
  - ➢ Sd^a epitope

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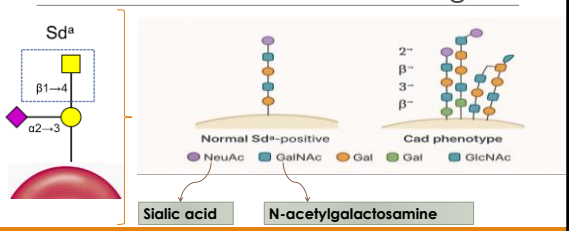
Structure of Sd^a and Cad antigens



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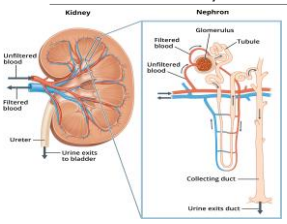
Biochemistry of SID in kidneys



- Detected in distal convoluted tubule and collecting ducts
- Carried by Tamm-Horsfall glycoproteins (uromodulin)
  - Most abundant protein in human urine
  - Hyaline casts in urine

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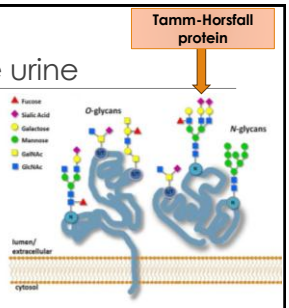
Sd^a antigen in the urine



- **B4GALNT2** enzyme highly expressed in kidney
- Adds GalNAc onto sialic acid residues of Tamm-Horsfall proteins
- Then secreted into urine
- >90% of the population

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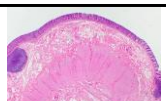

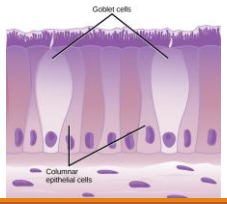

Biochemistry in colon

- Highly expressed on healthy colon tissue
  - Brush borders
  - Goblet cells
- Thought to be additional main source of antigen in human body

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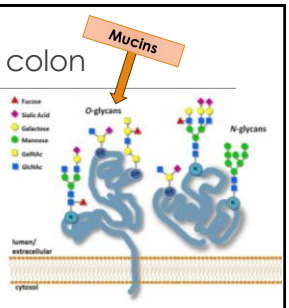
Sd^a antigen in the colon



- **B4GALNT2** enzyme highly expressed in colon
- Adds GalNAc onto sialic acid residues of mucins produced by goblet cells

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B4GALNT2 in other species

- 1950s – 90s: antigen recognized in other species like guinea pig, moles, hedgehogs, and mice
- Studies continue in others like pigs and sheep
- **Sialic acid found in all vertebrates**

J.A. Morton, M.M. Pickles, A.M. Terry; The Sda Blood Group Antigen in Tissues and Body Fluids. *Vox Sanguinis* 31 December 1970; 19 (5-6): 472–482. https://doi.org/10.1159/000466036

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So how and why the GUINEA PIG?!

- In 1951, researchers investigating blood group antigens found weird reactivity after RBCs were treated with guinea pig urine
- In 1963, another group purified the substance, proving it is carbohydrate in nature
- Guinea pigs express a more "Sd^a-active" version of Tamm-Horsfall proteins
  - Richest source of Sd^a antigen known!

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Genetic origin solved!

- 1960s – 70s: Sd^a antigen known
- 1980s – 90s: extracts from colon and kidney tissues could generate Sd^a antigen → ENZYME elucidated
- 2001 – 2003: researchers found new gene on CHROMOSOME 17 (*B4GALNT2*)
- 2019: Missense mutation gives rise to Sd(a=) phenotype

Sid gets his own blood group system!

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Disease associations

- Since Sd^a expressed on products of renal and colon epithelial cells, speculation that it contributes to:
  - Epithelial barrier function
  - Microbial adhesion and recognition
  - Cell-cell communication
  - Immune regulation via glycoprotein structure

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Sd^a may act as a decoy receptor

- Many pathogens exploit sialic acid residues gain entry and cause infection
  - Influenza A & B, Parainfluenza, Rotavirus, Parvovirus
  - Many bacteria use sialic acid as "molecular mimicry"
  - *Plasmodium falciparum* bind to sialic acid
  - *Trypanosoma cruzi* steals sialic acid from host
  - *Toxoplasma gondii* use sialic acid for adhesion

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Studies in other animals

- In mice, expression of Sd^a on von Willebrand factor leads to accelerated clearance, and thus bleeding
- Overexpression of *B4galnt2* in mice with muscular dystrophy reduced the pathology
- In sheep, the gene is expressed in ovaries and impacts fertility and litter size
- Knockout mice show significant changes in microbiome diversity, especially in large intestine

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Sd^a and colon cancer

- In health colon mucosa, Sd^a is "signature" of mature, differentiated epithelial cells
- In colon cancer, Sd^a is markedly downregulated or completely lost
  - Shift to production of oncofetal antigens
  - Immune evasion, altered microbial interaction
  - Metastatic potential increases
  - Loss of normal epithelial identity
  - Gain of malignant behavior

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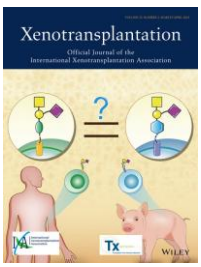

Clinical significance for transplant research

- *B4GALNT2* orthologue in pigs important for xenotransplant research
  - Pig gene codes for an enzyme that is 76% identical to human enzyme
  - Builds similar Sd^a/Cad epitopes
  - Most humans have IgM and IgG to these however, probably due to different sialic acid residue

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Clinical significance for transfusion

- Antigen is high prevalence, so anti-Sd^a rarely detected
- anti-Sd^a is most often IgM (sometimes IgG) and not significant
- May go undetected because screen cells lack high expression
- Rare cases of hemolytic transfusion reactions reported, usually when Sd(a+++) donor cells are transfused to recipient with anti-Sd^a

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Case Study of anti-Sd^a

- 91-year-old English female with myelodysplastic syndrome
- Multiply transfused; currently receiving 2 pRBC every 3 – 4 weeks
- Suffered a hemolytic reaction (HTR) during latest transfusion

Thornton, Nicole. Immunohematology Case Studies 2016 – 2. International Blood Group Reference Laboratory (IBGRL). NHS Blood and Transplant. Bristol, United Kingdom.

