IMGT/V-QUEST and IMGT/JunctionAnalysis:

The standardized approach for IG and TR rearrangement analysis

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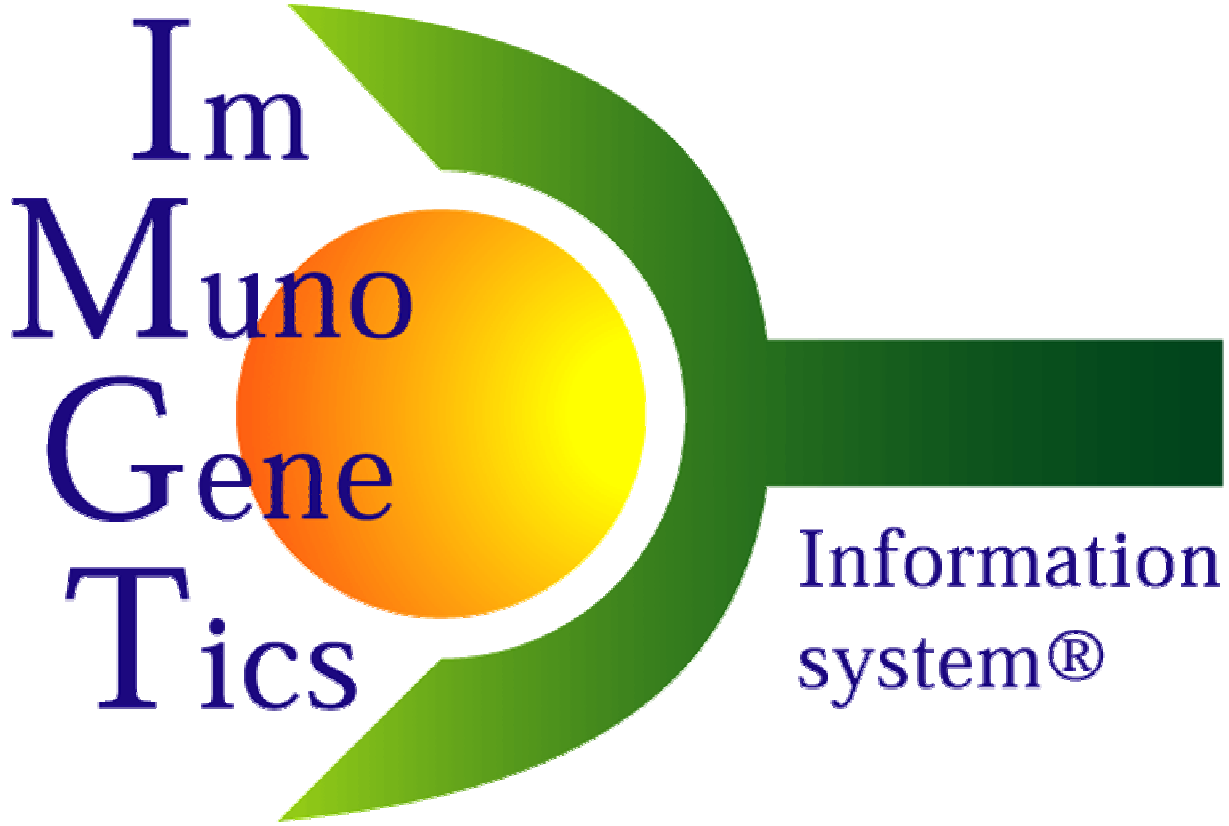
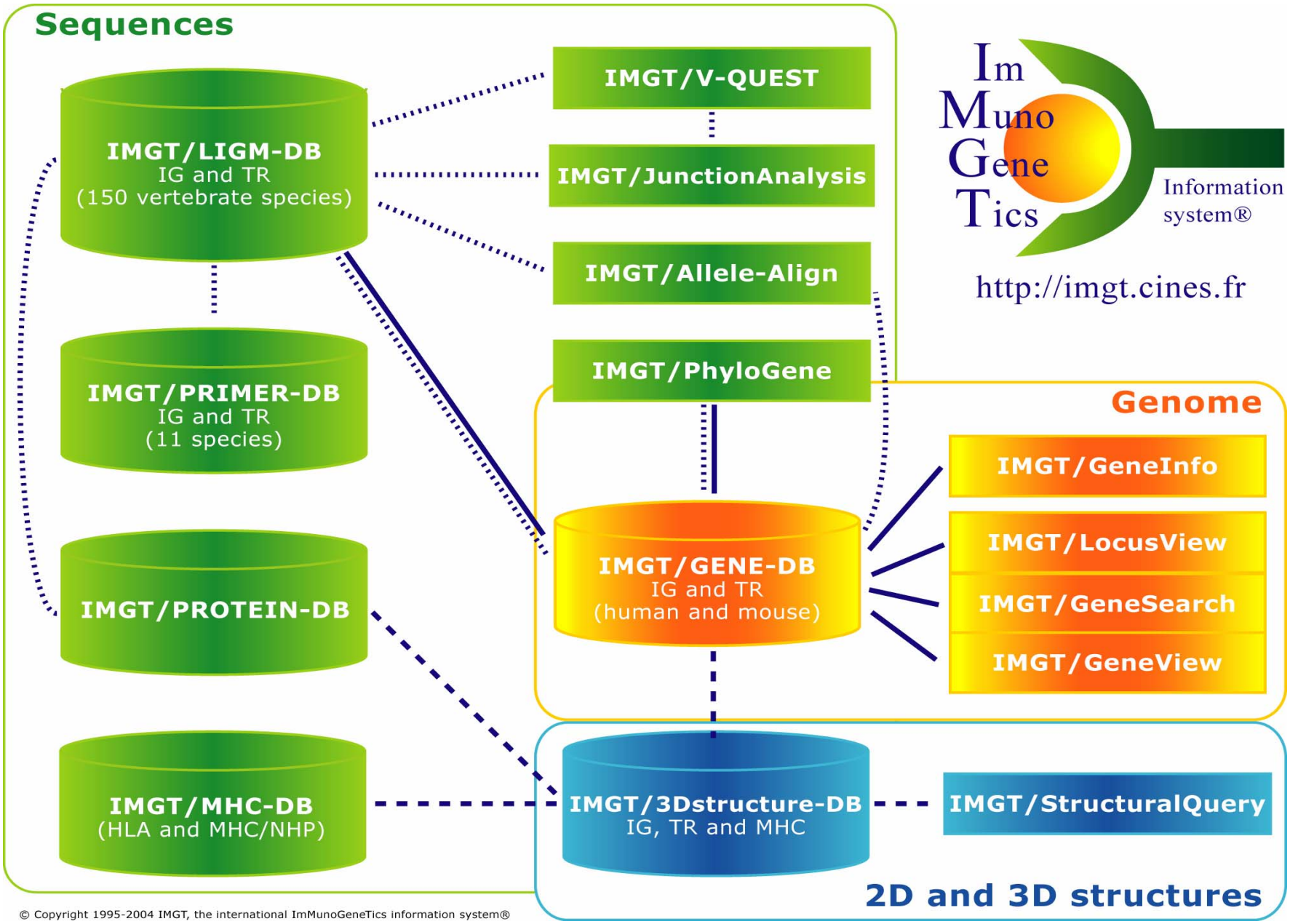
IMGT was created in June **1989**, during the Human Genome Meeting HGM 10

