# Properties of Antibodies

There are three major **functions** of antibodies:

1. **Opsonization**
2. **Neutralization**
3. **Complement activation**

## 1. Opsonization

- **Opsonization** refers to the **coating of antigen with antibody** that results in **enhanced uptake of the antigen**.

- There are **two receptors** on phagocytes capable of binding **opsonized antigen:**

  -- One is the **FcγR**,

  -- The other is the receptor for one of the complement components **C3b.**

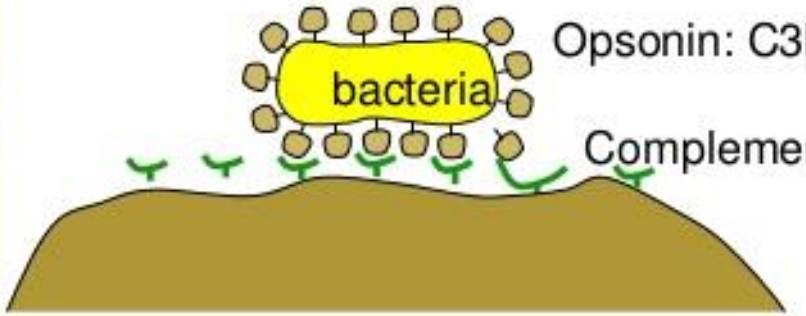
- If an antigen is **coated with IgM antibody** which has **C3b** bound to it then it will be **opsonized via the C3b receptor.**

- If an antigen is **coated with IgG antibody** it will be **opsonized via FcγR** on phagocytes.

- **Antigen-antibody complex** attachment on **FcγR** on phagocytes leads to their activation and the **enhanced uptake and degradation of antigen.**

# Opsonin Receptors

1. Complement receptors (CRs)

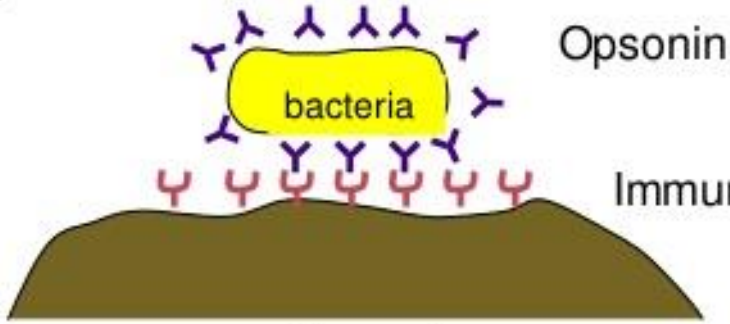



Opsonin: C3b or its cleavage products (i C3b

Complement receptor (CR: CR1, CR3, CR4

Macrophage

2. Immunoglobulin Fc receptors




Opsonin : antibody

Immunoglobulin $F_C$ receptor

Macropha ge

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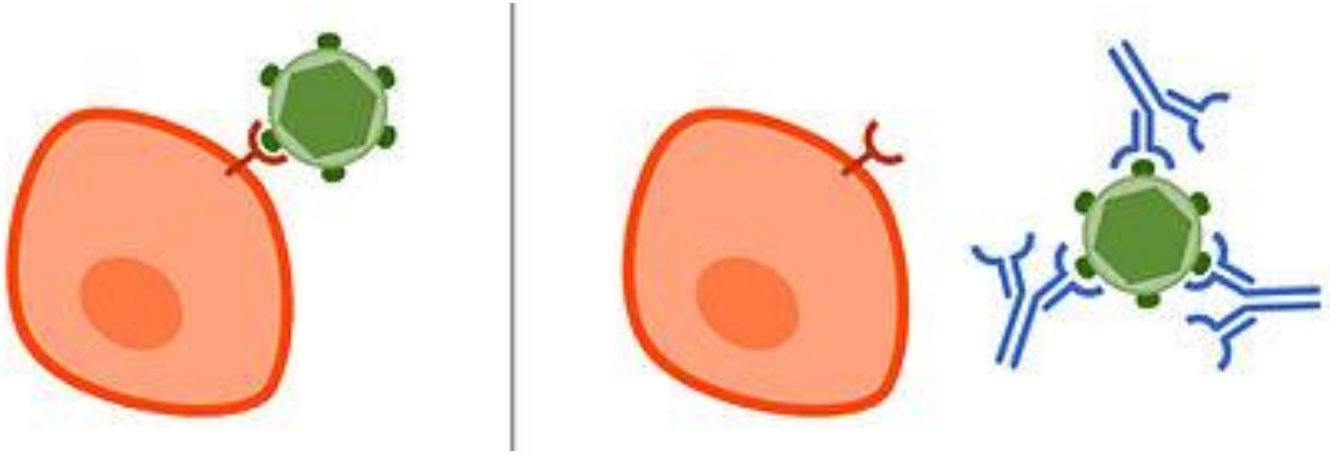
## 2. Neutralization

