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MHC Genes Class I, Class II in Invertebrates: The Echinodermata

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ABSTRACT

For the first time MHC Class I and ClassII genes were described in Echinodermata, (Invertebrates).That seems incredible for developmental immunobiology studies.

HLA-DRB1, HLA -DQB1 genes from Class II; HLA-E, HLA-B genes from Class I have been found in genomes of Ophiocomina nigra(Ophuirids) and Antedon bifida (Crinoïds) : Two Echinodermata which present the IPA (Invertebrate Primitive Antibody).

INTRODUCTION

As C.A Janeway wrote in 2001 (Ref 1) :

"The function of MHC molecules is to bind peptide fragments derived from pathogens and display them on the cell surface for recognition by the appropriate T cells. The consequences are almost always deleterious to the pathogen—virus-infected cells are killed, macrophages are activated to kill bacteria living in their intracellular vesicles, and B cells are activated to produce antibodies that eliminate or neutralize extracellular pathogens. Thus, there is strong selective pressure in favor of any pathogen that has mutated in such a way that it escapes presentation by an MHC molecule.

Two separate properties of the MHC make it difficult for pathogens to evade immune responses in this way. First, the MHC is polygenic: it contains several different MHC class I and MHC class II genes, so that every individual possesses a set of MHC molecules with different ranges of peptide-binding specificities. Second, the MHC is highlypolymorphic; that is, there are multiple variants of each gene within the population as a whole. The MHC genes are, in fact, the most polymorphic genes known

Because of the polygeny of the MHC, every person will express at least three different antigen-presenting MHC class I molecules and three (or sometimes four) MHC class II molecules on his or her cells . In fact, the number of different MHC molecules expressed on the cells of most people is greater because of the extreme polymorphism of the MHC and the codominant expression of MHC gene products.

The term polymorphism comes from the Greek *poly*, meaning many, and *morphe*, meaning shape or structure. As used here, it means within-species variation at a gene locus, and thus in its protein product; the variant genes that can occupy the locus are termed alleles. There are more than 200 alleles of some human MHC class I and class II genes, each allele being present at a relatively high frequency in the population. So there is only a small chance that the corresponding MHC locus on both the homologous chromosomes of an individual will have the same allele; most individuals will be heterozygous at MHC loci. The particular combination of MHC alleles found on a single chromosome is known as an MHC haplotype. Expression of MHC alleles is codominant, with the protein products of both the alleles at a locus being expressed in the cell, and both gene products being able to present antigens to T cells. The extensive polymorphism at each locus thus has the potential to double the number of different MHC molecules expressed in an individual and thereby increases the diversity already available through polygeny .

In addition to the highly polymorphic 'classical' MHC class I and class II genes, there are many genes encoding MHC class I-type molecules that show little polymorphism; most of these have yet to be assigned a function. They are linked to the class I region of the MHC and their exact number varies greatly between species and even between members of the same species. These genes have been termed MHC class IB genes; like MHC class I genes, they encode β2-microglobulin-associated cell-surface molecules. Their expression on cells is variable,

both in the amount expressed at the cell surface and in the tissue distribution"

In human, the main function of major histocompatibility complex (MHC) Class II molecules, is to present processed antigens which are derived primarily, from exogeneous sources.

Constitutive expression of MHC Class II molecules, is also confined to professional antigen- presenting cells (APC) of the immune system (Ref.2)

Since we have discovered the IPA (Invertebrate Primitive Antibody)(Ref. 3,4,5,6), to acquire a better understanding of the invertebrate immune system , it seemed useful to look for MHC class I, class II genes in invertebrates with Ophiocomina nigra (Ophuirids), Antedon bifida (Crinoïds) as model of studies.

MATERIALS AND METHODS

Animals

Ophiocomina nigra (Ophuirid) Antedon bifida(Crinoïd) were obtained at the station « Of Biologie Marine of Roscoff » France.