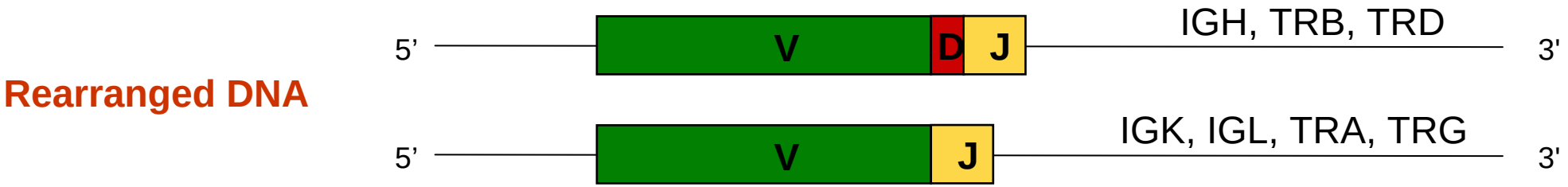


IMGT® was created to answer the need to manage the immunoglobulin (IG) and T cell receptor (TR) related data.



IMGT/V-QUEST and IMGT/JunctionAnalysis

IMGT/V-QUEST and **IMGT/JunctionAnalysis**, part of IMGT®, the international ImMunoGeneTics information system®, analyse the IG and TR rearranged V-D-J and V-J sequences.



1) identify the V, (D) and J genes and alleles involved in the rearrangements.
2) provide a detailed and accurate description of the sequences.

Results are according to the IMGT Scientific chart rules, based on the IMGT-ONTOLOGY axioms.

IMGT/V-QUEST and IMGT/JunctionAnalysis

IMGT/V-QUEST

- compares the user's sequences with the IMGT reference sequences (IMGT reference directory: CLASSIFICATION axiom)

- displays the nucleotide and protein alignments according to the IMGT Scientific Chart rules (IMGT labels: DESCRIPTION axiom)

- provides an extensive analysis of the mutations (IMGT unique numbering: NUMEROTATION axiom)

IMGT/JunctionAnalysis

- analyses accurately the junctions of IG and TR rearranged sequences: identifies IGHD, TRBD and TRDD genes and alleles, N- and P REGION, "gc" content, amino acid physicochemical properties, PI, etc.

Giudicelli et al. Nucleic Acids Res 32:435:440 (2004)
Yousfi Monod et al. Bioinformatics 20:379-385 (2004)

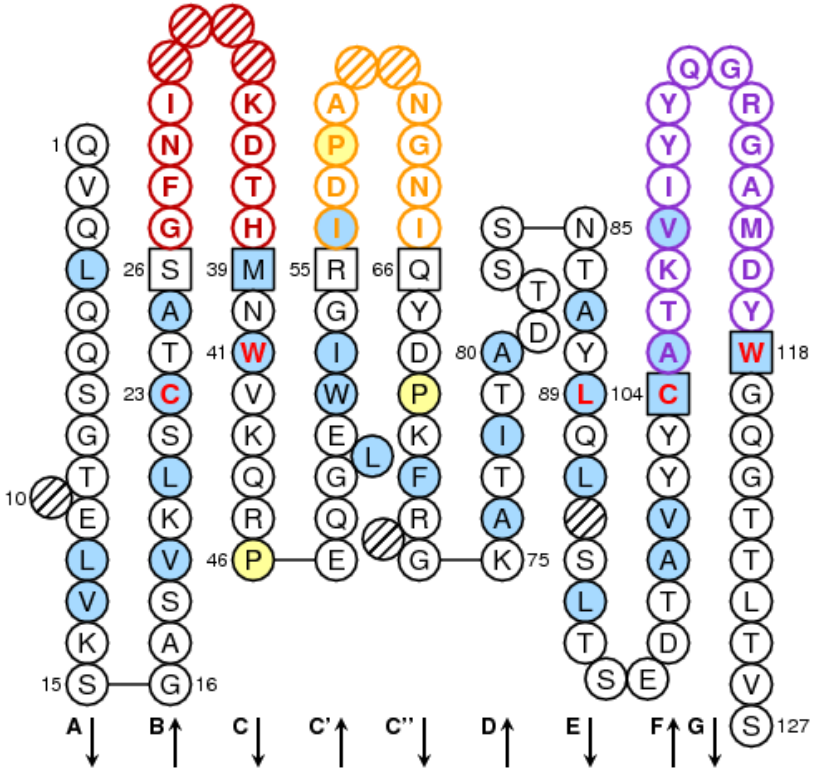
Specificities of IMGT/V-QUEST algorithm

IMGT unique numbering rules

- maintain conserved AA, and therefore codons, at the same positions
- standardize the delimitations of FR-IMGT and CDR-IMGT

