Lymphocyte circulation and homing

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An overview of lymphocytes activation

• Lymphocytes activation denotes an ordered series of events through which a resting lymphocyte is stimulated to divide and produce progeny, some of which become effectors cells.

• The full response thus include
  - Cell proliferation
  - Expression of immunologic functions
- Lymphocyte become activated when specific ligands bind to receptor on their surfaces.

Events of lymphocytes activation:

- Activation of PTK
  - Ligand induced clustering of receptors on the B or T cells appear to be a key event in triggering PTK activation.
  - The activated PTK appear to be either directly or indirectly responsible for triggering all subsequent events in lymphocytes activation.
- Soa cytosolic enzyme, phospholipase C-δ1, is activated which then acts to hydrolyze a specific class of phospholipids called phosphatidylinositides which give
- diacylglycerol (DAG)
- Inositol 1,4,5, triphosphate (IP3) serve as second messengers to trigger additional changes in cellular physiology
- DAG bind and activate PKC while IP3 released to cytoplasm and trigger calcium ions
- PKC and increased calcium are thought to be critical for initiating the subsequent events in activation
Trafficking, homing and adhesion

Trafficking

Non-random movement of cells from tissues, blood or lymph.
Includes migration to and from sites of lymphocyte maturation as well as homing.

Adhesion:

Binding of cells to other cells or extracellular matrix

Homing:

Tendency of lymphocytes activated in a particular region of the body to preferentially return to the same region.
Includes localisation of cells in distinct regions of lymphoid tissue.
Quantitative aspects of lymphocyte migration

- Traffic between lymphoid/non-lymphoid tissues involves ~5 x 10^{11} cells per day
- Only ~2% (1 x 10^{10}) of these cells are in the blood at any one time
- Lymphocytes only stay in the blood for ~30 minutes
- Circulating blood pool of lymphocytes is exchanged 48 times a day
- *However*……
- Less than 10% of blood lymphocytes migrate into lymph nodes, tonsils & Peyer’s patches.
- ~90% of lymphocytes leave the blood to enter organs such as the liver, lung spleen and bone marrow.
- Traffic is 5 times faster than traffic through lymphoid tissue
Why is lymphocyte homing necessary?

Tendency of lymphocytes activated in a particular region of the body to preferentially return to the same region.

Gut pathogen e.g. rotavirus

Anti-rotavirus T cells will never be needed in the skin

Anti-rotavirus T cells will be needed in the gut

Response resolves, lymphocytes non-randomly redistributed
• Lymphocytes are migratory cells; their distribution in the body reflects the rate at which they enter and depart particular sites as well as their local replication.

• Individual lymphocyte in a lymph node for example, linger there for an average of only 12 hours before detaching from the reticulin matrix and exiting through efferent lymphatics, swept along by the flowing lymph, these emigrating cell eventually are carried into the bloodstream, which disperses them throughout the body but they generally remain in circulation for only few minutes or hours before again taking up residence temporarily another lymphoid organ.
In most types of lymphoid organs lymphocytes enter via blood vessels and exit through lymphatics but in spleen they enter and exit directly from the blood.

Lymphocytes traffic among organs is not random. Resting naïve lymphocyte shuttle almost exclusively among the LN, Peyer’s patches, tonsils and spleen. Memory and effector cells can invade not only those sites but also the diffuse submucosal lymphoid tissues of the gut and the lung, the pulomnary interstitium and inflamed or infected sites in any organ.
• The effector and memory cells once activated show a very strong preference to return to the same type of tissue in which activation originally occurred.

• Ex. a memory cell originally activated in a lymphoid organ in the gut will tend to home preferentially to other gut associated lymphoid tissues for the rest of its life
• These tissue–selective homing patterns result from interactions between surface molecules on lymphocytes, homing receptors, (e.g. Selectin) and endothelial cells (HEV) (adressin)
Selectins & addressins

- SELECTINS
  - Leucocytes inc. Naive T cells: L SELECTIN
  - Endothelial cells: P SELECTIN & E SELECTIN
  - P selectin: Weibel-Palade bodies. E selectin: TNF & IL-1 induced

- A common core with different extracellular C type lectin domains that bind carbohydrates in a Ca2+ dependent manner.