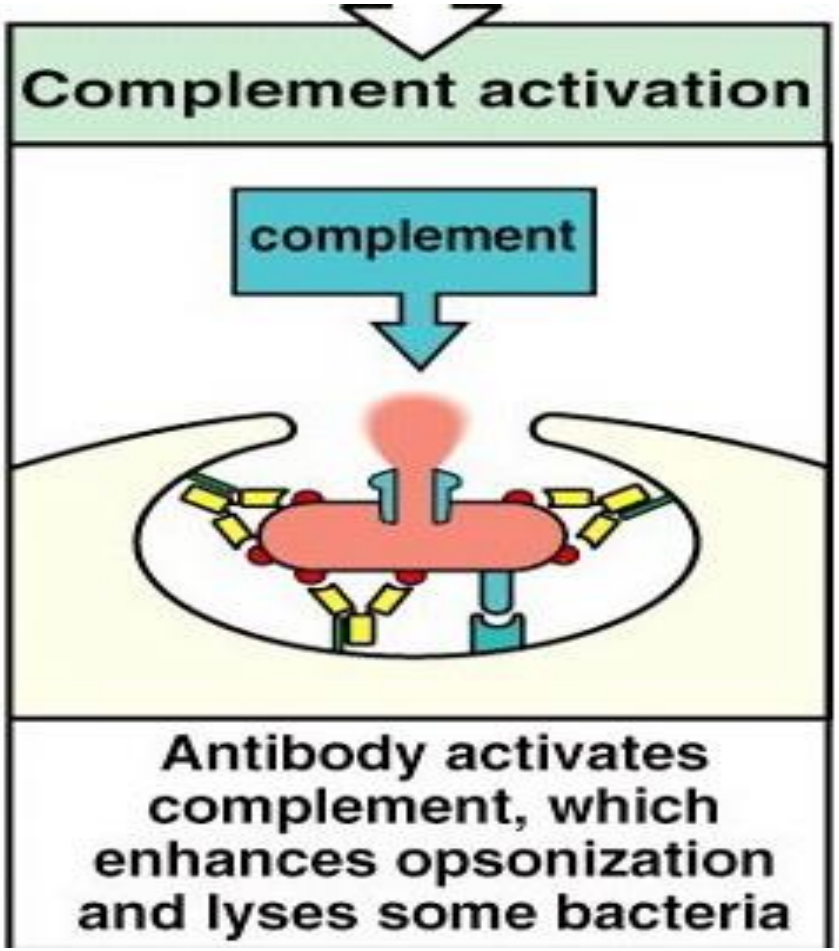
**Prevents adherence of:**

- Bacteria
- Virus
- Toxins



## 3. Complement activation

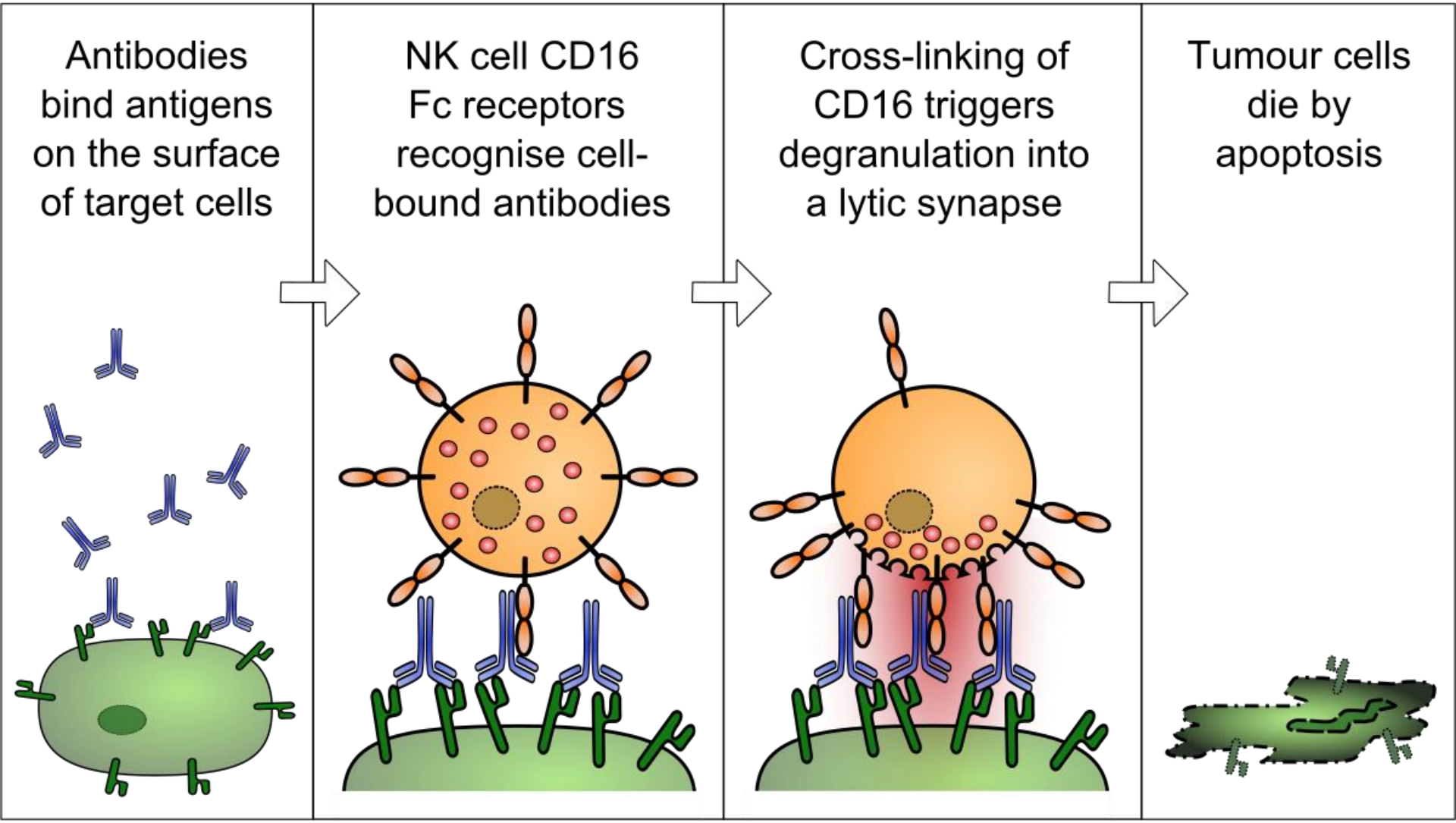
- Facilitates immunity
- Enhances uptake of antigens
- Kills susceptible bacteria



