
بسم الله الرحمن الرحيم

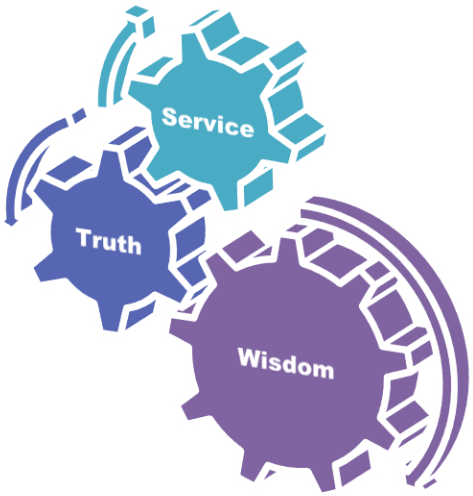
MSK- II MODULE
SKILL LAB /Physiology PRACTICAL
FIRST YEAR MBBS BATCH 50
DETERMINATION OF ABO BLOOD GROUPS

Dr. Ali Zain
FCPS PGT
Dept. Of Physiology

Date: 8 june 2023

Table of contents

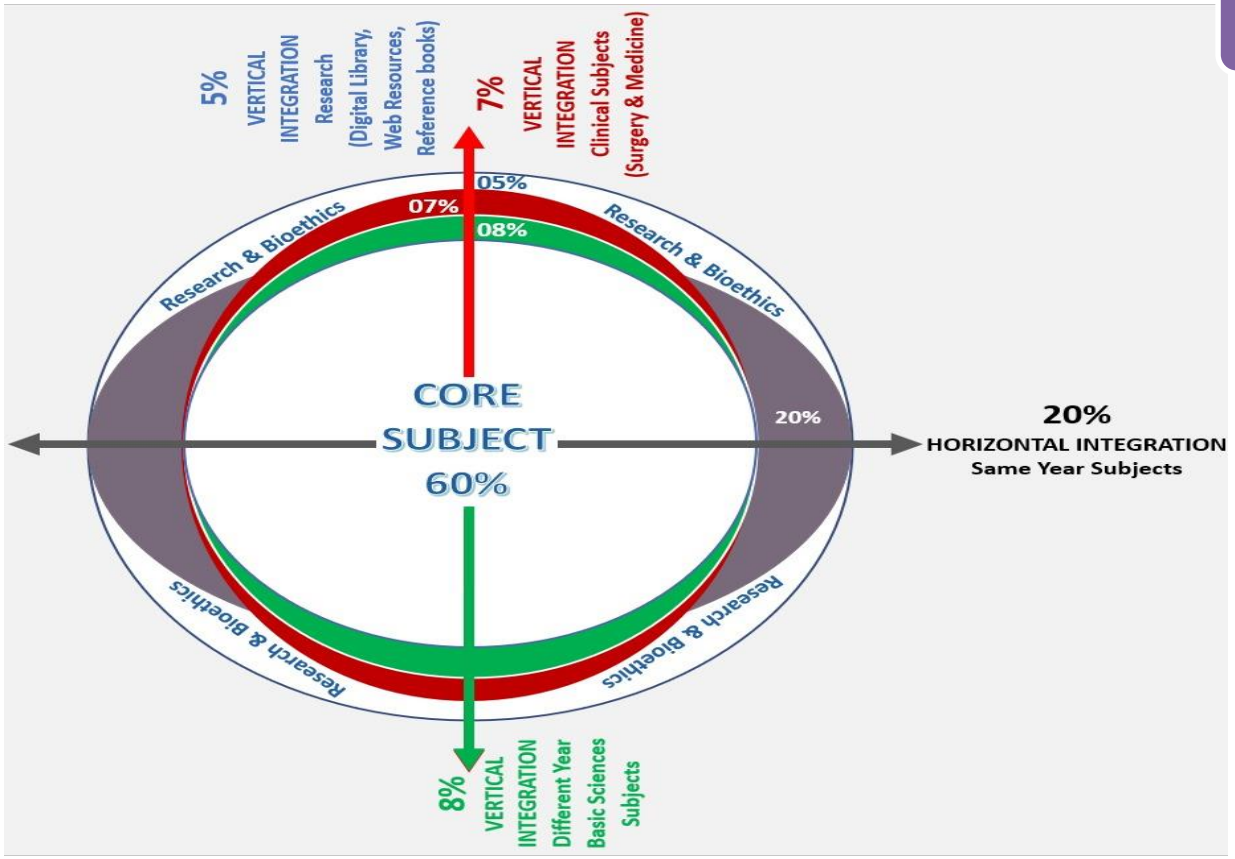
Motto



Vision; The Dream/Tomorrow

- To impart evidence based research oriented medical education
- To provide best possible patient care
- To inculcate the values of mutual respect and ethical practice of medicine

