Evidence of *Il2* and *Il6* Genes in the Crinoïd: *Antedon bifida* (Echinodermata)

Michel Leclerc^{1*}, Ariane Jolly², Pierre de la Grange²

- 1. Immunology of Invertebrates, University of Orleans, Sandillon, France.
- 2. GenoSplice Technology, iPEPS-ICM, Hôpital de la Pitié Salpêtrière, Paris, France.

Submitted 10 Jun 2018; Accepted 142 Jul 2018; Published 17 Aug 2018

For the first time, in 1984, the concept of lymphokine-like substances was evoked in an Echinodermata: the sea star *Asterias rubens* (1). Later, in 1996, it was spoken of inteleukin (*Il*)2, *Il4*, and *Il6* in the same animal (2).

Immunogenomics contributed to determine evidence of the presence of *Il2* (3) and *Il4* genes in Echinodermata but not of *Il6* gene.

The aim of this work, was the research of this last one, in an ancestral Echinodermata: the *Antedon bifida* which belongs to the class of Crinoïds.

Antedon bifida was obtained at the station of Biologie Marine of Roscoff, France. Digestive coeca were excised from the A. bifida body. mRNA was obtained from Uptizol (Interchim, France), and quality control was operated. 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 (4) 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+ (5) with parameter word size 7.

Table 1 summarizes the obtained results with sea star *Il2* and *Il6* transcriptomes, when compared to Homo sapiens and Mus musculus ones. Figure 1 shows the corresponding sequences in 5'-3'direction.

Although *Il2* gene appears like something well-known in Echinodermata, *Il6* gene evidence was not yet related in invertebrates. Beck and Habicht spoke of Il6-like protein in 1996 in invertebrates (6). So it is the first time that *Il6* gene is found in invertebrates. Our work in *Antedon bifida*, a Crinoïd, an ancestral Echinodermata shows that in

Table 1. Il2 and Il6 transcriptome characteristics										
Query ID	Query Symbol	Species	Subject ID	Identity (%)	Length	Mismatch	Gapopen	Query cover (%)	E-value	Bitscore
NM_000 586.3	IL2	Home sapiens	TRINITY_DN20229 _c2_g8_i1	97,06	34	1	0	4	5,00E-08	58,4
NM_001 314054.1	IL6	Mus musculus	TRINITY_DN19147 _c3_g2_i1	96	25	0	1	2	0,026	39,9

^{*}Correspondence: Immunology of Invertebrates, University of Orleans, Sandillon, France. E-mail: mleclerc45@gmail.com

TRINITY DN20229 c2 g8 i1 (IL2)

5'ACACCCTCCATCTTGTAGCATAATTACTACAAAGATTAAGAAAAATCAAGCTGCTTAAG A

>TRINITY_DN19147_c3_g2_i1 (IL6)

5'AGTGAATTACGCAAAGTTTCTGATTGGTTCCAAATCAATAAATTATCGCTAAATGAATCC AAATCAAATTATATATATATAAACATAAAAACAAAAAATCCATCTCTCAAAACTTAAAA GTTTTAATTAATGACAAGGTAATCACACGTGTTGAGGAAACCAAATTTCTTGGCGTCTCC ATTGACTCAAAATTATCTTGGAAACATCATATCAATACAATCTGCAACACACTTGCGAGA TATAATACTTTAATTCTTCCGTACATATCATACTGCAACATTGCCTGGGCCGTTACACCT GGCAAGTTCAGCACTTTATGCCCCTGGACTAGTTTCGAAACTACCAATATTGACCGAATT TTTAAATTGCAAAAACGAGCAATCCGAATCTGTTCAGGGGCATCATTCAGATCTCATTCA AAACACTTATTTTTAACTTTAAATCTCTCAACGTCTTTGATGTTAATAAACTTCAAACT GCATTATTCATGTACCGACTTACAAATAATCTACTTCCGACCCACATATGTTCACTCTTT ACTAAACCTTCTGATACCCATACACATTTTACCCGTTTTTCCAATACTAATGTATACACT GTTAAAAACTGCGACTCGCACATTAGACGTCATTCAATACTTTATACCGGACCTAAACTT TGGGAAAGTCTTTCACATTCACTGATAAAGAAACCTTCACTTTTTAGTTTTAAAATTACA TATAAGCAATCTCTAATCCAAGCATATGATGAATAAGGTGTTATGATTATTTTTAATAAT GTTAATTTATTGTTATTCTATTTTTTATATATTATAATTTAGTCACAACTATTACTGATA TTTGTTATTCTCTAATTTTTACTAGGCATTTTGTACAGCAAATATCAAT3'

Figure 1. Il2 and Il6 sequences.

an original way.

In summary cytokine genes such as *Il1*, *Il2*, *Il4*, *Il6* exist in invertebrates. The present finding highlights the novelty of our work.

Conflict of interest

The authors declared no conflict of interest.

References

- 1. Leclerc M, Brillouet C, Luquet G, et al. Properties of cell subpopulations of starfish axial organ: in vitro effect of pokeweed mitogen and evidence of lymphokine-like substances. Scand J Immunol. 1981;14:281-4.
- 2. Legac E, Vaugier G L, Bousquet F, et al. Primitive cytokines

- and cytokine receptors in invertebrates: the sea star Asterias rubens as a model of study. Scand J Immunol. 1996;44:375-80.
- Leclerc M. The Problem of II2 in Echinodermata. Int J Vaccines Vaccin. 2018;5:17-8.
- 4. Grabherr M G, Haas B J, Yassour M, et al. Full-length transcriptome assembly from RNA-Seq data without a reference genome. Nat Biotechnol. 2011;29:644-52.
- 5. Altschul S F, Gish W, Miller W, et al. Basic local alignment search tool. J Mol Biol. 1990;215:403-10.
- 6. Beck G and Habicht G S. Characterization of an IL-6-like molecule from an echinoderm (Asterias forbesi). Cytokine. 1996:8:507-12.