IMGT, the Informatics Framework to Support Effective Collaboration in Complex Environments

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IMGT®, the international ImMunoGeneTics information system®
http://imgt.cines.fr
Outline

• IMGT® domain of expertise
• IMGT-ONTOLEGY axioms and concepts
• IMGT/V-QUEST, IMGT/JunctionAnalysis, IMGT/3Dstructure-DB
• Conclusions and Perspectives
Vertebrates

Immunoglobulin (IG)

T lymphocyte

B lymphocyte

T cell receptor (TR)

MHC

Trimolecular complex

IMGT® domain: the adaptive immune response
Bone marrow

Blood

Lymph nodes, spleen

V-D-J and V-J rearrangements

Hypermutations, selection

http://imgt.cines.fr
Spacefill 3D representation of an IgG

- VH: V-DOMAIN of the immunoglobulin heavy chain
- VL: V-DOMAIN of the immunoglobulin light chain
- CH1, CH2, CH3: C-DOMAIN of the immunoglobulin heavy chain
- CL: C-DOMAIN of the immunoglobulin light chain

VH corresponds to the V-D-J-REGION (in green (V), orange (D)) of the heavy chain. VL corresponds to the V-J-REGION (in green (V) and yellow (J)) of the light chain.

The Immunoglobulin FactsBook, 2001
Structural domains

IG and TR

V-DOMAIN

C-DOMAIN

MHC

G-DOMAINs

http://imgt.cines.fr
Contribution of the 2 V-DOMAINs to the antigen binding site

**Immunoglobulin (IG)**

- **V-DOMAIN**
  - **V-J-REGION**
  - **V-D-J-REGION**
- **Heavy chain**
- **Light chain**
- **Membrane IgM**

**T cell receptor (TR)**

- **V-J-REGION**
- **V-DJ-REGION**
- **Alpha** - **Beta**
- **Gamma** - **Delta**

*The Immunoglobulin FactsBook, 2001*
Immunoglobulin IgG

Immunoglobulin (IG) synthesis

Genomic DNA (IGH Locus 14q32)

5'   V   D   J   C   3'

Rearranged DNA

mRNA

5'   2 x 10^{12} different IG per individual   3'

2 x 10^{12} different IG per individual

Immunoglobulin (IG) synthesis

150
FUNCTIONAL IG GENES

HEAVY CHAIN

V  D  J  C
5'  38 - 46  x  23  x  6  3'

LIGHT CHAIN

V  J  C
5'  30 - 35  x  5  29 - 33  x  4 - 5  Kappa  Lambda  3'

6300  POTENTIAL RECOMBINATIONS

N-DIVERSITY
SOMATIC MUTATIONS

x 1000

ABOUT 6.3 x 10^6 POSSIBILITIES

185 +165  POTENTIAL RECOMBINATIONS

ABOUT 3.5 x 10^5 POSSIBILITIES

2 x 10^12
DIFFERENT ANTIBODIES

IMGT-ONTOLOGY seven axioms:

To share, reuse and represent knowledge in Immunogenetics and Life Sciences

Giudicelli and Lefranc, Bioinformatics 1999
CLASSIFICATION axiom

- group
  - subgroup
    - gene
      - allele
  - locus
    - IGLV
      - IGLV2
        - IGLV2-11
          - IGLV2-11*02
    - human IGL (22q11.2)

« Concepts »

« Instances »

http://imgt.cines.fr
Homo sapiens
IGH locus on chromosome 14
at 14q32.33
IG and TR: 1538 genes and 2523 alleles (human, mouse)
The IMGT-ONTOLOGY main concepts of classification include ‘group’, ‘subgroup’, ‘gene’, ‘allele’.

They allowed to set up the nomenclature for IG and TR genes (V, D, J, C genes).

IMGT gene names were approved by HGNC in 1999 and entered in GDB, LocusLink and Entrez Gene (NCBI).

IMGT/GENE-DB is the international reference database for IG and TR genes (direct links from Entrez Gene NCBI).

