ABO Blood Group System

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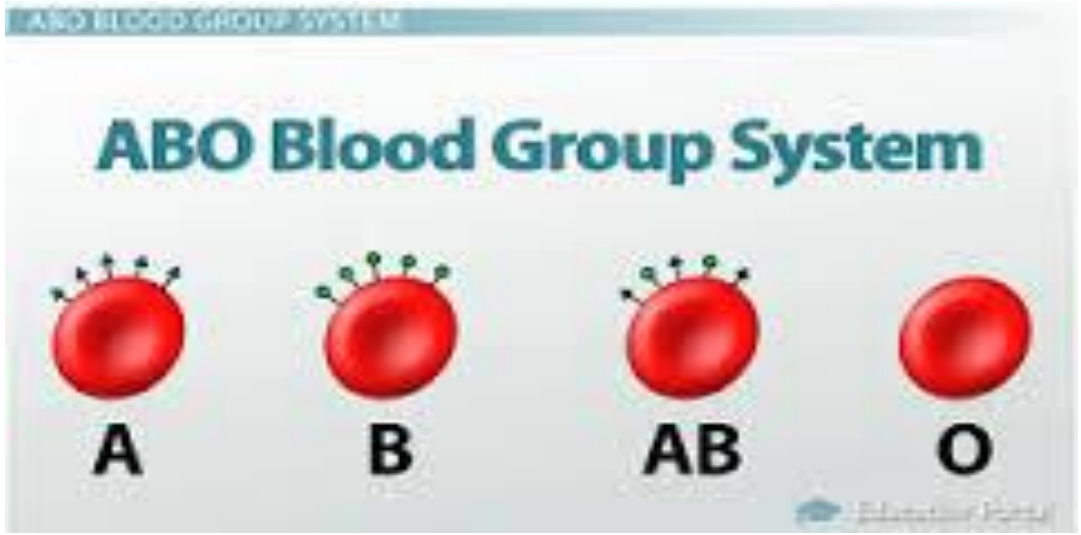
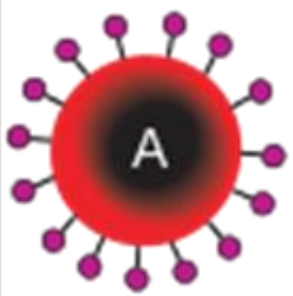
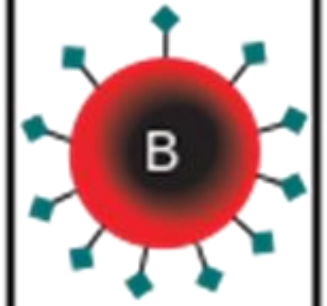
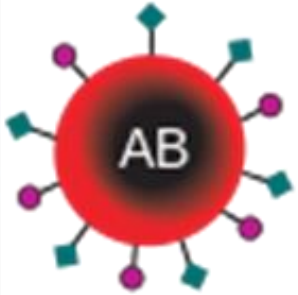
Discovery of ABO Blood Group System

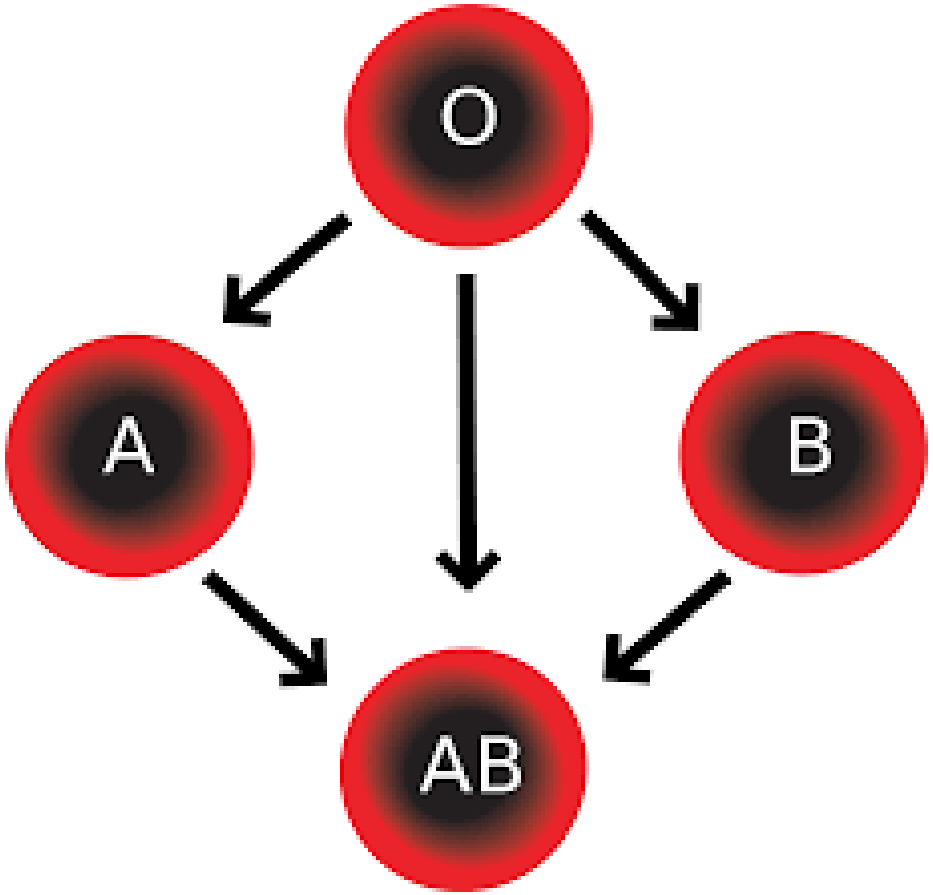
- 1901 by Karl Landsteiner
- Mixed cells + serum of associates
- Performed first forward and reverse grouping



