# Immunology

## B lymphocytes & Antibodies

**17. June 2026, Ruhr-Universität Bochum**

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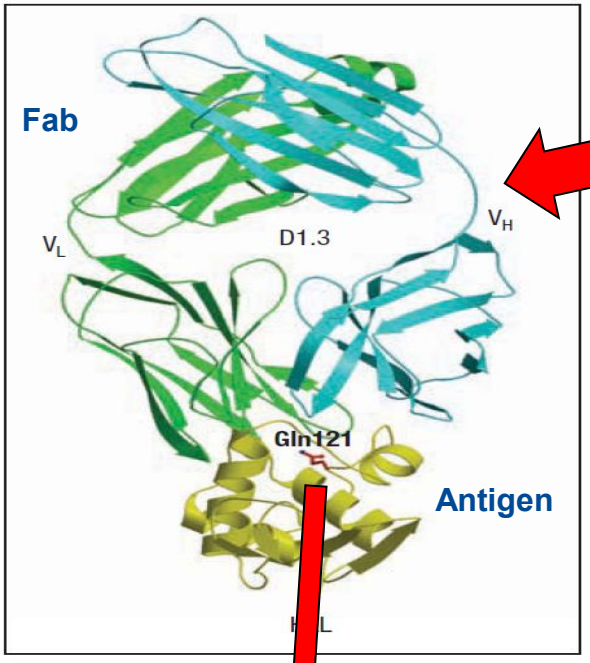
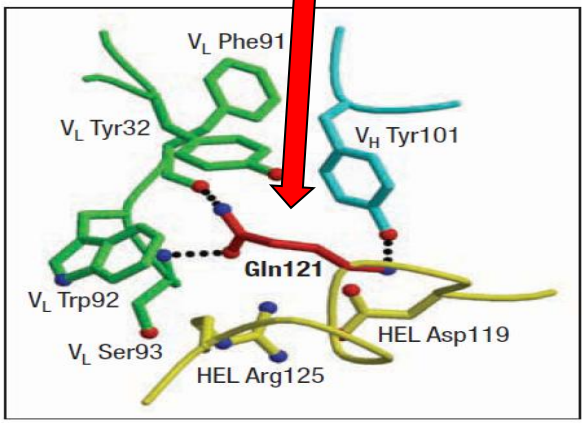
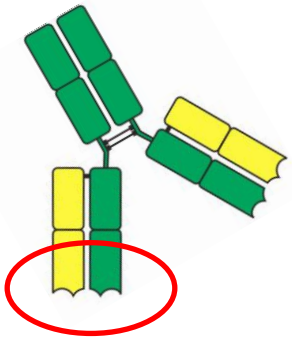
RUB

# The structure of a typical antibody molecule



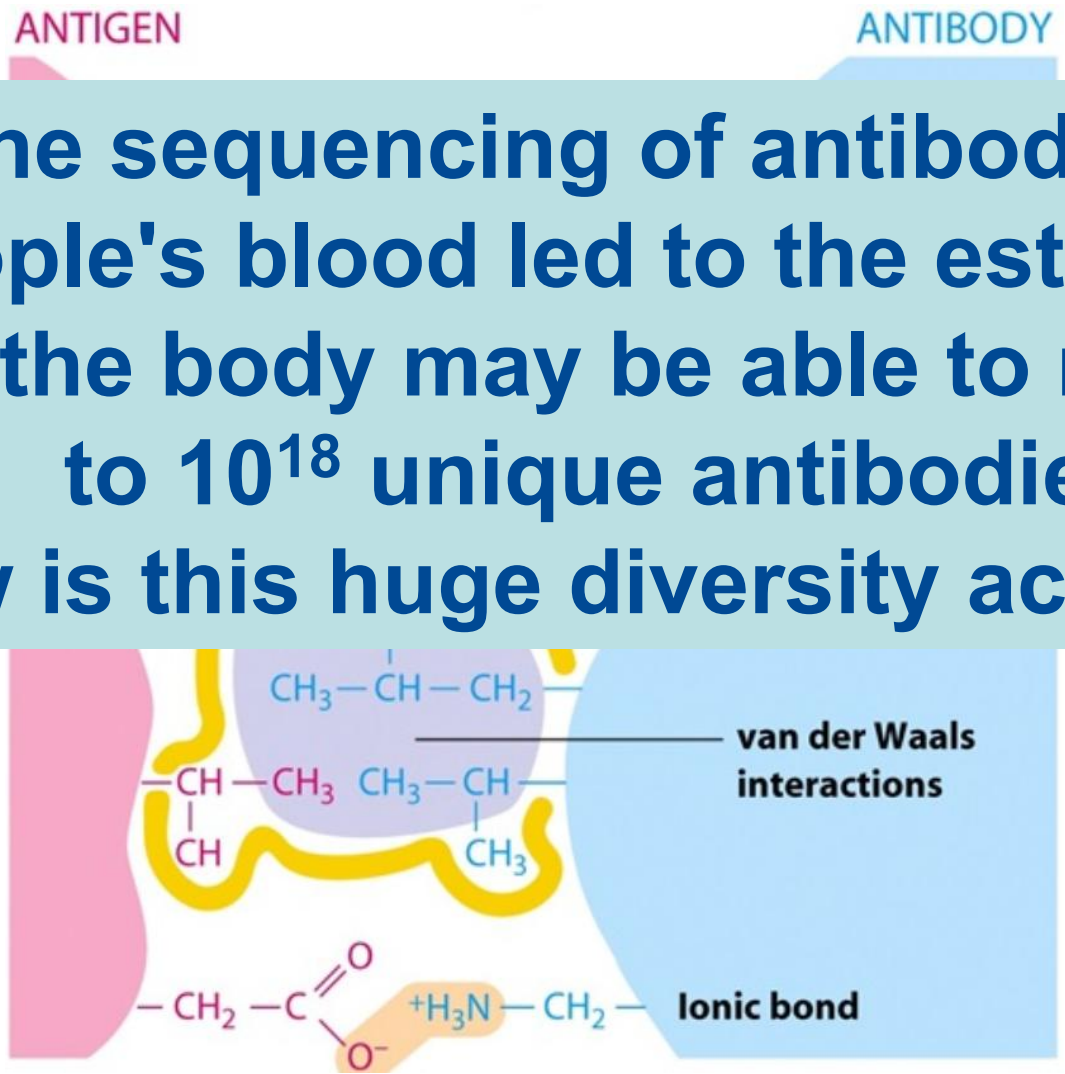
2 heavy chains ($C_H$), 4-5 domains

2 light chains ($C_L$), 2 domains

**The specific sequence of amino acids in the hyper-variable region of the antibody Fab determines the interaction with the antigen. In total 15-22 amino acids from the antibody may participate in the interaction with the antigen.**

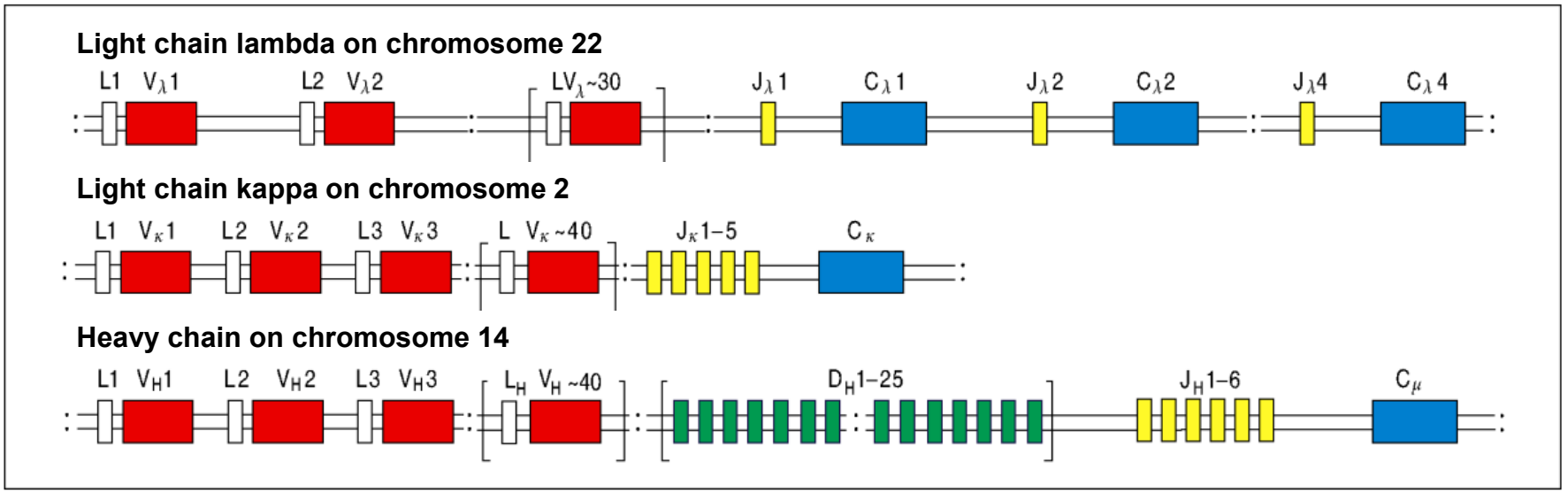
# The binding of an antigen

The binding of an antigen by an antibody is **never** covalent.

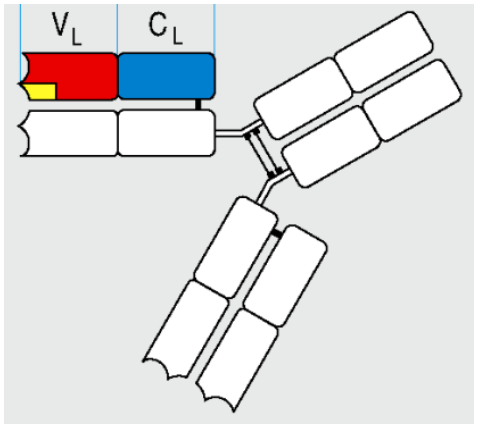


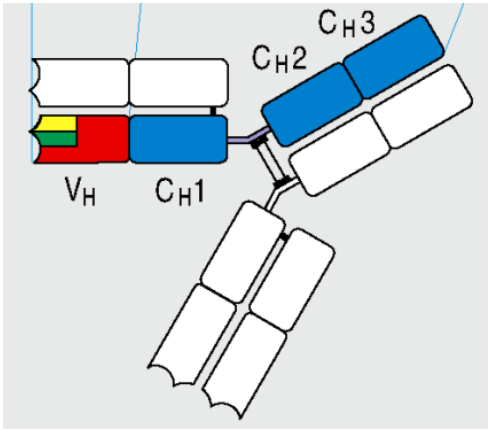

**The sequencing of antibodies in people's blood led to the estimation that the body may be able to make up to $10^{18}$ unique antibodies.**
**How is this huge diversity achieved?**

# Germline configuration of genes for the heavy and light chain




Aus: Murphy, Travers, Walport, *Janeway Immunologie*, 7. Aufl. © Spektrum Akademischer Verlag 2010



