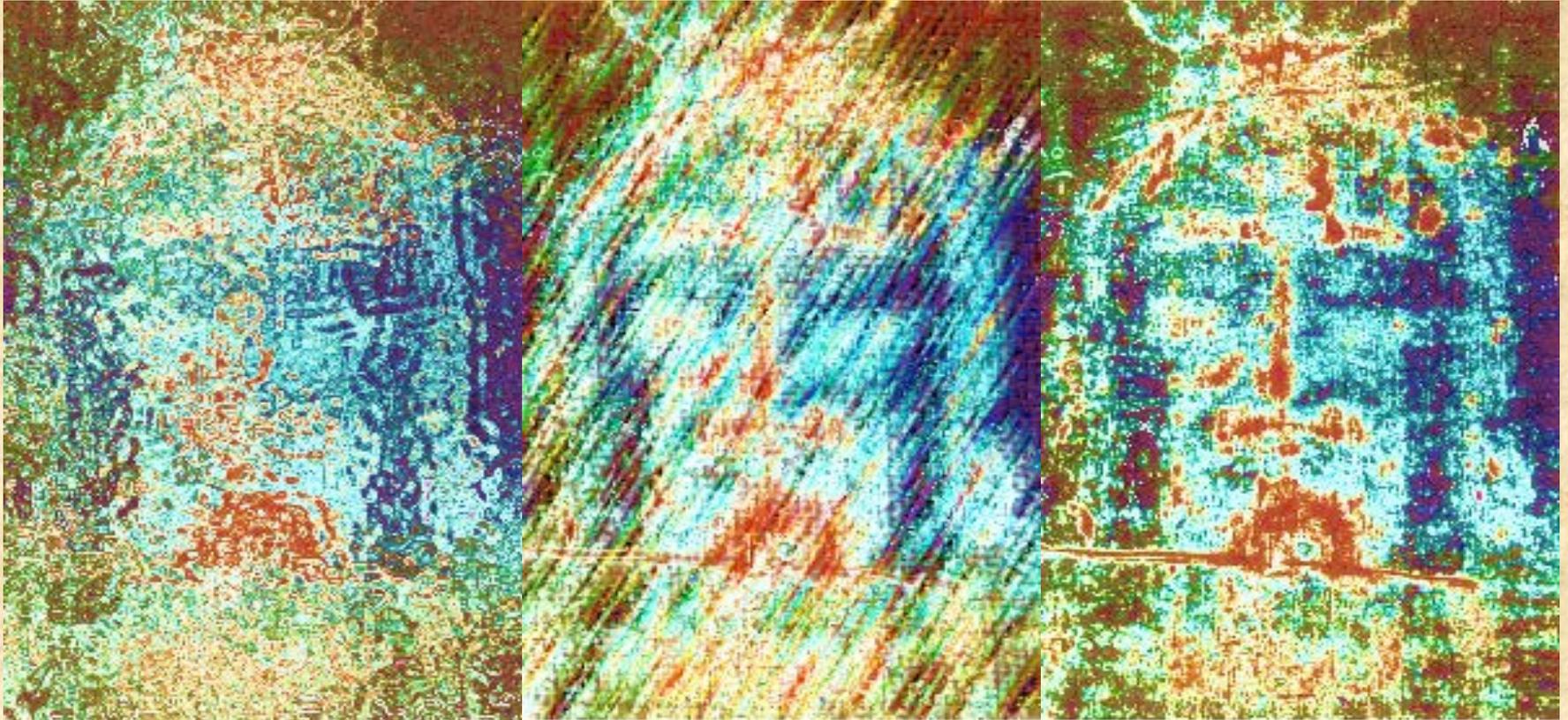


A Critical (Re)evaluation of the Shroud of Turin Blood Data:

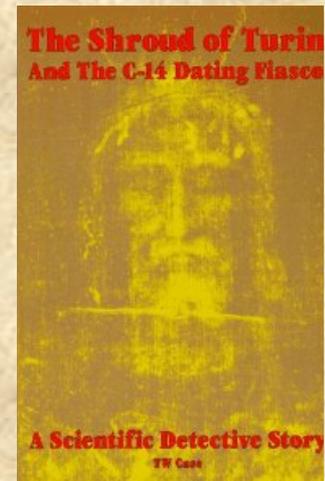


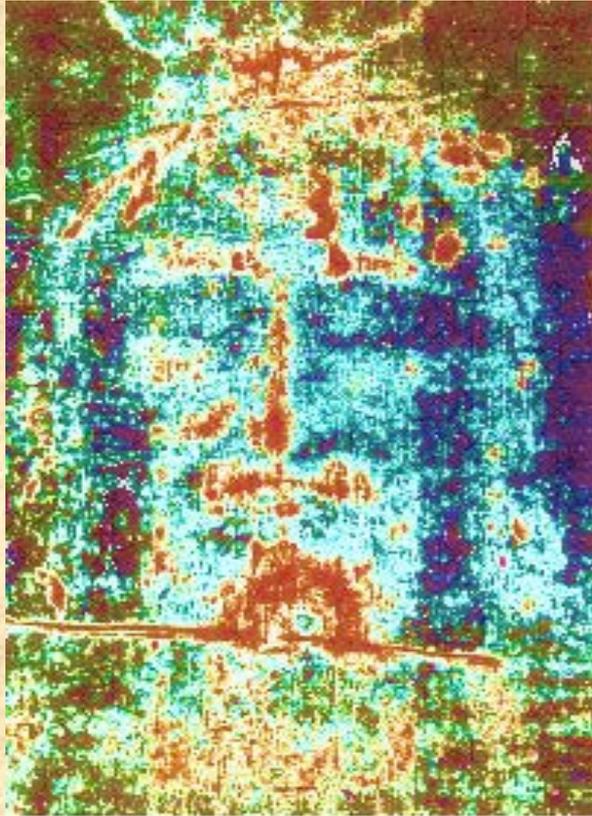
Strength of Evidence in the
Characterization of the Bloodstains

“The fact is, it meets a lot of the test criteria; it meets the descriptions that were required by the medical people, as to what it should look like.

But the most interesting thing is now there is immunological evidence that it is primate blood.”

-Heller & Adler interview 1995



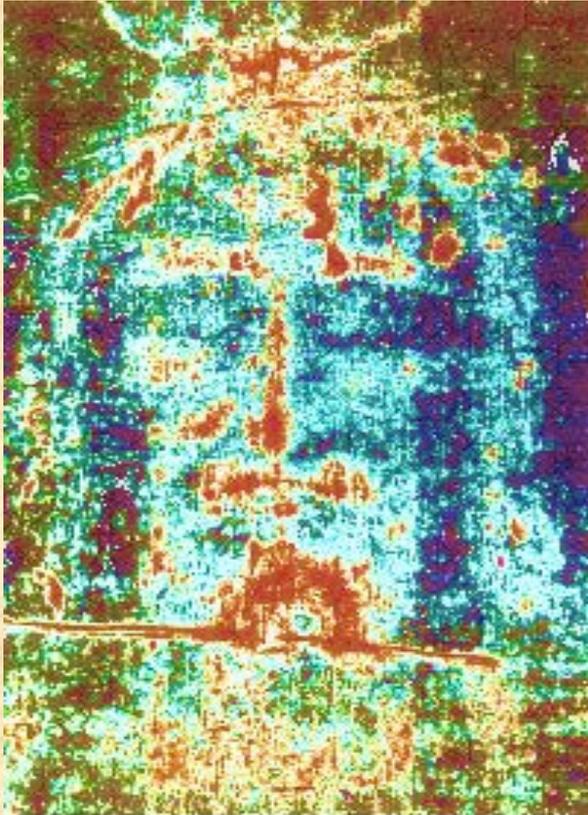


Is blood really present?

What type of blood is it?

Whose blood is it?

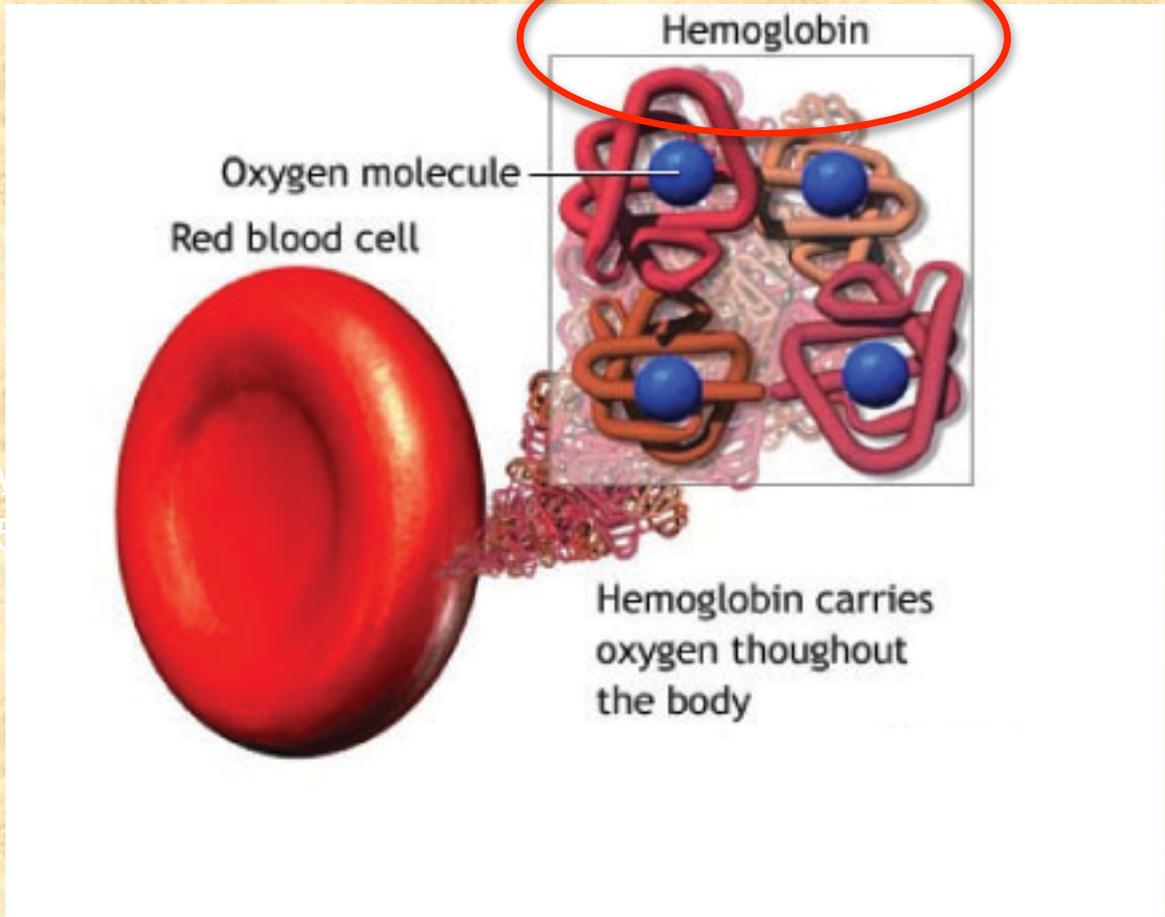
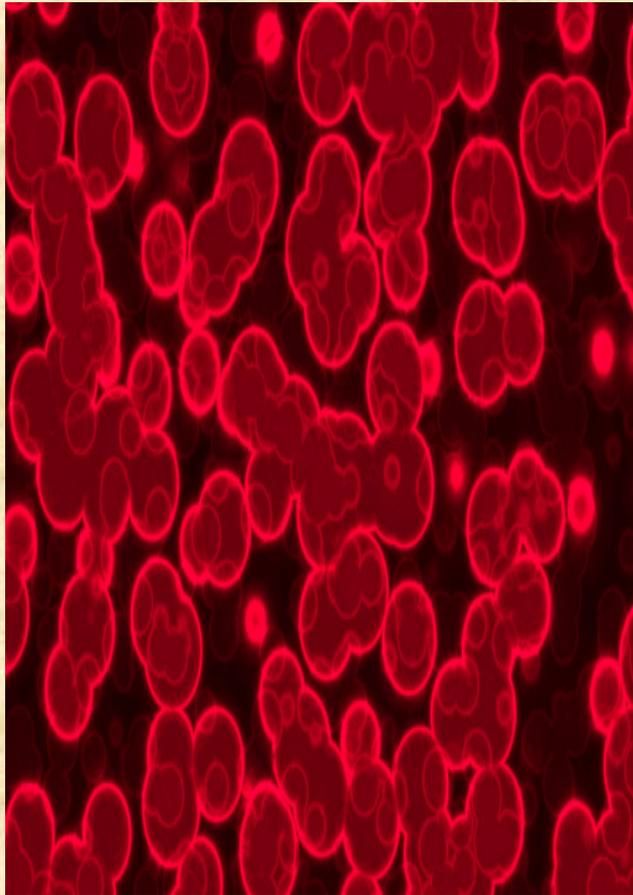
Is blood really present?



Chemical Methods
Spectroscopy Methods

Immunological Methods
Molecular Biology Methods
(DNA Techniques)

Is blood really present?



A single RBC contains
~ 250 million Hb molecules

Chemical Methods

Heme → Substrate

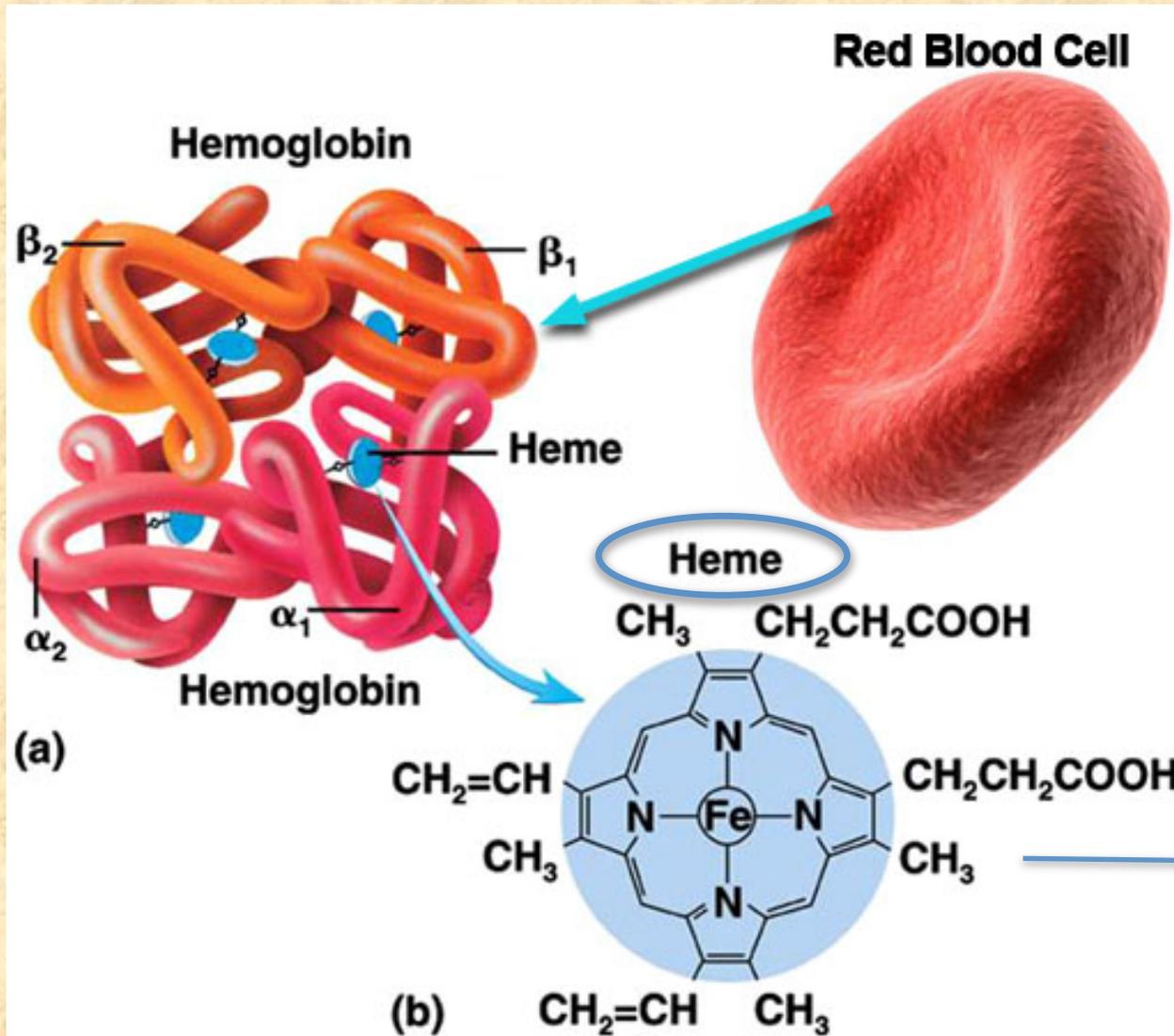


Chemical Methods

Heme → Substrate

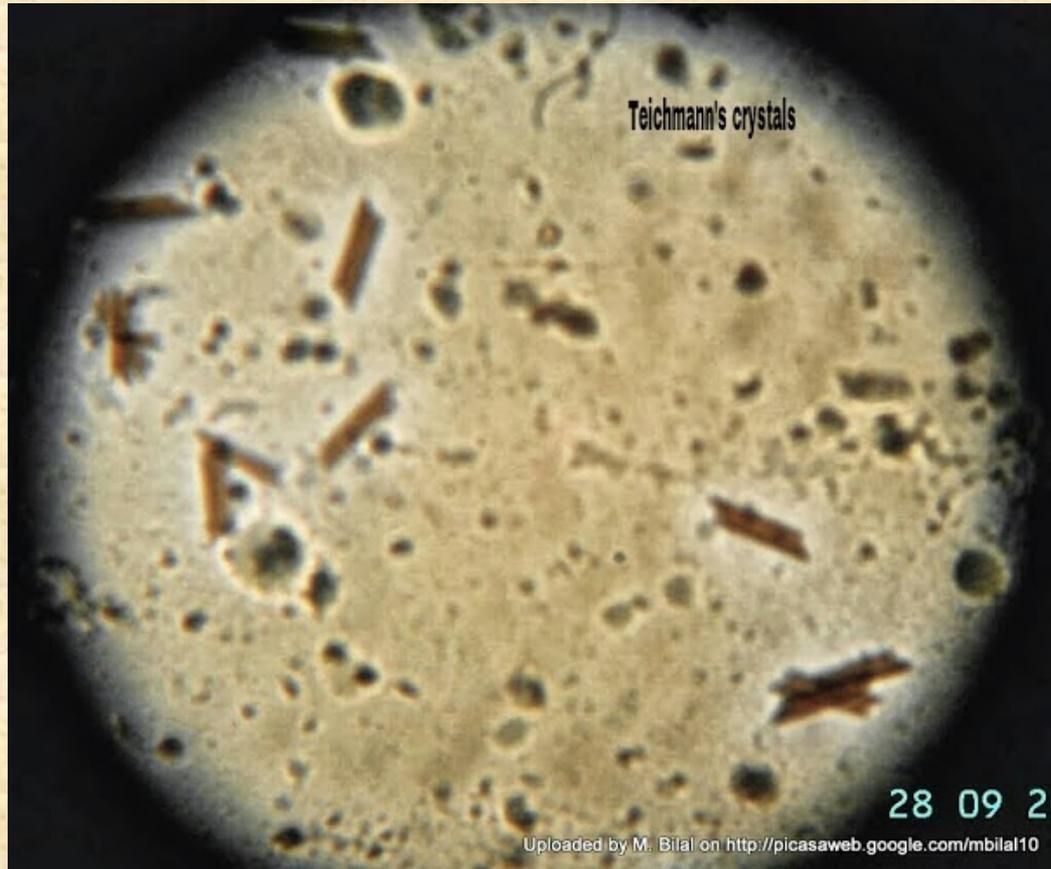


Chemical Methods



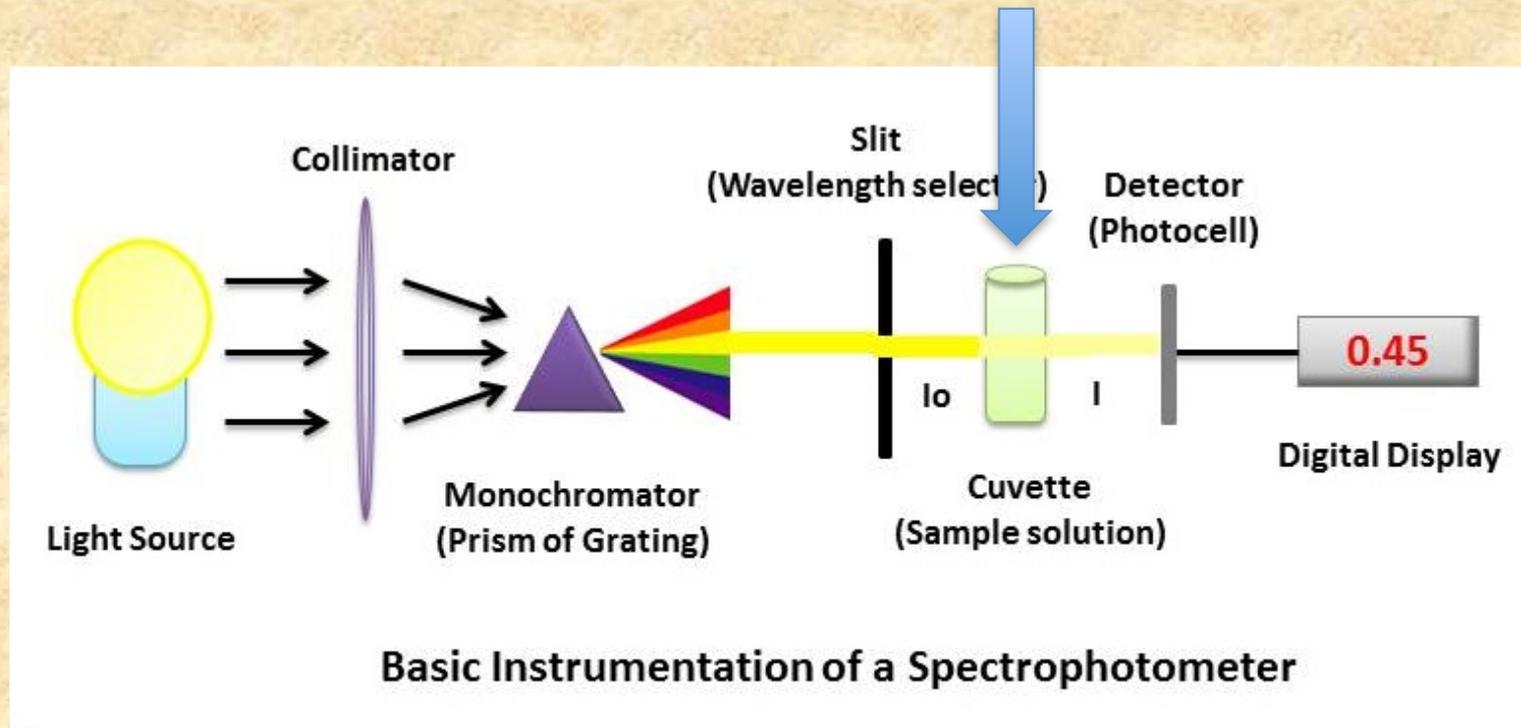
Chemical Methods

Heme → **AA + Salt** → **Crystal Formation**

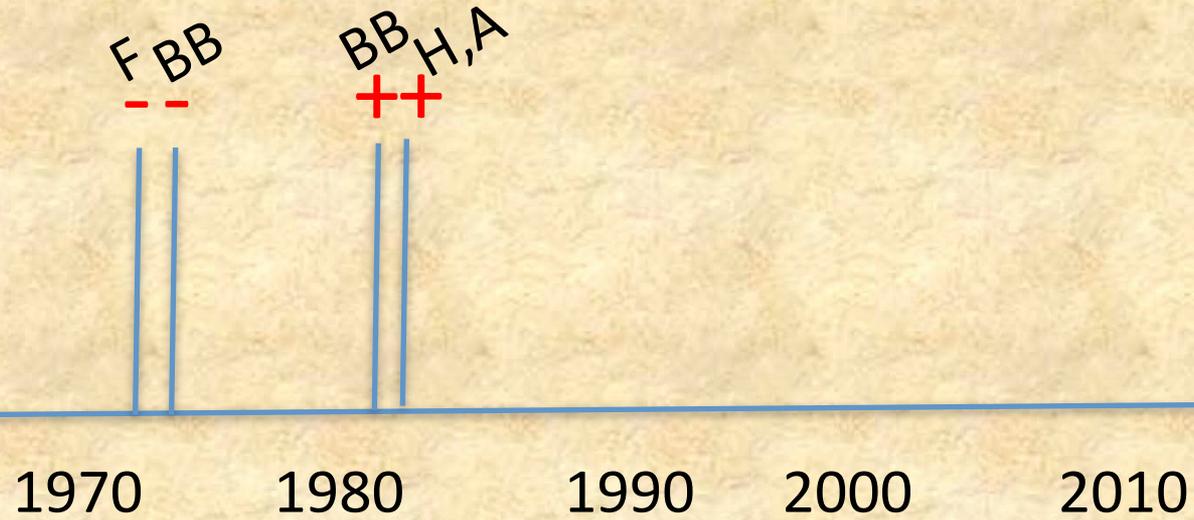
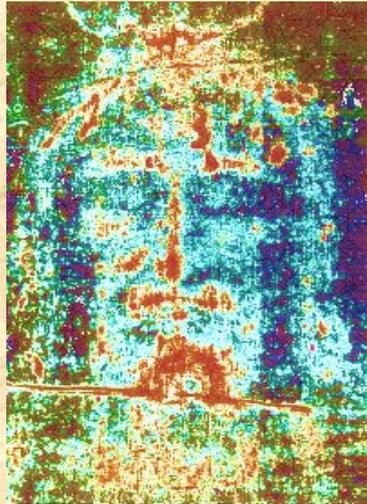


Spectroscopy Methods

Sample

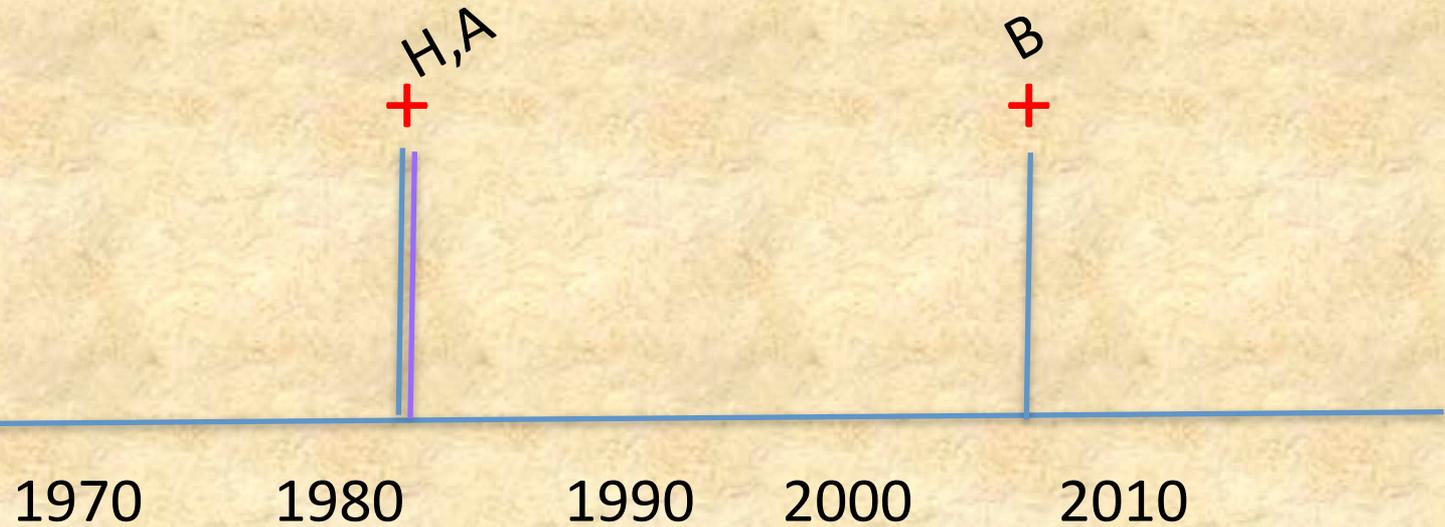
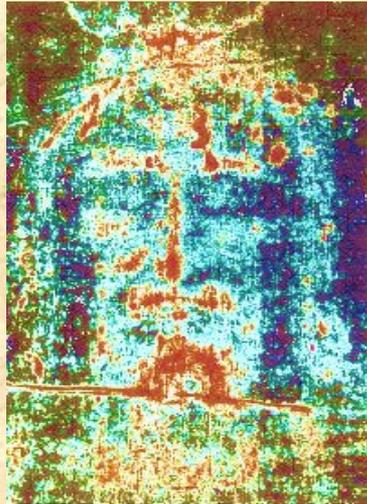


Detection of Hemoglobin (Heme)



Chemical Methods

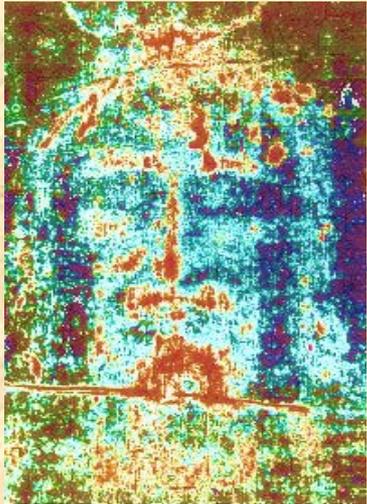
Detection of Hemoglobin (Heme)



Spectroscopy Methods

Breakdown products: biverdin, bilirubin
(Chemical methods)

Detection of Hemoglobin (Heme)



1970

1980

1990

2000

2010

GV
+

Immunochemical Methods

Is blood really present?

