2 Phases of B-Cell Development at 2 Locations

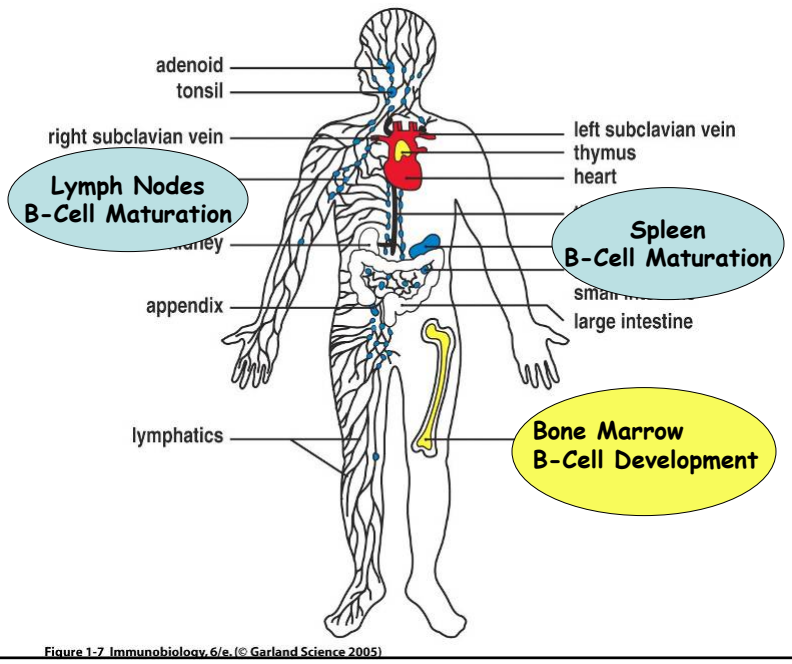


Figure 1-7 Immunobiology, 6/e, (© Garland Science 2005)

Antigen-Independent B-Cell Development

Bone Marrow

1. DNA rearrangements establish the primary repertoire, creating *diversity*

2. Allelic exclusion ensures that each clone expresses a single antibody on the surface, establishing *specificity*

3. Deletion of self-reactive clones establishes *tolerance*

Bone Marrow Stromal Cells Support Early B Lymphopoiesis

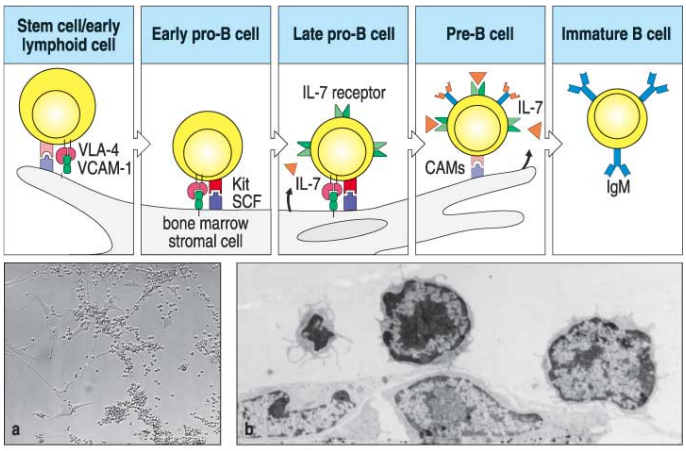
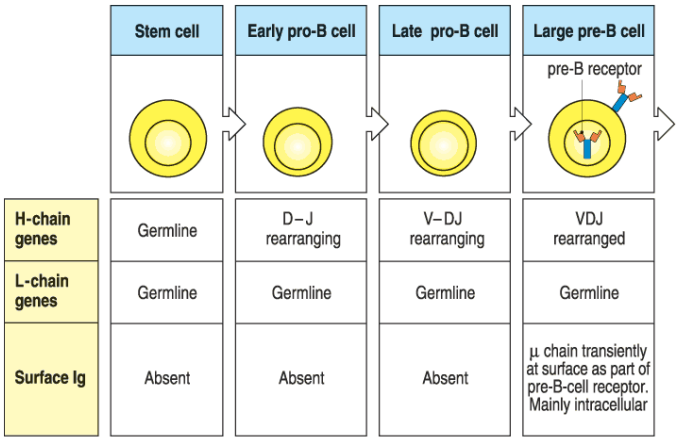