# Somatic recombination of the light chain

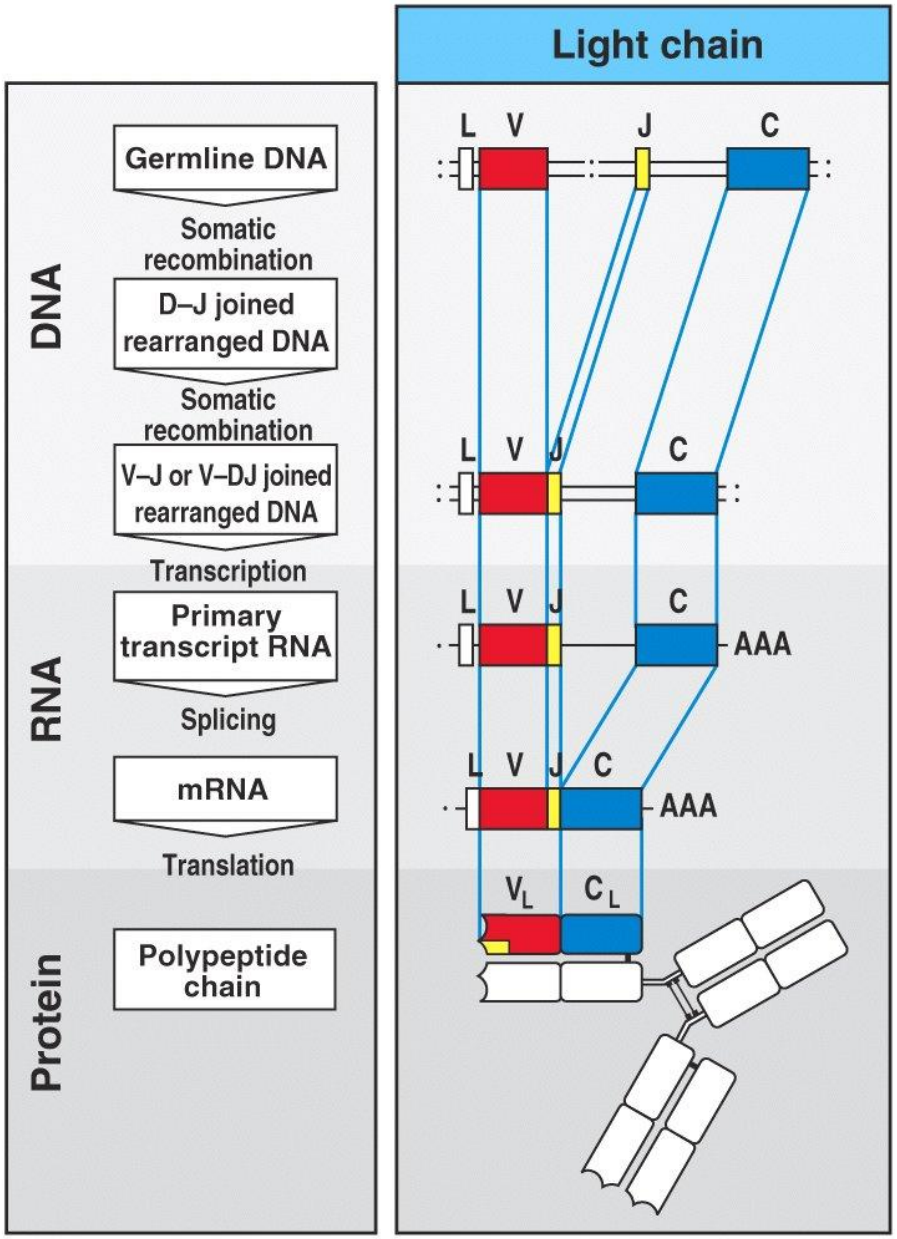



Figure 4-2 Immunobiology, 6/e. (© Garland Science 2005)

Combinatorial diversity:

**Light** chain κ: 40 $V_L$-segments
5 $J_L$-segments

40 x 5 = **200**

Combinatorial diversity:

**Light** chain λ: 30 $V_L$-segments
4 $J_L$-segments

30 x 4 = **120**

**320 possible Light chains**

# Somatic recombination of the heavy chain

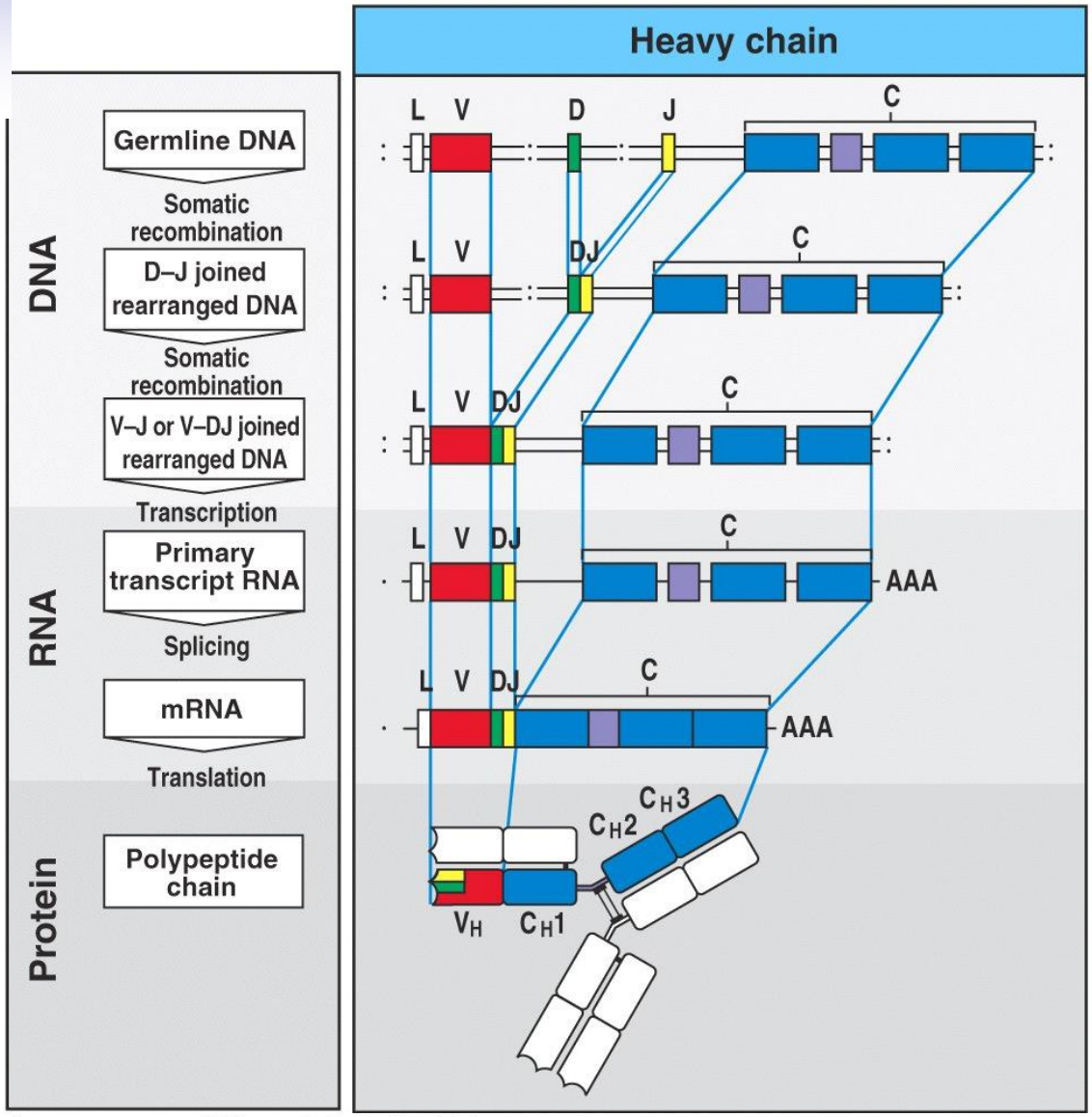



Figure 4-2 Immunobiology, 6/e. (© Garland Science 2005)

Combinatorial diversity:

**Heavy** chain:

40 $V_H$-segments
25 $D_H$-segments
6 $J_H$-segments

40 x 25 x 6 = **6000**

**6000 possible heavy chains**

# The diversity of the antibody repertoire

Combinatorial diversity of antibodies:

6000 different heavy chains times

320 different light chains

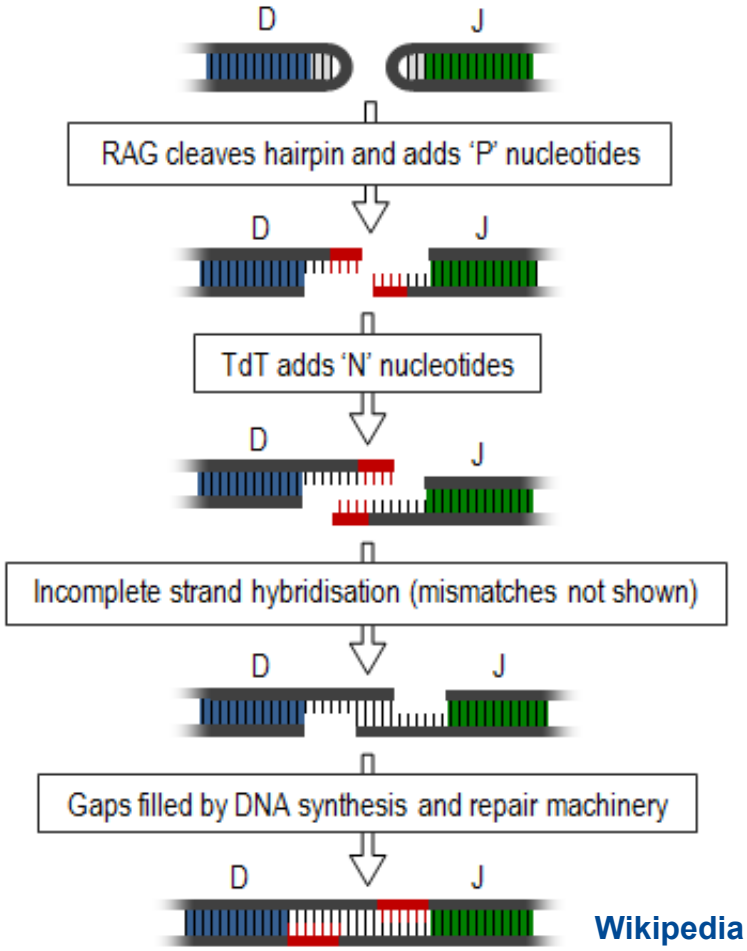
$\longrightarrow$ 1,9 x $10^6$ antibodies

**However total diversity is approximately $10^{18}$ different antibodies!**
**Additional mechanisms exist that further increase heterogenecity, namely:**
**junctional diversification, somatic hypermutation, class switching**

# Junctional diversity

During VDJ recombination the enzyme “recombination activating gene” (RAG1&2) induces palindromic nucleotides (1 to 4 P-nucleotides)

At the junction of the gene segments nucleotides are introduced by the enzyme “Terminal deoxynucleotidyl transferase” (2 to 10 N-nucleotides).




Wikipedia

Due to this process there is a high chance for the induction of frame shift mutation!

# Somatic hypermutation

Affinity maturation of the antigen binding sites takes place in the secondary lymphatic organs (only in activated B-cells).

Mutations occur in the whole variable region of the antibody.

Mutations concentrate on so-called hot spot regions.