DESCRIPTION axiom

PROTOTYPE for a V-GENE

Label 1         Label 2          Relations entre Labels
V-GENE          V-EXON
FR3-IMGT        CDR3-IMGT
L-PART1         DONOR-SPLICE
V-REGION        FR1-IMGT
V-REGION        CDR3-IMGT
IMGT/LIGM-DB

114,673 sequences from 210 species

IMGT-ONTOLOGY:

- 277 IMGT labels for sequences
- 285 IMGT labels for 3D structures

SO (Sequence ontology):

- 67 IMGT labels
DESCRIPTION axiom

• The IMGT-ONTOLOGY concepts of description comprise the standardized IMGT labels and relations.

• They have allowed to describe the IG, TR and MHC sequences and 3D structures, whatever the receptor type, the chain type, or the species.

• They are particularly useful to describe IG, TR, and MHC and their complexes (IG/antigen, TR/pMHC).

• It is possible to query the IMGT® databases (IMGT/LIGM-DB for sequences, IMGT/3Dstructure-DB for 3D structures) with IMGT labels.

• Sequence Ontology (SO) includes IMGT labels.
NUMEROTATION axiom

IMGT
Collier de Perles

CDR-IMGT lengths
[8.10.12]

alemtuzumab VH

IMGT Collier de Perles on two layers

CDR-IMGT lengths [8.10.12]

alemtuzumab VH

IMGT Collier de Perles

IMGT Web resources: 10 000 pages HTML

http://imgt.cines.fr
The eleven IMGT amino acid classes according to the physicochemical properties

<table>
<thead>
<tr>
<th>Volume classes</th>
<th>'Hydrophobic' classes</th>
<th>'Hydropathy' classes</th>
<th>Hydrophilic</th>
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<tbody>
<tr>
<td></td>
<td>in Å³</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Very large</td>
<td>189-228</td>
<td>F</td>
<td>K</td>
</tr>
<tr>
<td>Large</td>
<td>162-174</td>
<td>W, L</td>
<td>R</td>
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<tr>
<td>Medium</td>
<td>138-154</td>
<td>V, M</td>
<td>H, E</td>
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<tr>
<td>Small</td>
<td>108-117</td>
<td>C</td>
<td>D, Q</td>
</tr>
<tr>
<td>Very small</td>
<td>60-90</td>
<td>A</td>
<td>N</td>
</tr>
</tbody>
</table>

- **Aliphatic**
- **Sulfur**
- **Hydroxy**
- **Basic**
- **Acidic**
- **Amide**

Pommié et al. J. Mol Recognit. 17, 17-32, 2004
IMGT Collier de Perles amino acid profile

CDR-IMGT lengths
[8.10.12]

alemtuzumab VH

Pommié et al. J. Mol Recognit. 17, 17-32, 2004
Towards «Potential immunogenicity evaluation »

Comparison with human germline genes:

- Percentage of identity of the V-REGION
- Percentage of identity of the 4 FR-IMGT (91 VH positions, 89 V-KAPPA positions)
- Number of amino acids with IMGT class change in the 4 FR-IMGT ('class' refers to the 11 IMGT physicochemical properties aa classes)

<table>
<thead>
<tr>
<th>Antibody</th>
<th>IGHV Gene</th>
<th>Percentage</th>
<th>Identity (%)</th>
<th>Class Change</th>
<th>Count</th>
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</thead>
<tbody>
<tr>
<td>alemtuzumab</td>
<td>IGHV4-59*01</td>
<td>73 %</td>
<td>84.61 % (77/91)</td>
<td>2/91</td>
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<tr>
<td></td>
<td>IGKV1-33*01</td>
<td>86.32 %</td>
<td>97.75 % (87/89)</td>
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<tr>
<td>bevacizumab</td>
<td>IGHV7-4-1*02</td>
<td>72.40 %</td>
<td>74.72 % (68/91)</td>
<td>1/91</td>
<td></td>
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<tr>
<td></td>
<td>IGKV1-33*01</td>
<td>87.40 %</td>
<td>92.13 % (82/89)</td>
<td>2/89</td>
<td></td>
</tr>
<tr>
<td>trastuzumab</td>
<td>IGHV3-66*01</td>
<td>81.63 %</td>
<td>90.10 % (82/91)</td>
<td>0/91</td>
<td></td>
</tr>
<tr>
<td></td>
<td>IGKV1-39*01</td>
<td>86.32 %</td>
<td>93.25 % (83/89)</td>
<td>1/89</td>
<td></td>
</tr>
</tbody>
</table>
NUMEROTATION axiom

• The IMGT-ONTOLOGY concepts of numerotation include IMGT unique numbering and IMGT Collier de Perles for V-DOMAIN (IG and TR).

