History of Immunology

Molecular Immunology (MIR 511)

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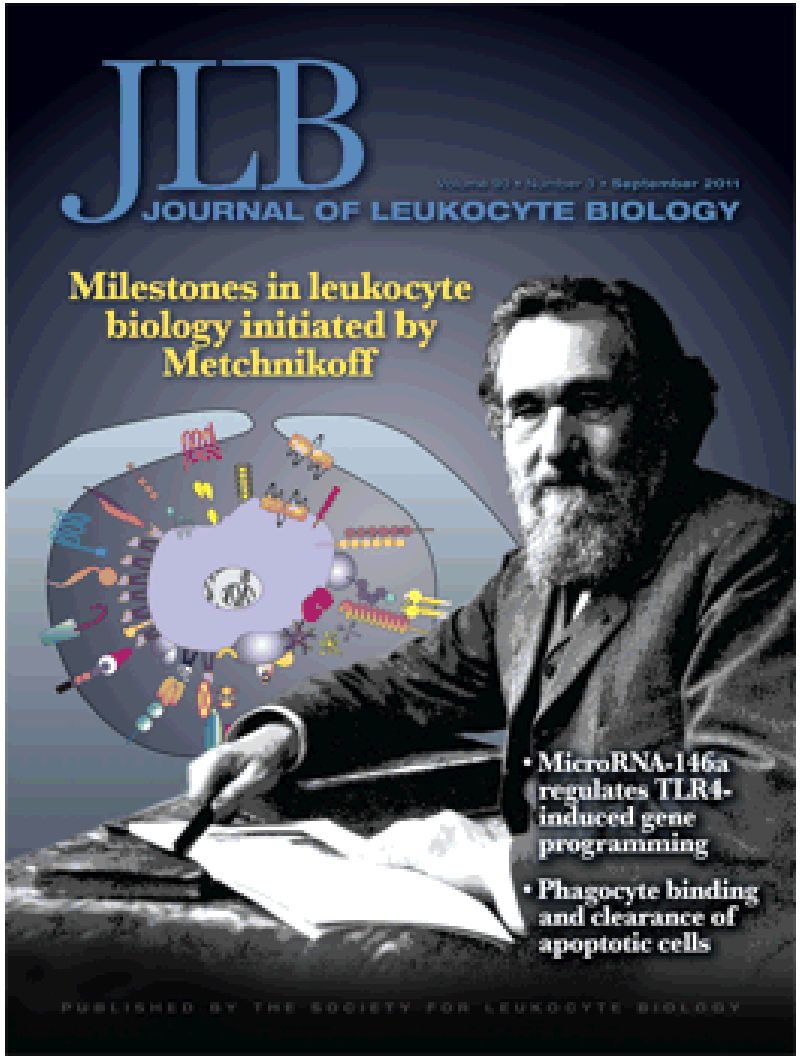
Required reading: Owens; Immunology(7TH Edition)
Chapter 1 – Overview of the Immune System:
A Historical Perspective of Immunity

Objectives

1. To gain a historical perspective of seminal research that provided underpinnings of immunology discipline.

2. To introduce key concepts of tumor immunology.

Assigned Reading



- Arthur M. Silverstein, Ilya Metchnikoff, the Phogocytic theory, and how things often work in science. *J Leuk Biol* 90:409, 2011.

- Jen-Marc Cavaillon, The historical milestones in the undersanding of leukocyte biology initiated by Elie Metchnikoff. *J Leuk Biol* 90:413, 2011.

Historical Paradigms in General Immunology and Tumor Immunology

Recognition of Active Immunity/ Protection from Infectious Agents

Molecular Mechanisms of Immunity (Ab, cells, cytokines)

Tumor Immunity

Survival of Species Depends on Defense Mechanisms

- **Fight/flight**
- **Barriers - skin**
- **Immune response-complexity depends on organism**

  Vertebrates:

  - **Organized lymphoid organs (spleen, thymus, bone marrow, lymph nodes, Peyer's patches)**
  - **Complex circulatory system (lymphocyte trafficking)**

Immunity (Latin)-*immunis*

Legal term = free from tax burden

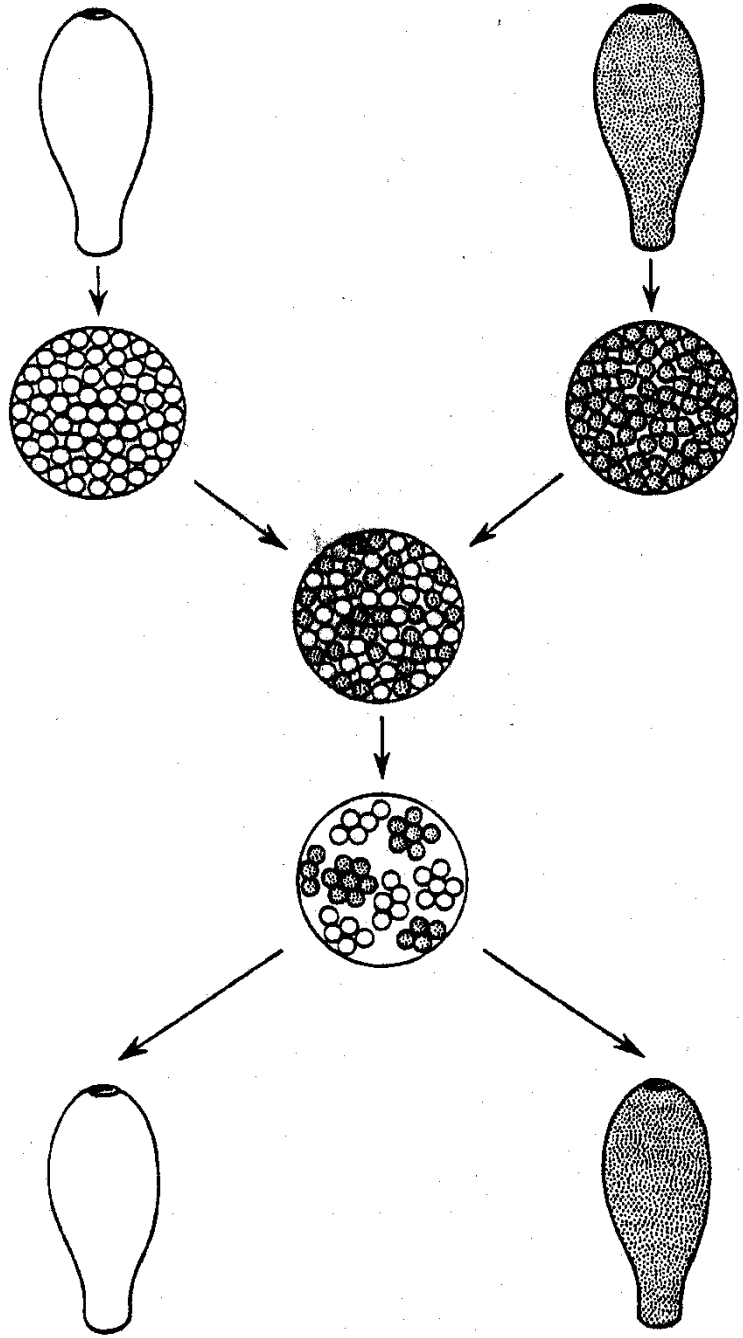
General Properties of Immune Response:

Protect, defend organism from infectious agents

- **Innate immunity (NK, PMN, MØ, megakaryocytes)**
  - **Primitive, higher organism**
- **Adaptive immunity (B, T cells)**
  - **Only vertebrates**

Recognize self from non-self

- **Primitive and higher organisms (Wilson 1907)**



Nonaggressive Incompatibility Reaction in Sponges

(Wilson 1907)

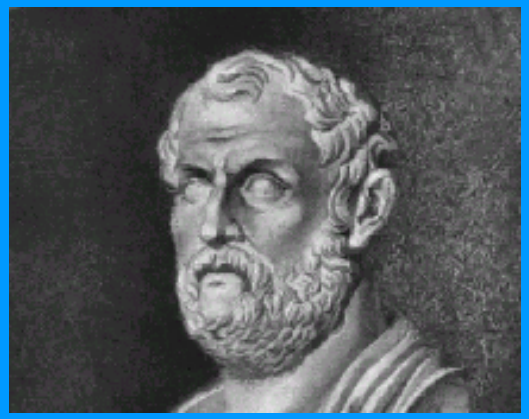
A mixture of dissociated cells obtained from two different species of sponge sorts itself out, and the cells aggregate to form parental body types. (Simplified and highly schematic)

Early Observations of Immunity (epidemics)

- **Examples of people resistant, protected from disease**
- **Attempts to actively induce immunity**

Thucydides (500 B.C.)
Observations on the plague (typhus fever) during the Peloponnesian War

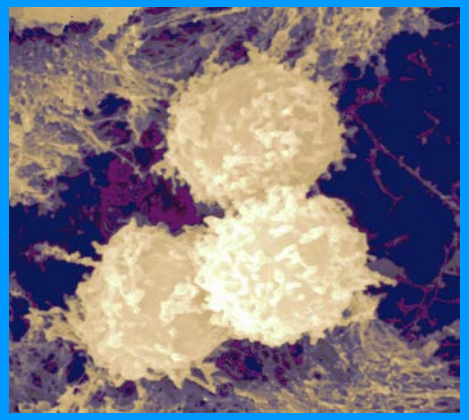
"Yet still the ones who felt most pity for the sick and the dying were those who had had the plague themselves and had recovered from it. They knew what it was like and at the same time felt themselves to be safe, *for no one caught the disease twice*, or, if he did, the second attack was never fatal. Such people were congratulated on all sides, and they themselves were so elated at the time of their recovery that they fondly imagined that *they could never die of any other disease in the future*."



Significance of Thucydides' Recognition of Fundamental Concepts of Immune Response

1. Exposure to disease could result in subsequent immunity (*memory*)

2. Protection to one disease did not confer general protection (*specificity*)



Hallmark Characteristics of the Immune Response

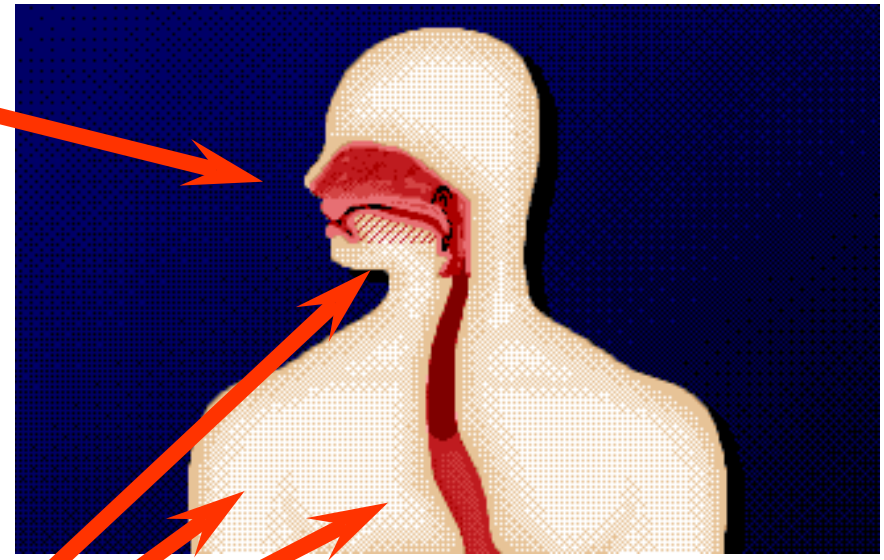
- **Specificity (distinguish subtle differences in Ag)**
- **Immunologic memory (recall response)**
- **Discrimination of self/non-self**
- **Diversity (discriminate 10^9 distinct Ag determinants)**
- **Self-regulation (positive and negative control)**

Smallpox

- **Earliest disease clinically identified**
- **Numerous epidemics (1st evidence on faces of Egyptian mummies - 1570 - 1085 BC)**
- **Led to *first* defined immunology experiments**

Smallpox Etiology

- Inhaled small pox virus infects epithelial cells lining trachea
- Virus spreads via blood to skin epithelium
- Small pox lesions occur on face, body



★ 40% Mortality rate (affects children, young adults)

Early Attempts to Actively Induce Protection Against Smallpox

- **Ancient Chinese dried postules, children inhale through nostril using silver tube (left - male, right - female (B.C.)**

- **Colonies - Cotton Mather (1660s - 1720s)**
  Native Indians, George Washington

Mary Pierrepont Montagu
credited with bringing first awareness of "variolation" process to England

Described method in Turkey to *variolate* healthy individuals using postules from less ill patients.

***Variola* (Latin) = smallpox**
***Variolation* = artificial exposure to small pox**

Letter from Lady Montagu to Sarah Chriswell (1717)

"I am going to tell you a thing that I am sure will make you wish yourself here. The small-pox, so fatal, and so general amongst us, is here entirely *harmless* by the invention of ingrafting.....I am patriot enough to take pains to bring this useful invention into fashion in England and I should not fail to write to some of our doctors very particularly about it, if I knew any one of them that I thought had virtue enough to destroy such a considerable branch of their revenue for the good of mankind!"