Professor Umar Model of Integrated Lecture



60% CORE SUBJECT

20% HORIZONTAL INTEGRATION Physiology biochemistry

8% VERTICAL INTEGRATION Pathology pharmacology

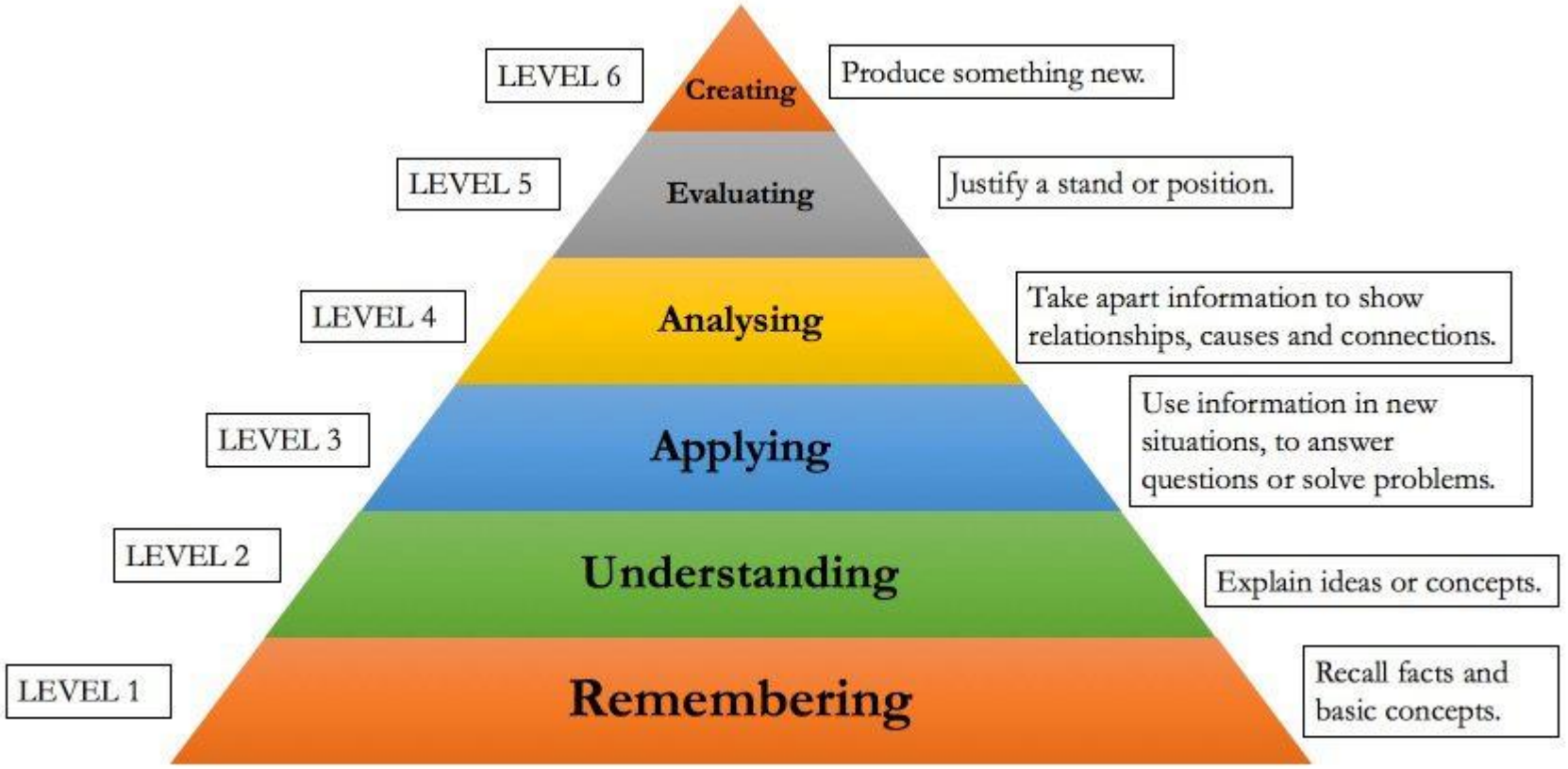
7% VERTICAL INTEGRATION Clinical integration

5% VERTICAL INTEGRATION Research, professionalism Ethics Digital library

BLOOM'S TAXONOMY : DOMAINS OF LEARNING

Sr. #	Domain of learning	Abbreviation	Levels of the domain	Meaning
1	cognition	C	C1	Recall / Remembering
2			C2	Understanding
3			C3	Applying / Problem solving
4	Psychomotor	P	P1	Imitation / copying
5			P2	Manipulation / Follows instructions
6			P3	Precision / Can perform accurately
7	Attitude	A	A1	Receiving / Learning
8			A2	Respond / Starts responding to the learned attitude
9			A3	Valuing / starts behaving according to the learned attitude

