

The Sea Star Igkappa Gene: Effects against Human Cancerous Cells. New Aspects

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Abstract

It was shown 32 years ago that the sea star axial organ cells (AO cells) produced a spontaneous cytotoxicity against mouse cancerous cells.

Recently, we discovered a sea star Igkappa gene with immune properties. This gene was inserted in a CMV (cytomegalovirus) and finally in a plasmid called « young » plasmid.

The induced « young » protein exerted a spontaneous cytotoxicity against osteosarcoma cells (U2oS cells) and recently against Hela cells (cervix carcinoma cells).

Keywords: Sea Star; Igkappa Gene; Human Cancerous Cells

Introduction

In 1983, Luquet and Leclerc [1] shown that the axial organ cells (AO cells), exerted a spontaneous and induced cytotoxicity against mouse SP2 myeloma cells and MBL2 cells.

The AO cells included essentially lymphocytes and phagocytes [1].

30 years later, we discovered a sea star Igkappa gene [2], with immune properties [3].

The aim of the present work was to study the behaviour of the « young » protein secreted by the sea star Igkappa gene, an anti HRP protein, in front of human malignant (U2oS, Hela cells) and healthy cells (human dendritic cells), by the use of plasmids.

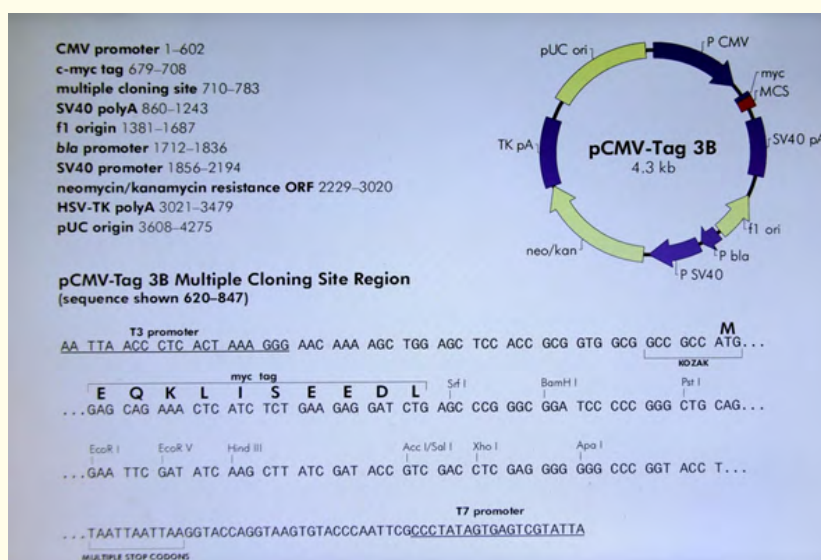
Materials and Methods

Gene cloning in a cytomegalovirus (CMV) was done in Germany (Eurofins Genomics, figures 1 and 2), from the sea star Igkappa gene [2], an anti-HRP gene. It constitutes the « promoter ». We recall the importance of anti-HRP epitopes synthesis and their relevance in Invertebrates [3].

Following steps as plasmid realization in correlation with the promoter, plasmid amplifications, transfections [4] were performed in the laboratory of DR S. Ruchaud° (CNRS, FRANCE) and Pr C. Pichon°° (CNRS, FRANCE) U2oS human cells, Hela human cells (cervix carcinoma cells) Dendritic cells as controls were used. They were transfected by plasmids, after electroporation, at time t = 0 they were observed at t = 24 h at t = 48 h: reactions were blocked to be estimated in western blots, as already described [5].

Cloning in N-terminal pCMV-Tag3B (c-mic tag)/ BamH1-EcoR1

GGA TCC GGA GGA **ATG** CGTGGCAACATGGCGTCTCTATGGATGTTCTTCTT
 TGTCGTGGGGATAACTTTACAACGGAGTTTGGCGATTACACGTTTCGCG
 AGCAACCGTCGGACACTAGCGCGTTGAGGGGAGCACAGTGGTGCTTCAC
 TGCTCCGTTGAGCAGTACATAAACACCACGGCCATCGTTTGGTGAGCCG
 TGACTCGGTCATCAGCCACAACAAGACCTGAAACTGTCCAGTCTAAACA
 CCGACCAGCTCAAAGGTACTCGATTTTCAGGCGACGCATCTCGGGGGAA
 TTCAACCTTAAAATAGTGAACCTTACCGCCACAGACGCCGCCAGTTACCG
CTGTCAGATG TAA GAA TTC



Figures 1 and 2: The pCMV-Tag 3B.

Results

The protein « young », also named: invertebrate primitive antibody seem to exert a spontaneous cytotoxicity 24 hours after transfection against osteosarcom cells (U2oS cells) against Hela cancerous cells (48 hours after transfection: 50 % cytolysis).

Western blots do not confirm, in the present time, at the level of transfected cells, the protein expression, for unknown reasons. But we estimated that the protein anti-HRP may be present in the supernatant of transfected cells. Further experiment is necessary to valid this hypothesis. From another point of view, the young protein exerts also a weaker spontaneous cytotoxicity (30 % cytolysis) against dendritic cells used as controls.

Conclusion

These results have proven to be of particular importance and could open the way to immune field in human cancer therapy.

It is a preliminary work and we attempt, now, to test other lineages of human malignant cells and healthy cells. About these last ones, we suggest to use another lineage of human control cells because dendritic cells are implicated in immune response (Antigen presenta-

tion) in man and sea star Igkappa gene and young protein belong to the sea star primitive antibody. So, the possible interaction between this type of cells and a primitive antibody could be problematic.

The cytolysis of human cancerous cells by the « young protein » would be spectacular if we could spread this property to other types of human cancerous cells, as it was shown for mouse malignant cells [1] with AO cells.

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