• They have been extended to the C-DOMAIN (IG and TR) and G-DOMAIN (MHC).

• They have allowed to bridge the gap between sequences and 3D structures in IMGT/3Dstructure-DB.

• They are used for mutations, polymorphisms, CDR-IMGT lengths, contact analysis, potential immunogenicity evaluation and paratope definition.

• WHO-INN programme requires the CDR-IMGT lengths for antibody.
IMGT/V-QUEST: analysis of IG and TR sequences

WELCOME!

to the IMGT/V-QUEST Search page

THE
INTERNATIONAL
IMMUNOGENETICS
INFORMATION SYSTEM®


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

Analyse your Immunoglobulin nucleotide sequences

- Human
- Mouse
- Chondrichthyes

- Teleostei
  - Atlantic cod
  - Channel catfish
  - Rainbow trout
- Sheep

Analyse your T cell Receptor nucleotide sequences

- Human
- Mouse
- Non-human primates
IMGT/V-QUEST: analysis of IG and TR sequences

Analyse your Immunoglobulin sequences

Your selection: Human

Your sequences are compared to the Human IG set from the IMGT/V-QUEST reference directory sets

Nucleotide sequences

Enter your sequence(s) in FASTA format (FASTA format is required):

>AY393054
gtggttttctttttcattttatgaaggtcgccgagtgctgtgggaccagggcttggtacagccagggcgggtccctgagactccctgtgcagctcttggattgacctttttggttatctttagctgtgctgccaccgctcaggggaaagggactggaatggtagtcataaatagacaggtgggaacagaaatatttggtatgataagggcagattatcatcactccctgagagattttccaaaaggtctgctattttgcaatgtgacccggagacacagcatatattctgaatgactgtgcttatctaccagggacatcttccgagctgtgggccctggaccctcaagggctcttgggggccacagcgccttggtctggttcctggtcaaggaactacccc

Or give the path access to a local file containing your sequence(s) in FASTA format (FASTA format is required):

Start | Clear the form

Analysis by batches of up to 50 sequences in a single run

http://imgt.cines.fr
IMGT/V-QUEST: analysis of IG and TR sequences

Selection of parameters for the results

Display type: HTML
Nb of nucleotides per line in alignment: 60

A. Detailed view
1. Alignment for V-GENE
2. Alignment for D-GENE
3. Alignment for J-GENE
4. Results of IMGT/JunctionAnalysis
   - with full list of eligible D-GENE
   - without list of eligible D-GENE
5. Sequence of the JUNCTION CDR1 and CDR2
6. V-REGION alignment according to the IMGT numbering
7. V-REGION translation
8. V-REGION mutation table
9. V-REGION mutation statistics
10. V-REGION mutation hot spots
11. IMGT Collier de Perles
   - Links to IMGT Collier de Perles
   - IMGT Collier de Perles (PNG format, slow)
   - no IMGT Collier de Perles
12. Sequences of (V-J or V-D-J) REGION with gaps in FASTA
Access to IMGT/PhyloGene for V-REGION
13. Annotations by IMGT/Automat

B. Synthesis view
1. Alignment for V-GENE
2. V-REGION alignment according to the IMGT numbering
3. V-REGION translation
4. V-REGION protein display
5. V-REGION protein display (with color)
6. V-REGION protein display (mutations displayed)
7. V-REGION most frequently occurring AA
8. Results of IMGT/JunctionAnalysis

Advanced parameters
Selection of IMGT reference directory set
- F:GRF in frame P
- With all alleles
- With allele *01 only