Fig 7.3 © 2001 Garland Science

Ordered Rearrangement of Ig Genes During B-Cell Development in the Bone Marrow

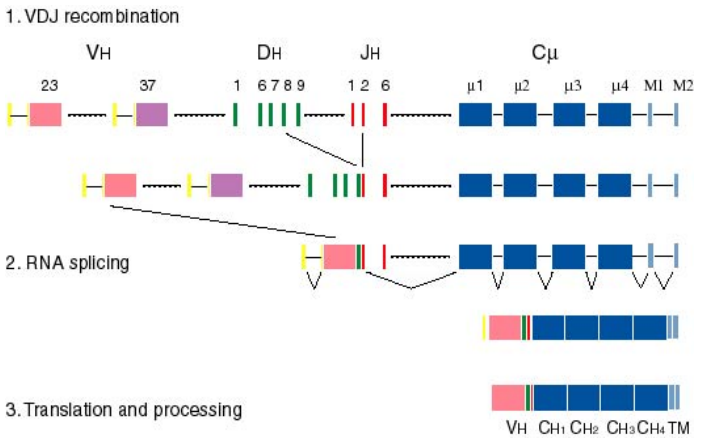


	Stem cell	Early pro-B cell	Late pro-B cell	Large pre-B cell
H-chain genes	Germline	D–J rearranging	V–DJ rearranging	VDJ rearranged
L-chain genes	Germline	Germline	Germline	Germline
Surface Ig	Absent	Absent	Absent	μ chain transiently at surface as part of pre-B-cell receptor. Mainly intracellular

Fig 7.5 part 1 of 2 © 2001 Garland Science

pBCR (Mu heavy chain + surrogate light chains) **signals**

Ig Heavy Chain VDJ Recombination--2 DNA Deletions, DJ and VD



Heavy chain rearrangement occurs first:

DJ on both alleles

V-DJ on one allele

Productive rearrangement (1/9)

Non-productive rearr. (8/9)

preBCR (Mu and surrogate L.C.)

V-DJ on second allele

1. STOP H.C.rearrangement
2. Proliferation
3. Begin L.C. rearrangement

pBCR

Non-prod.

DEATH

(H.C. Alleles)	PR D-J	NPR PR	NPR NPR

Light Chain Rearrangement: 4 possible alleles, each with 1/3 chance of a productive rearrangement