B cells become apoptotic when somatic hypermutation resulted in reduced binding affinity of the antibody (during affinity maturation)

# Summary

Antibodies can be raised against almost every molecule

The antibody diversity is based on three different mechanisms:

1) **Combinatorial diversity**
   (Somatic recombination)

2) **Junctional diversity**
   (Inaccurate junction)

3) **Somatic hypermutation**
   (Affinity maturing in germinal centres)

# B-cell development

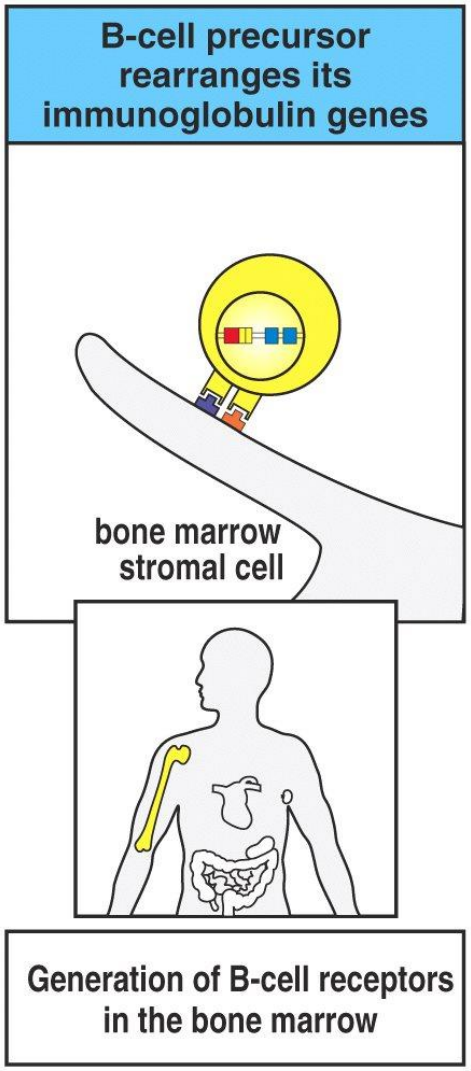



Figure 7-1 part 1 of 2 Immunobiology, 6/e. (© G

- B-cells develop continuously in the **b**one marrow, they derive from lymphatic progenitor cells
- The environment (stromal cells of the marrow) delivers the **necessary milieu** (surface molecules and cytokines) for the **development**
- The **immunoglobulin genes** are **rearranged**

# B-cell development

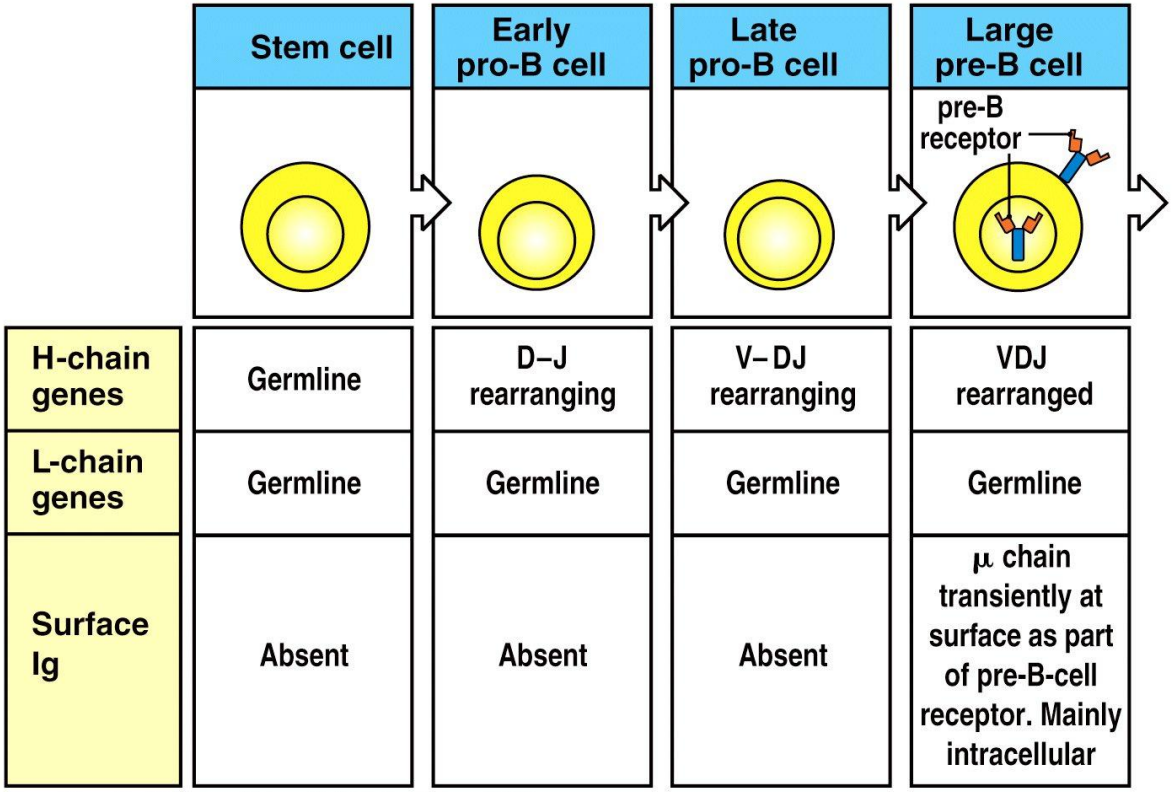



Figure 7-6 part 1 of 2 Immunobiology, 6/e. (© Garland Science 2005)

- The heavy chain is expressed with a surrogate L-chain to stabilize the pre-B-cell-receptor
- After successful expression of the heavy chain the pre-B-cell divides, before the rearrangement of the light chain starts

# B-cell development

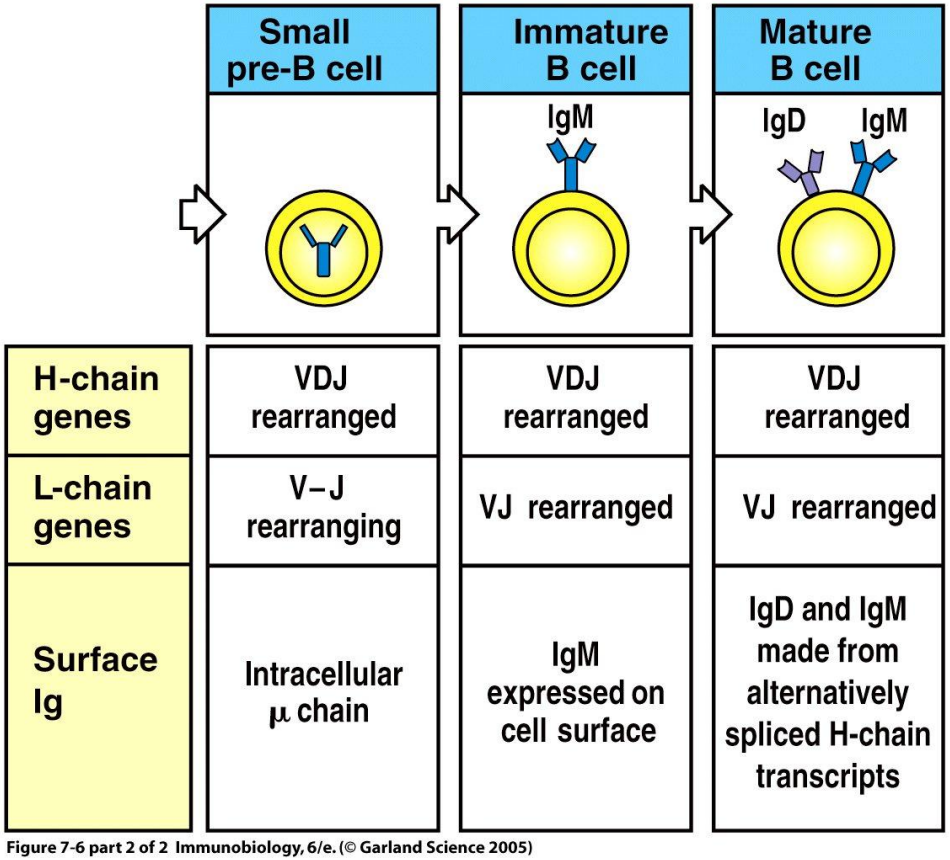
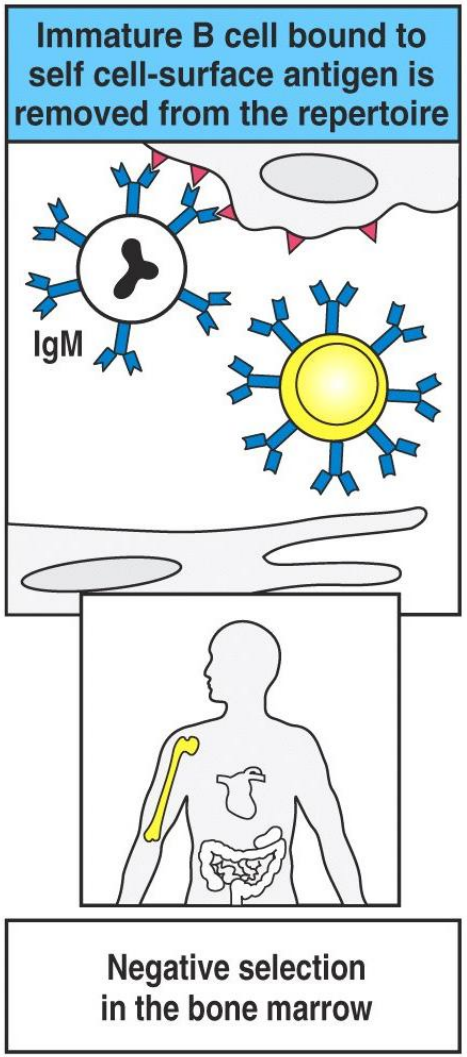



Figure 7-6 part 2 of 2 Immunobiology, 6/e. (© Garland Science 2005)

- Beginning of the V-J-rearrangement of the κ-locus of the light chain, in case of an unsuccessful rearrangement, the λ-Locus becomes rearranged

# B-cell development




Garland Science 2005)

In the bone marrow:

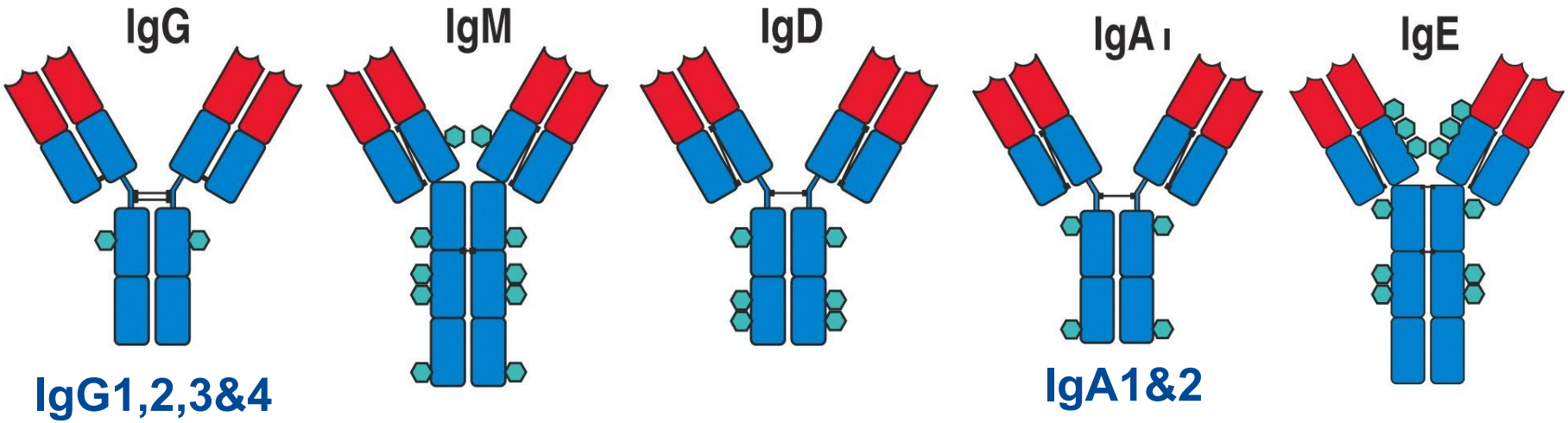
**Selection on self-tolerance**

- Contact to antigens in the environment of the bone marrow is leading to apoptosis or anergy in the immature B-cell

# B-cell development after leaving the bone marrow

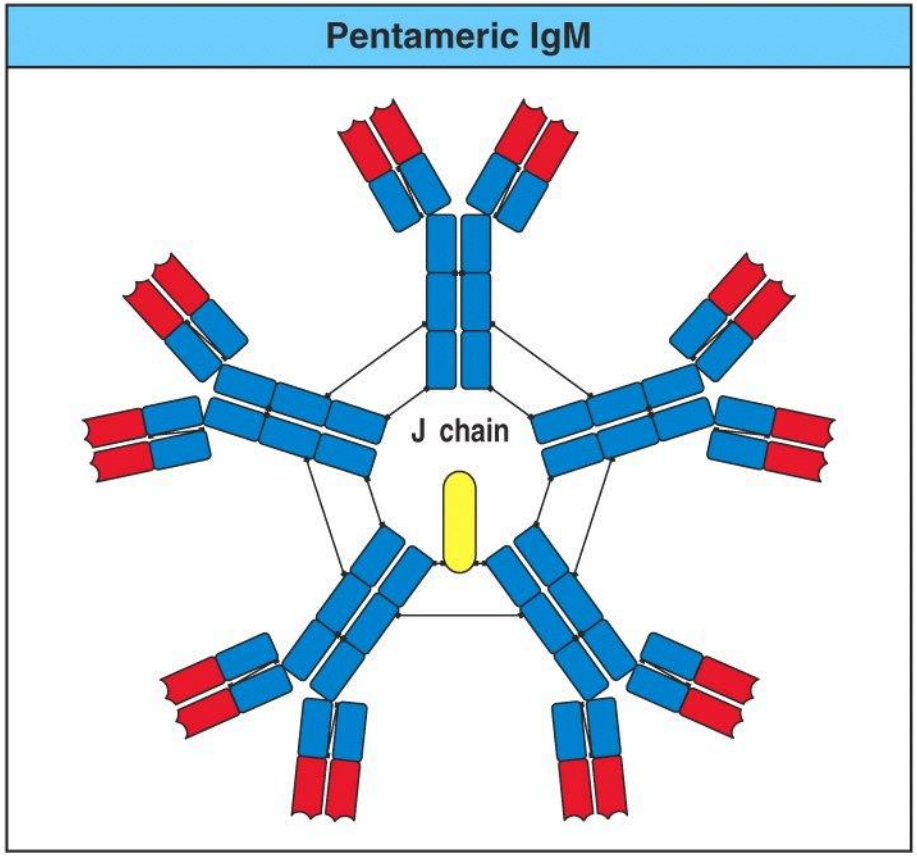
- Self-tolerant, naive B-cells leave the bone marrow.
- Naive B-cells circulate through the blood into the secondary lymphatic organs.
- If the B-cells encounter their specific antigen, they are activated and can differentiate into plasma cells or memory cells.

# Antibody classes - Isotypes




| Ig | IgG | IgM | IgA | IgD | IgE |
|---|---|---|---|---|---|
| Serum concentration (mg/dl) | 800-1700 | 50-190 | 140-420 | 0.3-0.4 | <0.001 |

# Antibody classes - Isotypes




The first antibody class produced by each B cell

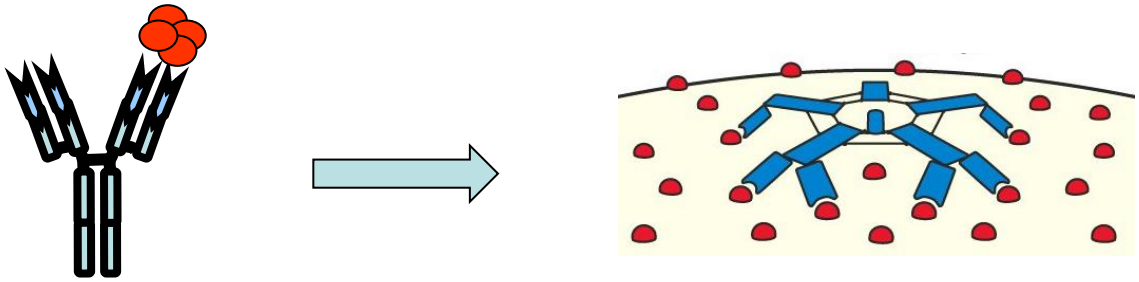
High avidity due to pentamer formation!

# Affinity & Avidity

**IgM compensates its low affinity with high avidity.**

**Affinity** = Binding strength between one antigen binding side and the antigen.

**Avidity** = Total binding strength between more antigen binding sides and one multivalent antigen. The absolute binding strength is potentiated



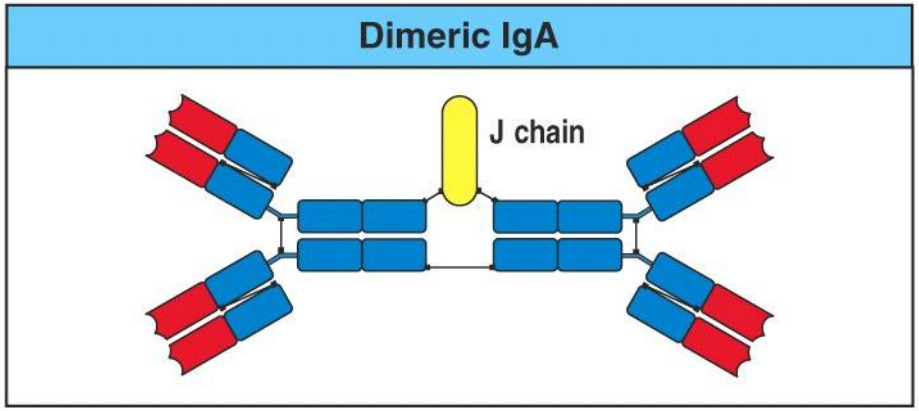
**Affinity**

**e.g. $K_D=10^{-4}$**

**Avidity**

**e.g. $K_D=10^{-10}$**

# Antibody classes - Isotypes

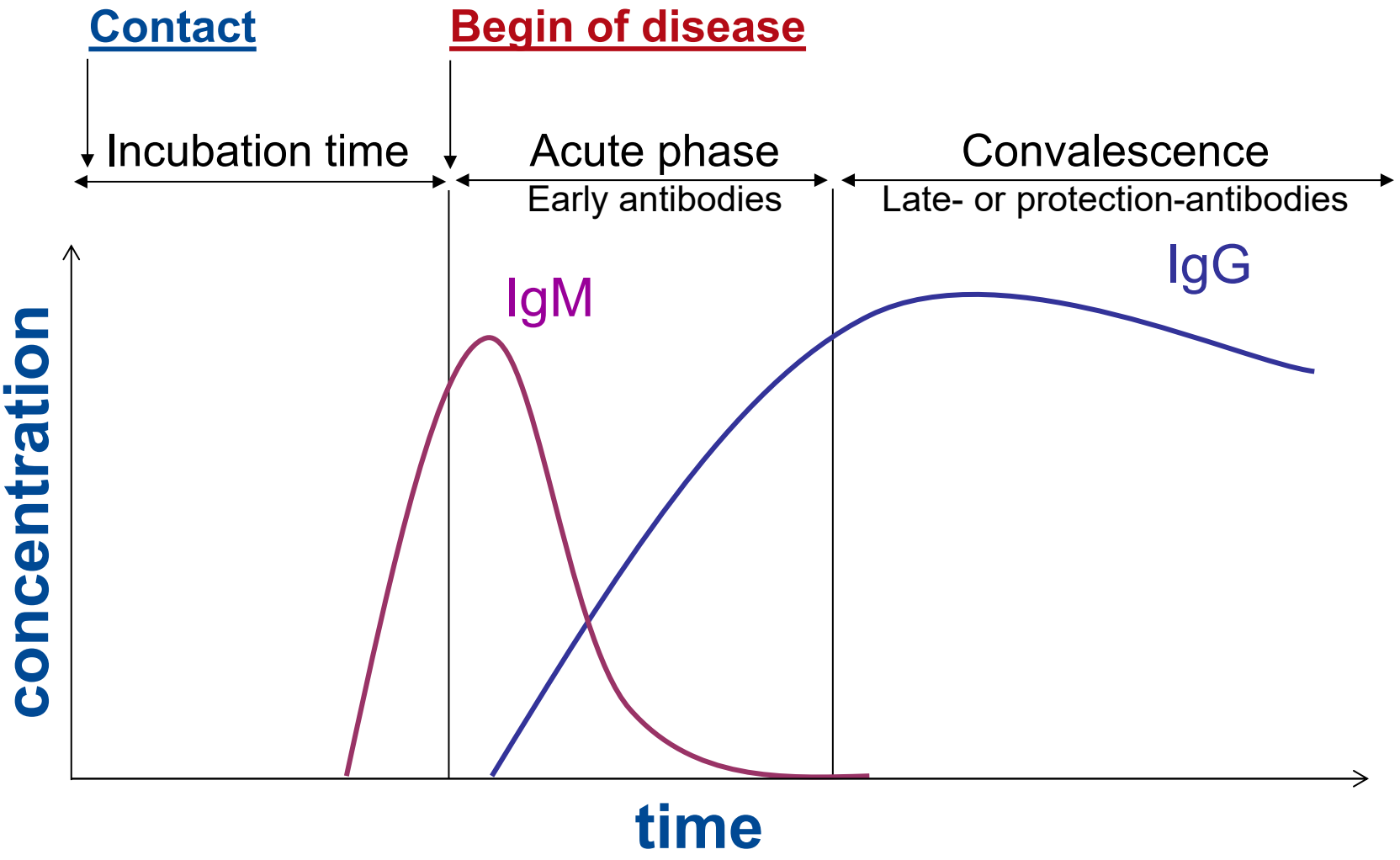



(Garland Science 2005)

