

Acquired alterations of IG and TCR loci in lymphoproliferative disorders

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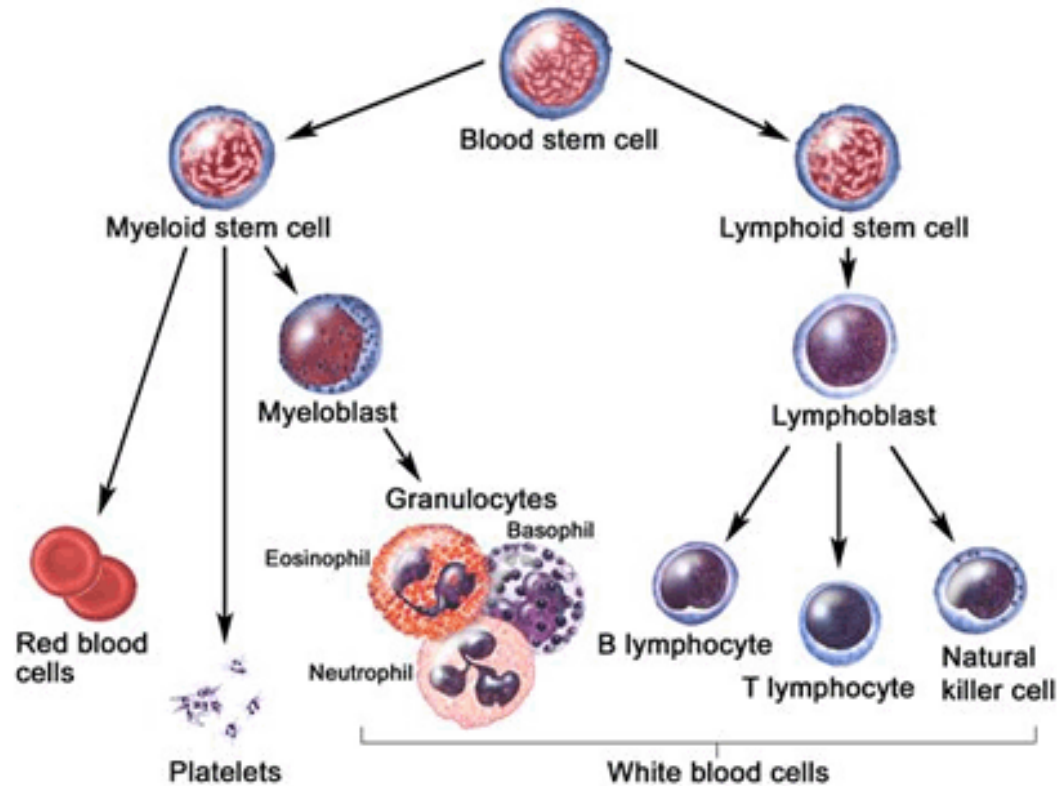
Overview

- What are lymphoproliferative disorders?
- What are 'IG' and 'TCR'?
- What are IG/TCR alterations?
- How to detect IG/TCR alterations?

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Blood cell development



major types
of lymphocytes

Blood cell development: A blood stem cell goes through several steps to become a red blood cell, platelet, or white blood cell.

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Three major types of lymphocytes

- B cells
- T cells
- Natural killer (NK) cells

B, T, and NK lineage of lymphoid malignancies

Different lymphoid malignancies of different lineage origin

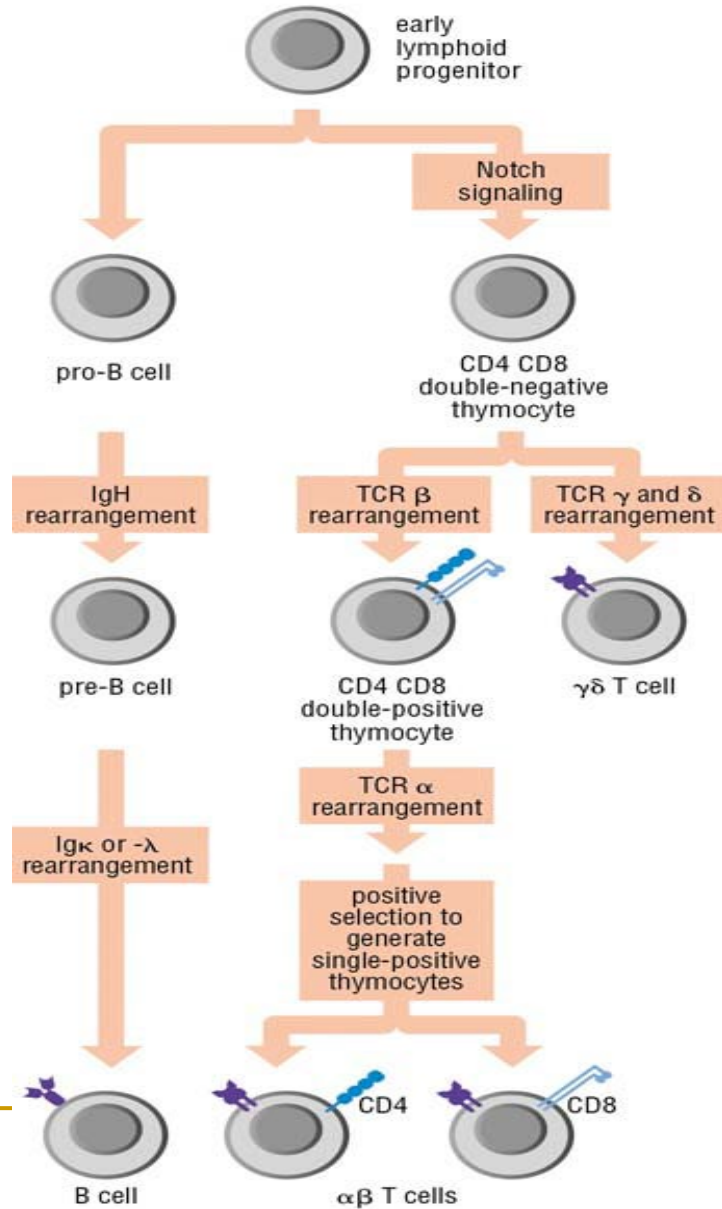
Lymphoid malignancy	B lineage	T lineage	NK lineage
Acute lymphoblastic leukemia – children – adults	82 – 86% 75 – 80%	14 – 18% 20 – 25%	< 1% < 1%
Chronic lymphocytic leukemias	95 – 97%	3 – 5%	1 – 2%
Non-Hodgkin lymphomas – nodal NHL – extranodal NHL – cutaneous NHL	95 – 97% 90 – 95% 30 – 40%	3 – 5% 5 – 10% 60 – 70%	< 2% < 2% < 2%
Multiple myeloma	100%	0%	0%

Function of B and T cells

- B cells are primary responsible for humoral immunity (relating to antibodies)
- T cells are involved in cell-mediated immunity.

B-cells

- Millions of different types of B cells each day
- In the blood and lymphatic system role of immune surveillance
- No production of antibodies until fully activated



Gene rearrangements of the antigen receptor genes occur during the lymphoid proliferation

Lymphoproliferative disorders (LPDs)

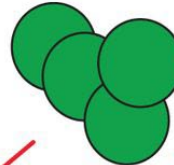
LPDs refer to several conditions in which lymphocytes are produced in excessive quantities.

- **Chronic lymphocytic leukemia** (most frequent) 25%
- Acute lymphoblastic leukemia
- Hairy cell leukemia
- lymphomas
- Multiple myeloma
- Waldenstrom's macroglobulinemia
- Wiskott-Aldrich syndrome
- Post-transplant lymphoproliferative disorder
- Autoimmune lymphoproliferative syndrome (ALPS)
- 'Lymphoid interstitial pneumonia'

pro B
heavy chain
rearrangement



pre B1
heavy chain
selection



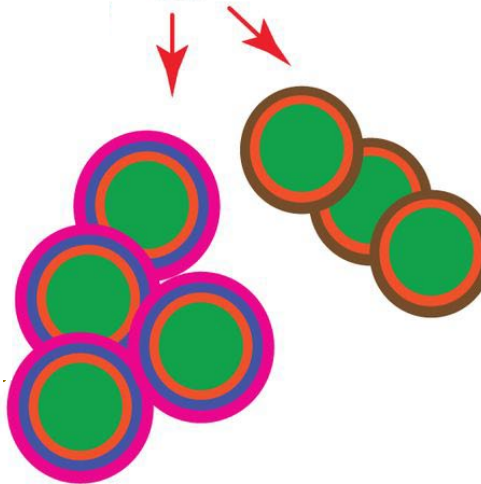
pre B2
light chain
rearrangement



naive B cell
receptor
selection



GC B cell
proliferation,
mutation,
selection



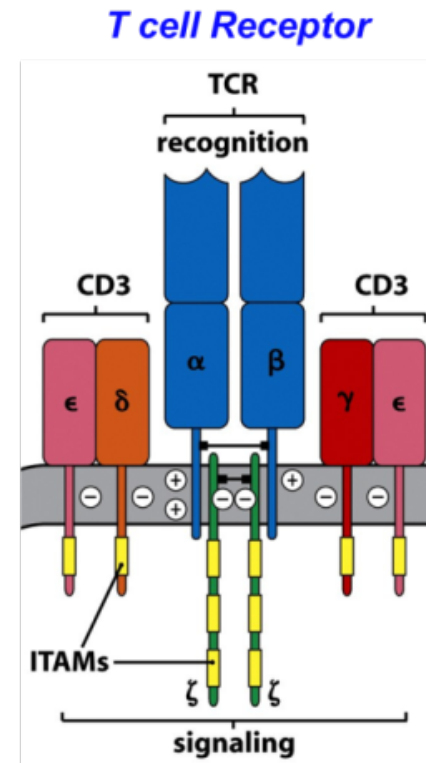
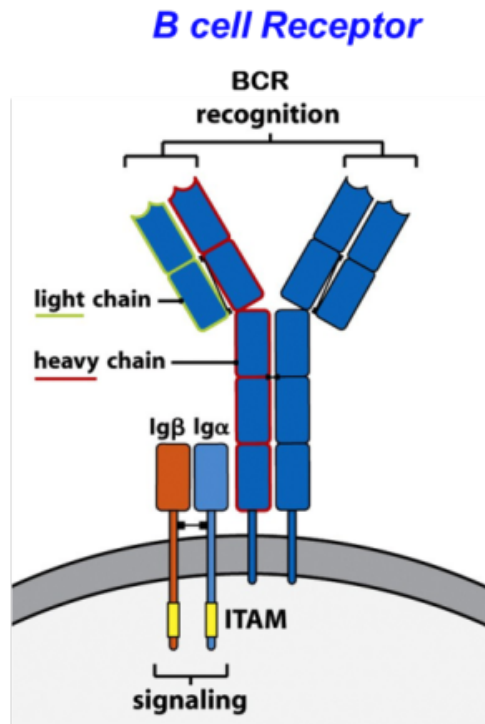
memory B cell
proliferation, mutation,
selection and class switch

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- What are IG/TCR alterations?
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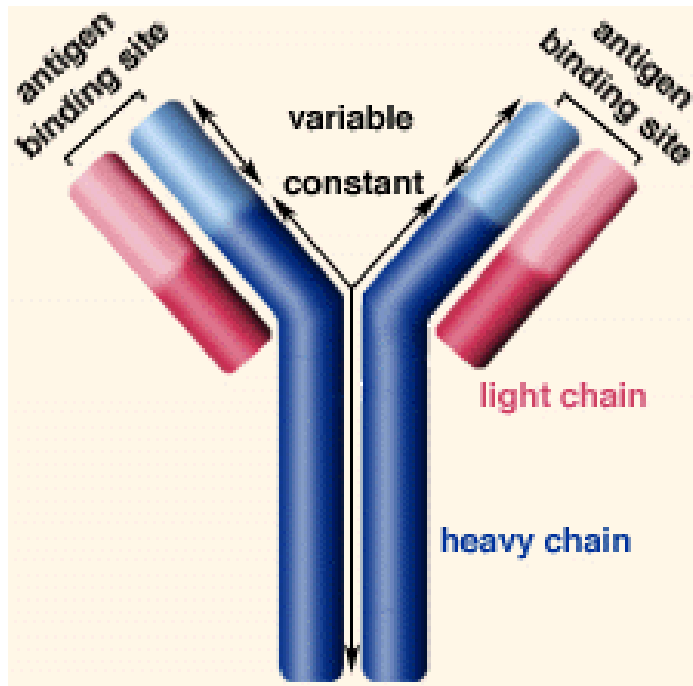
B-cell and T-cell receptors

T Cell and B Cell Antigen Receptors (TCR and BCR)



The variable domain of both the TCR α -chain and β -chain have three hypervariable regions

The ability to produce billions of different antibodies in humans results from the production of variable regions of light and heavy antibody genes by DNA rearrangement.