Kappa usually precedes lambda

Productive rearrangement produces IgM and the B CELL RECEPTOR on the surface

STOP further L.C. rearrangement

κ	PR,GL	NPR,PR	NPR,NPR	NPR,NPR	NPR,NPR
λ	GL,GL	GL,GL	PR,GL	NPR,PR	NPR,NPR
	↓				↓
	CONTINUE DEVELOPMENT				DEATH

Checkpoints which confer allelic exclusion

pBCR

BCR

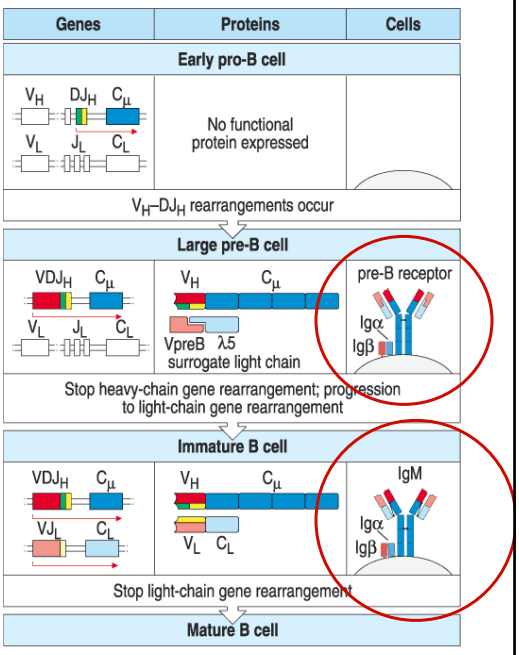


Fig 7.15 © 2001 Garland Science

THE B CELL RECEPTOR

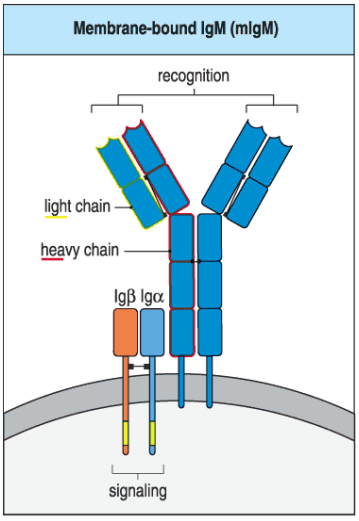


Fig 6.7 © 2001 Garland Science

Bound antigen gives signals to the B cell to proliferate and differentiate.

Signalling from the pBCR and BCR

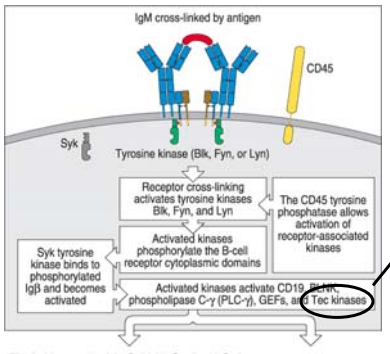


Fig 6.14 part 1 of 2 © 2001 Garland Science

Lack of Brutons' Tyrosine Kinase causes Bruton's XLA (X-linked agammaglobulinemia)

B cell development is blocked at the preB stage, due to failure of pBCR. No B cells, no antibodies, causing recurrent bacterial and viral infections.

CENTRAL B-CELL TOLERANCE: EDITING AND CLONAL DELETION (in Bone Marrow)

IgM on B Cell Surface

Recognition of self

L.C. editing

Recognition of self

DEATH

No self recognition

1. Proliferation
2. Maturation
3. Exit to periphery

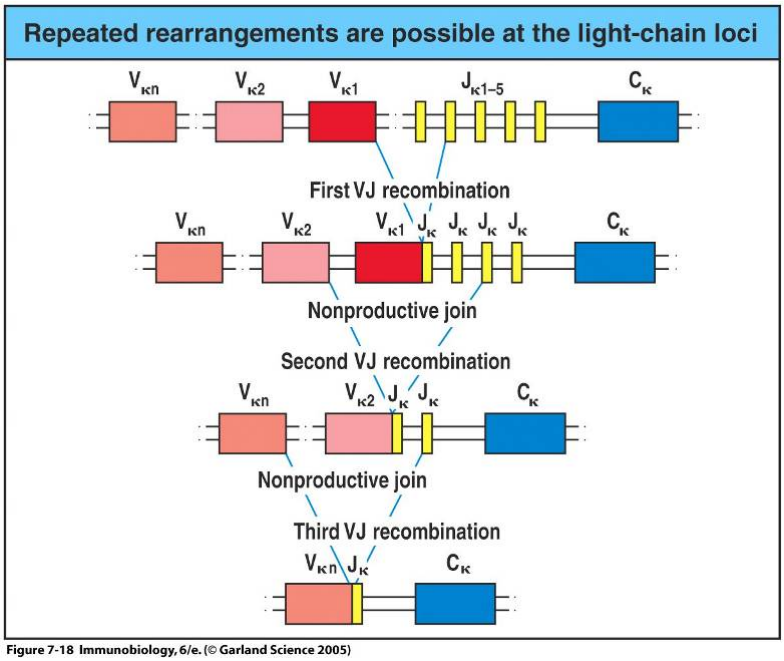


Figure 7-18 Immunobiology, 6/e. (© Garland Science 2005)