IgA forms **dimers**

- Secretion of the antibody on mucosal surfaces

# The serum-concentration of specific antibodies generated during infection to fight the infectious agent



# Antibody classes - Isotypes

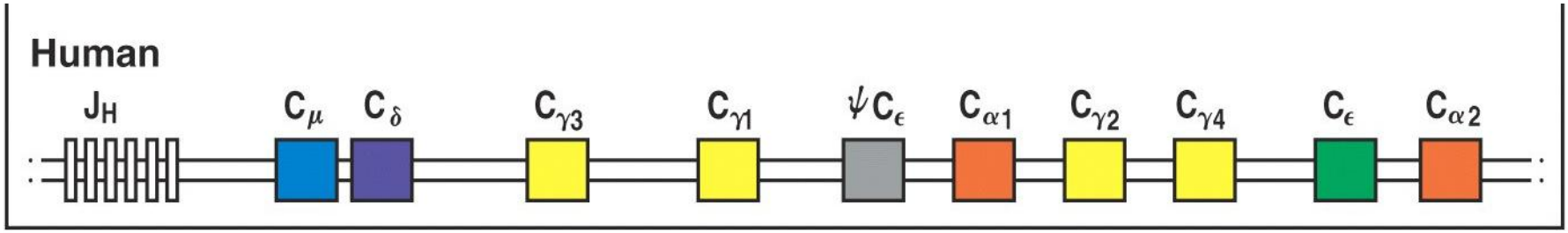



Figure 4-19 Immunobiology, 6/e. (© Garland Science 2005)

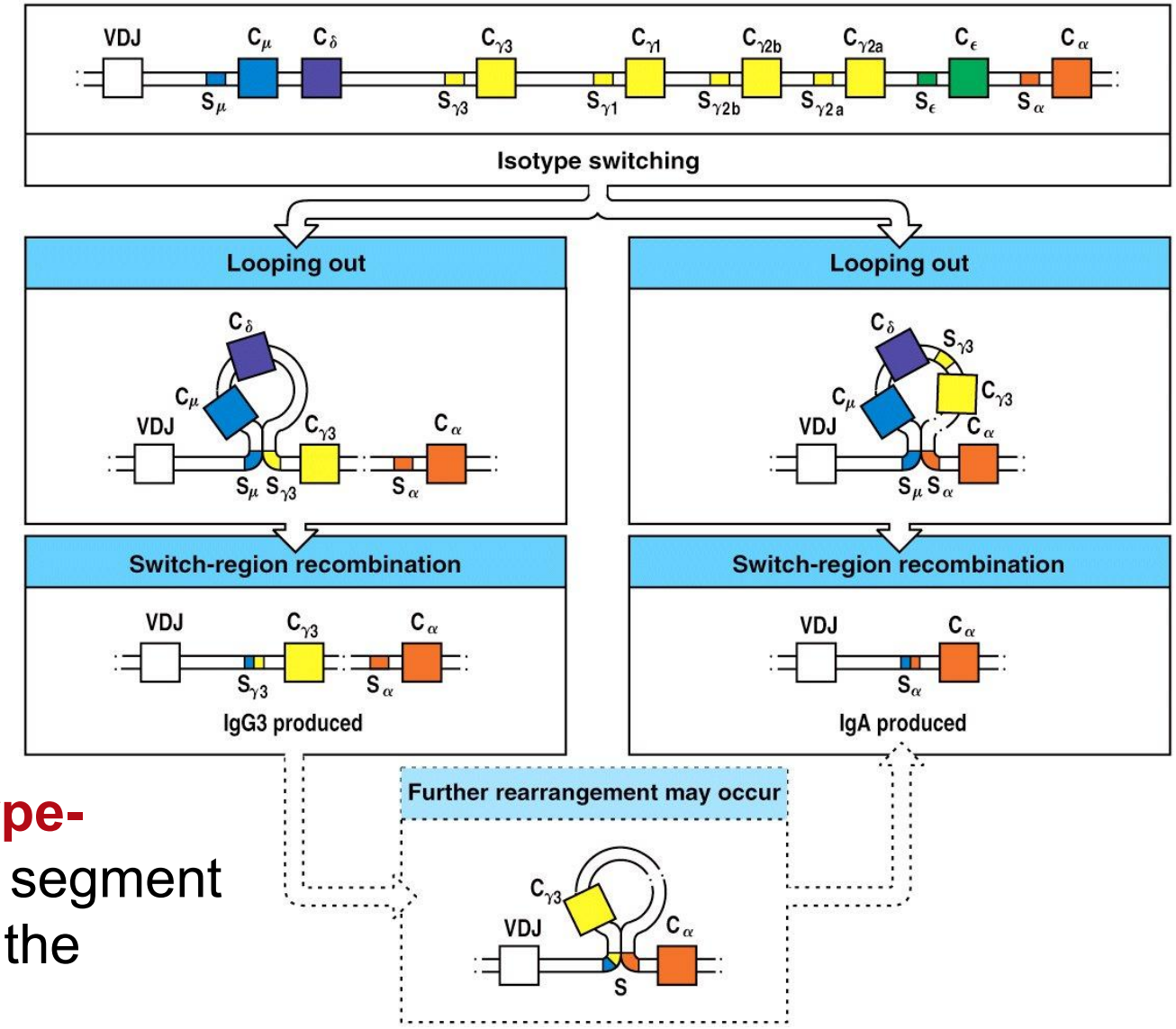
cμ = IgM, cδ = IgD, cγ = IgG, cε = IgE, cα = IgA

Ψ = Pseudogene

The expression of the genes of the constant region changes during the maturation of the B-cell =

Isotype-Switch

# Antibody classes - Isotypes



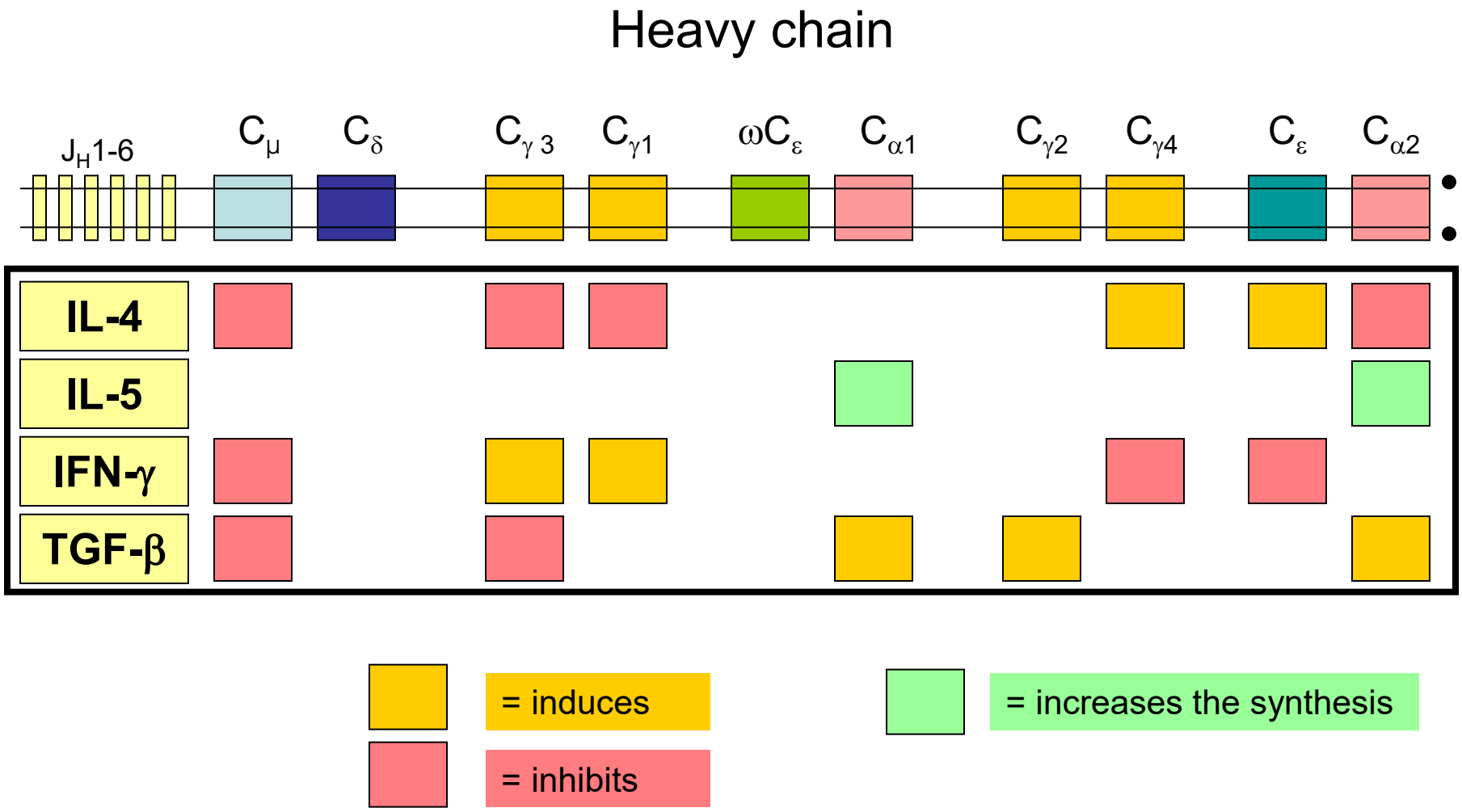

During the **Isotype-Switch** the gene segment located between the switching signal sequences is excised

→ develoment just in one direction

# Antibody classes - Isotypes

- Specific DNA-sequences (**Switch regions**) regulate the recombination
- The isotype-switch takes place in the lymph node (**germinal centre**)
- It requires a specific milieu of Cytokines, which induce transcription of enzymes involved in the switching process: **transcription factors**

# Cytokines involved in isotype-switching



# Antibody classes - function

There is a specific role for each antibody class during the immune response

| Function | IgM | IgD | IgG | IgA | IgE |
|---|---|---|---|---|---|
| Neutralisation | + | - | ++ | ++ | - |
| Opsonisation | - | - | ++ | + | - |
| Mast cell sensitization | - | - | - | - | +++ |
| Complement activation | +++ | - | + | + | - |
| Transport via epithelia | + | - | - | +++ | - |
| Transport via placenta | - | - | +++ | - | - |
| Diffusion into the tissue | +/- | - | +++ | ++ | + |

# B-cell activation

Naive B-cells need **2 signals** for activation:

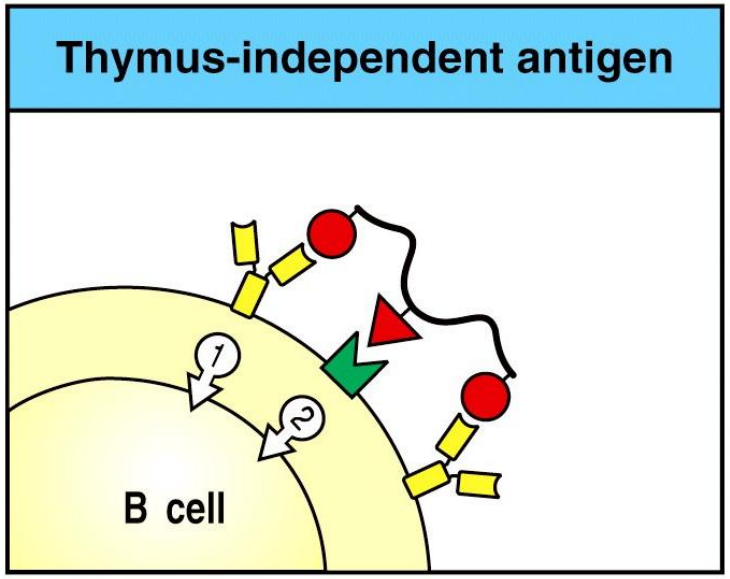
**First signal**: signalling through the B-cell receptor (BCR) = **Surface-Immunoglobulin**