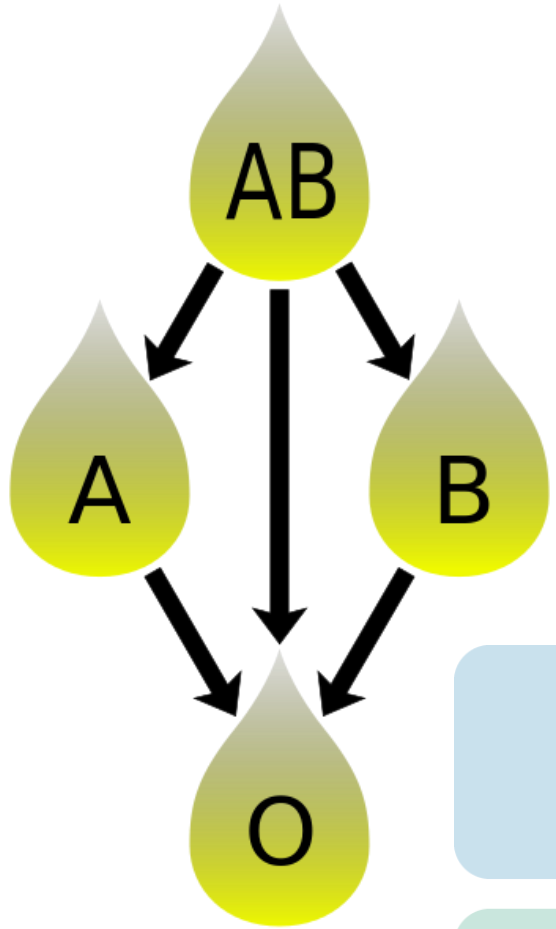
ABO Antigens and Antibodies

	Group A	Group B	Group AB	Group O
Red blood cell type				
Antibodies in Plasma	 Anti-B	 Anti-A	None	 Anti-A and Anti-B
Antigens in Red Blood Cell	 A antigen	 B antigen	 A and B antigens	None

ABO Compatibility for RBCs



ABO Compatibility for Plasma



Forward Grouping (Front Type)

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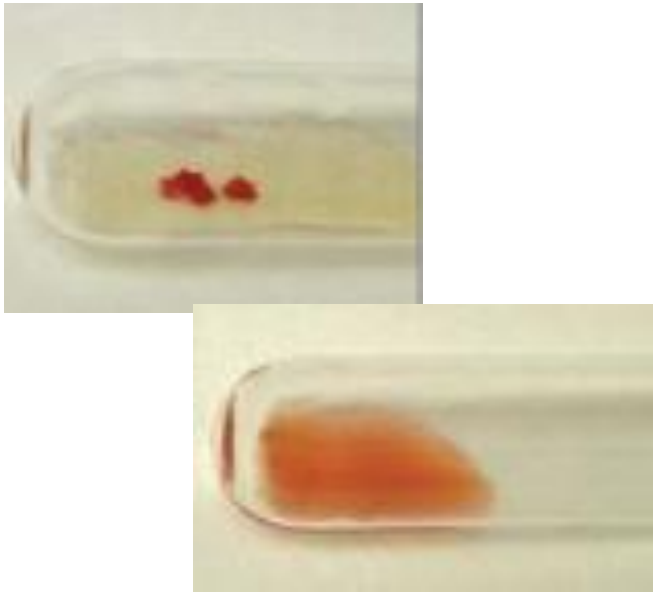


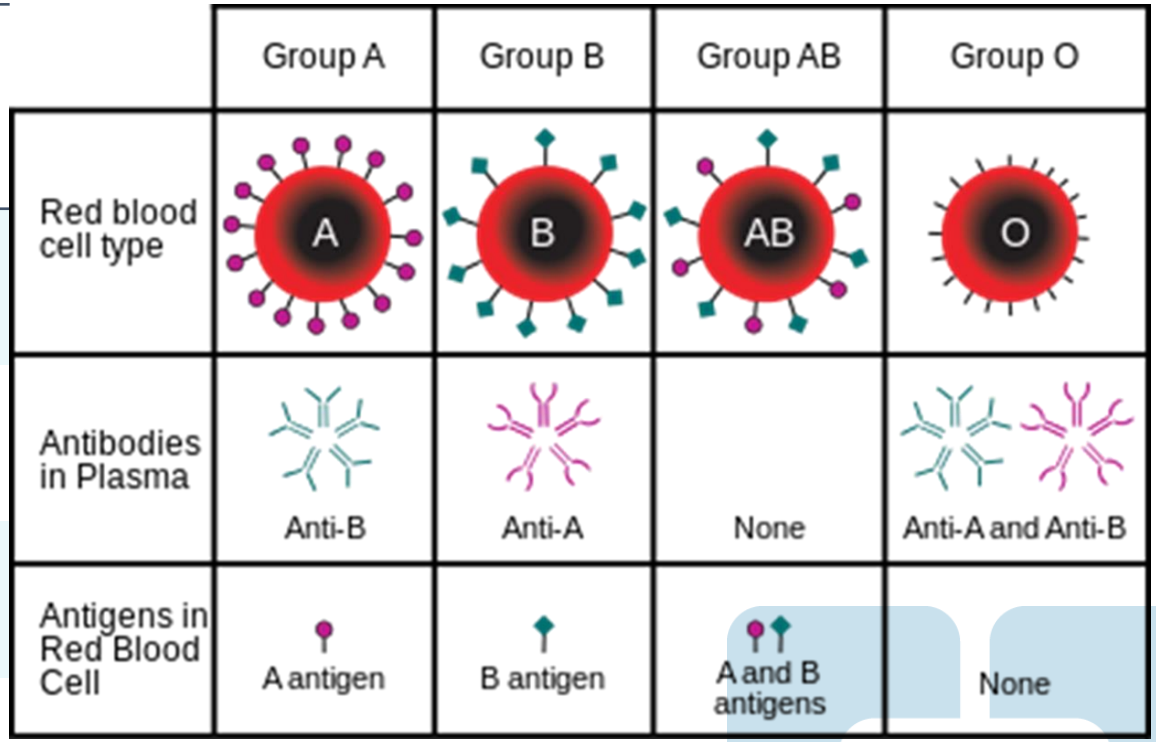
1 drop reagent (Anti-A,B, or D)

1 drop 3-5% patient red cell suspension

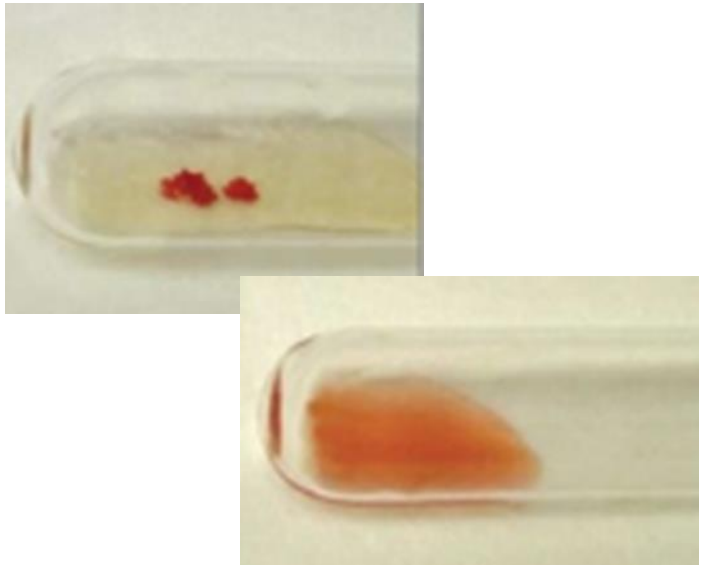
Centrifuge 30 seconds

Forwarding Grouping Results

Blood Type	Anti-A + Patient RBCs	Anti-B + Patient RBCs
O	0	0
A	4+	0
B	0	4+
AB	4+	4+



