PKR binds dsRNA

part of “Genomik der Genregulation”

Sonja Prohaska

Computational EvoDevo
University Leipzig

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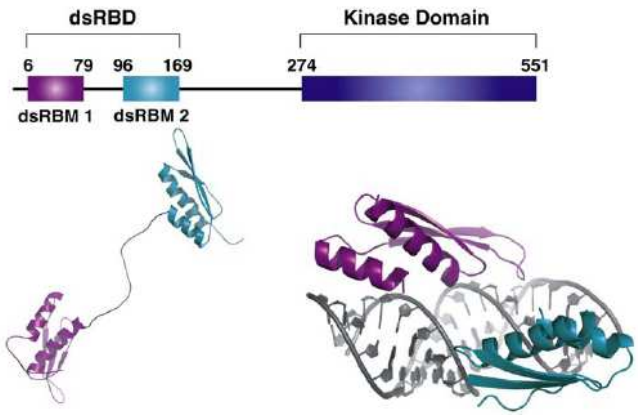
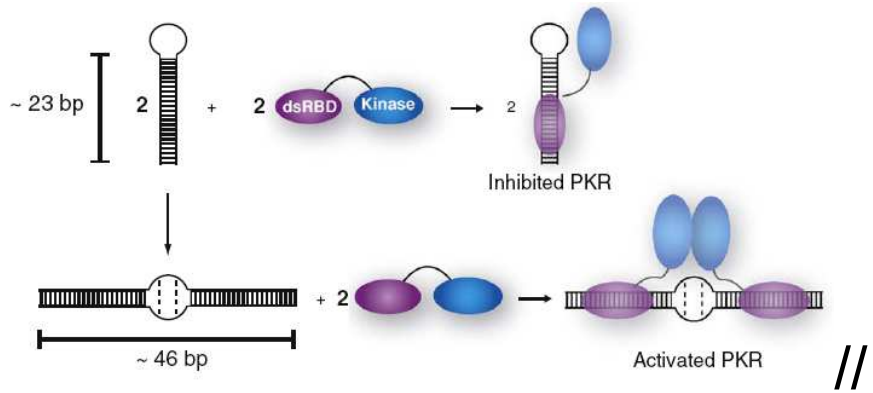
PKR – Protein Kinase RNA-activated

- tandem copies of dsRBD (double-stranded RNA binding domain) and kinase domain (which catalyzes phosphorylation)
- in general, the dsRBDs bind into the wide, minor groove of A-form RNA-helix and multiple dsRBDs can pack along the length of a helix
- min. 33bp RNA helices are needed for activation of PKR
- shorter dsRNAs inhibit PKR through competitive binding





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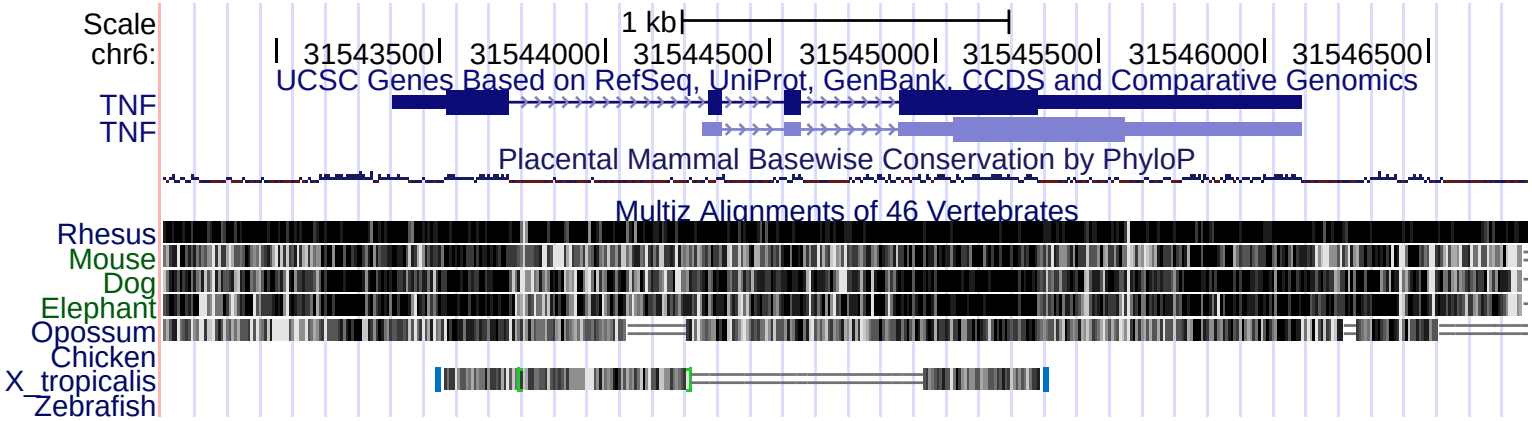
Function of PKR