Gaps are inserted in IMGT reference sequences, taking into acount the variable lengths of the CDR-IMGT

[8.8.15]



IMGT proposes for the first time a standardized description of the V-DOMAIN, whatever the antigen receptor, the chain type and the species.

IMGT/V-QUEST algorithm

In order to respect the IMGT unique numbering rules:

➔ The IMGT/V-QUEST algorithm is based on a global alignment which does not accept any insertion and/or deletion

➔ Gaps are integrated into the user sequence, according to the IMGT unique numbering, by comparison with a model sequence of the IMGT reference directory.

IMGT/V-QUEST web interface (http://imgt.cines.fr)

WELCOME !
to the IMGT/V-QUEST Search page

THE
INTERNATIONAL
IMMUNOGENETICS
INFORMATION SYSTEM®

Citing IMGT/V-QUEST: Giudicelli, V. et al. Nucl. Acids Res. 2004, 32, W435-440 PMID: 15215425 PDF

☞ You are in the new IMGT/V-QUEST, *upgraded for multiple sequences and with new functionalities.* NEW!

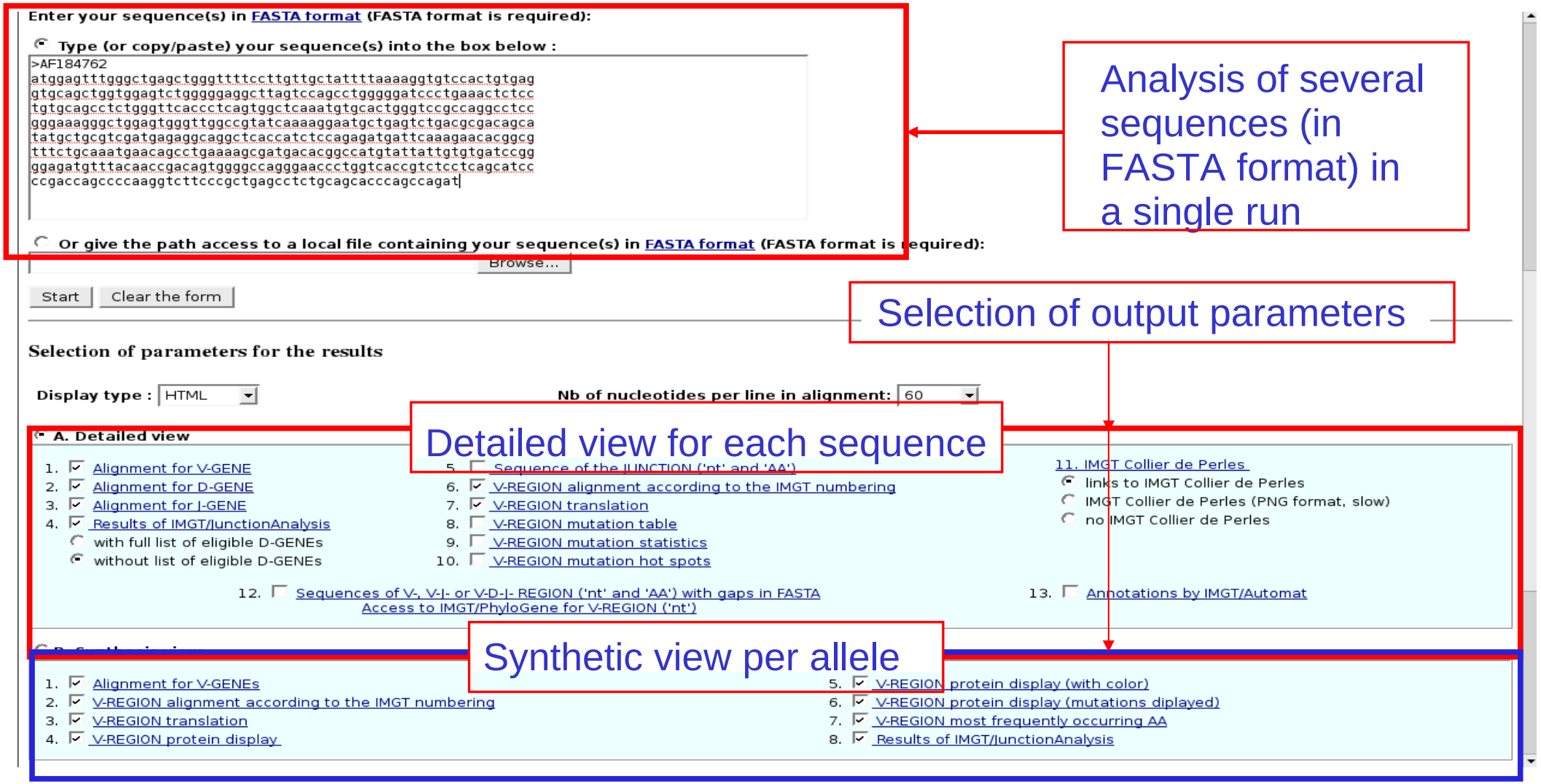
Analyse your Immunoglobulin nucleotide sequences