BLOOM'S TAXONOMY OF THE COGNITIVE DOMAIN



LEARNING OBJECTIVES

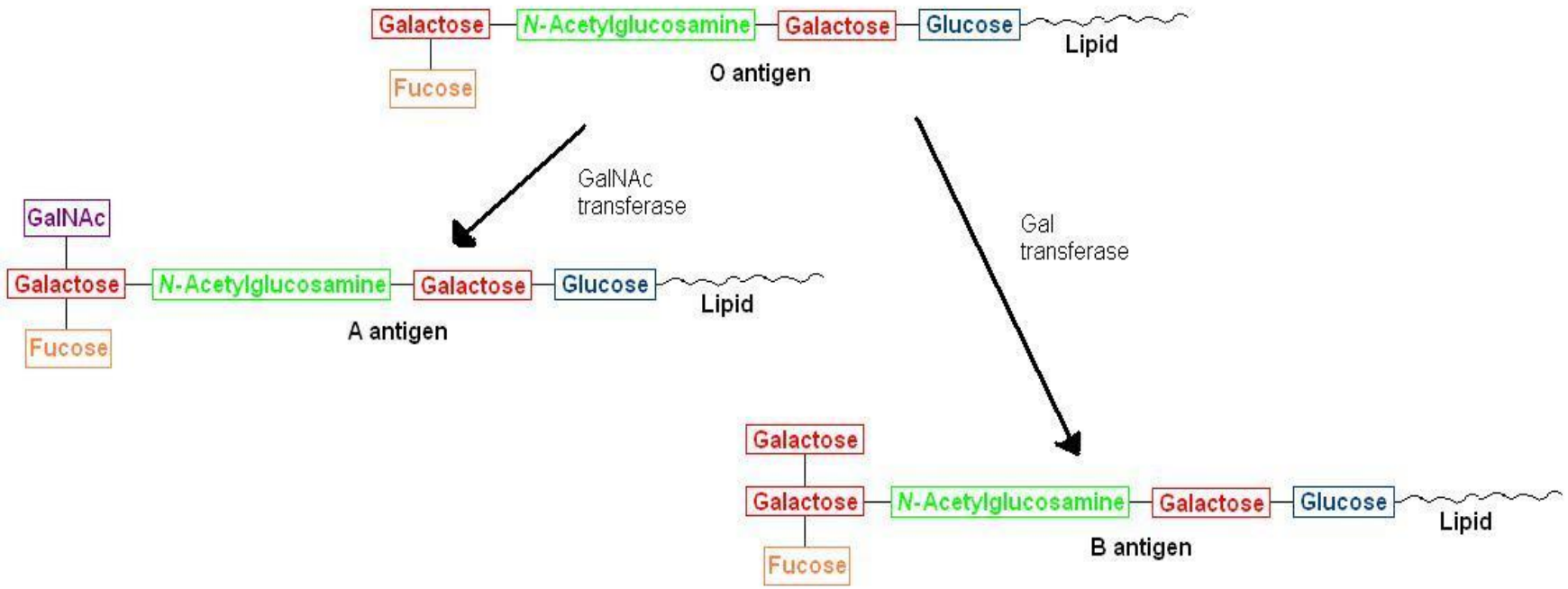
Sr. #	Learning Objective	Domain of Learning
1	To describe the relevance of doing ABO blood grouping.	C1
2	To perform step by step the ABO typing of Blood practical.	P3
3	To understand the reason of doing ABO blood grouping.	C1
4	To explain different types of Anti-serum	C2
5	To correlate Clinically with Acute Hemolytic Reaction and its signs and symptoms.	C3
6	To assess the neoborn with jaundice due to ABO incompatibility (vertical Integration)	C3

Horizontal integration

Horizontal integration

Horizontal integration (Biochemistry)

