Comparative genomics of RNA-binding proteins RBFOX

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INTRODUCTION

RBFOX2 is a tissue specific factor regulating alternative splicing. The RNA-binding domain (RRM) of this protein recognizes the sequence **5'-UGCAUGU-3'** in the intron flanking the regulated exon. The RBFOX family also includes homologues of RBFOX2 - RBFOX1 and RBFOX3, which have different levels of expression in different tissues.

Human RBFOX2 gene is described to contain three so-called poison exons. The inclusion of such exons leads to nonsense-mediated mRNA decay of the transcript. There is no such data for its homologues

AIMS

The aims of this work were

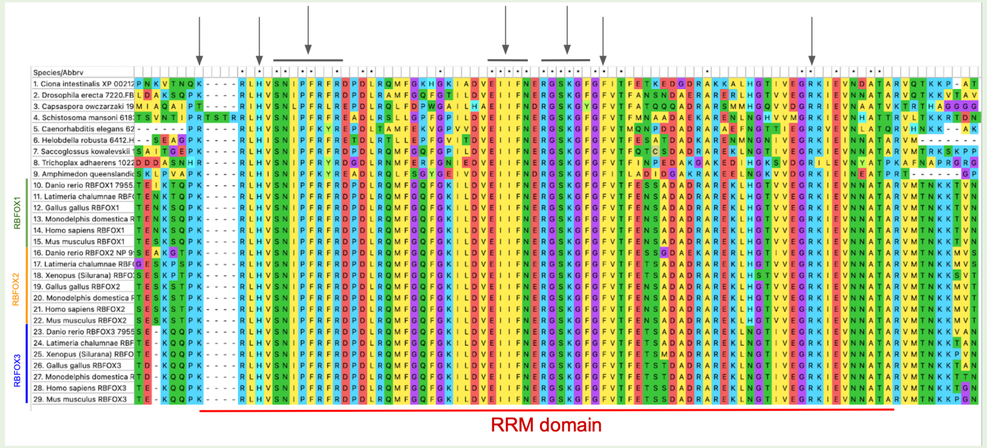
- to analyze the phylogeny of the RNA-binding proteins RBFOX
- to study the evolutionary conserved poison exons in those proteins

METHODS

- eggNOG 5.0
- Python
- PyMOL
- Jalview
- GenBank

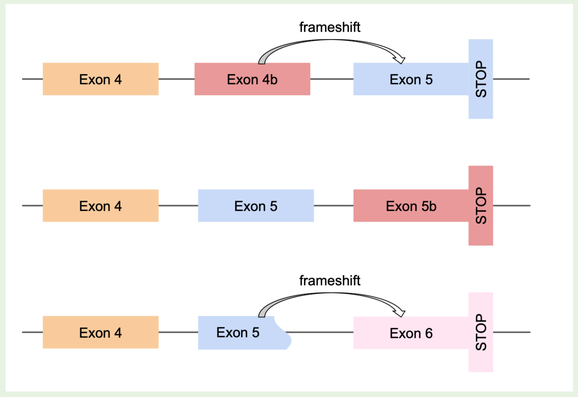
Generated by ImageFX for the request “A poison exon hiding in the ocean of intron”

RESULTS

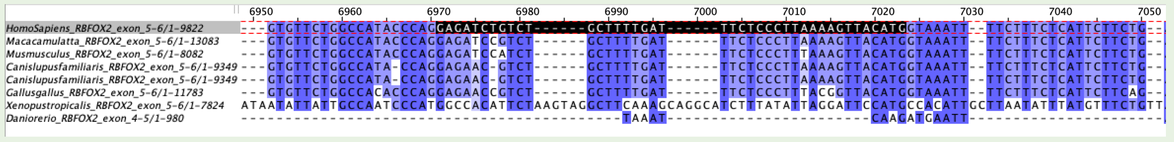


Amino acid sequence alignment showed that the proteins are very conserved among vertebrates: only 4 substitutions occurred in the RNA-binding domain in RBFOX3 compared to RBFOX1 and RBFOX2. In invertebrates, the amino acid sequence is much more variable. Gray arrows indicate amino acid residues involved in RNA binding.

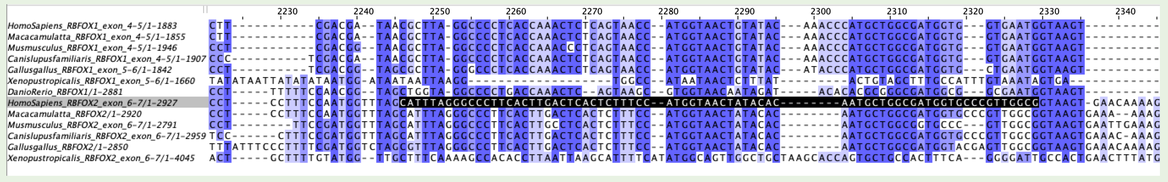
Representation of variable positions (red) on the three-dimensional structure of the RRM domain in complex with RNA target (purple)



In human RBFOX2, three NMD (nonsense-mediated mRNA decay) isoforms are annotated: in one case, NMD is caused by shortening of the fifth exon, which leads to a reading frame shift; in the other two, poison exons are inserted after the fourth and fifth exons, respectively.



The alignment of the introns, homologous to the fourth intron of human RBFOX2, revealed a conserved region in mammals and birds, but not in other classes of vertebrates. It suggests that the poison exon might have appeared in the common ancestor of Vertebrata, but was lost by amphibian and fish. *The poisonous exon 4b annotated in human RBFOX2 is highlighted in black.*



Moreover, in RBFOX1 there also seems to be a poison exon, homologous to 5b in human RBFOX2, judging by their high identity. *The poison exon 5b annotated in human RBFOX2 is highlighted in black.*

CONCLUSIONS

RBFOX proteins are highly conserved in vertebrates with only 4 varying position in the RNA-binding domain. These positions do not participate in the RNA binding.

The poison exon 4b annotated in human RBFOX2 was traced back to mammals and birds, but not amphibian and fish. As for 5b, it was also found in mammalian and avian RBFOX1.