The **second signal** can be given in two ways:

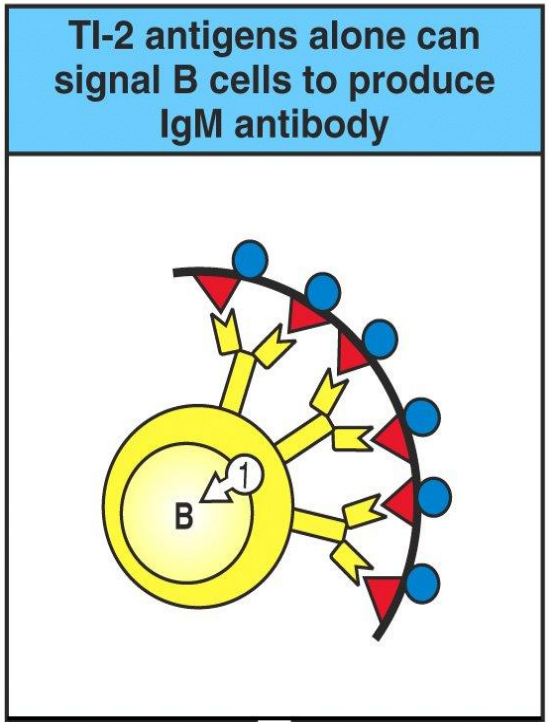
1) **Thymus-dependent** (via already activated T-helper-cells) = TD-antigens (thymus dependent)

2) **Thymus-independent** (activation without Tcell help) = TI- antigen (thymus independent)

# Thymus-independent (TI)-B-cell activation



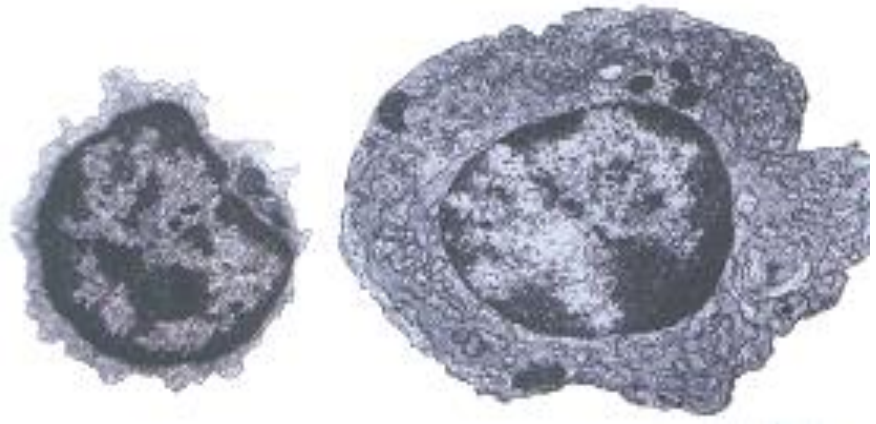

**TI-1-antigens** activate a further receptor on the surface, e.g. TLR-4 against LPS or a complement receptor = co-stimulatory signal




**TI-2-antigens** activate the B-cell by crosslinking the BCR via a large molecule containing antigens without costimulation

# TI-B-cell activation

## TI (1+2)-activation : B-cell → Plasma cell



- no germinal centre
- no memory cells
- no affinity maturation

# Thymus-dependent-B-cell activation

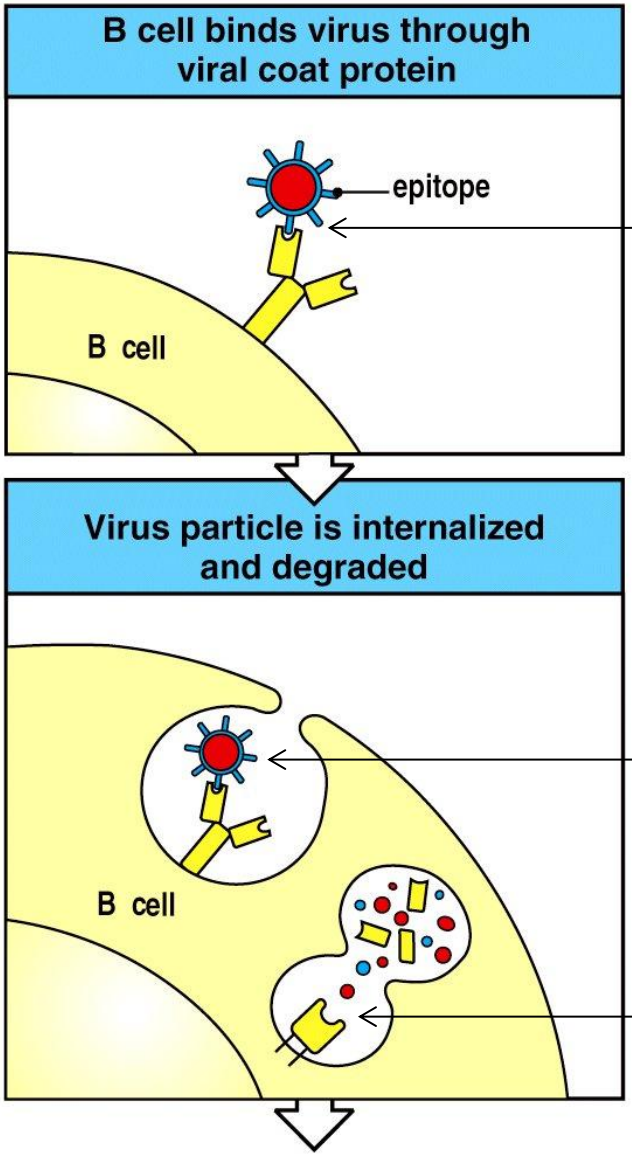



Figure 9-3 Immunobiology, 6/e. (© Garland Science 2005)

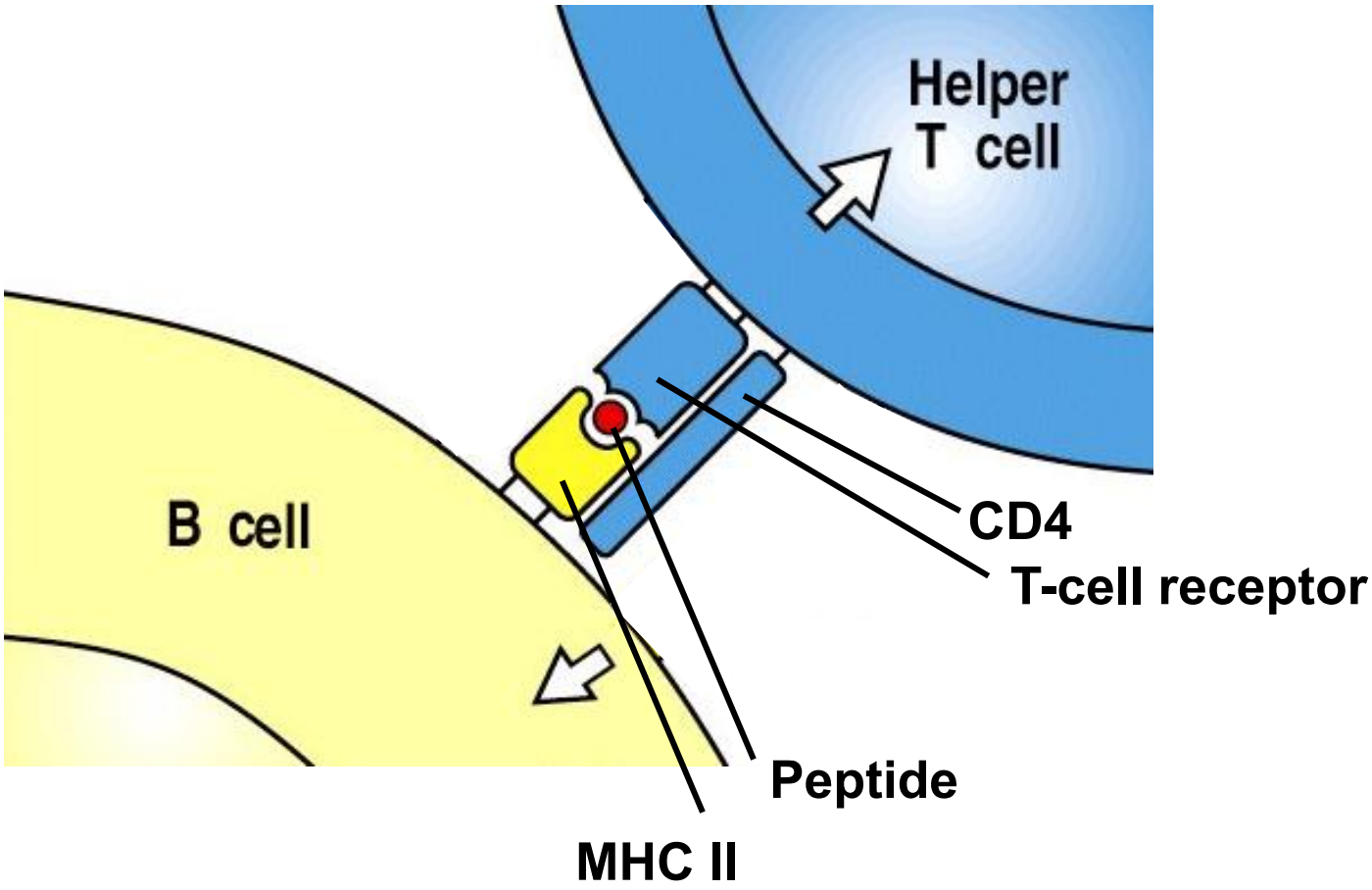
Binding of an antigen to its specific BCR