# Antibody Dependent Cellular Cytotoxicity (ADCC)

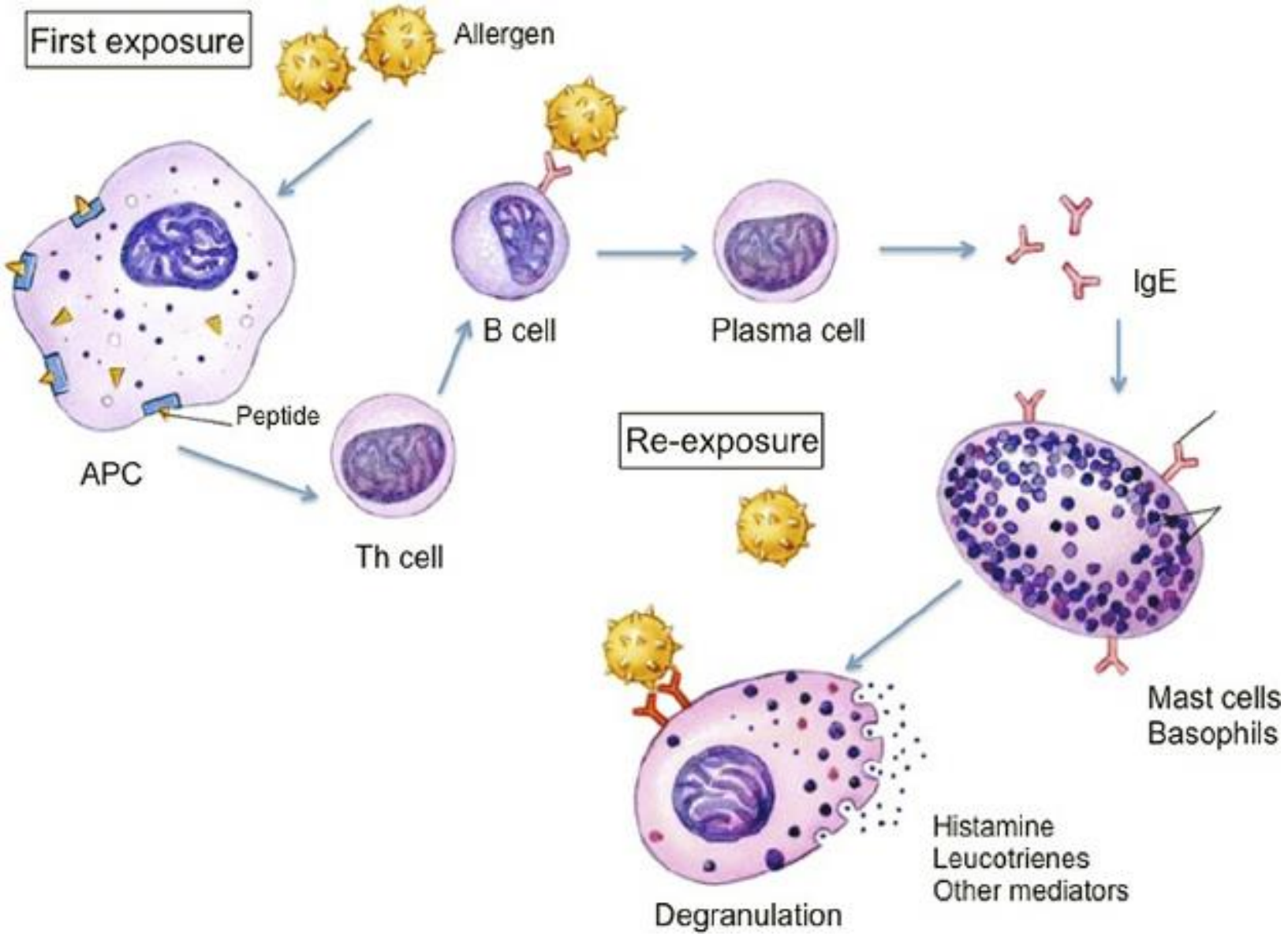
- **Phagocytes (esp. neutrophils), eosinophils,** and **NK cells** all mediate Antibody Dependent Cellular Cytotoxicity (**ADCC**).
- Attachment of **antigen-antibody complex** to **FcγRIII (CD16)** molecule attract the above-mentioned cells, all of which have granules.
- These cells **attach** to target cell and **transfer their granules** to activate the lytic machinery of the cell.
- ADCC is only triggered by **complexes of antigen and antibody.**

* CD or Cluster of Differentiation is a nomenclature for immune molecules.


Antibodies bind antigens on the surface of target cells
NK cell CD16 Fc receptors recognise cell-bound antibodies
Cross-linking of CD16 triggers degranulation into a lytic synapse
Tumour cells die by apoptosis

# Immediate (type I) hypersensitivity

- It is caused by **IgE antibodies**.
- It is due to attachment of **antigen-antibody complex** on **FcεR1** receptor on **mast cells** and **basophils**.
- It leads to the release of granules containing **inflammatory** and **vasoactive mediators** (histamine), **lipid derived mediators** (leukotrines, prostaglandins, platelet activating factor etc.) and **cytokines**.
- This phenomenon may result in **allergy** but is clearly of benefit in clearance of **extracellular parasites**.


First exposure
Allergen
B cell
Plasma cell
IgE
Peptide
APC
Th cell
Re-exposure
Mast cells
Basophils
Histamine
Leucotrienes
Other mediators
Degranulation

## Secretory Ig

- Epithelial cells in the **intestine** and other **secretory epithelia** (tear duct, salivary gland, lactating mammary gland etc.) possess a **polymeric Ig receptor (plgR)** which responds only to IgA or IgM (the polymeric antibodies) by recognizing the J chain.