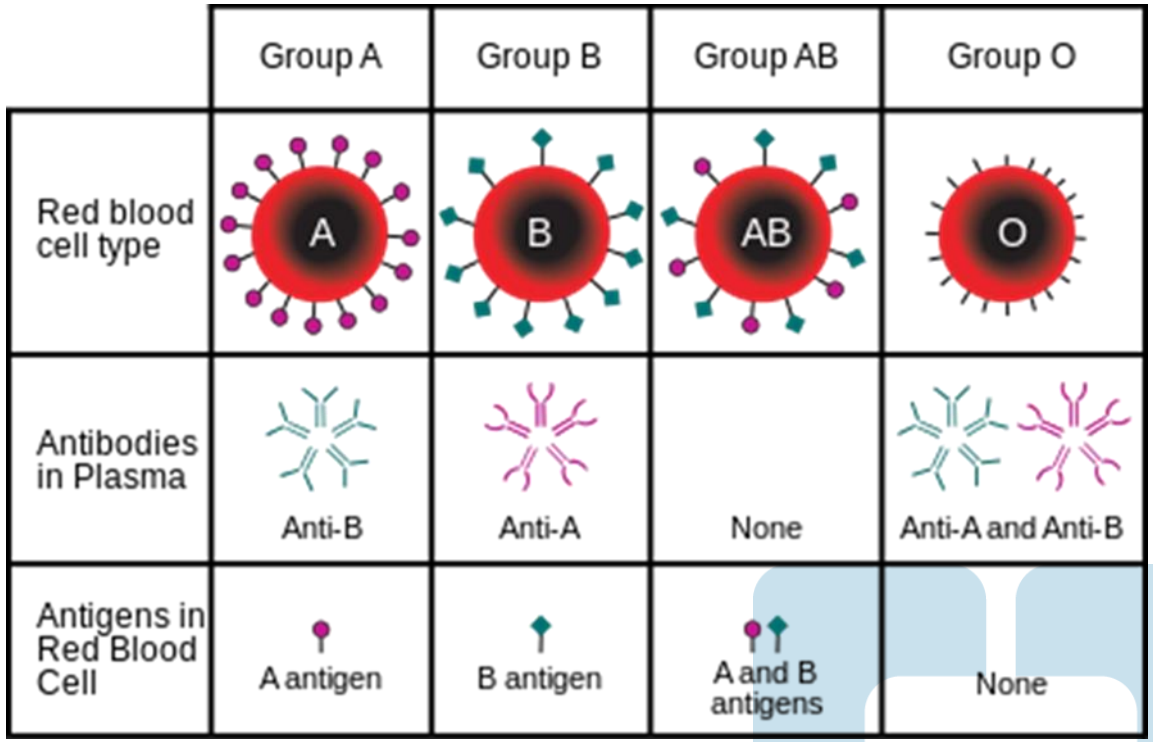
Reverse Grouping (Back Type)



1 drop A1 or B cells → 2 drops patient plasma → Centrifuge 30 seconds

Reverse Grouping Results

Blood Type	A1 cells + Serum	B cells + Serum
O	4+	4+
A	0	4+
B	4+	0
AB	0	0



Full ABO Blood Type Results

	Forward Grouping		Reverse Grouping	
Blood Type	Anti-A	Anti-B	A1 cells	B cells
O	0	0	4+	4+
A	4+	0	0	4+
B	0	4+	4+	0
AB	4+	4+	0	0

Frequency of ABO Blood Types in the Population

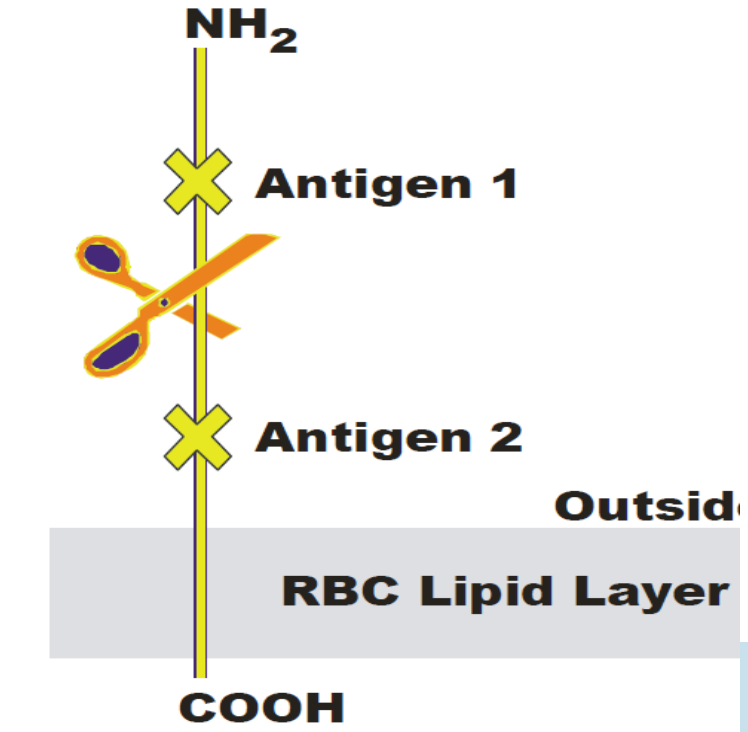
Most Common

Blood Group	Whites	African American	Hispanic	Asian
O	45%	50%	56%	40%
A	40%	26%	31%	28%
B	11%	20%	10%	25%
AB	4%	4%	3%	7%