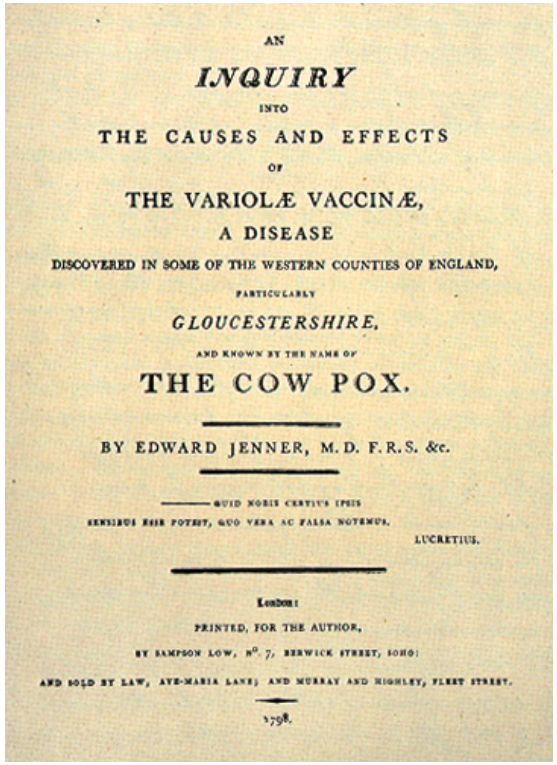
Result of Lady Montagu's efforts in England

Prince & Princess of Whales (& children) were variolated in 1722

Widespread Variolation for Smallpox

- **Danger - high risk of contracting disease (use viable virus to variolate)**

Edward Jenner performed 1st defined immunological experiment



AN

INQUIRY

INTO

THE CAUSES AND EFFECTS

OF

THE VARIOLÆ VACCINÆ,

A DISEASE

DISCOVERED IN SOME OF THE WESTERN COUNTIES OF ENGLAND,

PARTICULARLY

GLOUCESTERSHIRE,

AND KNOWN BY THE NAME OF

THE COW POX.

BY EDWARD JENNER, M.D. F.R.S. &c.

——QUID NOBIS CERTIUS IPSIS

SENSIBUS HAEC POTENT, QUO VERA AC FALSA NOTEMUS.

LUCRETIUS.

London:

PRINTED, FOR THE AUTHOR,

BY SAMPSON LOW, N°. 7, BERWICK STREET, SOHO:

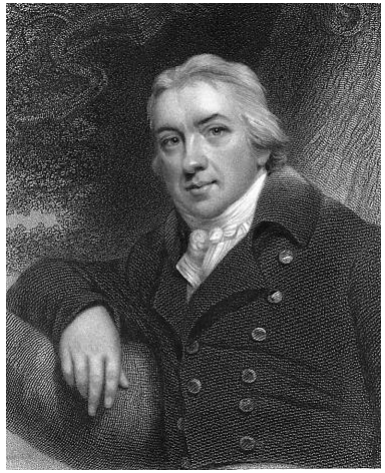
AND SOLD BY LAW, AVE-MARIA LANE; AND MURRAY AND HIGHLEY, FLEET STREET.

1798.

"An Inquiry into the Causes and Effects of Variola Vaccinae" (1798)

***Vaccus* (Latin) = cow (vaccination)**

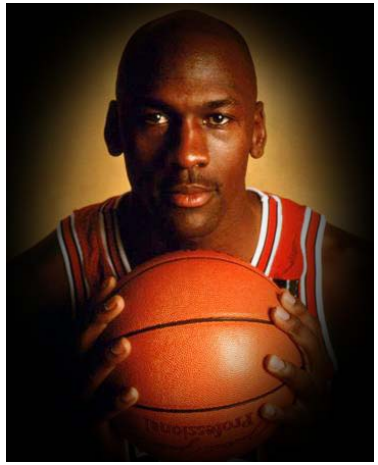
Hypothesis: Pre-exposure to cowpox protects against smallpox infection.



Why Think?

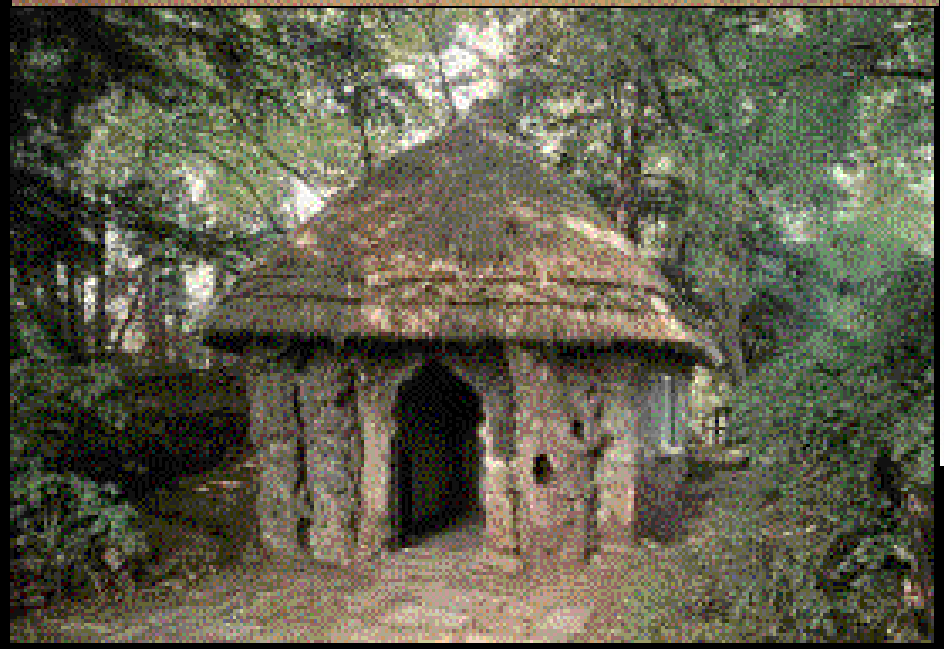
Why Not Try the Experiment?

John Hunter (teacher of Edward Jenner)



Just Do It!

Nike



Thomas Jefferson letter to Edward Jenner (1800s)

"Yours is the comfortable reflection that mankind can never forget that you have lived. Future nations will know by history only that the loathsome smallpox existed."

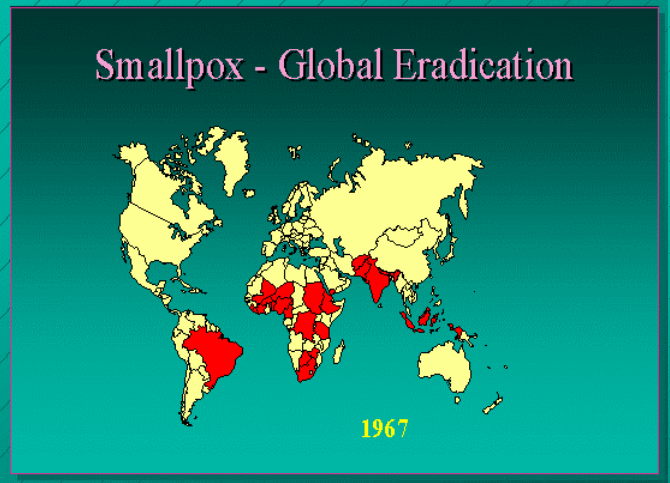
WHO OMS

World Health Organization - Organisation Mondiale de Lasante

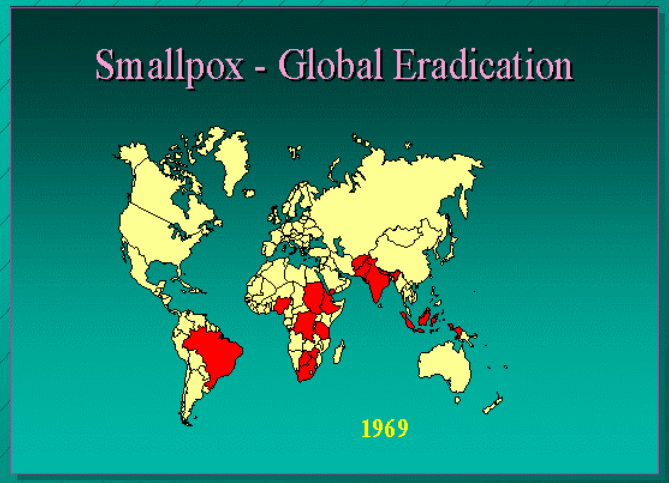
- **Cure for smallpox never found, only protection**

- **1966 > 10 million infected/year**
- **1966 - 1977 - Initiative to eradicate smallpox by vaccination**

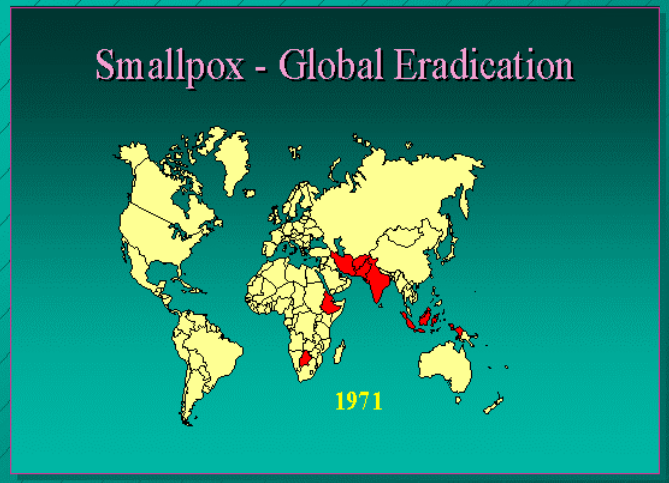
1967



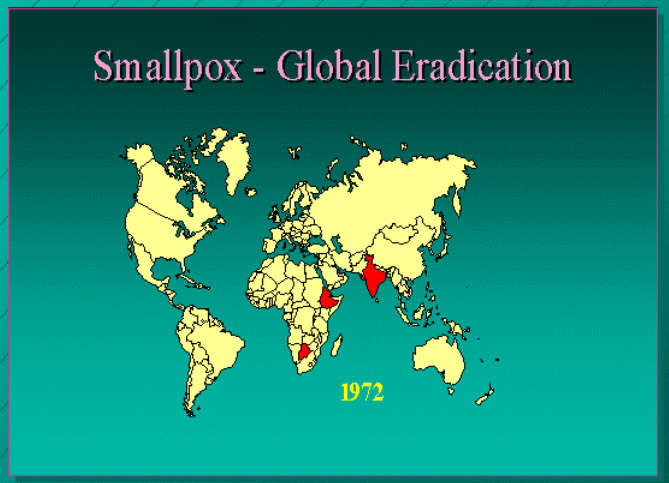
1968



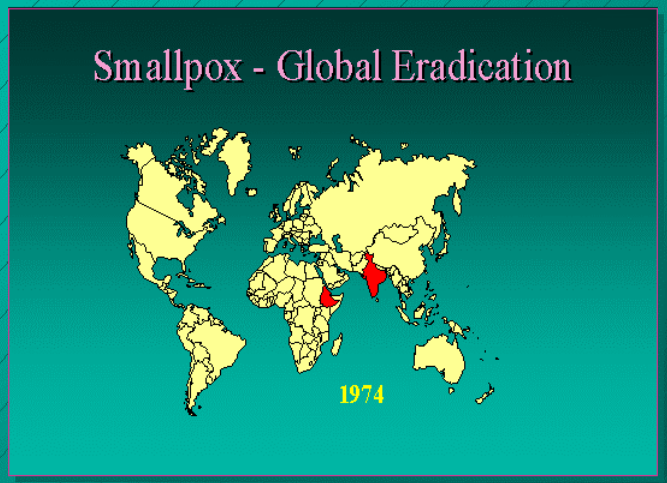
1971



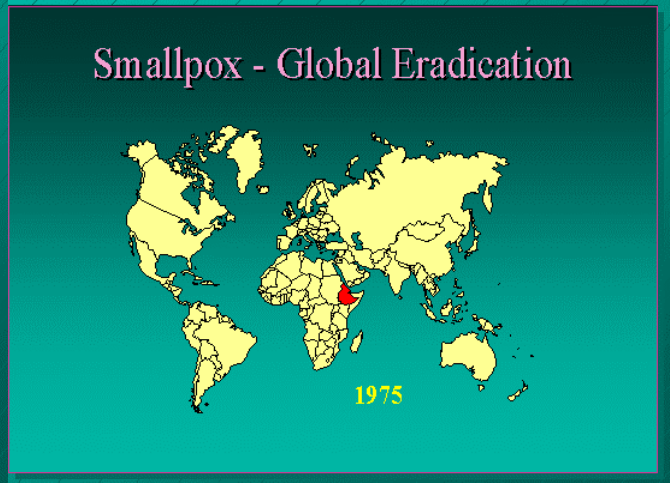
1972

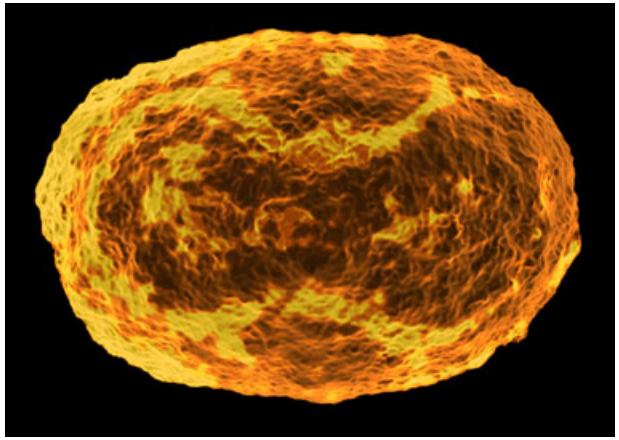


1974



1975





WHO 1980

- Smallpox is the first infectious disease to be eradicated by worldwide program of vaccination

- Ethical debate over destruction of remaining vials
  - Virulent smallpox too dangerous to keep (germ war-fare)
  - May be necessary to use virus to develop anti-viral reagents (humans only host)

Impact of Jenner Study on Immunology

- **Widespread acceptance of method for inducing immunity to infectious disease. Safer than variolation using smallpox.**

- **Thought only living organisms could confer immunity (not immediately adaptable to other diseases).**

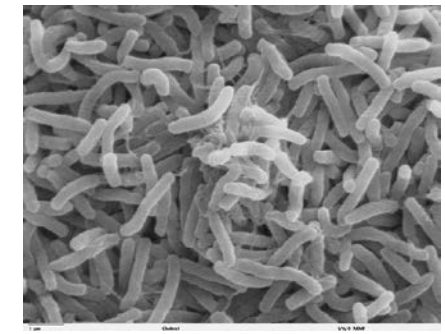
- **Protection not passed from generation to generation. Studies not directed toward understanding mechanisms.**