- Each selectin binds to specific carbohydrates and is able to transduces signals into the cell
• VASCULAR ADDRESSINS
• *On high endothelial venules in lymphoid tissue:*
  • Carbohydrates that “decorate” CD34 and GlyCAM-1
  • Sialyl LewisX molecules
  • Peripheral Node addressins (PNAd)
• *Mucosal endothelium:*
  • MAAdCAM-1
• Guides lymphocyte entry into lymphoid tissues
High endothelial venules

Post capillary venules in 2º lymphoid tissue HIGH ENDOTHELIAL VENULES. Specialised to allow lymphocytes and nothing else into the lymph node.

Post capillary venules in other tissues are lined by simple squamous epithelium.
Role of endothelial cells in trafficking and recirculation

Endothelial are involved in:
Vasomotor tone, vascular permeability, regulation of coagulation, immune modulation and lymphocyte extravasations

High endothelial venules
Constitutively present in secondary lymphoid tissue
Need to allow egress of naïve cells from the circulation

Post-capillary venules
Present in non-lymphoid tissues

Molecules expressed by endothelial cells regulate trafficking and recirculation through lymphoid and non-lymphoid tissues
Phases of lymphocytes binding and penetration of the vessel wall

• **Primary adhesion phase**

• An initial interaction of most nonintegrin – homing receptors with a vascular addressin is relatively weak and short lived so that the cell continue to roll along the HEV, last few seconds.
Cytokine activated endothelial cells express adhesion molecules.

Li
• **Secondary adhesion phase**

In this phase the cell rapidly translocate presynthesized integrin onto its surface (Leucocyte function associated antigen, LFA-1) stop rolling and become firmly anchored to the endothelium, secondary lymphoid-tissue chemokine (SLC) on HEV.

• **Diapedesis**

After adhering to HEV a lymphocyte can then pass between high endothelial cells to enter the surrounding tissue, this depend on the continued expression of surface integrins that provide attachment to adjacent cells and to the extracellular matrix.
• After exiting the blood stream each lymphocyte must then continue its migration within the lymphoid organ until it reaches an appropriate resting place
e.g.- Para cortical region for most T cells
• - Lymphoid follicles for B cells of a L.N.
• This controlled by chemokines
• For example :
• SLC and ELC are expressed in the T-cell zone of lymphoid organs and are responsible for attracting naïve T cells to these area
• BLC attrack B cells to follicles
Function of lymphocytes migration

- 1- They can survey the entire body from foci of infection or foreign body
- 2- Such movement help maintain a balance overall distribution of lymphocytes among the tissues
- 3- Modulate and optimize a cell growth and function
- 4- This process places strong pressure on the population as the migrating lymphocytes are forced to compete with one another for a limited space available in each tissue
THE IMMUNE RESPONSE
Immunity

The ability of the body to fight infection and/or foreign invaders by producing antibodies or killing infected cells.

Immune System

The system in the body responsible for maintaining homeostasis by recognizing harmful from nonharmful organisms and produces an appropriate response.
• **Pathogens**
  – Viruses, bacteria or other living thing that causes disease/immune response.

• **Antigens**
  – Toxins that pathogens produce that cause harm to an organism.
Parts of the Immune System

2. Lymph nodes
3. Thymus Gland – Produces T Lymphocytes
4. Bone Marrow – Produces B Lymphocytes
Nonspecific Immune Response

These are defenses the body uses no matter what the invader may be. These defenses include:

- **Phagocytosis** – done by Macrophages
- **Natural Cell Killers**
- **Inflammation** - caused by release of Histamine from leukocytes
- **Fever** – caused by histamines. The fever (high temp) kills invaders by denaturing their proteins.
Specific Immune Response

This is a specific response to a specific pathogen/antigen.