Hemoglobin (Heme)



Non-heme constituents

What type of blood is it?

Immunological Methods

Species: Animal, Human

Blood type: A, B, AB, O

What type of blood is it?

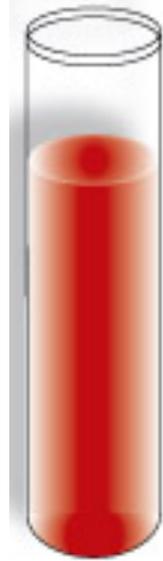
Immunological Methods

Species: Animal, Human

Blood type: A, B, AB, O

DNA Methods

Above plus if Male or Female

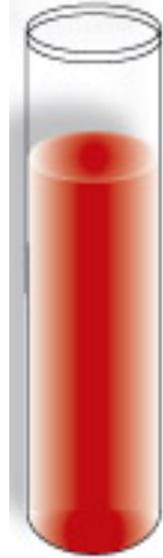


Whole
Blood



Serum

Cells

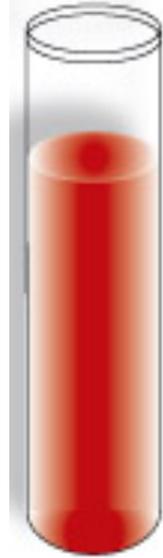


Whole
Blood



Serum → Proteins

Cells



Whole
Blood



Serum → Proteins

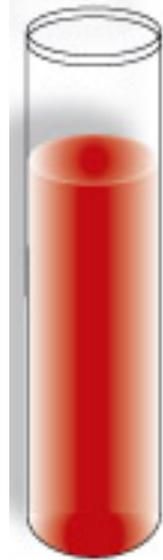
Cells



Albumin



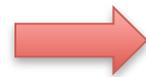
Antibody



Whole
Blood



Serum



Proteins

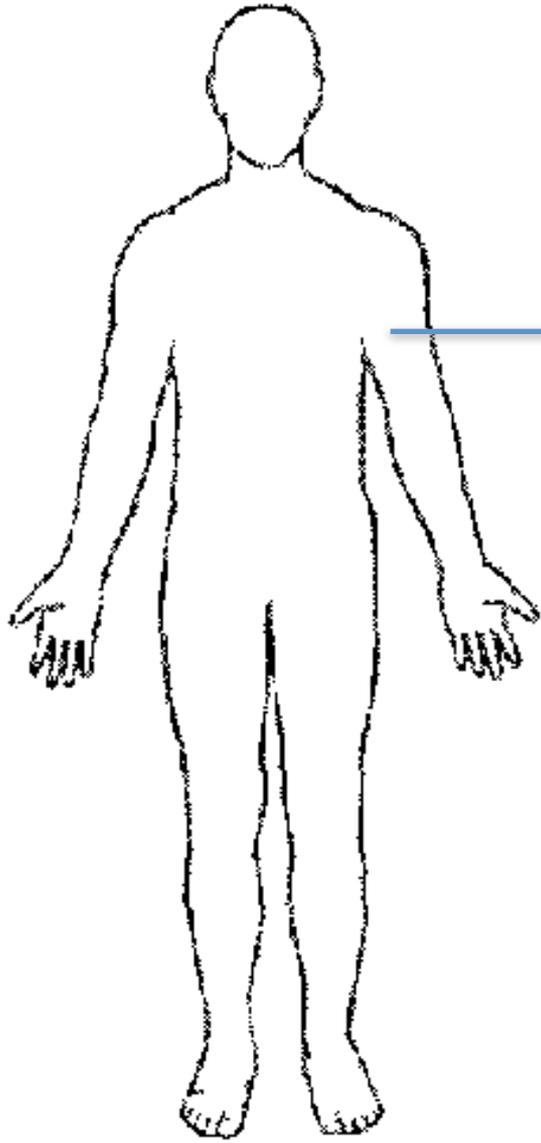
Cells



Albumin



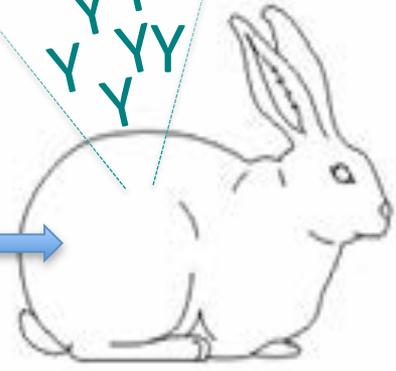
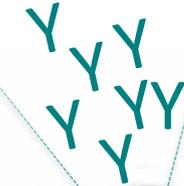
Antibody



Rabbit anti-human albumin antibodies



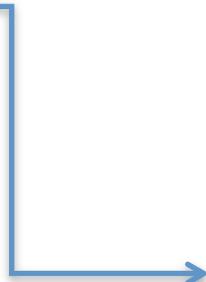
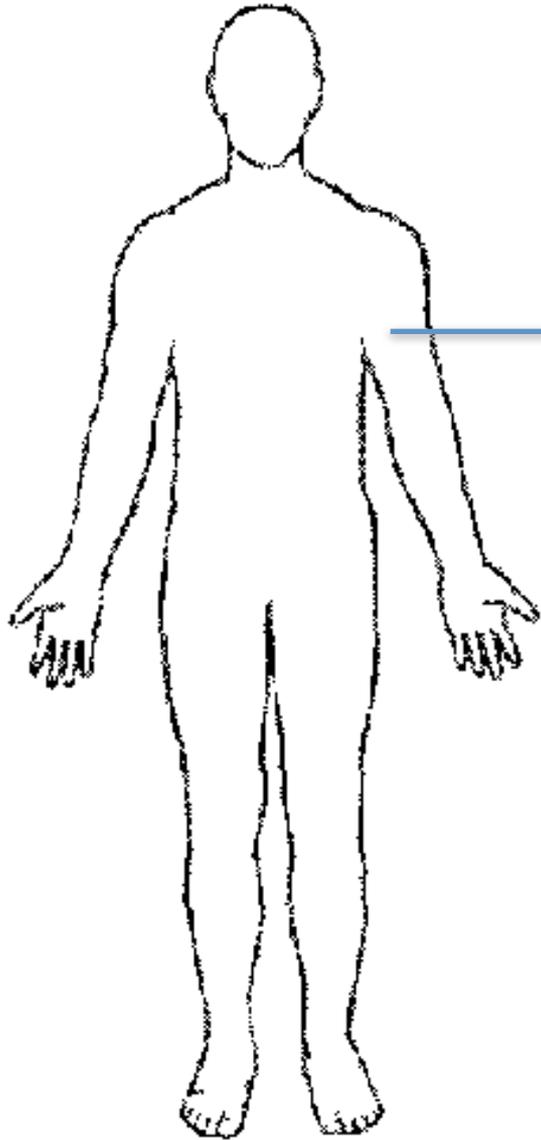
Albumin



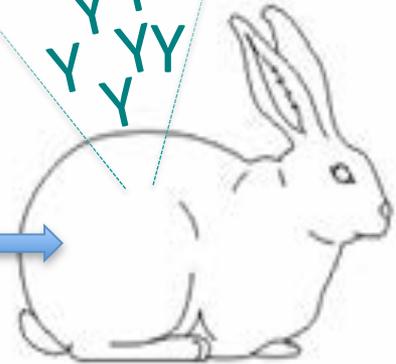
Made in

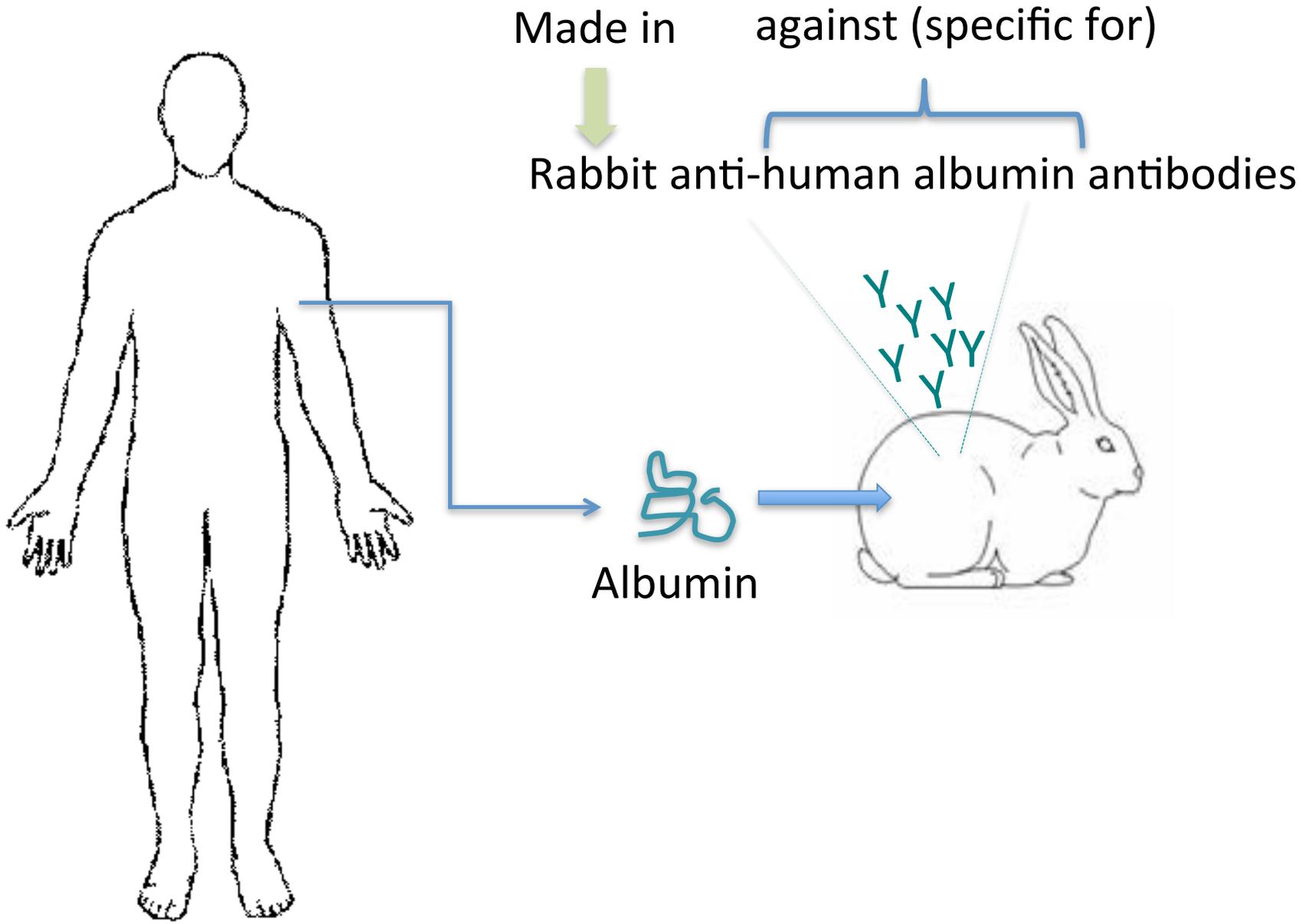


Rabbit anti-human albumin antibodies



Albumin





Immunohistochemistry



Antibody

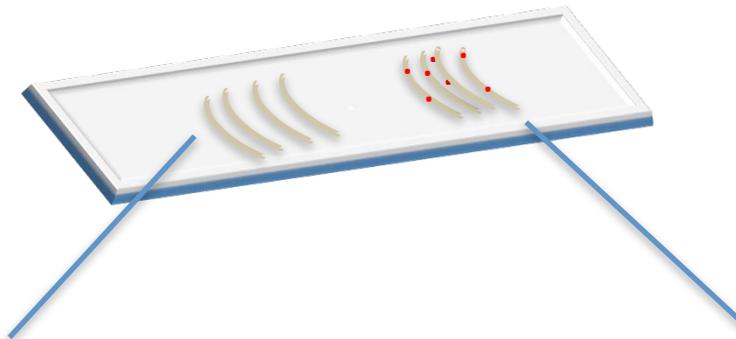
Immunohistochemistry



Antibody

Labeled with Fluorescent Tag or Chemical Tag

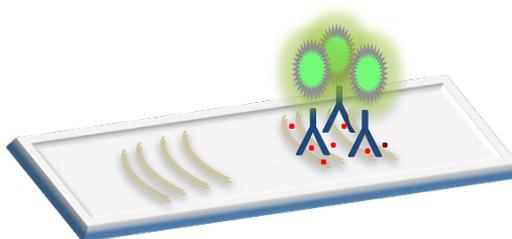
Immunohistochemistry



Unstained fibers
(control)

Bloodstained fibers

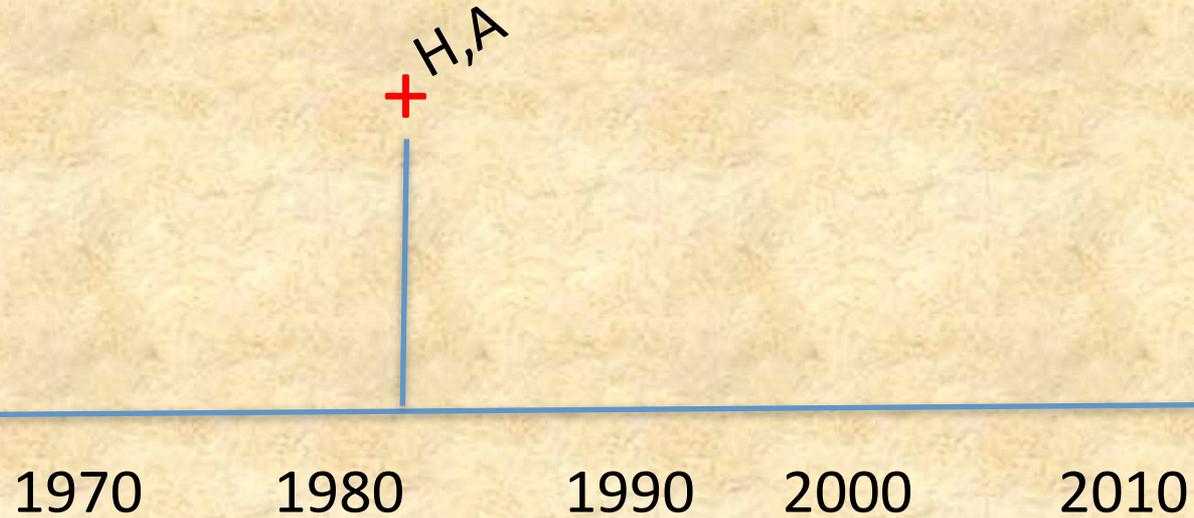
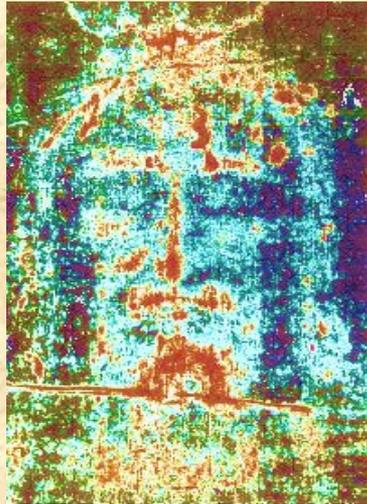
Immunochemistry



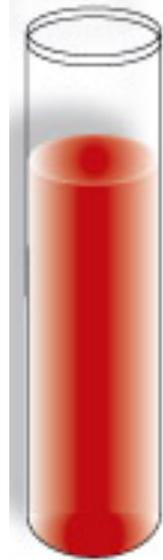
Control fibers: Negative
Bloodstained fibers: Positive

Results suggest albumin is
present in bloodstained fibers

Detection of Serum Albumin



Immunological Methods
(Chemical Methods)



Whole
Blood



Serum



Proteins

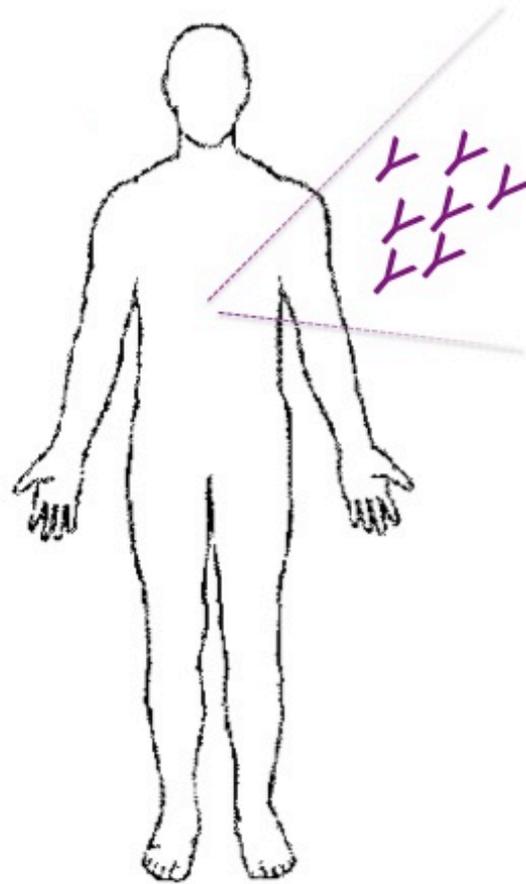
Cells



Albumin



Antibody



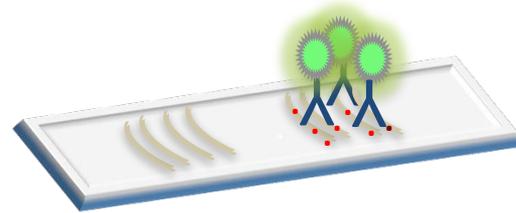
IgG
IgM
IgA
IgD
IgE

Antibody = Immunoglobulin (Ig)

Immunochemistry



Labeled Specific Antibody
Specific for Human Ig

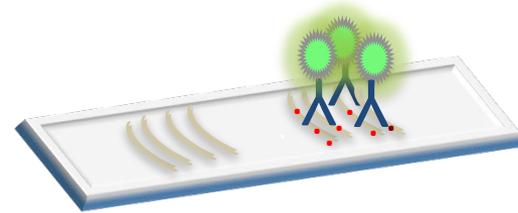


Immunofluorescence observed
(Only on bloodstained fibers)

Immunochemistry



Labeled Specific Antibody



Immunofluorescence observed

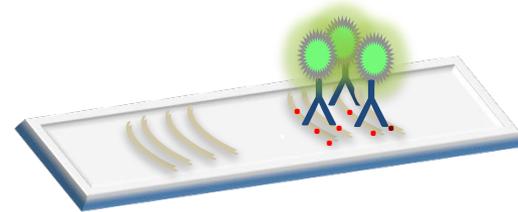


Labeled Nonspecific Antibody

Immunochemistry



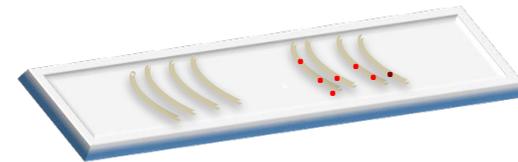
Labeled Specific Antibody



Immunofluorescence observed



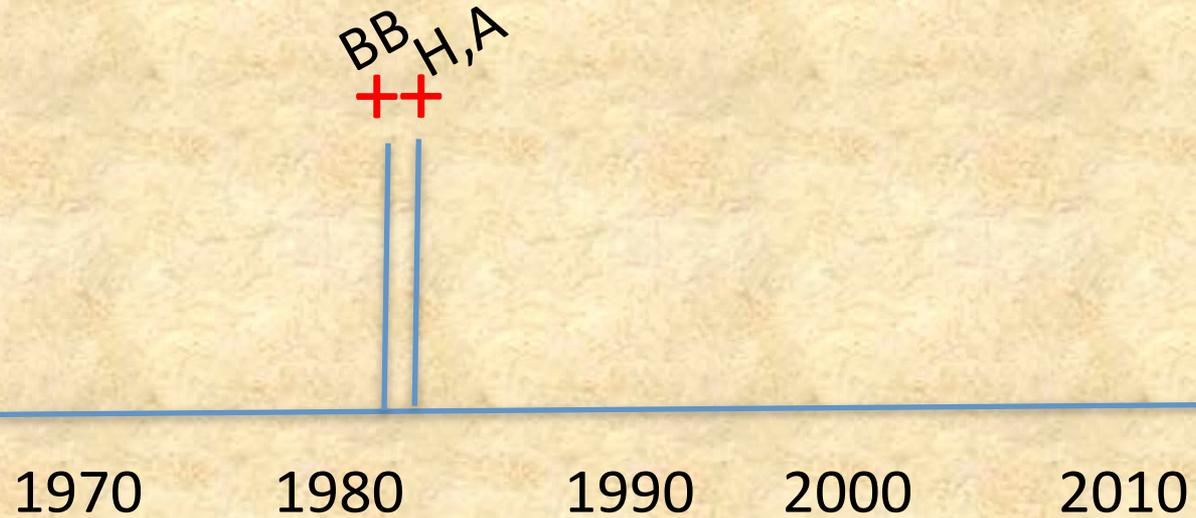
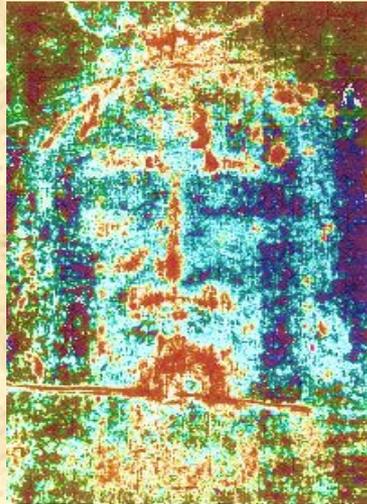
Labeled Nonspecific Antibody



