DASH extensions (Immunology)

Protein-RNA interactions

DASH

T cell – APC binding

BCR/TCR modeling

Antibody– Antigen binding
DASH Extension 1: protein-RNA interactions
Example: protein-RNA interactions from DASH?

Query → DASH hits bound to RNA → Query superimposed on hits → Overall distribution of RNA on query
Overall distribution of nucleotides on Regnase-1

Nucleotide-abundant

Nucleotide-scarce
Is this asymmetric distribution a coincidence?

High

Low

Not a coincidence!
RNA-binding Regnase-1-DASH hits are all ribosome components

<table>
<thead>
<tr>
<th>DASH Score</th>
<th>PDB ID</th>
<th>Protein Chains</th>
<th>Complex</th>
</tr>
</thead>
<tbody>
<tr>
<td>37</td>
<td>5OQL_P</td>
<td>C,R,T,Z,a,d,r,w,0</td>
<td>Pre-Ribosome</td>
</tr>
<tr>
<td>32</td>
<td>5WLC_SL</td>
<td>L9,LE,LU,NB,NC,NE,SA,SC, SF, SI</td>
<td>Processome</td>
</tr>
<tr>
<td>23</td>
<td>4A2I_V</td>
<td>L</td>
<td>Ribosome</td>
</tr>
<tr>
<td>20</td>
<td>2YKR_W</td>
<td>K,L</td>
<td>Ribosome</td>
</tr>
</tbody>
</table>
What are these components?

Cleaving rRNA (utp24)

5OQL_P: Pre-ribosome

5WLC_SL: Processome

4A2I_V: ribosome

2YKR_W: ribosome
UTP24: a Regnase-1 homolog in ribosomes

The PIN domain endonuclease Utp24 cleaves pre-ribosomal RNA at two coupled sites in yeast and humans

Graeme R. Wells, 1 Franziska Weichmann, 1 David Colvin, 1 Katherine E. Sloan, 1 Grzegorz Kudla, 2, 3 David Tollervey, 2 Nicholas J. Watkins, 1 and Claudia Schneider 1,*

Architecture of the 90S Pre-ribosome: A Structural View on the Birth of the Eukaryotic Ribosome

Markus Koreprobst 1, 2, Martin Turk 1, 2, Nikola Kellner 1, Jingdong Cheng 1, Dirk Flemming 1, Isabelle Koi-Braun 1, Martin Koi 1, Matthias Thoms 1, Otto Berninghausen 1, Roland Beckmann 2, 3, 4, 5, Ed Hurt 1, 2, 3, 4, 5

3.2-Å-resolution structure of the 90S preribosome before A1 pre-rRNA cleavage

Jingdong Cheng, Nikola Kellner, Otto Berninghausen, Ed Hurt & Roland Beckmann

The complete structure of the small-subunit processome

Jonas Barandun, Malik Chaker-Margot, Mirjam Hunziker, Kelly R Molloy, Brian T Chait & Sebastian Klinge
acidic residues (E98, D131, D150 and D152)\textsuperscript{43}. Importantly, we were also able to trace the RNA chain between the 5’ end of the pre-18S rRNA and the 3’ end of the 5’ ETS, visualizing the A1 cleavage site. The 3’ end of the 5’ ETS ‘walks’ along the surface of the 90S particle and points to the penultimate H9 of the 5’ ETS. Unexpectedly, we found the Utp24 catalytic site close to the box A heteroduplex, which is more than 35 Å away from the A1 cleavage site (U588) (Fig. 7a–c). Such a distance is a clear indication that the 90S preribosome is still not yet in the precessome modus regarding A1 cleavage (Fig. 7c,d). We suggest that after the 90S is in a release-competent state, a currently unknown splitting factor/helicase may be activated to unwind the box A/A’ helices and unlock Utp24 together with the rRNA, thus resulting in endonucleolytic cleavage at site A1 (Fig. 7e).
Functional similarity: Regnase-1 also helicase to unwind target RNAs

UPF1 unwinds in the 5’ to 3’ direction

- Reg1 bound to RNA but unable to cleave stem
- Reg1-upf1 interaction activates helicase, unwinding stem
- Reg1 cleaves unwound stem

Therefore, Reg1 expected to cleave on 3’ side of stem
MSAs encode structural information

Traditional AB modeling is Slow

Approach used by Rosetta, Schrodinger, etc. (thousands of hours!)