Least Common

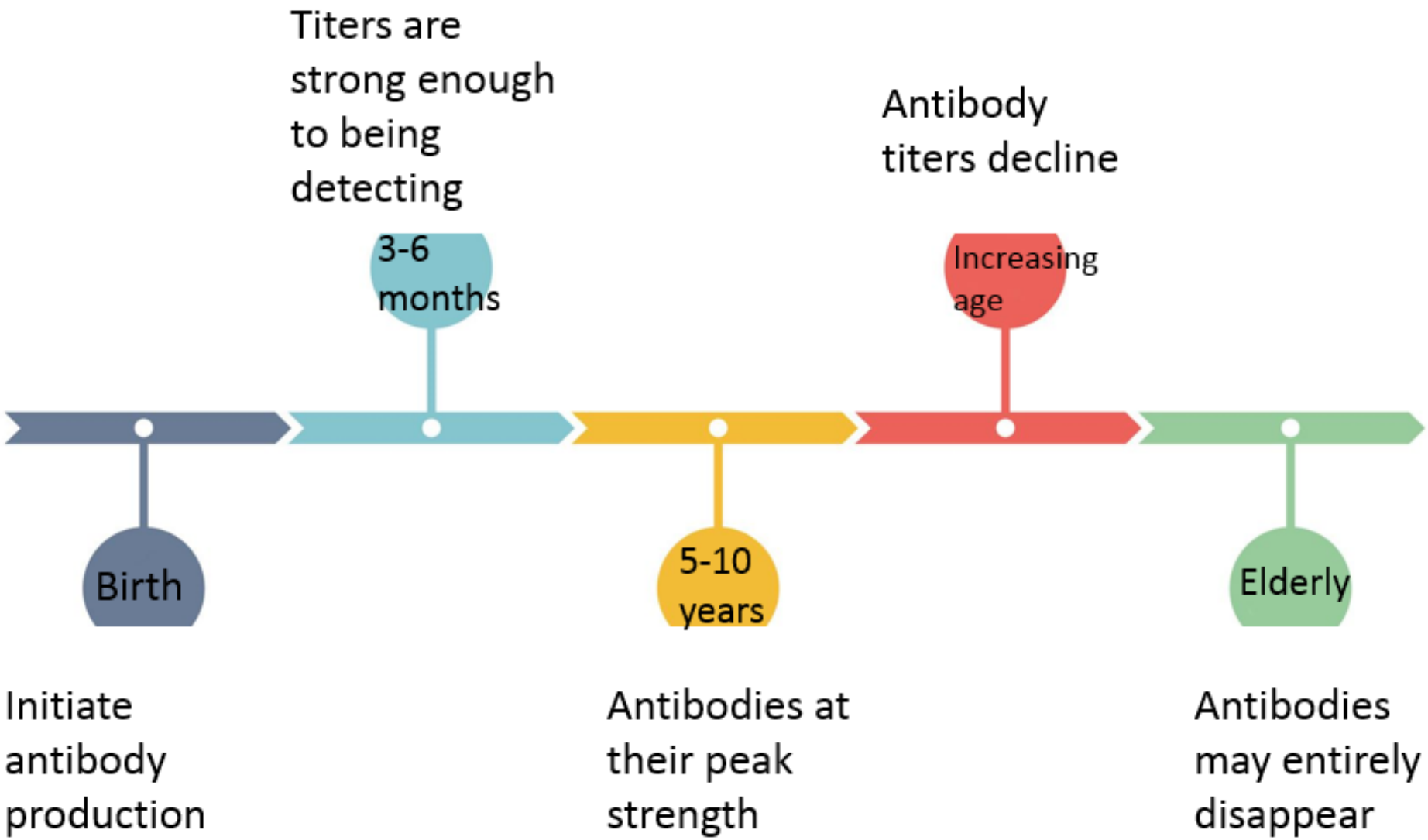
ABO Antibody Characteristics

	ABO
Enzymes	Increased
IgM vs. IgG	Anti-A and Anti-B = IgM (Anti-A,B = IgG)
Cold or Warm Reacting	4°C (some 37)
Natural vs. Immune	Natural
Hemolytic Transfusion Reaction (HTR)	Yes
Hemolytic Disease of the Fetus and Newborn (HDFN)	Yes
Binds Complement	Yes
Dosage	No



Enzyme Treatment

ABO Antibody Levels Throughout Life

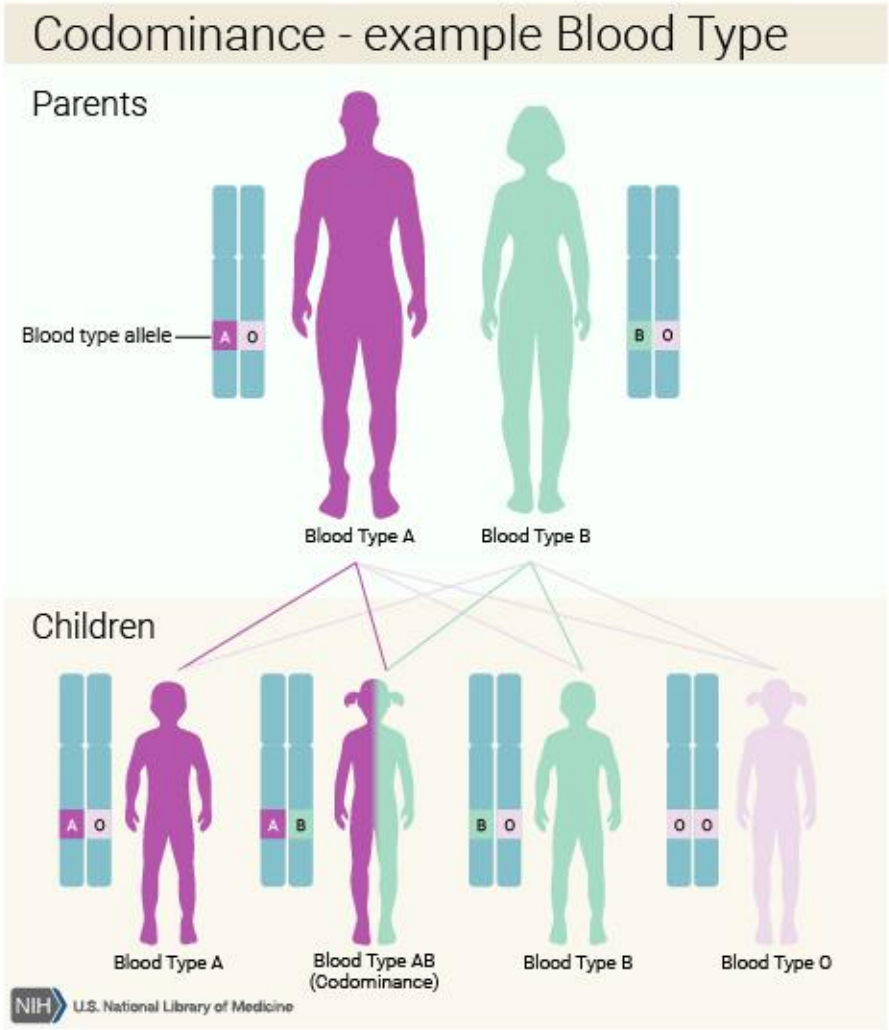


ABO Antigen Characteristics

- Develop in early fetal life
- Newborns = 20-50% of antigen sites
- A and B expression fully developed at 2-4 years
- Formed on:
  - RBC membranes
  - Endothelial cells
  - Platelets
  - Lymphocytes
  - Epithelial cells

