Course Title : Monoclonal Antibodies (mAbs)

– From Theory to Therapy

1. Definition and Fundamental Principles

Monoclonal Antibodies (mAbs) are laboratory-produced molecules engineered to serve as substitute antibodies that can restore, enhance, or mimic the immune system's attack on cells.

Unlike **polyclonal antibodies**, which are a mixture of antibodies secreted by different B-cell lineages and recognize multiple epitopes (binding sites) on an antigen, **monoclonal antibodies** are:

- **Homogeneous:** Derived from a single clone of a B-cell.
- **Monospecific:** They bind to one, and only one, specific epitope with high affinity.

2. The Production Breakthrough: Hybridoma Technology

The production of mAbs was revolutionized in 1975 by Georges Köhler and César Milstein. The challenge was that B-cells produce specific antibodies but die quickly in a lab culture, while myeloma (cancer) cells are "immortal" but do not produce specific antibodies.

The Step-by-Step Process:

1. **Immunization:** A laboratory animal (usually a mouse) is injected with a specific target antigen.
2. **B-cell Extraction:** Once an immune response is triggered, the animal's spleen is removed to harvest the B-lymphocytes.
3. **Fusion:** These B-cells are fused with **Myeloma cells** using polyethylene glycol (PEG). The resulting hybrid cell is called a **Hybridoma**.
4. **Selection (HAT Medium):** The cells are placed in a special medium where only the hybrid cells can survive. Unfused B-cells die naturally, and unfused myeloma cells are blocked by the medium's chemistry.
5. **Screening and Cloning:** The surviving hybridomas are screened to identify which ones produce the most effective antibody. Once identified, that single cell is cloned to produce an "immortal" line of identical antibodies.

3. The Evolution of Antibody Engineering

To reduce the risk of the human body rejecting mouse antibodies (a reaction known as **HAMA** - Human Anti-Mouse Antibodies), scientists developed four generations of mAbs:

- **Murine (-omab):** Derived entirely from mouse proteins. They have high immunogenicity and a short half-life in humans.
- **Chimeric (-ximab):** A "molecular chimera" where the variable regions (the part that grabs the antigen) are mouse-derived, and the constant regions (the structural part) are human.

- **Humanized (-zumab):** Only the very tips of the antibody (the CDRs) are mouse-derived; the rest (approx. 95%) is human.
- **Fully Human (-umab):** Created using transgenic mice or phage display libraries, these are 100% human proteins and are the best tolerated by patients.

4. Therapeutic Mechanisms of Action

How do these "magic bullets" actually work? There are three primary methods:

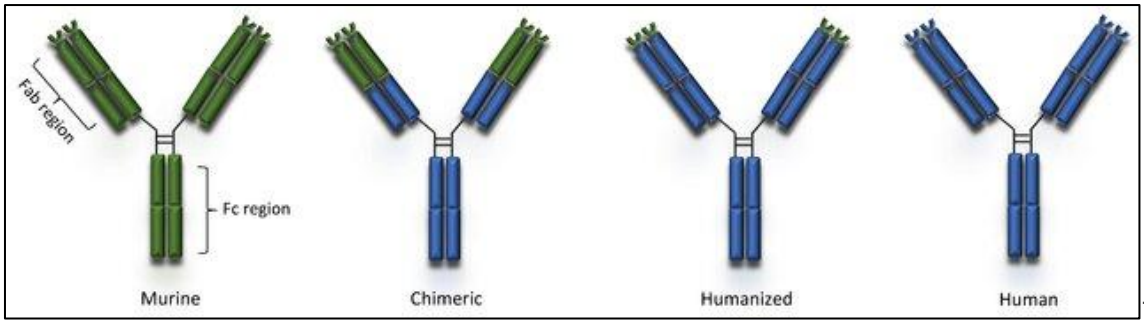
1. **Signaling Blockade:** The mAb binds to a growth factor or a receptor on a cell surface, "plugging" it so the cell can no longer receive instructions to divide or grow.
2. **Immune System Recruitment: * ADCC (Antibody-Dependent Cellular Cytotoxicity):** The antibody acts as a "flag," attracting Natural Killer (NK) cells to destroy the target cell.
    - **CDC (Complement-Dependent Cytotoxicity):** The antibody triggers a cascade of proteins that punch holes in the target cell's membrane.
3. **Payload Delivery (Conjugates):** mAbs can be chemically linked to a drug, toxin, or radioactive particle. The mAb finds the cancer cell, enters it, and releases the "payload," killing the diseased cell while sparing healthy ones.