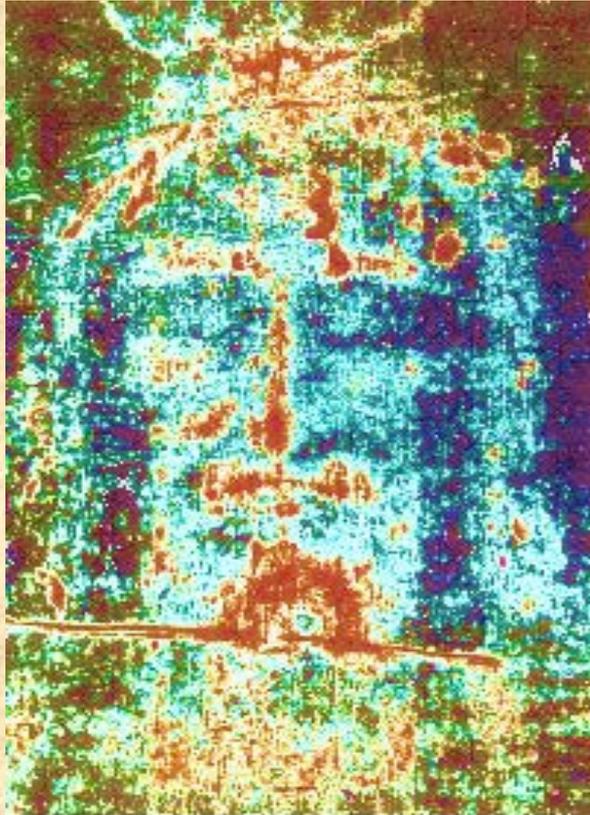
No Immunofluorescence observed

Controls demonstrate that
antibody binding was specific

Detection of Serum Immunoglobulin

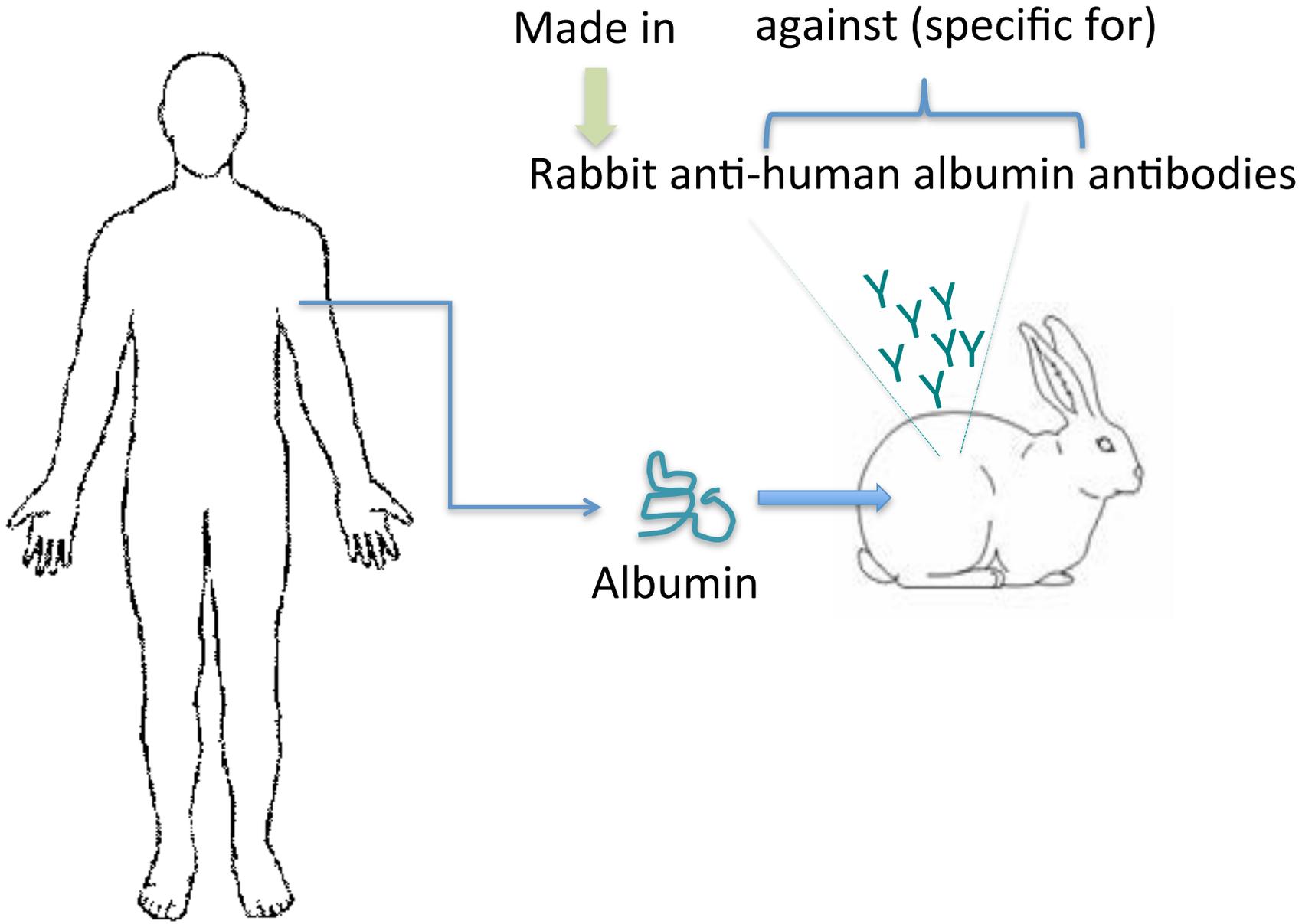


Immunochemical Methods

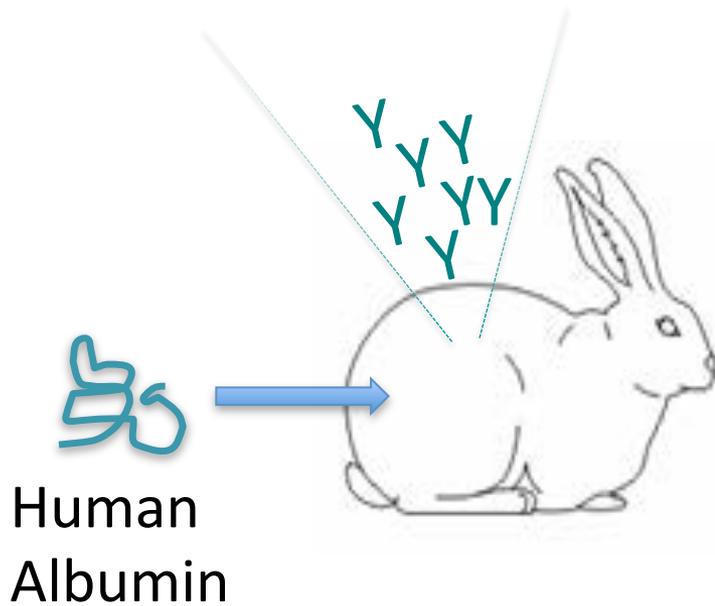


What type of blood is it?

Anti-human albumin
Anti-human immunoglobulin



Rabbit anti-human albumin



  Human Albumin

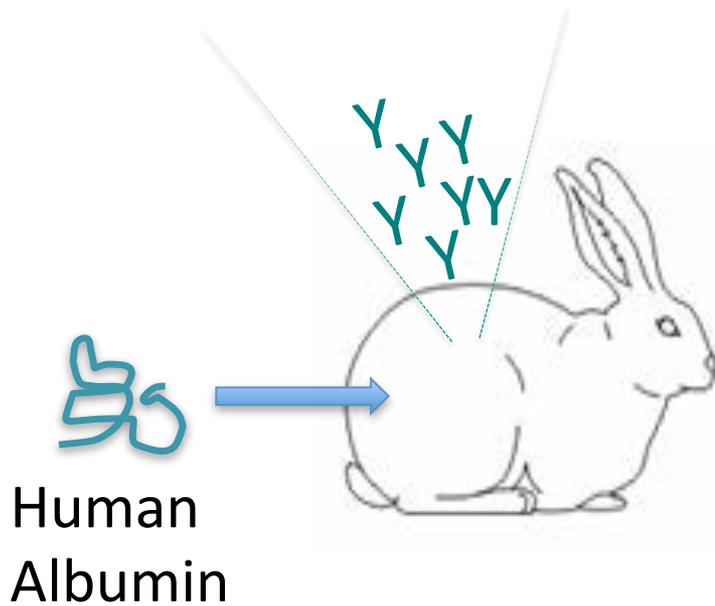
 Chimp Albumin

 Baboon Albumin

 Cow Albumin

 Horse Albumin

Rabbit anti-human albumin



✓  Human Albumin

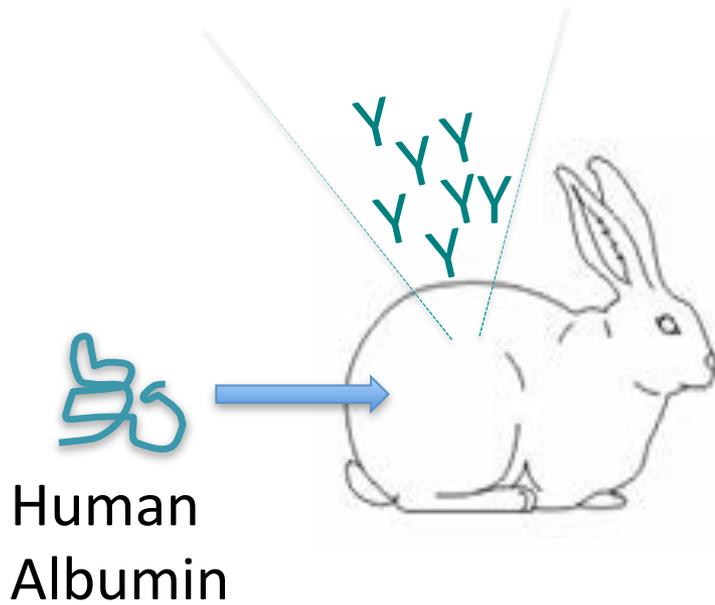
✓  Chimp Albumin

 Baboon Albumin

 Cow Albumin

 Horse Albumin

Rabbit anti-human albumin



  Human Albumin

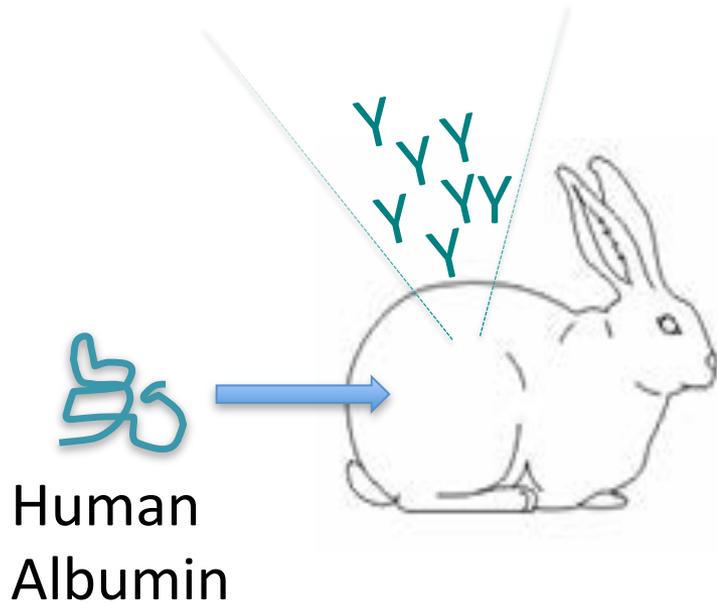
  Chimp Albumin

  Baboon Albumin

 Cow Albumin

 Horse Albumin

Rabbit anti-human albumin



✓  Human Albumin

✓  Chimp Albumin

✓  Baboon Albumin

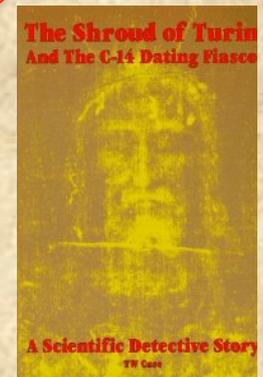
—  Cow Albumin

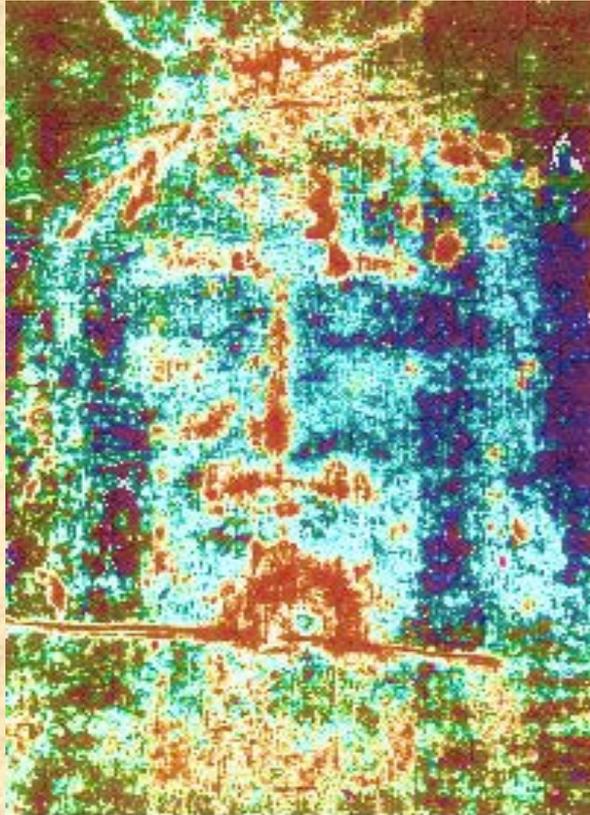
—  Horse Albumin

“The fact is, it meets a lot of the test criteria; it meets the descriptions that were required by the medical people, as to what it should look like.

But the most interesting thing is now there is immunological evidence that it is primate blood.”

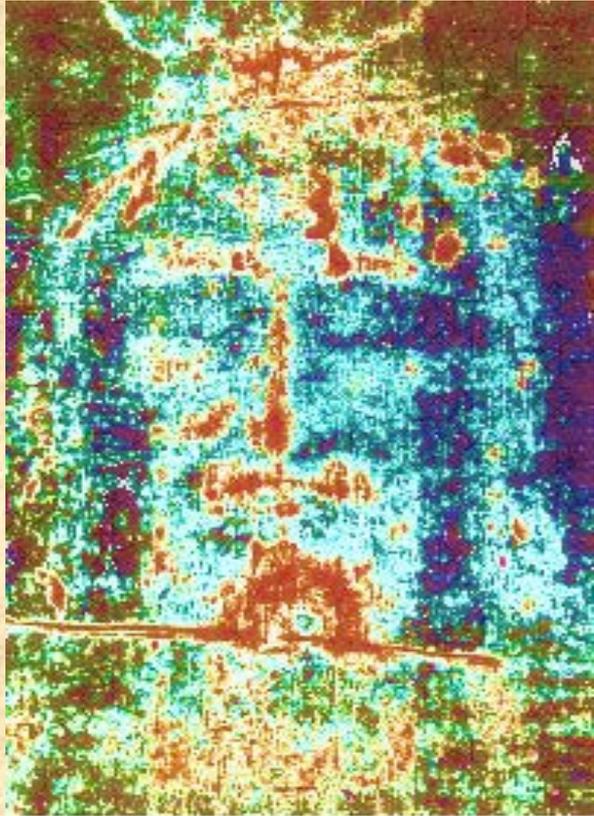
-Heller & Adler interview 1995





What type of blood is it?

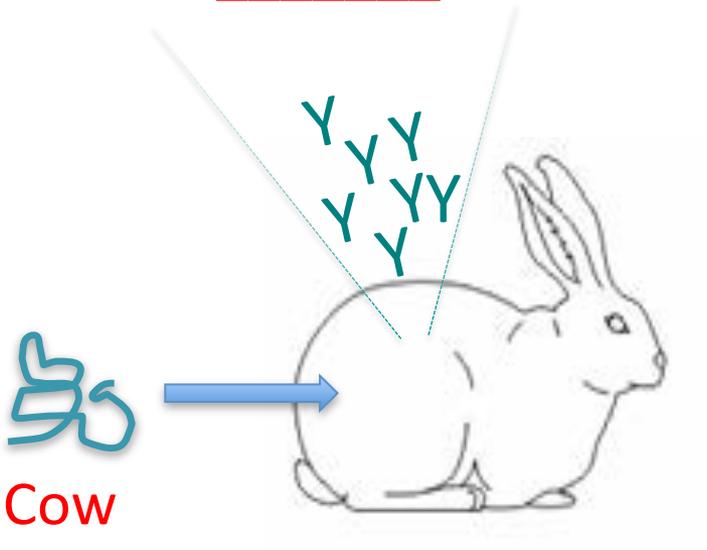
Cross-reactivity
precludes a definitive
answer (human blood)
from these results



What type of blood is it?

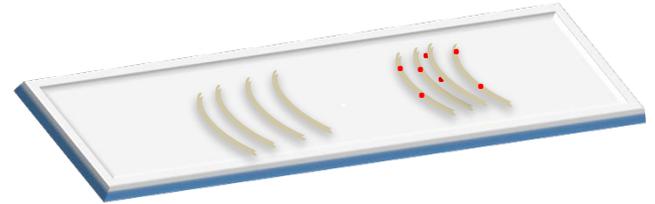
Important to note that while studies have examined the possibility that human (primate) blood may be present, technically it is unknown if blood from other animal types may also exist

Rabbit anti-_____ albumin



- Cow
- Chicken
- Goat
- Dog
- Other

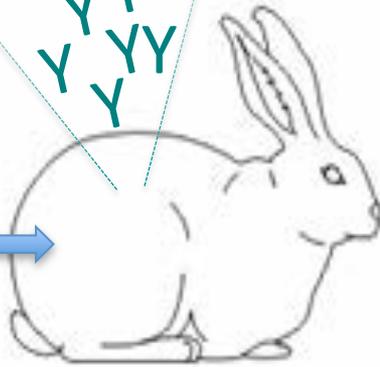
Rabbit anti-_____ albumin



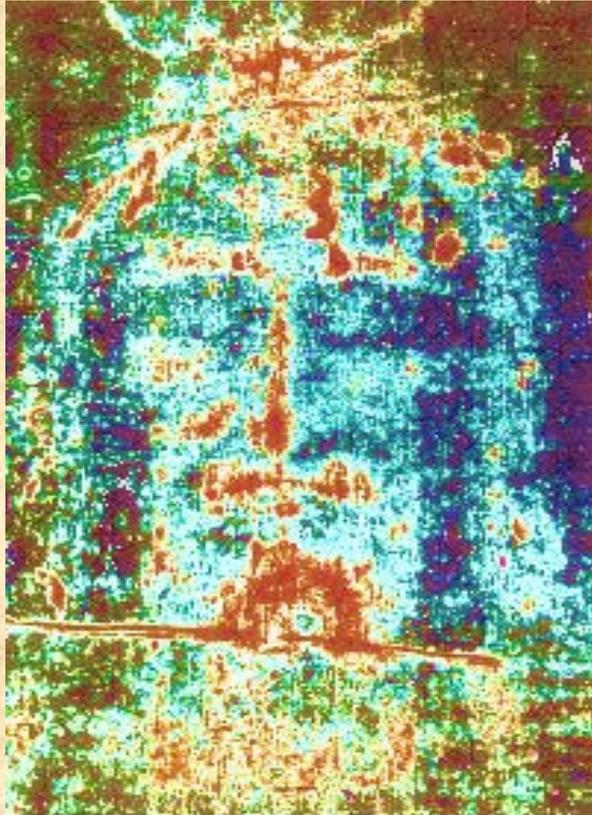
Shroud fibers



Y
Y
Y
Y
Y
Y
Y
Y



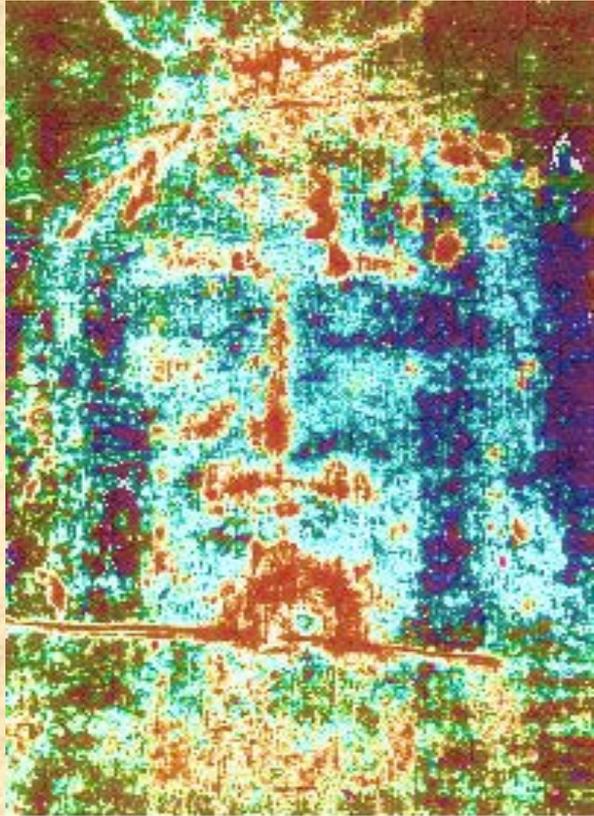
- Cow
- Chicken
- Goat
- Dog
- Other



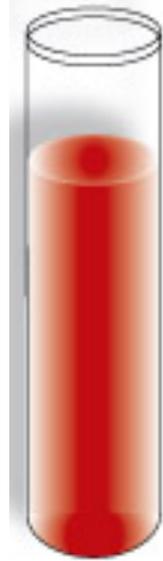
What type of blood is it?

Primate blood has been detected

Primate blood components are present



ABO Blood Typing



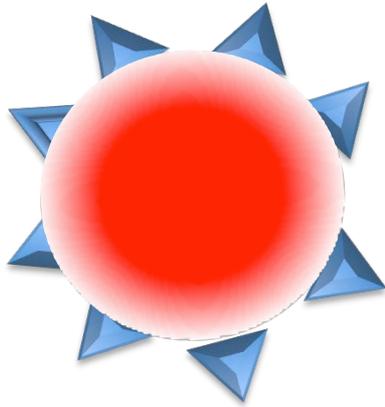
Whole
Blood



Serum

Cells

RBC



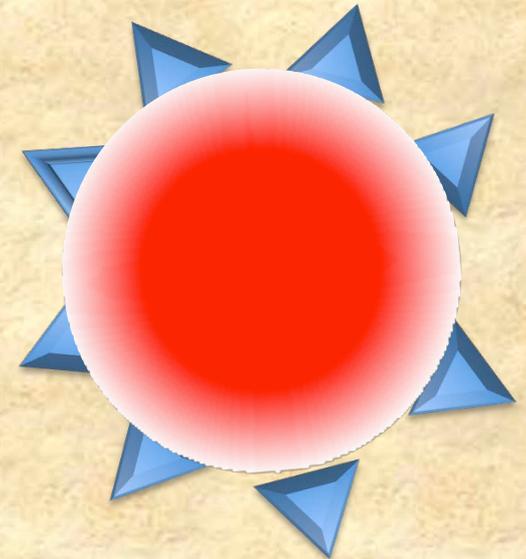
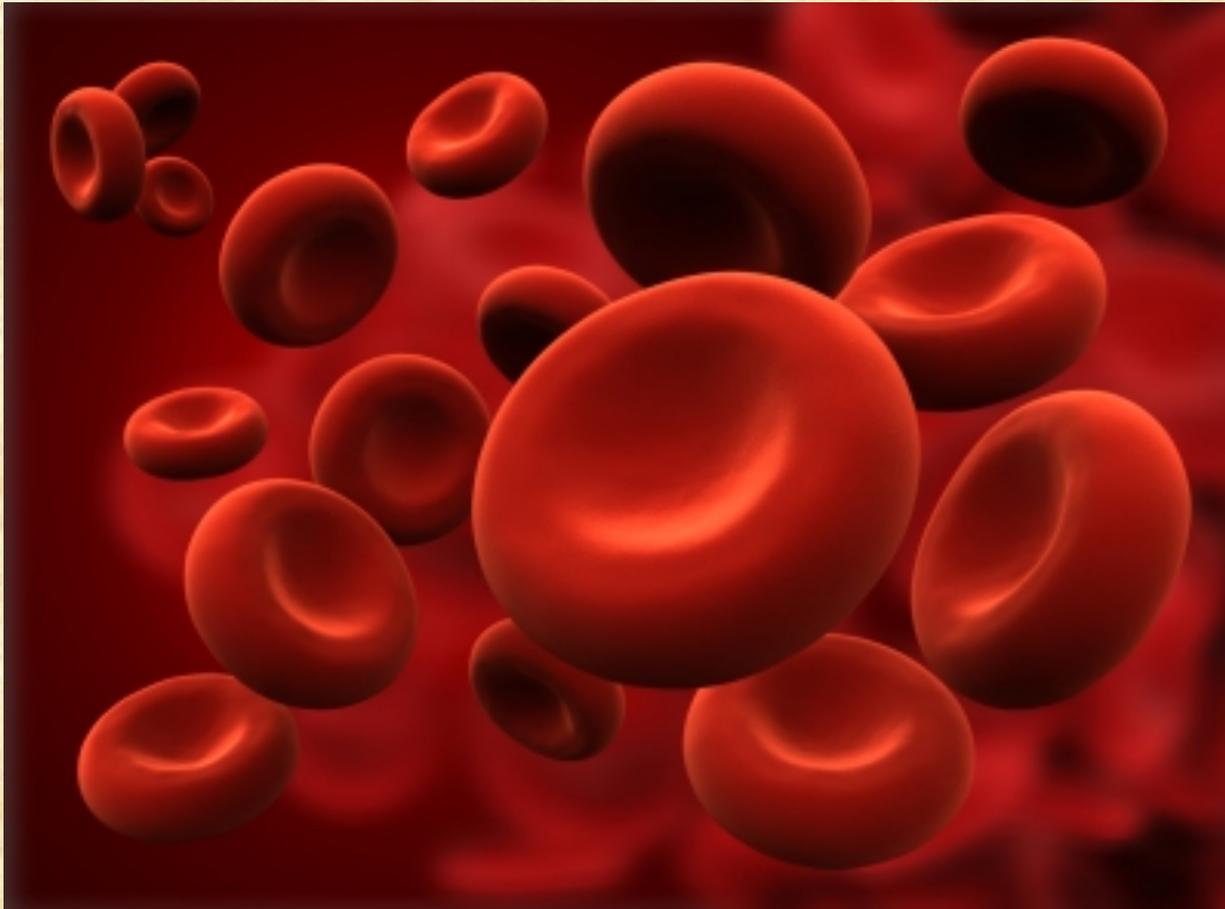
Forward
Typing

Serum

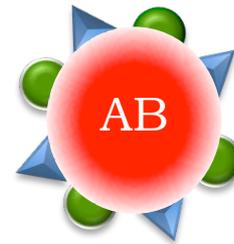
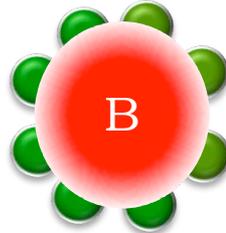
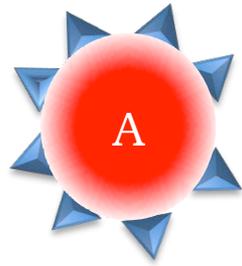