Selection of parameters for IMGT/JunctionAnalysis
- Nb of D-GENE in IGJ JUNCTIONs (default is 1)
- Number of accepted mutations
  - default in 3V-REGION
  - default in D-REGION
  - default in 5U-REGION

More options for Detailed view
- Nb of nucleotides to exclude in 5' of the V-REGION for the evaluation of the nb of mutations (in results 8 and 9)
- Nb of nucleotides to add (or exclude) in 3' of the V-REGION for the evaluation of the alignment score (in result 1)
3. V-REGION translation

```
GI  5  10  15
E  Q  L  V  Q  S  C  A  E  K  K  P
X56368 IGHV5-51*03
gag gta gta gta gta gta gga 
seq1 --- --- -gc --- --- --- --- ... --- --- --- ---
--- --- --- --- --- --- --- --- ... --- --- --- ---
--- --- --- --- --- --- --- --- ... --- --- --- ---
seq3 --- -g- --- --- -a- --- --- --- ... -e --- --- --- ---
--- --- --- --- --- --- --- --- ... --- --- --- ---
seq4 --- --- --- --- --- --- --- --- ... --- --- --- ---
--- --- --- --- --- --- --- --- ... --- --- --- ---
--- --- --- --- --- --- --- --- ... --- --- --- ---
seq5 --- -g- --- --- -a- --- --- --- ... -e --- --- --- ---
--- --- --- --- --- --- --- --- ... --- --- --- ---
--- --- --- --- --- --- --- --- ... --- --- --- ---

-------------------------- FR1 - IMGT
20  25  30
G  E  S  L  K  I  S  C  K  G  S  G  Y  S  F
X56368 IGHV5-51*03
gyy gag tct gty ayt tcc tgt ayt gtt tct gga tac aqc tgt
seq1 --- --- --- --- --- --- --- --- --- --- --- ---
seq2 --- --- --- --- --- --- --- --- --- --- --- ---
seq3 --- --- --- --- --- --- --- --- --- --- --- ---
seq4 --- --- --- --- --- --- --- --- --- --- --- ---
seq5 --- --- --- --- --- --- --- --- --- --- --- ---
seq6 --- --- --- --- --- --- --- --- --- --- --- ---
```
**IMGT/V-QUEST: advanced parameters**

### Selection of parameters for the results

**Display type:** HTML

- **Nb of nucleotides per line in alignment:** 60

#### A. Detailed view

1. **Alignment for V-GENE**
2. **Alignment for D-GENE**
3. **Alignment for J-GENE**
4. **Results of IMGT/JunctionAnalysis**
   - with full list of eligible D-GENEs
   - without list of eligible D-GENEs
5. **Sequence of the JUNCTION (nt and AA)**
6. **V-REGION alignment according to the IMGT numbering**
7. **V-REGION translation**
8. **V-REGION mutation table**
9. **V-REGION mutation statistics**
10. **V-REGION mutation hot spots**
11. **IMGT Collier de Perles**
   - Links to IMGT Collier de Perles
   - IMGT Collier de Perles (P16 format, slow)
   - no IMGT Collier de Perles
12. **Sequences of V-, D- and V(D)J-REGION (nt and AA) with gaps in FASTA**
   - Access to IMGT/Phylotree for V-REGION (nt)
13. **Annotations by IMGTautomat**

#### B. Synthesis view

1. **Alignment for V-GENE**
2. **V-REGION alignment according to the IMGT numbering**
3. **V-REGION translation**
4. **V-REGION protein display**
5. **V-REGION protein display with color**
6. **V-REGION protein display (mutations displayed)**
7. **V-REGION most frequently occurring AA**
8. **Results of IMGT/JunctionAnalysis**

### Advanced parameters

**Selection of IMGT reference directory set:** FORF+ in frame P

- **With all alleles**
- **With allele *01 only**

**Selection of parameters for IMGT/JunctionAnalysis**

- **Nb of D-GENE in VH JUNCTIONs (default 1)** Default: 5
- **Number of accepted mutations**
  - in 3V-REGION: Default
  - in D-REGION: Default
  - in 5V-REGION: Default