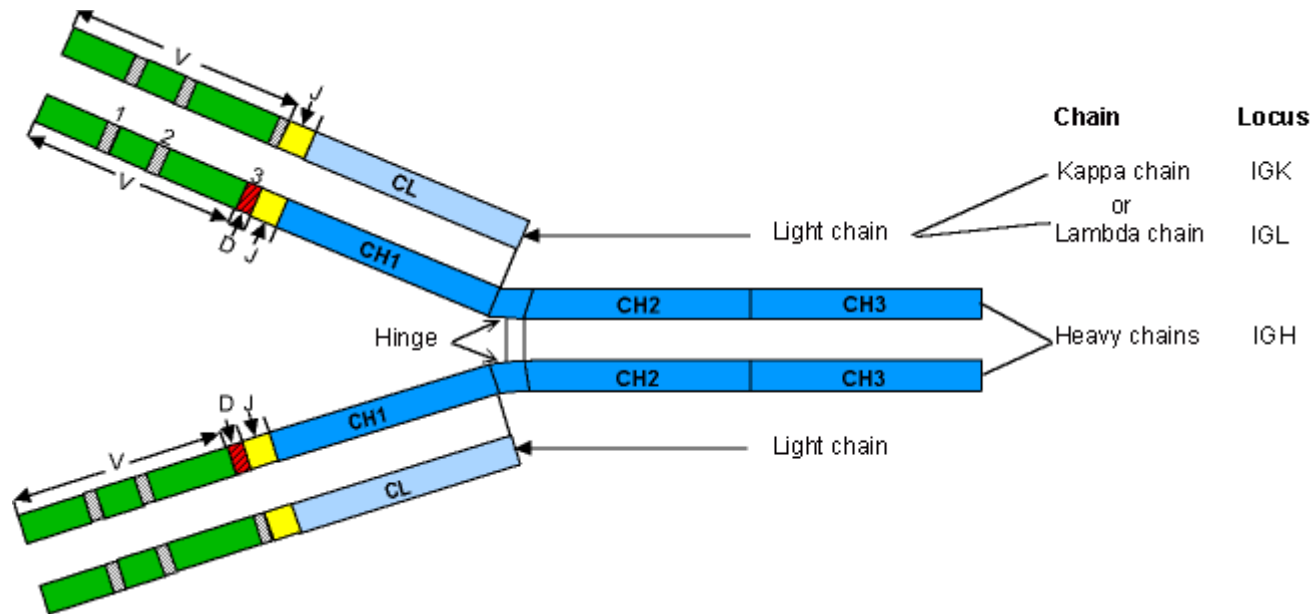


<http://www.biology.arizona.edu>

Heavy chain (IGH)
Light chain (IGL)

Ig molecules are composed of
4 polypeptide chains:
2 identical heavy chains
2 identical light chains
Ig protein consists of 2 segments
C and V regions

Schematic representation of an Immunoglobulin (IG)



Overview

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Cell development.

Stage	Heavy chain	Light chain
Progenitor (or pre-pro) B cells	germline	germline
Early Pro (or pre-pre)-B cells	undergoes D-J rearrangement	germline
Late Pro (or pre-pre)-B cells	undergoes V-DJ rearrangement	germline
Large Pre-B-cells	is VDJ rearranged	germline
Small Pre-B cells	is VDJ rearranged	undergoes V-J rearrangement
Immature B cells	is VDJ rearranged	VJ rearranged
Mature B cells	is VDJ rearranged	VJ rearranged

B cell development

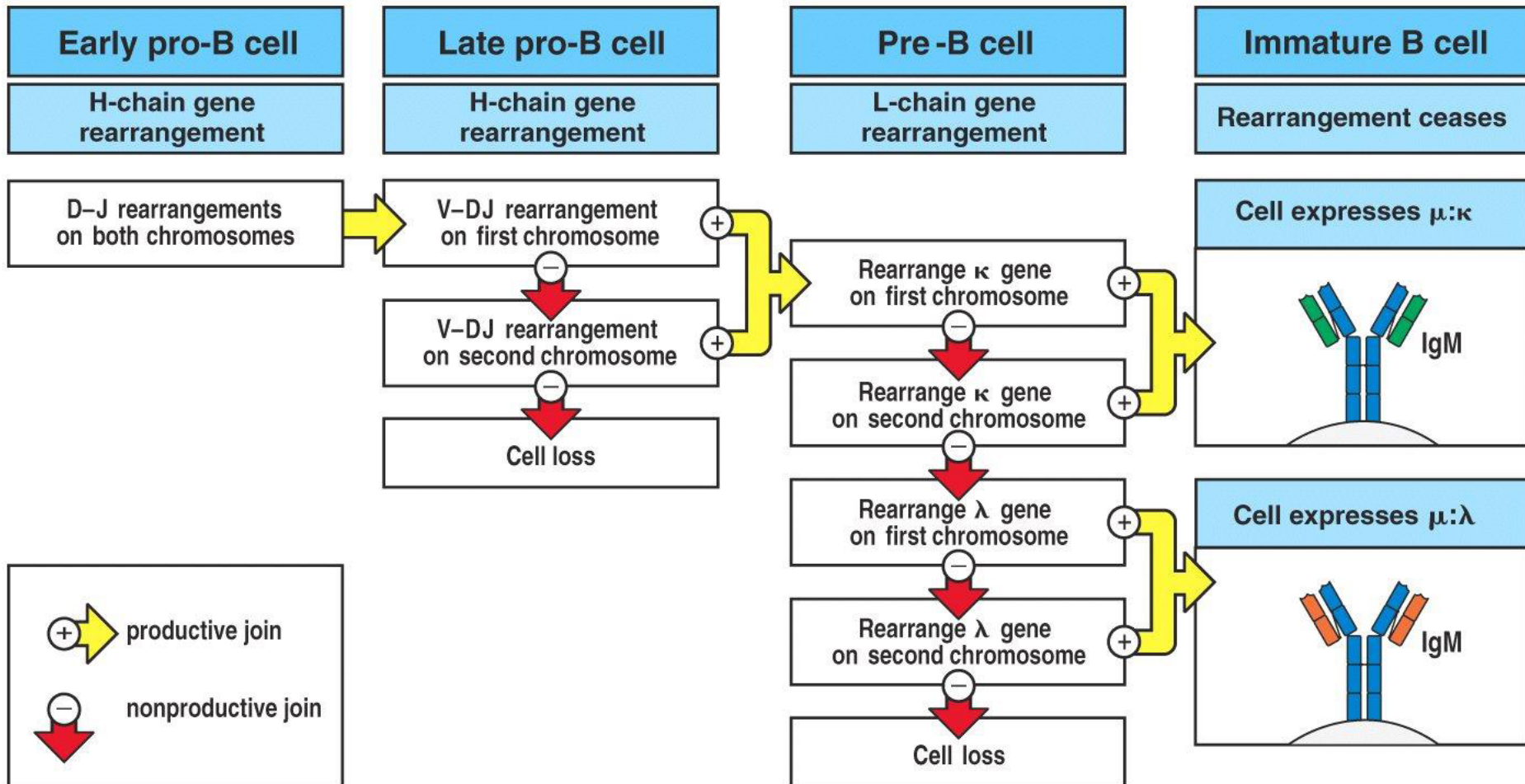
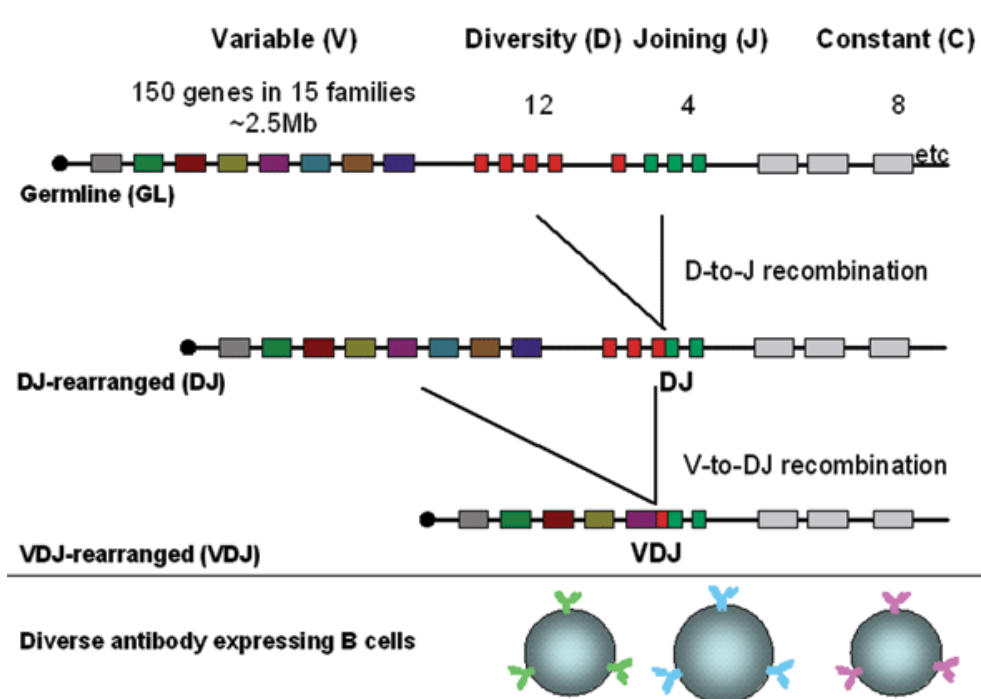


Figure 7-15 Immunobiology, 6/e. (© Garland Science 2005)

B-cells: Stepwise rearrangement of V, D, and J gene segments

The production of variable regions of heavy antibody genes by DNA rearrangement.