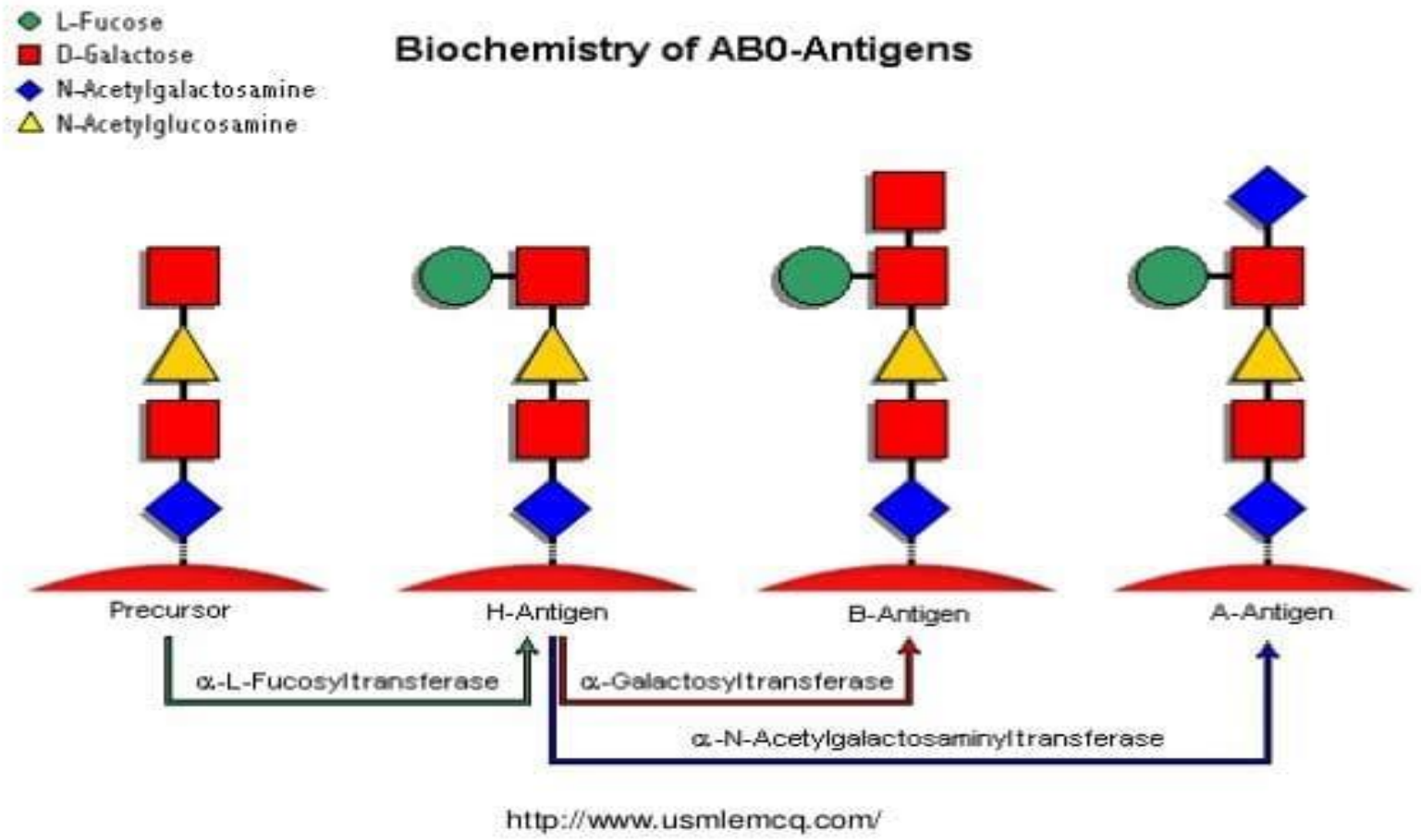
Human blood groups depends on the functioning of glycosyltransferases, enzymes that catalyze the formation of glycosidic bond between the Structure and function of the human blood. Specific oligosaccharide antigens attach to the proteins and lipids on the surface of erythrocytes.



Reference:- Wikipedia image of ABO biochemistry

Horizontal integration

Horizontal integration (Biochemistry)

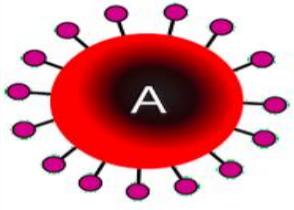
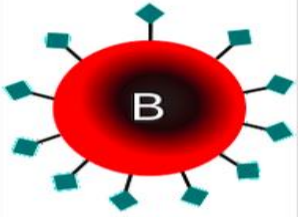
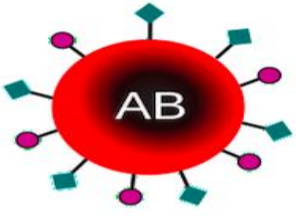
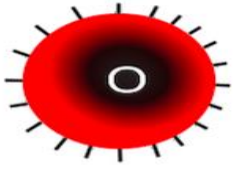


Core Concept

METHOD FOR DETERMINATION OF Total Platelet Count.

Core Concept

- Landsteiner discovered the ABO system of blood groups in 1900. By now, more than 400 blood group's antigens have been recognized.
- ABO system of blood groups consists of 3 allelic genes A, B and O. A person may have A, B or O type antigens on his red blood cells or any two of these together. O antigens are weak antigens, as are the anti-0 antibodies; hence they rarely cause any agglutination reaction.

	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

Core Concept

Table showing genetics of ABO system:-

It is inherited from parents to offspring on the basis of Mendel's law. That group inheritance depends upon the genes received from parents. Genes controlling blood group in man are 3 instead of two and are known as multiple alleles.