Reverse
Typing

A single red blood cell expresses
~ 2 million ABO molecules
on its surface



ABO Blood Groups

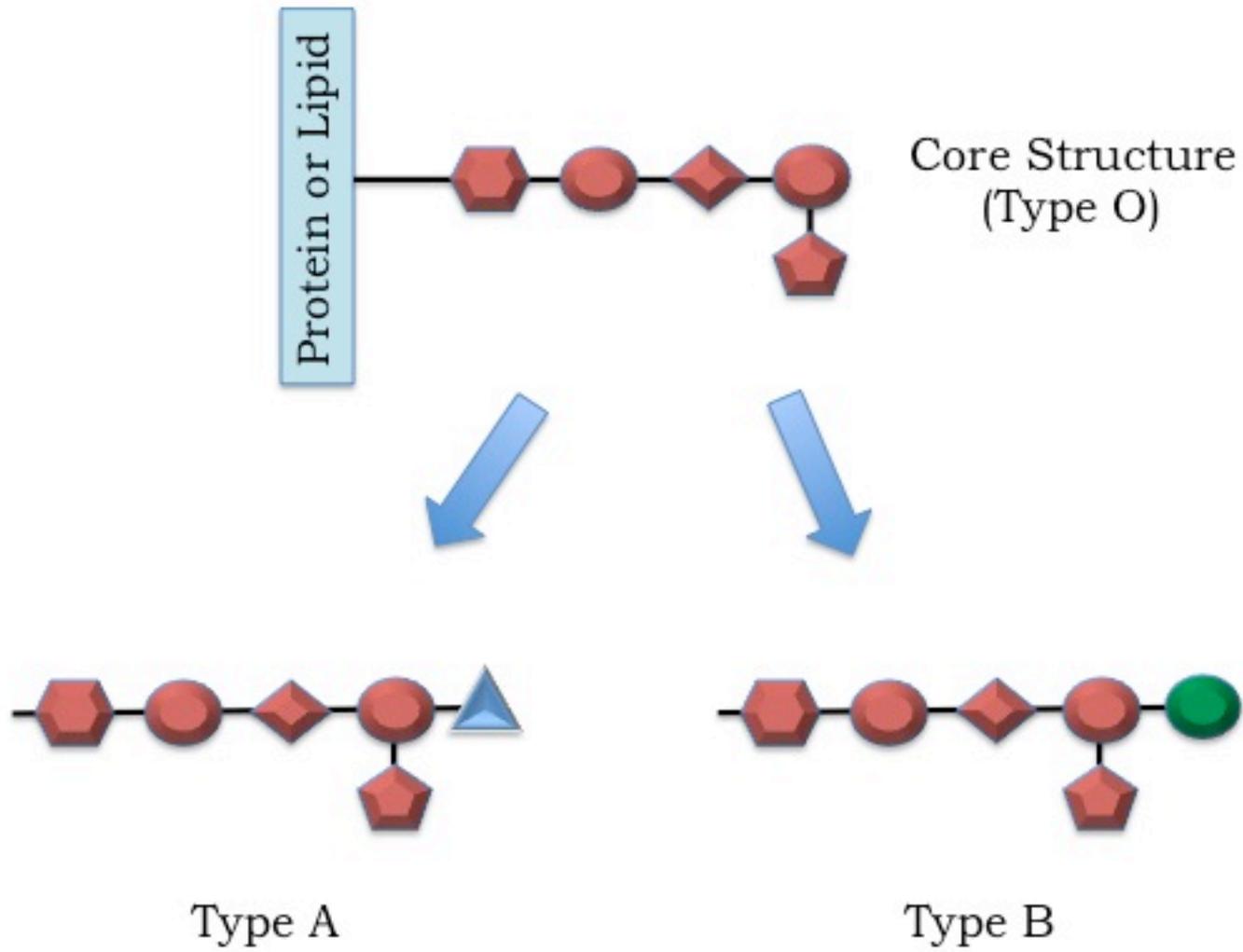


Antigens Present: A

B

A & B

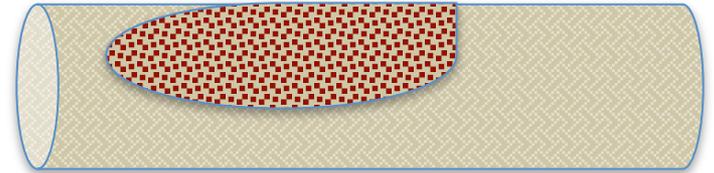
-



Immunohistochemistry



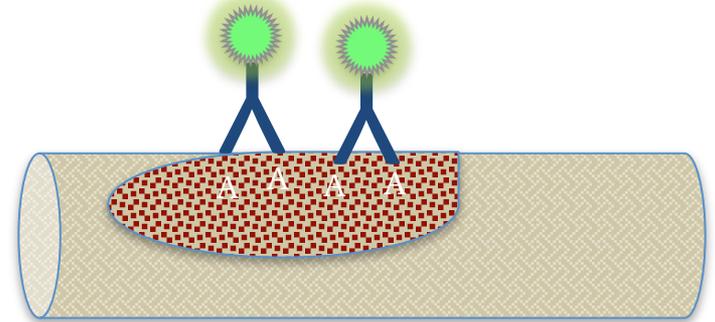
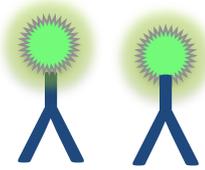
White (control) fiber



Bloodstained fiber

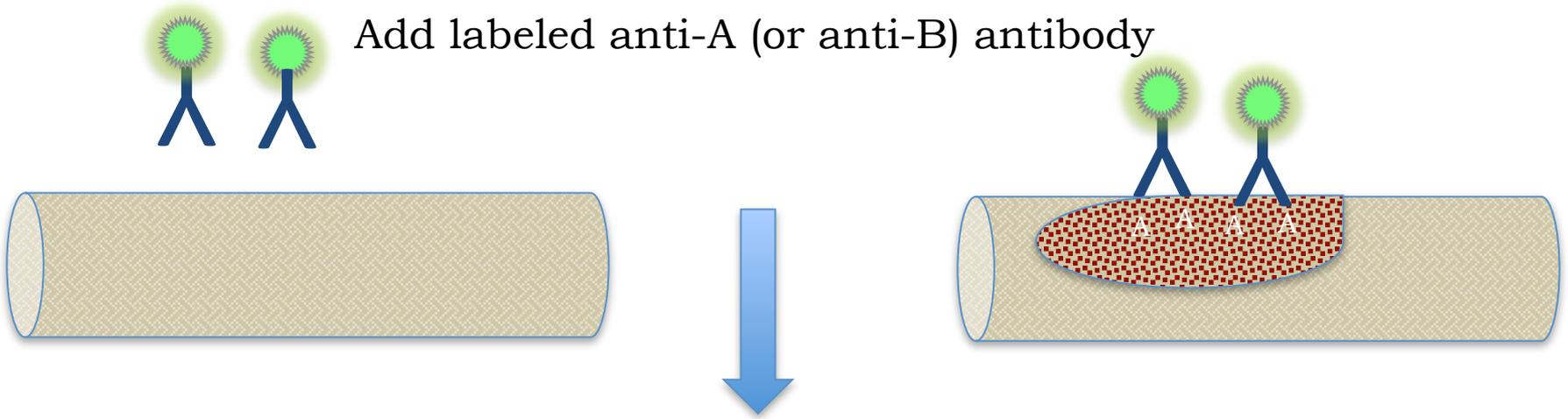
Immunohistochemistry

Add labeled anti-A (or anti-B) antibody



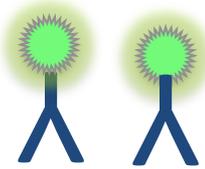
Immunohistochemistry

Add labeled anti-A (or anti-B) antibody

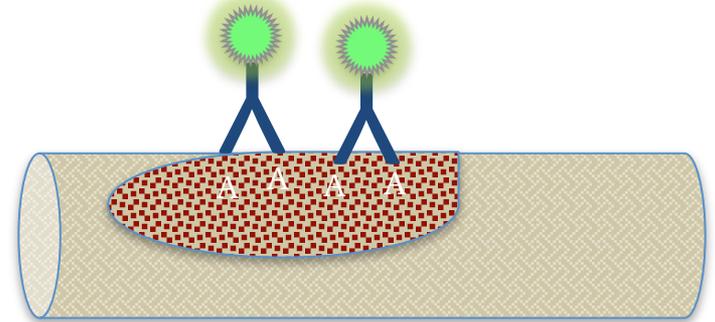


Wash to remove unbound antibody

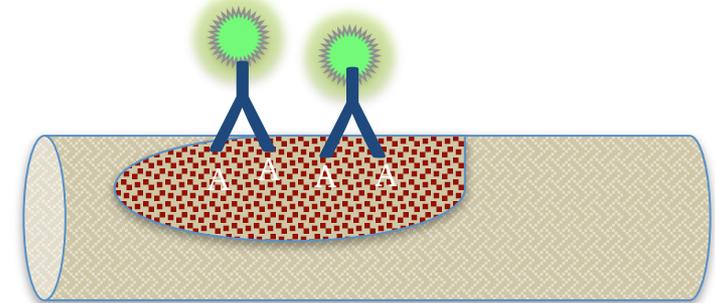
Immunohistochemistry



Add labeled anti-A (or anti-B) antibody



Wash to remove unbound antibody

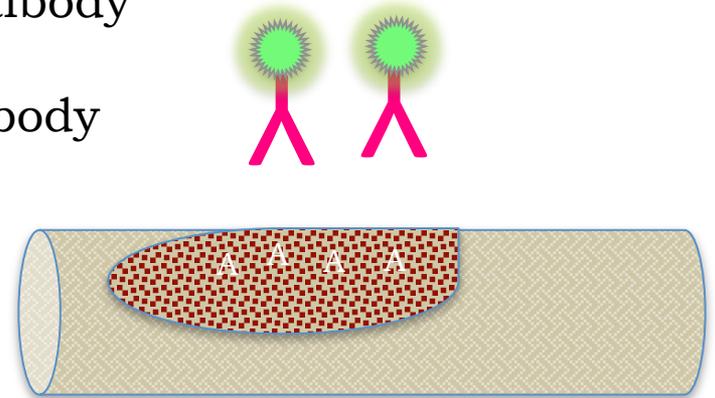
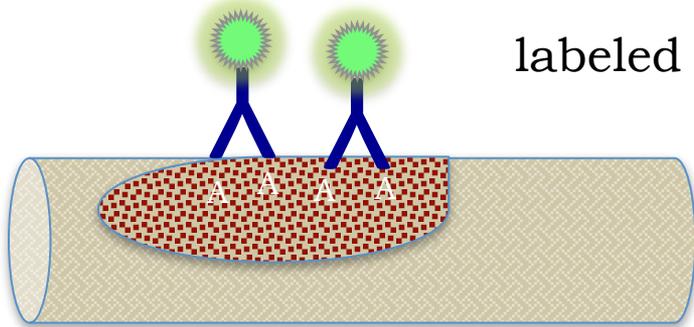


Evaluate Microscopically



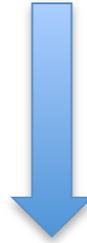
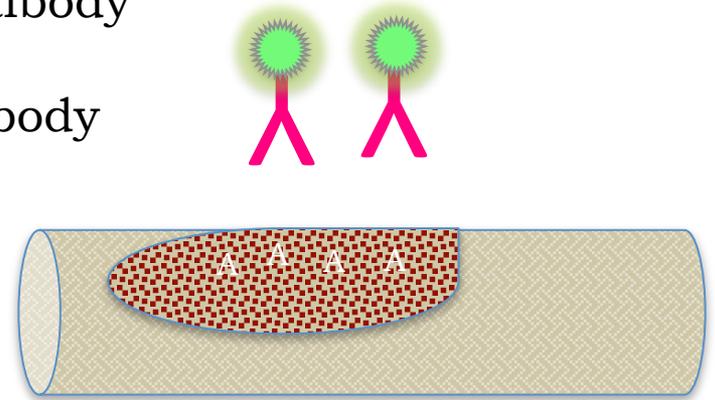
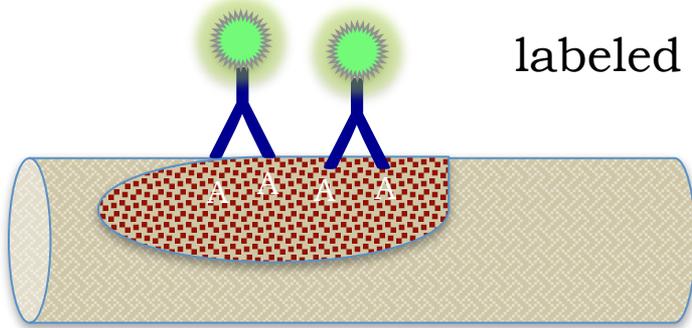
Immunohistochemistry: Specificity Controls

Add labeled specific antibody
or
labeled irrelevant antibody

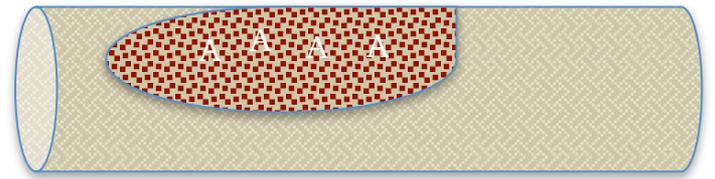
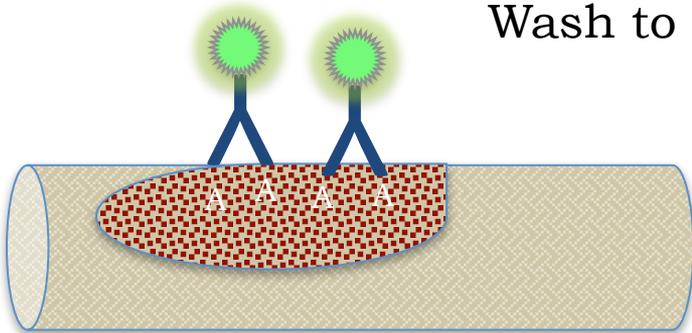


Immunohistochemistry: Specificity Controls

Add labeled specific antibody
or
labeled irrelevant antibody



Wash to remove unbound antibody



Evaluate Microscopically



Bloodstained fibers:

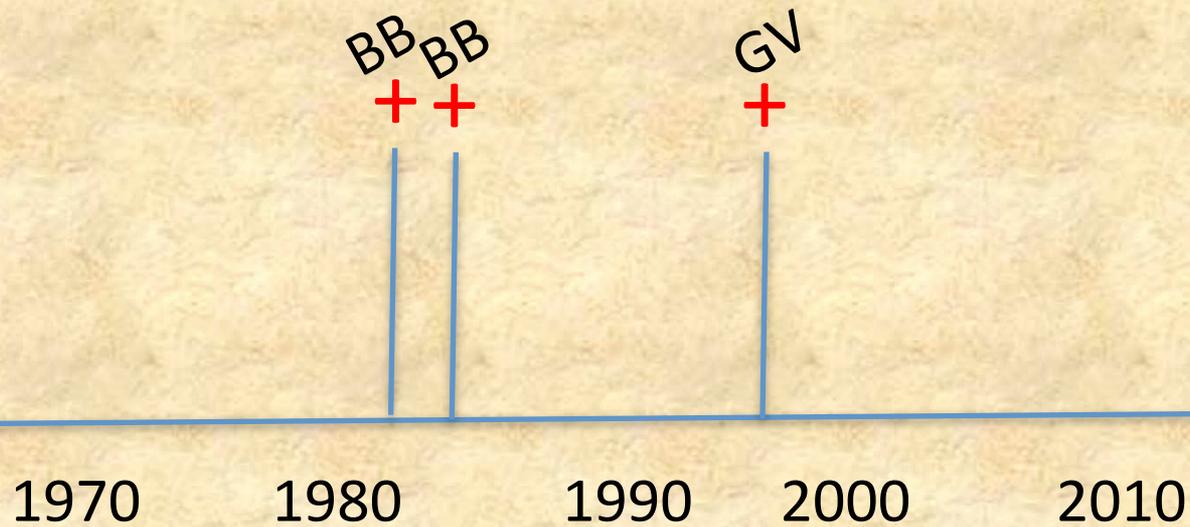
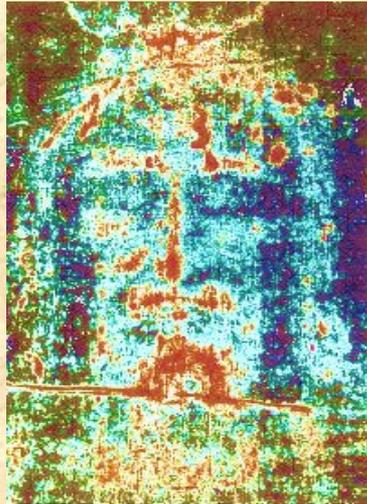
Positive for both A and B (equal intensities)

Negative reactivity with anti-O antibody

Control fibers: Negative for all



Assignment of AB blood type

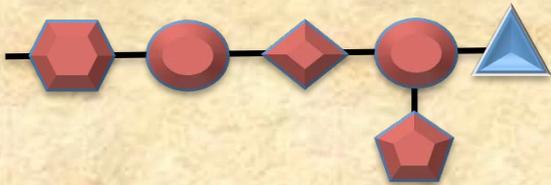
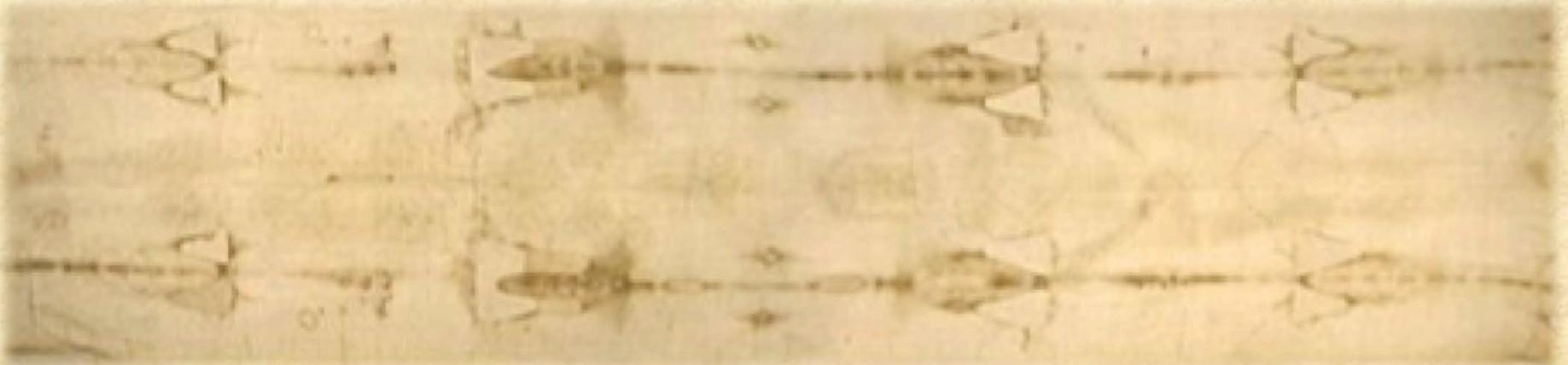
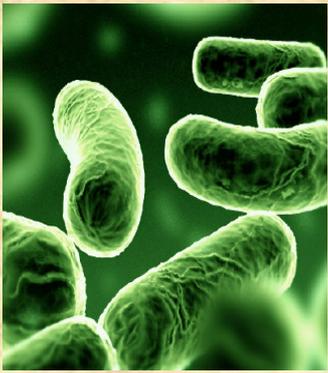


Forward Typing Methods

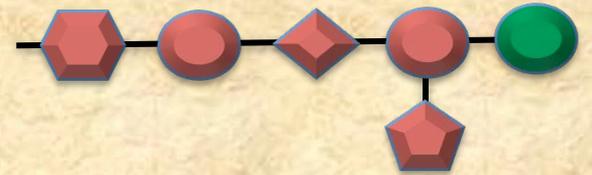


*“A lot of old material
types as AB”*





Type A



Type B



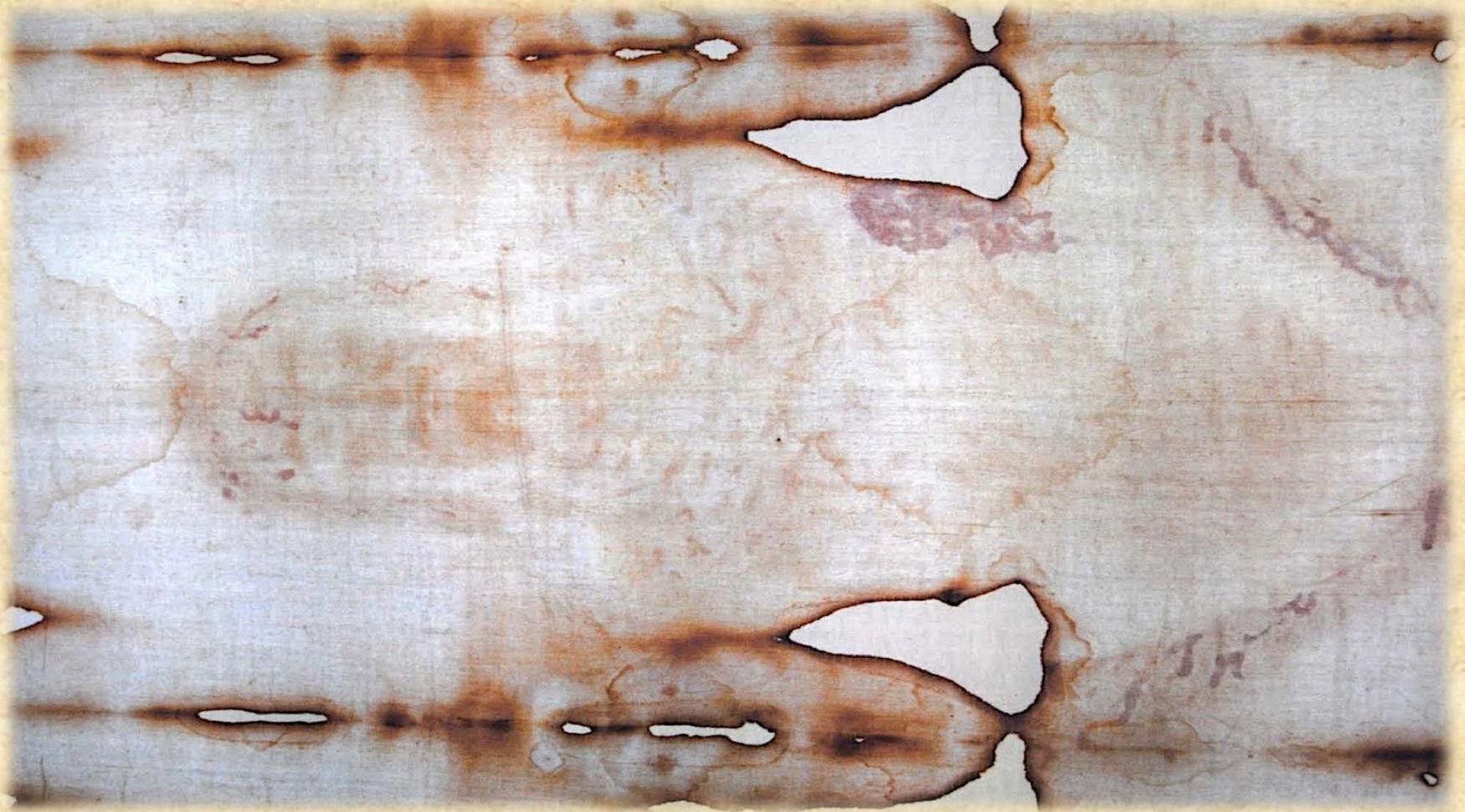
R. K. Connolly *Nature* 224: 325 (1969)
Hawass, et al. *JAMA* 303:638 (2010)

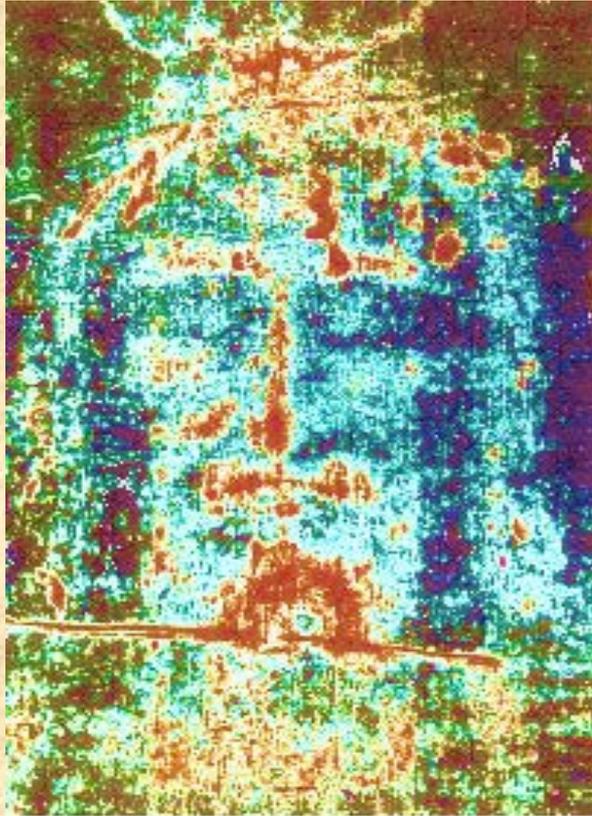
King Tutankhamen

Blood type: A



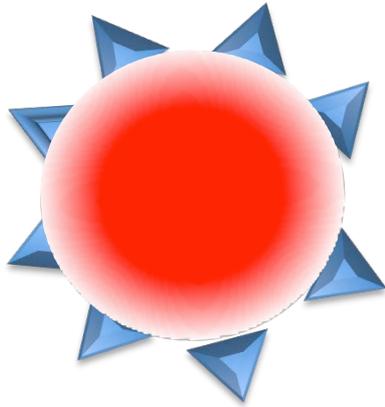
Fibers at the bed of the bloodstain were negative for reactivity with anti-A or anti-B antibodies





Do we really know that
the blood is AB?