Inheritance of ABO Genes

- Genes: *A*, *B*, or *O*
- Chromosome 9
- O = amorph
- Codominant Expression



ABO Genotype vs. Phenotype

Genotype	AA	AO	BB	BO	AB	OO
Phenotype	A		B		AB	O

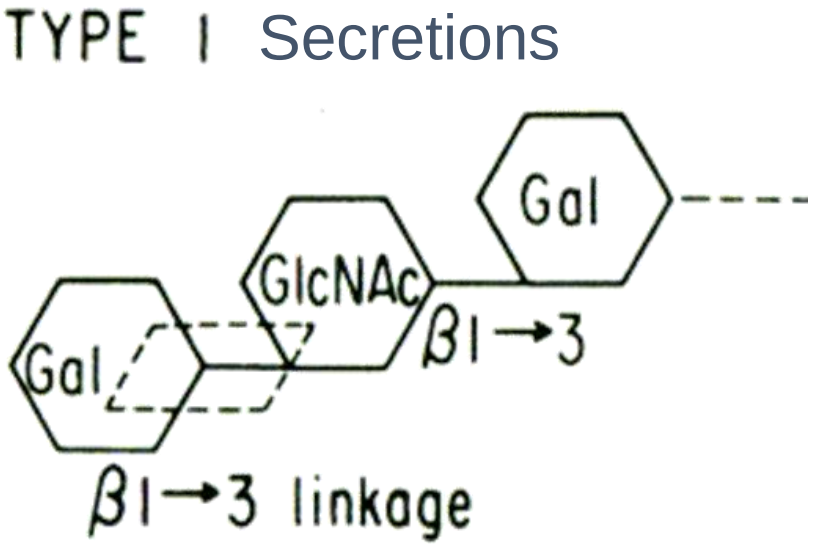
Example: same phenotypes, different genotypes:

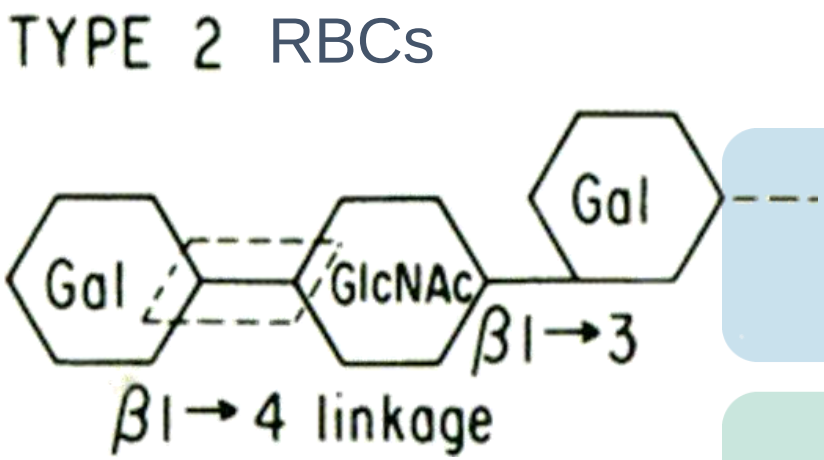
		A	A
O	O	AO	AO
	O	AO	AO

		A	O
O	O	AO	OO
	O	AO	OO

Inheritance of Hh and Se genes

- FUT1 (H gene)
- FUT2 (Se gene)
- Closely linked on Chromosome 19
- Code for glycosyltransferases

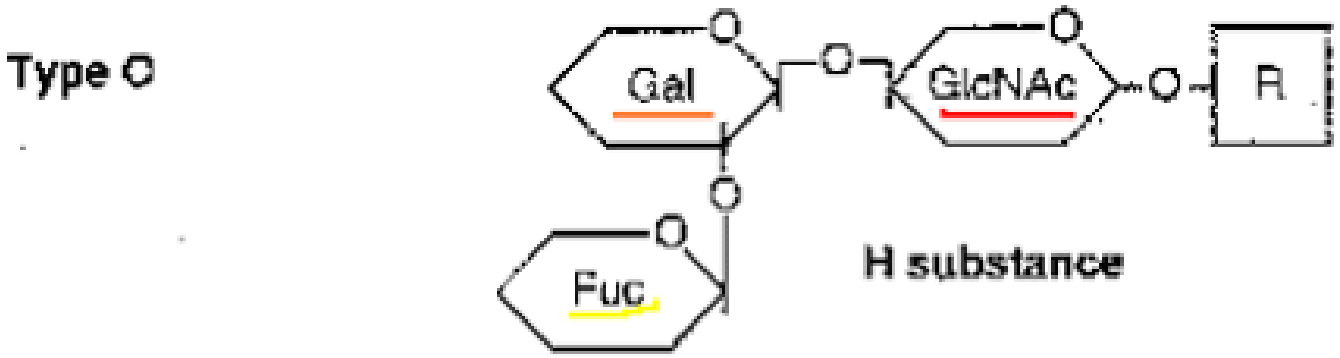




Formation of “O” Blood Type (RBCs)

Genotype: *HH* or *Hh*, *OO*

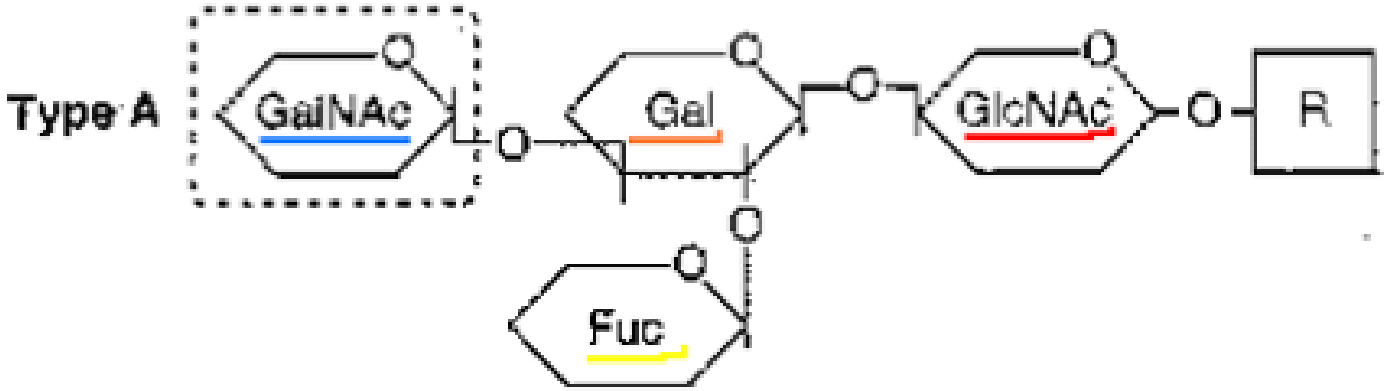
Gene	Glycosyltransferase	Immunodominant sugar	Antigen
H (FUT1)	α-2-L-fucosyltransferase	L-fucose	H