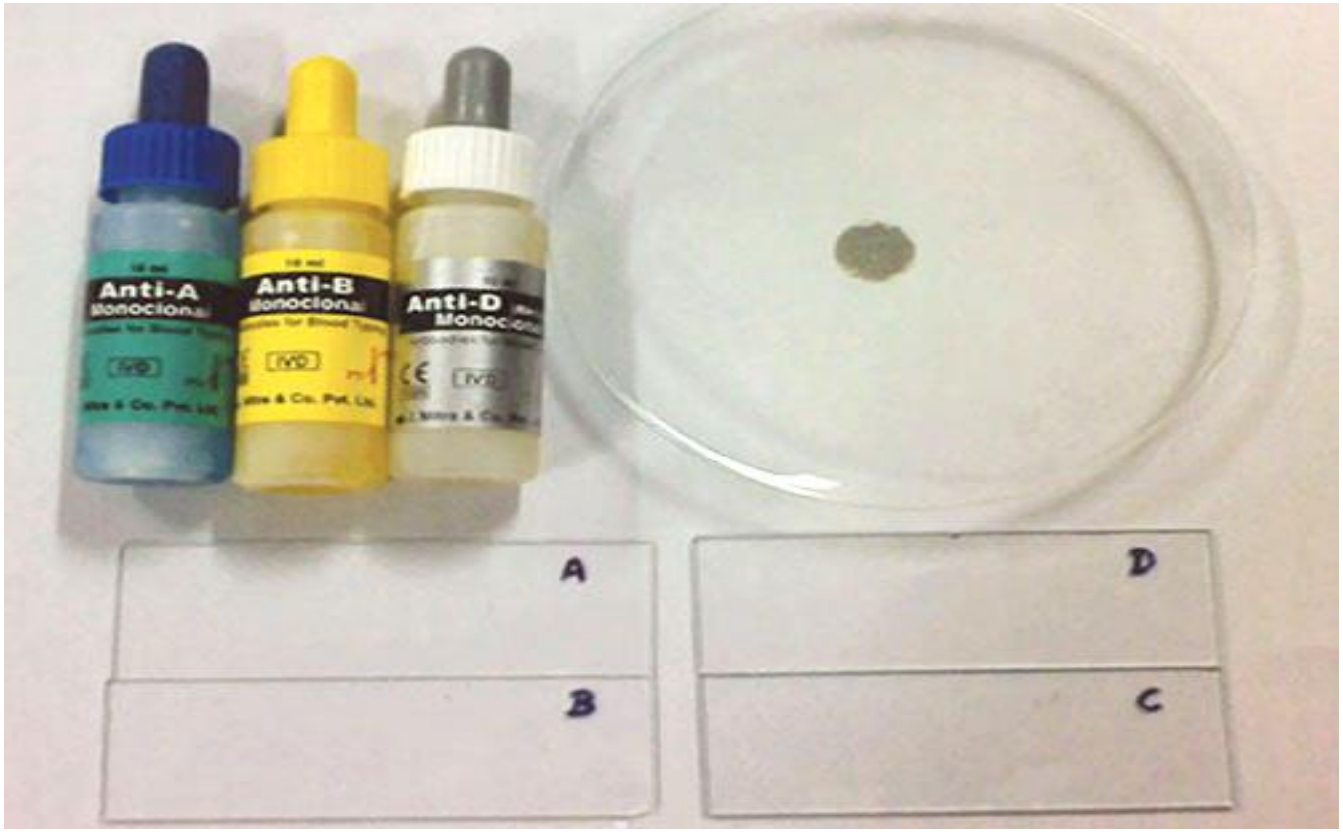
Allele from Parent 1	Allele from Parent 2	Genotype of Offspring	Blood-types of Offspring
I^A	I^A	$I^A I^A$	A
I^A	I^B	$I^A I^B$	AB
I^A	*i*	I^A i	A
I^B	I^A	$I^A I^B$	AB
I^B	I^B	$I^B I^B$	B
I^B	*i*	I^B i	B
i	*i*-	*ii*	O

Method using formal citrate:-

Core Concept

Apparatus and reagents:-

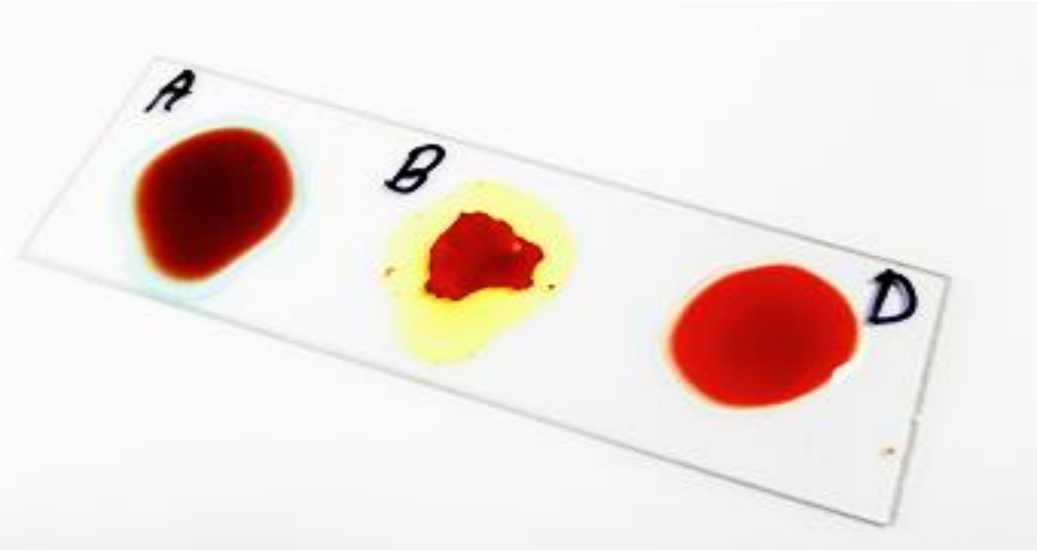
a. Glass slides or ceramic plate
b. Tooth pick
c. Anti A antiserum
d. Anti B antiserum