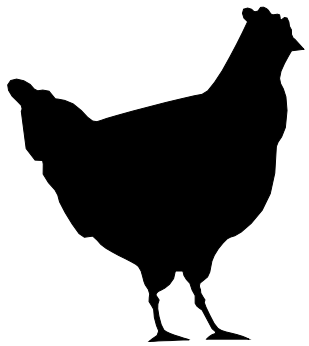
Louis Pasteur

'Father of immunology'

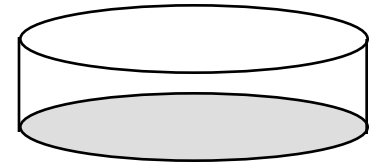
"Chance favors only the prepared mind."
1878

Vibrio cholerae

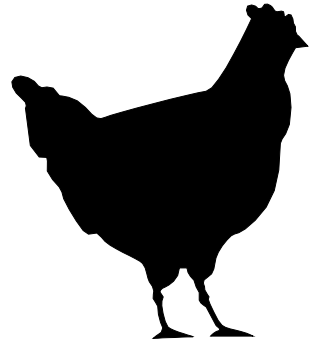




Isolate cholera (bacteria)



Bacterium dies
(accidentally)



1. Inoculate with attenuated bacteria:
Animal lives

2. Challenge with lethal dose cholera toxin:
Animal protected

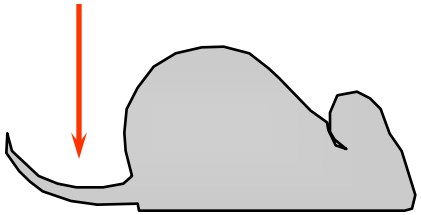
Significance of Pasteur's Findings

Process called *vaccination* in homage to Jenner

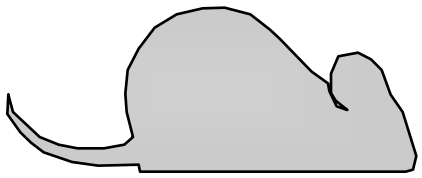
- Demonstrated weakened, attenuated bacteria can serve as vaccine
- Safer ⟶ concept of prophylactic therapy
- Infectious disease had specific identifiable causative agents
- Field dominated toward isolating infectious agents

Emil von Behring - Landmark Experiment Demonstrating "Anti-toxin" Basis of Immune Response (*1898*) – *Awarded 1st Nobel Prize 1901*

Immunize DT (attenuated bacteria)



Remove serum
Adoptively transfer to naïve recipient



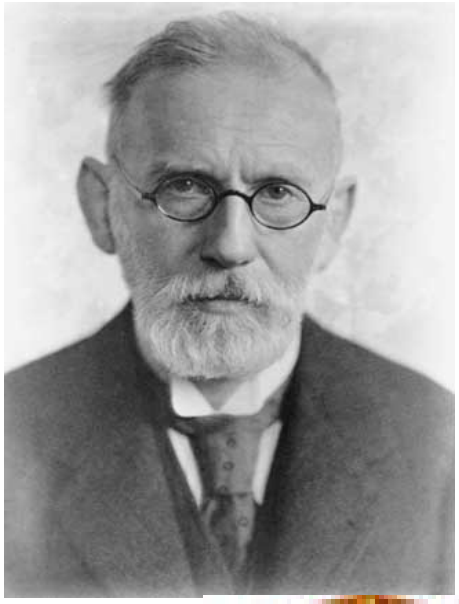
Challenge with DT

Resistant **to DT,**
not **other infectious agents**

Conclusions

- **Antitoxin in serum can neutralize toxic effects of infectious agent (DT)**
- **Specificity - neutralize DT but not other bacterial toxins**

Paul Ehrlich Scientific Contributions
Nobel Prize in Immunology - 1908



- Founder of scientific discipline of immunology
- Impact broader than immunology

"The immune substances…..in the manner of magic bullets, seek out the enemy."

–Paul Ehrlich

Paul Ehrlich

Robert Koch
Nobel Prize – 1905
Discovered causative agent and testing methods for tuberculosis, anthrax, cholera, pink eye→ revolutionized bacteriology

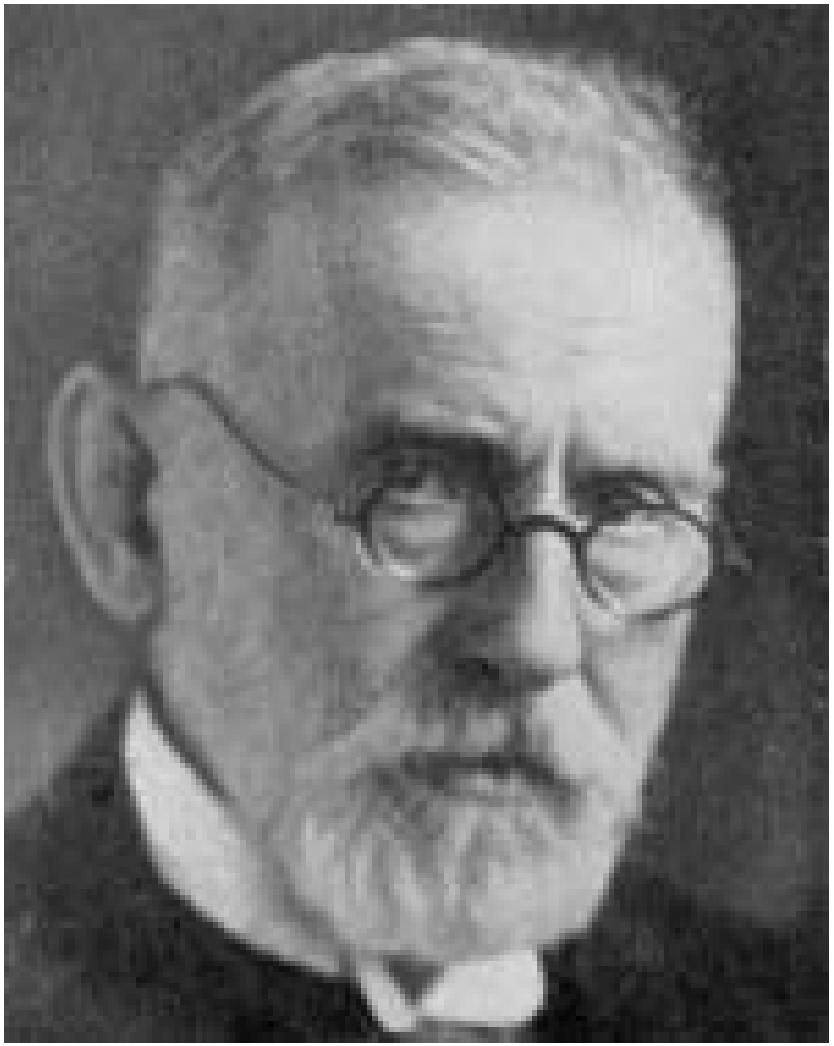
- 1891 – Koch hired Ehrlich at the Institute for Infectious Disease
- Brought together Paul Ehrlich and Emil von Behring who was working on anti-diphtheria serum immunotherapy
- Ehrlich applied quantitative analysis to immunology

Limitations of von Behring's Serum Therapy

- Serum production difficult, yield of serum insufficient
- Failed to standardize the sera so not reproducible
- Ehrlich developed quantitative methods to measure sera activity - *"I made it my task to introduce measures and figures into investigations regarding the relations existing between toxine and antitoxine" P Ehrlich, 1900*
- Produced huge quantities of standardized serum, provided to pediatric clinics

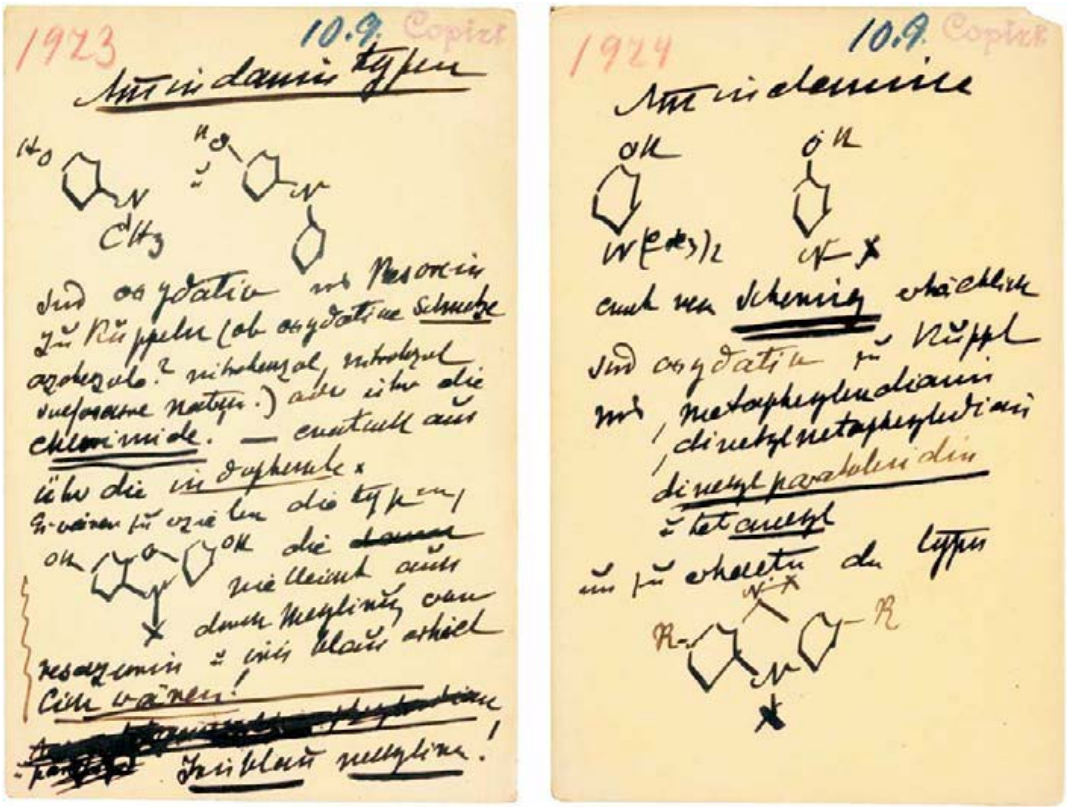
  1895 – German Congress of Internal Medicine – *'New remedy unequivocally assessed the best treatment every realized'*

Paul Ehrlich – Guiding Principles



- Systematic experimentation
- Quantitative, chemical basis of all questions
- Important to test biological responses in vitro & in vivo

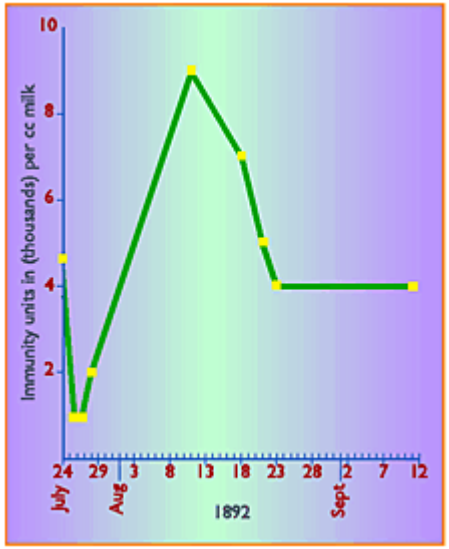
Paul Ehrlich – Guiding Principles



- Obsessed with organic chemistry
- Believed in rigorous recording ideas, experiments-developed own orthography
- Daily handed out carbon copies with ideas written on it

Kaufmann, Nature Reviews Drug Discovery, 7, 2008

The Most Elegant Immunological Experiment of the 19th Century

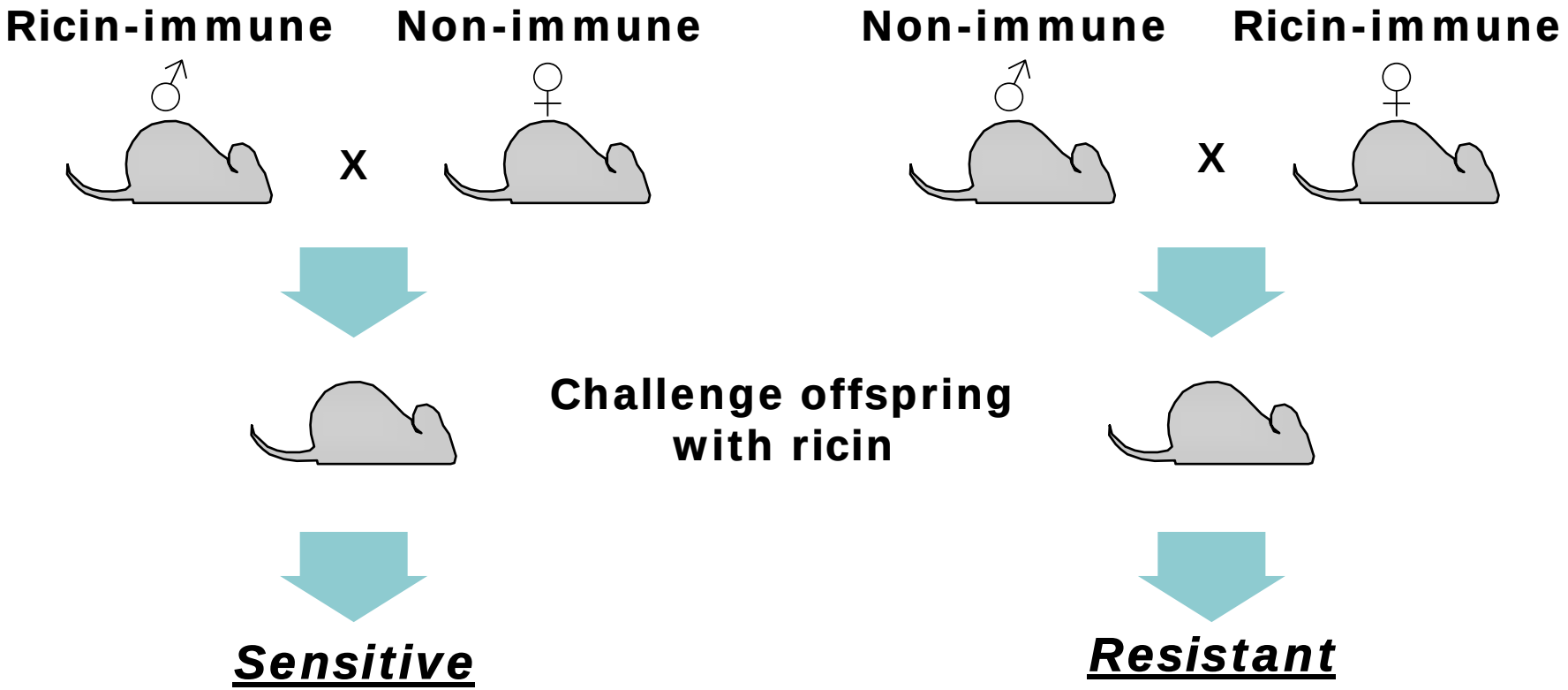


Question: Does antibody in neonates derive from the mother (passive transfer) or father (genetic transfer)?