- Human
- Mouse
- Rat (test for IGHV2 and IGHV5 subgroups)
- Chondrichthyes
- Teleostei
  - Atlantic cod
  - Channel catfish
  - Rainbow trout
- Sheep

Analyse your T cell Receptor nucleotide sequences

- Human
- Mouse
- Non-human primates

IMGT/VQUEST+JCTA QUERY PAGE



1. “Detailed view” results: Summary

Sequence number 1: AF184762

Sequence compared with the human IG set from the IMGT reference directory

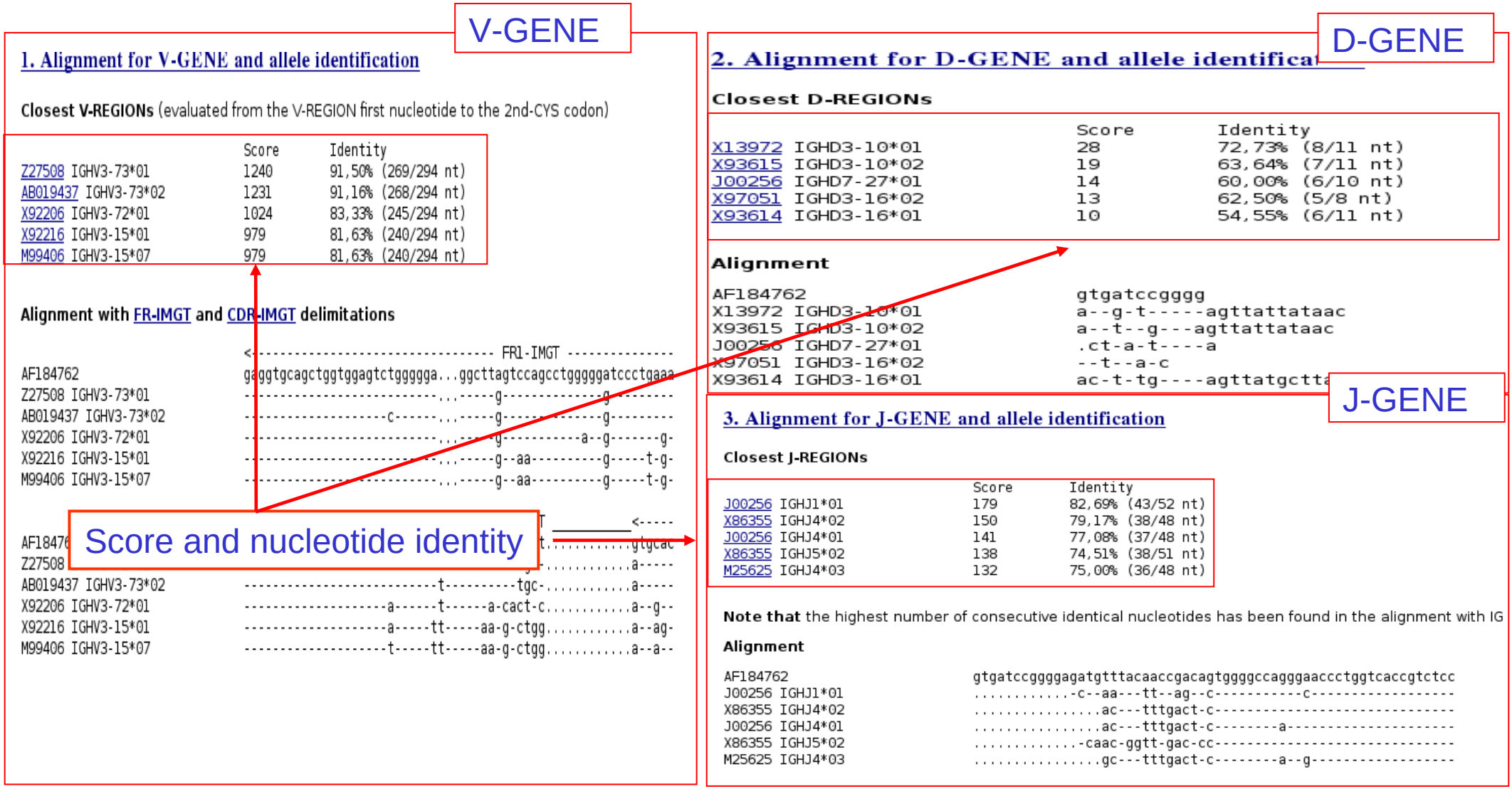
>AF184762
atggagtttgggctgagctgggttttccttgttgctattttaaaaggtgtccactgtgag
gtgcagctggtggagtctgggggaggcttagtccagcctgggggatccctgaaactctcc
tgtgcagcctctgggttcaccctcagtggctcaaatgtgcactgggtccgccaggcctcc
gggaaagggctggagtgggttggccgtatcaaaaggaatgctgagtctgacgcgacagca
tatgctgcgtcgatgagaggcaggctcaccatctccagagatgattcaaagaacacggcg
tttctgcaaatgaacagcctgaaaagcgatgacacggccatgtattattgtgtgatccgg
ggagatgtttacaaccgacagtggggccagggaaccctggtcaccgtctcctcagcatcc
ccgaccagccccaaggtcttcccgctgagcctctgcagcacccagccagat

Automatic interpretation

Result summary:	**Productive IGH rearranged sequence** (no stop codon and in frame junction)		
V-GENE and allele	IGHV3-73*01	score = 1240	identity = **91,50%** (269/294 nt)
J-GENE and allele	IGHJ1*01 (a)	score = 179	identity = 82,69% (43/52 nt)
D-GENE and allele by IMGT/JunctionAnalysis	IGHD3-10*01	D-REGION is in reading frame 3	
CDR-IMGT lengths and AA JUNCTION	[8,10,10]	CVIRGDVYNRQW	

(a) Other possibilities: IGHJ4*02 and IGHJ5*02 (highest number of consecutive identical nucleotides)

2. “Detailed view” results: V,D and J alignments



3. “Detailed view” results: IMGT/JunctionAnalysis

4. Results of IMGT/JunctionAnalysis

Maximum number of accepted mutations in: 3'V-REGION = 2, D-REGION = 4, 5'J-REGION = 2

Analysis of the JUNCTION

D-REGION is in reading frame 3.