- **plgR** is a trans-membrane protein that in humans is encoded by the *PIGR* gene. It is an **Fc receptor**.

- **Polymeric Ig receptor** mediates secretion (transcytosis) of antibody, predominantly **IgA (also IgM)** into the external secretions.

- In vivo, **sIgA** is the product of **two different cell types**, the **plasma cell** and the **epithelial cell**.

- Secreted IgA forms a very important **defence against infection.**

# Neonatal immunity

- In humans, **maternal IgG can cross the placenta**.
- Babies have a **high level** of maternal IgG at birth.
- They also acquire **IgA (and some IgG)** post-natally from **breast milk**, however these remain in the gut lumen.

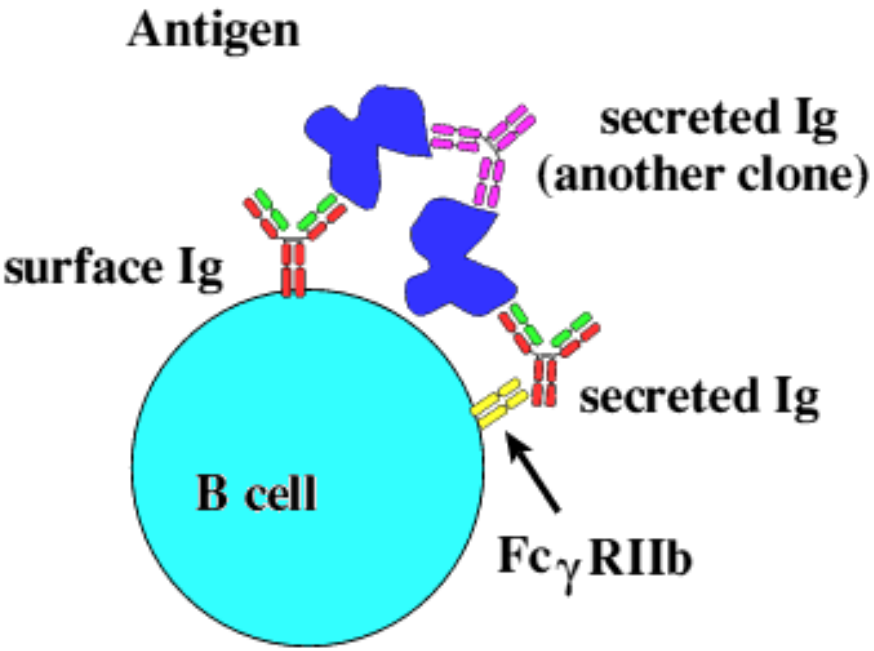
- In some other species, for example, ruminants and rodents, **IgG does not cross the placenta**.

- Instead high levels of IgG are secreted in the milk produced peri-natally (**colostrum**) and IgG is specifically taken up from the gut into blood of the neonate via an **IgG receptor**, whereas IgA remains in the gut.

- In all species **maternal IgG and IgA are important** both locally and systemically in protecting the neonate during the development of it's own immune system.

# Antibody regulation

Co-ligation of the **FcγRIIb receptor (CD 32)** and **surface Ig** on B cells inhibits B cell activation and resulting antibody production thereby acting as a **negative feedback loop**.



# Antibody diversity

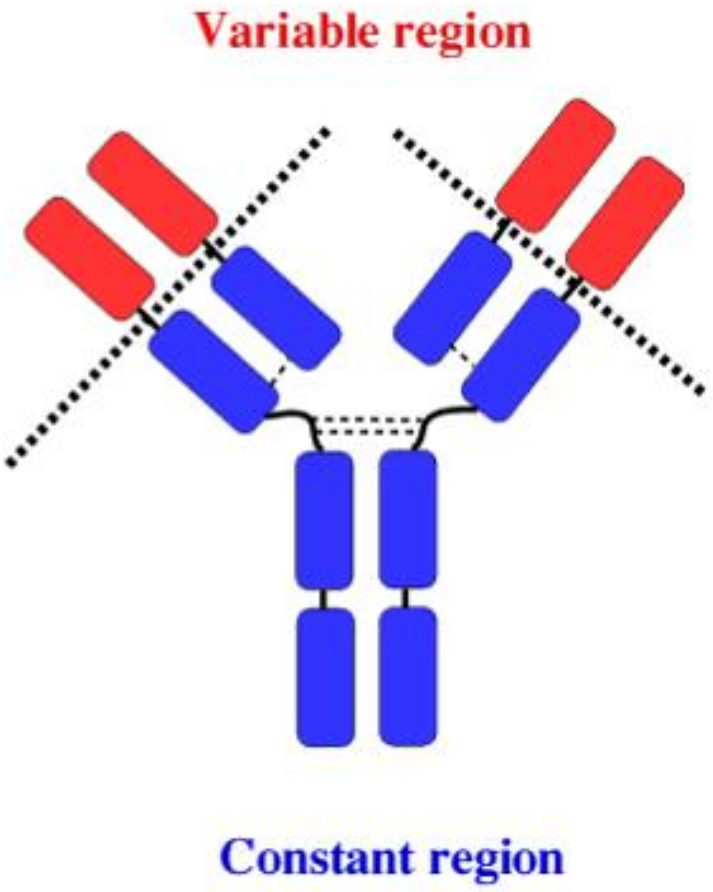
- There are an estimated **$10^9$** different antibody molecules, and thus **$10^9$ distinct B cell clones** in a single individual.

How is this astonishing diversity generated?

- This problem has been resolved recently by the demonstration that **Ig genes rearrange during B cell development.**

- The heavy chain genes have **no complete exon** encoding the variable region domain.