RBC



Forward
Typing

Serum



Reverse
Typing

Gideon: Fleece Experiment

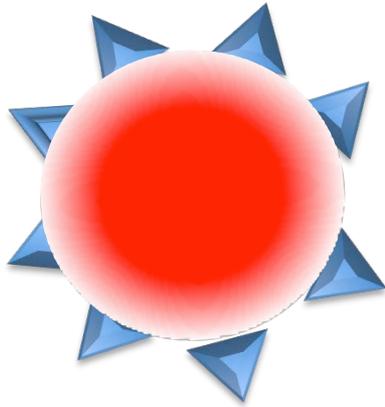
1162 BC

Fleece: Wet Ground: Dry

Fleece: Dry Ground: Wet



RBC



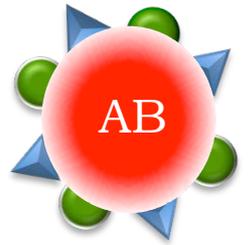
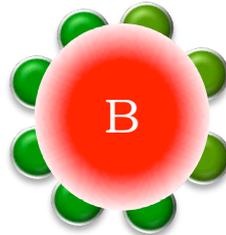
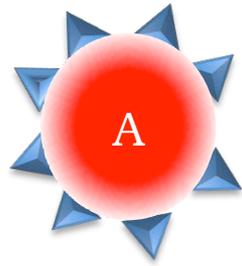
Forward
Typing

Serum



Reverse
Typing

ABO Blood Groups



RBC Surface:

A

B

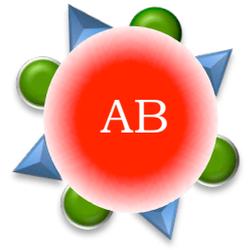
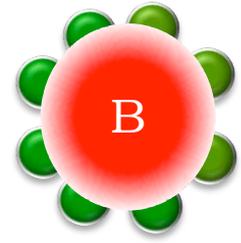
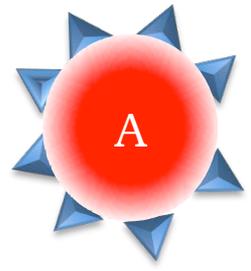
-

A & B

Serum:

**anti-B
Antibodies**

ABO Blood Groups



RBC Surface:

A

B

-

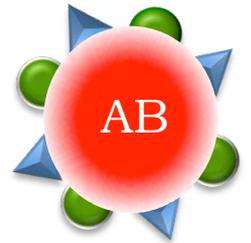
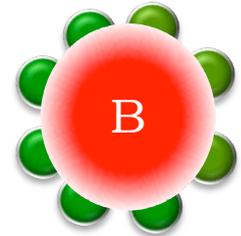
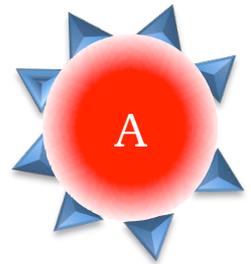
A & B

Serum:

**anti-B
Antibodies**

**anti-A
Antibodies**

ABO Blood Groups



RBC Surface:

A

B

-

A & B

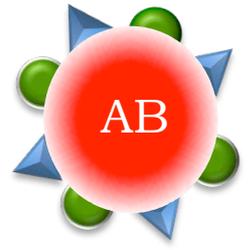
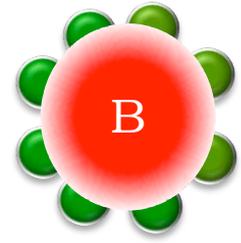
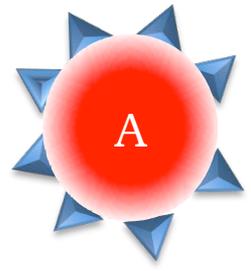
Serum:

**anti-B
Antibodies**

**anti-A
Antibodies**

**anti-A & B
Antibodies**

ABO Blood Groups



RBC Surface:

A

B

-

A & B

Serum:

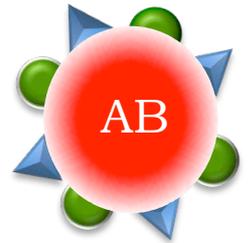
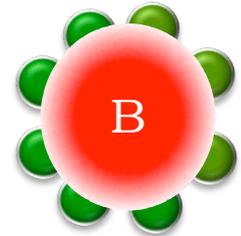
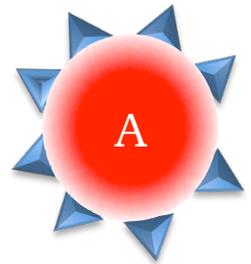
**anti-B
Antibodies**

**anti-A
Antibodies**

**anti-A & B
Antibodies**

-

ABO Blood Groups



RBC Surface:

A

B

-

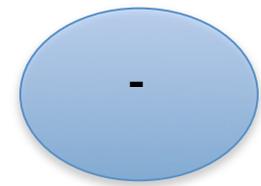
A & B

Serum:

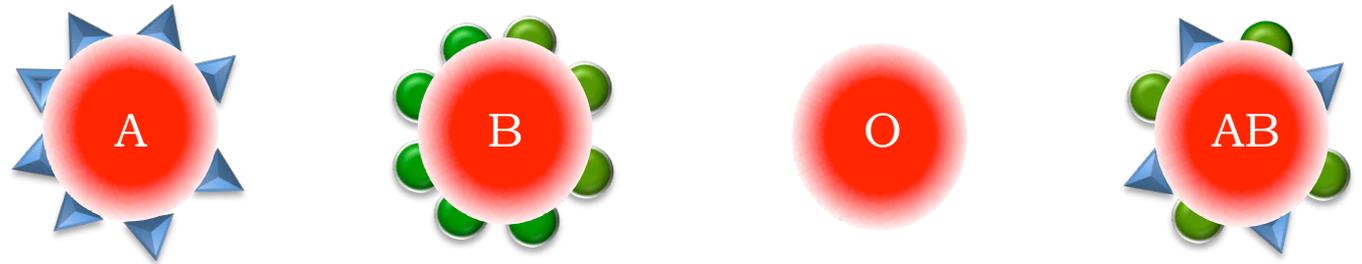
**anti-B
Antibodies**

**anti-A
Antibodies**

**anti-A & B
Antibodies**



ABO Blood Groups



RBC Surface:

A

B

-

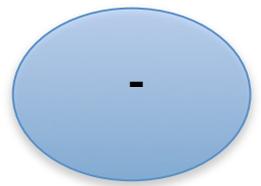
A & B

Serum:

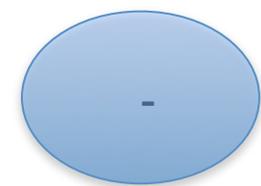
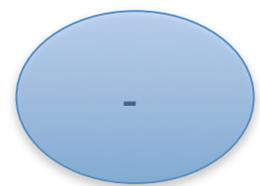
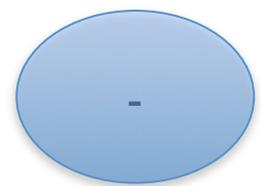
**anti-B
Antibodies**

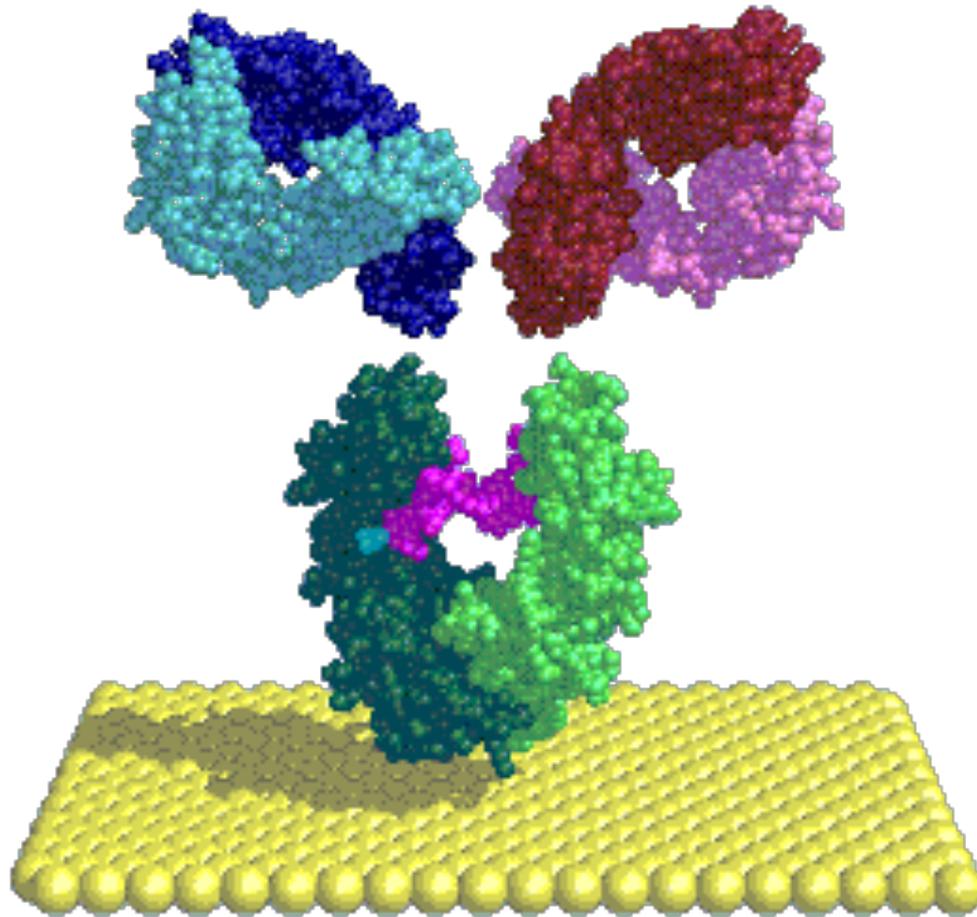
**anti-A
Antibodies**

**anti-A & B
Antibodies**



Aged Samples: Degradation/Loss of Function



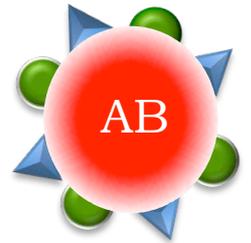
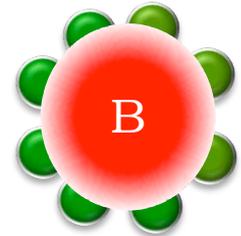
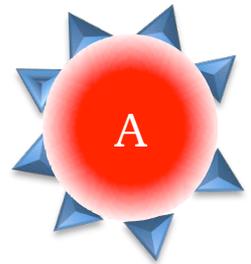


Reverse typing:
Serum Antibodies must be functional
Maintain proper 3-D conformation

Over time, such antibodies might still exist
but not be functional



ABO Blood Groups



RBC Surface:

A

B

-

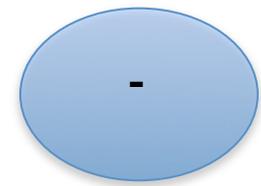
A & B

Serum:

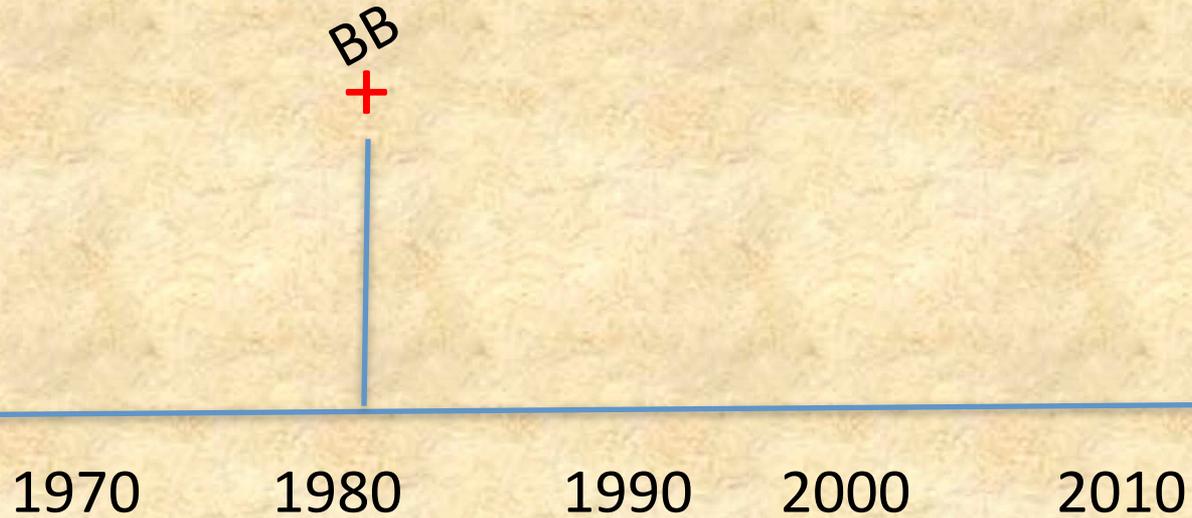
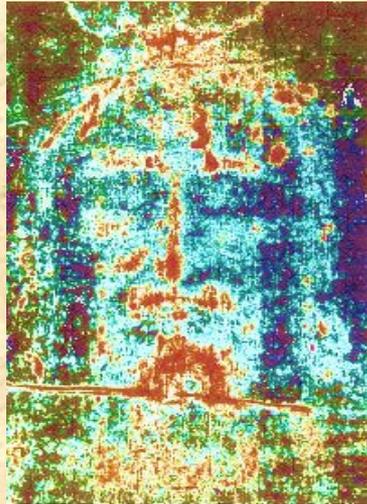
**anti-B
Antibodies**

**anti-A
Antibodies**

**anti-A & B
Antibodies**

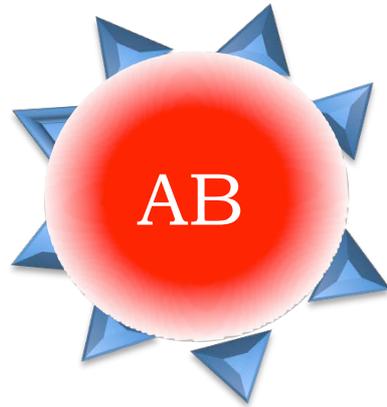


Assignment of AB blood type



Reverse Typing Methods
Results Inconclusive at Best

RBC



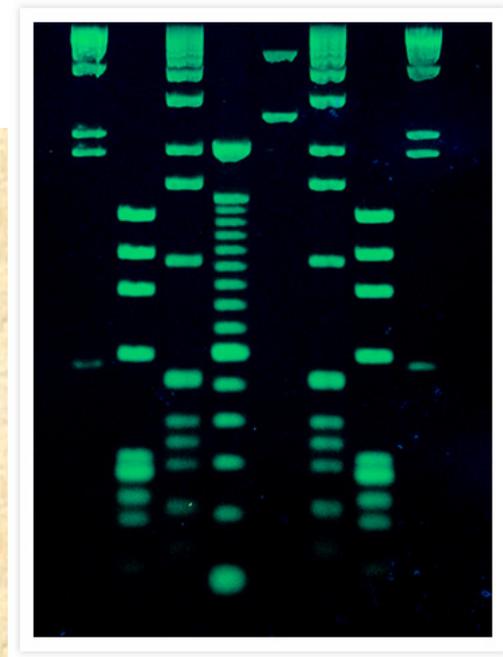
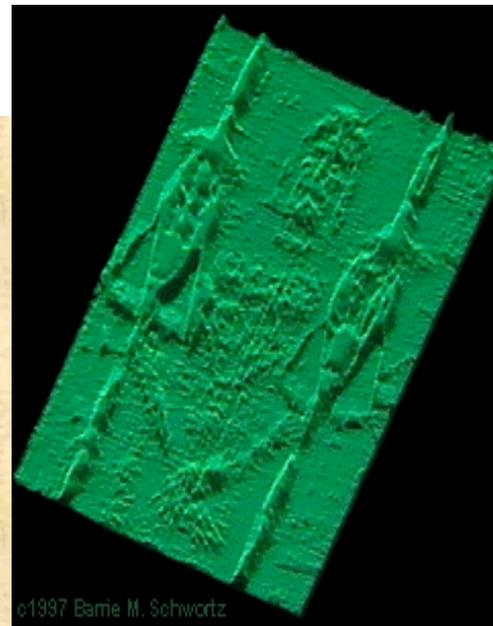
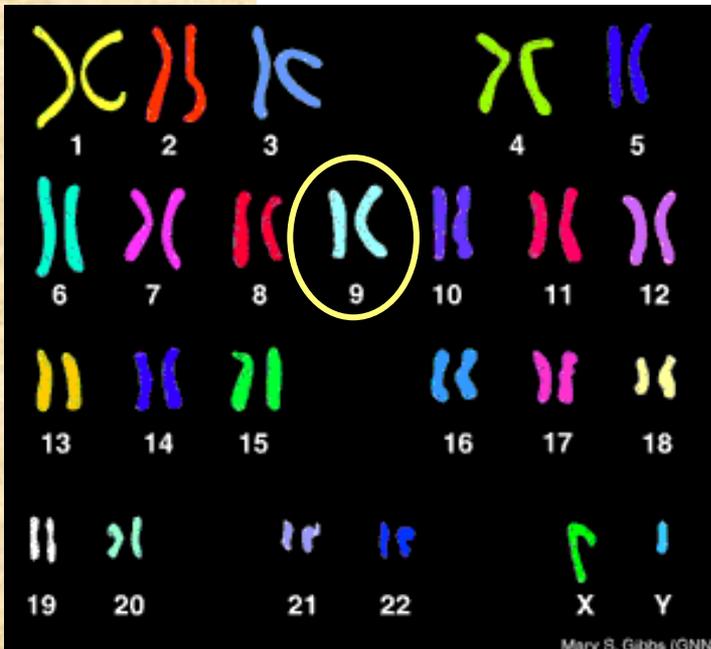
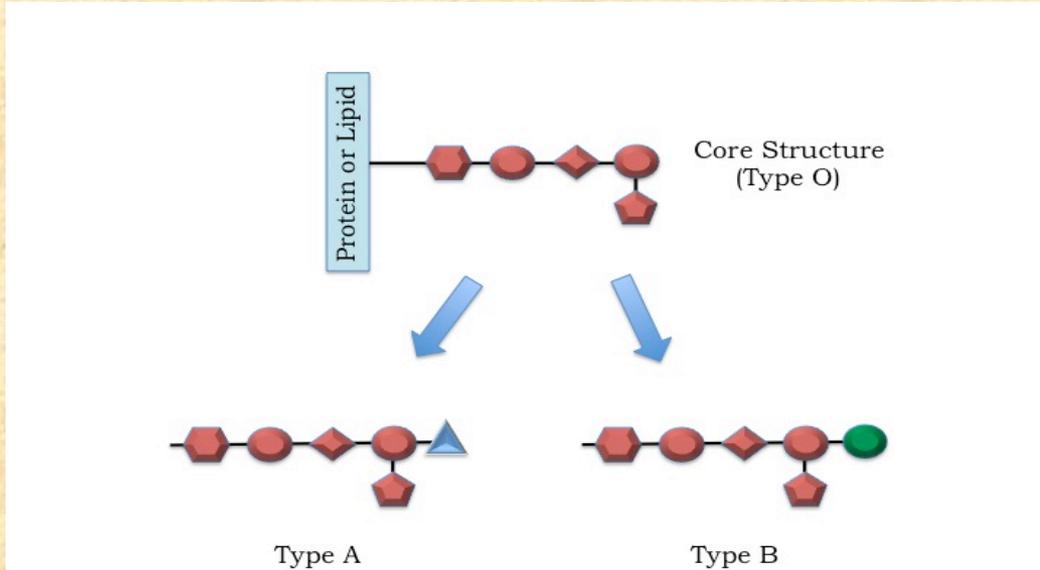
Forward
Typing

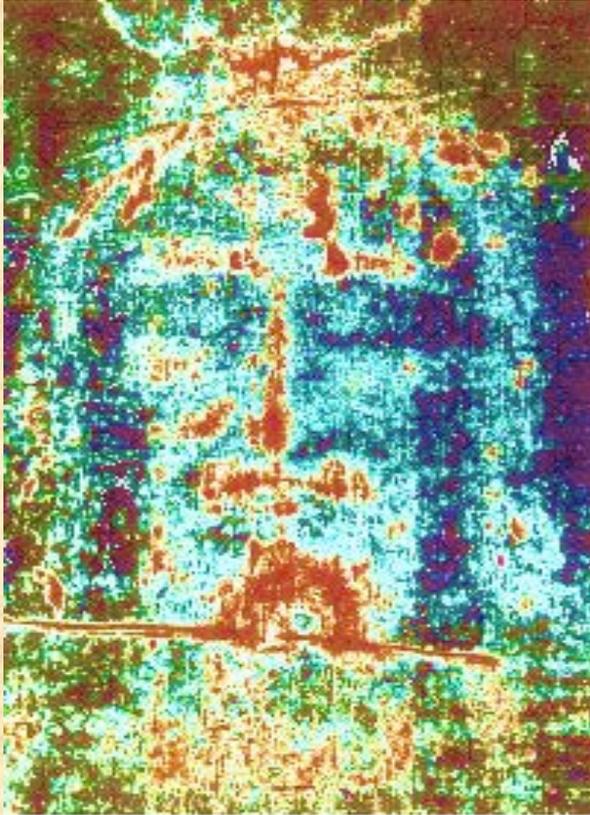
Serum



Reverse
Typing

Molecular (DNA) Analysis



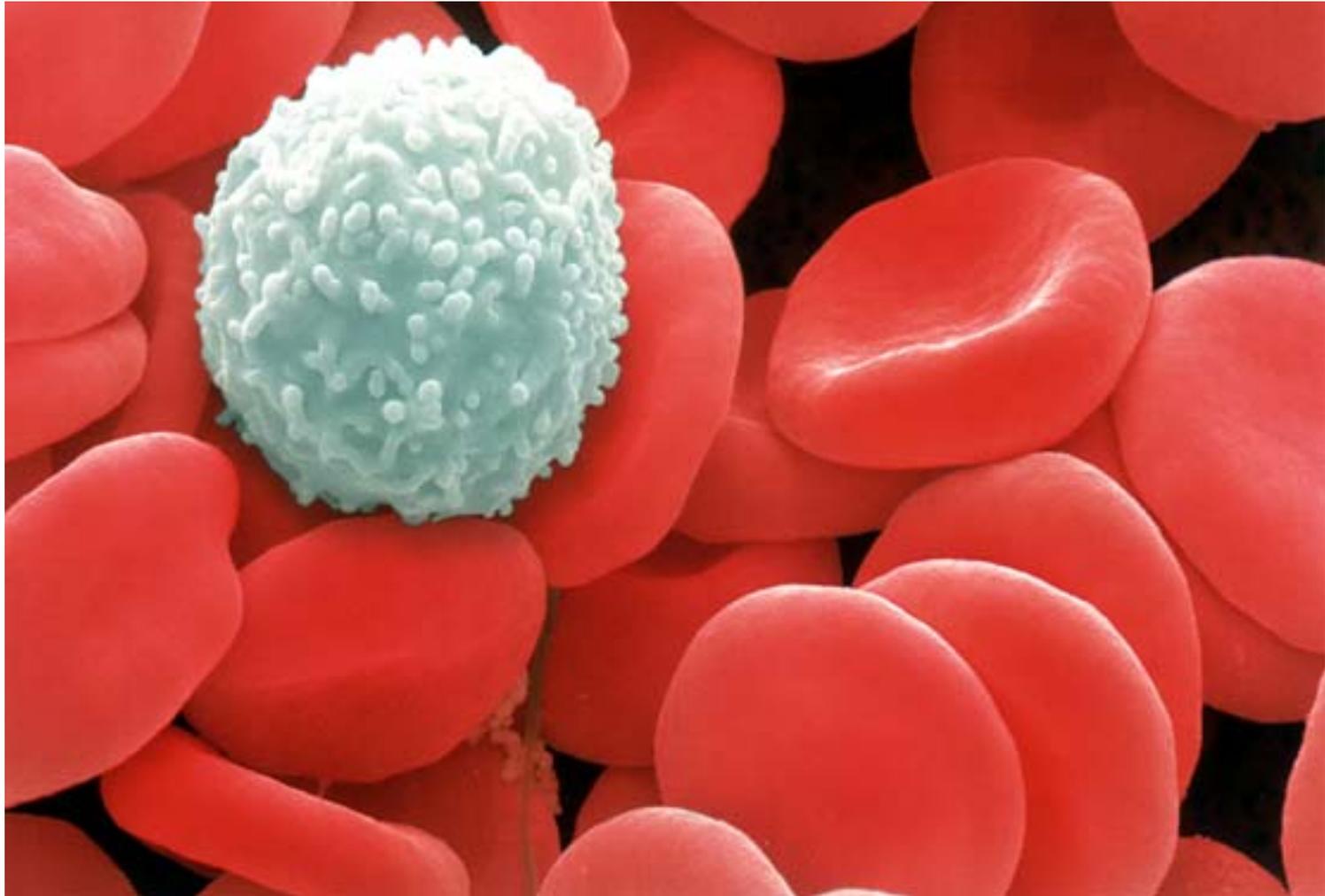


Is the blood really AB?

