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The TNFR In *Asterias rubens* Sea Star: Genomic Studies

Michel Leclerc[1] and Nicolas Kresdorn[2]*

[1]*Department of Cell Biology, Developmental Biology, Immunology, University of Orléans, Orléans, France*
[2]*GenXPro, Frankfurt, Germany*

Abstract

The sea star *Asterias rubens* includes TNFR, in its genome, when compared to mouse one: mouse isoform 2 of TNF AIP3-interacting protein 1, mouse TNF receptor-associated factor 2, mouse TNF receptor-associated factor 3, mouse TNF receptor-associated factor 4, mouse TNF receptor-associated factor 6. Many of these factors are bound to NF Kappa -B genes which have recently been isolated in the *Asterias rubens* genome. They play a rôle in the regulation of the sea star immune system.

Keywords: *Asterias rubens*; Genome; Sea stars

Introduction

Tumor necrosis factor receptor (TNFR)- associated factors belong to a family of intracellular adaptor proteins that mediate signaling downstream of various cell surface receptors, including members of the TNFR superfamily [1].

TNFR have been described in mammals, Drosophila and Caenorhabditis elegans and are characterized by conserved structural motifs [2].

TNFR are related to detective activation of kinases and/or transcription factors such as NF Kappa-B. NF Kappa-B genes have been discovered recently in *Asterias rubens* [3].

In the present paper, we research TNFR, in immunized and non-immunized sea stars to HRP, in their genome, when compared to mouse genome.

Materials and Methods

a) Sea stars were obtained from the Biology Institute (Gothenbugh University). Immunizations to HRP, Genomic studies were already described [4].

b) After ligation of adapters for Illumina 's GSII sequencing system, the cDNA was sequenced on the Illumina GSII sequencing.

c) 100 bp from one side of the approximately 200 bp fragments sequences were assembled using Velvet [5].

Results

First results concern non-immunized sea stars to HRP: (a) Evidence of isoform 2 of TNFAIP3-interacting protein 1 in sea star *Asterias rubens*:

One contig (Contig2216) could be annotated via BLASTX to Mus musculus "Isoform 2 of TNFAIP3-interacting protein 1" from the Swissprot database (TNIP1_MOUSE), with an e-value of 2.33e-17. On an aligned region of 406 amino acids, 193 positive and 101 identical amino acids were found.

5'ACGTCCTTAGCAACACCCTTGACAACAGCCGTAGCCGT

AGCCGGCAATTCCAACACGGCTTGGCTTGAGTCTGCAAC

AATGCGTTGTACTTCTCTTTGCAAGTTTCTACATTTGTCT

AGACAGCTGCAATTAGTTGACGCAAAGATAACCTTTATA

AAACAATTTCGTAACTAATCAACATATTTGCTAGACTTG

ATTTCAAGTCTGAGACTGCGGTTATTTCTCTGCATCAGCC

ATGAAGAATTCC 3'

b) Evidence of TNF receptor-associated factor 2 in sea star *Asterias rubens*:

One contig (Contig15544) could be annotated via BLASTX to Mus musculus "TNF receptor-associated factor 2" from the Swissprot database (TRAF2_MOUSE), with an e-value of 5.52e-49. On an aligned region of 137 amino acids, 112 positive and 86 identical amino acids were found.