Obtention of Ophuirid and Crinoïd mRNA

Digestive coeca were excised from their bodies and mRNA were obtained from Uptizol (Interchim) then quality controls were operated. (Ref.7)

Sequencing

Sequencing was made on Illumina Next Seq 500 with paired-end : 2. 75 bp

Transcriptome was assembled from RNA-Seq fastq files using Trinity v2.1.1 (Ref.8) with default parameters. A BLAST database was created with the assembled transcripts using makeblastdb application from ncbi-blast+ (v2.2.31+). The sequences of transcripts of interest were then blasted against this database using blastn application from ncbi-blast+ (Ref.9) with parameter word_size 7.

RESULTS

First,it is shown a table in which are presented characteristics of HLA-E (MHC Class I)

HLA-DQB1 (MHC Class II) genes in Ophiocomina nigra (Ophuirids).Sequences are following

QueryID	Query Name	SubjectID	Identity (%)	Length	Mismatch	Gapopen	Query cover (%)	E-value	Bitscore
NM_005516.6	HLA-E	TRINITY_DN6320_c0_g1_i1	78,63	117	22	2	4,00	2,00E-12	75,00
NM_002123.4	HLA-DQB1	TRINITY_DN20883_c0_g1_i1	90,91	33	3	0	2,00	8,00E-04	45,40

>TRINITY_DN6320_c0_g1_i1 HLA-E

5'AGAGGACACGTCATTCTGAGCGTAAGG
GCCGCAGCGAAAGGTGGCAGGGCCCGC
GCTTTT
AAAGGCTGAAATCCCGGCGGCTCAGGCCT
GTCGTTTCCAGCACTTTGGGAGGCCCAGG
AA
GATGGATCGCTTGAGGCCAGGAGTTCGG
GACTAGCCTGGCCAACATGGTGAACACC
CGTC
TCTACTAAAAATAGATCGGAAGAGCGTC
GTGTT3'

>TRINITY_DN20883_c0_g1_i1 HLA-DQB1

5'GTAAAACAGCATTTCATCTGAAAAGAAA
TTCAATGTCCAAAGTTCAAAAACTCTGTG
AAG
ACTTGAATGCAAAAAGTACTCAAGTCCAT
CACATATTTGGCATTTTTAGATATGATCTTC
CAAAGATTTTAAAATAAAACAAAAGAAAA
ACCAAAAGAAGAAAAAAATTTAACAAAA
AAA
TAAAGGGCCAAAAAAAATTTTAAAAAAA
AAAAAACCCCCATTTTTTTTGGGTCTAAA
AAA
AAAAAAAAAAAAAAAATCGC3'

Two genes:HLA-E, HLA-B (ClassI) appear in the following table in Antedon bifida

QueryID	Query Name	SubjectID	Identity (%)	Length	Mismatch	Gapopen	Query cover (%)	E-value	Bitscore
NM_005516.6	HLA-E	TRINITY_DN19334_c8_g2_i1	88,15	287	28	4	11,00	2,00E-91	337,00
NM_005514.8	HLA-B	TRINITY_DN15013_c0_g1_i1	100,00	21	0	0	1,00	3,70E-02	39,90

The sequences in 5'-3' follow:

>TRINITY_DN19334_c8_g2_i1 HLA-E

5'TGTAATCCCAGCACTTTGGGAGGCCGA
GGCGGGCGGATCACGAGGTCAGGAGATC
GAGAC
CATCCTGGCTAACACAGTGAAACCCCGT
CTCTACTAAAAATACAAAAAATTAGCCGG
GCG
TGGTGGCGGGCGCCTGTAGTCCCAGCTAC

TCGGGAGGCTGAGGCAGGAGAATGGCGT
GAA
CCCGGGAGGCGGAGCTTGCAGTGAGCCG
AGATCGCGCCACTGCACTCCAGCCTGGGC
GAC
AGAGCGAGACTCTGTCTCAAAAAAAAAA
AAAAAAAAAAAAA3'