• The response involves the creation of Antibodies. And cell mediated
The Pathway of Specific Immune Response

Step 1
Pathogens eaten by Macrophage

Step 2
Displays portion of Pathogen on surface

Step 3
Helper-T cell recognizes Pathogen
Step 1: Pathogens eaten by Macrophage

Step 2: Displays portion of Pathogen on surface

Step 3: Helper-T cell recognizes Pathogen
Immune Response Explained

1. Antigen infects cells.
2. Macrophage ingests antigen and displays portion on its surface.
3. Helper T-Cell recognizes antigen on the surface of the macrophage and becomes active.
4. Active Helper T-Cell activates Cytotoxic T-Cells and B-Cells.
5. Cytotoxic T-Cells divide into Active Cytotoxic T-cells and Memory T-Cells.
6. Active Cytotoxic T-Cells kill infected cells.
7. At the same time, B-Cells divide into Plasma Cells and Memory B-Cells.
8. Plasma cells produce antibodies that deactivate pathogen.
9. Memory T and Memory B cells remain in the body to speed up the response if the same antigen reappears.
10. Supressor T-Cells stop the immune response when all antigens have been destroyed.
Primary vs. Secondary Immune Response

- **Primary Immune Response**
  - This is a response to an invader the First time the invader infects the body.
    - No measurable immune response for first few days.
    - Next 10 – 15 days antibody production grows steadily

- **Secondary Immune Response**
  - A more rapid response to an invader the 2nd time it invades the body.
    - Antibody production increases dramatically and in a much shorter time period.
Passive vs. Active Immunity

1. **Active Immunity**
   This is immunity where the body is “actively” producing antibodies to fight infection.
   Ex: You have a throat infection and you are actively creating antibodies to fight it.
   
   **Vaccination:** An injection of a *weakened strain* of an infectious microbe (pathogen) that causes the body to undergo active immunity (produce antibodies).

2. **Passive Immunity**
   This is immunity where antibodies are given to a person from the blood of another person or animal.
   This immunity only lasts for a short period of time.
   ex: Breastfeeding mothers pass antibodies to their children through the milk.
Lymphocyte Subsets

Lymphocyte subsets

CLP

T CELLS

Common lymphoid precursor

B CELLS

T CELLS

T

Th

Activate B cells and macrophages
THETAPEL CELLS

CTL

Kill virus infected cells
CYTOTOXIC T LYMPHOCYTES

PC

Produce antibodies
PLASMA CELLS
• All the progeny cells derived from any single naïve lymphocyte constitute a lymphocyte clone
• Some members of each clone differentiate into effector cells whereas the reminder are memory cells
• Clonal restriction
• Each lymphocyte or clone of lymphocyte has a uniquely restricted specificity for antigen
• The lymphopoeitic system is able to produce lymphocytes with approximately $10^8$ alternative antigen specificity (primary lymphocyte repertoire)
• Clonal selection
• Exposure to an antigen selectively promotes the growth of any clones that recognize it without affecting other cells in the population
Clonal Selection

Memory Cells

Antibody-Producing Plasma Cells
B Lymphocytes

- First identified as being produced in the bursa of Fabricius in birds
- In humans, B cells are produced in the bone marrow (in adults) and fetal liver
- Primary effectors of humoral immunity (via antibody production)
- Two main groups of B cells: antibody producing (plasma cells) and memory cells
- Recognize antigen by means of surface-expressed antigen receptors
- Generally dependent on “T cell help”
Antigens and Antibodies - I

- **Antigen** - derived from “antibody generating”; now understood to mean a compound which elicits either a cellular or humoral immune response.

- Can be protein or large polysaccharide.

- Smaller molecules (e.g. lipids, DNA) can be antigenic when coupled to “carrier proteins”. These antigens (called *haptens*) can then bind antibody free from carrier.