Antigen-Independent B-Cell Development

Bone Marrow

1. DNA rearrangements establish the primary repertoire, creating *diversity*

2. Allelic exclusion ensures that each clone expresses a single antibody on the surface, establishing *specificity*

3. Deletion and editing of self-reactive clones establishes *tolerance*

Antigen-Dependent B Cell Maturation

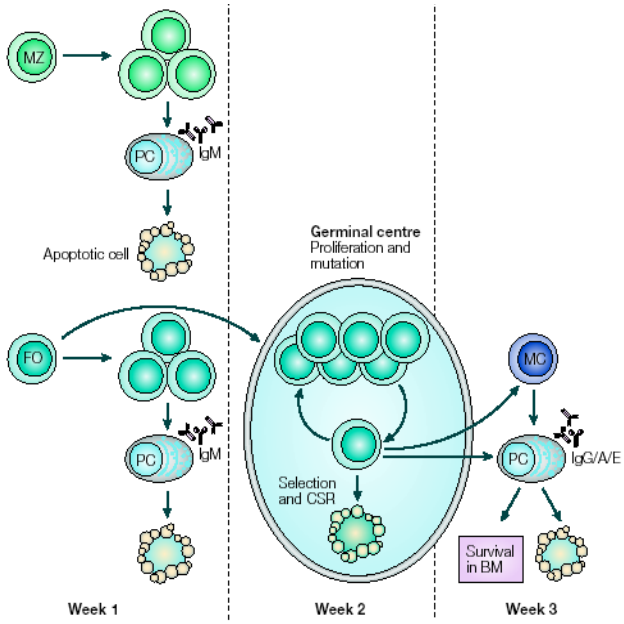
In Periphery (spleen and LN)

Antigen and T_H cells give B cells two signals:
1) proliferate 2) differentiate

T-cell dependent responses are refined two ways: 1) higher affinity antibodies
2) IgG/A/E ("switched") isotypes

Two products of B cell development:
1) plasma cells secrete Ig (final effector)
2) memory cells respond to II° antigen