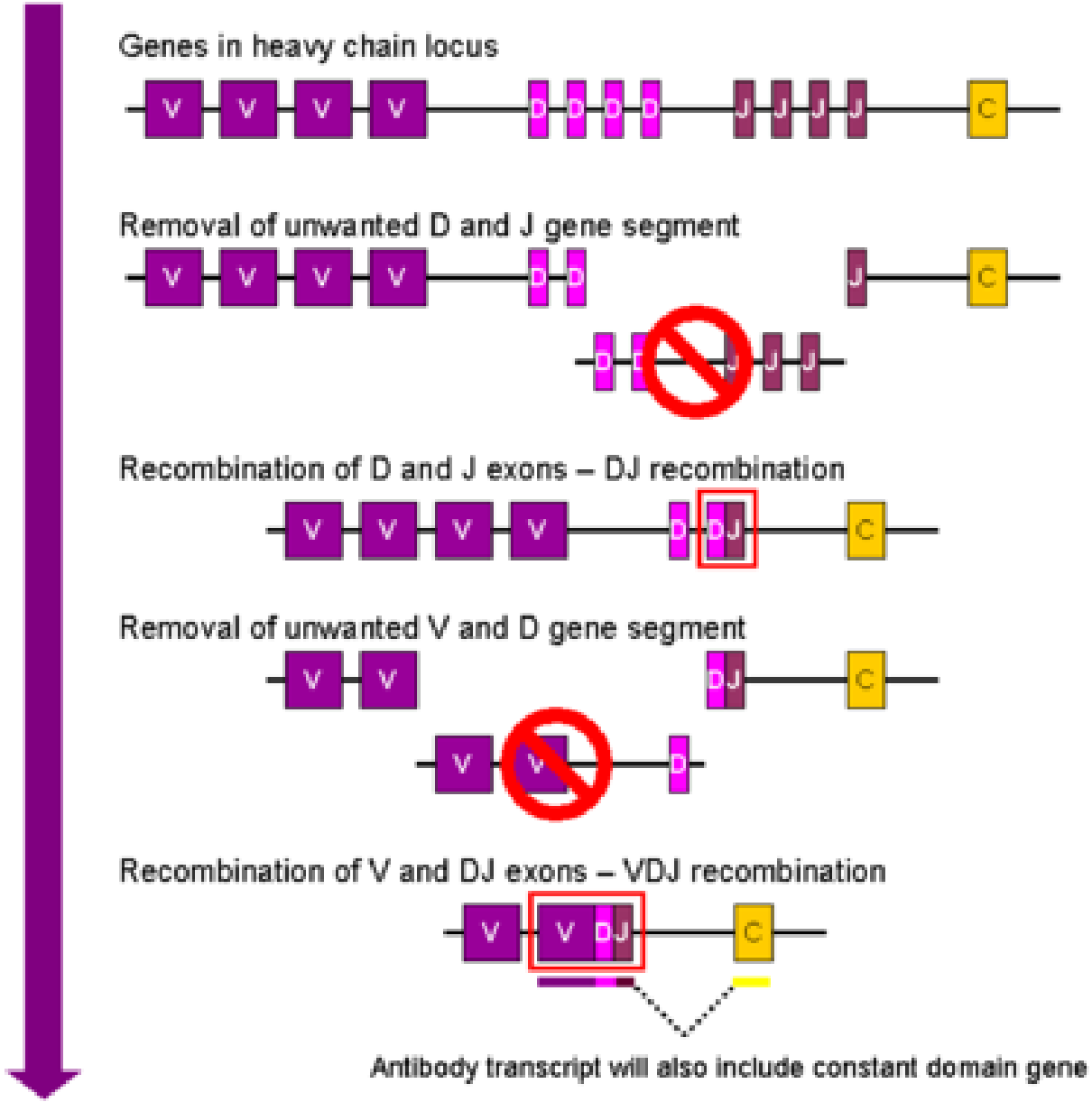
- Instead this is **split into arrays of gene segments on different chromosomes.**



- These are rearranged by a unique process called **V(D)J recombination.**

- The **variable (V), joining (J),** and in some cases, **diversity (D) gene segments** are rearranged in a **nearly random fashion**.

- V(D)J recombination occurs in the **primary lymphoid organs** (bone marrow for B cells and thymus for T cells).


Genes in heavy chain locus
V V V V D D D D J J J J C
Removal of unwanted D and J gene segment
V V V V D D J C
D J J J
Recombination of D and J exons – DJ recombination
V V V V D DJ C
Removal of unwanted V and D gene segment
V V DJ C
V V D
Recombination of V and DJ exons – VDJ recombination
V V DJ C
Antibody transcript will also include constant domain gene

- The process ultimately results in **novel amino acid sequences** in the antigen-binding regions of **immunoglobulins** and **TCRs (T cell receptors)**.

- It allows for the **recognition** of antigens from **nearly all pathogens** including bacteria, viruses, parasites, and worms as well as **"altered self cells"** as seen in cancer.

- This recognition can also be **allergic** in nature (e.g. to pollen or other allergens) or **may match host tissues** and **lead to autoimmunity.**

*TCRs are receptors on T cells.

- Light chain genes are **similarly** organized on different chromosomes but they have **no D (diversity) gene segments.**

- There are **51 functional Vl** genes and **41 Vk genes**.

- **D and J (junctional)** genes code for **amino acids at the carboxyl end of V regions** including **CDR3**.

- It was first noted that the organization of Ig genes in mature B cells was **different** from that in embryonic tissues.

