Who ARE you?

In search for novel AUBP targets

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Key Points

- ARE & AUBPs
- Diplomathesis & AREsite
- Findings so far
- PhD-thesis brain-dump

ARE

- AU-rich element
- Cis-acting regulatory elements found at 3' UTR of approximately 8% of human protein coding genes
- Motifs range from AUUUA to WWWWAUUUAWWWW
- AMD (ARE mediated decay) ⇒ mRNA stability control

ARE

- Class I ARE contain several dispersed copies of the AUUUA motif within U-rich regions
- Class II ARE contain at least 2 overlapping UUAUUUA(U/A)(U/A) nonamers
- Class III ARE are U-rich regions that do NOT contain the AUUUA pentamere

AMD

- ARE mediated decay
- AUBP bind ARE, recruit deadenylation factors ⇒ destabilizing
- Inhibit interaction of factors/AUBPs ⇒ stabilizing

AMD

- Degradation of an AU-rich binding protein AMD targeted mRNA begins with deadenylation
- A missing poly-A tail prevents translation of a mRNA ⇒ exonucleolytic attack
- Followed by decapping, the attack of a 3' - 5' exonuclease-complex (exosome)
  or 5' - 3' exonucleases via processing-bodies (p-bodies)
  degrades the AUBP target

AUBPs

- AUF1, HuR, TTP

AUF1

- AUF1 (A+U rich RNA-binding factor1, heterogeneous nuclear ribonucleoprotein D)
- Plays a role in mRNA decay and translation control
- Binds class I and class II AREs via RRMs
- Shuttles between the cytoplasm and the nucleus
- Four protein isoforms, lots of protein-protein interactions
- Hard to link destabilizing or stabilizing effects to one of them

HuR

- HuR (human antigen R)
- Binds the nonamer NNUUNNUUU
- Shuttles between the cytoplasm and the nucleus

HuR

- Contains three RRM motifs
- Two N-terminal binding to ARE motifs
- The remaining C-terminal motif binding to poly-A tails
- Stabilizing the RNA-protein complex

TTP

- Tristetraprolin
- Destabilizing effect on mRNA target
- Predominantly found in the cytoplasm
- Binds core UUAUUUAUU of class II AREs, promotes deadenylation and degradation
- Tandem zinc fingers can bind to adjacent 5' -UAUU- 3' subsites on the single-stranded RNA

AUBPs in general

All three AUBPs can be found to interact or compete with each other for single stranded target sites, sometimes having an agonistic and sometimes an antagonistic effect on the stability of their targets.

They can have a great influence on the half-life of mRNAs, providing the cell with a fast response mechanism to environmental or developmental conditions

Diplomathesis Goals

- Create AREsite database and webserver
- Published ⇒ rna.tbi.univie.ac.at/AREsite

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- Only tip of the iceberg

The Plan

- Have a look at human first ⇒ more known targets
- Focus on AUUUA ⇒ ARE core motif

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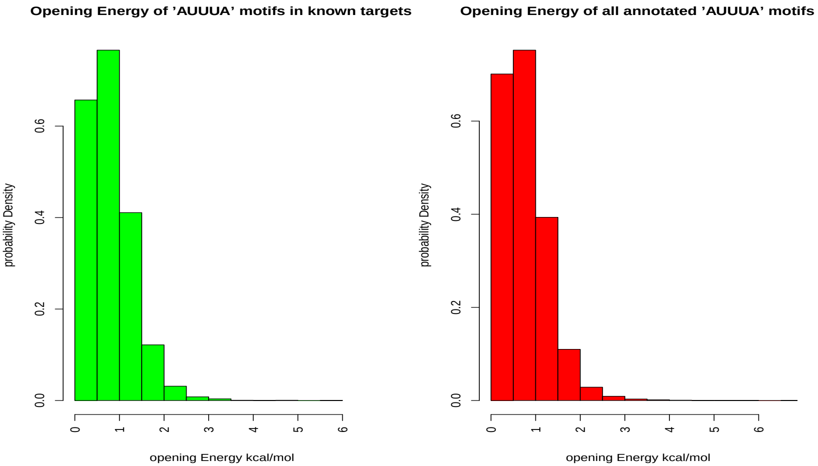
- Focus on AUUUA ⇒ ARE core motif
- Use accessibility as filter for regulatory ARE motifs
- Compare known targets to the rest

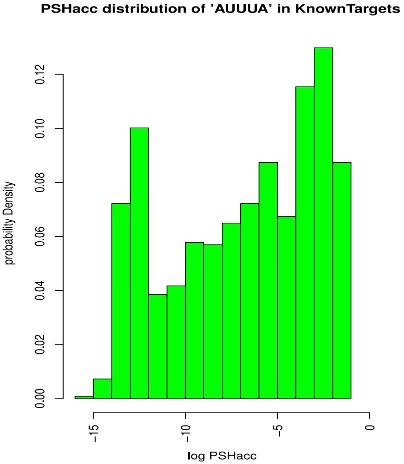
Accessibility

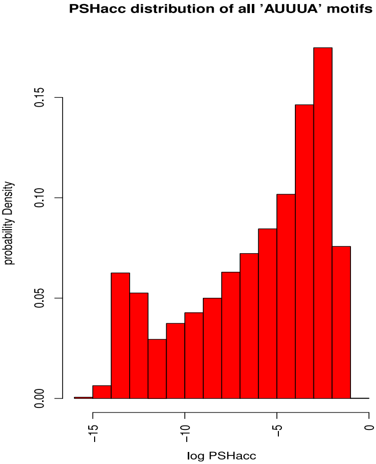


New Plan

- Marín and Vanícek ⇒ P-value
- Use over-representation as filter

P-value


PSHacc distribution of 'AUUUA' in KnownTargets


PSHacc distribution of all 'AUUUA' motifs

Conclusion

The ARE core motif 'AUUUA' as well as ARE flanking regions are over-represented in the known target transcripts

Conclusion 2

We need an other filter

PhD-thesis

- Still lot of statistics to do
- Motif- position/distance/flanking regions
- Overlap with other motifs?

- Structural context that embeds ARE motifs ⇒ RNAup
- Both 2D/3D if possible
- Decouple structure from sequence

- Analyze consensus sequences specific for (each) AUBP ⇒ HuR targets U-rich seq
- Get distance of zinc-fingers/RRM from X-ray
- Any relationship of findings with AUBP effect

Conclusion

- Accessibility of core-motif insufficient filter
- Over-representation better
- Combination with sequence based statistics?

- Check structures
- Single stranded vs. stem/loop
- Use information on protein motifs
  ⇒ Distance relevant?
- Cluster targets according to AUBP

- Correlate decay rates with findings
- Predict decay of 'housekeeping' genes
- Integrate conservation information

Thanks to

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Xtof for driving,organization
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