T-DEPENDENT B-CELL MATURATION IN GERMINAL CENTERS



B Cell Activation By T-Cell Dependent And T-Cell Independent Antigens

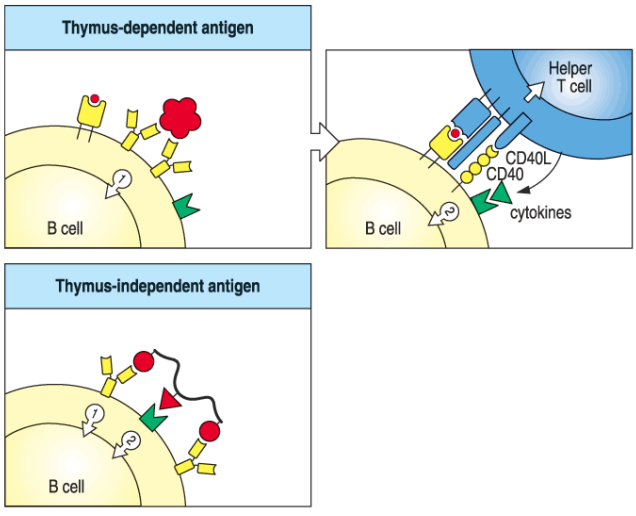
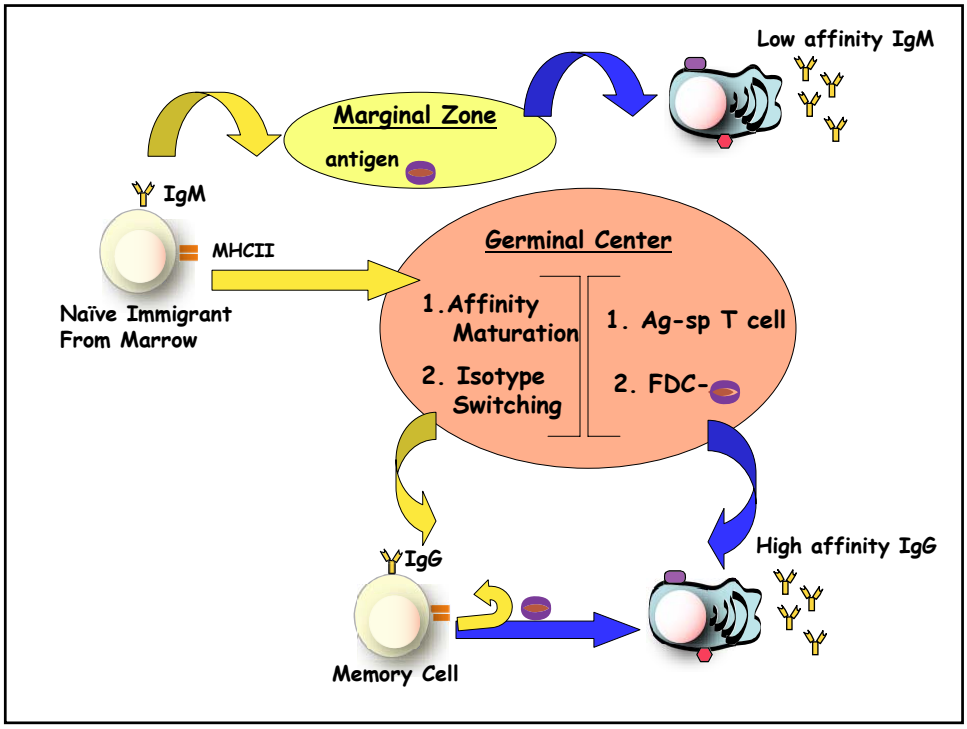


Fig 9.2 © 2001 Garland Science



T Cell Help Is Required for GC Reactions

(B cells signal T cells by presenting Ag in association with MHC II)

T cells provide 2 kinds of help to B cells:

1. Cell-cell signals from CD40L/CD40 and other surface molecules.

2. Secreted cytokines

The Germinal Center

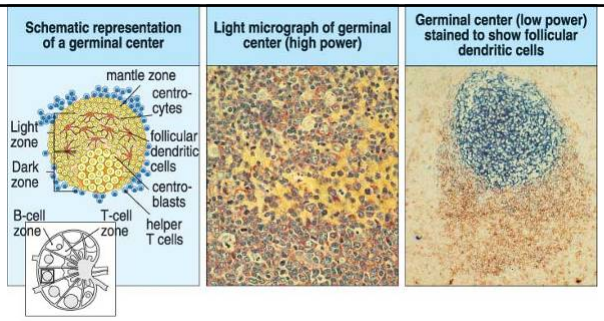


Fig 9.12 © 2001 Garland Science

1. Affinity maturation
   a. Somatic hypermutation-requires AID
   b. Selection for high affinity clones
2. Isotype switch recombination-requires AID
3. Peripheral tolerance
4. Final maturation to memory or plasma cell.

AFFINITY MATURATION IN THE GC

Proliferation + Somatic Hypermutation — Dark zone

(Iterative cycles)

Ag(FDC) + T cell help → SURVIVAL/ SELECTION — Light zone

T help and no Ag binding
(eliminates *low affinity* clones)

or → DEATH

Ag binding and no T help
(eliminates *self-reactive* clones, giving tolerance)

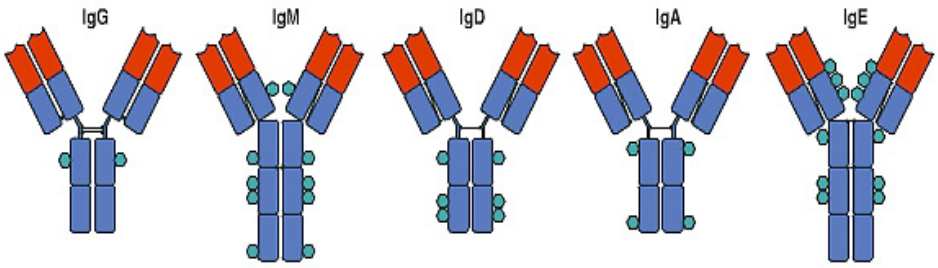
SELECTIVE SURVIVAL IN GC

Requires: a.High affinity surface Ig
b. Ag-specific T cell help, esp. via CD40/CD40L