Internalization of the Antigen/BCR complex

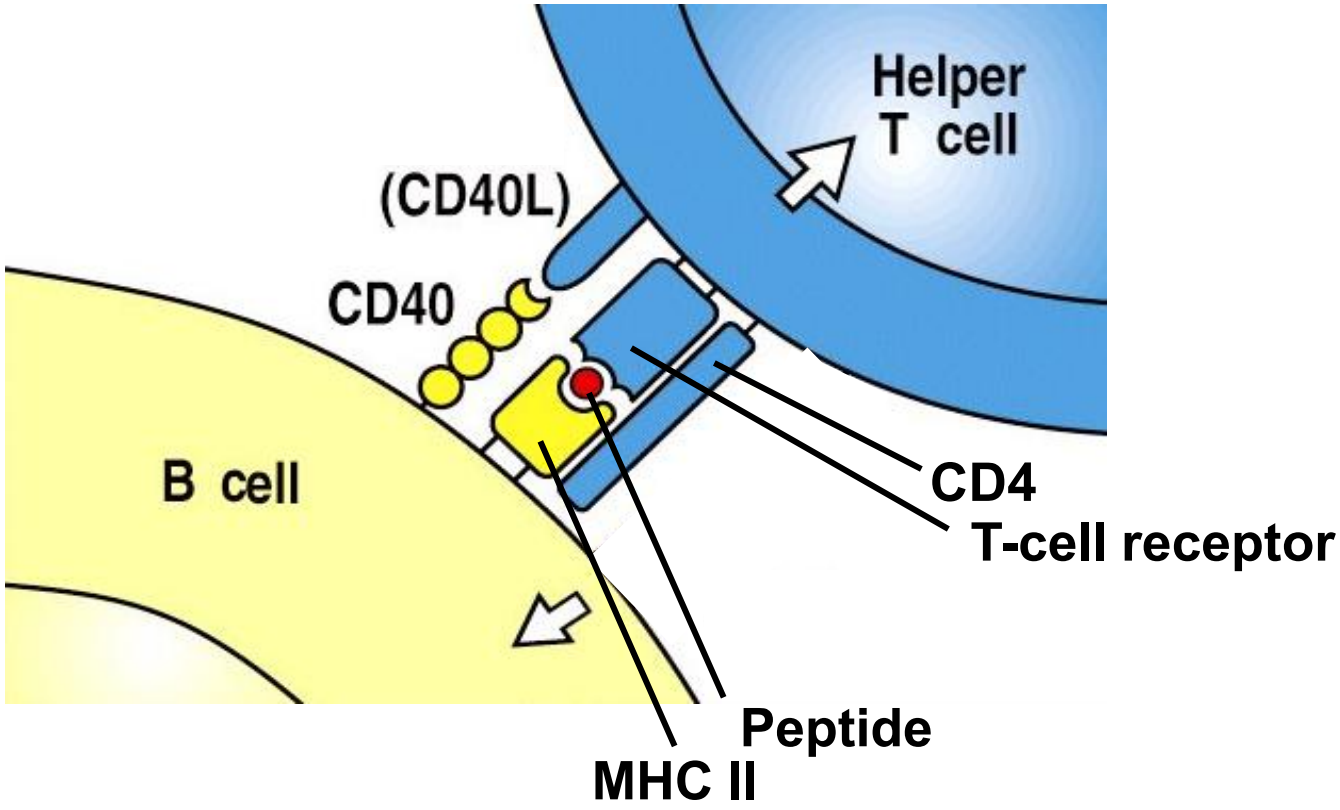
Degradation of proteins and binding to MHCII

# Thymus-dependent-B-cell activation

**Linked recognition** = T-cell and B-cell recognise the same antigen (the B cell via the BCR; and the T-cell via the TCR)

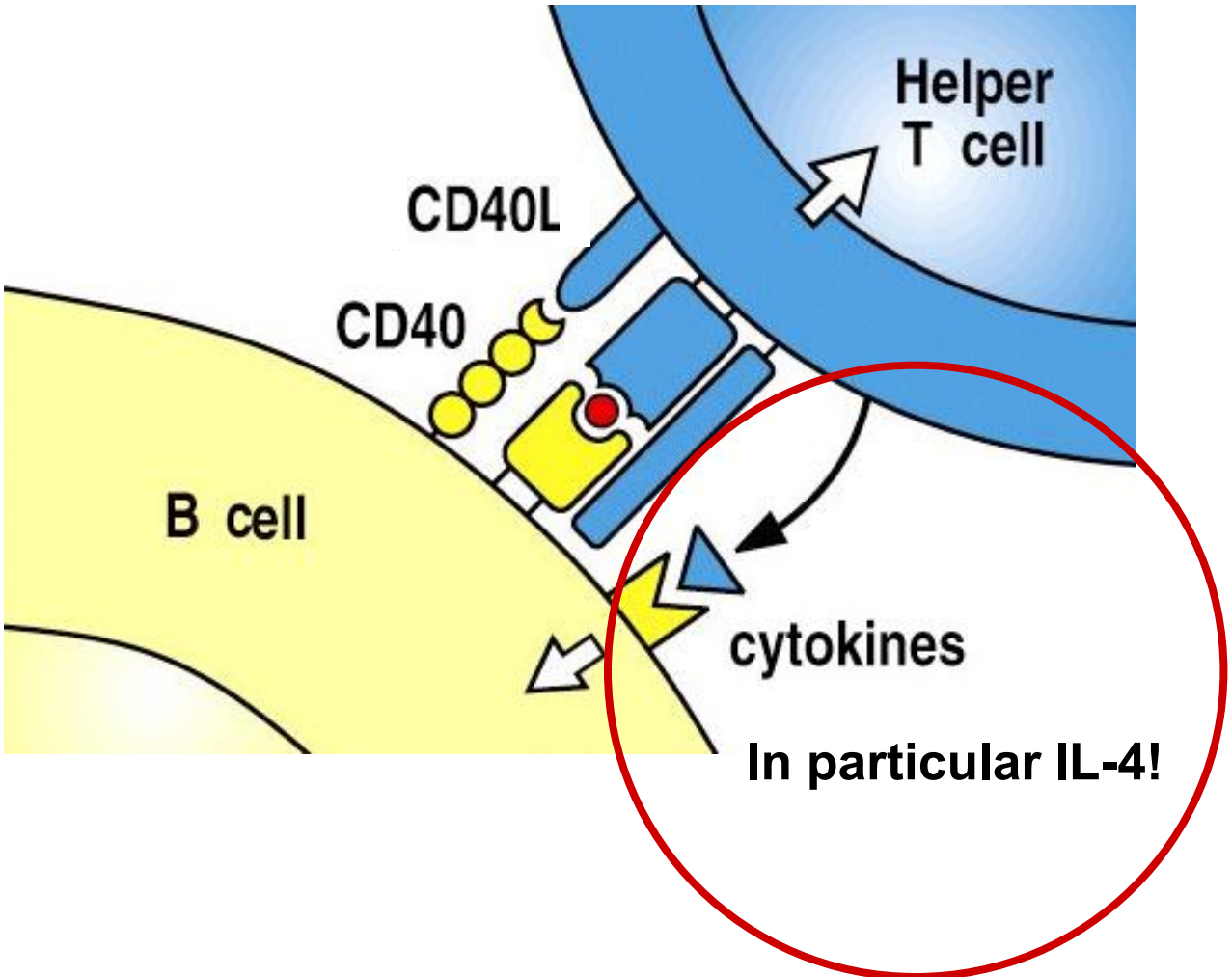


# Thymus-dependent-B-cell activation




A T-cell does not express co-stimulatory signals for B cells until the cell is **activated** by a **dendritic cell!**

# Thymus-dependent-B-cell activation



# Summary: TD-B-cell activation

## Signals for the B-cell activation:

1) Binding of an antigen to the BCR
   (B-cell receptor = membranous immunoglobulin)

2) Signals given by a T-cell, which recognised the MHC-II-presented peptide via TCR and the co-receptor CD4:

   - Stimulation of CD40 (B-cell) by CD40-ligand (T-cell)
   - Cytokines

# TD-B-cell activation

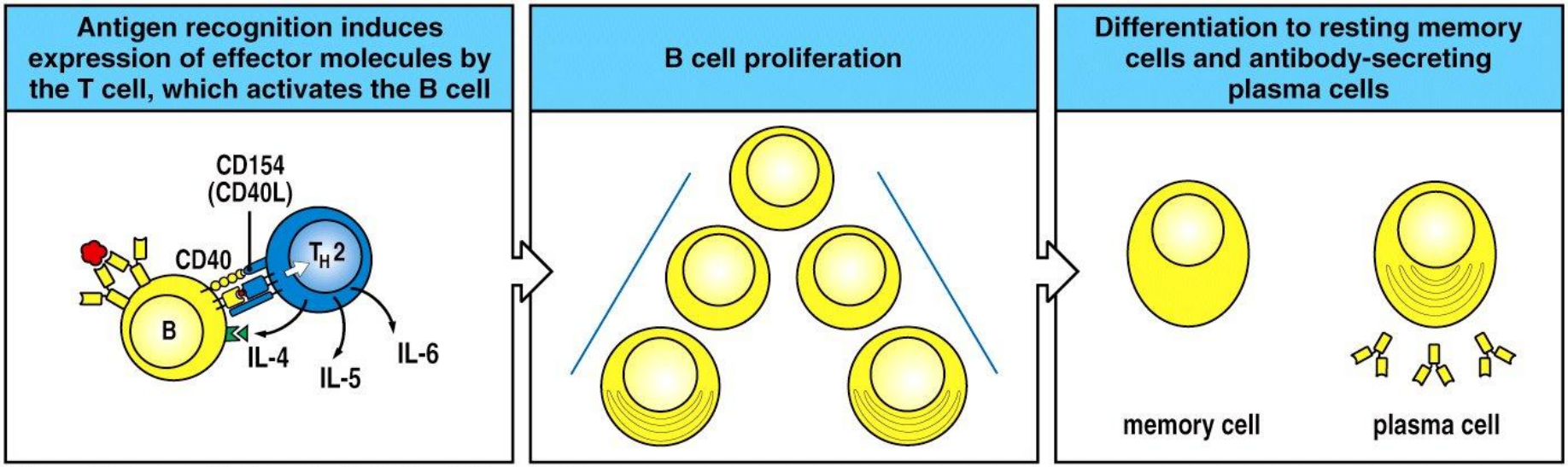



Figure 9-5 Immunobiology, 6/e. (© Garland Science 2005)

Function of the effector cytokines:

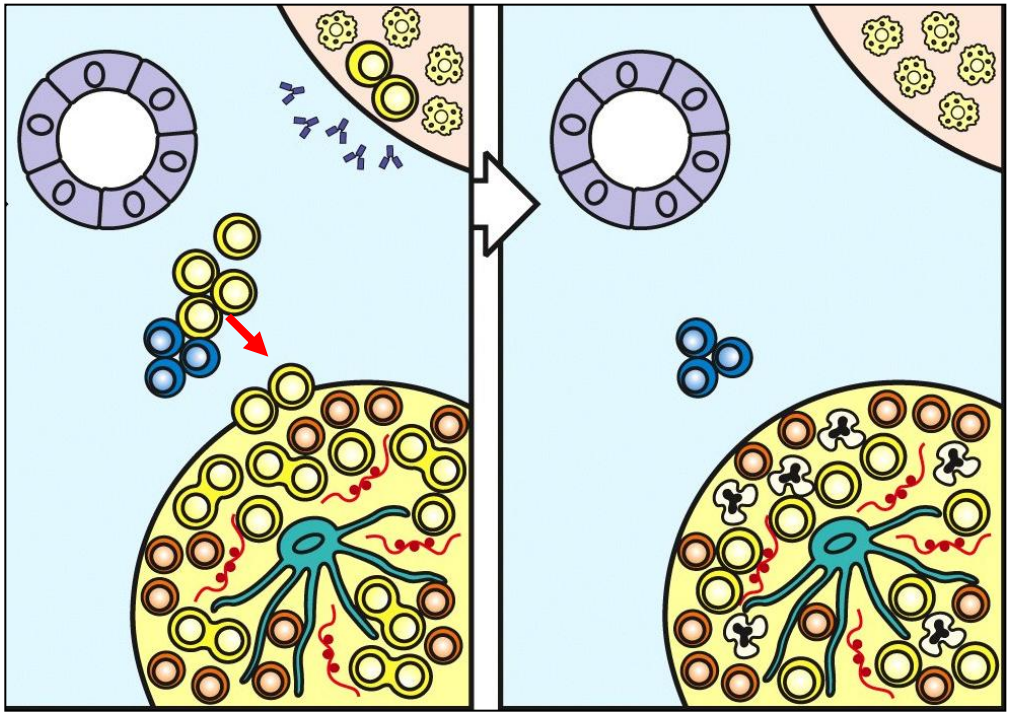
- Proliferation of the B-cell
- Isotype-switch

After activation B cells develop to:

- B-memory cells
- Antibody-secreting plasma cells

# Affinity maturation in the germinal center

Some B- and T-cells leave the primary proliferation foci and move in a **germinal centres**. The B-cells proliferate quickly (**centroblasts**) and pass through the **somatic hyper mutation**.



