RNA 2D to 3D Modeling with RNA2D3D rev. 5.6 and newer

- System Installation
- Basic Organization of the System
- 2D to 3D Modeling Example
- Nano Design Example

Last revision: 6/14/2011

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RNA2D3D rev. 5.6+ Installation & Getting Started

First Steps:

1) The RNA2D3D package is distributed as a compressed TAR file.
2) After uncompressing (gunzip filename.tar.gz), open the tar file: tar –xvf filename.tar
3) A subdirectory named rna_2d3d_vLT5.x will be created. (In the file name L – indicates Linux, I – Irix/SGI binaries. 5.x indicates system revision number).
4) Change directory to the rna_2d3d_vLT5.x and run the set-up/installation script CONFIG-RNA2D3D. The file cshrc_2d3d will be created that contain definitions for a self contained environment.
5) Execute: source ./cshrc_2d3d. This is for the csh shell. (For other shells you will have to define equivalent commands. Note: we have not extensively tested this system under other shells.)
6) Start the system by its alias name: rna_2d3d (or any alias you chose to give it in the cshrc file.

Important Notes: Please, read the RNA2D3D-README.txt file for information on the libraries needed to run RNA2D3D.

RNA2D3D has been compiled with the following **libraries**. Most of the **OpenGL libraries are shared**, and you may need to install them on your Linux environment: Makefile's LDFLAGS = -L/usr/X11R6/lib –l**GLw** –l**GL** –l**GLU** –l**Xext** –l**X11** –l**Xm** –l**Xt** –lm –lmmalloc

Refinements of the structure elements and the whole structure are performed by the **Tinker** package binary modules (minimize, optimize, pdbxyz, and xyzpdb), which were downloaded to the subdirectory **Tinker-sta.dir** by "web-get" calls: wget ftp://dasher.wustl.edu/pub/tinker-bin/linux/minimize.gz, etc., with kind permission of prof. Jay Ponder. In case you experience problems with any of these modules, you may have to download new binaries or the source files and recompile them on your machine.

Prepared by: W. Kasprzak, BSP, SAIC-Frederick, Inc.

RNA2D3D rev. 5.6+ System Organization

System Organization Notes:

The sample input files are provided in the **sample.dir** subdirectory, which is subdivided into subdirectories listed below, based on the file types. Identical organization is assumed for the **RNA_2D3D** subdirectory, where the user files are to be stored (and will be searched for by the system).

BPLfiles directory contains rna_2d3d-specific base pair lists where sequence and all the base pairs, including pseudoknots, are listed. Negative values given to base pairs denote tertiary interactions, that will be marked with linking lines but no attempt will be made to impose realistic geometry on them. In case of pseudoknots, for which geometry cannot be predicted automatically, the user may apply the tertiary (negative) designations and attempt to manually model them (non-trivial, but achievable).

PDBfiles directory contains PDB format (AMBER, STANDARD, TINKER, MSI) files. These can be the rna_2d3d output files based on the predicted structures, or imported from outside databases. If you read in a PDB file, the 3D view of it shows only as the backbone geometry, and the 2D view as the whole sequence in a circle. Pairing data is not parsed out of the PDB input file!

3DModels directory contains binary 2D and 3D information files. These are “state dumps” that can be used for saving the state of your project and restoring it for further work. They are also necessary as the nano-structure building blocks.

TPLfiles directory contains pairing lists (plain text files), which specify interactions between building blocks assumed to be available in the 3DModels directory, explicitly called by their names.

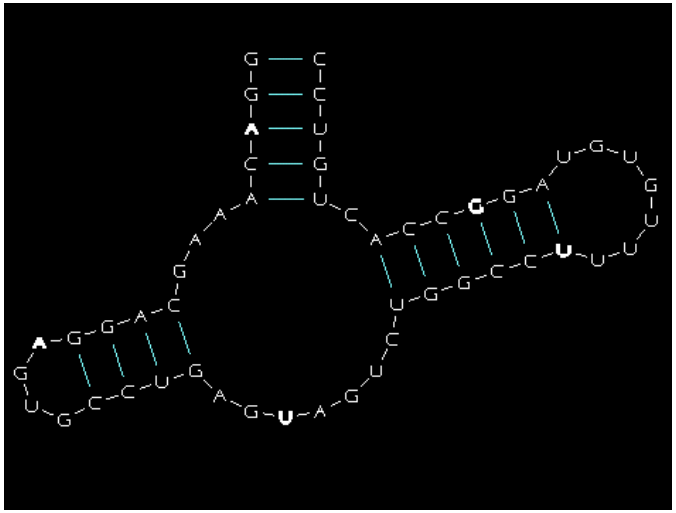
From RNA Secondary Structure to 3D Structure with RNA2D3D