>TRINITY_DN15013_c0_g1_i1 HLA-B

5'GCCGAATATGATGCAGAGGTATCAGGGG
GTGAAGCATCTGGAGGTGAGGTATCGGC
AGGA
GAGGCATCTGGGGGAGAAGCTGAACAAT
CTGACAATGAAAGCGATTAGATAACATTT
TTT
TAATTCTAGTTGCAGCCTAAATATTTCGAT
ATTACTTTTTTTTACTAGTTGAATGATTAA
CAAAAGAAAGCAACAACTGTGGTAATAT
TGCTAATTATGAAATGAAAAATGTTTAAT
GTG
GCCCTGACACTAAATTGTAAACTGTTTTT
TAGTAATAAGAATTTCAATAGCTTCTCTG
AA
AGAAGATGTCTCTGAGAGAGTAATATTTG
ACAGGTTCAGTGTATTTAAAGACTTATAA
TG
TAAAGCAGAGATGTAACTAGAGAAACCT
AGATATTGATGTCAACAAACTAAGGGTGC
ATG
GAAAATGTGAAAGACTTTAAGAGTGGGT
GACCCTGCCTACCAACACAATTCAATCCA
TGT
TTGAGGCTTTTTTTCATTAGCCTAATAGTG
AAGTCAGTGGCGTAAGGCCCCCTTGTTTA
G
CACTCCTAAGGGTCCCTAATGATGGATAA
TTGTATTGGGCTCTTCATGCTCTGGGGCC
CT
GGGTTTAGCTAGTGAGTGCTCATAGCAGT
TGGCTGGGCAAGGTTAGAAAGCAATGGT
TCT
GTGCAGACATTTGCATTTAATTGACCAAT
ATTTCAAATCGTGTGTTACACAGGAATCA
TA
ACCTAATCAGCAGTTGTTTTTAATAAACA
TTGCATCTTGGTCGACGTAATATTGTTATGG
ACTGTCTGTGAAACCATGTGAATCTAAAC
TCTTAAAAATGCCTGCCTCTCCTGTCCTT
GC
TAAATATAAATTTGTTTTCTCAATTAGGCG

GCCCTGACACTAAATTGTAAACTGTTTTT
TAGTAATAAGAATTTCAATAGCTTCTCTG
AA
AGAAGATGTCTCTGAGAGAGTAATATTTG
ACAGGTTCAGTGTATTTAAAGACTTATAA
TG
TAAAGCAGAGATGTAACTAGAGAAACCT
AGATATTGATGTCAACAAACTAAGGGTGC
ATG
GAAAATGTGAAAGACTTTAAGAGTGGGT
GACCCTGCCTACCAACACAATTCAATCCA
TGT
TTGAGGCTTTTTTTCATTAGCCTAATAGTG
AAGTCAGTGGCGTAAGGCCCCCTTGTTTA
G
CACTCCTAAGGGTCCCTAATGATGGATAA
TTGTATTGGGCTCTTCATGCTCTGGGGCC
CT
GGGTTTAGCTAGTGAGTGCTCATAGCAGT
TGGCTGGGCAAGGTTAGAAAGCAATGGT
TCT
GTGCAGACATTTGCATTTAATTGACCAAT
ATTTCAAATCGTGTGTTACACAGGAATCA
TA
ACCTAATCAGCAGTTGTTTTTAATAAACA
TTGCATCTTGGTCGACGTAATATTGTTATGG
ACTGTCTGTGAAACCATGTGAATCTAAAC
TCTTAAAAATGCCTGCCTCTCCTGTCCTT
GC
TAAATATAAATTTGTTTTCTCAATTAGGCG3

CONCLUSION

MHC class I and classII genes exist in Echinodermata, at least in Echinodermata which possess a sophisticated immune system as Asterids, Ophuirids and Crinoïds. It would be interesting also to study MHC system in Echinodermata which present only innate immune response as Echinids, Holothurids (Ref.10)

The HLA-DQB1 gene is a part of a family of genes called the human leukocyte antigen (HLA) complex.

The found HLA-B gene and HLA-E gene belong also to the HLA complex (MHC Class I): it helps the immune system to distinguish the body own proteins from proteins made by foreign invaders such as viruses and bacteria (Ref . 11, 12, 13)

Although all efforts to find in Invertebrates a Major Histocompatibility Complex System have been unsuccessful, we find to day, for the first time, a MHC class II gene (HLA-DQB1 gene) a MHC class I (HLA-B HLA-E genes)in Echinodermata: It's of great novelty.It's a fundamental discovery in the domain of Comparative and Adaptative Immunology.

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