

Platelets, Pecam1 Gene, Thromboxane Genes, in Invertebrates

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ABSTRACT

Three main points assert the evidence: of « platelets » in Echinodermata (Asterids, Ophuirids and Crinoïds (Invertebrates), of a new cytokine (Thromboxane A2) :

- a) the evidence of thromboxane genes.
- b) the appearance of T.E.M platelets, in the asterid, *Asterias rubens*.
- c) the presence of PECAM1 gene in ophuirids and crinoïds.

INTRODUCTION

The appearance of platelets, in *Asterias rubens* has been described (Ref.1 Fig 1) in invertebrates: They resemble blood platelets of vertebrates.

In the present work, we recall genes which are implicated in the physiology of vertebrate platelets:

Thromboxane A synthetase gene, Thromboxane A2 receptor gene, Pecam1 gene are the main genes which represent thromboxane activity in vertebrates.

This study reports T.E.M observations of sea star platelet, genomic research of Thromboxane genes and Pecam1 gene in Echinodermata genomes

MATERIALS AND METHODS

1) *Ophiocomina nigra* and *Antedon bifida* were purchased by the Laboratory of Roscoff (France). Digestive coeca were excised. RNA was extracted.

2a) *Ophiocomina nigra* and *Antedon bifida* mRNA were obtained (Ref.2) .Furthermore quality controls were made.

2b) Sequencing : Transcriptome was assembled from RNA-Seq fastq files using Trinity v2.1.1 (Ref.3)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 .4) with parameter word_size 7.

RESULTS

We observe a sea star *Asterias rubens* platelet in T.E.M (Ref.1, Fig.1) Evidence of Platelets in Echinodermata

On the other hand, a table summarizes the genomic results. TBXA2R represents the human thromboxane A2 receptor gene, TBXAS1 the human synthetase one

Query ID	Query Name	Subject ID	Identity (%)	Length	Mismatch	Gapopen	Query cover	E-value	Bitscore
NM_00106 0.5	TBXA2R	TRINITY_D N38594_c0_ g1_i1	77,93	222,00	34,00	9,00	13,00	2,00E-27	124,00
NM_00106 1.5	TBXAS1	TRINITY_D N22549_c0_ g1_i1	84,21	38,00	6,00	0,00	2,00	1,60E-01	38,10

The sequence of the TBXA2R transcriptome is following :

```
>TRINITY_DN38594_c0_g1_i1
5'ATATATCATATATGATATAGTACCTTTGTT
ATATATCATAATACATATAAATGTGTATTA
```

```
TGTTATCTATAATTATATAATTTTCATATATAA
GATGTATAATATGTATCATATATTATAT
ATGTTATGTAATATATATAGTATATATAAGA
TGACACAGGATAAATATTATATACTATGA
CATATAAATATATGAGGTTATATGTTACAT
ATAAGGCATAGCACATAACATGTAATATA
```

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TATCATATATAATTTTTTTTTTAGACAGAATC
TTGTCCTGTTGCACAGGGTGGGGTACAAT
GGCGCCATCTTTGCTCACTGCAACTTCTG
CCTCACGGGTCCAAGCGATTGTCCTCCCT
CA
GCCTCCCAGGTAGCTGGGACTACACCAC
ACTGGGACTACACCAGCTGCCACCATGC
CTAG
CTAATTTTTGTATTTTTGGTAGAGACAGG
GTTTTGCCGTGTTGCCAGGCTGGTAGAT
CG3'

Second TBXS1 transcriptome is given:

>TRINITY_DN22549_c0_g1_i1
5'AAATAAGCATACGCATGGAAGAATCAC
TCAGATTTTTATGTTAAATAGGAGGA
ACTTAGA
AAACACCAAGTGTGGATTTGGAGAATTT
TGTAATAACTTAACCAAAGACAATGCCTAA
TCA
CATTGAGGGCAACATAAGTGGCACTATGT
GTGTCATCGGCTCAACAGTTCATTCATCA
TC
ATCGGGATCTAACAAAATGACACATTGTA
GGCATAATCATAACAGGACTCGGCGTAGG
TT
ATCAGCAACAGCTATGATTGGAGTACTCG
GAGGA3'

At last we have a special look on the PECAM1 transcriptome sequence which was found in *Ophiocoma nigrum* genome, It exists also in *Antedon bifida* one :

5'ATATATCATATATGATATAGTACCTTT
GTTATATATCATAATACATATAAATGTGT
ATTA
TGTTATCTATAATTATATAATTTTCATATA
TAAGATGTATAATATGTATCATATATTAT
AT
ATGTTATGTAATATATATAGTATATATAA
GATGACACAGGATAAATATTATATACTA
TGA

Evidence of Platelets in Echinodermata

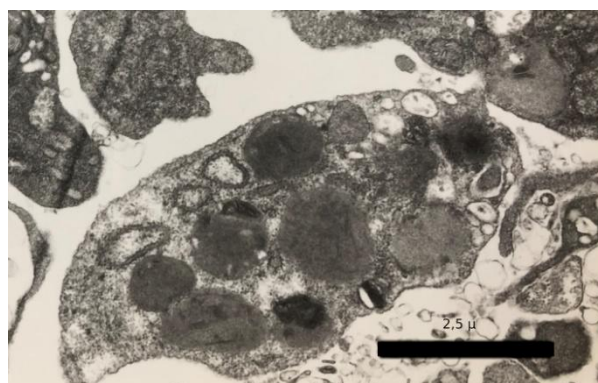


Figure 1

CATATAAAATATATGAGGTTATATGTTA
CATATAAGGCATAGCACATAACATGTAA
TATA
TATCATATATAATTTTTTTTTTAGACAGAA
TCTTGTCTGTTGCACAGGGTGGGGTAC
AAT
GGCGCCATCTTTGCTCACTGCAACTTCTG
CCTCACGGGTCCAAGCGATTGTCCTCCCT
CA
GCCTCCCAGGTAGCTGGGACTACACCAC
ACTGGGACTACACCAGCTGCCACCATGC
CTAG
CTAATTTTTGTATTTTTGGTAGAGACAGG
GTTTTGCCGTGTTGCCAGGCTGGTAGAT
CG
G3'

DISCUSSION AND CONCLUSION

Thromboxane A2 gene, we found in ophiurids (Echinodermata) induces a cytokine in human (Ref 5). Thromboxane A2 produced by activated platelets, has prothrombotic properties. It stimulates activation of new platelets as well as increases platelet aggregation (Ref.5). Pecam1, we found also in ophiurids and crinoïds, has a special rôle in vascular biology and platelet function in human (Ref.6).

Genomic results assert the evidence of :

- a) a new cytokine in invertebrate : the thromboxane
- b) a new gene in these last ones : the Pecam1 gene

Furthermore TEM results (Ref.1 Fig.1) show structures which resemble blood platelets. In conclusion, it was clearly shown that platelets, Pecam1 gene and thromboxane cytokine coexist in Echinodermata : It's of special interest in Comparative Hematology.

At our knowledge red blood cells don't exist in Invertebrates but just lymphocytes and platelets!

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