5. Major Clinical Applications

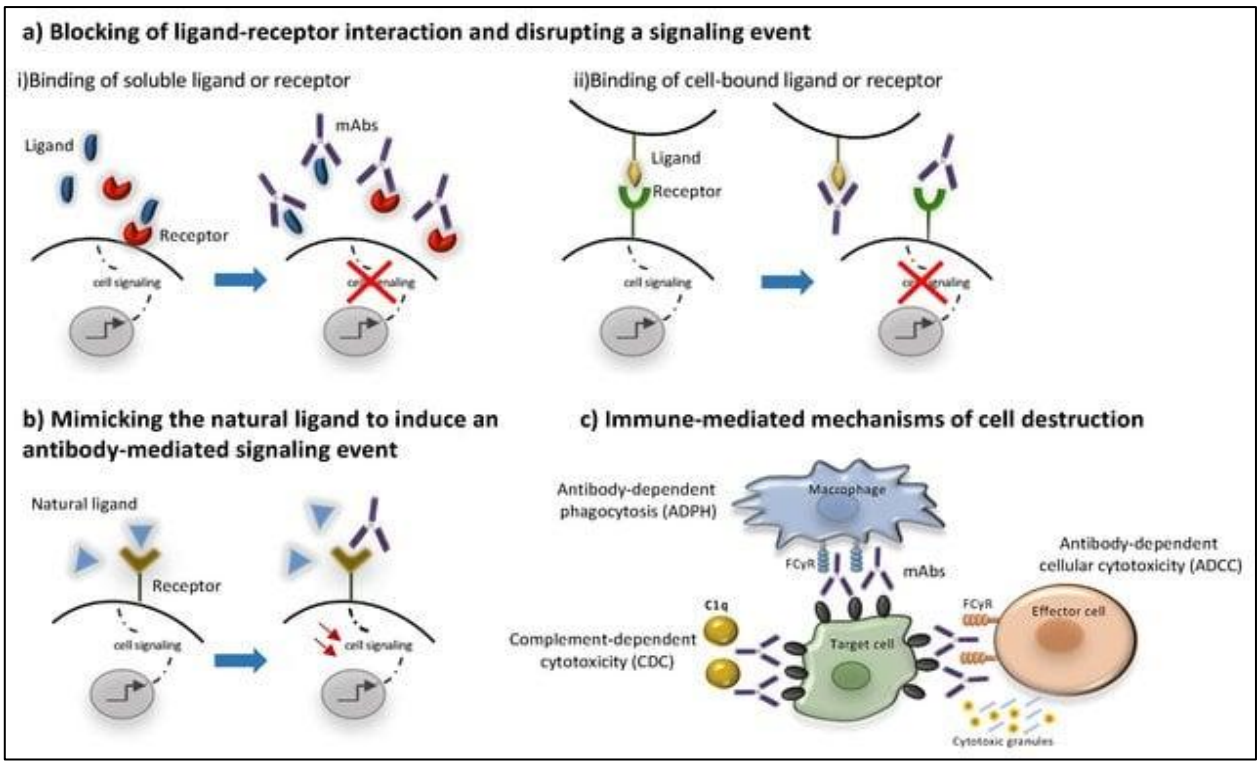
- **Oncology:** mAbs like *Trastuzumab* (Herceptin) for breast cancer or *Rituximab* for non-Hodgkin lymphoma have transformed survival rates.
- **Autoimmune Diseases:** By neutralizing inflammatory proteins like TNF-alpha, drugs such as *Adalimumab* (Humira) treat rheumatoid arthritis and Crohn’s disease.
- **Infectious Diseases:** Used to neutralize toxins or prevent viral entry (e.g., treatments for RSV or COVID-19).
- **Diagnostics:** They are the core technology behind pregnancy tests, COVID-19 rapid tests, and advanced imaging in pathology labs.

6. Conclusion

Monoclonal antibodies represent the pinnacle of **Targeted Therapy**. By moving away from the "carpet bombing" approach of traditional chemotherapy and toward the "sniper" precision of immunology, they have opened new doors for treating previously incurable diseases. Despite their high cost and complex manufacturing, they remain the fastest-growing class of drugs in the pharmaceutical industry today.



Types of monoclonal antibodies.



Mechanisms of action of monoclonal antibodies. MAbs may act through direct (a,b) or indirect mechanisms (c). The direct mechanisms include: (a) blocking ligand-receptor interactions through binding to (i) a soluble ligand or receptor or (ii) to a cell-bound ligand or receptor leading to inhibition of downstream signaling events, (b) agonism through binding to a receptor by mimicking its natural ligand leading to the activation of signaling pathways. Indirect mechanisms are immune -mediated as they involve the activation of certain types of immune cells and molecules to kill target cells (c). Most mAbs bear a human IgG1 Fc region that can activate effector cells, such as natural killer (NK) cells to induce antibody-dependent immune cell cytotoxicity (ADCC), or macrophage inducing antibody-dependent phagocytosis (ADPH), through the interaction with their FCγ receptors. Moreover, the Fc region of mAbs can activate the complement leading to complement-dependent cytotoxicity (CDC).