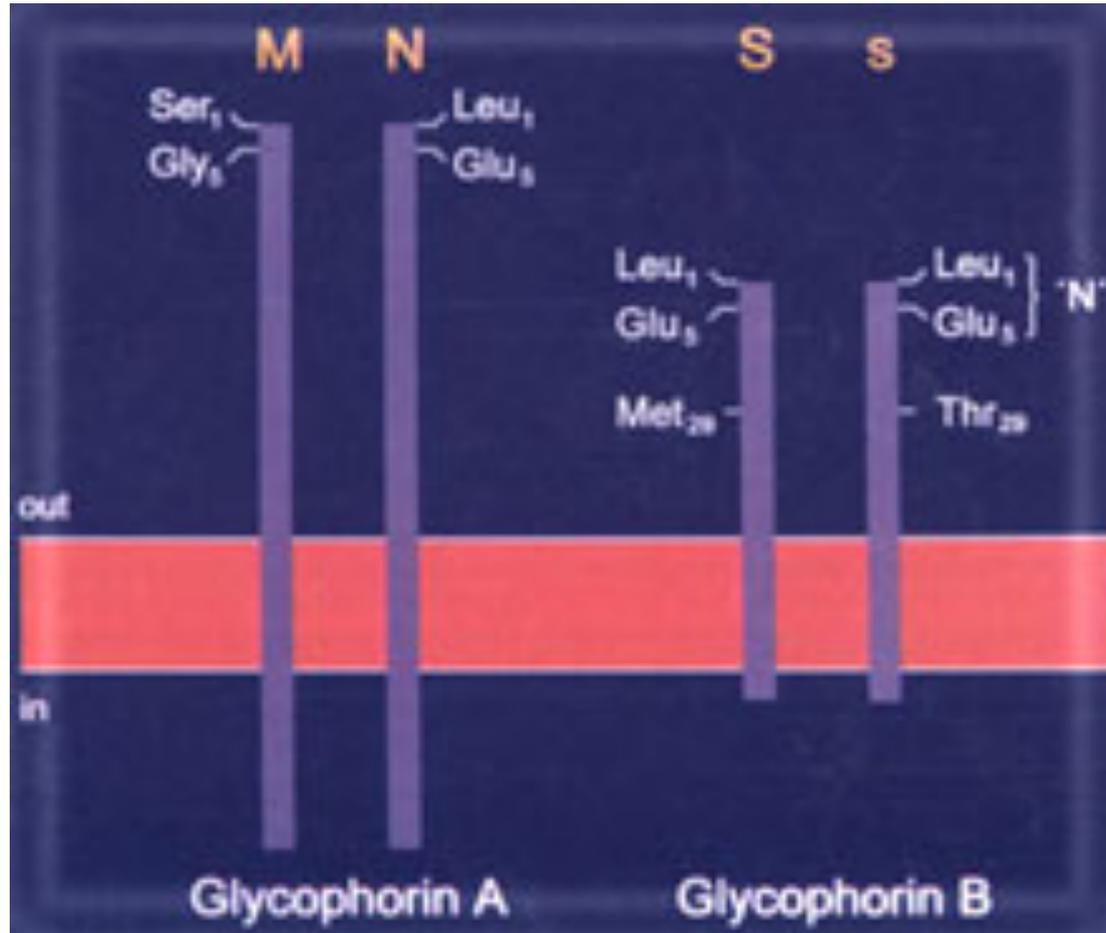
Forward typing results
suggest yes

A 2nd confirmatory
test is important
(essential)

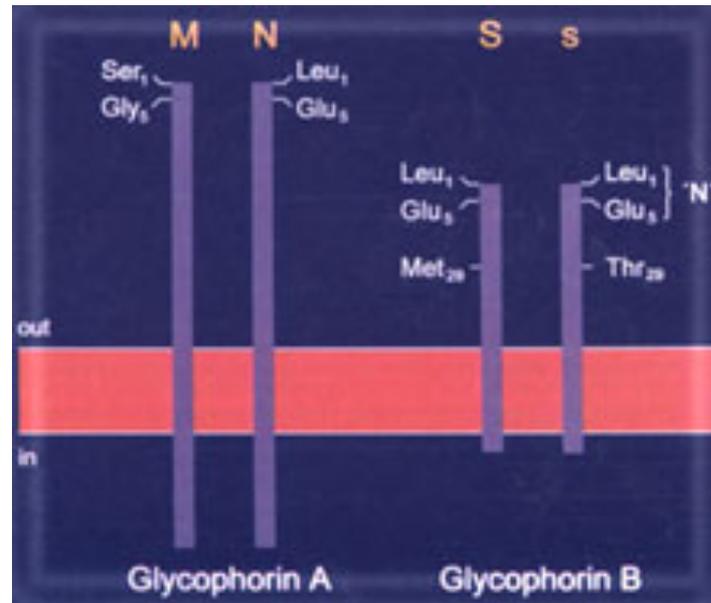
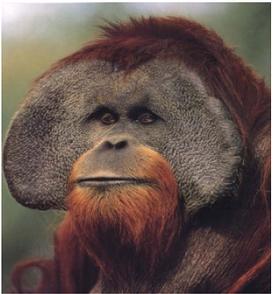
Other Red Blood Cell Surface Markers



MNS System



MNS System



MN antigens

RBCs of anthropoid apes, humans

S antigens

Human RBCs only

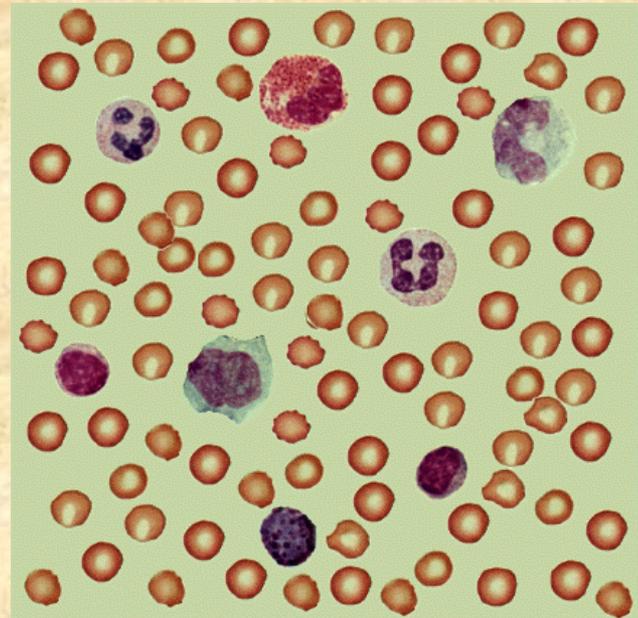
Unstained fibers

MNS negative

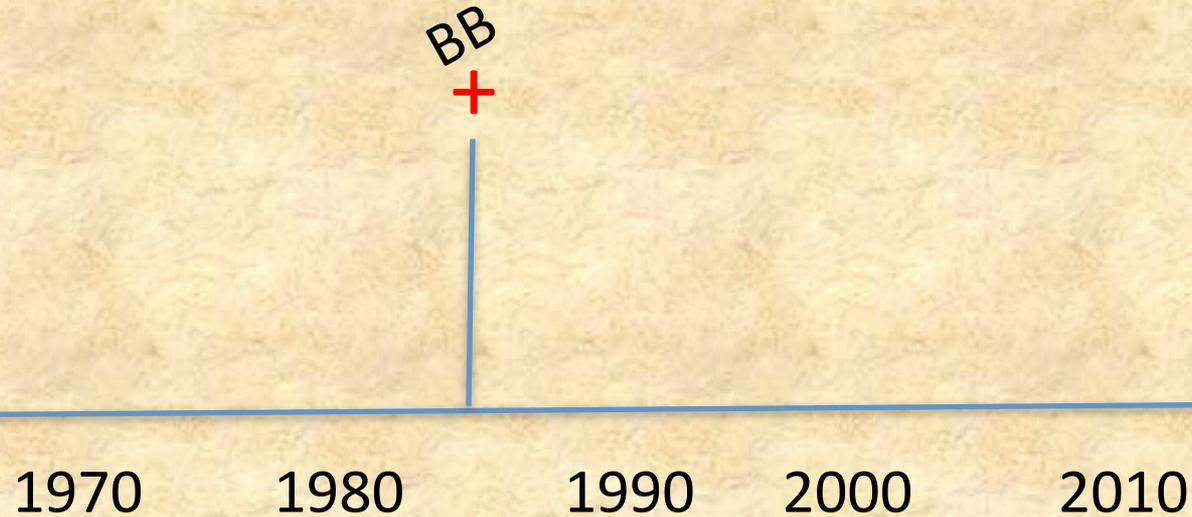
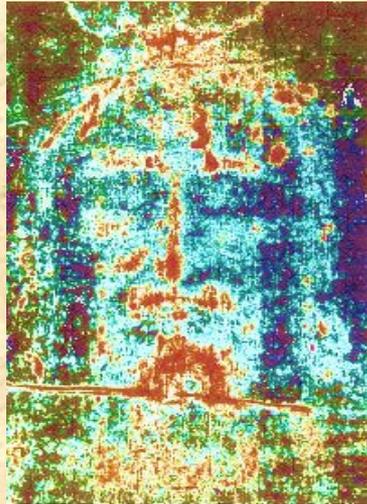


Bloodstained fibers

MNS positive

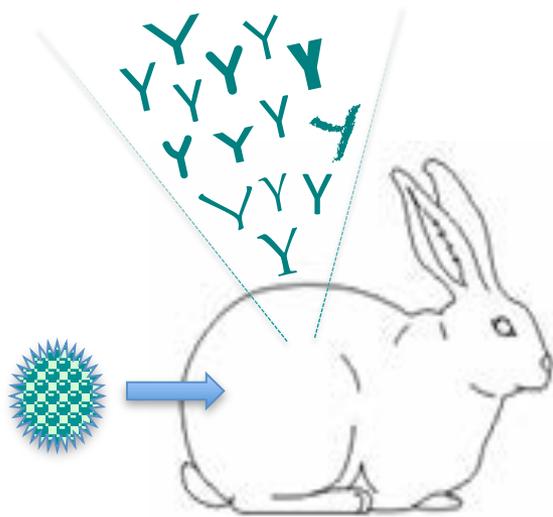


Shroud fibers are MNS positive



Immunochemical Methods
(Brief report: “fairly good binding”)

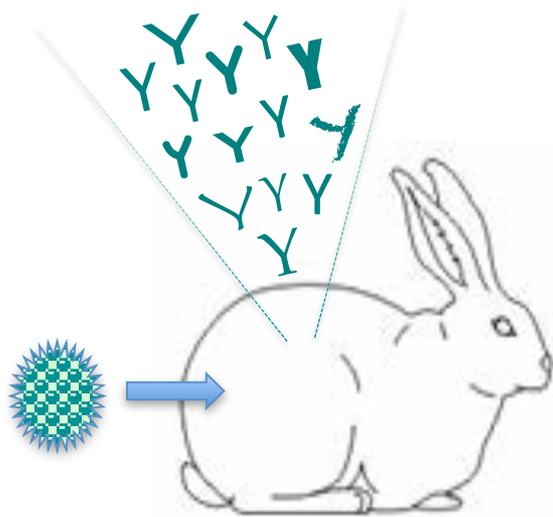
Blood serum harvested



Polyclonal Antibodies
(Mixed type)

Before the '80s

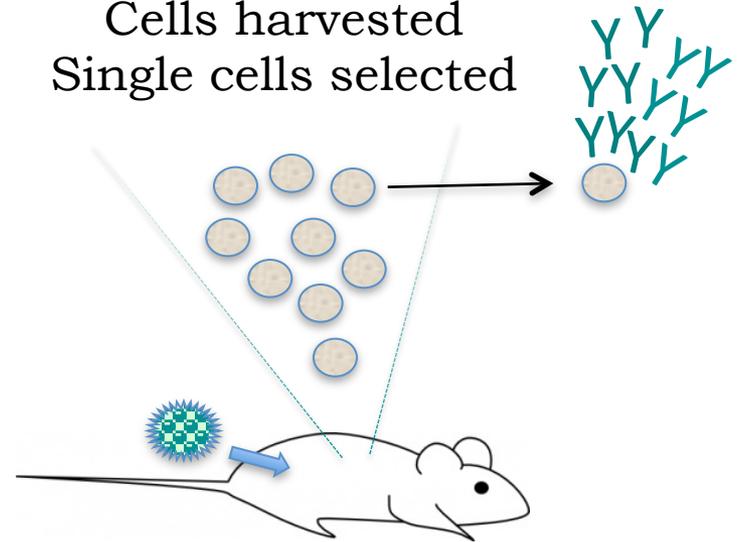
Blood serum harvested



Polyclonal Antibodies
(Mixed type)

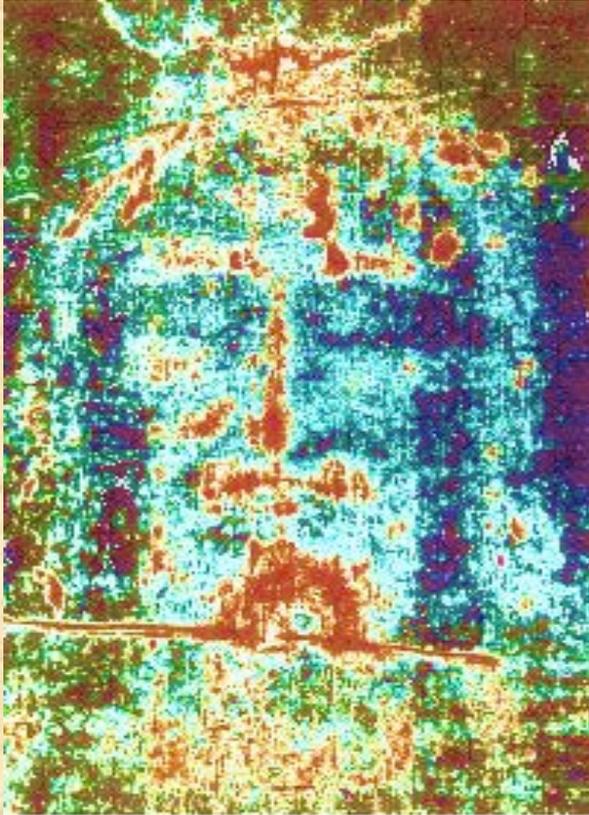
Before the '80s

Cells harvested
Single cells selected

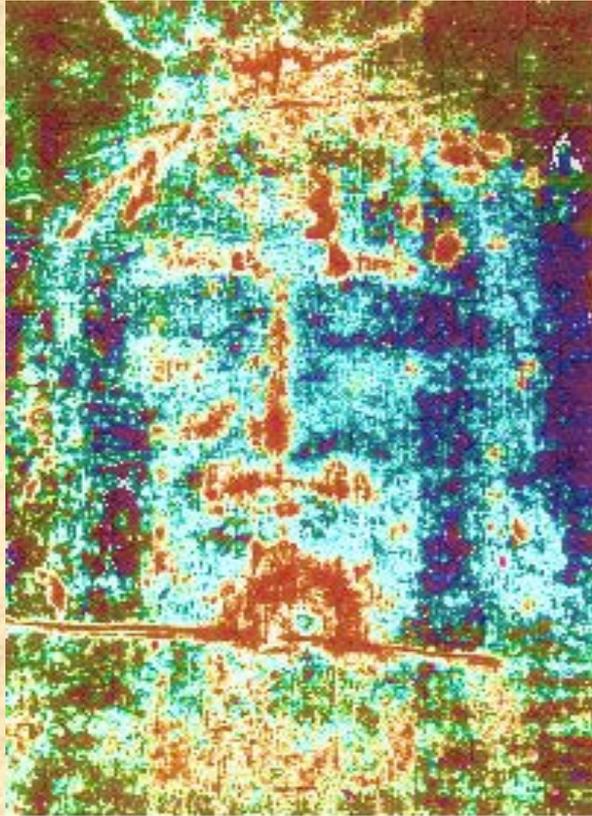


Monoclonal Antibodies
(Identical)

Mid-Post '80s



**In 2010, highly specific
monoclonal antibodies
that effectively
distinguish human blood
were developed**

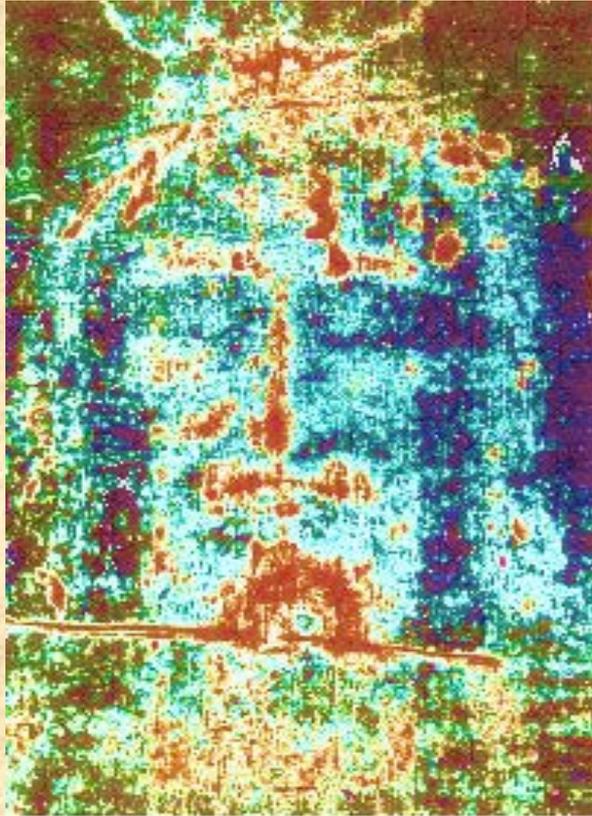


What type of blood is it?

Summary

At least Primate
(MNS suggests possibly human)
Other types?

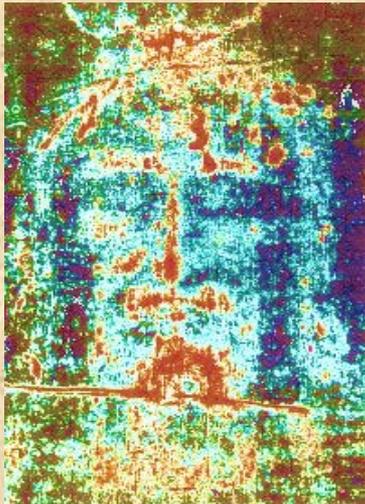
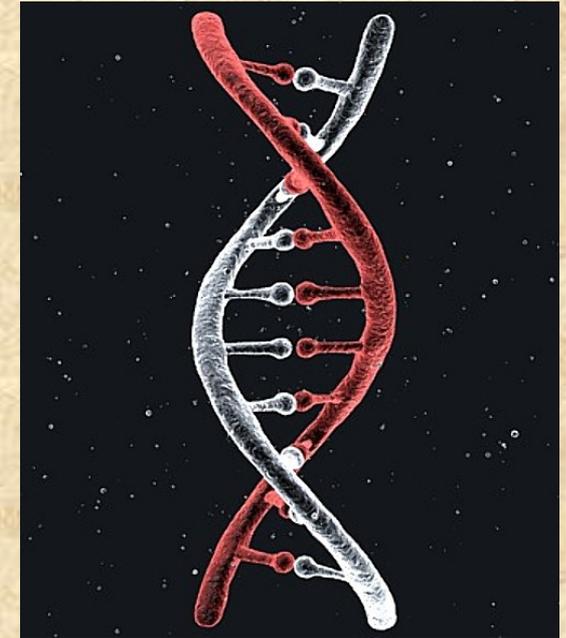
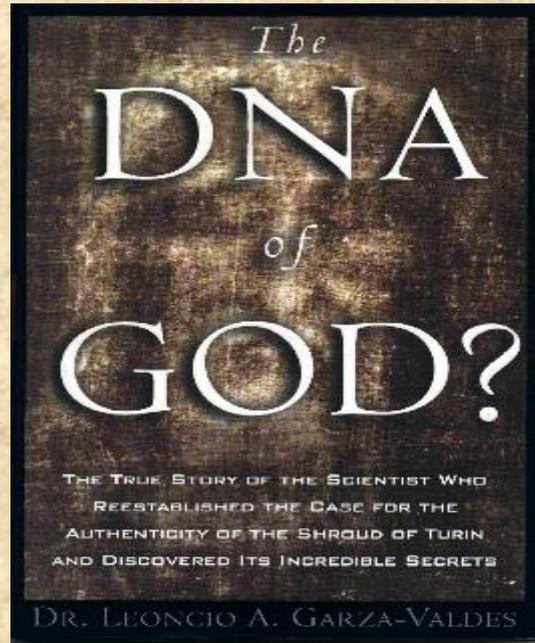
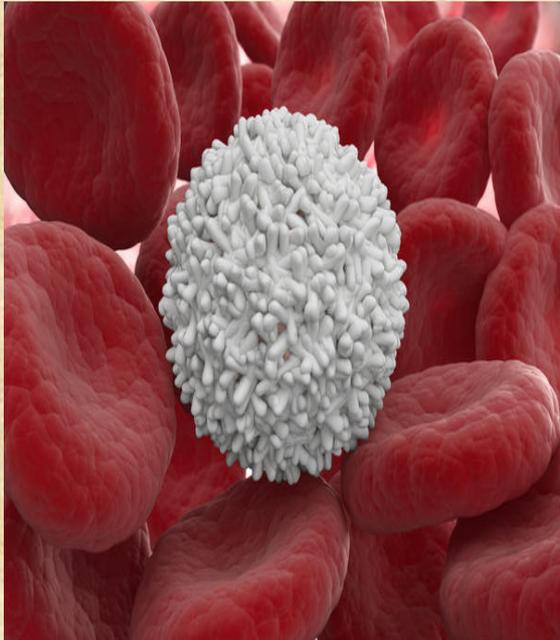
AB by forward typing
(confirmatory test important)



Is the blood from a male?

Is the blood from a single individual?

Human DNA has been isolated from bloodstained fibers of the Shroud



CGV

++

1970

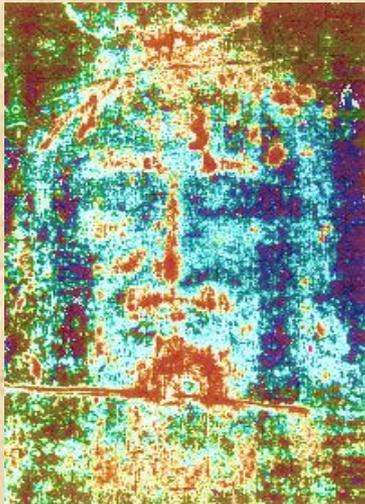
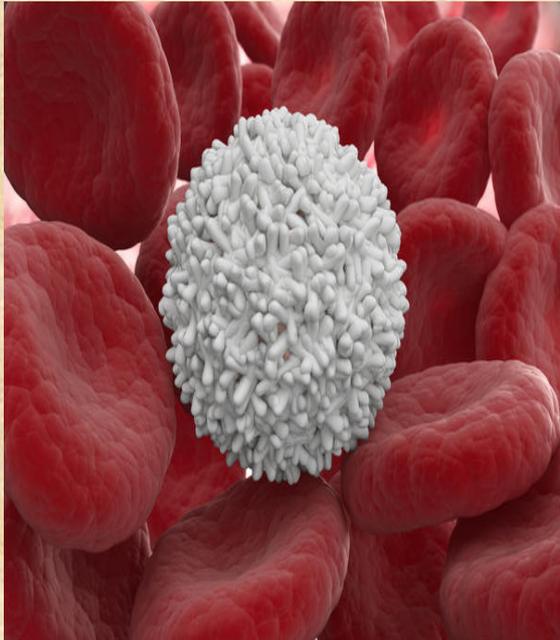
1980

1990

2000

2010

Plant, human DNA has been isolated from dust particles on the Shroud



1970

1980

1990

2000

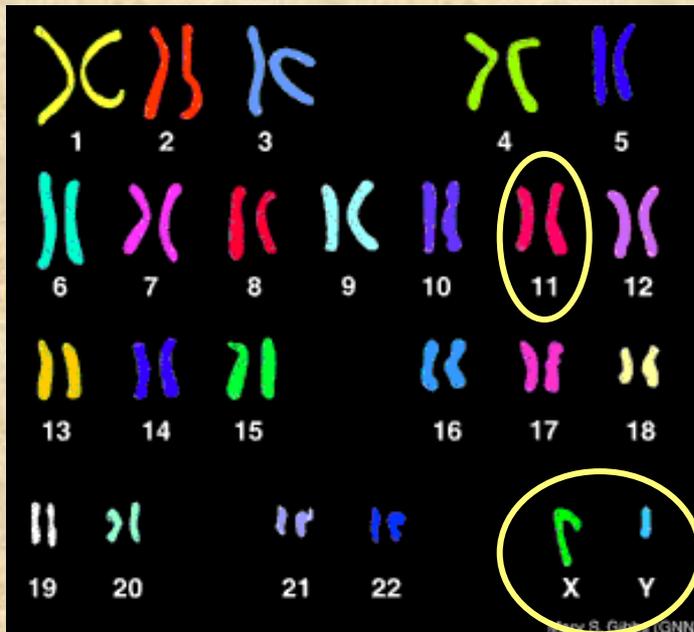
2010

2014

B

+

Human DNA and the Shroud



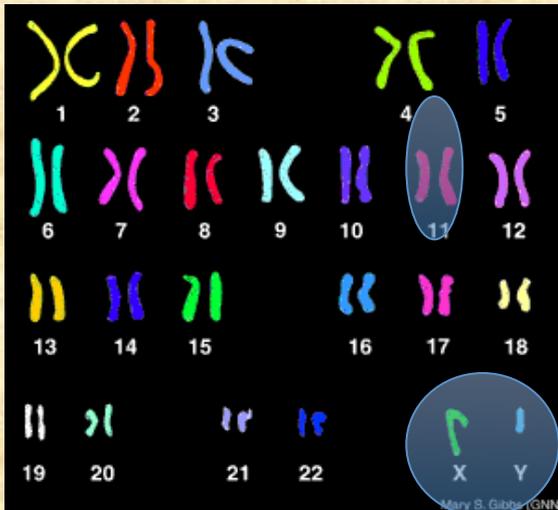
Betaglobin gene
Amelogenin X gene
Amelogenin Y gene



Endogenous vs. Exogenous DNA?



**It is unknown if
the DNA truly originates
from blood cells**



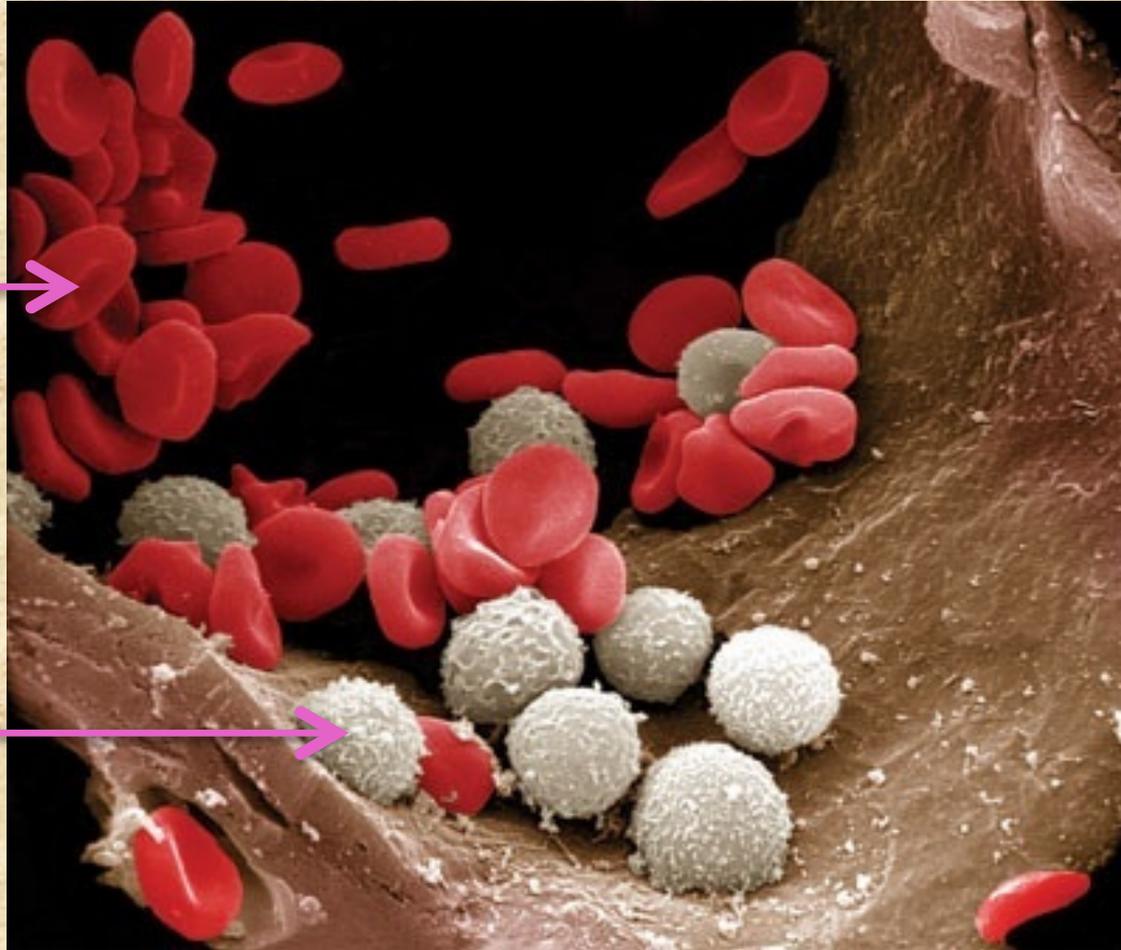
Human Blood Cells

DNA

No



Yes



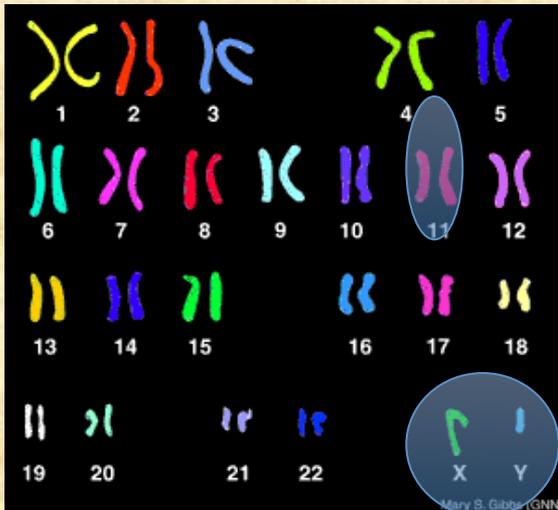
**White blood cells of the immune system
are unique among all cells of the body**



**They exhibit DNA rearrangement
of specific receptor genes**

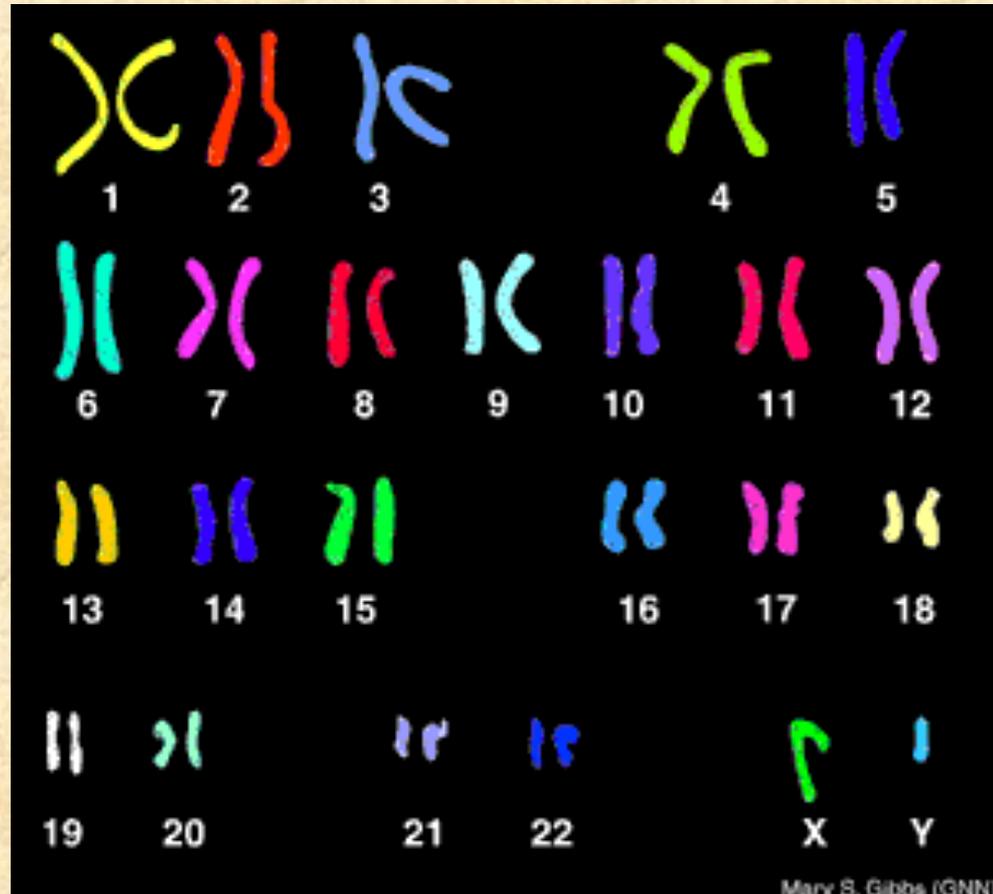


Is it possible to analyze additional DNA segments?