Click on mutated (underlined) nucleotide to see the original one:

Input	V name	3'V-REGION	N1	D-REGION	N2	5'J-REGION	J name	D name	Vmut	Dmut	Jmut	Ngc
AF184762	IGHV3-73*01	tgt.....	g	.....tgatccggggagatgtt.....	tacaaccga	.....cagtgg	IGHJ1*01	IGHD3-10*01	0	4	1	5/10

Translation of the JUNCTION

Click on mutated (underlined) amino acid to see the original one:

	104	105	106	107	108	109	113	114	115	116	117	118	Frame	CDR3-IMGT length	Molecular mass	pI
	C	V	I	R	G	D	V	Y	N	R	Q	W				
AF184762	tgt	gtg	atc	cgg	gga	gat	gtt	tac	aac	cga	cag	tgg	+	10	1,508.72	9.24

Be aware that some allele reference sequences may be incomplete or from cDNAs. In those cases, IMGT/JunctionAnalysis uses automatically the allele *01 for the analysis of the JUNCTION.

5. Sequence of the JUNCTION ('nt' and 'AA')

```
104 105 106 107 108 109 113 114 115 116 117 118
 C   V   I   R   G   D   V   Y   N   R   Q   W
tgt gtg atc cgg gga gat gtt tac aac cga cag tgg
```

4. “Detailed view” results: mutations analysis

V-REGION mutation table

FR1-IMGT	CDR1-IMGT	FR2-IMGT	CDR2-IMGT	FR3-IMGT	CDR3-IMGT
g36>a	t88>c, F30>L (+ - -)	a115>g, M39>V (+ - -)	t168>c	g211>a, V71>M (+ - -)	a313>g, T105>V (- - -)
g51>a	t99>a	t135>c	g170>a, R57>K (+ + +)	a215>g, K72>R (+ + +)	c314>t, T105>V (- - -)
	g100>a, A34>N (- - -)		c174>g, S58>R (- - -)	t226>c, F76>L (+ - -)	t315>g, T105>V (- - -)

Mutation hotspot (motifs and positions in germline V-REGION)

(a/t)a (wa)

Motif	Positions
aa	58-59 (FR1)
aa	142-143 (FR2)
ta	165-166 (FR2-CDR2)
ta	168-169 (CDR2)
aa	171-172 (CDR2)
aa	175-176 (CDR2)
ta	180-181 (CDR2)
ta	187-188 (CDR2)
ta	199-200 (FR3)
aa	214-215 (FR3)
aa	249-250 (FR3)
aa	253-254 (FR3)
ta	262-263 (FR3)
aa	269-270 (FR3)
aa	274-275 (FR3)
aa	283-284 (FR3)
aa	285-286 (FR3)
ta	304-305 (FR3)
ta	307-308 (FR3)
ta	312-313 (FR3)
ta	315-316 (CDR3)

(a/g)g(c/t)(a/t) (rgyw)

Motif	Positions
agct	8-11 (FR1)
ggct	31-34 (FR1)
ggtt	80-83 (CDR1)
ggct	94-97 (CDR1)
ggct	132-135 (FR2)
ggct	146-149 (FR2)
ggtt	156-159 (FR2)
agca	172-175 (CDR2)
agct	177-180 (CDR2)
agtt	184-187 (CDR2)
agca	195-198 (CDR2)
ggca	220-223 (FR3)
ggtt	224-227 (FR3)

(a/t)(a/g)c(c/t) (wrcy)

Motif	Positions
agct	8-11 (FR1)
agcc	41-44 (FR1)
aact	59-62 (FR1)
agcc	72-75 (FR1)
tgct	99-102 (CDR1)
agct	177-180 (CDR2)
tgct	201-204 (FR3)
agcc	277-280 (FR3)
aacc	285-288 (FR3)
tact	307-310 (FR3)
tact	312-315 (FR3)

t(a/t) (tw)