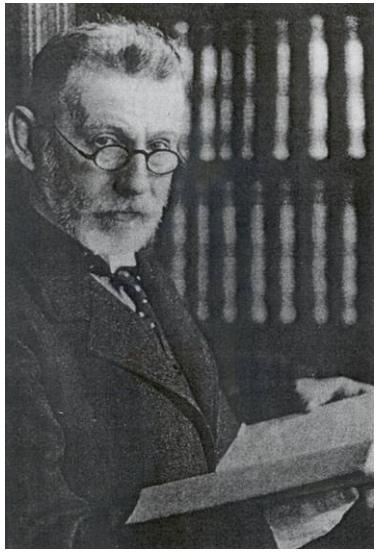
"I have been able to succeed in finding a simple research plan..." *Paul Ehrlich 1892*

Nature Immunol 1:93, 2000

Ehrlich's Experimental Plan



'It is not to be doubted that the immunity that we have observed in the offspring of immune mothers....depends on the transfer of maternal antibody.' (1892) P Ehrlich



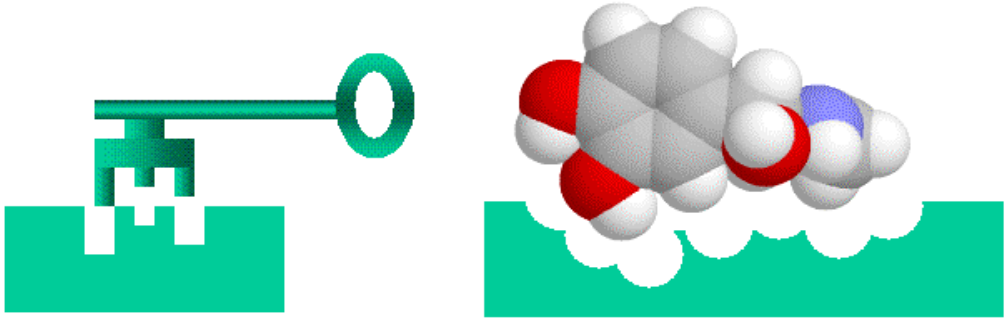
Paul Ehrlich – Side-Chain Theory

- 1st comprehensive theory of antibody formation
  Side-chain theory

- Addresses question of how immune response (anti-toxins) distinguishes so many antigens with such specificity

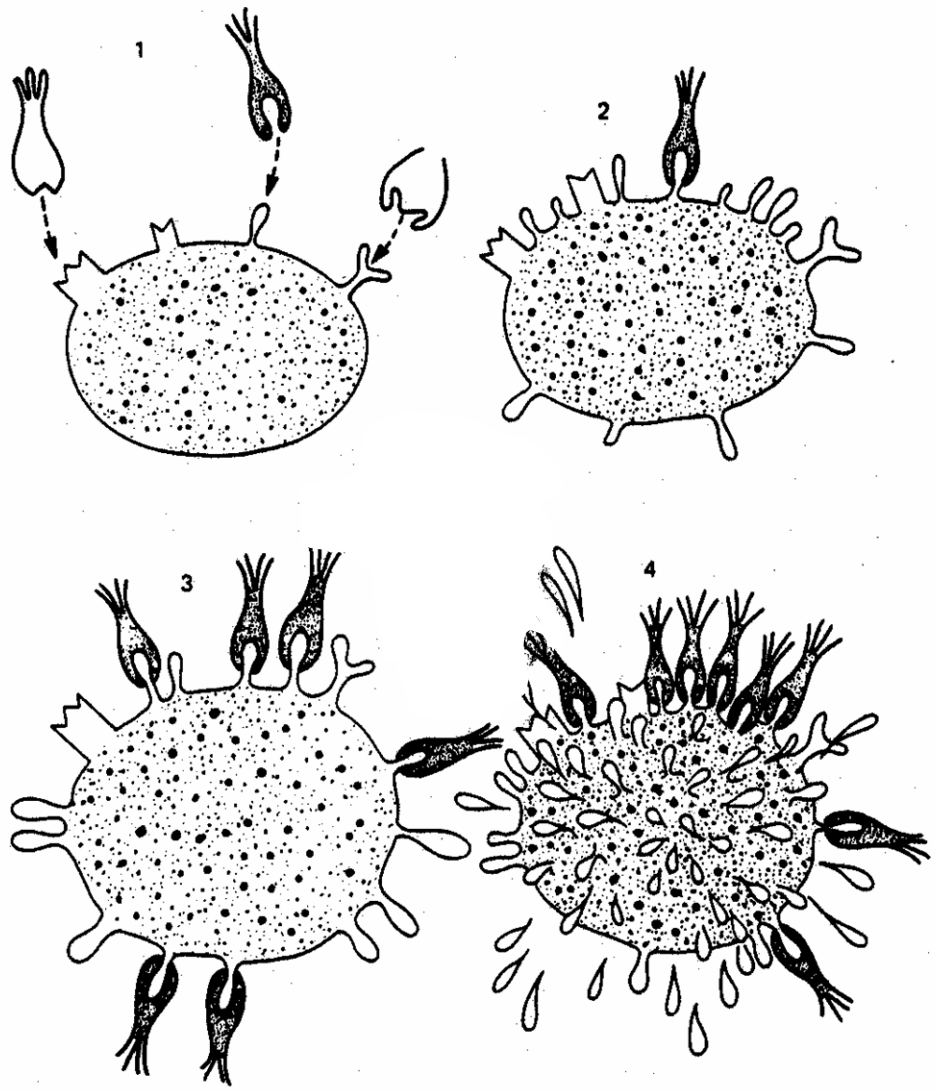
Emile Fischer – Lock and Key hypothesis for enzyme-substrate interactions

- Nobel Prize in chemistry (1902) for research on sugars, proteins, fats and enzymes



Paul Ehrlich's Side-chain Theory of Antibody Formation (1897)

- No physical evidence for existence of antibodies
- Innovation of using diagrams to illustrate hypothetical molecules
- New way of thinking about immunology – first coined term 'antibody'; receptor novel concept.
- Antigens bind to pre-existing cell surface receptors, stimulate cells to synthesize more receptors and to secrete them into the extracellular fluid.



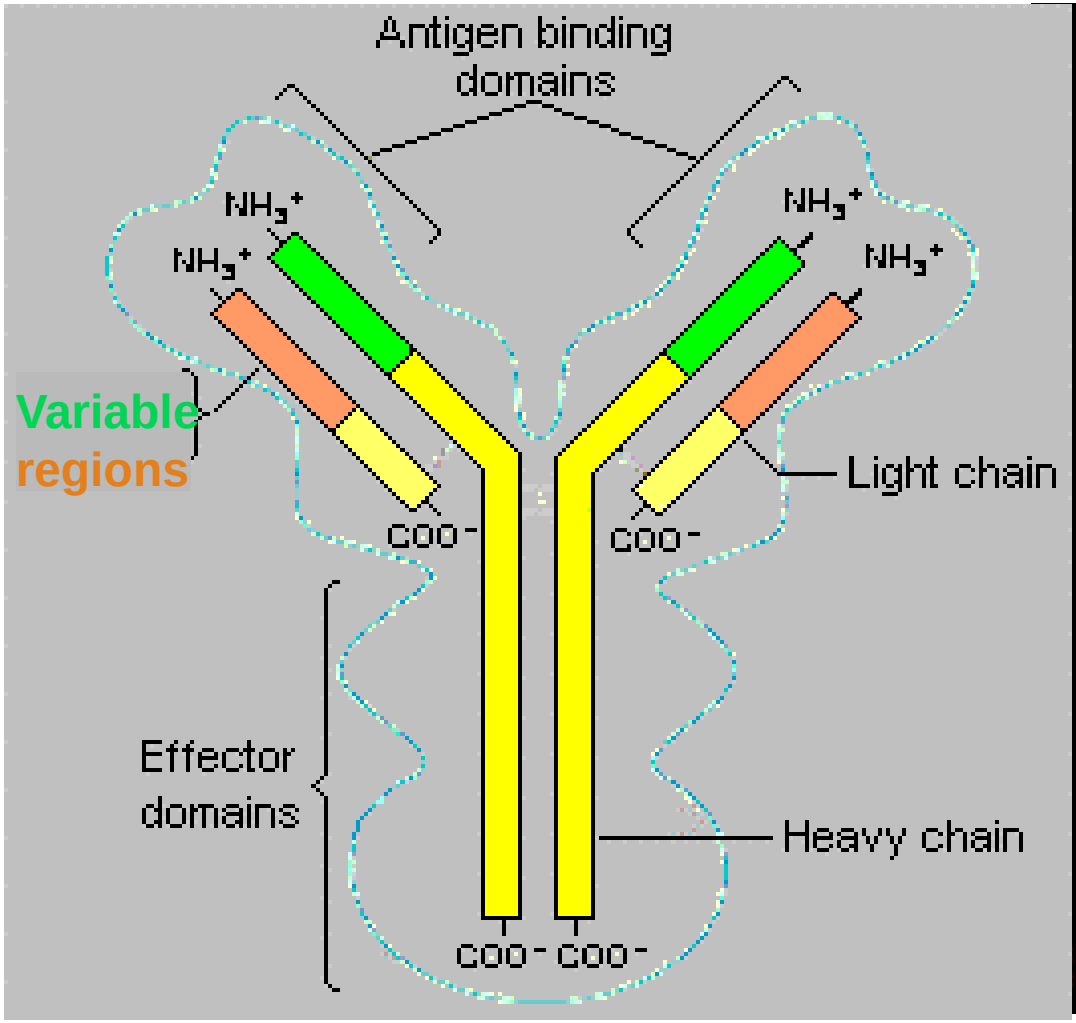
Draft of side chain theory in office of Paul Ehrlich

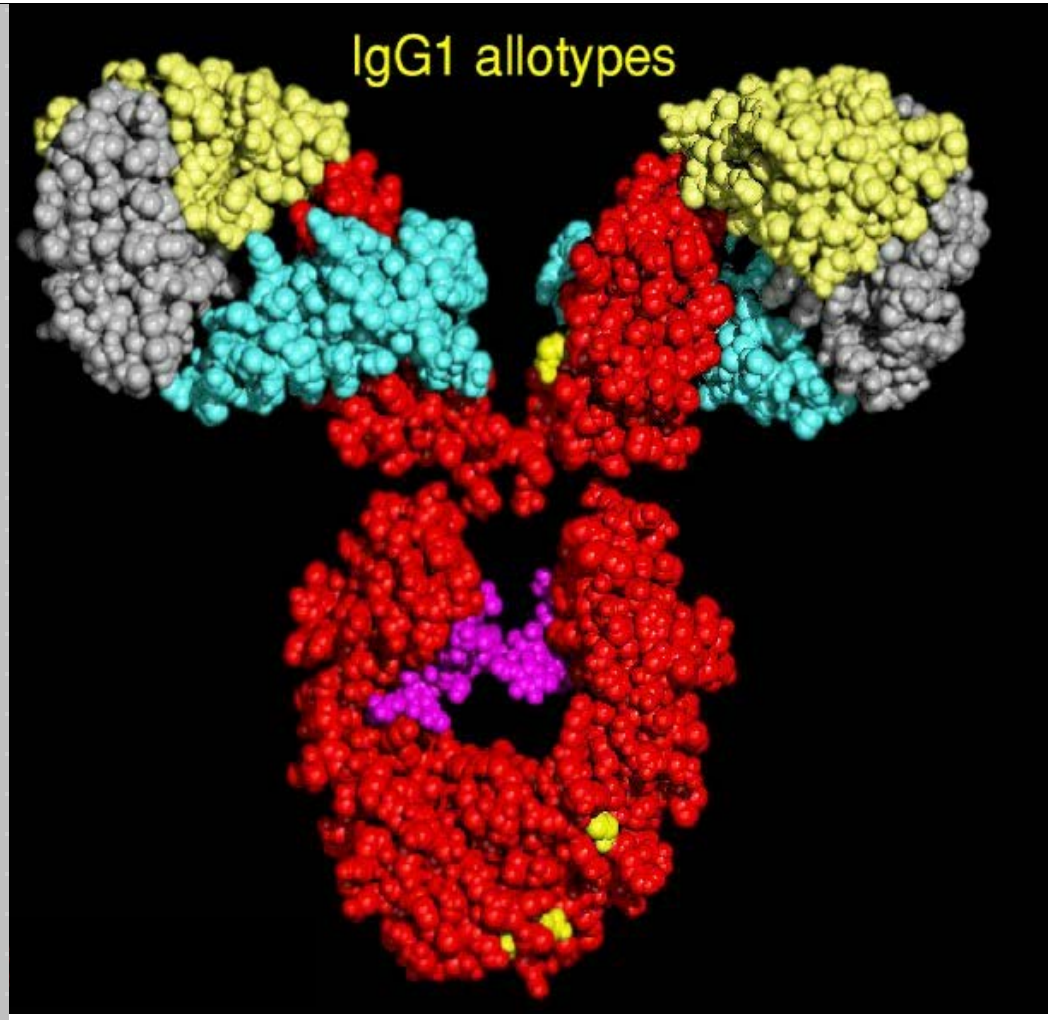
Microbes and Infection, 6, 2004

Humoral (Ab/serum) vs. Cellular Immunity

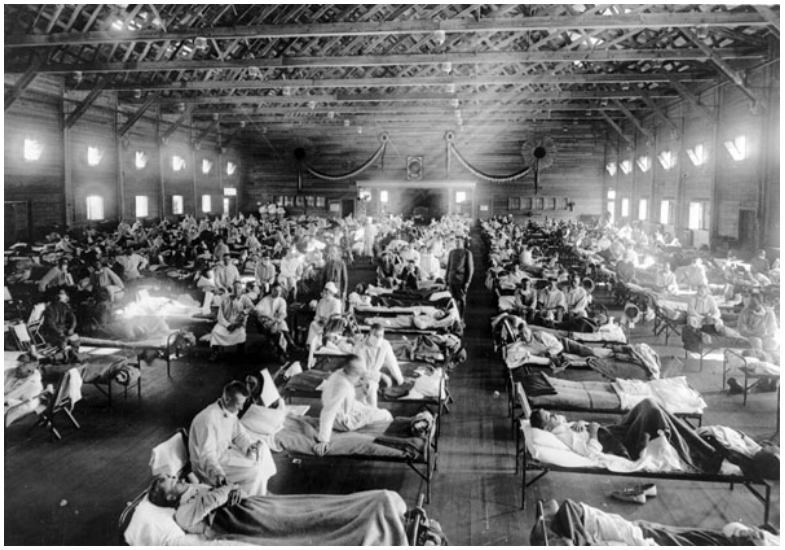
- New paradigm - reports by Ehrlich, von Behring support concept Ab responsible for immunity, i.e., cells not necessary.
- Next 50 years dominated by study of *Immunochemistry* (Ab structure, Ab/Ag interactions, cellular source of Ab)
- Study of cellular immunity largely ignored (Metchnikoff)