stepwise rearrangement of V, D, and J gene segments

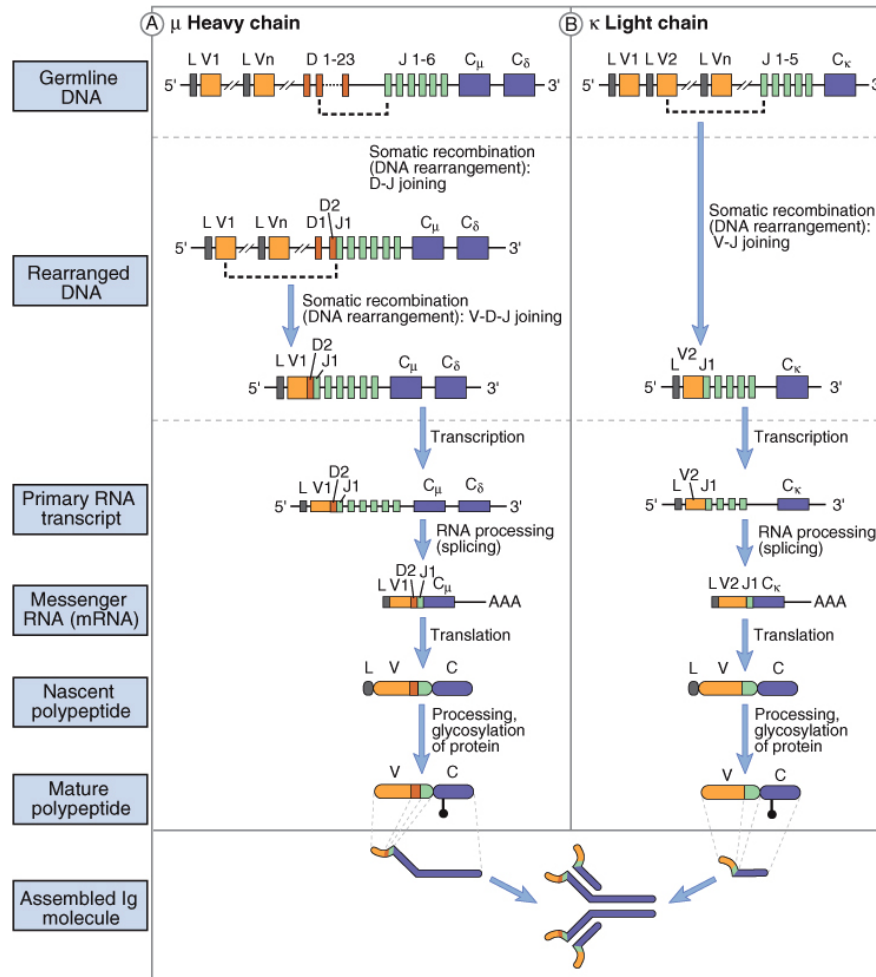


Genes encoding antigen receptors are unique:
-high diversity
-developing lymphocytes through V(D)J rearrangement.

Gene rearrangements of the antigen receptor genes occur during the lymphoid proliferation

These V-J gene rearrangements generate products that are unique in length and sequence in each cell resulting in diverse antigen expressing B cells.

The production of variable regions of light and heavy antibody genes by DNA rearrangement.



Estimated diversity of human Ig and TCR molecules

	IgH	Igα	Igλ	TCR α	β	γ	δ
		molecules		molecules			
Number gene segments							
V gene segments	~44	~43	~38	~46	~47	~6	~6
D gene segments	27				2		3
J gene segments	6	5	4	53	13	5	4
Combination diversity	$>2 \times 10^6$			2×10^6	<5000		
Junctional diversity	++	±	±	+	++	++	++++
Total diversity	$>10^{12}$			$>10^{12}$	$>10^{12}$		

Unique products of V(D)J rearrangements resulting in diverse antigen expressing B-cells

Possible variation through recombining gene fragments?

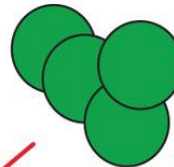
Over 15,000,000 combinations of variable, diversity and joining gene segments are possible.

Imprecise recombination and mutation increase the variability into billions of possible combinations.

pro B
heavy chain
rearrangement



pre B1
heavy chain
selection



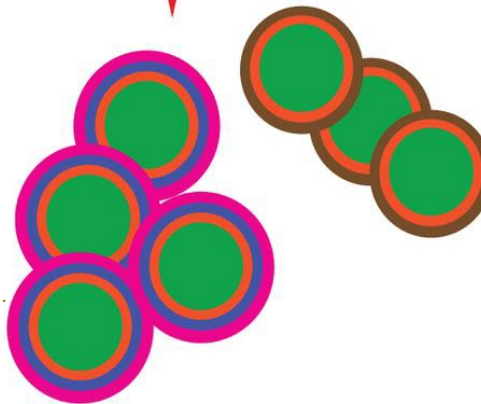
pre B2
light chain
rearrangement



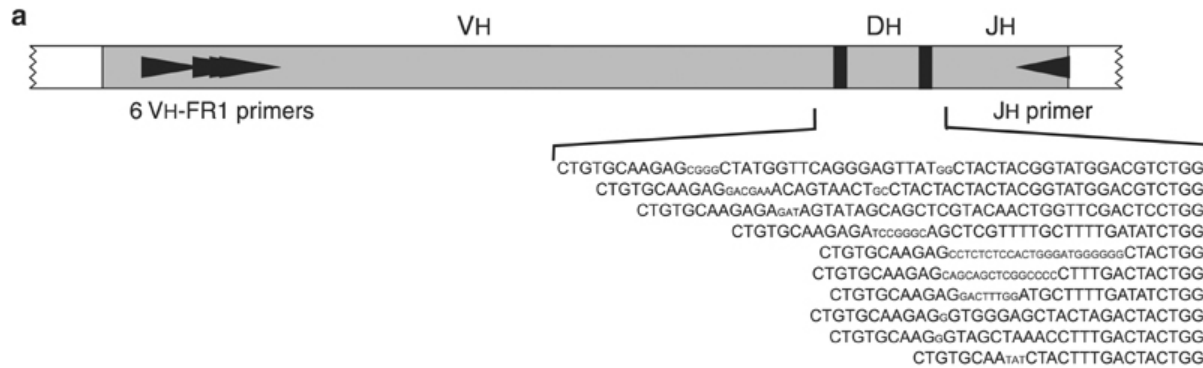
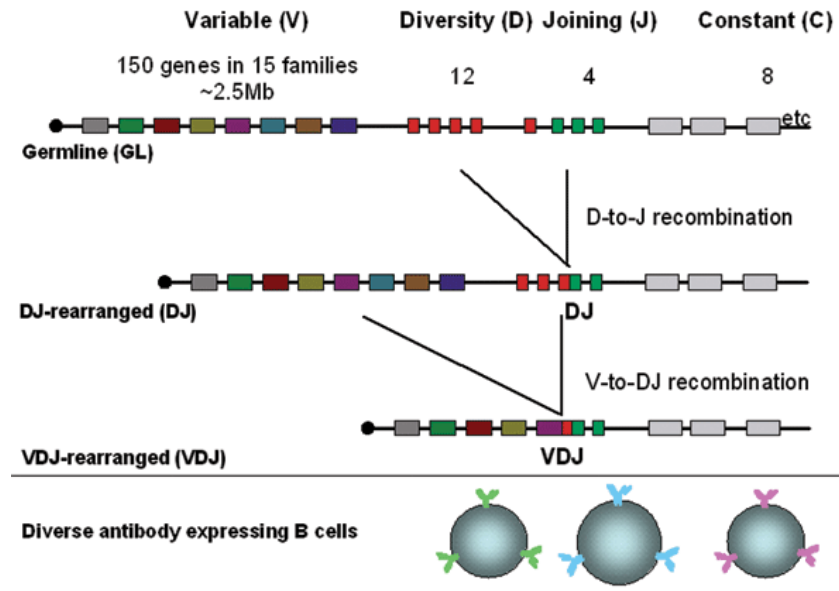
naive B cell
receptor
selection