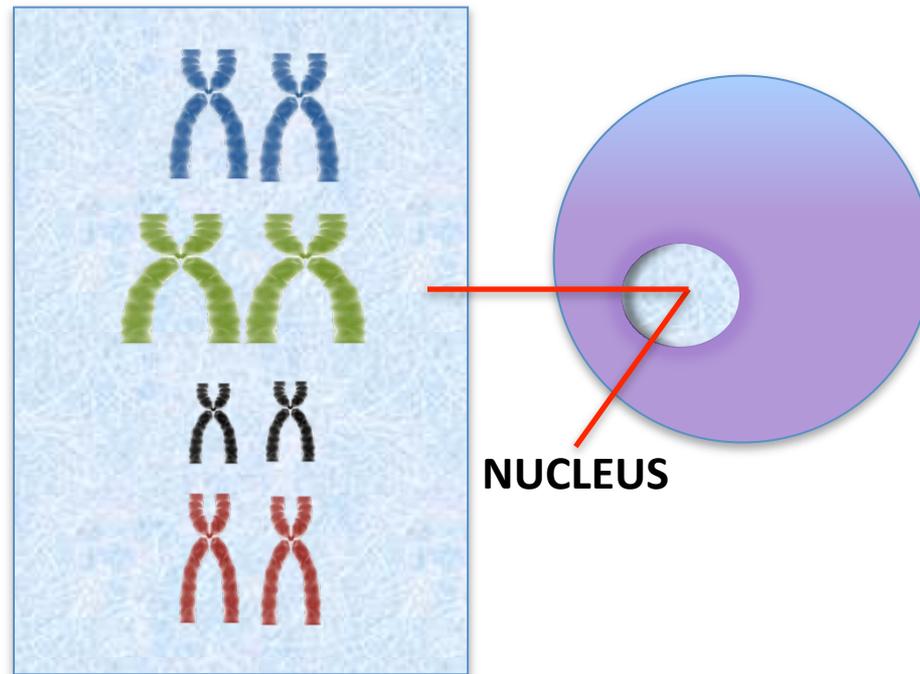


“Portions of the X & Y chromosomes have been found, but the rest of the DNA is really too degraded to be useful...”



Mary S. Gibbs (GNN)

The human genome consists of
20,000-30,000 genes



NUCLEUS

NUCLEAR DNA

The Polymerase Chain Reaction

Targeting & Amplification of Specific Genes



Sample
(DNA)

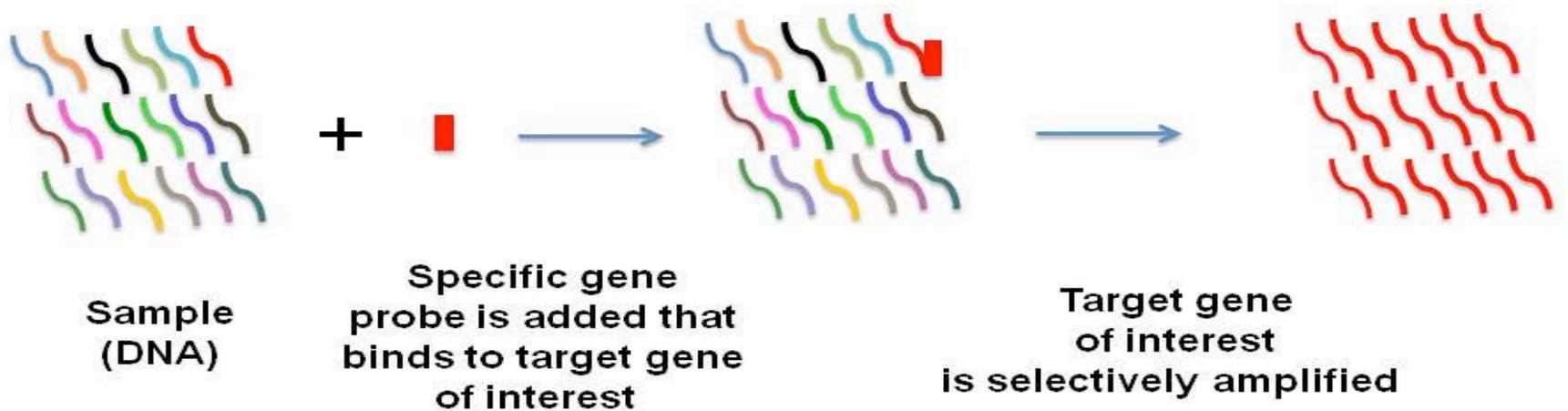
+



Specific gene
probe is added that
binds to target gene
of interest

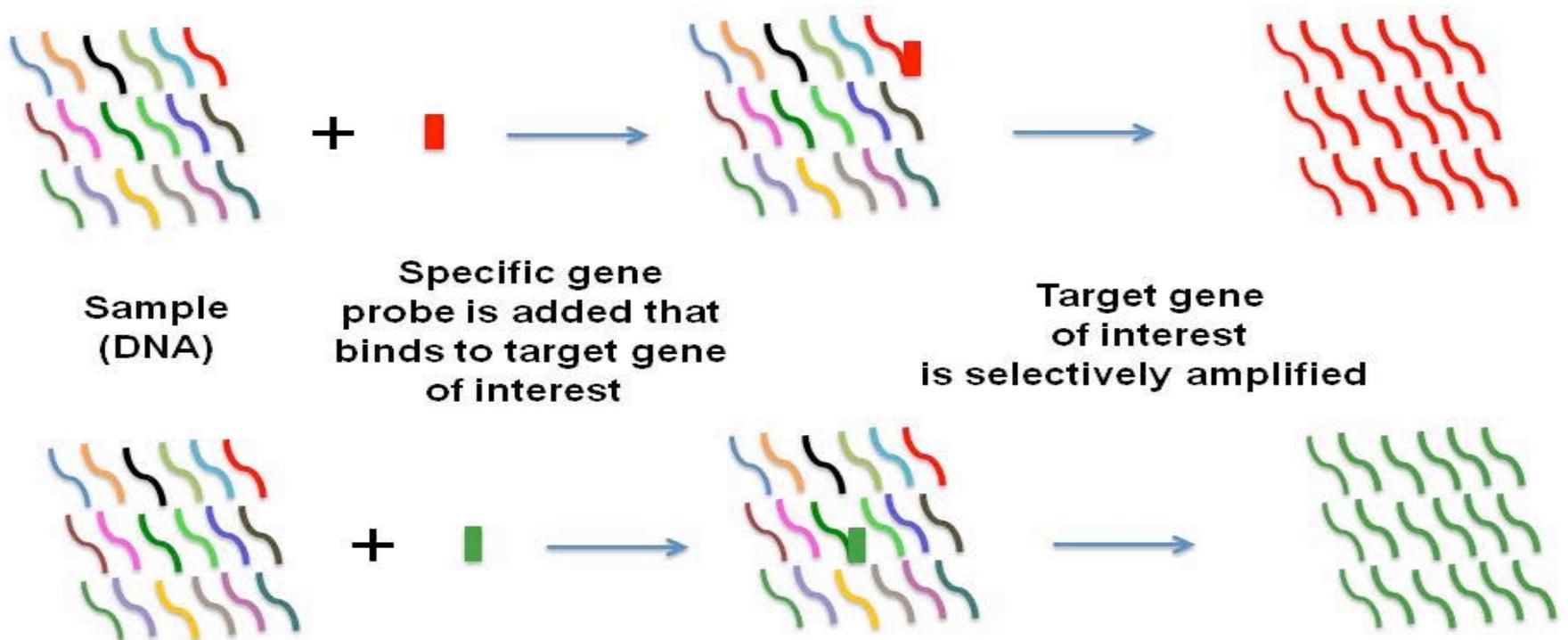
The Polymerase Chain Reaction

Targeting & Amplification of Specific Genes



The Polymerase Chain Reaction

Targeting & Amplification of Specific Genes



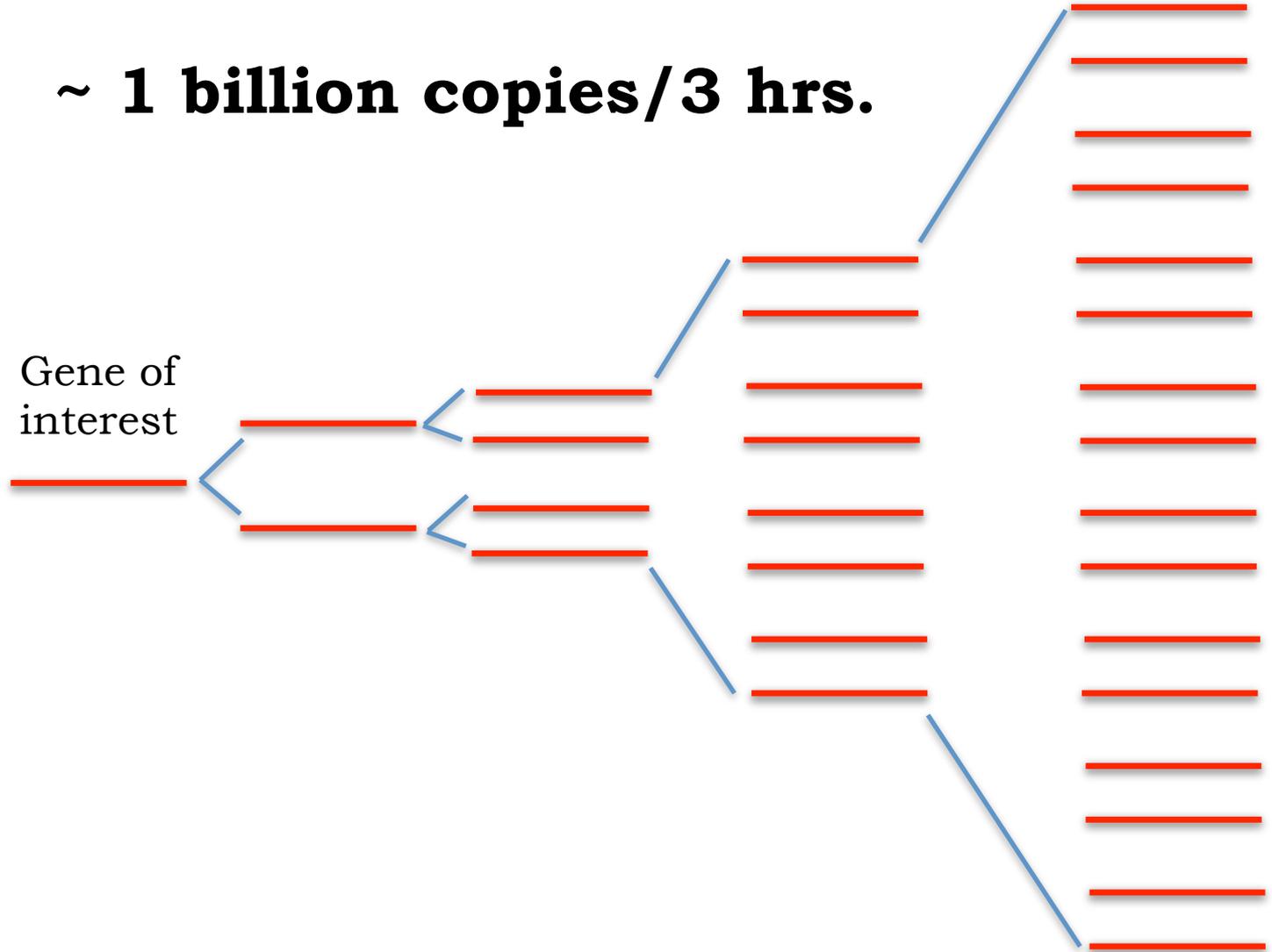
Polymerase Chain Reaction

DNA

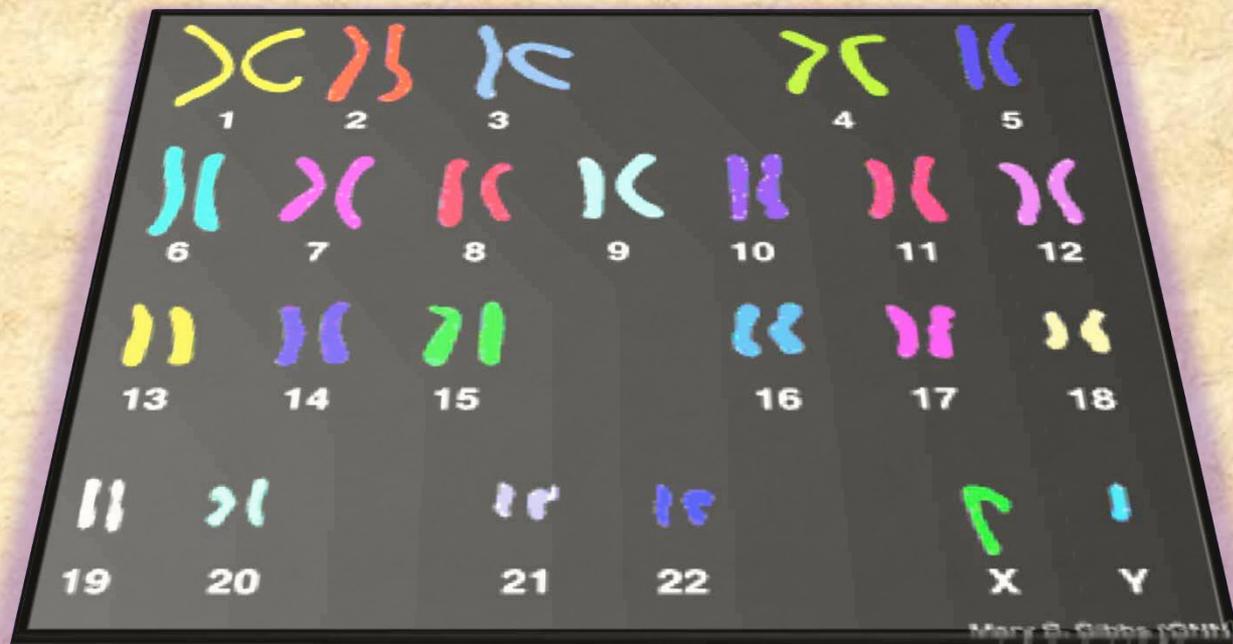
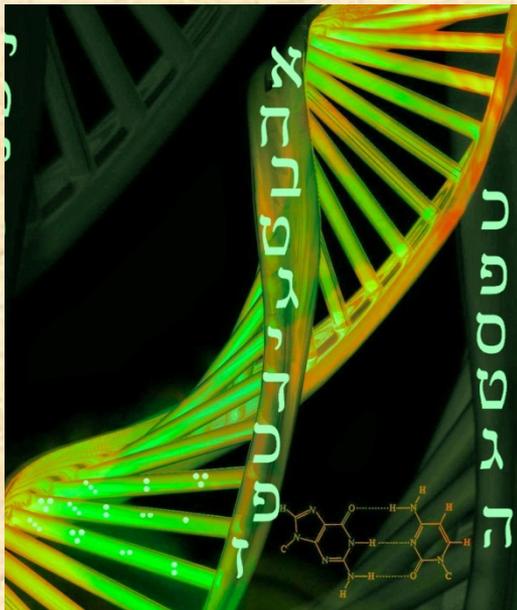


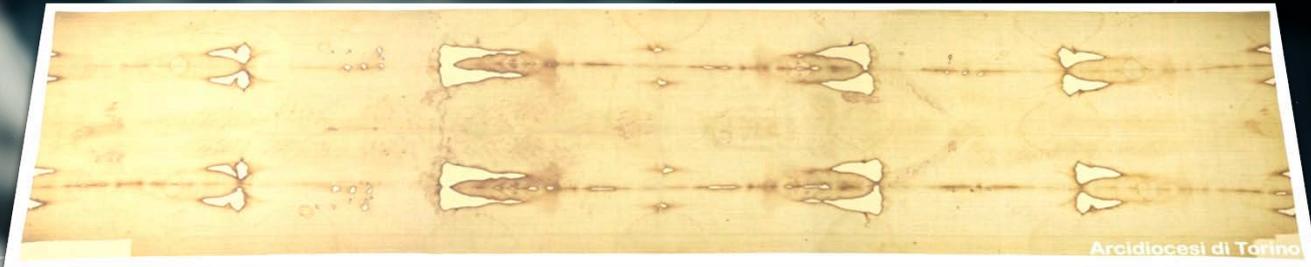
~ 1 billion copies/3 hrs.

Gene of
interest



Is the DNA too degraded for analysis?





Arcidiocesi di Torino



Modern DNA Analysis

nanoschematic

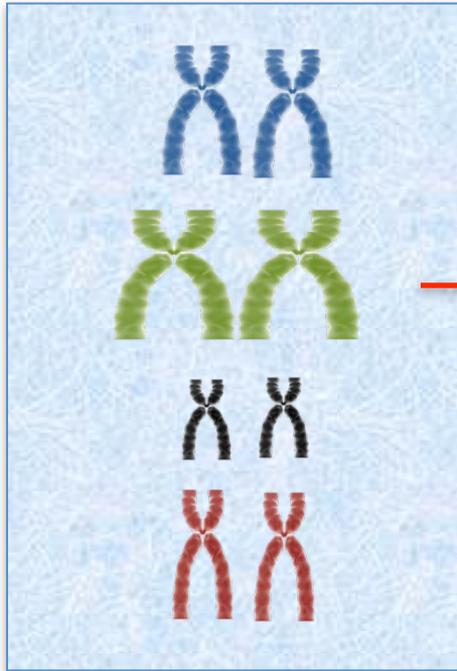
DNA contains the genetic information that allows all modern living things to function, grow and reproduce. However, it is unclear how long in the 4-billion-year history of life DNA has performed this function, as it has been assumed that the earliest forms of life may have used RNA as their genetic material. (1) (2) DNA may have acted as the central part of early cell structures as it can both transmit genetic information and carry the majority of the cell's proteins. (3) This ancient DNA world where nucleic acid would have been used for both structure and genetics may have influenced the evolution of the current world. 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Modern DNA analysis

What type of heterogeneity is present?

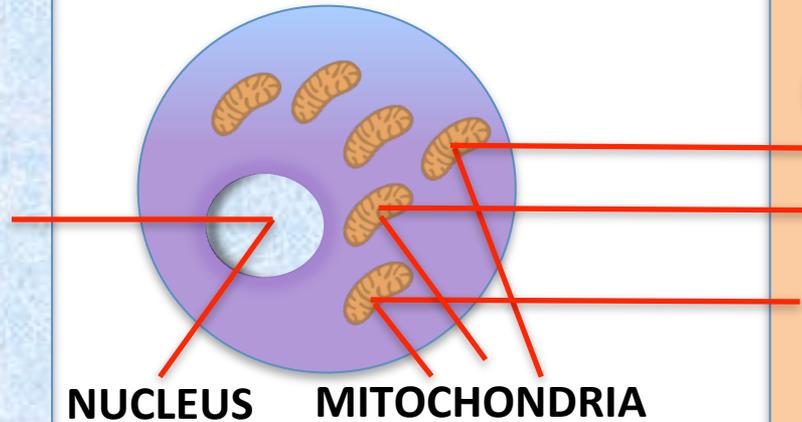
Is the DNA pattern consistent with blood from a single individual?





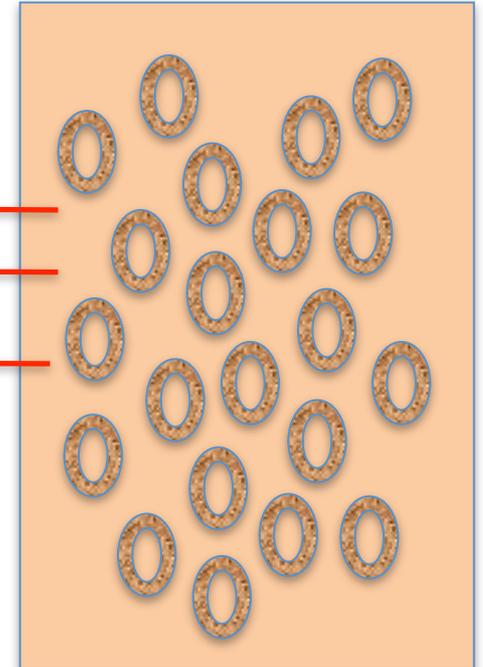
NUCLEAR DNA

2 COPIES PER CELL
20,000-30,000 TOTAL GENES



NUCLEUS

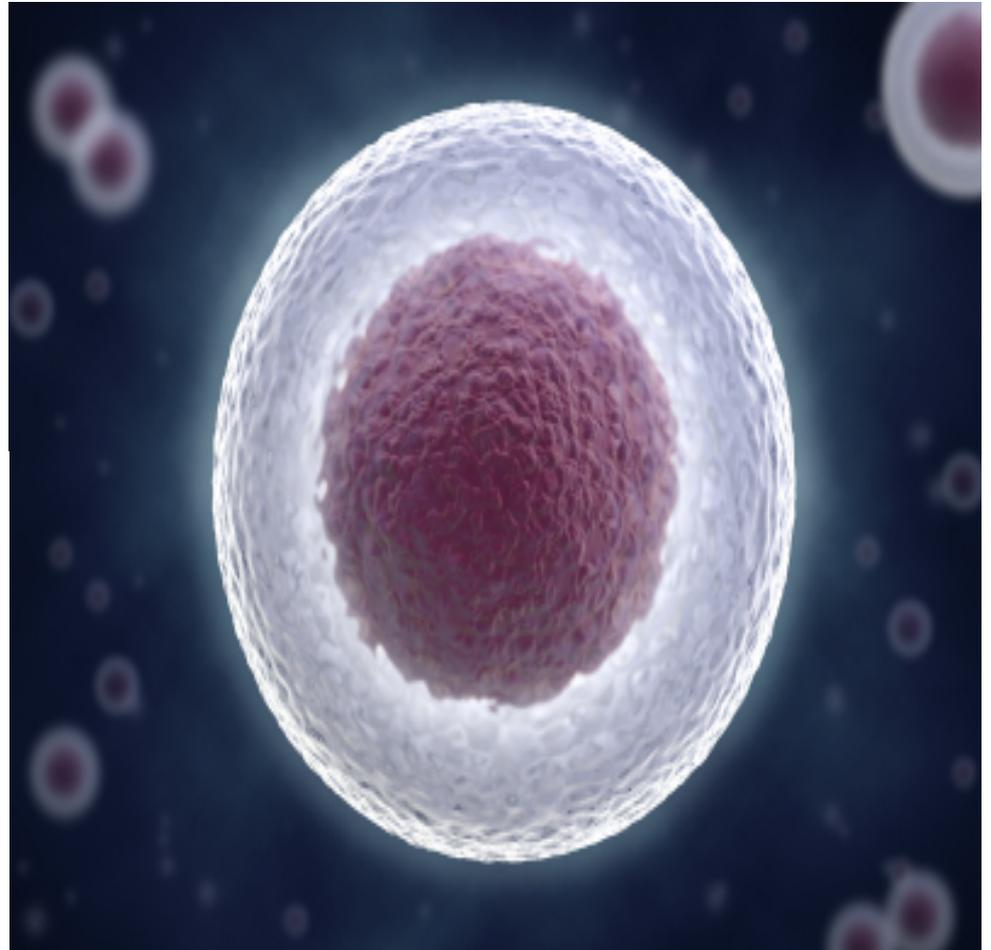
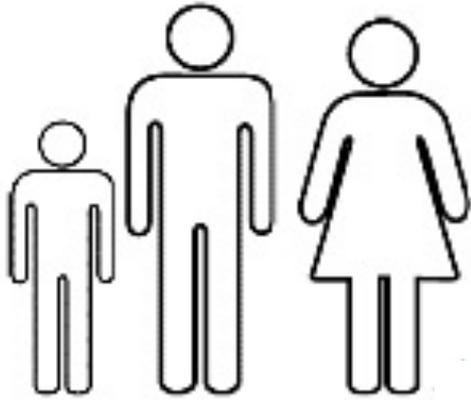
MITOCHONDRIA