Immunoglobulin Structure





Durability of Ab Response

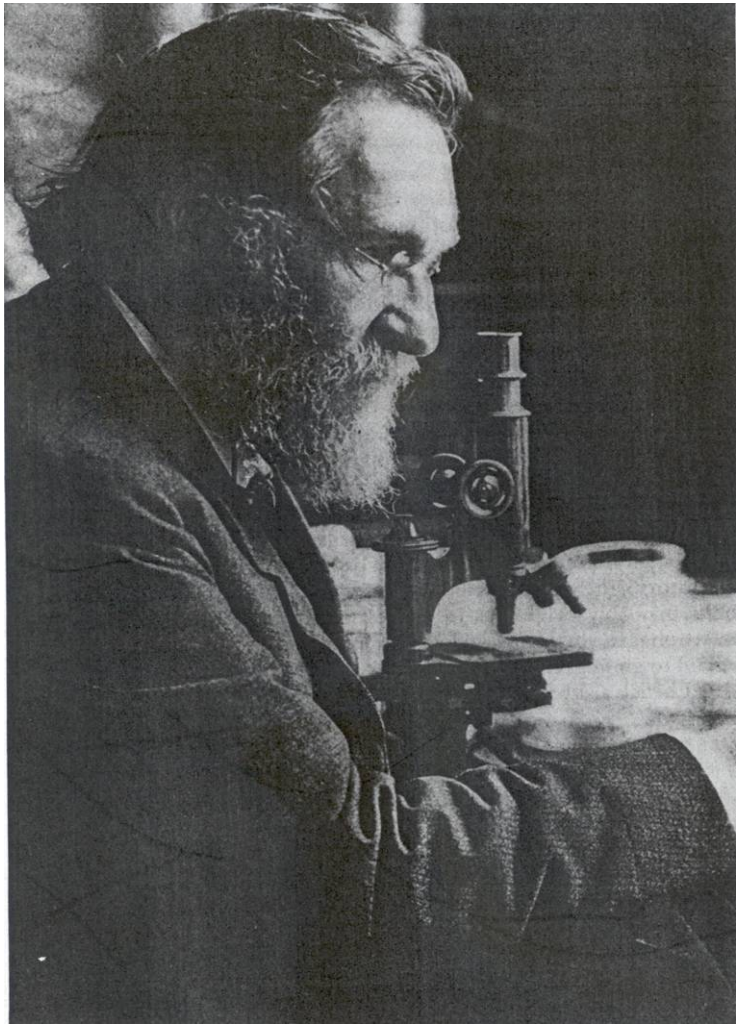


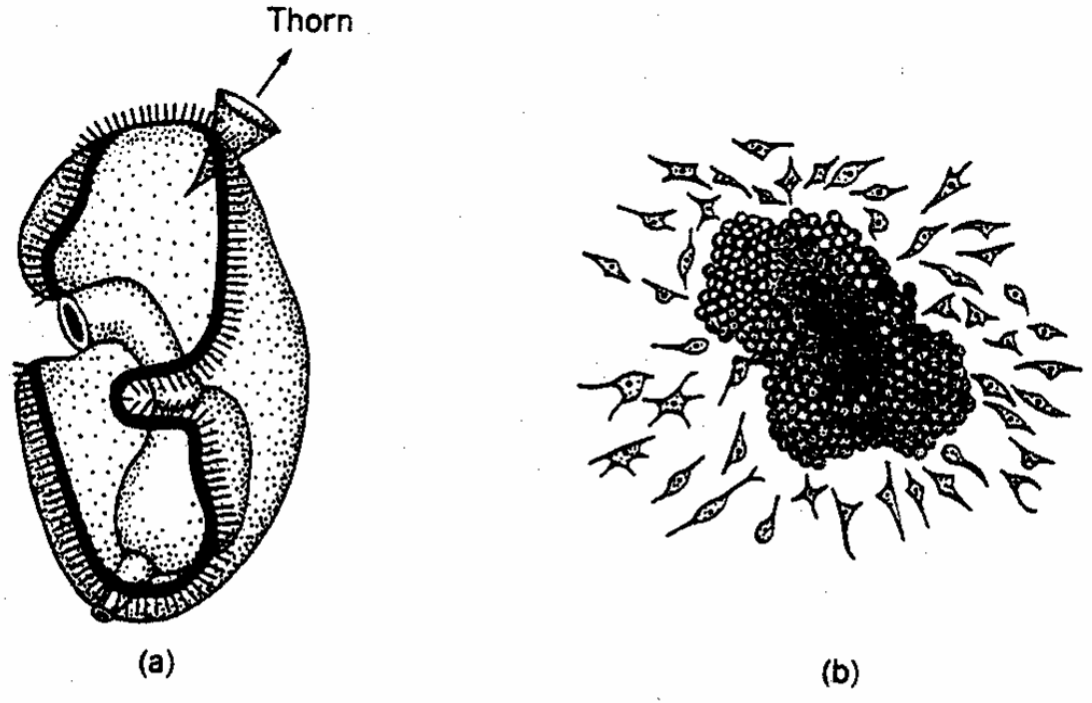
- Measles infection on the Faroe Islands in 1781 protected patients from re-infection in 1846 (Panum, 1847).
- Survivors of 1918 influenza pandemic have Ag-specific Ab titers to HA protein in 2008 (Yu, 2008).

- Persistent protective Ab is found in people vaccinated against yellow fever (75 years), smallpox (50 years), and polio (40 years) (Cooney, 1991; Crotty, 2003; Paul, 1951).

- Longitudinal analysis of Ag specific Ab titers in humans calculated $t_{1/2}$ of those Abs against measles to be 3014 yrs (Amanna, 2007).

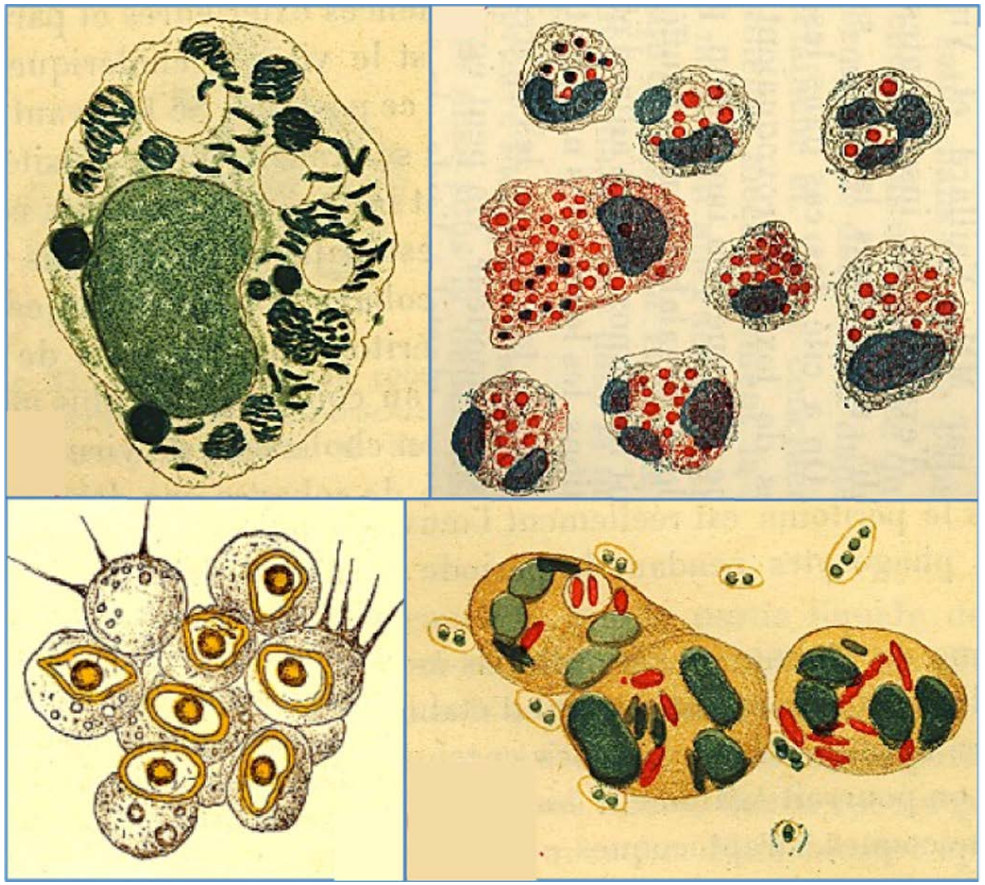
Elie Metchnikoff
Contribution to Cellular Immunity





In a famous experiment, the Russian immunologist Elie Metchnikoff stuck a splinter into a starfish larva *(a)*. The next day the foreign body was surrounded by macrophages *(b)*. Metchnikoff concluded that the body defends itself against foreign particles that threaten its integrity by mobilizing cells of a special type, which attempt to eliminate the foreign matter.

Elie Metchnikoff
Father of Innate & Cellular Immunity



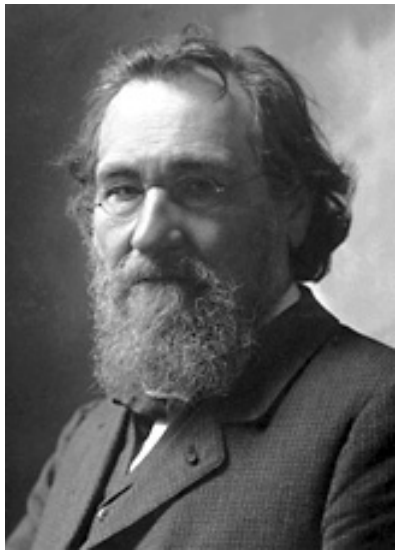
Metchnikoff, E (1901) L'immunite dans les maladies infectieuses. Paris, Mason.
Cavaillon, J-M, J Leuk Biol 90:413, 2011

Elie Metchnikoff - Host Cells Responsible for Immunity (*1893*)

- 1st evidence that cells respond to foreign antigens
- Unable to demonstrate *specificity*
- Revealed basic tenet of inflammation & protective function of recruited leukocytes
- Not until 1940s-1950s that cellular immunology becomes in vogue

1908 – Nobel Prize

In recognition of their work on immunity



Paul Ehrlich and
Elie Metchnikoff jointly awarded Nobel Prize for contributions to immunology

Basis of immunological research for next century

1898

- Roswell Park founded 1st institute dedicated to cancer research

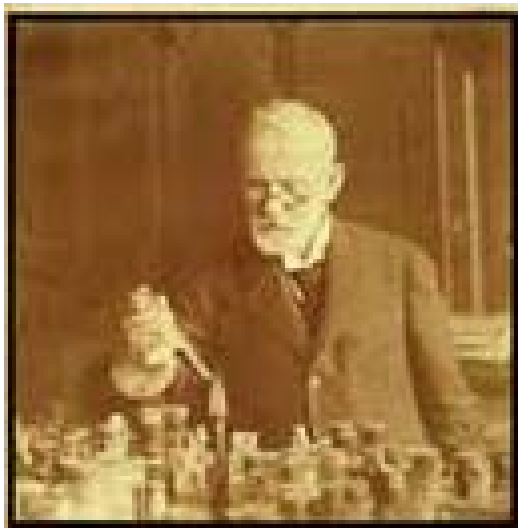
1904

- First scientific observations implicating immunological reactions to malignancy (Gaylord, Clowes, Baeslack)
- Dr. G.H.A. Clowes, driven by the fact that his son had leukemia, initiates the first cancer chemotherapy program in the United States