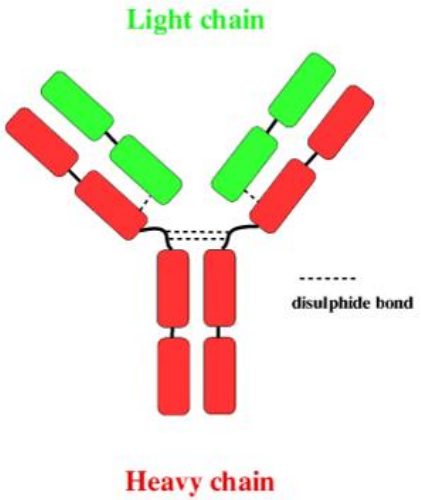
- The pieces of genomic DNA containing antibody genes in embryonic tissue, or immature B cells, are **larger** that those from mature B cells.

- This implies that the DNA in mature B cells has been **rearranged** resulting in the **excision** of some DNA.

# Class switching

- **Class switching** (or **isotype switching** or **isotypic commutation** or **class switch recombination**) is a biological mechanism that **changes an antibody from one class to another**, for example from IgM to IgG.

- During this process, the **constant region portion of the antibody heavy chain is changed**, but the **variable region of the heavy chain stays the same.**



- Since the variable region does not change, class switching **does not affect antigen specificity**.

- The antibody retains **affinity for the same antigens**, but can **interact with different effector molecules** e.g., on cell surface.

* An effector molecule is usually a small molecule that selectively binds to a protein and regulates its biological activity. In this manner, effector molecules act as ligands that can increase or decrease enzyme activity, gene expression, or cell signaling.

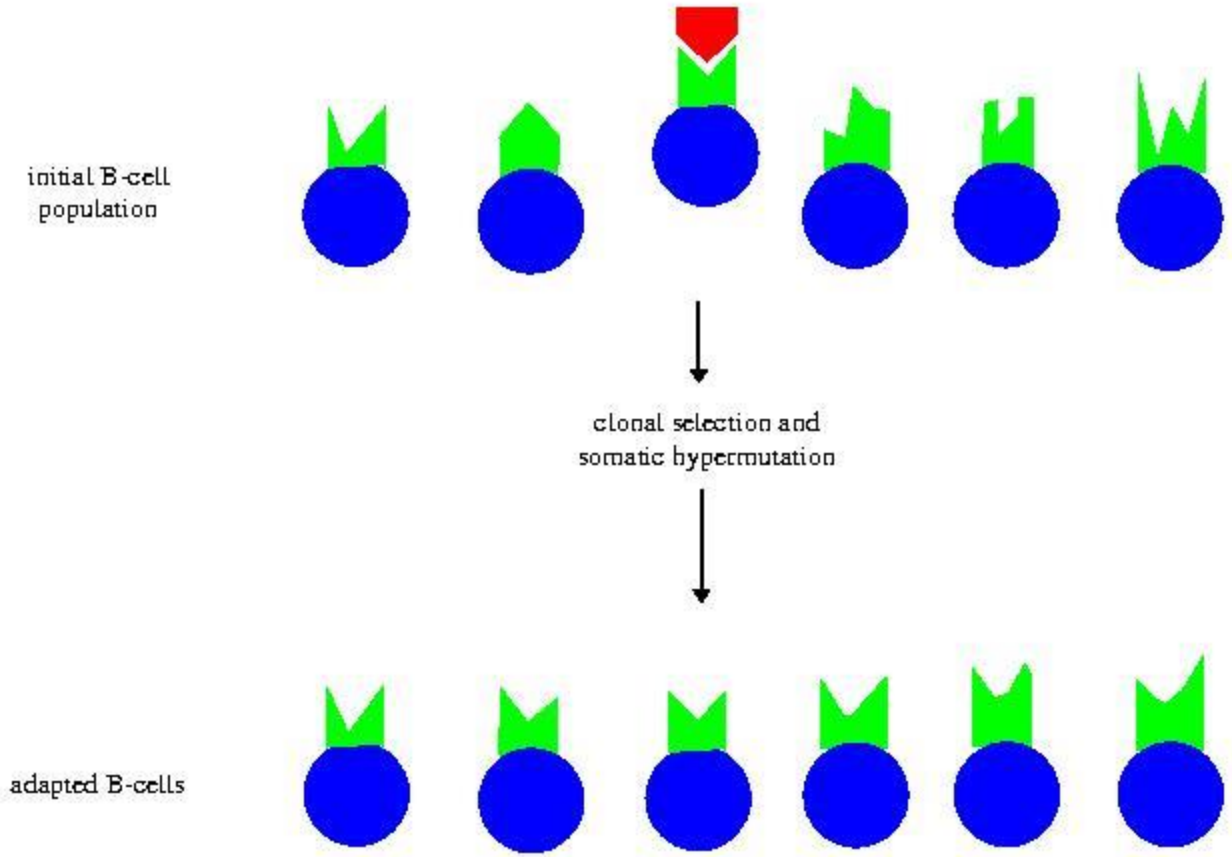
# Affinity maturation

- Affinity maturation is a consequence of **somatic mutation in Ig gene segments.**

- There is a special mechanism for inducing mutations in **rearranged V regions (VDJ).**

- Cells expressing mutations which **improve antigen binding** are **favored** in terms of **activation** and **co-operation with T cells**.

## AFFINITY MATURATION II



- Thus during an immune response (particularly as the free concentration of antigen falls), the average affinity of antibodies produced increases as **higher affinity clones** are selected.

- The rate of mutation is **1 per 1000 for each V gene base pair** per cell division compared to **1 per 1,000,000 for each base pair** per cell division for **normal DNA**.

# T dependent and T independent response

- **Protein antigens** do not induce antibody responses in the absence of T lymphocytes. So they are called **T dependent**.