1. Selects clones producing high affinity antibody--i.e.affinity maturation

2. Eliminates self-reactive clones--peripheral tolerance.

Switching to Different Ig Isotypes



Ig Isotypes Have Different Functions and Distributions

Functional activity	IgM	IgD	IgG1	IgG2	IgG3	IgG4	IgA	IgE
Neutralization	+	–	++	++	++	++	++	–
Opsonization	–	–	+++	*	++	+	+	–
Sensitization for killing by NK cells	–	–	++	–	++	–	–	–
Sensitization of mast cells	–	–	+	–	+	–	–	+++
Activates complement system	+++	–	++	+	+++	–	+	–

Distribution	IgM	IgD	IgG1	IgG2	IgG3	IgG4	IgA	IgE
Transport across epithelium	+	–	–	–	–	–	+++ (dimer)	–
Transport across placenta	–	–	+++	+	++	+/–	–	–
Diffusion into extravascular sites	+/–	–	+++	+++	+++	+++	++ (monomer)	+
Mean serum level (mg ml^{-1})	1.5	0.04	9	3	1	0.5	2.1	$3x10^{-5}$

Fig 9.19 © 2001 Garland Science

mRNA Splicing

DNA rearrangement: CSR

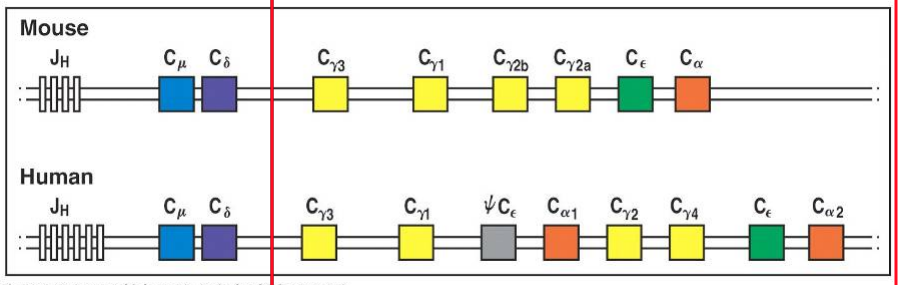
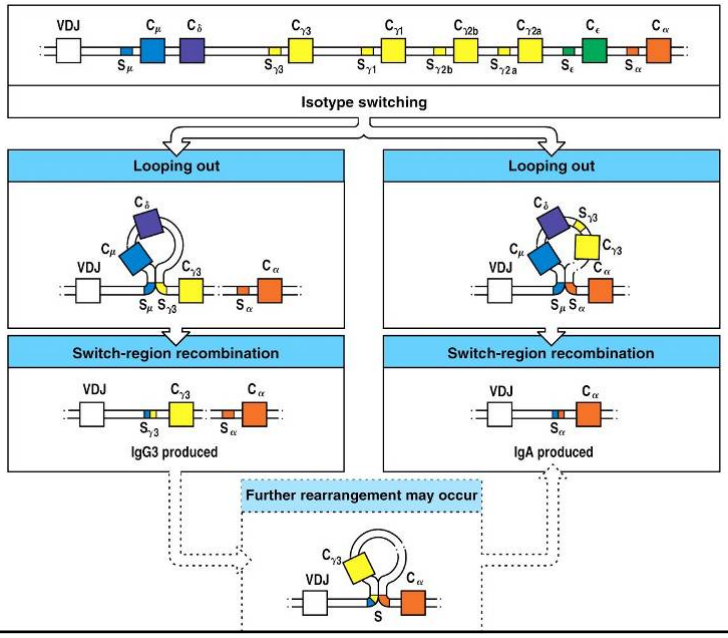