From Ehrlich to Burnet

1900s

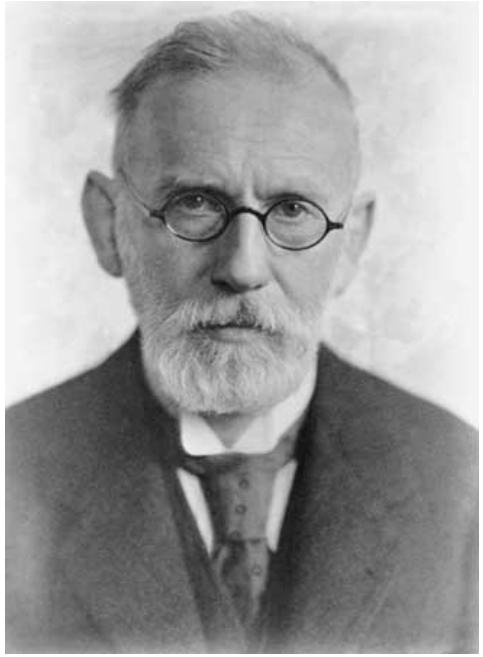
1950s



Paul Ehrlich

MacFarlane Burnet

Awarded Nobel Prize for Contributions to Immunology



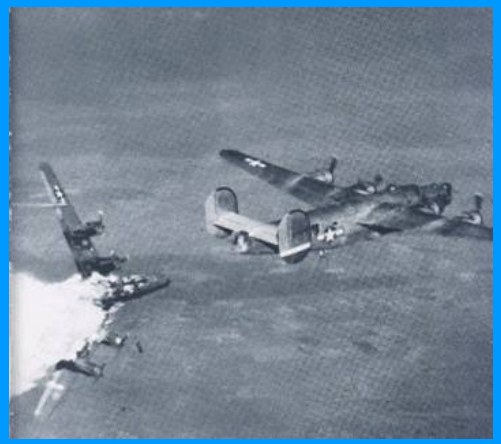
- Paul Ehrlich – 1908

  100th year anniversary

- Frank MacFarlane Burnet – 1960

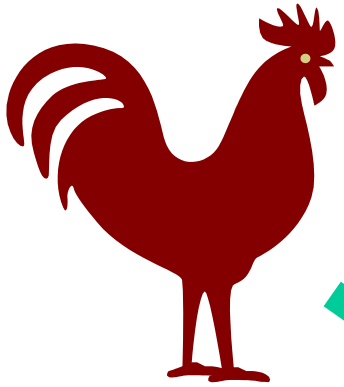
  50th anniversary of clonal selection theory (1957)

Unifying theme – specificity of immune response



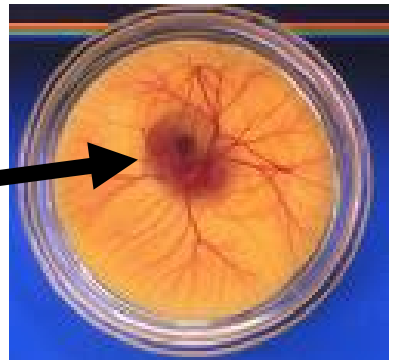
Research in Transplantation/Graft Rejection Mechanisms Advanced by WWII

Macfarlane Burnet and Peter Medawar



Inoculate $2x10^6$ leukocytes

Opaque foci: cellular proliferation, immune response



Chick chorioallantoic membrane

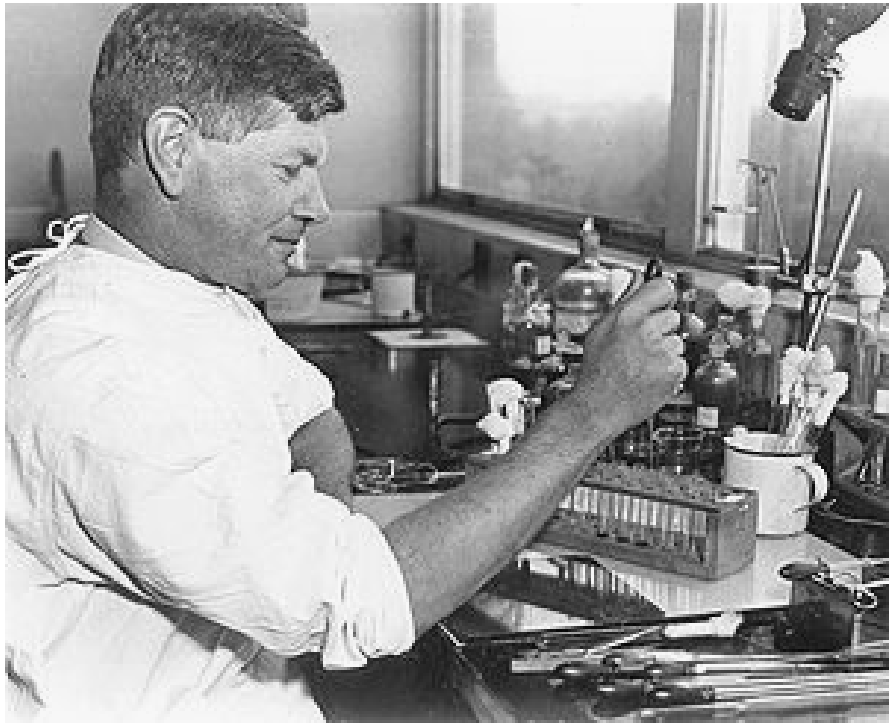
Conclusions

- Mediated by cellular arm (T cells)
- Self vs. non-self recognition (MHC I-dependent)

Nobel Prize (1960)
Immunological Recognition of Self

1960 - Macfarlane Burnet (center) and Peter Medawar (second from right) were awarded the Nobel Prize for the discovery of immunological tolerance.

Abiding Passion-Scientific Work



- Found benchwork an excellent ‘occupational therapy’ that allowed his mind to wander and wonder while his hands were occupied with pipettes and eggs
- Unexpected results would not be dismissed as technical mistakes
- Rarely used statistics; biologically important data should be obvious

Abiding Passion-Scientific Work

- Worked alone
- One or sometimes two graduate students
- Careful in selection of graduate students
- Succession of highly competent and devoted women

First Draft of Clonal Selection Theory

1st draft of clonal selection theory of antibody production
[6.9.57]

Suppose we set out to produce a theory type theory from first principles.

A. The body must produce enough types of globulin to correspond to all potential antigenic determinants in and out of the body. During the functional phase all those which react with body components must be removed to account for tolerance findings. This fits with our present theory.

B.i Either the capacity to produce the removed type of globulin is eliminated i.e. the clone of that pattern dies in descendants -

B.ii The corresponding globulin molecules of an living produced but circumstances are such that non contact with antigenic determinant does not result in antigenic reaction in contrast to the behaviour of a foreign determinant.

C. There is no escape from the requirement that antibody is specifically produced by cells which normally & immediately genetically determined. One cannot assume for instance that a given antibody or globulin molecule has perhaps 100 exact sites on its surface corresponding to 50 antigenic determinants and averaging two of each. On second thoughts it is hard to exclude the possibility of one producing in addition that there are 1000 different molecular patterns each of 50 ± ag. det.

D If we postulate as there is some basis for, that the lymphocyte has a surface capable of reacting with appropriate antigen and that after reaction the lymphocyte has special ability to multiply into an appropriate number and become a parent, we have a powerful mechanism for known effects.

E In the first the lymphocyte accepts this capacity to proliferate only when it is ± 11 days post hatching

F. The most therefore postulate a period during which the lymphocytes are not being selected to tolerate self components, or non simply they switch off & stay dormant.

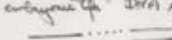
1st draft 6.9.57.

2.

F. The next problem in line is the early stage of development a randomization mechanism involves the appearance of a wider range of globulin patterns sufficiently wide to cover all determinants.

Suppose for instance there is some aspect of protein synthesis by which a segment or set of segments can be fitted in at random - or the same thing for RNA or even the DNA blueprint.

In the ancestors of normal gamma cells for a certain period in ontogeny the DNA replication is of the nature

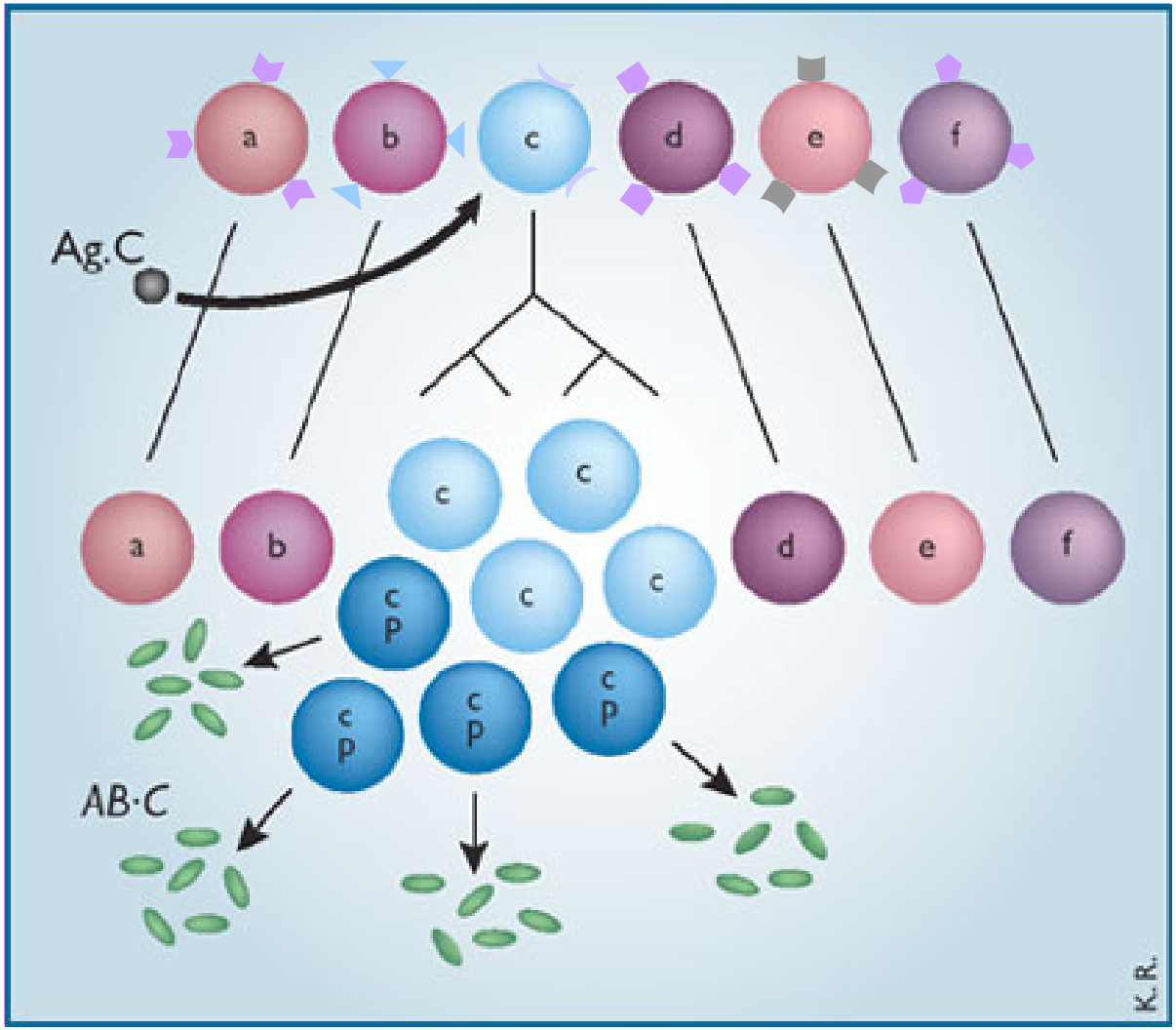


where — are defined codes ..... is randomized for the production of globulins.

At a later stage  stabilization of the random segment.

The same process may occur as a result of somatic mutation in later life giving A1C1* segment.

Macfarlane Burnet – 1957
Clonal Selection Theory



- Antigen selects antibody-forming cell by binding to surface receptors
- Proliferation of the selected clone & release of soluble antibody
- Early exposure to antigen (at birth) leads to tolerance

Theory of Immune Surveillance in Tumor Immunology

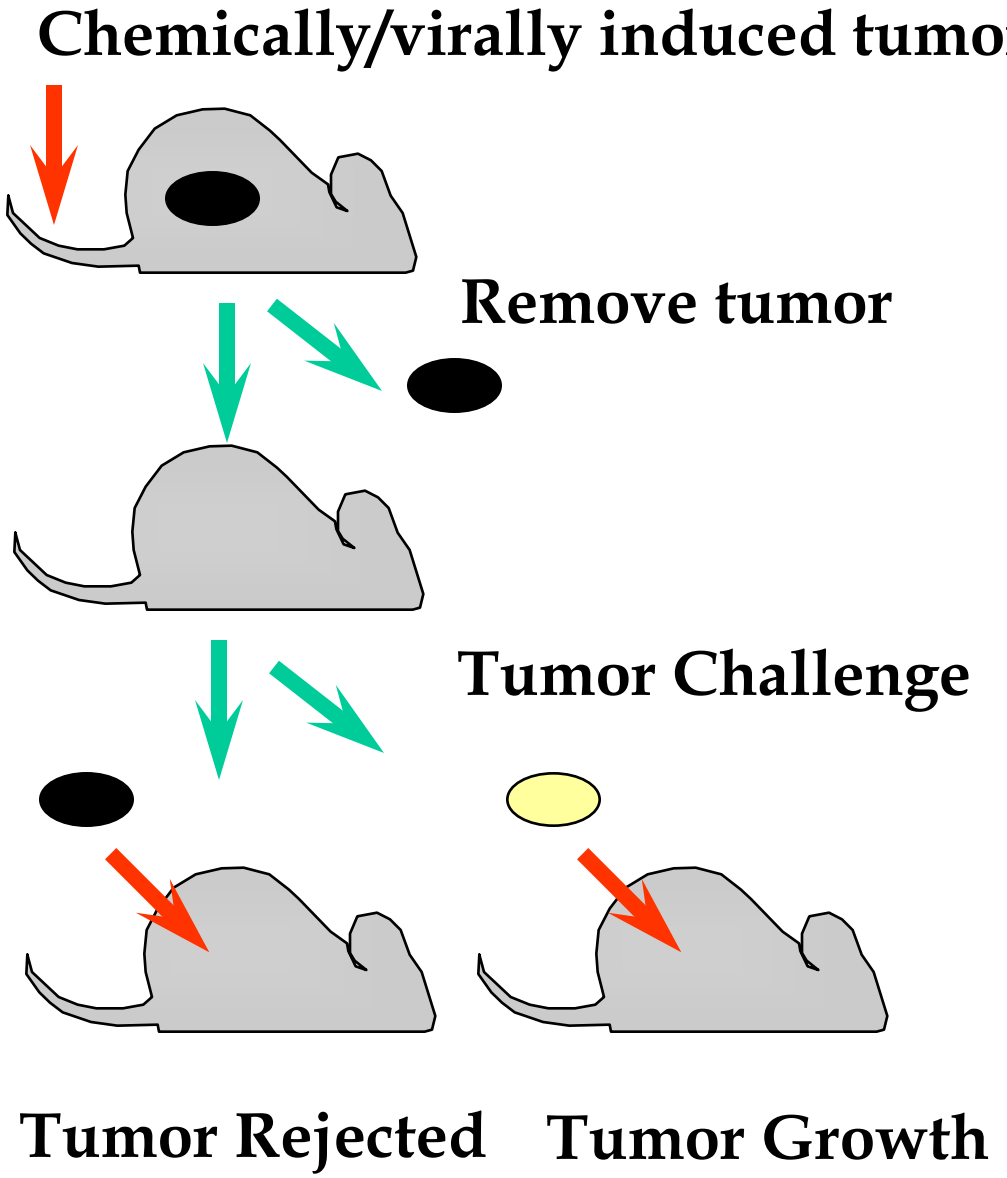
Macfarlane Burnet

- Immune system recognizes tumor Ag as “foreign” and rejects emerging cancer cells continuously.