1. Start with RNA secondary and possible tertiary interaction data.
2. Apply RNA2D3D to this data to produce a rough approximation of the 3D model.
3. Check for steric clashes and adjust if necessary.
4. Apply molecular mechanics minimization to spot any missed steric clashes and to improve bonded and non-bonded interactions.
5. Check for conservation of given secondary and tertiary structure interaction and adjust if necessary.
6. For complex structures – check PDB database for similar structures. If necessary adjust model manually and/or splice in a database motif.
7. If additional tertiary structure interactions found, manually adjust and go to 4 else go to 8.
8. Apply molecular dynamics. If satisfied STOP, else go to 7.

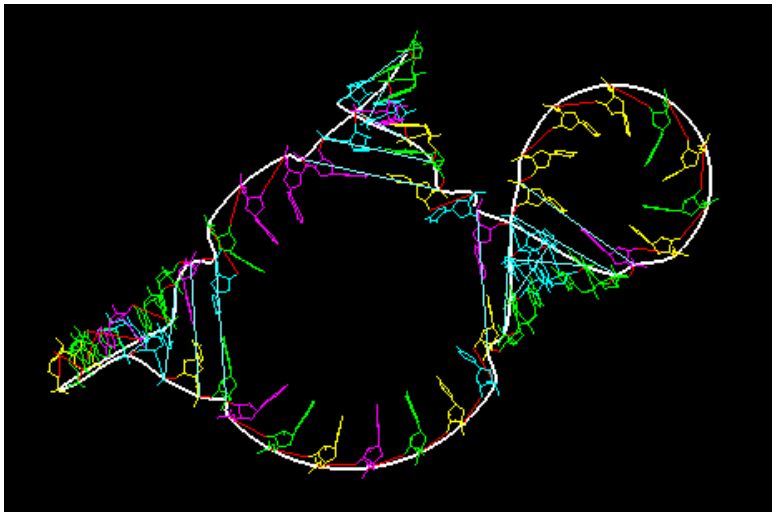
2D & 3D Modeling Example 1: Hammerhead Rybozyme