Formation of “A” Blood Type (RBCs)

Genotype: *HH* or *Hh*, *AA* or *AO*

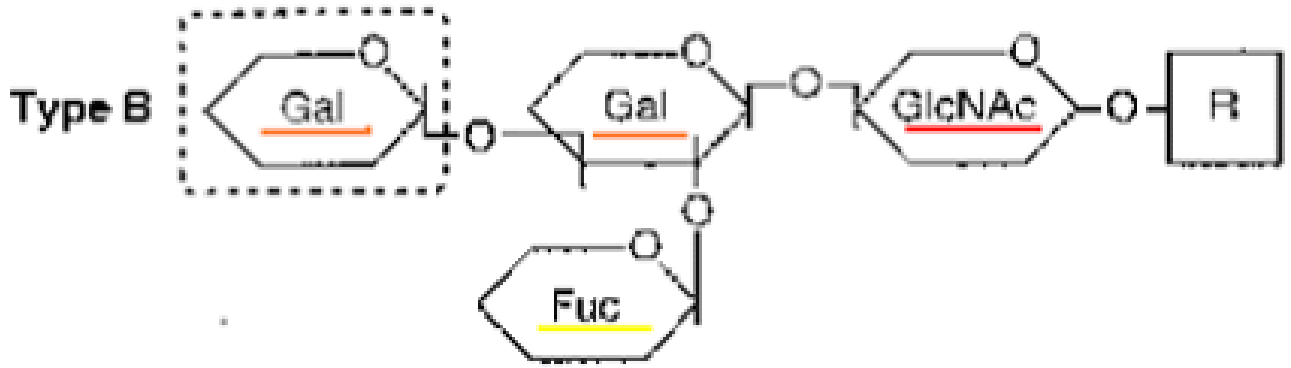
Gene	Glycosyltransferase	Immunodominant sugar	Antigen
H (FUT1)	α-2-L-fucosyltransferase	L-fucose	H
A	α-3-N-acetylgalactosaminly-transferase	N-acetyl-D-galactosamine (GalNAc)	A



Formation of “B” Blood Type (RBCs)

Genotype: *HH* or *Hh*, *BB* or *BO*

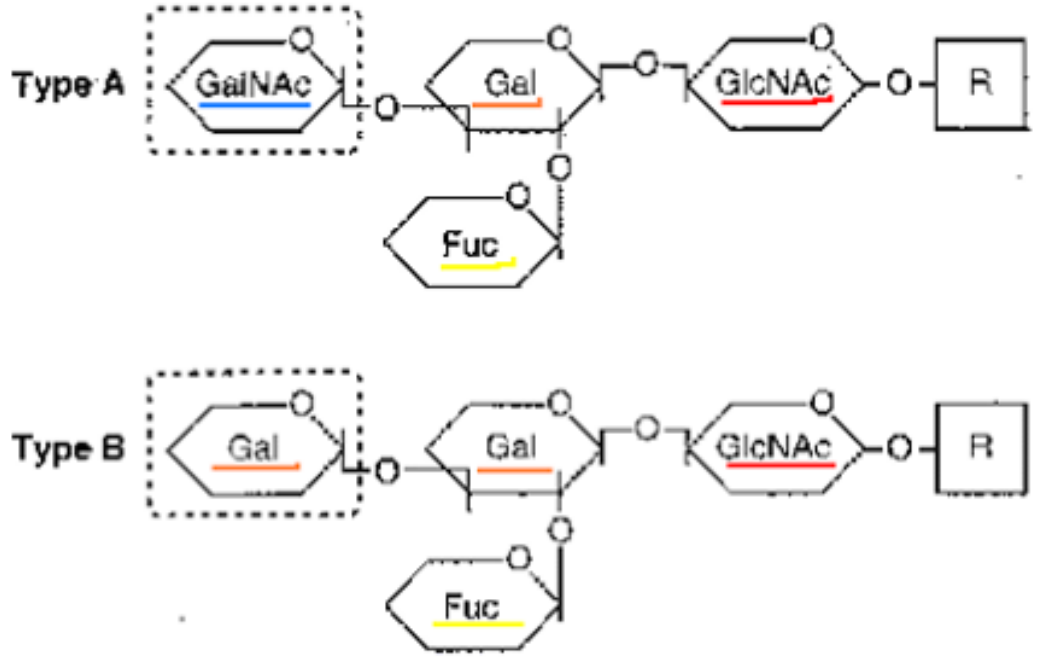
Gene	Glycosyltransferase	Immunodominant sugar	Antigen
H (FUT1)	α-2-L-fucosyltransferase	L-fucose	H
B	α-3-D-galactosyltransferase	D-galactose	B



Formation of “AB” Blood Type (RBCs)

Genotype: *HH* or *Hh*, *AB*

Gene	Glycosyltransferase	Immunodominant sugar	Antigen
H (FUT1)	α-2-L-fucosyltransferase	L-fucose	H
A	α-3-N-acetylgalactosaminly-transferase	N-acetyl-D-galactosamine (GalNAc)	A
B	α-3-D-galactosyltransferase	D-galactose	B



A antigen sites: 600,000

B antigen sites: 720,000

Formation of Blood Type in Secretions

- Glycoproteins
- Inherit *SeSe* or *Sese*

80% Secretors (*SeSe/Sese*)
20% Nonsecretors (*sese*)

Gene	Glycosyltransferase	Immunodominant sugar	Antigen
Se (FUT2)	α-2-L-fucosyltransferase	L-fucose	Soluble H
A or B	α-3-N-acetylgalactosaminly-transferase α-3-D-galactosyltransferase	N-acetyl-D-galactosamine (GalNAc) D-galactose	Soluble A or B

Precursor Substance: Type 1 —Se→ H soluble antigen —A or B→ A and B soluble antigens

H Antigen Present by Blood Type

O > A2 > B > A2B > A1 > A1B

Greatest
Amount of H

Least Amount
of H

Anti-H in A_1 and A_1B Individuals

- Most H antigen is converted to A and B
- H is so well hidden patients can make anti-H
- Reacts best with O cells
- Interferes with antibody screens due to type O reagents