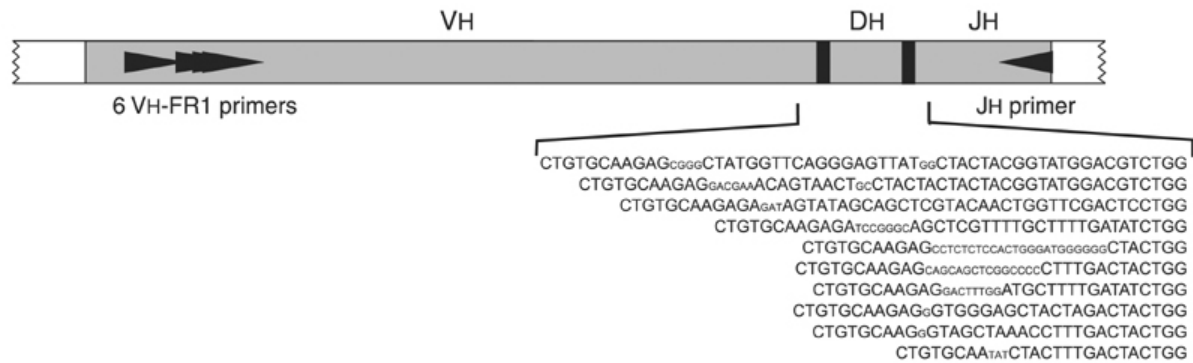


GC B cell
proliferation,
mutation,
selection

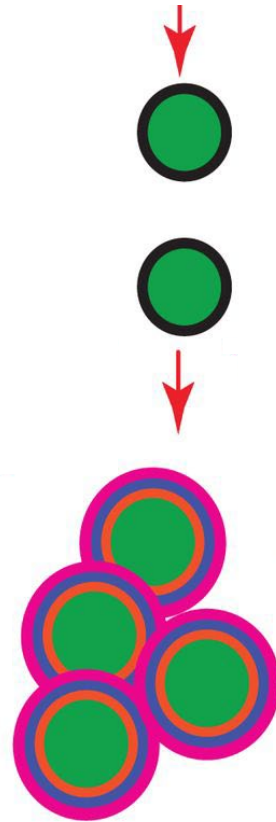


memory B cell
proliferation, mutation,
selection and class switch





- These V-J gene rearrangements generate products that are unique in length and sequence in each cell resulting in diverse antigen expressing B cells.



```

CTGTGCAAGAGCCTCTCTCCACTGGGATCGGGGCCCTTTGACTACTGG
CTGTGCAAGAGCCTCTCTCCACTGGGATCGGGGCCCTTTGACTACTGG
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CTGTGCAAGAGCCTCTCTCCACTGGGATCGGGGCCCTTTGACTACTGG
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Clinically relevant testing

- B-cell versus T-cell malignancy (origine)
- Reactive versus malignant
- New lymphoma versus recurrence
- Assessment of remission and relapse
- Bone marrow involvement
- Evaluation of treatment effectiveness

Analysis Techniques

- Detecting gene rearrangements of the antigen receptor genes
- Discrimination between clonal and polyclonal Ig/TCR gene products

Discrimination between clonal and polyclonal Ig/TCR gene PCR products

GeneScanning analysis

- ↳ Fast, accurate, sensitive, monitoring of clonal proliferations
- ↳ Need sequence equipment

Heteroduplex analysis

- ↳ Available to most laboratories
- ↳ Sensitivity ~5-10%

NGS (Next Generation Sequencing) analysis

- ↳ Accurate, easy interpretation, very sensitive, quantitative monitoring of clonal proliferations possible
- ↳ Need NGS equipment, expensive

Design of novel primer sets for detection of Ig/TCR rearrangements

BIOMED-2 study (multiplex PCR)

Ig genes: *IGH*: V_H-J_H and D_H-J_H
IGK: V_κ-J_κ and Kde rearrangements
IGL: V_λ-J_λ

TCR genes: *TCRB*: V_β-J_β and D_β-J_β
TCRG: V_γ-J_γ
TCRD: V_δ-J_δ, D_δ-D_δ, D_δ-J_δ, and V_δ-D_δ

BIOMED-2 clonality strategy

Suspected
B-cell lymphoma



IGH V(D)J FR1, FR2, FR3
IGH DJ(A)
IGK-VJ and DE

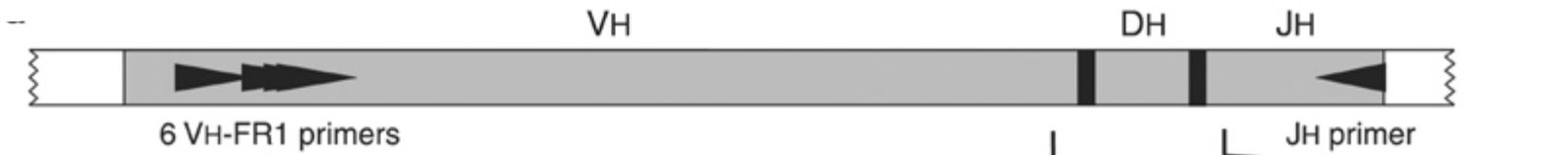
Suspected lymphoma
of unknown origin



Suspected
T-cell lymphoma



TCRGVJ (A and B)
TCRB V(D)J (A and B)
TCRB DJ



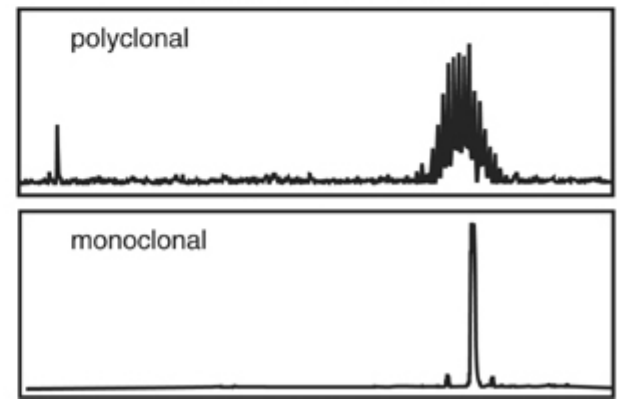
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GeneScanning



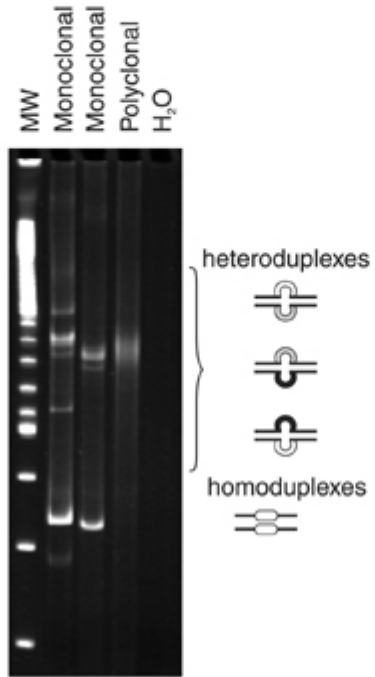
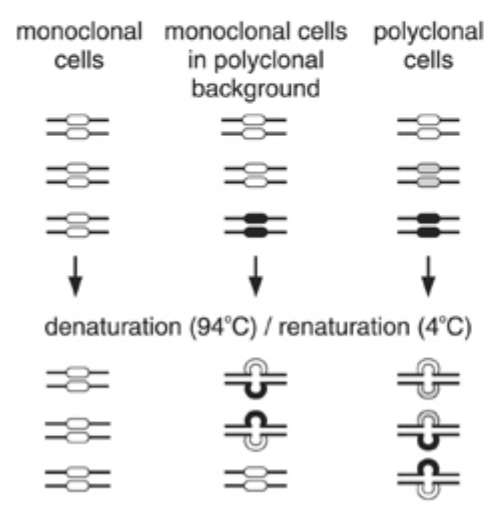
Van Dongen et al Leucemia 2003



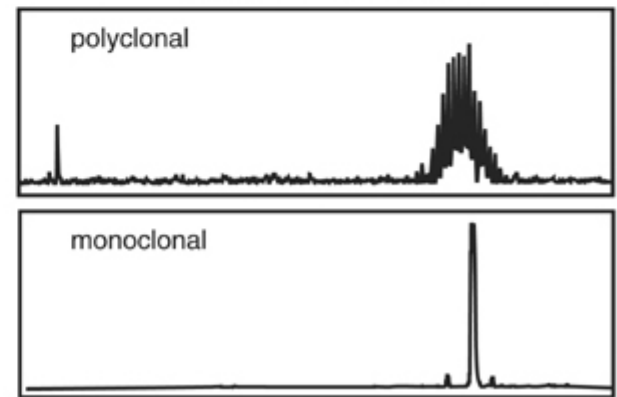
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CTGTGCATATCTACTTTGACTACTGG
  
```

b Heteroduplex analysis



c GeneScanning



Van Dongen et al Leucemia 2003

Analysis of *TCRB* gene rearrangements



TCRB tubes A and B

Vβ	Offset	5'	3'
Vβ2	(-204)	AACTATGTTTTGGTATCGTCA	3'
Vβ4	(-201)	CACGATGTTCTGGTACCGTCAGCA	
Vβ5/1	(-197)	CAGTGTGTCCTGGTACCAACAG	
Vβ6a/11	(-201)	AACCCTTTATTGGTACCGACA	
Vβ6b/25	(-201)	ATCCCTTTTTGGTACCAACAG	
Vβ6c	(-201)	AACCCTTTATTGGTATCAACAG	
Vβ7	(-198)	CGCTATGTATTGGTACAAGCA	
Vβ8a	(-201)	CTCCCGTTTTCTGGTACAGACAGAC	
Vβ9	(-198)	CGCTATGTATTGGTATAAAGCA	
Vβ10	(-201)	TTATGTTTACTGGTATCGTAAGAAGC	
Vβ11	(-198)	CAAAATGTACTGGTATCAACAA	
Vβ12a/3/13a/15	(-198)	ATACATGTACTGGTATCGACAAGAC	
Vβ13b	(-198)	GGCCATGTACTGGTATAGACAAG	
Vβ13c/12b/14	(-198)	GTATATGTCCTGGTATCGACAAGA	
Vβ16	(-201)	TAACCTTTATTGGTATCGACGTGT	
Vβ17	(-198)	GGCCATGTACTGGTACCGACA	
Vβ18	(-201)	TCATGTTTACTGGTATCGGCAG	
Vβ19	(-201)	TTATGTTTATTGGTATCAACAGAATCA	
Vβ20	(-193, inv)	CAACCTATACTGGTACCGACA	
Vβ21	(-201)	TACCCTTTACTGGTACCGGCAG	
Vβ22	(-201)	ATACTTCTATTGGTACAGACAAATCT	
Vβ23/8b	(-201)	CACGGTCTACTGGTACAGCA	
Vβ24	(-197)	CGTCATGTACTGGTACAGCA	

TCRB tubes A and C: Jβ A primers

3'	5'	Jβ
GTGGTCTAAGTGTCAACATCCATTC	(+53)	Jβ1.1
CTGGTCCAATTGGCAACATCCATTC	(+53)	Jβ1.2
TTCAACCGAGTGACAACATCCATTC	(+55)	Jβ1.3
CTTGGGTCGAGAGACAGAACCCATAC	(+56)	Jβ1.4
CTGAGCTGAGAGGTAGGATCCATTC	(+55)	Jβ1.5
GTCCGAGTGACACTGTCCATAC	(+58)	Jβ1.6
TCCGACTGGCATGACCCATTC	(+56)	Jβ2.2
TCCGACTGGCACGACCCGCTC	(+58)	Jβ2.6
GTCCGAGTGCCAATGTCCATTC	(+52)	Jβ2.7

TCRB tubes B and C: Jβ B primers

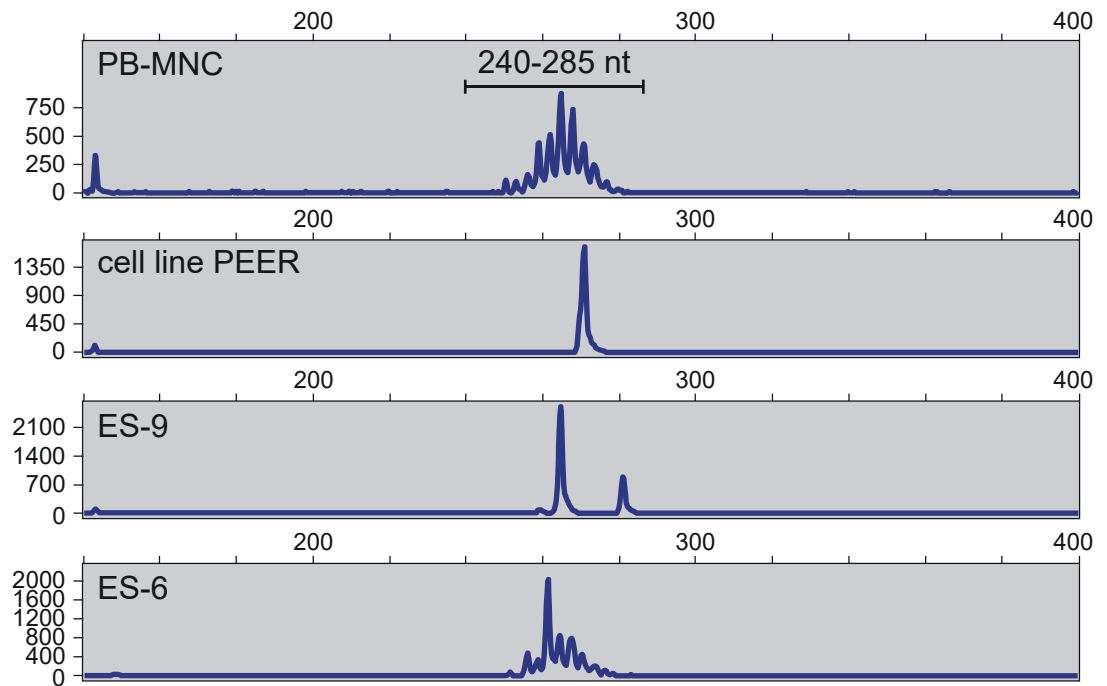
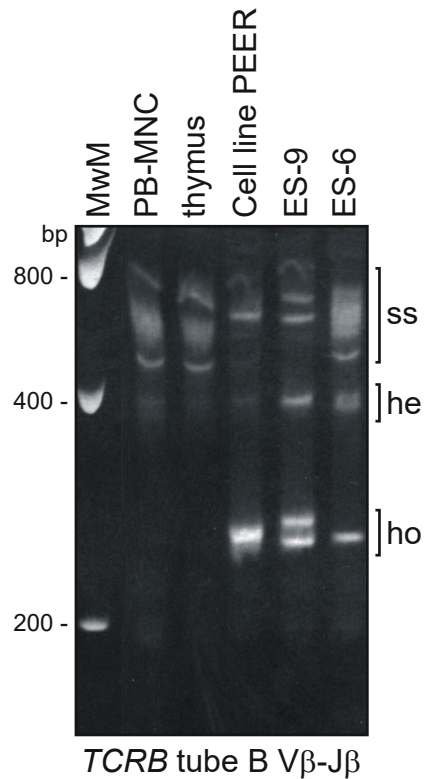
3'	5'	Jβ
AGTGGCAGCATCCATTCTTCC	(+59)	Jβ2.1
ACTGTCACGAGCCATTCGCC	(+58)	Jβ2.3
AGAGTCACGACCCATTCGACC	(+59)	Jβ2.4
CACGAGCCACACGCGC	(+57)	Jβ2.5