**More options for Detailed view**

- **Nb of nucleotides to exclude in 5’ of the V-REGION for the evaluation of the nb of mutations (results 8 and 9)** Default
- **Nb of nucleotides to add (or exclude) in 3’ of the V-REGION for the evaluation of the alignment score (result 1)** Default
V-DOMAINs: VH and V-KAPPA

VH
V-D-J junction

V-KAPPA
V-J junction

Mouse (Mus musculus) E5.2Fv

CDR3-IMGT = Complementarity determining region (105-117)
V-J junction (104-118)
V-D-J junction (104-118)
Immunoglobulin V-D-J generation
of sequence diversity

3′V-REGION  N-REGION  D-REGION  N-REGION  5′J-REGION

tgtgcaaaaaga tac agcatatttggt gtggactgtat tcc gat acaactgttccg actccctgg

JUNCTION

C A P Y R G D T Y D Y S W
tgt gcg cca tac cgg ggt gac act tat gat tac tcc tgg

http://imgt.cines.fr
## IMGT/JunctionAnalysis Results

**Locus**
- IGH

**Species**
- Homo sapiens

**IMGT repertoire link**
- Locus representation

### Maximum number of mutations:
- V-REGION: 2
- D-REGION: 4
- J-REGION: 2

### Deletion criterion:
- Using patterns

### Best D gene choice for a same score:
- Less mutations

## Description of the JUNCTIONs

Click on mutated (underscored) nucleotid to see the original one.

<table>
<thead>
<tr>
<th>Input</th>
<th>V name</th>
<th>V-REGION</th>
<th>D-REGION</th>
<th>N1</th>
<th>N2</th>
<th>J-REGION</th>
<th>J name</th>
<th>D name</th>
<th>Vmut</th>
<th>Dmut</th>
<th>Jmut</th>
<th>Mgc</th>
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**http://imgt.cines.fr**
### JUNCTION alignments with translation and IMGT AA classes

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

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<th>104</th>
<th>105</th>
<th>106</th>
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**#1 seq1**

tct tgt acc gcc gtc gct tat ... ... ... ... tgc gac gca gac gcc tcc cag cag cag tgg

| C   | Y   | K   | P   | T   | B   | D   | D   | D   | G   | H   | P   | A   | E   | Y   | F   | Q   | Q   | V   |     |

**#2 seq2**

tct tgt acc gcc gtc gat gat gat gcc ... ... ... ... gac gac gca tgc tcc cag cag tgg

| C   | Y   | G   | G   | S   | A   | Y   |     |     |     |     |     |     |     |     |     |     |     |     |     |

**#3 seq3**

tct tgt acc gcc gtt ccc tat gat gat gcc ... ... ... ... tgc gac gca gcc gcc tcc cag cag tgg

| C   | Y   | G   | G   | S   | A   | Y   |     |     |     |     |     |     |     |     |     |     |     |     |     |

**#4 seq4**

tct tgt acc gcc gtt ccc tat gat gat gcc ... ... ... ... tgc gac gca gcc gcc tcc cag cag tgg

| C   | A   | K   | Q   | B   | P   | P   | E   | Y   | S   | S   | A   | Y   | H   | D   | G   | W   | F   | D   | P   | V   |

**#5 seq5**

tct gac aca aca aac ccc ccc gcc tat gct gac gca tat gat gat gcc gcc tcc cag cag cag tgg

| C   | A   | F   | E   | M   | Y   | S   | S   | G   | G   | S   | Y   | Y   | P   | P   | D   | A   | F   | E   | L   | V   |

**#6 seq6**

tct gac aca gac aty ctc tat gct tgg ggc ggt tat gcc gcc tcc ccc gtt gct gcc tgg cgg cgg cgg tgg

| C   | A   | F   | Q   | B   | P   | P   | E   | Y   | S   | S   | A   | Y   | H   | D   | G   | W   | F   | D   | P   | V   |