- Cancer develops if imbalance between host immune response and tumor environment.

Br Med J, 1:841, 1957

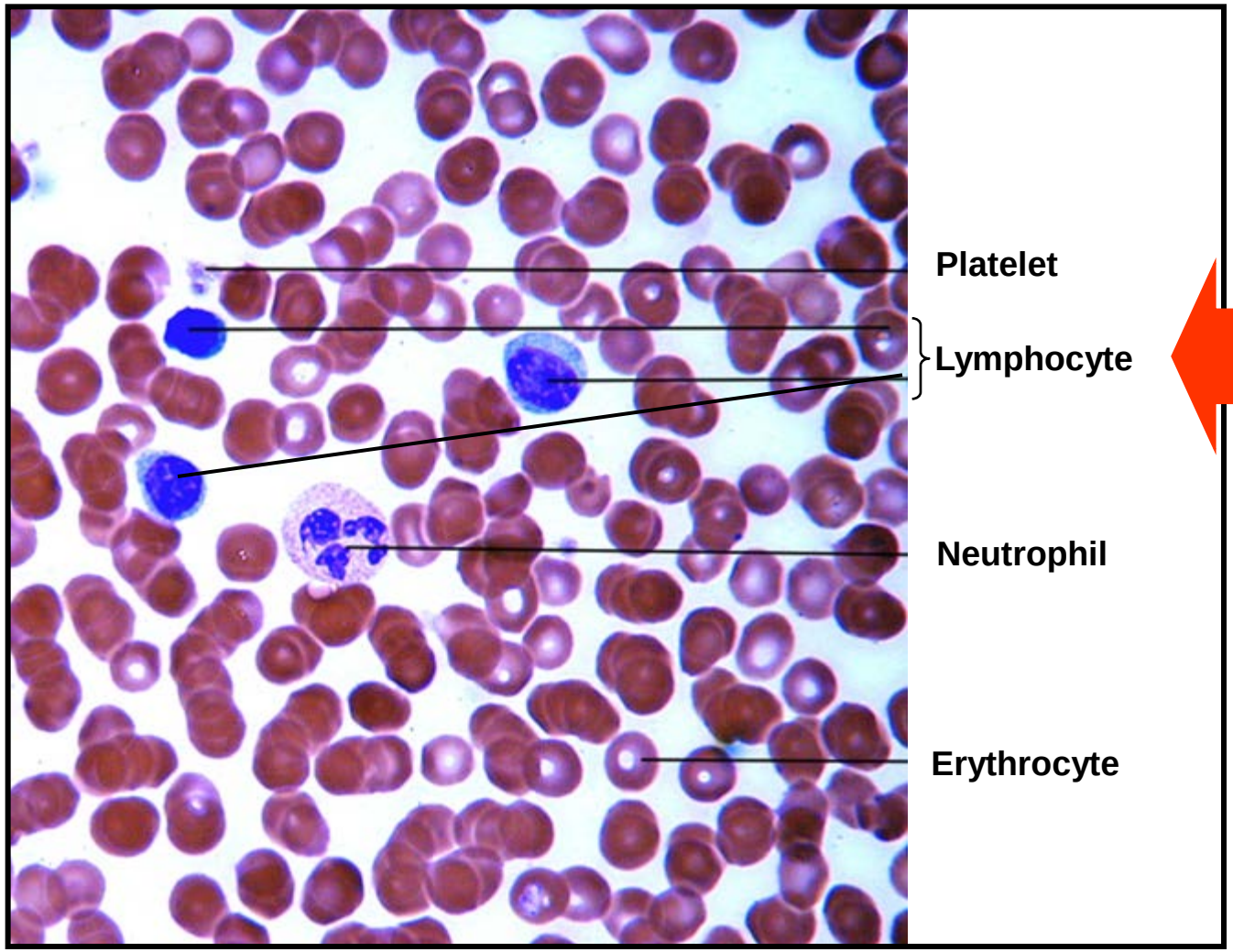
Principles of Tumor Immunity *(Gross, 1943)*



Conclusions

- Evidence for tumor rejection antigen
- Specificity of anti-tumor immune response
- Immunologic memory
- Cell mediated response (Subsequently showed T cell dependent; Ab fail to transfer tumor immunity)

Human Blood (1,000x)



?

Are antibody production & cell-mediated immunity performed by same cell?

*Paul Ehrlich developed staining techniques; identified leukocyte subtypes in blood – basis of hemotology

Humoral Immunity

1956 - Glick

Identification of "B cells" as source of Ab

- **Surgically remove Bursa of Fabricius in chickens**
- **Assistant mistakenly used to demonstrate Ab response**
- **Unable to make Ab, still reject skin graft**

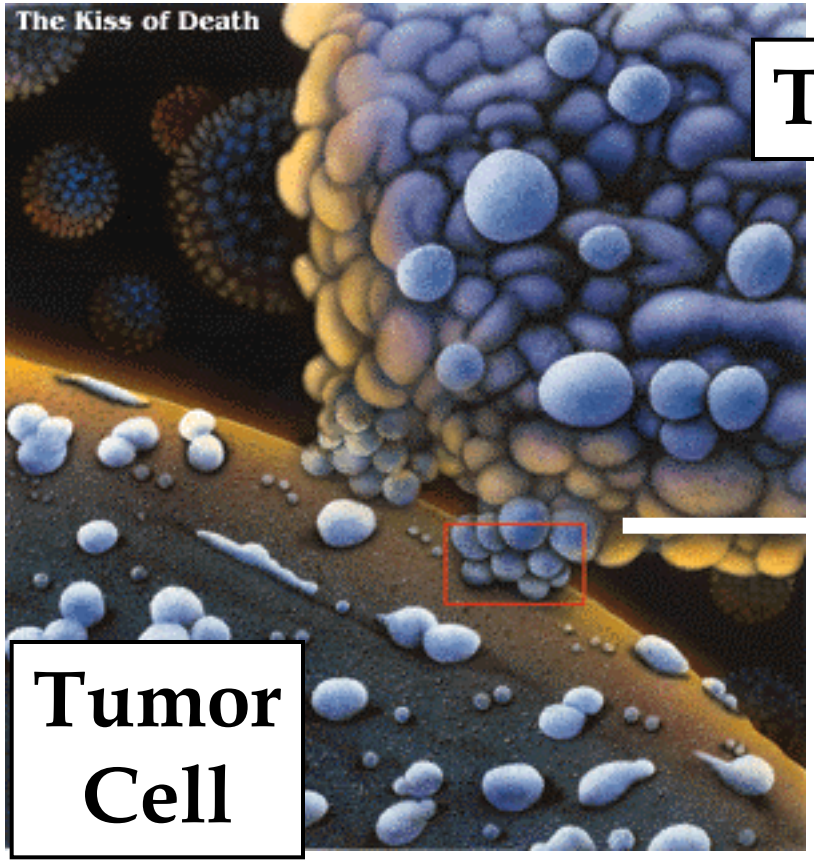
Cellular Immunity

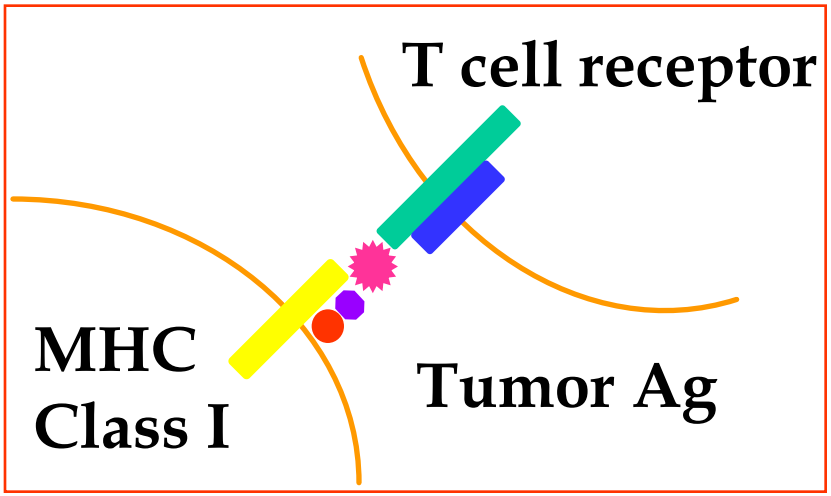
1961 - Miller and Good

Identification of "T cells" as mediator of self/non-self recognition

- **Thymectomize animals at birth**
- **Challenge with foreign graft**
- **Increased survival time of graft**

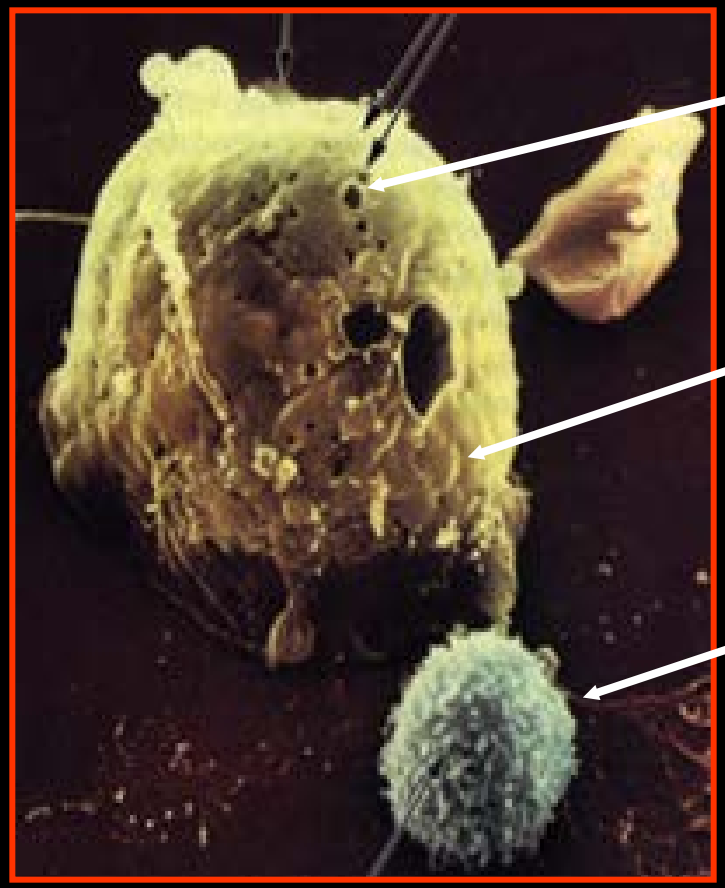
T Cell Mediated Cytotoxicity





- Perforin/cytotoxic granules
- Fas/FasL mediated killing

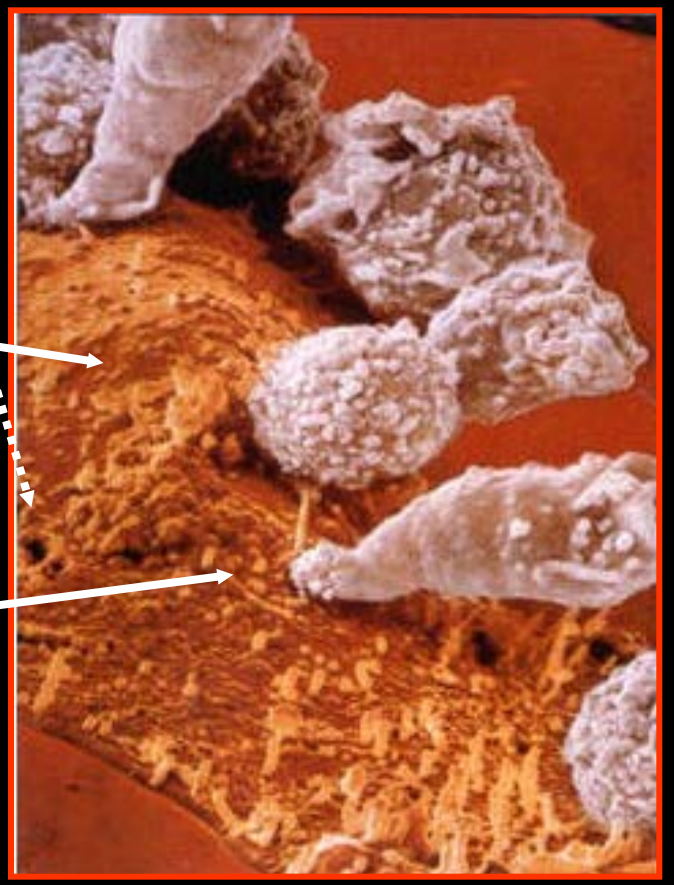
Cytotoxic T Lymphocytes Attacking Cancer Cell