Figure 4-19 Immunobiology, 6/e. (© Garland Science 2005)

CSR Involves DNA Deletion and Loss



"Germline" (I region) Transcripts Are Necessary For Isotype Switch Recombination

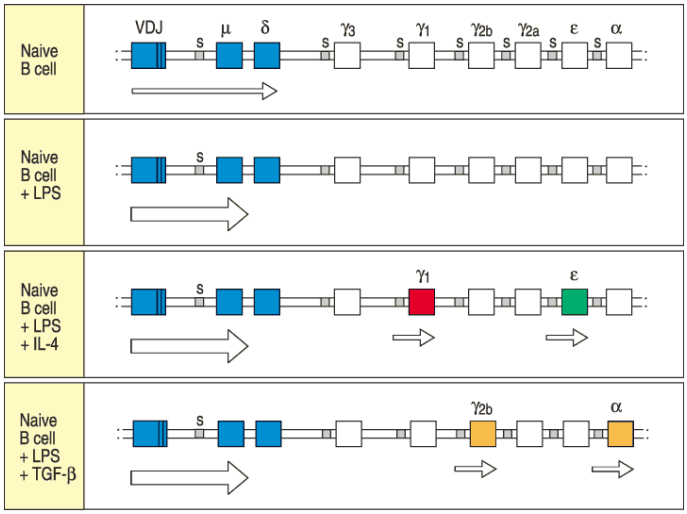


Fig 9.8 © 2001 Garland Science

T Cell Cytokines Instruct Choice of Isotype

	Role of cytokines in regulating Ig isotype expression						
Cytokines	IgM	IgG3	IgG1	IgG2b	IgG2a	IgE	IgA
IL-4	Inhibits	Inhibits	Induces		Inhibits	Induces	
IL-5							Augments production
IFN-γ	Inhibits	Induces	Inhibits		Induces	Inhibits	
TGF-β	Inhibits	Inhibits		Induces			Induces

Figure 9-7 Immunobiology, 6/e. (© Garland Science 2005)

T cell secretes cytokines

↓

Specific I region transcription

↓

Isotype switch recombination to specific C_H gene segment

TGFβ

↓

IαSαCαRNA

↓

Cut and join Sμ and Sα DNA

↓

VDJCα mRNA

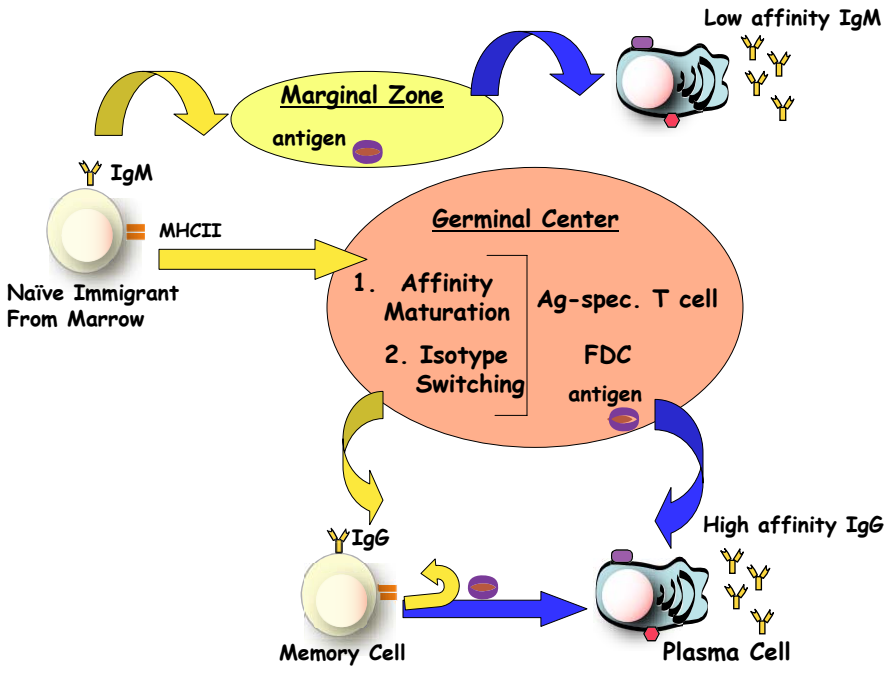
↓

IgA

Hyper IgM Syndrome

(Increased susceptibility to specific pathogens)