Core Concept

Procedure:-

1. Obtain clean microscope slides. Using a glass marking pencil, mark one end A and the other end B.
2. Prick the tip of the finger under sterile conditions to obtain blood and place one drop of blood on each end of the marked slide.
3. Add one drop of anti-A serum to the A side. Add one drop of anti-B serum to the side B.
4. Mix the antiserum and blood on each side with a tooth pick, using always a new stick for each side. Gently rock the slide back and forth and observe for any agglutination of red cells.



Core Concept

Procedure:-

5. If agglutination occurs on side 'A' only then blood type is group ’A’. and if - agglutination occurs on side 'B' only then blood group is 'B'. If agglutination occurs on both the sides, blood group is AB'. If no agglutination occurs on either side, the blood group is 'O'. Sometimes it is necessary to observe the cells under the microscope to ascertain the agglutination in doubtful cases. Agglutination generally looks like red pepper granules.

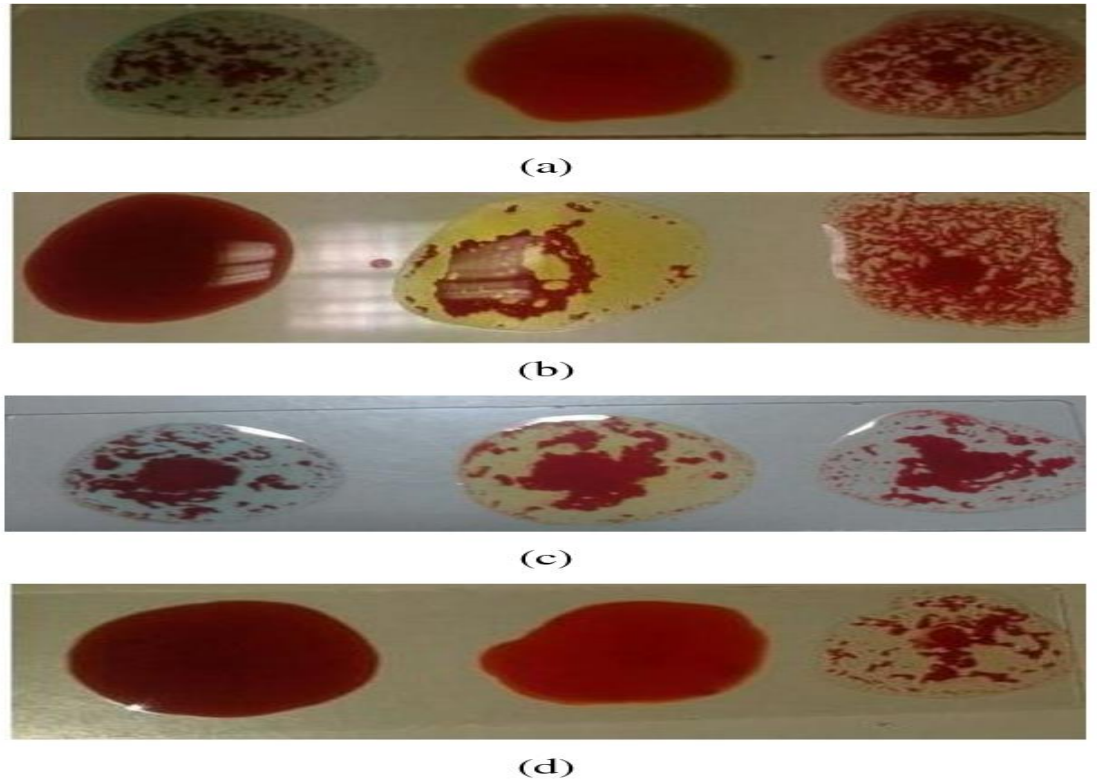


Fig.10. (a) Result of A+ (b) Result of B+
(c) Result of AB+(d) Result of O+

Core Concept

Precautions

a. Finger should be cleaned properly.

b. Prick should be moderately deep.

c. Avoid squeezing of finger and mixing of tissue fluid with the blood.

d. Lancet should not be greasy.

E. If clumping occurs then discard it and make a new slide.

F. Read the label for date of expiry on bottle of antisera.

G. The mixing of blood and serum should be quick to avoid coagulation. d. Make sure that you do not mix the anti-A and anti-B antisera.

Vertical integration

Vertical integration

Why are blood types important?