- PKR is involved in innate immune response and protects against viral infection
- dsRNA is produced during viral infection and is used to destinguish self from non-self
- binding to dsRNA activates PKR by inducing dimerization and a subsequent auto-phosphorylation reactions
- once activated PKR phosphorylates the translation initiation factor eIF2α causing inhibition of translation
- positive feedback loop: PKR activates NFκB, NFκB upregulates expression of interferon, interferon activates PKR

TNFα – tumor necrosis factor α

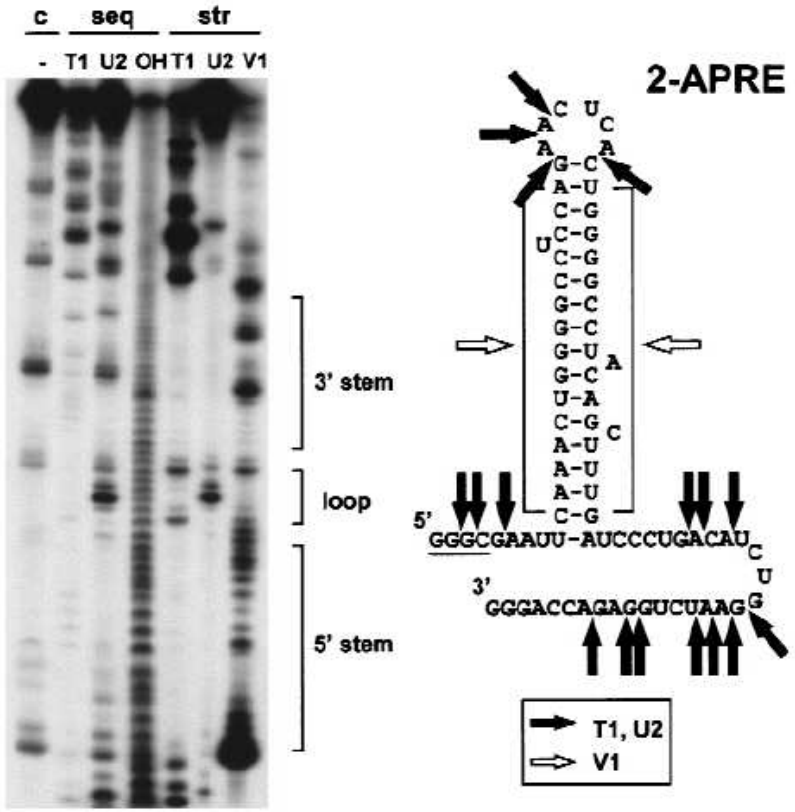


- a cytokine involved in systemic inflammation
- able to induce fever and apoptosis but inhibit viral replication
- has a RNA motif in its 3'-UTR that binds PKR

Splicing of TNFα is dependent on PKR binding

- *cis*-acting elemtent in the 3'-UTR is designated 2-APRE
- evidence
  - when 2-APRE is present, splicing is sensitive to inhibition by PKR inhibitors or transdominant-negative mutant PKR
  - increase expression of wtPKR enhances splicing efficiency of 2-APRE containing transcripts
  - in *vitro* 2-APRE forms a stable, 17bp stem-loop structure that strongly activates PKR
- physiological signals that cause activation of PKR should promote splicing of TNF-*alpha* precursor transcripts
- PKR regulates splicing of TNF*alpha*