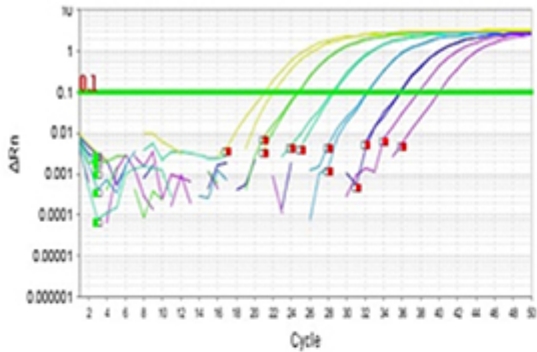
TCRB tube C

Dβ1	5'	3'	Dβ2	5'	3'
	(-252)	GCCAAACAGCCTTACAAAGAC		(-137)	TTTCCAAGCCCCACACAGTC

Analysis of *TCRB* gene rearrangements



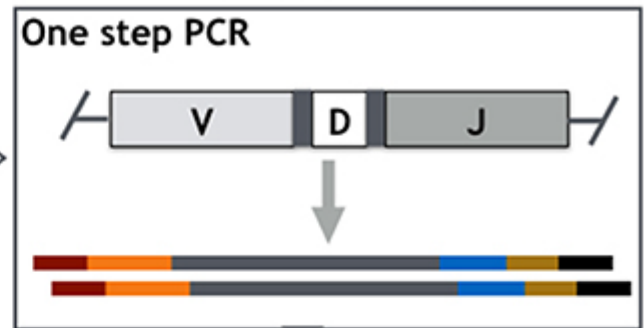
Quantitate 1 ug BMMC gDNA



Triplicates

1 ug BMMC gDNA
 plasmid A at 0.001%
 plasmid B at 0.01%

Library preparation



S_A sequencing reads

Plasmid A
 GCCTTTGGATTACCTTT...
 GCCTTTGGATTACCTTT...

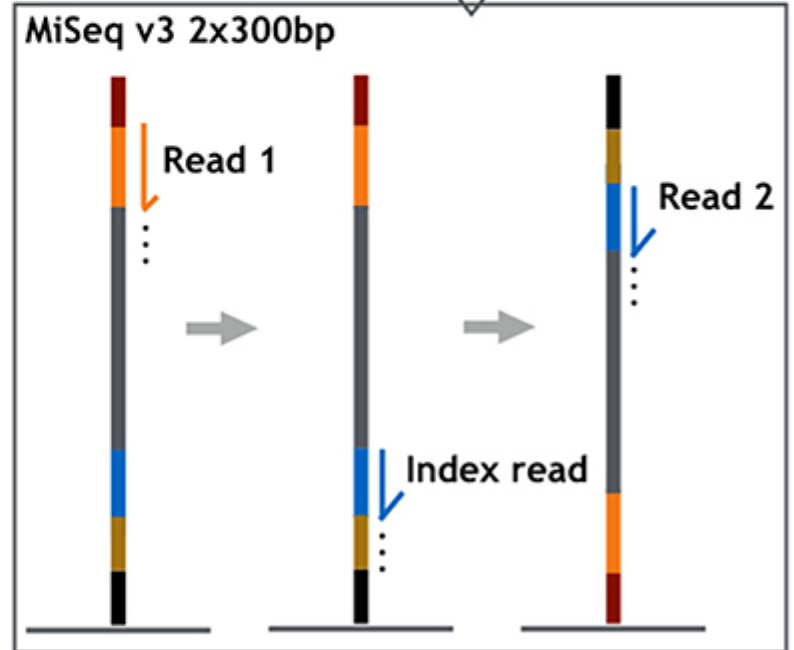
S_B sequencing reads

Plasmid B
 GGCTCTGGATTACAGGT...
 GGCTCTGGATTACAGGT...

S_{MRD} sequencing reads

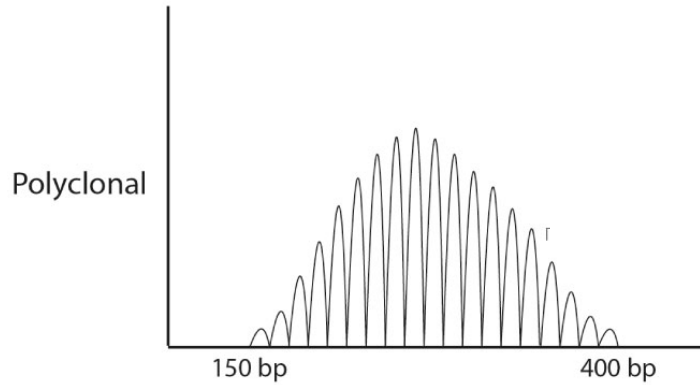
MRD target
 CACCTGCTCTGGATC...
 CACCTGCTCTGGATC...

$$MRD = S_{MRD} \times \frac{0.01\%}{S_B}$$

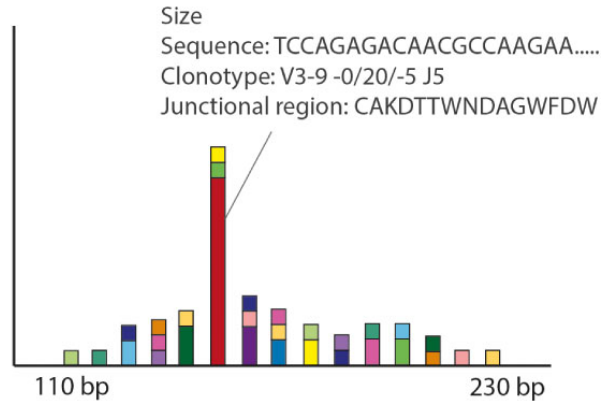
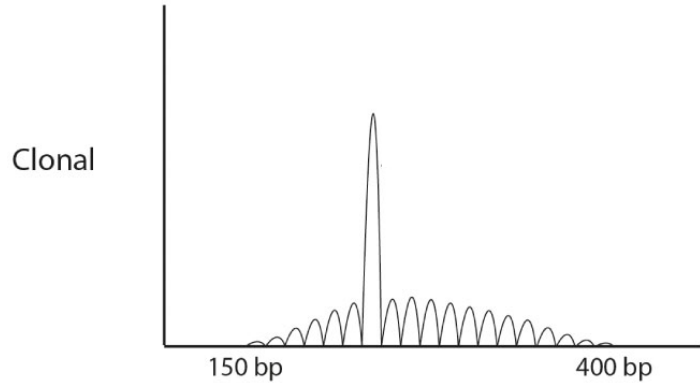
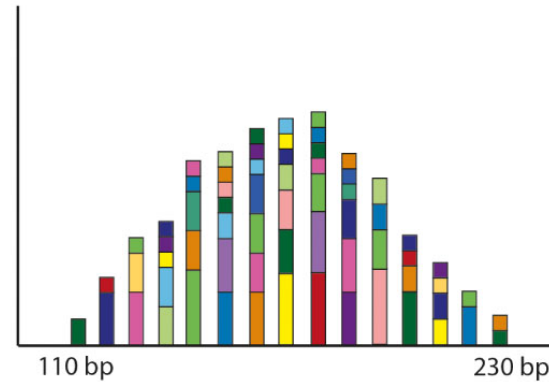


NGS analysis

GeneScan fragment size analysis

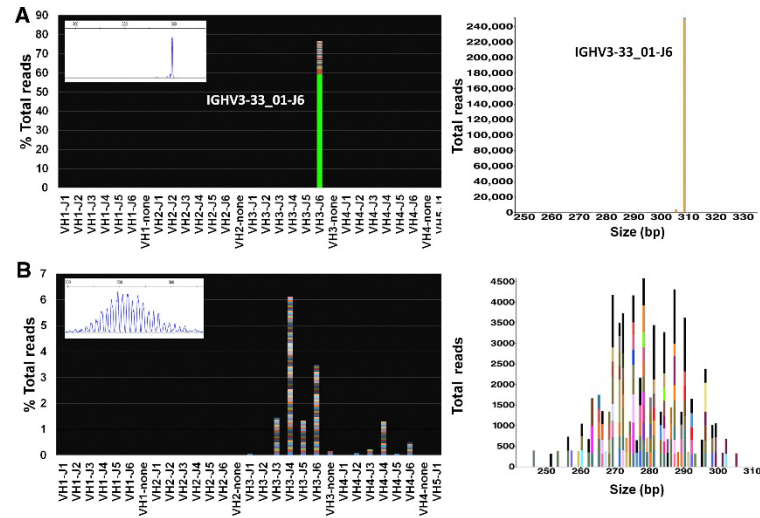


NGS-based analysis

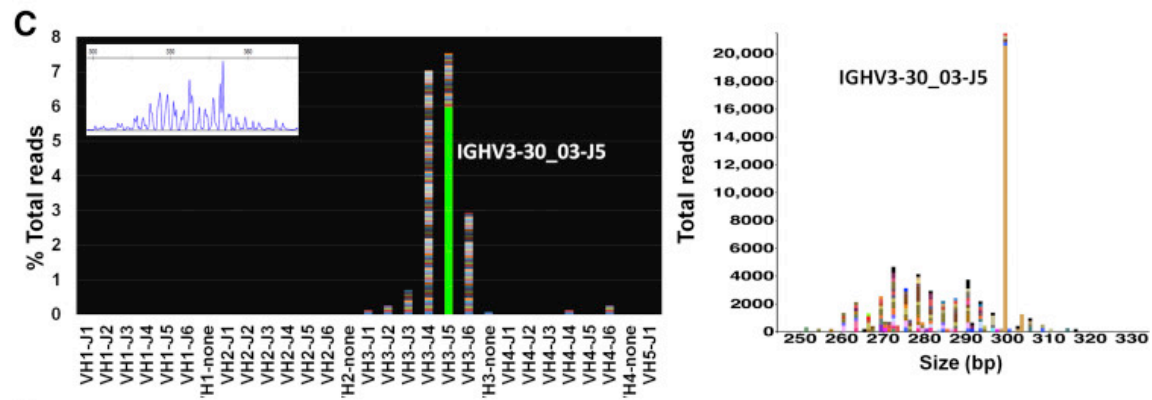
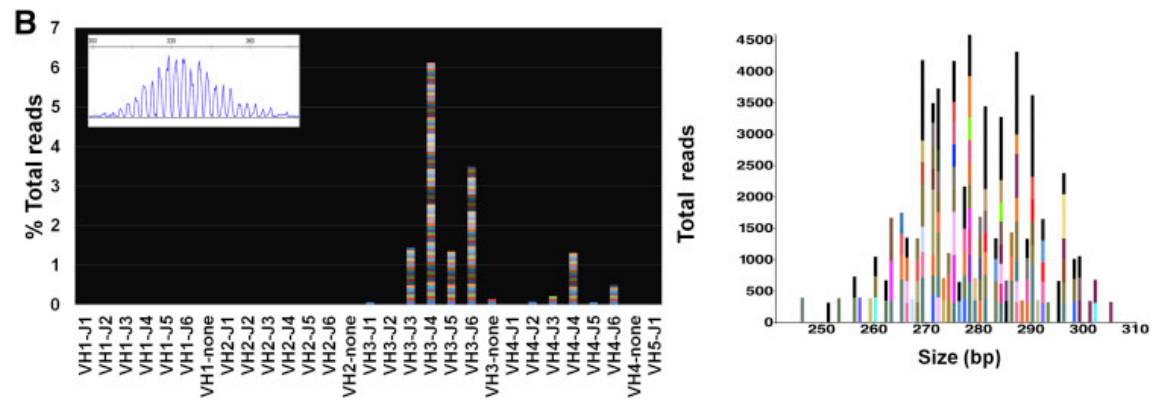
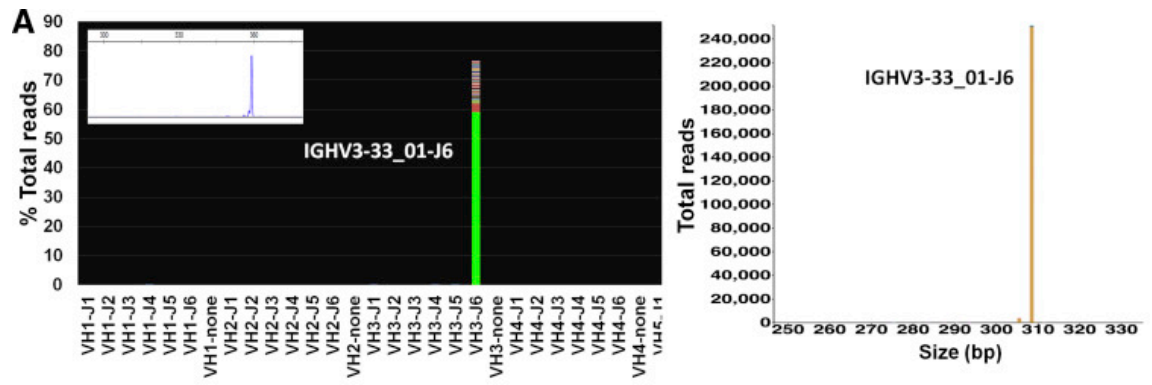