5'ATTGATCTTCCAGAGCAGCACCCCGTCGTAGGATGACG

CCAAGAGGGAGTATCGGGTAAAACCACTTCATTCTACTC

GCCTGTTTTCTACACCAGCCGACATGGCTATAAGCTCTC

TGCAAGGATCTACTTGAATGGAGATGGGTTAGGGAAAC

ACACGCACATGTCGCTTTTCTTTGTGGTCATGCGAGGGC

GCCATGACGCTCTCCTTCCGTGGCCTTTCCACCAGAAGG

TGACCTTCATGCTGATGAATCAGAACAACCGGGAGCACG

TCATTGACGCCTTCAGACCAGACCCTGCAAGCTCCTCGT

TCAAACGGCCCAACAGTGACATGAATGTTGCTTCCGGAT

GTCCGCTCTTCATTGCGCAGGAAGTCCTGGATTCAACTG

ACAGTTTCATCAAAGACGACACAATGTTTGTCAAAGTCG

TTGTCGATAATGTTGGATTGGACAACATTTGAACAATGA

CAGTGACAAAGCCGTGTCCAAATTGGTAATCACAGCTGT

GGCTACGACAGGATTTCTGCGTCATCATGTAATATAGCA

TCCTGTATAGCAACAGTTGTAGCAATAGCTGTAGCCAGC

***Corresponding author:** Nicolas Kresdorn, GenXpro de. Frankfurt, Germany, E-mail: mleclerc45@gmail.com

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AGTTCGAATATGACCCGACTTTGCTGGATACAAGTGGTG

CTTATTTTAAATAGTGGCAAGTTGCCCCCAAATGTCTTCG

TATGCATAATTGATGCGATGTGAAGATTTAACACAAGCT

GTGAGTTCGCACCAAATGTGTGACGCTCGGCTGAAGTTA

AACATTTTTTTCTGTAGTCCACGGGAAAAAAATTTATAAC

AGGTGACTCT 3'

c) Evidence of TNF receptor-associated factor 4 in immunized sea star *Asterias rubens* to HRP :

One contig (Contig12096|m.10269) could be annotated via BLASTX to Mus musculus "TNF receptor-associated factor 4" from the Swissprot database (TRAF4_MOUSE), with an e-value of 5.38e-14. On an aligned region of 132 amino acids, 67 positive and 41 identical amino acids were found.

5'CGAATGCAGTGGACTTGCATGCAGTCTTGCTCGTTTTT

CATGACATGGCCCTAGCCCCGGTGGTCTTACACTTTCGT

ACTGTATCTGTGTACAGAGGAATAGTCGTATAGGGCTAG

ACATGGCCCTAACATTCAATCGTGATCGCTGACCAGACA

TCAGACTTGACTTTATACACAGCGTGCATGACTTTGGAA

GAATACTGACCATTTTATGAAAACATCTTTCTTCTTCTTT

CTTCTTGCAGGTCAAAGATCACGTAGAGAACGACTGCCC

CAAGGCGAAGATACCCTGCCCATTTCAAACTATGGGTTG

TGACTTTAAGGGTGAACGTCAAAAGATGCAGAAACACGT

GCAAGACGAGCCAGTGGAGCATCTGACCTACGTAGGGG

ATTCGATGCTGAAGCACGCTACACTACTGGAGAAACACA

AGGAGATGCTTGTTGACCATAAGGAGTCTTTGGGCACGT

GCGTGCAGAAGGTCCGCGATCTTGAGATGCTGTACGGCT

CACAGCTCGTCTGGAAAATCGACCGCTACGCCGAGAGGA

TGCAGGAGGCAAAGACCAACAAGAAAGTGACCATCTTC

AGCCCGCCATTTCTAACCAGTCGACATGGTTACAAGATG

ACGGTATCGGCTTGTCTGAACGGCGAC 3'

Discussion and Conclusion

It is apparent that, with the exception of *Asterias rubens* and, to a lesser degree, Drosophila, our understanding of invertebrate TNFR system, is still lacking in detail.

It is true that Liu [2] has identified a Drosophila TNF receptor-associated factor:DTRAF1, but not really the mouse TNF receptor-associated factor 2 which is found in *Asterias rubens*: we may speak of homologies between these factors.

In the same manner a homolog of mouse isoform 2 of TNFAIP3-interacting protein 1, discovered in *A.rubens*, exists in Drosophila. This factor inhibits NF Kappa-B activation and TNF-induced NF Kappa-B dependent gene in vertebrates. We recall that NF Kappa-B genes have been discovered recently in *Asterias rubens* [3].

At last, an enigmatic TNF receptor-associated factor 4 appears in immunized sea stars to HRP and not in non-immunized sea stars. We have already observed such phenomenon due to the immunization : it remains unanswered.

In conclusion, such a sophisticated defense system(Invertebrate primitive antibody, TNFR system) has all saved the sea star to survive for more than 200 million years on the earth.

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