- Specific regions of antigen (known as *epitopes* or *antigenic determinants*) interact with/are recognized by antibodies.
Antibodies

- Y-shaped *protein* molecule.
- Made up of *variable* and *constant* regions.
- Made up of *Heavy* and *Light* chains.
- Produced by B-Lymphocytes
- **Function:** Recognize antigens, bind to and deactivate them.
  - **Note:** Variable region recognizes the antigens.
How an antibody operates/works

Deactivation of a bacterium by an antibody.
T Lymphocytes

- T cells derive from stem cells in the bone marrow, “mature” in the thymus, and then are released into the “periphery”

- Similarities between T and B cells:
  - Antigen receptor on surface (T cell receptor)
  - Recognize single, specific antigen
  - Expand through clonal selection
  - Some T cells exist as long-lived memory cells

- T cell production decreases during adolescence, due to *thymic involution*
T Cell Development and Thymic Maturation

Approximately 95% of all thymocytes die in the thymus!!!
T Cell Activation/Differentiation

- TCR-MHC/Ag interaction + costimulation $\rightarrow$ activation of naïve T cells
- “Primary response” occurs within 48 hours
- Involves increased transcription of IL-2, IL-2R (receptor)
- Why both?
- IL-2 $\rightarrow$ proliferation and differentiation into memory cells and effector cells ($T_h$ and $T_c$)
Effector T Cells

CD8 → TCR → Tc Cell

CD4 → TCR → Th Cell

Cytotoxic Activity

Activation

Th1

IL-2
IFN-γ

Th2

IL-4, IL-5
IL-6, IL-10
Memory T Cells

- Derived from both naïve and effector T cells
- Long-lived
- Secondary response – rapid response upon re-encountering antigen (less stringent activation requirements, little or no costimulation required)
- Phenotypically, they appear similar to effector cells
- Do they require persistent antigen stimulation?
Activation of CD8 cells

- Responds quickly giving rise to other T cells
- Cytotoxic T cells – seek out and destroy abnormal cells
  - lymphotoxin
- Memory T<sub>C</sub> cells – function during a second exposure to antigen
- Suppressor T cells – suppress the immune response
Antigen Recognition and the Activation of Cytotoxic T Cells
Unlike B cells, T cells cannot recognize “free” antigen; it must be:

- Processed (digested) in to smaller fragments within special “antigen presenting cells” (APC’s)
- Presented on the surface of APC’s in the context of MHC (major histocompatibility complex) proteins
Infected cell

Viral or bacterial antigen

Inactive cytotoxic (CD8) T cell

ACTIVATION AND CELL DIVISION

Memory T<sub>C</sub> cells (inactive)

Active T<sub>C</sub> (cytotoxic) cells

Lysed cell

Class I MHC

Antigen

Costimulation

CD8

Lymphotoxin release

Cytokine release

Perforin release

Disruption of cell metabolism

Stimulation of apoptosis

Destruction of cell membrane

DESTRUCTION OF TARGET CELL
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<tr>
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<th>Dendritic cell</th>
<th>Macrophage</th>
<th>B Lymphocyte</th>
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<tbody>
<tr>
<td><strong>Antigen uptake</strong></td>
<td>Endocytosis</td>
<td>Phagocytosis</td>
<td>Receptor-mediated endocytosis</td>
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<td><strong>Class II MHC</strong></td>
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<td><strong>T-cell activation</strong></td>
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<td>Effector T cells</td>
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Fig 10-18
In contrast to naïve T cells, effector T cells have:
  - Less stringent activation requirements (reduced costimulatory dependency)
  - Increased expression of adhesion molecules (why?)
  - Both membrane-bound and soluble effector molecules

<table>
<thead>
<tr>
<th>Table 14-2: Effector molecules produced by effector T cells</th>
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<tr>
<td><strong>Cell type</strong></td>
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<tr>
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<td>T_{H1}</td>
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<td>T_{H2}</td>
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Cytotoxic Activity of Tc Cells
(Cytotoxic T Lymphocytes or CTL’s)

Perforin – Pore-forming protein
Granzymes – Serine proteases

Fig. 14-4
Fig. 14-7

- Nucleus
- Granule
- Completed pore
- Polymerized perforin
- Perforin monomers
- CTL
- Target cell
Clonal Deletion

Foreign Antigen

Normal T or B Cell

Self Antigen

Autoreactive T or B Cell

Foreign Antigen

Normal T or B Cell
What Causes Autoimmunity?

- **Non-autoreactive T cell or B cell**: Normal Immune Response
- **Autoreactive T cell or B cell**: Death of Autoreactive T or B Cell (Clonal Deletion)
- **Autoreactive T cell or B cell**: Death by Apoptosis (PCD)