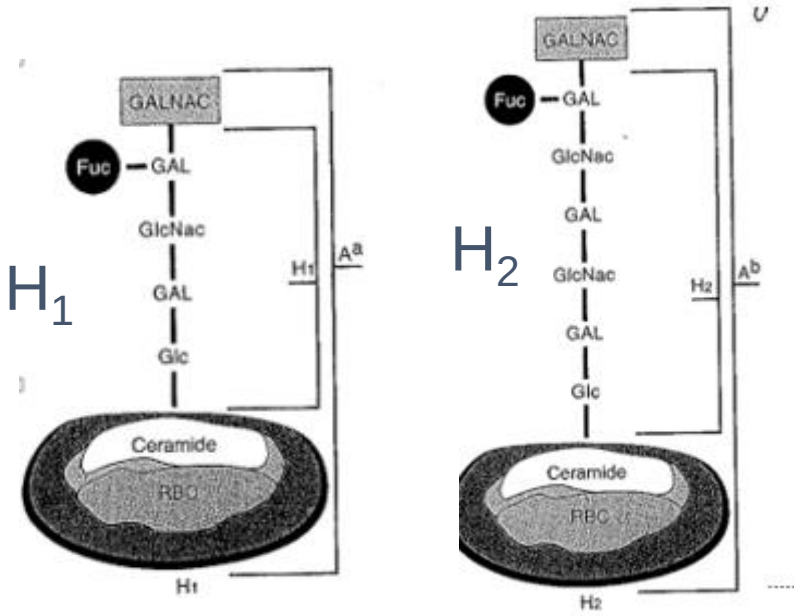
Anti-H (in A_1 and A_1B)
IgM
Reacts at 4^{o}C to room temp
Naturally occurring
No HTR - Insignificant
No HDFN

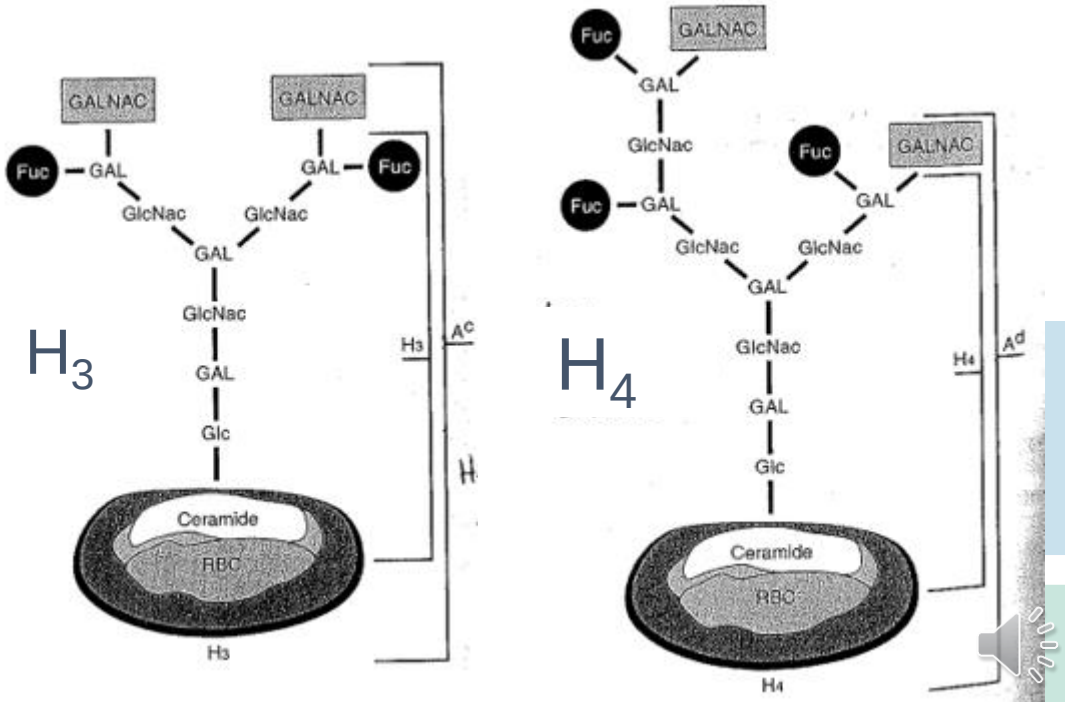
A Subgroups

	A_1	**A_2**
Reactivity	Anti-A and Anti-A_1	Anti-A only
Frequency	80%	20%
Inheritance	*A gene* normal	*A gene* mutated (single base substitution and single base deletion)
Enzyme	α-3-N-acetylgalactosaminly-transferase	α-3-N-acetylgalactosaminly-transferase with altered active site (less effective)
Immunodominant sugar	N-acetyl-D-galactosamine (GalNAc)	N-acetyl-D-galactosamine (GalNAc)
Produce Anti-A_1	No	1-8% of A_2 individuals 22-35% of A_2B individuals

A_1 vs A_2 Structure

- 4 different H antigens:
  - H_1 and H_2 = unbranched straight chains
  - H_3 and H_4 = complex branched chains
- Terminal sugars are identical
- H_1 and H_2 converted to A^a and A^b antigens by both A_1 and A_2 enzymes (A_2 is less efficient)
- H_3 and H_4 converted to A^c and A^d antigens by A_1 enzyme and very poorly by A_2 enzymes or not at all





A_1 and A_2 Quantitative and Qualitative Differences

Quantitative	Qualitative
A_2 has decreased # of antigen sites (A_1 = 1,170,000/RBC, A_2 = 290,000/RBC)	Differences in precursor chains
A_2 has decreased amount of transferase enzymes	Differences in transferase
A_2 has decreased amount of branched A antigens	A_2 individuals can form an anti-A_1

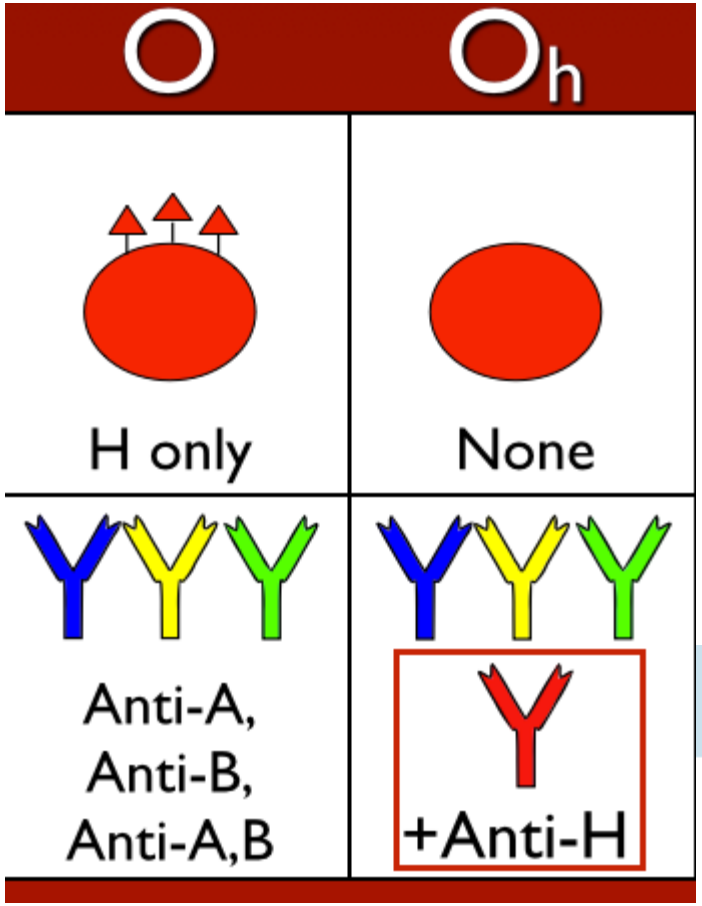
Anti-A1 Lectin Testing

- Anti-A_1 lectin: reagent made from seed extract of *Dolichos biflorus* plant
  - Agglutinates A_1 but not A_2 cells
- Reactions of Patients' RBCs with:

	Anti-A (Anti-A plus Anti-A_1)	Anti-A_1 lectin
A_1	+	+
A_2	+	0

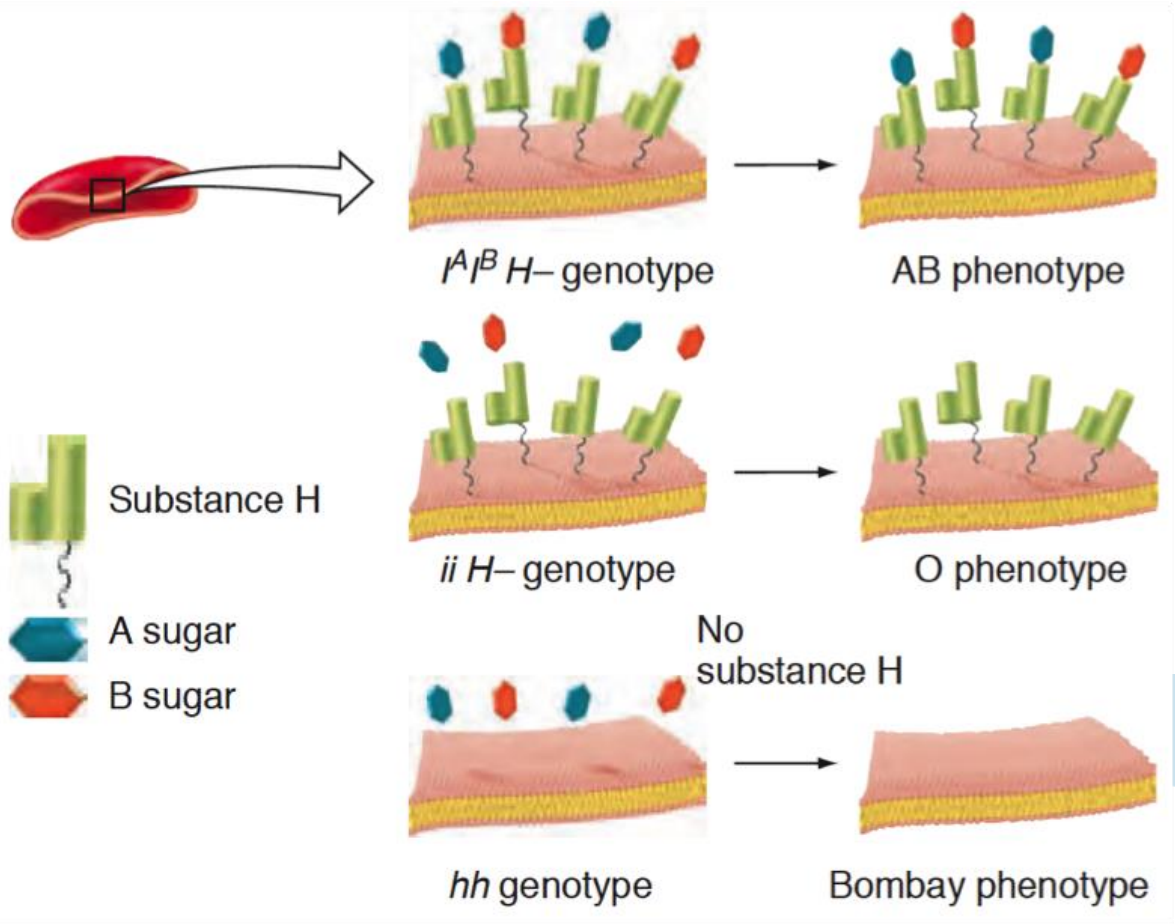
The Bombay Phenotype (O_h)

- Results from double dose of *h* gene (*hh)* – extremely rare
  - Prevalence in India = 0.048%
  - Prevalence in Europe = 0.0003%
- No *H* gene means no H antigens which means no ABO genes expressed
- No H antigen also means they form anti-H



Bombay Phenotype Genetics

- Mutation in *FUT1* (*H* gene)
  - Silenced – unable to code for fucosyltransferase
  - No α-2-L-fucosyltransferase, no L-fucose attached, no H substance present
- FUT2 gene (Se gene) also silenced
  - No ABO antigens form in secretions
- Normal ABO genes, but can't form ABO antigens due to lack of H
  - Genes written as Oh, Oh^A, Oh^B, Oh^{AB}



Bombay Phenotype Testing

- Phenotype as O blood group (lack all ABH antigens)
- Screen is all positive
- Use Anti-H lectin (extract of *Ulex europaeus*)
  - Reacts with type O, but not with Bombay

Blood Group	Forward Group			Reverse Group		
	Anti-A	Anti-B	Anti-A,B	A1 Cells	B Cells	O Cells
A	+	-	+	-	+	-
B	-	+	+	+	-	-
AB	+	+	+	-	-	-
O	-	-	-	+	+	-
Bombay	-	-	-	+	+	+

Anti-H in Bombay

	Anti-H (in A1 and A1B)	Anti-H in Bombay
IgM/IgG	IgM	IgM
Temperature of Reactivity	4°C and Room Temperature	Strongly at 37°C
Natural or Immune	Natural	Natural
Transfusion Reactions	No – Insignificant	Yes – binds complement & causes RBC lysis
HDFN	No	No

*Bombay patients must receive Bombay blood (H negative)

Para-Bombay

	Gene	RBC Antigens	Antigens in Secretions
Bombay	***hh sese*** Silenced *FUT1* and *FUT2*	None	None
Parabombay (Red cell H partially deficient, nonsecretor)	***hh* (weak variant) *sese*** Mutated *FUT1* gene With or without active *FUT2* gene	Very small amounts of H, A, and B	None
Parabombay (Red cell H deficient, secretor)	***hh Se*** Silenced *FUT1 (H)* gene Active *FUT2 (Se)* gene	Little to none of H, A, and B (absorbed onto RBC from plasma)	H, A, and B

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