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Case Study of anti-Sd^a

- ABO/Rh: O, Rh-positive
- Pre-transfusion screening: Negative
- Units issued by electronic crossmatch
- Following HTR, patient samples and suspect unit sent to regional red cell immunohematology reference laboratory

Thornton, Nicole. Immunohematology Case Studies 2016 – 2. International Blood Group Reference Laboratory (IBGRL). NHS Blood and Transplant. Bristol, United Kingdom.

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Case Study of anti-Sd^a

Regional Reference Lab results:

- Antibody screen: NEGATIVE
- 10-cell antibody panel: NEGATIVE
- Donor cells crossmatch INCOMPATIBLE with plasma
- DAT on suspect unit: NEGATIVE
- DAT on patient sample: POSITIVE, IgG
- Eluate from patient cells: NEGATIVE

Thornton, Nicole. Immunohematology Case Studies 2016 – 2. International Blood Group Reference Laboratory (IBGRL). NHS Blood and Transplant. Bristol, United Kingdom.

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Case Study of anti-Sd^a

- Patient stabilized
- New samples taken 1 week later and referred to IBGRL as non-urgent case
- Now only giving serological crossmatch compatible blood to patient
- Referring lab confirmed antibody screen still negative

Thornton, Nicole. Immunohematology Case Studies 2016 – 2. International Blood Group Reference Laboratory (IBGRL). NHS Blood and Transplant. Bristol, United Kingdom.

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Case Study of anti-Sd^a

IBGRL results:

- ABO/Rh: Group O, R1r (DCe/dce)
- DAT: Positive, IgG
- Antibody screens (gel and tube): NEGATIVE
- Antibody panel with papain treated cells:

POSITIVE, variable strength, mixed field reactivity

Thornton, Nicole. Immunohematology Case Studies 2016 – 2. International Blood Group Reference Laboratory (IBGRL). NHS Blood and Transplant. Bristol, United Kingdom.

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	D	C	c	E	e	S	s	K	Fya	Fyb	Jka	Jkb	IS	IAT	Papain
1	+	+	0	0	+	+	+	0	0	+	0	+	0	0	1+mf
2	+	0	+	+	0	+	0	0	+	+	+	0	w+	w+	2+mf
3	0	+	+	0	+	0	+	0	+	0	0	+	w+	w+	2+mf
4	0	0	+	+	+	+	+	0	+	+	+	+	0	0	1+mf
5	0	0	+	0	+	0	+	0	0	+	0	+	0	0	1+mf
6	0	0	+	0	+	0	+	+	+	+	+	+	w+	w+	2+mf
Unit	+	+	+	0	+			0					2+mf	3+mf	4+mf
Auto	+	+	+	0	+			0					0	0	w+

Agglutination "looked unusual, like small refractile bunches of grapes in a sea of negative cells"

Thornton, Nicole. Immunohematology Case Studies 2016 – 2. International Blood Group Reference Laboratory (IBGRL). NHS Blood and Transplant. Bristol, United Kingdom.

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All the clues point to anti-Sd^a

- Unusual mixed field (mf) agglutination
- Refractile agglutinates
- Reactivity stronger with papain treated cells
- Reactivity at IS and AHG
- Variation of expression

anti-Sd^a

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Further testing with patient plasma

- Testing against Sd(a++) and Sd(a=) cells

Cells	IS	IAT	Papain treated
Cad	3+mf	4+mf	4+mf
Sd(a++)	1+mf	3+mf	4+mf
Sd(a=)	0	0	0
Sd(a=)	0	0	0
Unit	2+mf	3+mf	4+mf
Autocontrol	0	w+	0

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Hemagglutination Inhibition

	PATIENT PLASMA			ANTI-Sd^a CONTROL		
Cells	With Sd(a+) urine	With Sd(a=) urine	With patient urine	With Sd(a+) urine	With Sd(a=) urine	With patient urine
Cad	0	4+mf	4+mf	0	4+mf	4+mf
Sd(a++)	0	3+mf	3+mf	0	3+mf	3+mf
Unit	0	3+mf	3+mf	0	3+mf	3+mf

Sd^a antigens in urine neutralized patient antibody, which prevents the antibody from reacting with antigens on the cells = CONFIRMATION

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The SID antigen and antibody, a most PEE-culiar blood group!

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THANK YOU!

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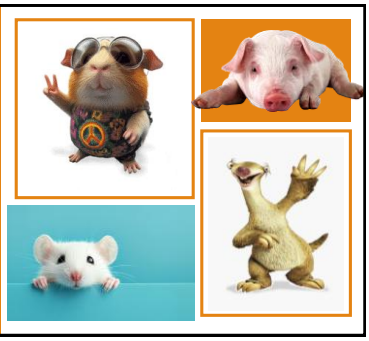

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