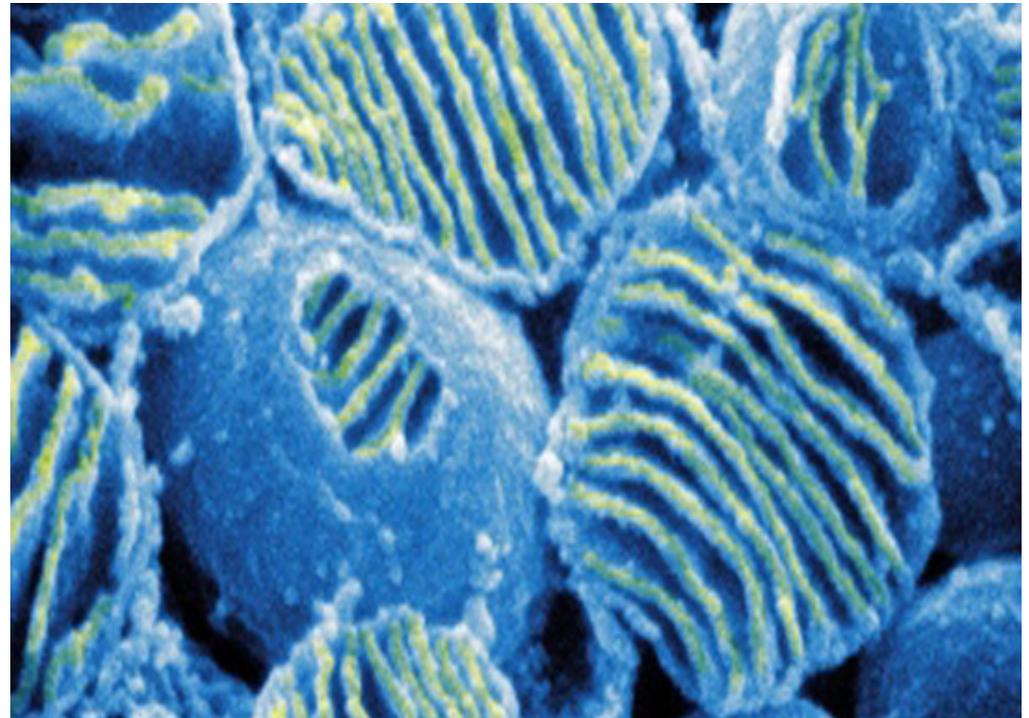
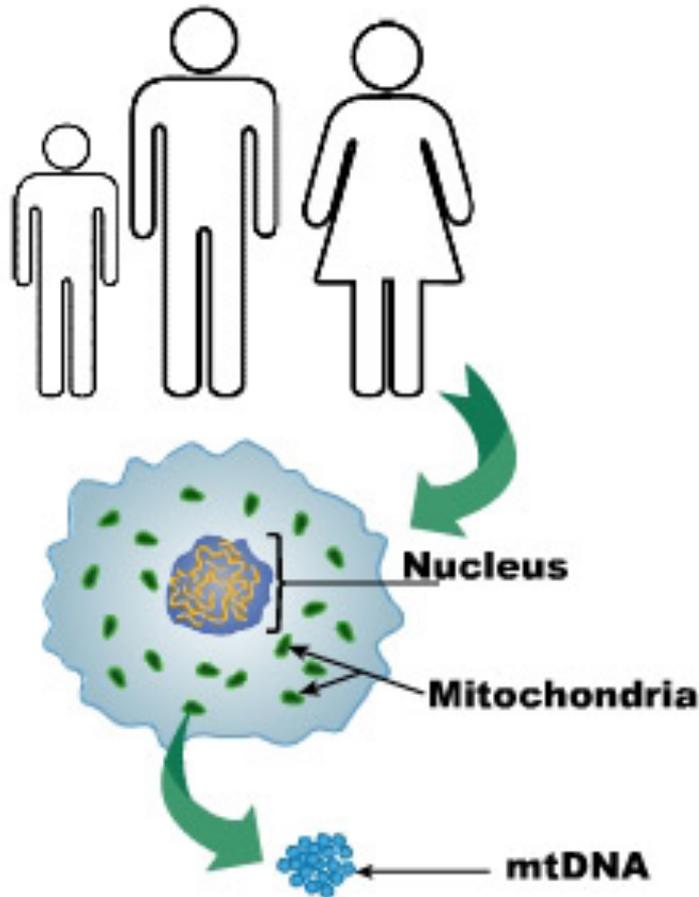
MITOCHONDRIAL DNA

100-10,000 COPIES PER CELL
37 TOTAL GENES

Nuclear DNA is inherited from both parents

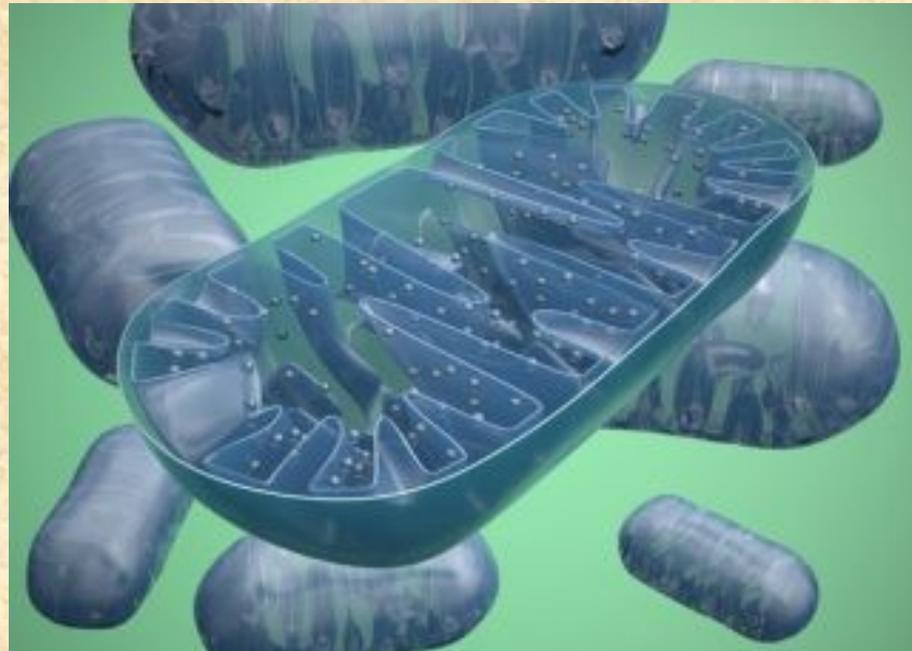


Mitochondrial DNA is maternal in origin



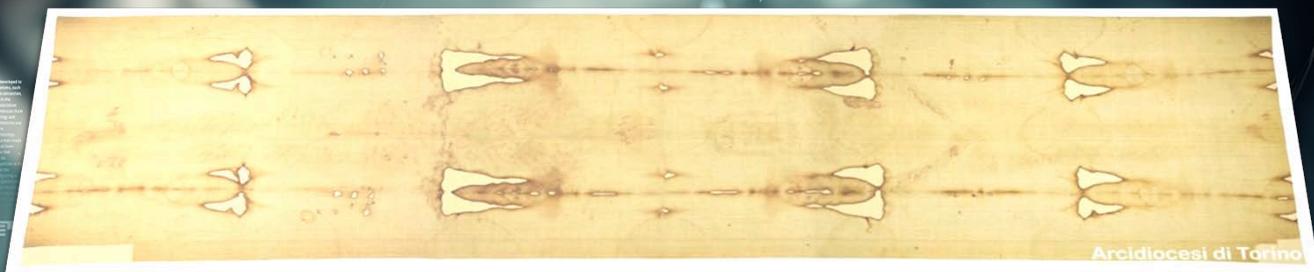
Modern DNA analysis

The chances of two unrelated individuals sharing the same mtDNA profile is extremely remote



Mitochondrial DNA



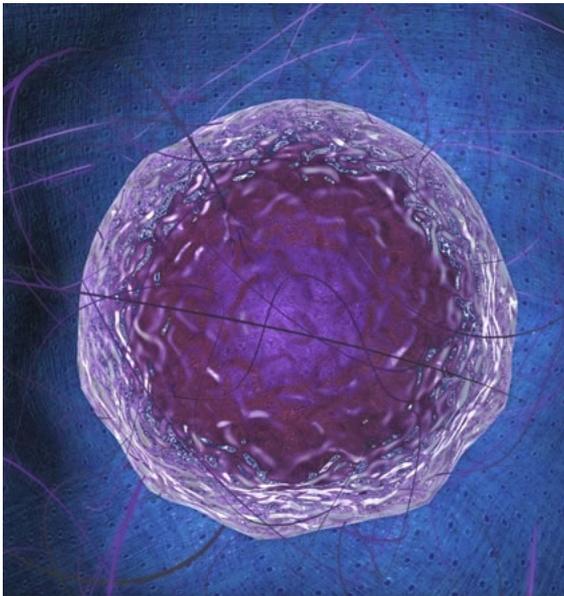


Mitochondrial DNA
Maternal inheritance
Higher copy number



DNA Heterogeneity

Certain DNA segments (genes, non-coding sequences) are identical among all individuals



Certain DNA segments are not identical

Nuclear DNA: 2 forms (max)

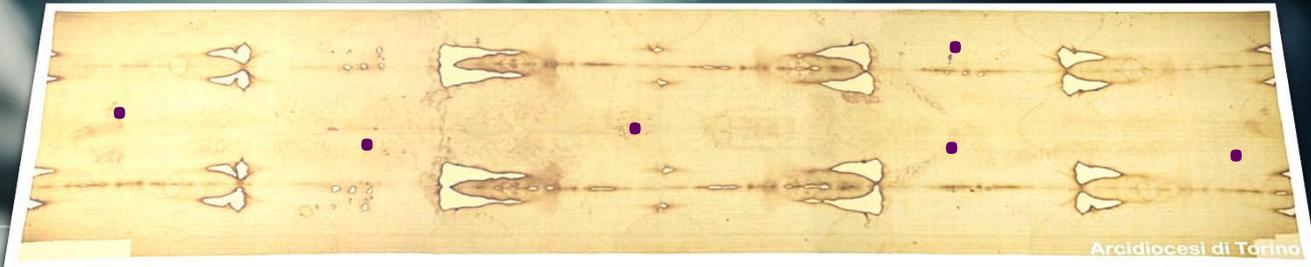
Mitochondrial DNA: 1 form

1: AATTCCGCCG

2: AATTCCGCTG

3: AATCCCGCCG

4: AATTCCACCG

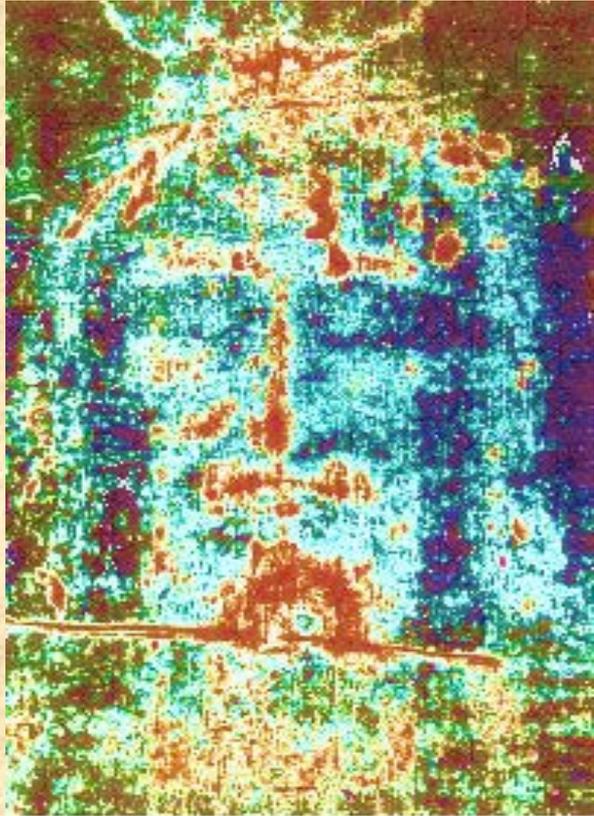


Shroud CODIS Combined Data Index System Multiple Bloodstains

nanoschematic

DNA contains the genetic information that allows all modern living things to function, grow and reproduce. However, it is unclear how long it takes a clone to replicate a copy of the DNA and perform this function. As it has been proposed that the earliest forms of life may have used RNA as their genetic material (ribonucleic acid) may have acted as the central part of many cells in establishing as it can both transmit genetic information and carry out catalysis as part of ribozymes (RNA) this ancient RNA world where genetic code would have been used for both catalysis and genetics may have influenced the evolution of the current genetic code based on four nucleotide bases. This would occur, since the number of different bases in RNA is dependent on a code of between 4 and 100 million bases. Increasing replication accuracy and a large number of 'readings' using the catalytic activity of ribozymes.

However, there is a direct relationship between the number of bases in the genetic code and the number of different amino acids that can be used to build proteins. The genetic code is a set of rules that defines how sequences of nucleotide bases (the DNA or RNA sequence) are translated into the amino acid sequences of proteins. It is a set of rules that defines how sequences of nucleotide bases (the DNA or RNA sequence) are translated into the amino acid sequences of proteins. It is a set of rules that defines how sequences of nucleotide bases (the DNA or RNA sequence) are translated into the amino acid sequences of proteins.

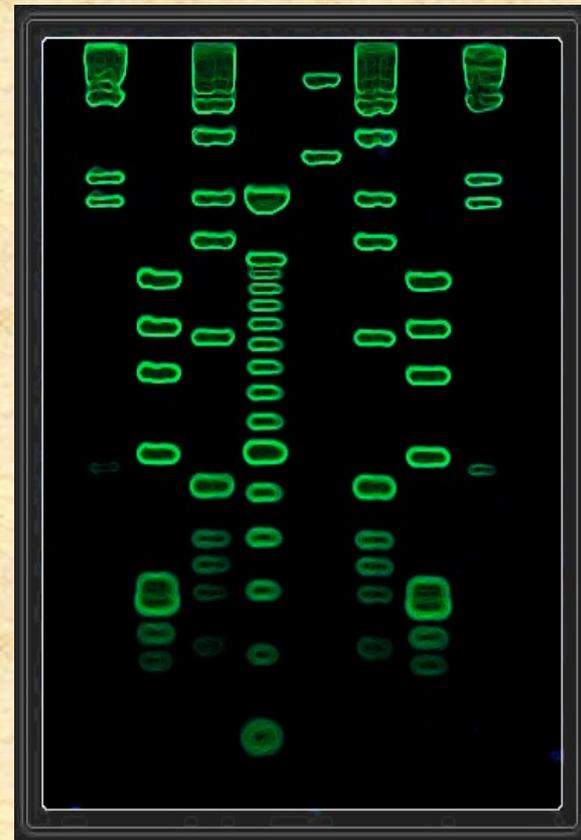
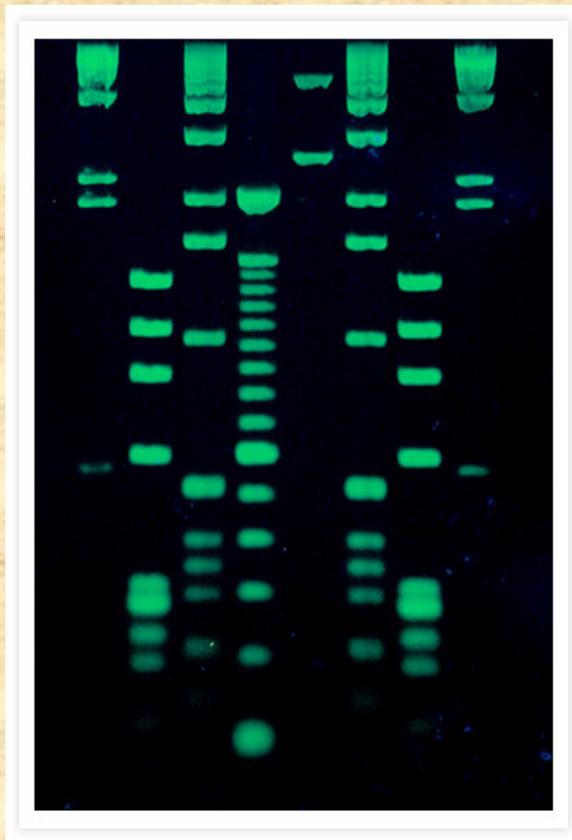
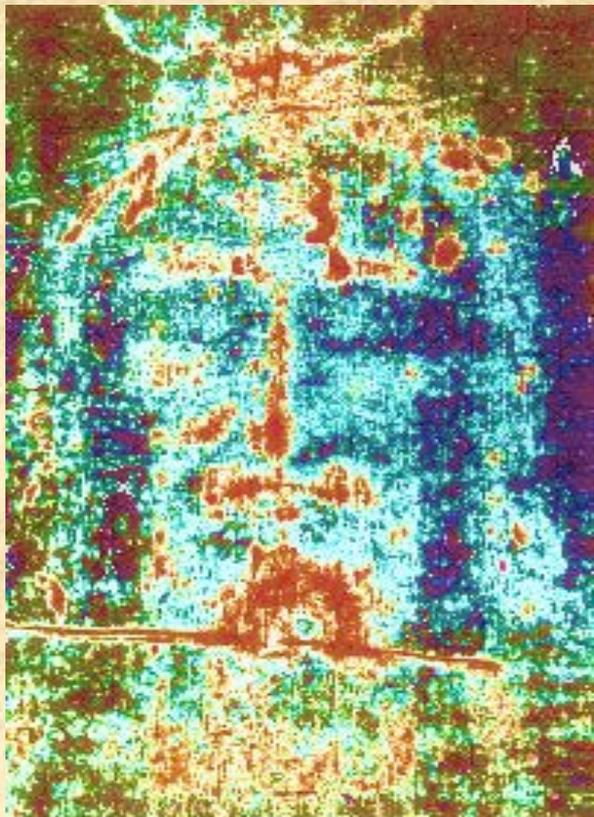


Whose blood is it?

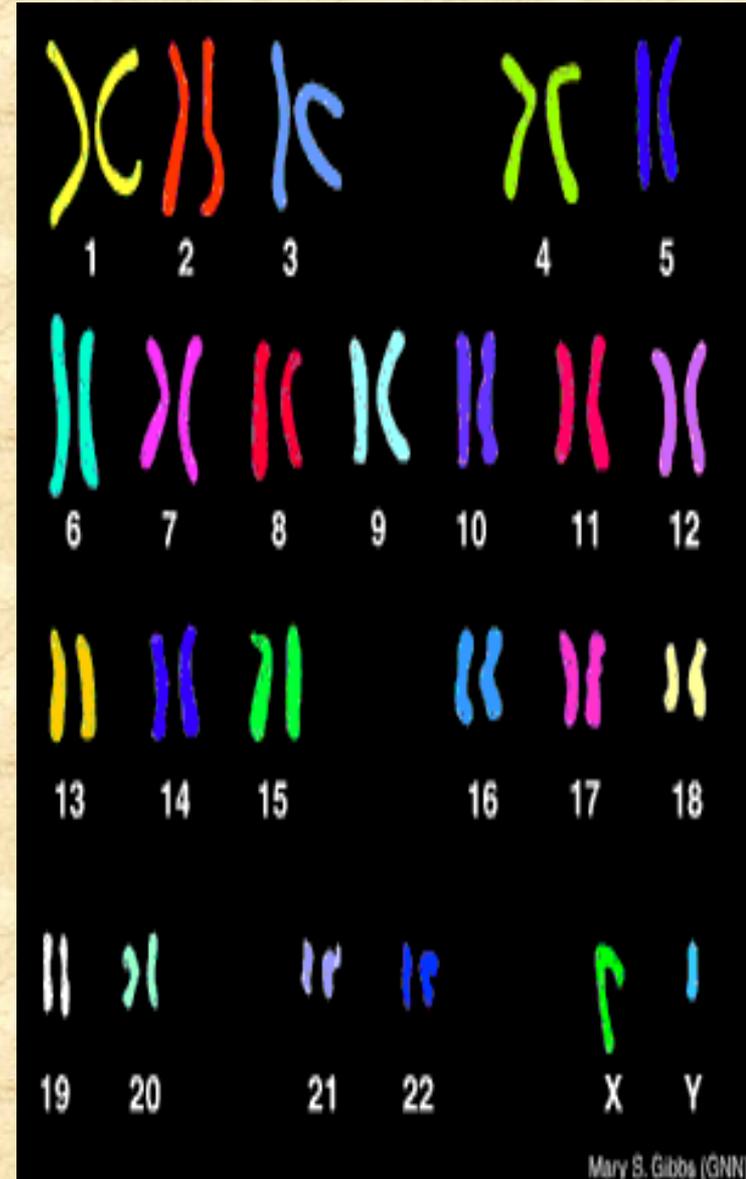
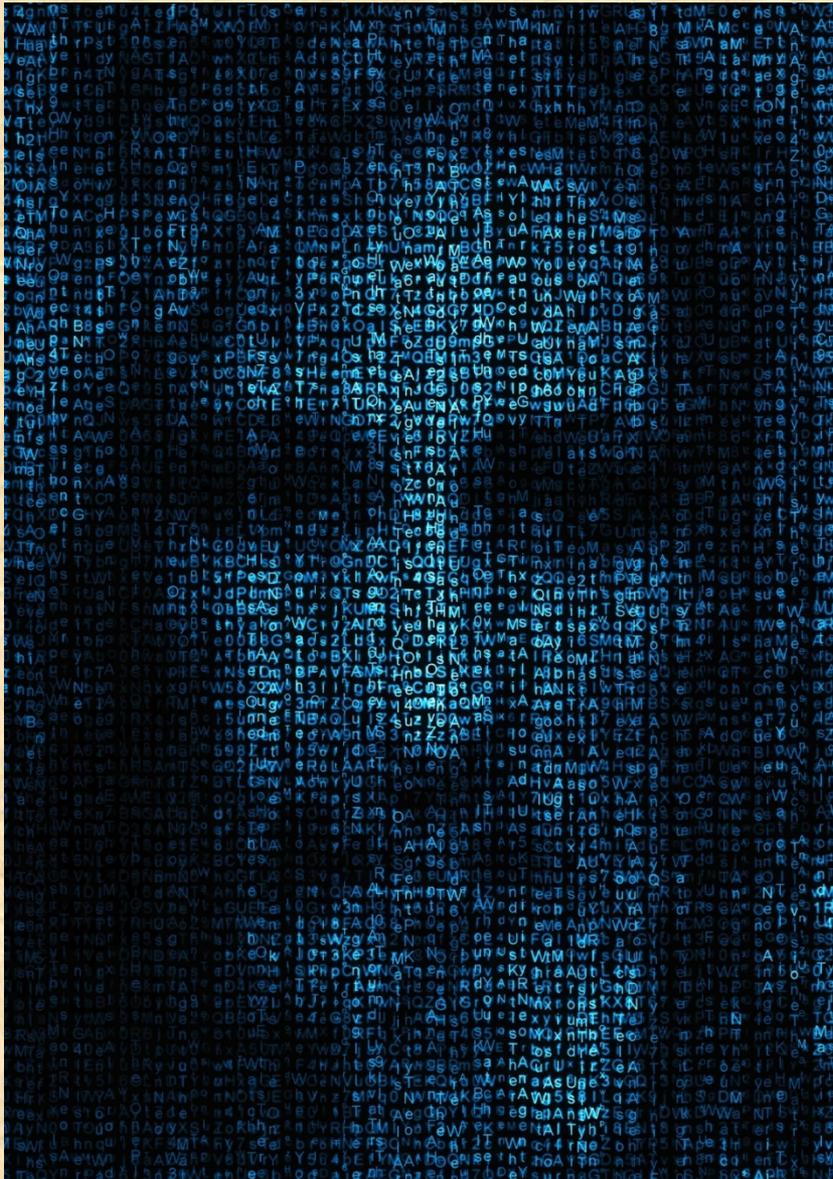
DNA tests are by nature, comparative

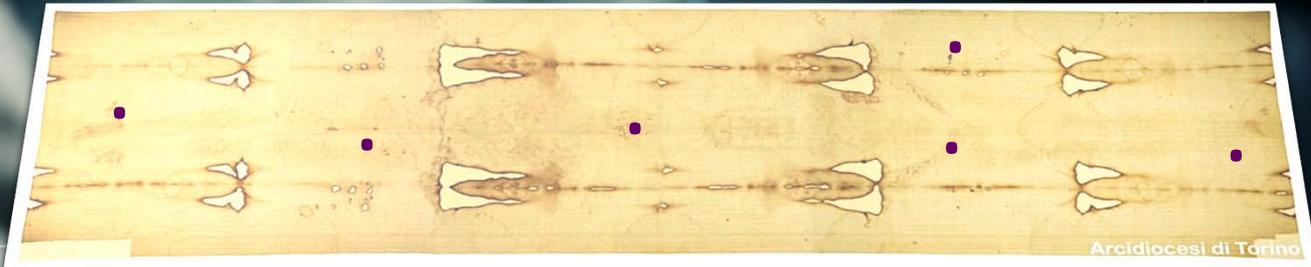
Name: Jesus
Insurance Group ID #: N/A

Sample



Cloning the Man on the Shroud





Shroud DNA Studies Representative of Single/Multiple Individuals? Originates from Blood Cells?

nanoschematic

DNA contains the genetic information that allows an organism living things to grow and reproduce. However, it is unclear how long in the 4-billion-year history of life DNA may have used DNA as their genetic material. Various DNA may have been the central part of early cell metabolism as it can both transfer genetic information and carry out reactions as part of ribosomes. [1] This ancient DNA world, which would have been used for both metabolic and genetic, may have influenced the evolution of the current genetic code based on their metabolic bases. This would occur since the number of preserved bases is an organism in a state of between 10-100 million of bases. The metabolic machinery and other molecules in DNA [2] using the catalytic activity of the enzymes [3].

However, there is a direct evidence of genetic information in the Shroud of Turin. The DNA in the Shroud of Turin is the same as the DNA in the blood of a single individual. This is the first time that DNA has been found in the Shroud of Turin. The DNA in the Shroud of Turin is the same as the DNA in the blood of a single individual. This is the first time that DNA has been found in the Shroud of Turin.