> Blood transfusion creating a wrong combination of donor-patient blood types could result in the death of the patient.

> Furthermore a wrong combination donor-patient of the rhesus factor could also provoke a rejection from the patient's body and result in death.

	CAN DONATE TO	CAN RECIEVE FROM
A+	A+, AB+	A+, A-, O+, O-
A-	A+, A-, AB+, AB-	A-, O-
B+	B+, AB+	B+, B-, O+, O-
B-	B+, B-, AB+, AB-	B-, O-
AB+ (universal recipient)	AB+	ALL GROUPS
AB-	AB+, AB-	A-, B-, AB-, O-
O+	A+, B+, AB+, O+	O+, O-
O- (universal donor)	ALL GROUPS	O-

Reference:- Google Image Blood Transfusion

Vertical integration

MOST IMPORTANT CLINICAL SIGNIFICANCE

BLOOD PRODUCT COMPATIBILITY

ABO Type	RBC Antigen	Plasma Antibodies	pRBCs	PLTs	FFP	Cryo
Type: Pt's blood + known antibodies → determines ABO/Rh type **Screen**: Pt's serum + RBCs with known antigens → finds common antibodies **Cross**: Pt's serum + donor cells mixed → agglutination means incompatible	ABO-gene derived cell surface **glycoproteins** which determine the ABO type. **Rh factor** is inherited separately from ABO.	Primarily **IgM** pentamers with **IgG** (more in type O) which can activate the **complement** cascade resulting in **hemolysis**.	~300 cc/bag stored at 1-6°C for 35-42 days	~50 cc/bag (pooled to 4-6 bags) stored at 20-24°C for 5 days.	~250 cc/bag stored at < -18°C for a year (use within 24 hours once thawed)	~15 cc/bag (pooled to 4-6 bags) stored at < -18°C for a year (use within 6 hours once thawed)
			The following ABO blood product **compatibilities** are shown as first and second choice(s) in the first and second rows, respectively. These can be adjusted based on blood bank inventory, antibody titers, etc. **Rh factor: Rh+ patients** can receive Rh+ OR Rh- products whereas **Rh- patients** ideally receive Rh- products.			
 40%			A O	A B, AB, O	A AB	A B, AB, O
 10%			B O	B A, AB, O	B AB	B A, AB, O
 5%		none	AB A, B, O	AB A, B, O	AB	AB A, B, O
 45%	none		O	O A, B, AB	O A, B, AB	O A, B, AB

Abbreviations: packed red blood cells (**RBCs**), platelets (**PLTs**), fresh frozen plasma (**FFP**), cryoprecipitate (**cryo**)
Source: Joint United Kingdom (UK) Blood Transfusion and Tissue Transplantation Services Professional Advisory Committee (JPAC)

Acute Hemolytic Transfusion Reaction: *Signs and Symptoms*

Vertical integration

ABO incompatibility
(can be due to error in patient identification, improper labelling of specimens/donations, or testing errors)

Non-ABO blood group incompatibilities
(commonly from exposure to prior transfusions or pregnancy)

Classical complement pathway activation on RBC surface

IgG antibody activation against antigens on donor RBC

RBCs bound and destroyed by macrophages in liver and spleen

Release of RBC stroma

Release of histamine from mast cells

Intravascular hemolysis

Extravascular hemolysis

Constriction of bronchial smooth muscle

Vasodilation, increased vascular permeability

Free hemoglobin is released into plasma

Release of IL-1, IL-6, IL-8, and TNF from leukocytes

Systemic endothelial damage and release of thromboplastic material

Activation of Factor XII and intrinsic clotting cascade

Respiratory distress

Skin flushing

Hypotension

Free hemoglobin passes through glomeruli

Increase in hypothalamic thermoregulatory set-point

DIC

Compensatory increase in heart rate

Reflex tachycardia

Anxiety

Diffuse bleeding

Ischemic tissue damage

Renal reabsorption capacity is exceeded

Fever

Chills

Hemoglobinuria

RBC stroma obstructs renal glomeruli

Flank pain

Renal failure

Abbreviations:
- **DIC:** disseminated intravascular coagulation
- **IgG:** immunoglobulin G
- **IL:** interleukin
- **RBC:** red blood cell
- **TNF:** tumor necrosis factor

Notes:
- *Reported incidence of 1 per 1.8 million transfusions*
- *Fever may be the only presenting sign of an acute hemolytic transfusion reaction*
- *Risk of death is less than 10% and is connected to volume of incompatible blood transfused*

Biomedical Ethics

Beneficence

> Obligations to preserve life, restore health, relieve suffering and maintain function.

> TO DO “GOOD”.

> NON ABANDONMENT – OBLIGATIONS TO PROVIDE ONGOING CARE

Conflict of interest :-

must not engage in activities that are not in patients best interest

Codes of Ethics for Laboratory

Testing laboratories have an obligation to adhere to high ethical standards in order to provide with the accurate and reliable test results needed to meet the requirements and reduce uncertainty in results.