Adapted from P. Groenen et al, Front. Oncol., 08 February 2023

NGS analysis



M.E. Arcile et al The Journal of Molecular Diagnostics



From the patient to the analysis

- Selection of material
- Protocol
- Check DNA quality
- Clonality analysis

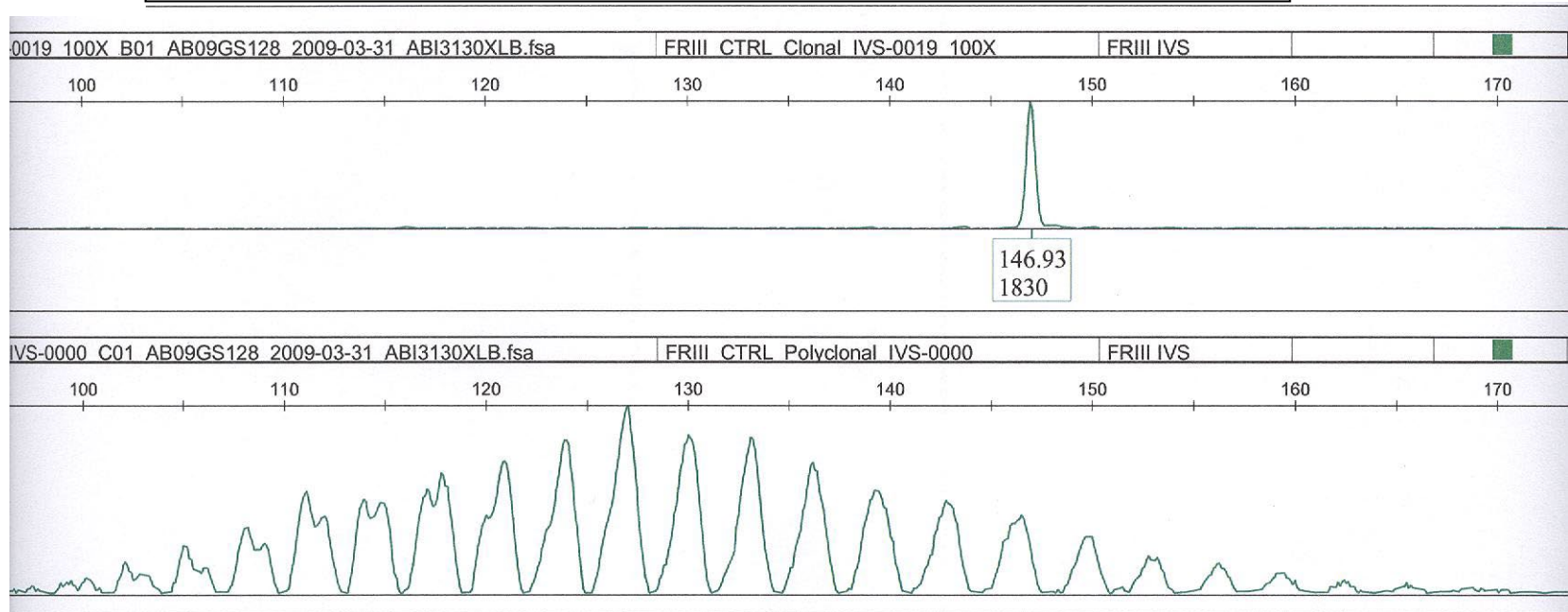
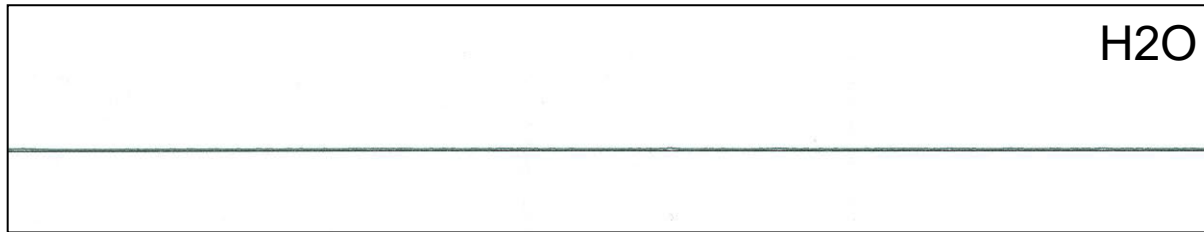
Examples of cases

- Case 1: Lymphoma? Reactive?
- Case 2: Relapse?
- Case 3: T-cell lymphoma

Case 1

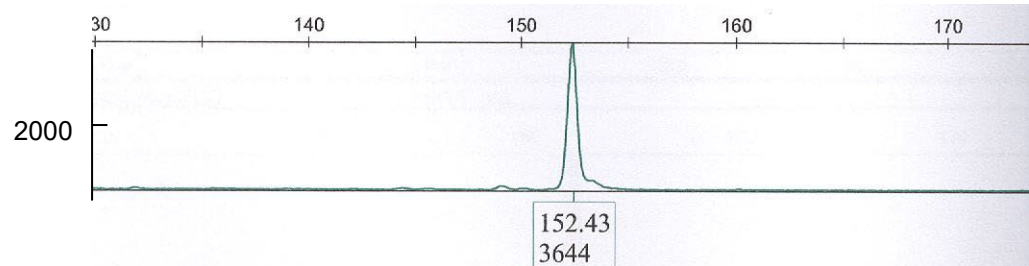
- Female
- 63 years
- Lymphoma in dec. 2004
- FR1 polyclonal, FR3 monoclonal
- Feb. 2009 biopsy
- Relapse?

Case 1: GeneScan analysis of controls



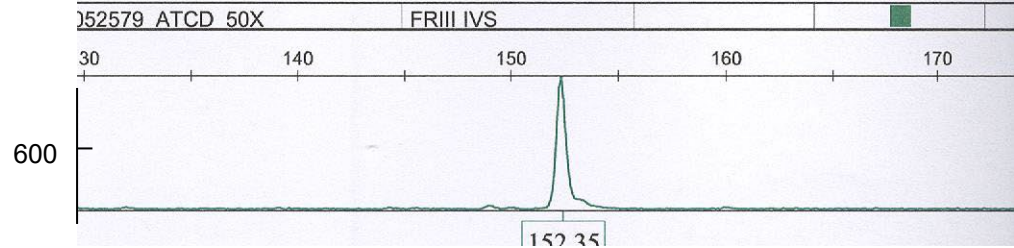
Case 1: Relapse?

2/2009
Biopsie
FR3: 152 bp



by: Administrator

12/2004
Biopsie
FR3: 152 bp



Case 1: Results

Controls ok

B-cell targets:

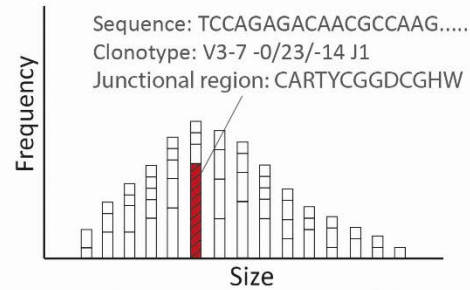
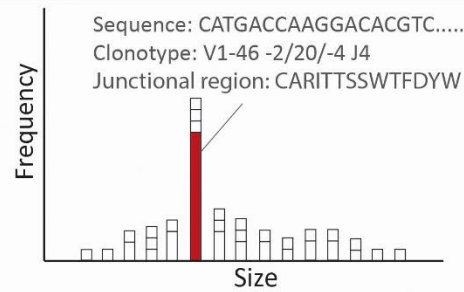
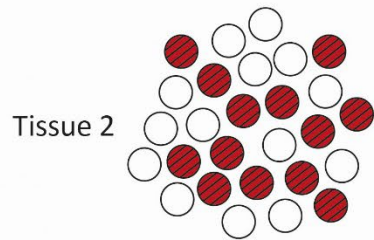
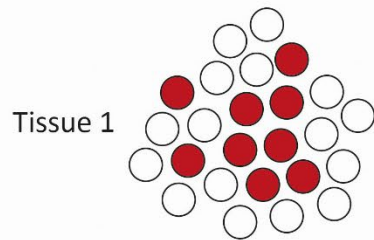
IGH(VDJ) FR3 clonal

Conclusion of the molecular results:

Clonal rearrangement of the IGH gene was detected in this specimen.

This gene rearrangement profile seems to be identical to the one detected in the biopsie of 12/2004 and could confirm the relapse of the disease.