Adding a sequence to an existing MSA is fast (<1 sec)!

Repertoire Builder: MAFFT-DASH MSA + MAFFT v7
10,000 models in 30 min!
Repertoire Builder uses MSA-based approach

A. Prepare template MSAs

B. CDR template MSAs binned by length

C. Extend template MSA
Repertoire Builder workflow

D. Rank templates

Query-template alignments

- q-t₁
- q-t₂
- q-t₃

Feature vectors (vᵢ)

- q-t₁
- q-t₂
- q-t₃

Weight vector (w)

E. Assemble 3D model

Scores

- \( t₁ \)
- \( t₂ \)
- \( t₃ \)

\( Sᵢ = w \cdot vᵢ \)
Repertoire Builder uses MSA-based approach

How important are MSAs?
Good MSAs are crucial
Good MSAs are crucial

<table>
<thead>
<tr>
<th>CDR1</th>
<th>CDR2</th>
<th>CDR3</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="image" alt="CDR1" /></td>
<td><img src="image" alt="CDR2" /></td>
<td><img src="image" alt="CDR3" /></td>
</tr>
</tbody>
</table>

No misalignment
Repertoire Builder error was lower than other tested methods.
Extension to Antibody-Antigen docking

Build 3D models

Antibody

Sequences

3D models

Repertoire Builder

Forte, Spanner, Scwrl
Extension to Antibody-Antigen docking

Predict initial paratope and epitope

3D models
Antibody → Separate features → Initial predictions
Antigen → Paratope

Epitope
Extension to Antibody-Antigen docking

Dock antibody and antigen

Initial epitope prediction → Patches → Poses

Hex
Extension to Antibody-Antigen docking

Prepare improved features and predict final epitope
Significant improvement over Hex sampling
Significant improvement over Hex scoring
Quantitative improvement over ClusPro
## Quantitative improvement over ClusPro

Docking performance comparison between Adapt and ClusPro

<table>
<thead>
<tr>
<th></th>
<th>Total query</th>
<th>Successful query</th>
<th>Total models</th>
<th>Successful models</th>
</tr>
</thead>
<tbody>
<tr>
<td>Adapt</td>
<td>430</td>
<td>156 (36.28%)</td>
<td>11218</td>
<td>343 (3.06%)</td>
</tr>
<tr>
<td>ClusPro</td>
<td>430</td>
<td>166 (38.60%)</td>
<td>11218</td>
<td>227 (2.02%)</td>
</tr>
</tbody>
</table>

Distribution of three quality classes among successful models

<table>
<thead>
<tr>
<th></th>
<th>Successful models</th>
<th>Acceptable</th>
<th>Medium</th>
<th>High</th>
</tr>
</thead>
<tbody>
<tr>
<td>Adapt</td>
<td>343</td>
<td>306/343</td>
<td>35/343</td>
<td>2/343</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(89.21%)</td>
<td>(10.20%)</td>
<td>(0.58%)</td>
</tr>
<tr>
<td>ClusPro</td>
<td>227</td>
<td>186/227</td>
<td>39/227</td>
<td>2/227</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(81.94%)</td>
<td>(17.18%)</td>
<td>(0.88%)</td>
</tr>
</tbody>
</table>
Adapt poses show low overlap with ClusPro
Antibody-specific epitope prediction

Initial AUC .76
Final AUC .80
Extension to TCR-epitope-MHC modeling

https://sysimm.org/immune-scape/

ImmuneScape is the first automated TCR-epitope-MHC modeling server of its kind

Li et al. . Meth Mol Biol (2019)
Extension to T cell-APC cell modeling

Antigen presenting cell (APC)

Peptide-MHC

TCR

T cell
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