- Always wear white coat in lab
- Handle the glassware gently
- Wait for your turn while working in groups
- Use gloves while handling chemicals
- Do not waste reagents or other lab supplies

MORAL DUTIES OF DOCTOR

- The duty to help cure
- The duty to promote and protect the patient's health.
- The duty to inform
- The duty to confidentiality
- The duty to protects patients life
- The duty to protect the patient's life
- The duty to respect the patient's autonomy
- The duty to protect privacy
- The duty to respect the patients dignity.

Brain Storming

Question & Answer

Brain Storming

Clinical Scenario

A 32-year-old woman presents to the hospital in labor. Her blood type is O negative, and she has received anti-D immunoglobulin during pregnancy due to Rh(D) incompatibility with her previous child. The father's blood type is A positive. After delivery, the newborn's blood type is determined to be A positive. The baby appears well initially but develops jaundice on the second day of life. Laboratory tests reveal elevated bilirubin levels. The Coombs test is negative.

Brain Storming

QUESTIONS

Question 1 : What is the most likely cause of the newborn's jaundice?

Answer:

The most likely cause of the newborn's jaundice is ABO incompatibility between the mother (O negative) and the newborn (A positive).

Brain Storming

QUESTIONS

Question 2 : How would you further investigate and manage this case?

Answer:

Further investigation and management may include:

- Monitoring bilirubin levels to assess the severity of jaundice.
- Phototherapy to reduce bilirubin levels and manage jaundice.
- Exchange transfusion if bilirubin levels become dangerously high or if other signs of hemolysis occur.
- Close monitoring of the newborn's clinical condition, including physical examination and laboratory tests.

Brain Storming

QUESTIONS

Question 3 : What potential complications might arise due to this ABO incompatibility?

Answer:

Potential complications of ABO incompatibility include:
Hyperbilirubinemia leading to severe jaundice, which can cause brain damage (kernicterus) if untreated.
Hemolytic disease of the newborn, although less severe compared to Rh(D) incompatibility.
Anemia, hepatosplenomegaly (enlargement of the liver and spleen), and generalized edema in severe cases.
Long-term effects are generally rare but may include learning disabilities or mild developmental delays in rare instances of severe ABO incompatibility.

Suggested Research Article

Related Research Article

https://www.hindawi.com/journals/bmri/2021/6629060/

Human ABO Blood Groups and Their Associations with Different Diseases

Silamlak Birhanu Abegaz [1]
Show more

Academic Editor: Sercan Erg n

Abstract

Introduction. Human ABO blood type antigens exhibit alternative phenotypes and genetically derived glycoconjugate structures that are located on the red cell surface which play an active role in the cells' physiology and pathology. Associations between the blood type and disease have been studied since the early 1900s when researchers determined that antibodies and antigens are inherited. However, due to lack of antigens of some blood groups, there have been some contentious issues with the association between the ABO blood group and vulnerability to certain infectious and noninfectious diseases. *Objective*. To review different literatures that show the association between ABO blood groups and different diseases. *Method*. Original, adequate, and recent articles on the same field were researched, and the researcher conducted a comprehensive review on this topic. Thus, taking out critical discussions, not only a descriptive summary of the topic but also contradictory ideas were fully retrieved and presented in a clear impression. In addition, some relevant scientific papers published in previous years were included. The article search was performed by matching the terms blood types/groups with a group of terms related to different diseases. The articles were screened and

HOW TO ACCESS DIGITAL LIBRARY

- **Steps to Access HEC Digital Library**

1. Go to the website of HEC National Digital Library.
2. On Home Page, click on the INSTITUTES.
3. A page will appear showing the universities from Public and Private Sector and other Institutes which have access to HEC National Digital Library HNDL.
4. Select your desired Institute.
5. A page will appear showing the resources of the institution
6. Journals and Researches will appear
7. You can find a Journal by clicking on JOURNALS AND DATABASE and enter a keyword to search for your desired journal.

Link:https://www.topstudyworld.com/2020/05/access-hec-digital-library.html?m=1

HOW TO APPLY FOR HEC DIGITAL LIBRARY IF YOU ARE NOT MEMBER

- If you are not a member of HEC national digital library then you can fill the application after downloading it from

http://www.digitallibrary.edu.pk/app_form.html

- Fill the form that will appear like this
- Mail it to mailto:

REFERENCES

1. Professor Saqib Sohail Manual of physiology.
2. Guyton and Hall text book of medical physiology, 14th edition
3. Human Physiology by Lauralee Sherwood, 9th edition
4. Ganong's Review of Medical Physiology, 26th Edition
5. Berne and Levy Physiology-7th Edition
6. Costanzo Physiology by Linda S. Costanzo, 7th edition
7. Human Physiology: An Integrated Approach, by Silverthorn-8th edition
8. Best & Taylors Physiological Basis of Medical Practice-13th Edition
9. Google Images And Wikipedia Illustrations for Understanding.


Thank You!