Clonal comparison

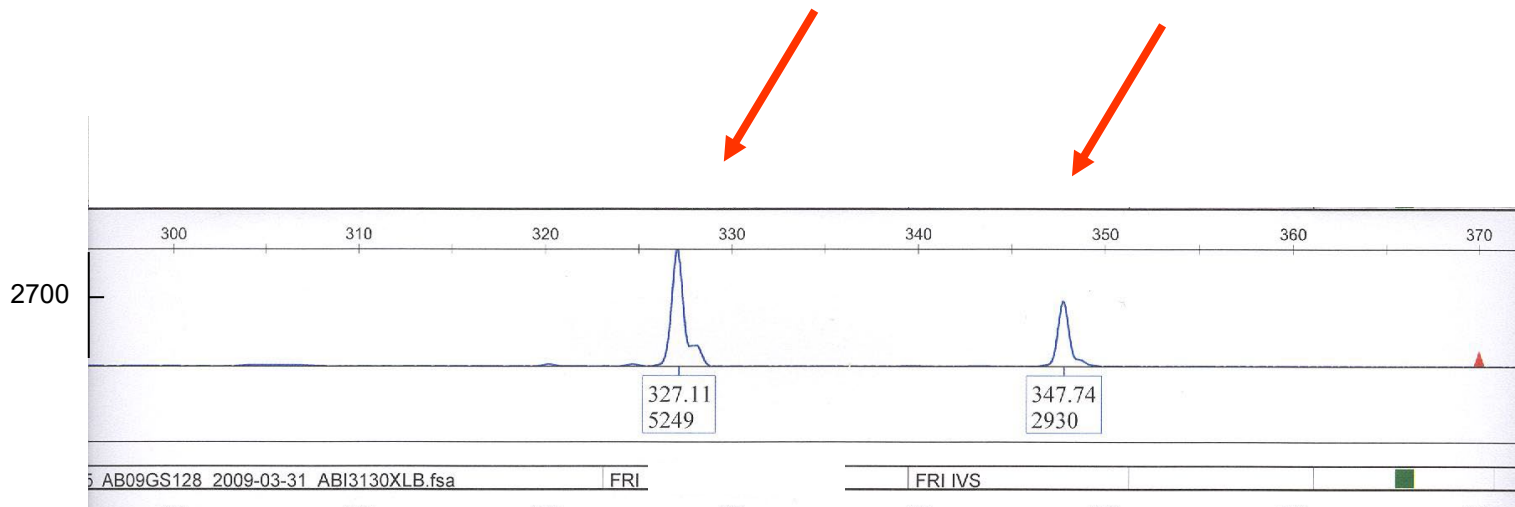


Adapted from P. Groenen et al, Front. Oncol., 08 February 2023

Case 2

- Female
- 81 years
- Biopsie
- Lymphoma?

Case 2



3/2009
biopsie
FR1
327,348 bp

Case 2: Results

Controls ok

B-cell targets:

IGH(VDJ) FR1 clonal

FR2 clonal

Conclusion of the molecular results:

Clonal rearrangements of the IGH gene were detected in this specimen.

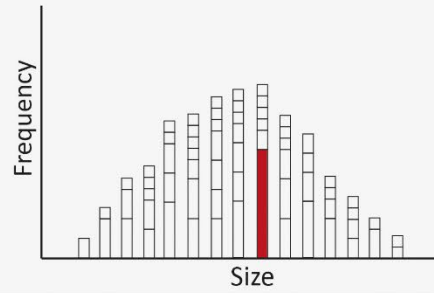
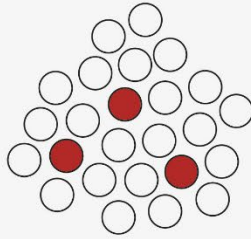
This gene rearrangement profile fits to the presence of one or two clonal B-cell population/B-NHL.

Advantage of NGS

Lymphocyte population

NGS-based analysis

Detection of small clones in polyclonal background



Adapted from P. Groenen et al, Front. Oncol., 08 February 2023

Case 3:

Male

75 years

Lymphoma origine?

2 biopsies

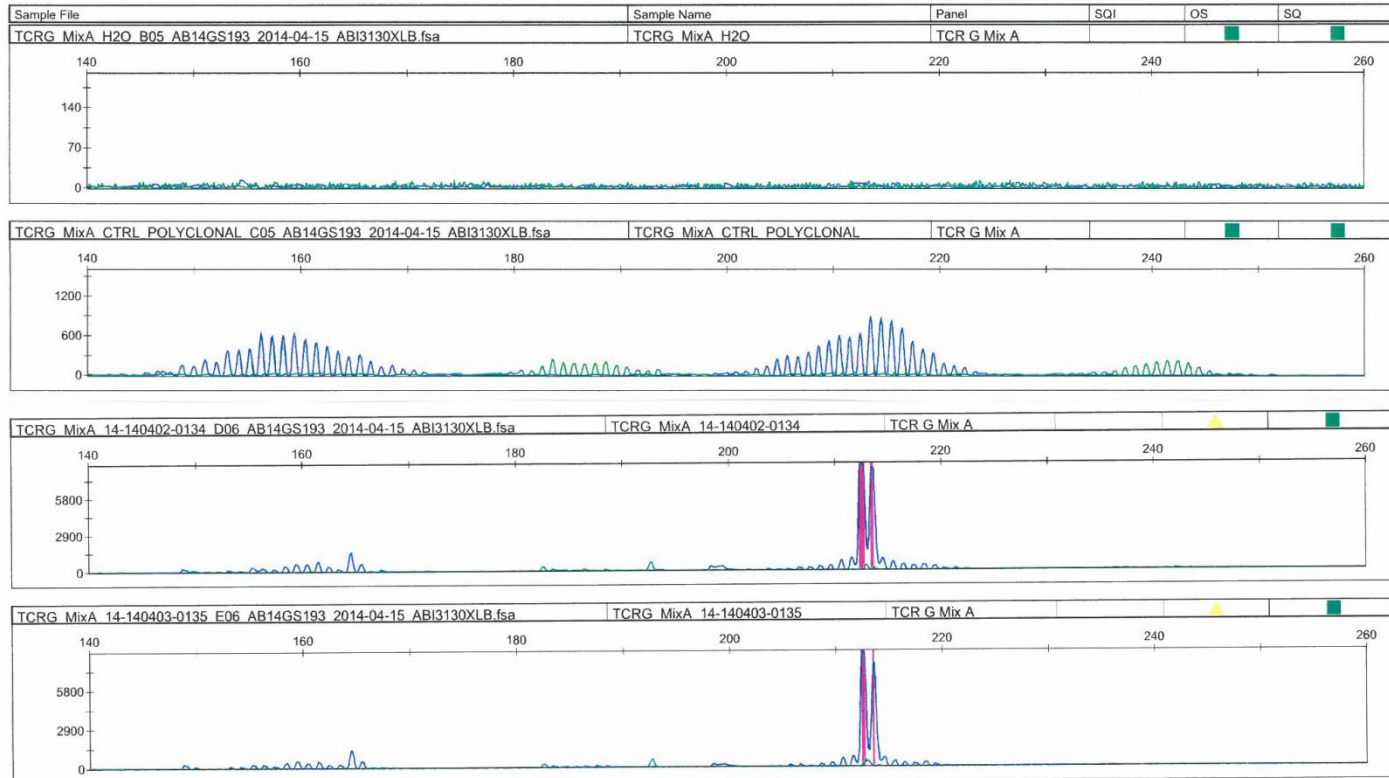
First results:

FR1, 2, 3 kappa polyclonal

Case 3: TCRG

Applied Biosystems
GeneMapper 4.1

TCRG PCR 14-342

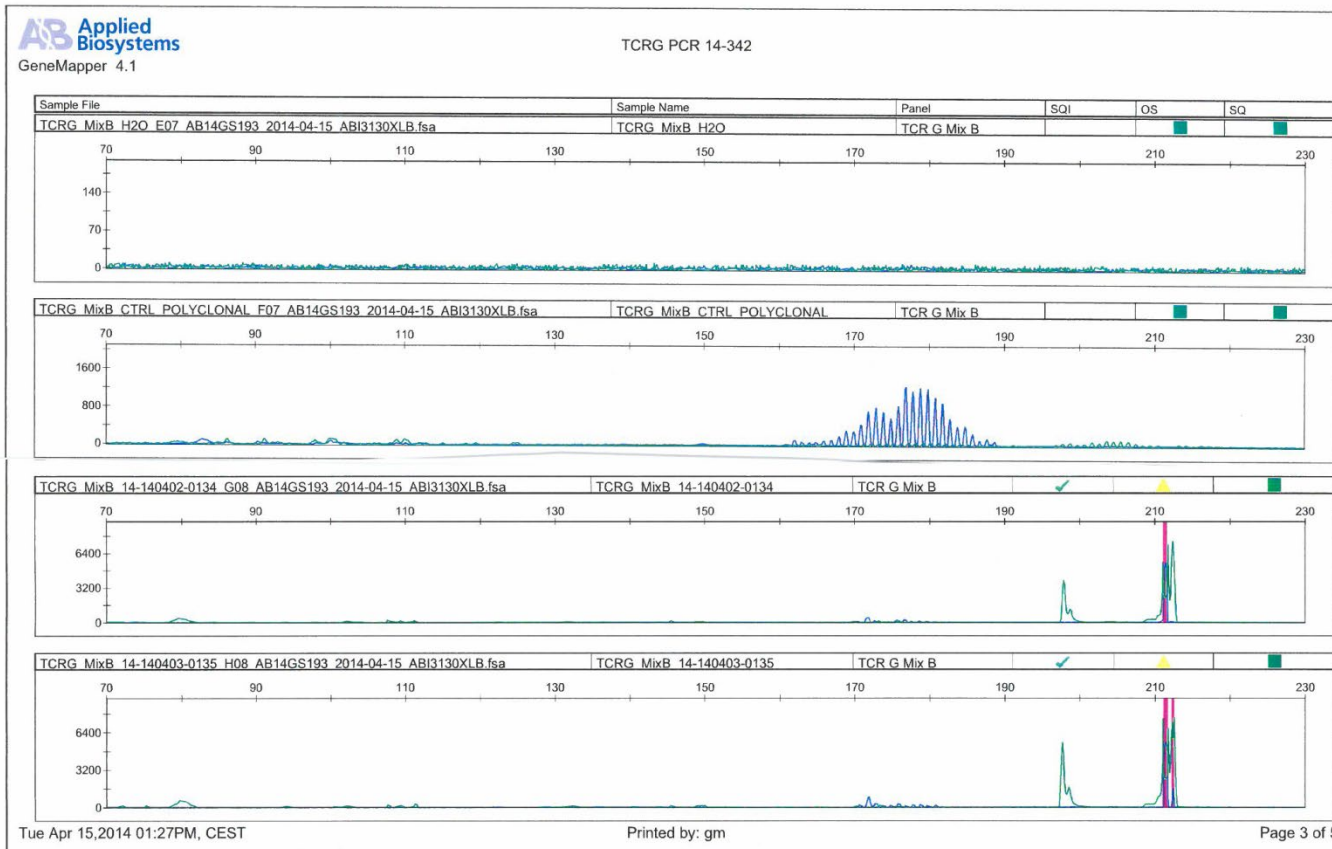


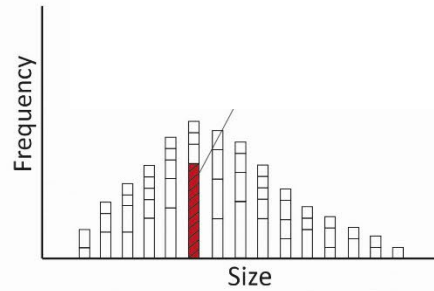
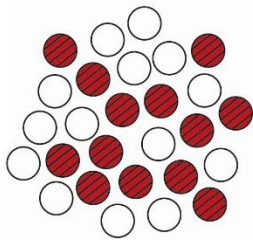
Tue Apr 15, 2014 12:26PM, CEST