**#7 seq7**

tct gac aca gac aty ctc tat gct tgg ggc ggt tat gcc gcc tcc ccc gtt gct gcc tgg cgg cgg cgg tgg

| C   | A   | F   | Q   | B   | P   | P   | E   | Y   | S   | S   | A   | Y   | H   | D   | G   | W   | F   | D   | P   | V   |

**#8 seq8**

tct gac aca gac aty ctc tat gct tgg ggc ggt tat gcc gcc tcc ccc gtt gct gcc tgg cgg cgg cgg tgg

| C   | A   | F   | Q   | B   | P   | P   | E   | Y   | S   | S   | A   | Y   | H   | D   | G   | W   | F   | D   | P   | V   |

**#9 seq9**

tct gac aca gac aty ctc tat gct tgg ggc ggt tat gcc gcc tcc ccc gtt gct gcc tgg cgg cgg cgg tgg

---

Analysis of the 3D structures

THANK YOU for using IMGT/3Dstructure-DB

IMGT/3Dstructure-DB card for: 1ce1

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<tr>
<th>IMGT protein name</th>
<th>IMGT receptor type</th>
<th>IMGT receptor description</th>
<th>Ligand(s)</th>
<th>Species</th>
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Experimental technique: X-ray diffraction
Resolution (in angstrom): 1.90
PDB release date: 25-JUN-99

Contact analysis:

IMGT/3Dstructure-DB Domain pair contacts (overview) of 1ce1

Atom contact types
- Non covalent
- Covalent
- (BB) Backbone/backbone
Access to atomic pair contacts in IMGT/3Dstructure-DB

Click on residue in IMGT Collier de Perles (or in amino acid sequence)

http://imgt.cines.fr
### Atomic pair contacts in IMGT/3Dstructure-DB

**41V - TRP (W)**

chain : 1u8k_B

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- **Tot**: Total number of atomic pair contacts
- **NCo**: Number of non covalent atomic pair contacts
- **Pol**: Number of polar atomic pair contacts
- **HB**: Number of hydrogen bonds
- **NPol**: Number of non polar atomic pair contacts
- **Cov**: Number of covalent links (other than chain covalent links)
- **SS**: Number of disulfide bridges
Hydrogen bonds (IMGT Collier de Perles on 2 layers)

Collier de Perles: HUMAN IGHV V-DOMAIN from B12 (PDB: 1hzh_H)

[8.8.20]

## Contacts VH-(Ligand), V-KAPPA-(Ligand)

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<th>IMGT description</th>
<th>Chain ID</th>
<th>IMGT chain description</th>
<th>Domain number</th>
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### Contact Table

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<th>Unit 2 Domain</th>
<th>Unit 2 Chain</th>
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</table>
Contacts VH-(Ligand)

Contacts of VH 1ce1_H with (Ligand) 1ce1_P

Atom contact types
- Non covalent
- Polar
- Hydrogen bond
- Non polar

Atom contact categories
- Non covalent
- Covalent
- Disulfide
- Backbone/backbone
- Side chain/side chain
- Backbone/side chain
- Side chain/backbone

Summary:

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List of the Residue@Position pair contacts:
Click 'R@P' for IMGT Residue@Position cards

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Click 'R@P' for IMGT Residue@Position cards

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Kaas Q. et al
IMGT unique numbering

**V-DOMAIN** (IG, TR)
AND
**V-LIKE-DOMAIN**
(other than IG, TR)

**C-DOMAIN** (IG, TR)
AND
**C-LIKE-DOMAIN**
(other than IG, TR)

**G-DOMAIN** (MHC)
AND
**G-LIKE-DOMAIN**
(other than MHC)

Immunoglobulin superfamily (IgSF)

MHC superfamily (MhcSF)
CONCLUSIONS and PERSPECTIVES

Three IMGT® biological approaches: genetic, genomic, structural. Knowledge is based on the IMGT-ONTOLOGY axioms and concepts.

On the informatics side: building of IMGT-Choreography on interactions between IMGT® components to answer biological questions.

The same axioms and concepts are valid for a multiscale (molecule, cell, organ, organism) and systemic approach (system immunobiology).

Duroux P. et al. Biochimie in press
Many thanks to the IMGT® team at Montpellier, France