Lethal holes

Cancer cell

Cytotoxic T cell



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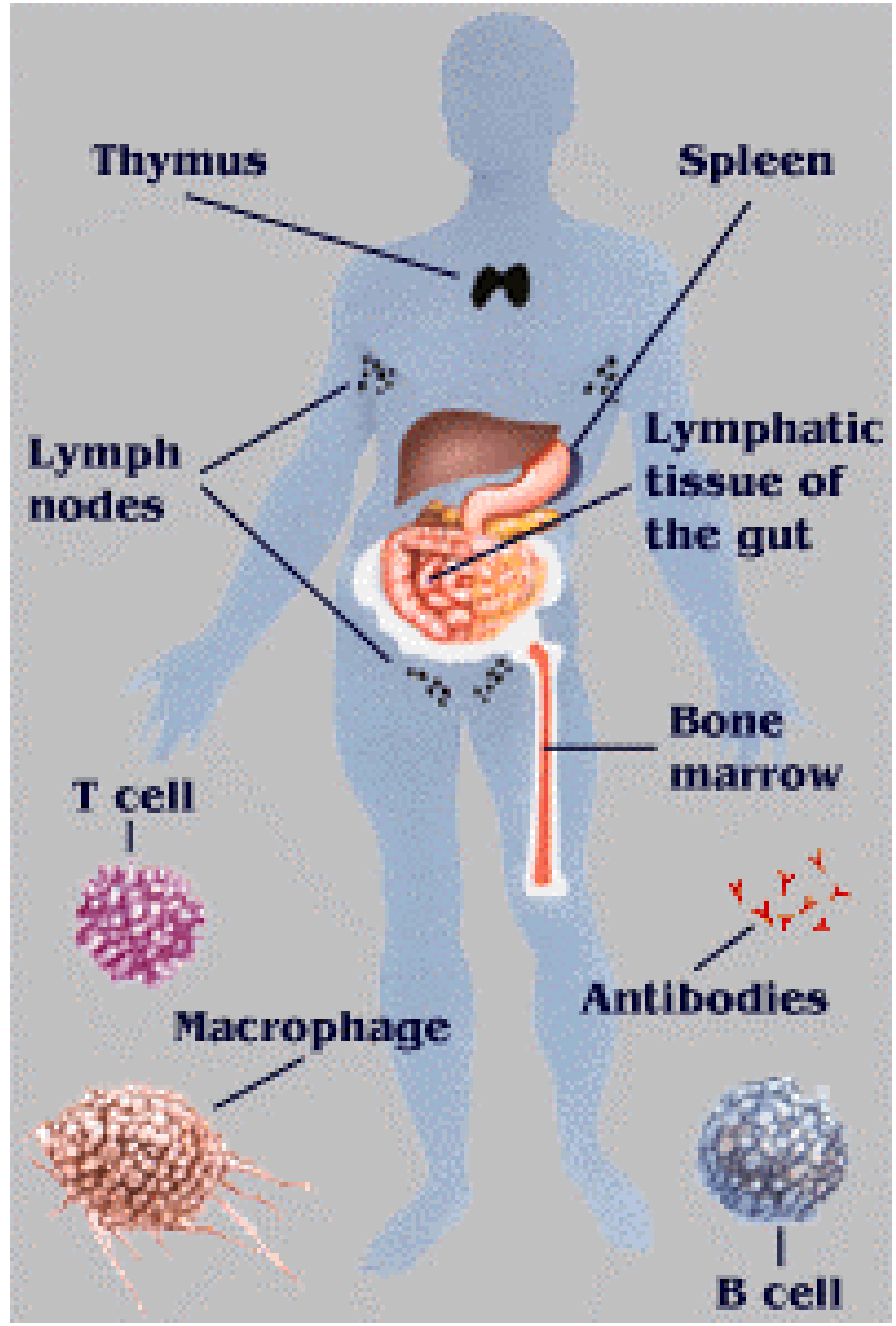
Cellular Mediators of the Adaptive and Passive Immune Response



1980s

- Molecular analysis of B, T cell receptors
- Identification of immunoregulatory cytokines
- Signal transduction pathways underlying B, T activation, cytokine regulation

1990s

- Molecular identification of co-stimulatory molecules, adhesion molecules
- Role of professional antigen presenting cells (APC; dendritic cells) in controlling T cell response

2000s

- Molecular understanding of host - tumor relationship

Nobel Laureates in Immunology

- Cesar Milstein and Georges F. Kohler (1984) *Development of Technique for Monoclonal Antibody Formation*

- Niels K. Jerne (1984) *Theories concerning the specificity in development (lymphocyte clonality) and control of the immune system*

- Peter Doherty and Rolf Zinkernagel (1996) *Discoveries Concerning the Specificity of the Cell Mediated Immune Defense*

> *"It was a wonderful example of how certain things cannot be planned,"* says Zinkernagel. *"Absolutely, this was a miracle of chance."*

Nobel Laureates in Immunology

- R. Yalow (1977)

Development of radioimmunoassays of peptide hormones

- B. Benacerraf, J. Dausset and G.D. Snell (1980)

Discoveries concerning genetically determined structures on the cell surface (major histocompatibility complex) that regulate immunological reactions.

- S. Tonegawa (1987)

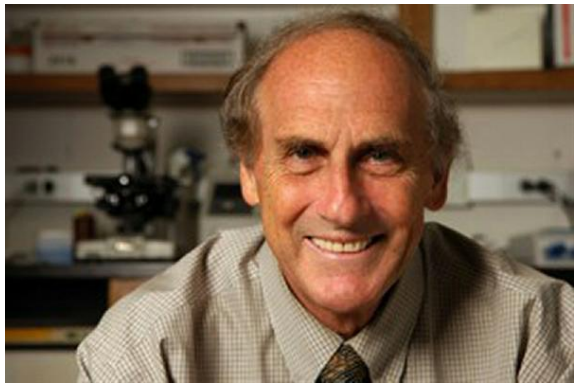
Discovery of the genetic principle for generation of antibody diversity.

- J.E. Murray and E.D. Thomas (1990)

Discovery concerning organ and cell transplantation in the treatment of human diseases; luck and collaborations critical.

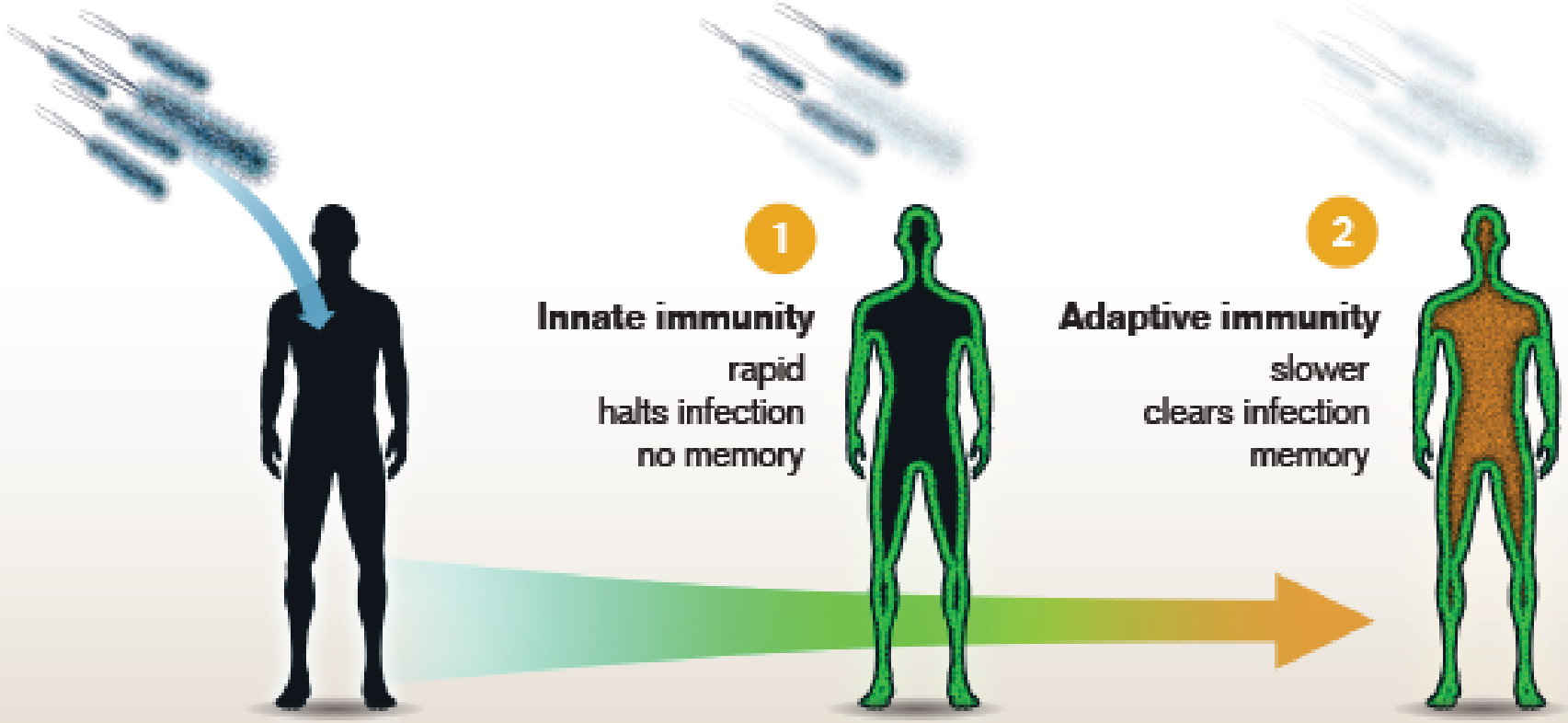
Nobel Laureates in Immunology

- B. Beutler and J. Hoffman (2011)
  Discovered sensors of innate immunity (Toll-like receptors, TLR)



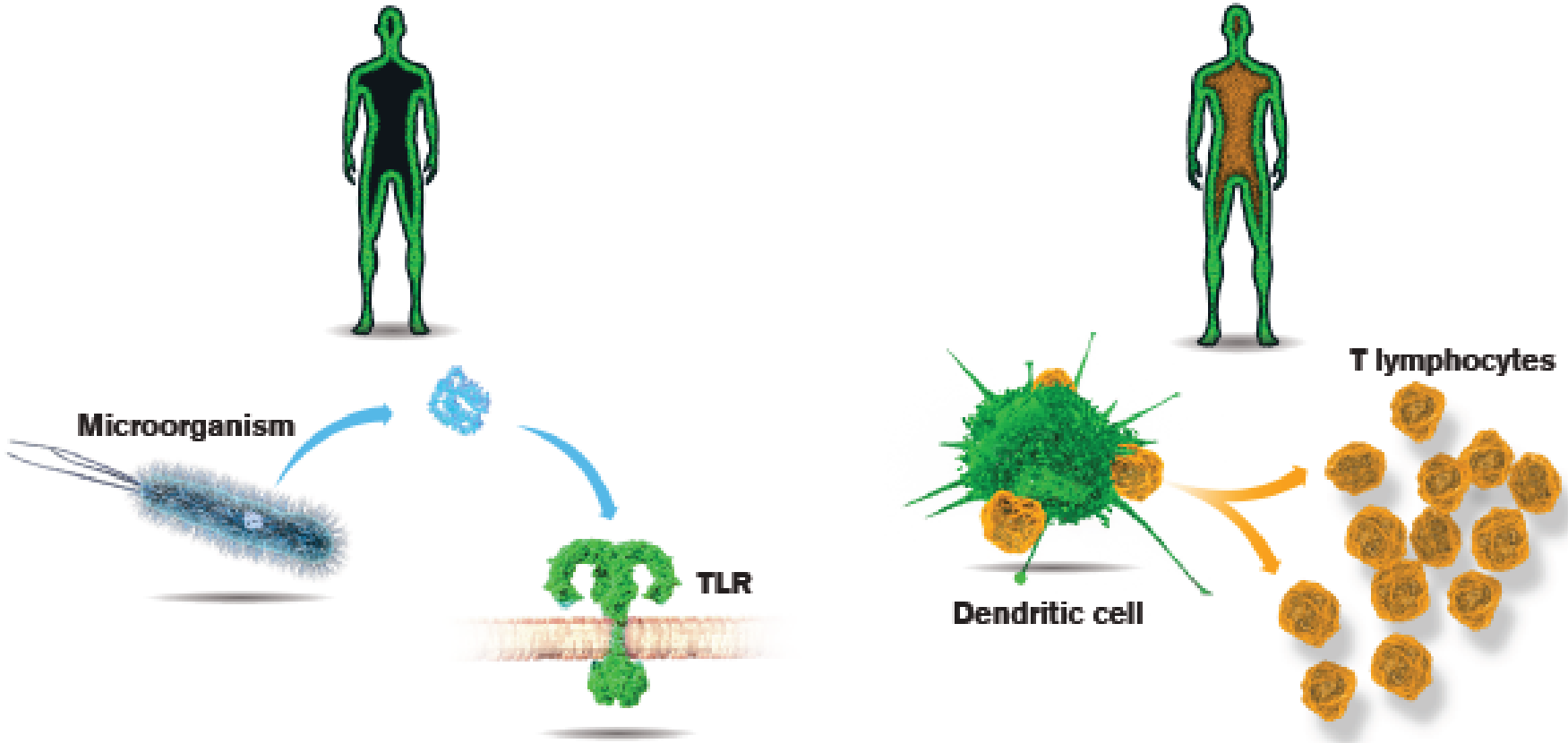
- R. Steinman (2011)
  Discovered new type of cell, dendritic cell, that controls adaptive immunity

The Nobel Prize in Physiology or Medicine 2011



The immune system

Infection of the human body by pathogenic microorganisms such as bacteria, viruses, parasites or fungi triggers the immune response. It occurs in a two-step process: innate immunity halts the infection, and adaptive immunity subsequently clears it.



1 Innate immunity

Components of microorganisms bind to Toll-like receptors located on many cells in the body. This activates innate immunity, which leads to inflammation and to the destruction of invading microorganisms.

2 Adaptive immunity

Dendritic cells activate T lymphocytes, which initiates adaptive immunity. A cascade of immune reactions follows, with formation of antibodies and killer cells.

Illustration: Mattias Karlén