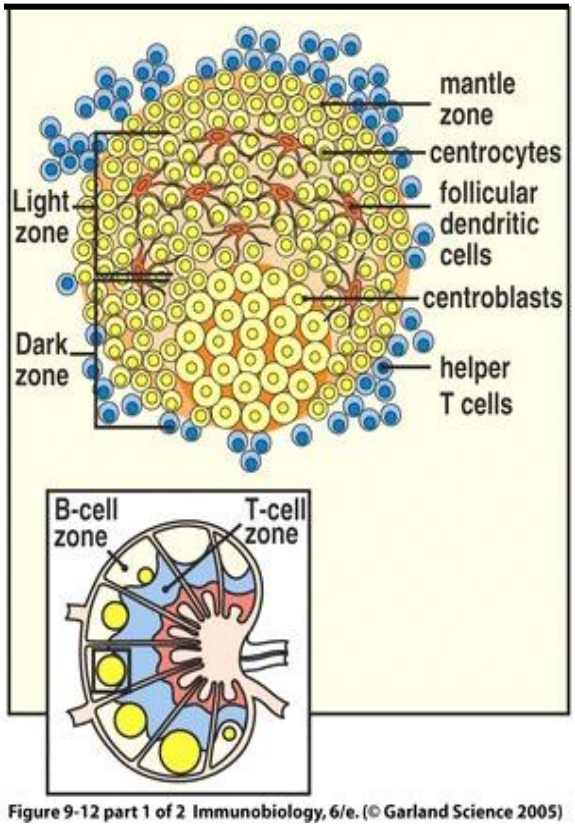
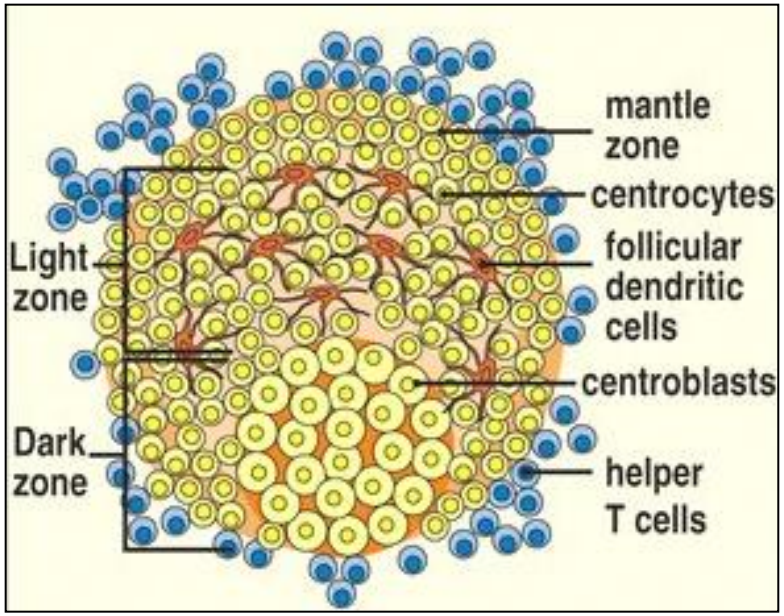



Figure 9-12 part 1 of 2 Immunobiology, 6/e. (© Garland Science 2005)

# FDCs play an important role in affinity maturation

1) FDC = Follicular dendritic cells



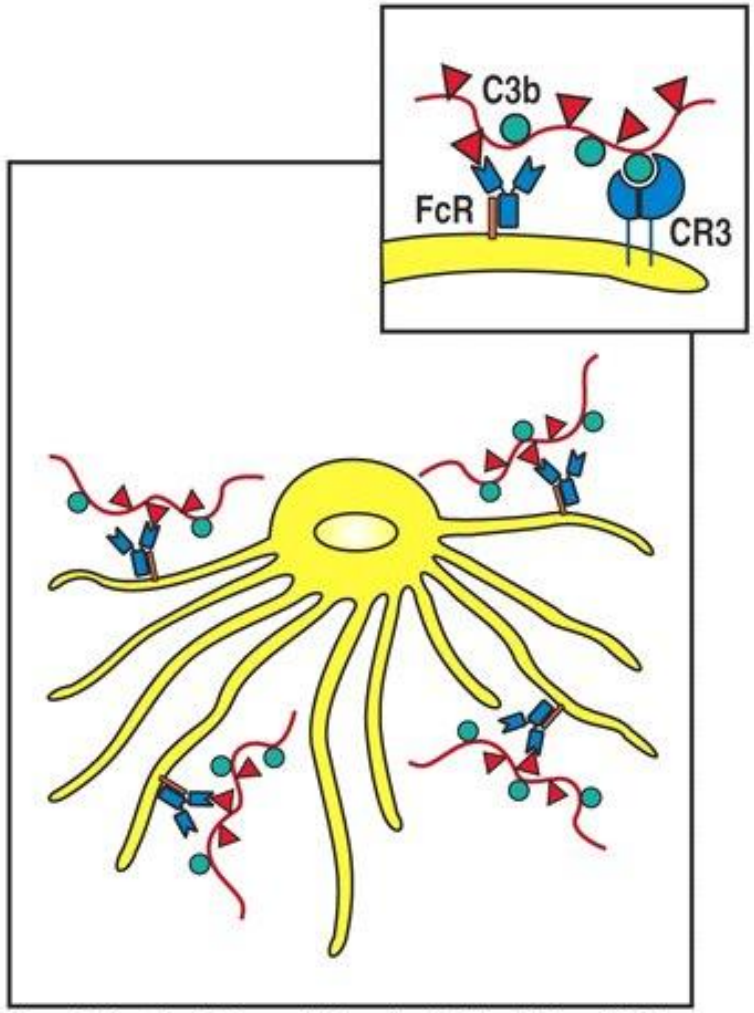



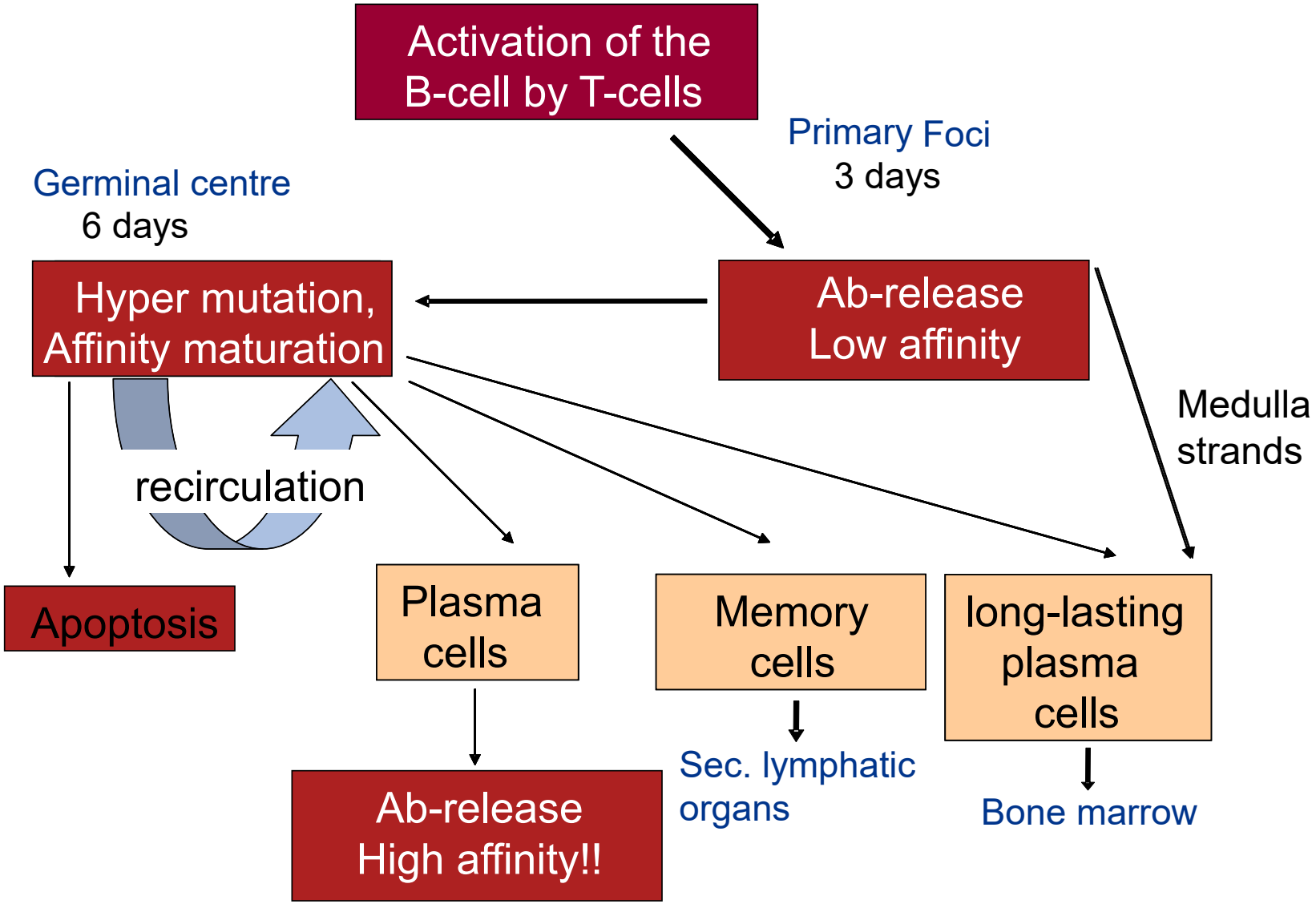

Figure 9-14 part 2 of 2 Immunobiology, 6/e. (© Garland Science 2005)

- FDCs are found in the **germinal centres** and attract B-cells by the release of **chemokines**.
- FDCs present antigens **not on MHC** but by immune complexes bound to **Fc- and complement receptors**.

# Tfh cells play an important role in affinity maturation


Follicular T helper cells (Tfh)

# Summary: Life cycle of mature B-lymphocytes



# Immunological memory

**Memory cells**

B-memory cells (Memory B-cells) circulate.

They need stimulation by an **antigen** for **antibody production** and divide continuously in low frequency.

**long-lasting Plasma cells**

Long-lasting plasma cells occur after the **second antigen contact**

They move into the **bone marrow** (important for their survival).

They need **no antigen** for **antibody production**.

They survive for years/decades.