Motif	Positions
tt	34-35 (FR1)
tt	82-83 (CDR1)
tt	88-89 (CDR1)
tt	135-136 (FR2)
tt	158-159 (FR2)
ta	165-166 (FR2-CDR2)
tt	167-168 (CDR2)
ta	180-181 (CDR2)
tt	186-187 (CDR2)
ta	199-200 (FR3)
tt	226-227 (FR3)
tt	246-247 (FR3)
ta	262-263 (FR3)
ta	304-305 (FR3)
tt	306-307 (FR3)
ta	312-313 (FR3)
ta	315-316 (CDR3)

V-REGION mutation statistics

Nucleotides

IMGT Label		V-REGION	FR1-IMGT	CDR1-IMGT	FR2-IMGT	CDR2-IMGT	FR3-IMGT	CDR3-IMGT
Total nucleotides with gaps		312 (318)	78	36	51	30	117	0 (6)
Mutations	Total	25 (30)	2	4	2	9	8	0 (5)
	Silent	8	2	1	1	3	1	0
	Non silent	17 (22)	0	3	1	6	7	0 (5)
Transitions	a>g	3 (4)	0	0	1	1	1	0 (1)
	g>a	6	2	1	0	1	2	0
	c>t	1 (2)	0	0	0	0	1	0 (1)
	t>c	4	0	1	1	1	1	0
	a>c	0 (1)	0	0	0	0	0	0 (1)
	c>a	1	0	1	0	0	0	0
	a>t	3	0	0	0	2	1	0
	t>a	1	0	1	0	0	0	0

V-REGION mutation statistics

Amino acids

IMGT labels			V-REGION	FR1-IMGT	CDR1-IMGT	FR2-IMGT	CDR2-IMGT	FR3-IMGT	CDR3-IMGT
Total amino acids (AA) with gaps			104 (106)	26	12	17	10	39	0 (2)
Identical AA			89	26	10	16	5	32	0
AA changes	Total		15 (17)	0	2	1	5	7	0 (2)
	Conserved IMGT AA classes (hydropathy, volume, chemical)	(- - -)	3 (4)	0	1	0	2	0	0 (1)
		(+ + +)	2	0	0	0	1	1	0
		(+ - -)	7	0	1	1	2	3	0
		(- + -)	1 (2)	0	0	0	0	1	0 (1)
		(- - +)	0	0	0	0	0	0	0
		(+ + -)	0	0	0	0	0	0	0
		(+ - +)	2	0	0	0	0	2	0

Who are the IMGT/V-QUEST users?

1. IG and TR genome diversity and evolution studies

- genome diversity and genome evolution studies of the adaptive immune responses
- veterinary research (analysis of IG and TR repertoires in farm, wild life and model animal species)

Comparison of IG and TR from newly sequenced genomes is done versus complete known loci. For example: non-human primates vs human, rat vs mouse, etc.

2. Biotechnology related to antibody engineering

- single chain Fragment variable (scFv)
- phage displays
- combinatorial libraries
- chimeric, humanized and human antibodies

Humanization of antibodies based on standardized human FR-IMGT and non-human CDR-IMGT allows to decrease immunogenicity and to maintain specificity.

Who are the IMGT/V-QUEST users?

3- Fundamental and medical research

Repertoire analysis of the IG antibody sites and of the TR recognition sites:

- in normal situations
- in pathological situations (autoimmune diseases, infectious diseases, AIDS, leukemias, lymphomas, myelomas)

Diagnostics

- clonalities in leukemias, lymphomas
- detection and follow up of residual diseases

Prognostic factor in Chronic Lymphocyte Leukemia (CLL)

- “unmutated” IGH V-REGION (identity > 98%): bad prognostic
- “mutated” IGH V-REGION (identity < 98%): good prognostic

ERIC, the European Research Initiative on CLL (www.ericll.org) recommands IMGT/V-QUEST for such studies

IMGT, the international ImMunoGeneTics information system®

http://imgt.cines.fr

The IMGT team at Montpellier, France

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- MARIE Network

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