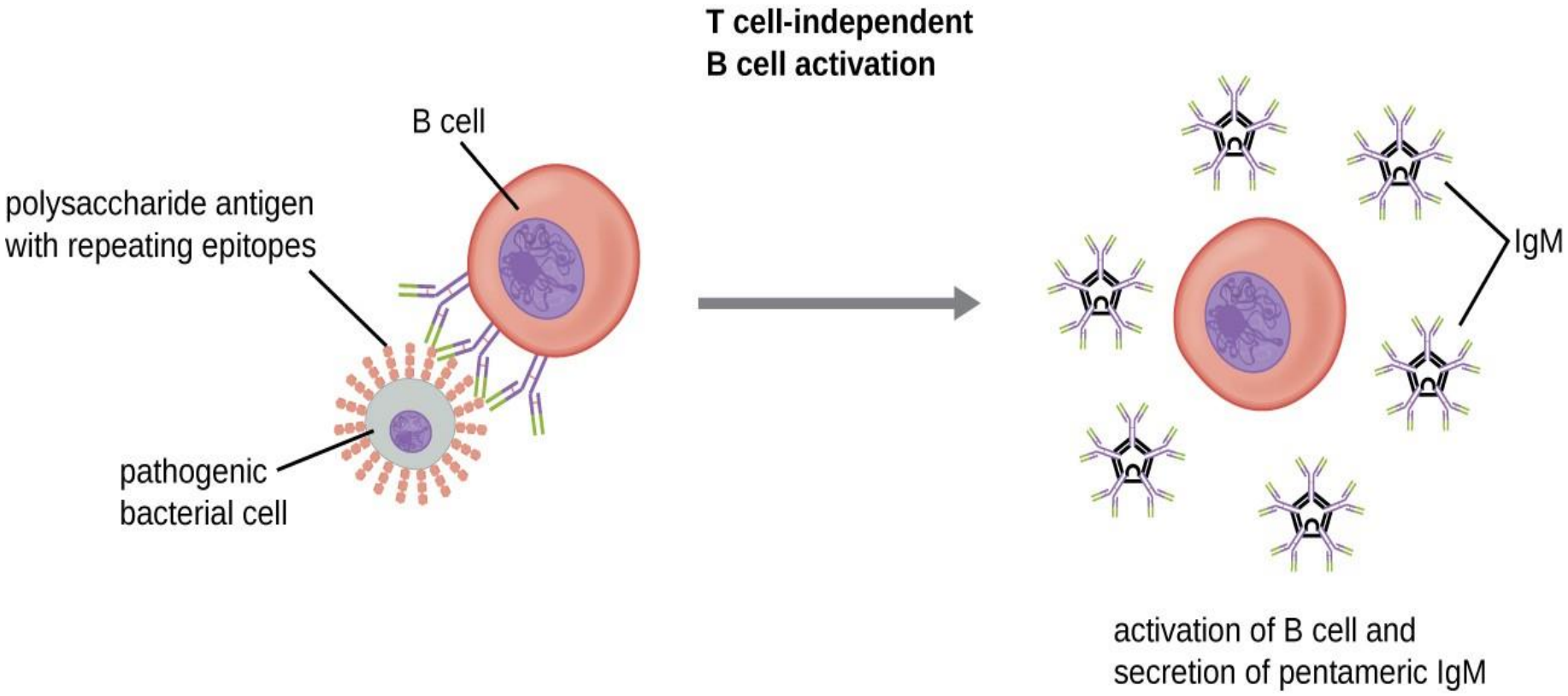
- **Non-protein antigens**, polysaccharides and lipids for example, can lead to antibody responses **without** T cells so they are called **T independent.**

- **T independent antigens** are **usually polymeric** and it is believed that they **cross link** membrane Ig on B cells sufficiently well to activate them without co-operation from T cells.

- The antibodies to these antigens are invariably **IgM** and **do not demonstrate affinity maturation.**

**Polymeric**: Same subunits repeated several times

**Cross linking:** It means that one antigen molecule is attached to both arms of two IgM molecules on the surface of a B cell.


T cell-independent
B cell activation
B cell
polysaccharide antigen
with repeating epitopes
pathogenic
bacterial cell
IgM
activation of B cell and
secretion of pentameric IgM