Printed by: gm

Page 3 of 5

Case 3: TCRG



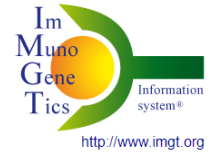


Adapted from P. Groenen et al, Front. Oncol., 08 February 2023

False sequences

N° Echantillon	reads	total Rank	Sequence	Length	Raw count	V-gene	J-gene	% total reads	Cumulative %
240326-0081	147489	1	CCTTCCCATTTTAATTCACT GCCTTTGTCTTTTCCAAGCC CCACACAGTCAGACTAACCT CTGCCACCTGCGCTTCTGT CCGCTGCCCAGTGGTTGGG GGAGGGGGACTAGCAGGG AGSAAACATTTTGTATCAT GGTGTACACTTGTGGGGAC TAGCGGGGGCTCAGAGAGA GATACGCAGTATTTTGGCC CAGGC	198	17806	Db2	Jb2-3	12,07	12,07
		2	GGAGGTGAGAAGGAAGCCC CCGGCCTGGTCCATACCCC ACCACCACTTGCAATATGG GGGGTGATGTCACCCACCC TGCACCTCCCTCAAAAGAG CAGCTGCTCTGGTGTCTC TCCCAGGCTCTGGGGGCG GACCCATGGGAGGGGCTGT TTTTGTACAAAAGCTGTAACA TTGTGGGACAGGGGGCG CATTAATGTACATGATGGC TACACCTCGGTTCCG	225	13458	Db1	Jb1-2	9,12	21,20
		3	GCTTAAGGCAGATCTACTAT TCAATGAATGTTGAGGTGA CTGATAAGGGAGATGTTCC TGAAGGSTACAAGTCTCT CGAAAAGAGAAGAGGAATT TCCCCTGATCCTGGAGTC GCCAGCCCCAACAGACC TCTGTACTTCTGTGGCAG CAGTCTCCATGGGAGAG ATAATTAATGGCTACACCTT CGGTTCCG	200	8600	Vb27	Jb1-2	5,83	27,03

WELCOME ! to **IMGT/V-QUEST**



IMGT®, the international ImMunoGeneTics information system®

Citing IMGT/V-QUEST:
Brochet, X., Lefranc, M.-P. and Giudicelli, V. Nucl. Acids Res. 36, W503-508 (2008). PMID: 18503082 **PDF**
Giudicelli, V., Brochet, X., Lefranc, M.-P. Cold Spring Harb Protoc. 2011 Jun 1;2011(6). pii: pdb.prot5633. doi: 10.1101/pdb.prot5633.
PMID: 21632778 **Abstract** also in *IMGT booklet with generous provision from Cold Spring Harbor (CSH) Protocols* **PDF** (**high res**) **PDF** (**lower res**)

IMGT/V-QUEST program version: [3.6.3](#) (30 January 2024) - IMGT/V-QUEST reference directory release: [202416-4](#) (18 April 2024)

Analyse your IG (or antibody) or TR nucleotide sequences

The list of the IMGT/V-QUEST reference directory sets to which your sequences can be compared is available [here](#).

Human sequence sets to test IMGT/V-QUEST are available [here](#).

Your selection

Species

Receptor type or locus

Sequence submission

Ⓞ Type (or copy/paste) your nucleotide sequence(s) in [FASTA](#) or in [FASTQ](#) format

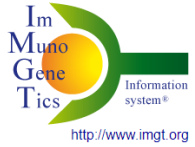
```
>24032d0081
CCTTCCCATTTTAAATTCAC16CCCTTTGTCTTTTCCAAGCCCCACACAGTCCAGACTAACCTCTGCCACCTGCGCTTCT16CCGCTG6CCAGT69TT69666
AGGGGGACTAGCAGGGAGGAAAACATTTTTGTATCATGGTGAACATTGTGGGGACTAGCGGGGGCTCAGAGAGAGATACGCAGTATTTTGGCCCAAGGC
```

Or give the path access to a local file containing your sequence(s) in [FASTA](#) or in [FASTQ](#) format

Aucun fichier n'a été sélectionné

Display results

THANK YOU for using IMGT/V-QUEST



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Citing IMGT/V-QUEST:
Brochet, X., Lefranc, M.-P. and Giudicelli, V. Nucl. Acids Res. 36, W503-508 (2008). PMID: 18503082 **★20★**
Giudicelli, V., Brochet, X., Lefranc, M.-P. Cold Spring Harb Protoc. 2011 Jun 1, 2011(6). pii: pdb.prot5633. doi: 10.1101/pdb.prot5633.
PMID: 21632778 **Abstract** also in *IMGT booklet with generous provision from Cold Spring Harbor (CSH) Protocols* **★20★** (high res) **★20★** (lower res)

IMGT/V-QUEST program version: [3.6.3](#) (30 January 2024) - IMGT/V-QUEST reference directory release: [2024.16-4](#) (18 April 2024)

Species	Homo sapiens (human)
Receptor type or locus	TR
IMGT directory reference set	F+ORF+ in-frame P
Search for insertions and deletions	yes

A. Detailed results for the IMGT/V-QUEST analysed sequences

Number of analysed sequences: **1**

1: [2403260081](#)

- 👉 This release of IMGT/V-QUEST uses [IMGT/JunctionAnalysis](#) for the analysis of the JUNCTION
- 👉 Hyphens (-) show nucleotide identity, dots (.) represent gaps

Sequence 1 2403260081: no results

Fri Apr 26, 2024 09:06:08 CEST

[IMGT/V-QUEST Documentation](#) [IMGT/V-QUEST Search page](#)



False sequences

N° Echantillon	reads	total Rank	Sequence	Length	Raw count	V-gene	J-gene	% total reads	Cumulative %
240326-0081	147489	1	CCTTCCCATTTTAATTCACT GCCTTTGTCTTTTCCAAGCC CCACACAGTCAGACTAACCT CTGCCACCTGCGCTTCTGT CCGCTGCCCAGTGGTTGGG GGAGGGGGACTAGCAGGG AGSAAACATTTTGTATGAT GGTGTACACTTGTGGGGAC TAGCGGGGGCTCAGAGAGA GATACGCAGTATTTTGGCC CAGGC	198	17806	Db2	Jb2-3	12,07	12,07
		2	GGAGGTGAGAAGGAAGCCC CCGGCCTGGTCCATACCCC ACCACCAACTTGCAATATGG GGGGTGATGTCACCCACCC TGCACCTCCCTCAAAAGAG CAGCTGCTCTGGTGTCTC TCCCAGGCTCTGGGGGCG GACCCATGGGAGGGGCTGT TTTTGTACAAAAGCTGTAACA TTGTGGGACAGGGGGCG CATTAATGTACATGATGGC TACACCTCGGTTCCG	225	13458	Db1	Jb1-2	9,12	21,20
		3	GCTTAAGGCAGATCTACTAT TCAATGAATGTTGAGGTGA CTGATAAGGGAGATGTTCC TGAAGGSTACAAGTCTCT CGAAAAGAGAAGAGGAATT TCCCCTGATCCTGGAGTC GCCAGCCCCAACCCAGACC TCTGTACTTCTGTGGCAG CAGTCTCCATGGGAGAG ATAATTAATGGCTACACCTT CGGTTCCG	200	8600	Vb27	Jb1-2	5,83	27,03

A. Detailed results for the IMG T/V-QUEST analysed sequences

Number of analysed sequences: **1**

1. 2403260081seq3

 This release of IMG T/V-QUEST uses [IMG T/JunctionAnalysis](#) for the analysis of the JUNCTION

 Hyphens (-) show nucleotide identity, dots (.) represent gaps

Sequence: 1 2403260081seq3

Analysed sequence length: 200.

Sequence analysis category: 2 (indel search & correction).

Sequence compared with the [Homo sapiens \(human\) TR set](#) from the [IMG T reference directory](#) (set: F+ORF+ in-frame P)

>2403260081seq3

```
gcttaaggcagatcctctattcaatgaatgttgaggctgactgataaggagatgttcctg
aagggtacaaagtcctcgaagaagagaagaggaaattccccctgatcctggagtcgccc
gccccaacccagaccctctgtactctctgctagcagctctctccatggagcagataatta
atggctacaccctcgggttcg
```

Result summary: 2403260081seq3	Productive TRB rearranged sequence (no stop codon and in-frame junction)		
V-GENE and allele	Homsap_TRBV27*01_F	score = 745	identity = 100.00% (149/149 nt) - (V-REGION partial in 5', missing nt nb= 124)
J-GENE and allele	Homsap_TRBJ1-2*01_F	score = 85	identity = 80.77% (21/26 nt) - (J-REGION partial in 3' missing nt nb= 21)
D-GENE and allele by IMG T/JunctionAnalysis	Homsap_TRBD1*01_F	D-REGION is in reading frame 2	
FR-IMG T lengths, CDR-IMG T lengths and AA JUNCTION	[X.6.37.3]	[X.6.14]	CASSLSMGQIINGYTF
JUNCTION length (in nt) and decryption	48 nt = (12)-5(8)0(7)-5(7)-6(14)	(3)V3(N1)5(D)3(N2)5(5)J	

1. Alignment for V-GENE and allele identification

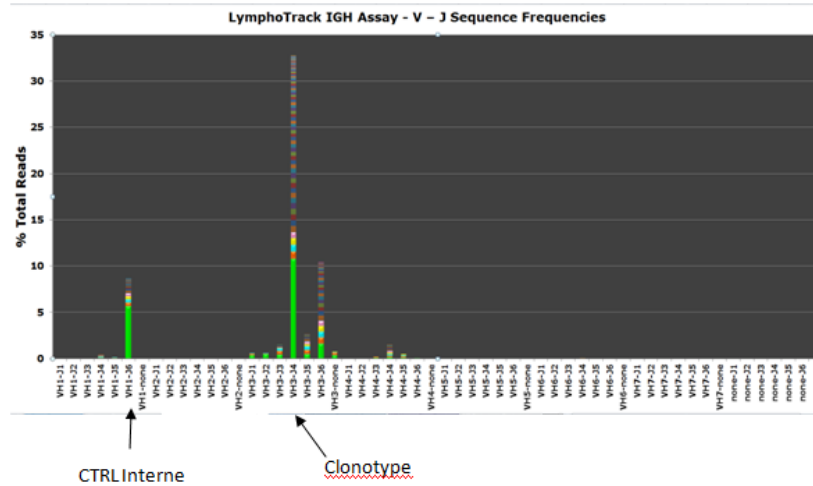
Closest V-REGIONS (evaluated from the V-REGION first nucleotide to the 2nd-CYS codon)

		Score	Identity
U88314	Homsap TRBV27*01 F	745	100.00% (149/149 nt)
U88314	Homsap TRBV28*01 F	394	73.83% (110/149 nt)
U11951	Homsap TRBV24-1*01 F	322	68.46% (102/149 nt)
IMG T000021	Homsap TRBV24-1*02 F	313	67.79% (101/149 nt)
IMG T000021	Homsap TRBV10-2*02 F	304	67.11% (100/149 nt)

Alignment with [FR-IMG T](#) and [CDR-IMG T](#) delimitations

MRD (minimal residual disease) analysis

170322-0041



LymphoTrack™

MRD Report

Sequence #1		Sequence Name:		Sample	
Replicate:	9	MRD Status	Sequence Detected (1)	Reads in Replicate	915981 (2)
CATCTGGATACACCTTCACCAGCTACTATATGCACTGGGTGCGACAGGCCCTGGACAAGGGCTTGAGTGGATG GGAATAATCAACCCTAGTGGTGGTAGCACAAGCTACGCACAGAAGTTCCAGGGCAGAGTCACCATGACCAGGGA CACGTCCACGAGCACAGTCTACATGGAGCTGAGCAGCCTGAGATCTGAGGACACGGCCGTGTATTACTGTGCTA GAGATCTCACAGTTGTATTAGTACCAGCTGCTATCCTCCGACTACTTTGACTACTGGGGCCAGGGAACCT					
Replicate Details:	Read Count	Cumulative Read Count:	Cumulative Read Frequency		
Exact Match	(3) 71	71	7.75E-5		
1 Mismatch	40	111	1.21E-4		
2 Mismatch	10	121	1.32E-4 (4)		
	Detection Limit:		% Confidence		
	1e-3		NA		
	1e-4		NA		
	1e-5		NA		
	1e-6		NA		