1. Mutations in CD40L
2. Mutations in CD40
3. Mutations in AID (or repair enzymes downstream of AID)
4. One or more other genes defined by human disease remain to be identified!



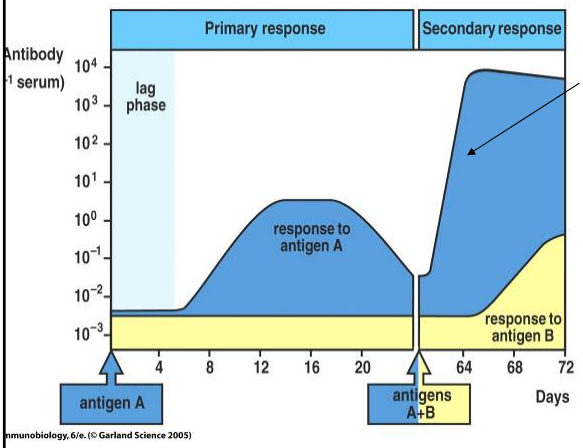
1. Memory B cells

Surface Ig, usually IgG
High affinity for antigen
Long-lived, even in the absence of antigen
Respond rapidly to secondary stimulation

2. Plasma Cells-final B cell effectors

Secrete copious amounts of Ig, no surface Ig
Non-dividing
Some are short-lived, some become long-lived in the bone marrow

MEMORY B CELLS and a MEMORY RESPONSE



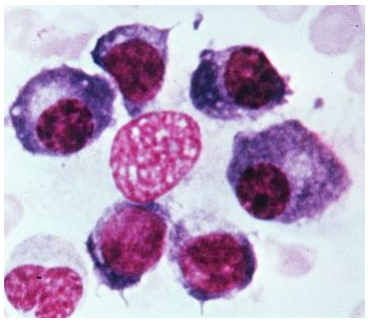
MEMORY CELLS

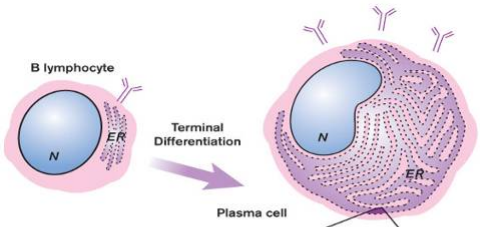
1. Memory cells are post GC
   a. High affinity
   b. Switched isotype

2. Memory cells differentiate into plasma cells rapidly

3. Long-lived in absence of antigen.

Plasma Cells-Final B-Cell Effectors





1. One job: secrete antibody
2. Terminally differentiated, post-mitotic
3. Limited half-life

ANTIGEN-DEPENDENT B CELL MATURATION

1. Occurs in periphery (spleen, lymph nodes)
2. Antigen selects specific clones for proliferation and maturation.
3. Bacterial polysaccharides are T-cell independent activators of B cells.
4. Protein antigens require T cells to help B cells mature.
5. T cells and B cells communicate
   - -B cells process antigen and present peptide-MHC to T cells, which stimulates the T cells.
   - -T cells provide cell-cell signals via CD40L/CD40
   - -T cells provide soluble cyotkine signals

6. T-cell dependent B cell maturation occurs in Germinal Centers
7. Affinity maturation in GCs results from somatic hypermutation + selection for high antigen-binding affinity
8. Class switch recombination occurs in GCs
9. Deletion of self-reactive clones provides peripheral tolerance.
10. Memory B cells and plasma cells emerge from the GC reaction.