2-APRE

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H.sapiens       GAAUUCAAACUGGGGCCUCCAGAACUCACUGGGGCCUACAGCUUU-GAUCC
S.scrofa        GAAUUGGAACUGGGGCUUCCAGA-CUCGCUGGGGUCCUUGGGUUUGGAUUC
O.cuniculus     GCAUUCAAACUGAGGCUUCCAGGACUCACUGGGGCCUUCAGAACUCCAUUC
B.taurus        ----------------UCCAGAACUCCCUGGGGUCCACAGCUU-------
C.hircus        ------------GGGCUCCAGAAGUUGCUGGUGCCU--------------
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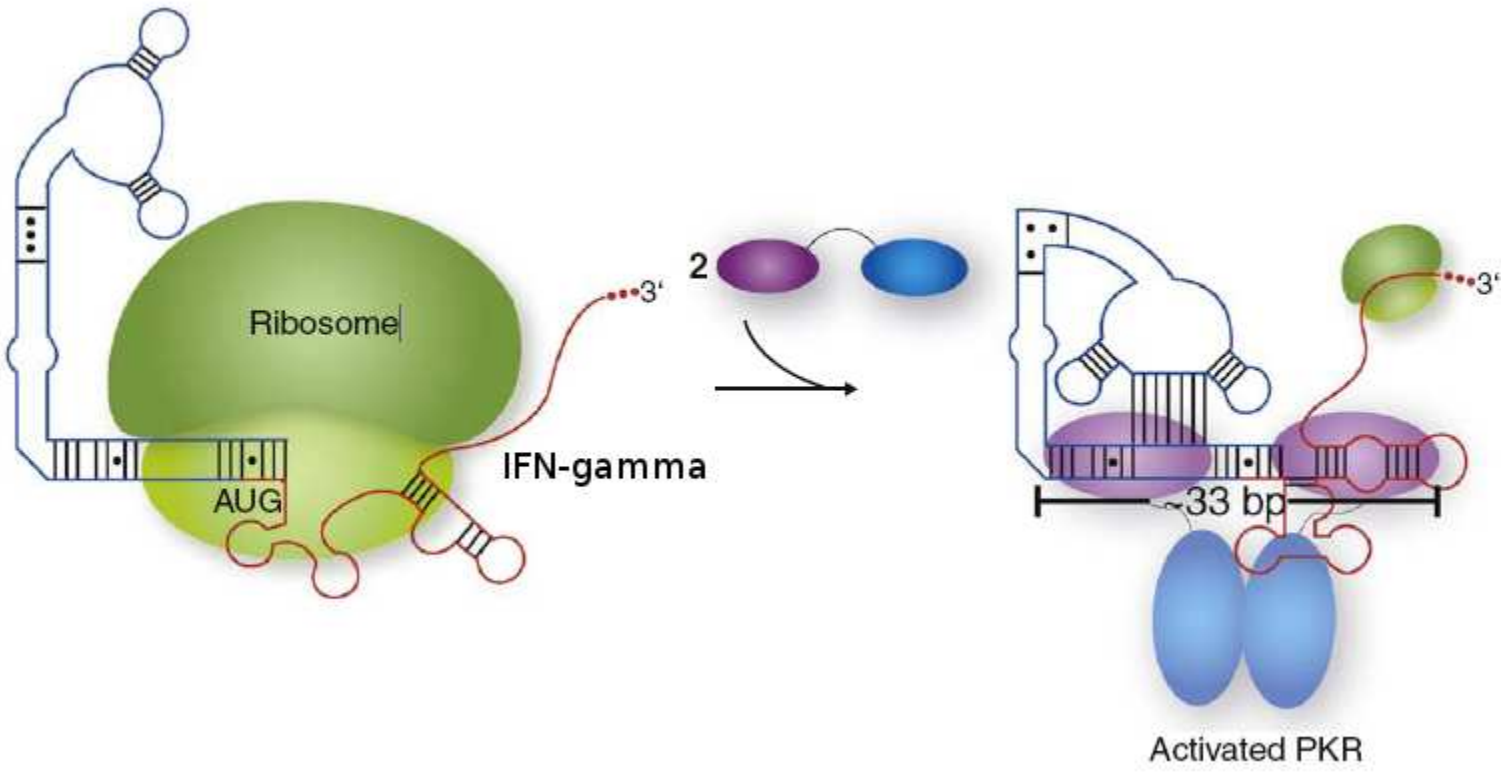
IFN-γ – interferon γ

- PKR and the ribosome compete for binding to the IFN-γ 5'-UTR
- the 5'-UTR forms a pseudoknot and coaxial stacking generates the binding interface for PKR



Literature

- [Nallagatla, 2011] Subba Rao Nallagatla, Rebecca Toroney and Philip C Bevilacqua. *Regulation of innate immunity through RNA structure and the protein kinase PKR*. Curr. Op. Struct. Biol. 2011. 21, p119-127.
- [Cohen-Chalamish, 2009] Smadar Cohen-Chalamish, Anat Hasson, Dahlia Weinberg, Lise Sarah Namer, Yona Banai, Farhat Osman and Raymond Kaempfer. *Dynamic refolding of IFN-γ mRNA enables it to function as PKR activator and translation template*. nature chem. biol. 2009. 5:12, p896-903.
- [Osman, 1999] Farhat Osman, Nayef Jarrous, Yitzhak Ben-Asouli and Raymond Kaempfer. *A cis-acting element in the 3'-untranslated region of human TNF-α mRNA renders splicing dependent on the activation of protein kinase PKR*. Genes and Development. 1999. 13, p3280-3293.