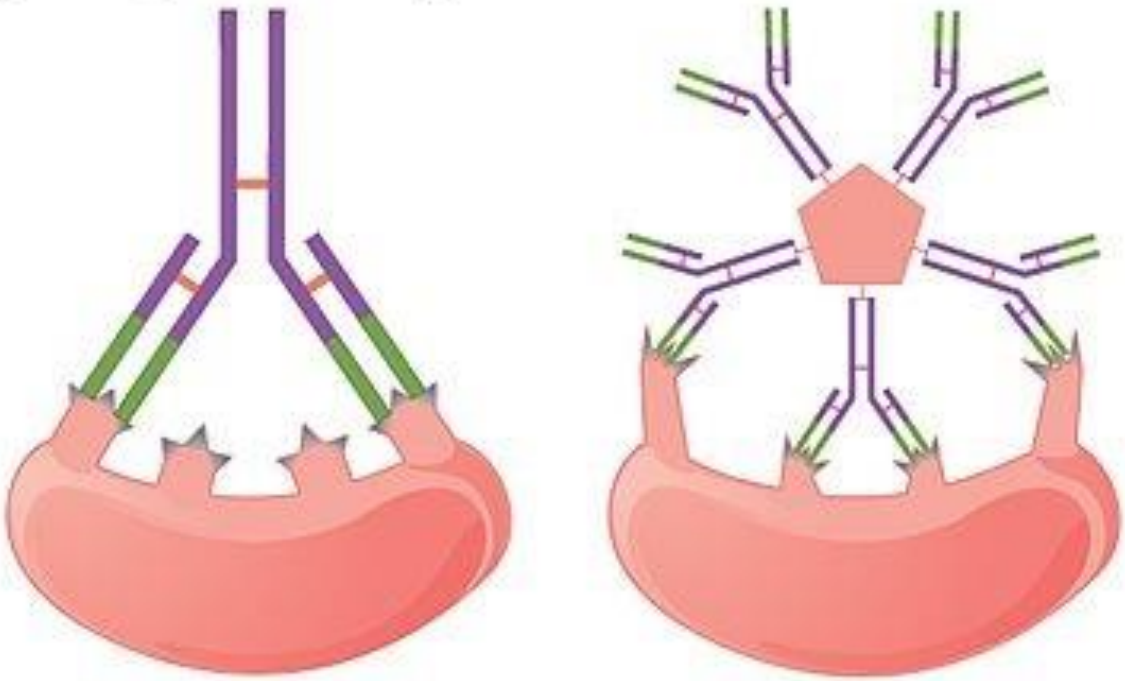
# Affinity and avidity

- **Affinity** of an antibody (or other receptor) is really the **strength of interaction between one antibody combining site and one corresponding epitope on its antigen.**

- **Avidity** is the **overall strength** of antigen-antibody reaction**.**

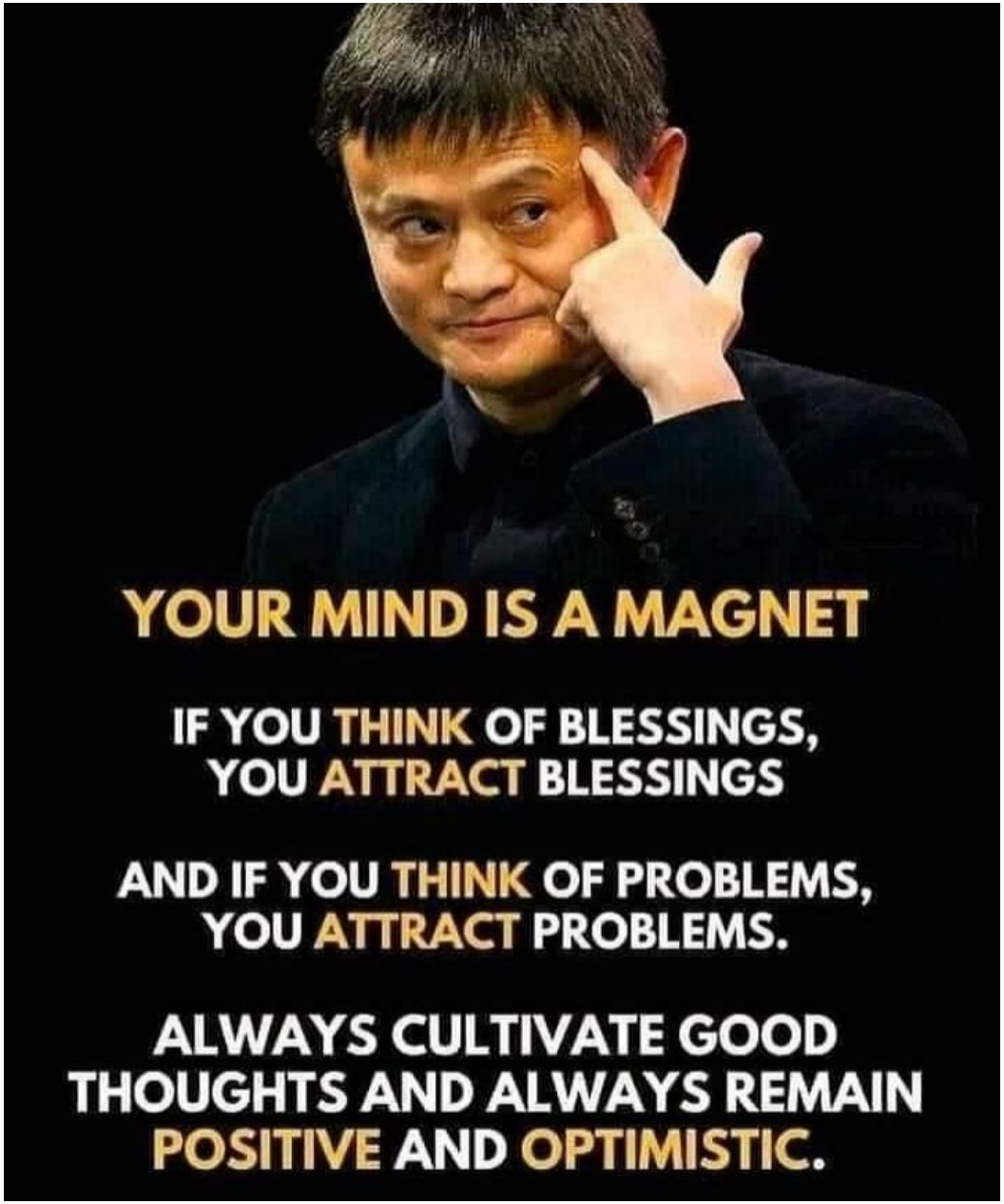
- Antibodies are **polyvalent** and so what is measured in practice is often the **avidity** of the antibody which is much greater than affinity.

**(a) Affinity versus avidity**



Affinity refers to the strength of a single antibody–antigen interaction. Each IgG antigen binding site typically has high affinity for its target.

Avidity refers to the strength of all interactions combined. IgM typically has low affinity antigen binding sites, but there are ten of them, so avidity is high.


YOUR MIND IS A MAGNET
IF YOU THINK OF BLESSINGS,
YOU ATTRACT BLESSINGS
AND IF YOU THINK OF PROBLEMS,
YOU ATTRACT PROBLEMS.
ALWAYS CULTIVATE GOOD
THOUGHTS AND ALWAYS REMAIN
POSITIVE AND OPTIMISTIC.


VianaArts


Thank You!