RNA2D3D Modeling

Hammerhead Ribozyme example; Illustration of some of the Tools

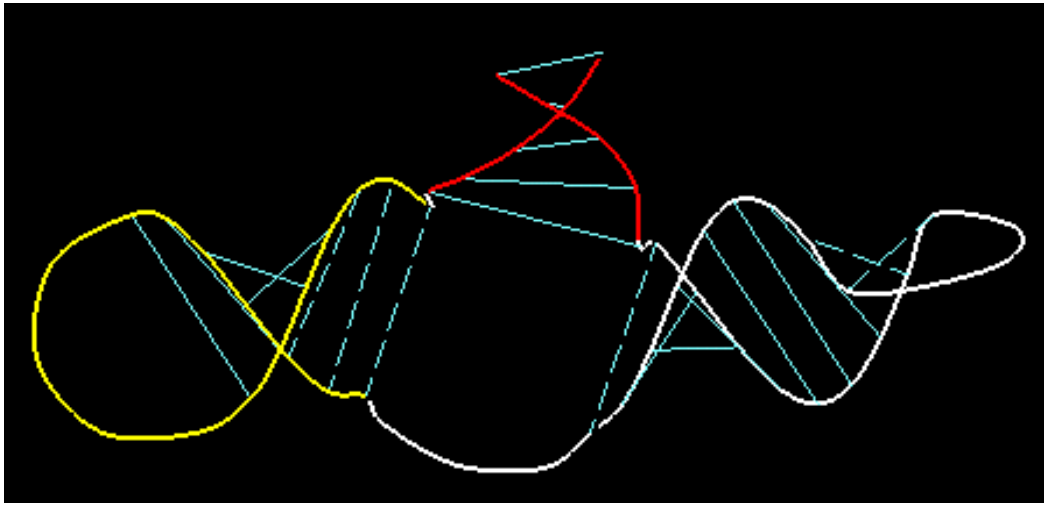


2D scaled secondary structure



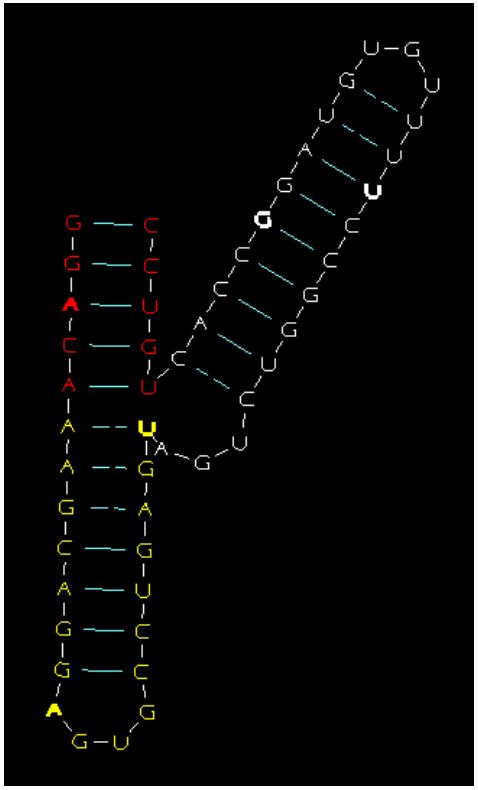
Planar 3D embedding and stems transformed into A-form Helices

Stem Compactification



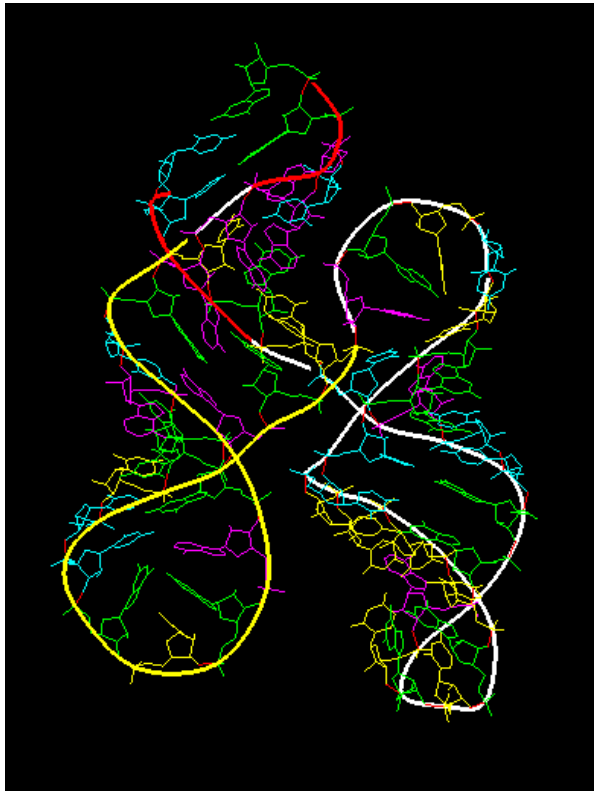
Martinez H.M., Maizel J.V. Jr., & Shapiro B.A. *JBSD*, 25, 2008

Further Improvement of the Model by Stacking, Segment Positioning and Energy Refinement



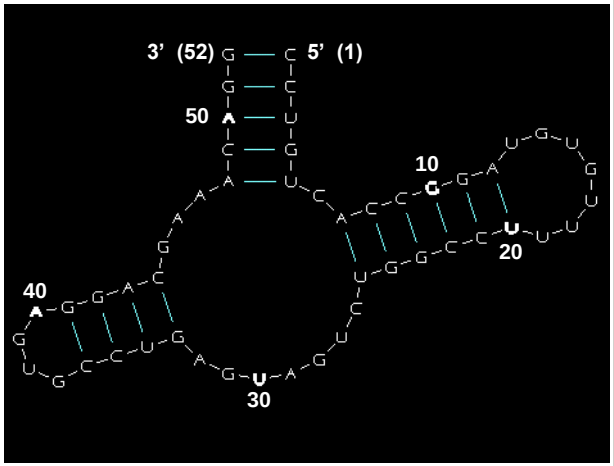
Stem Stacking
(Red and Yellow)

Corresponding 3D Model

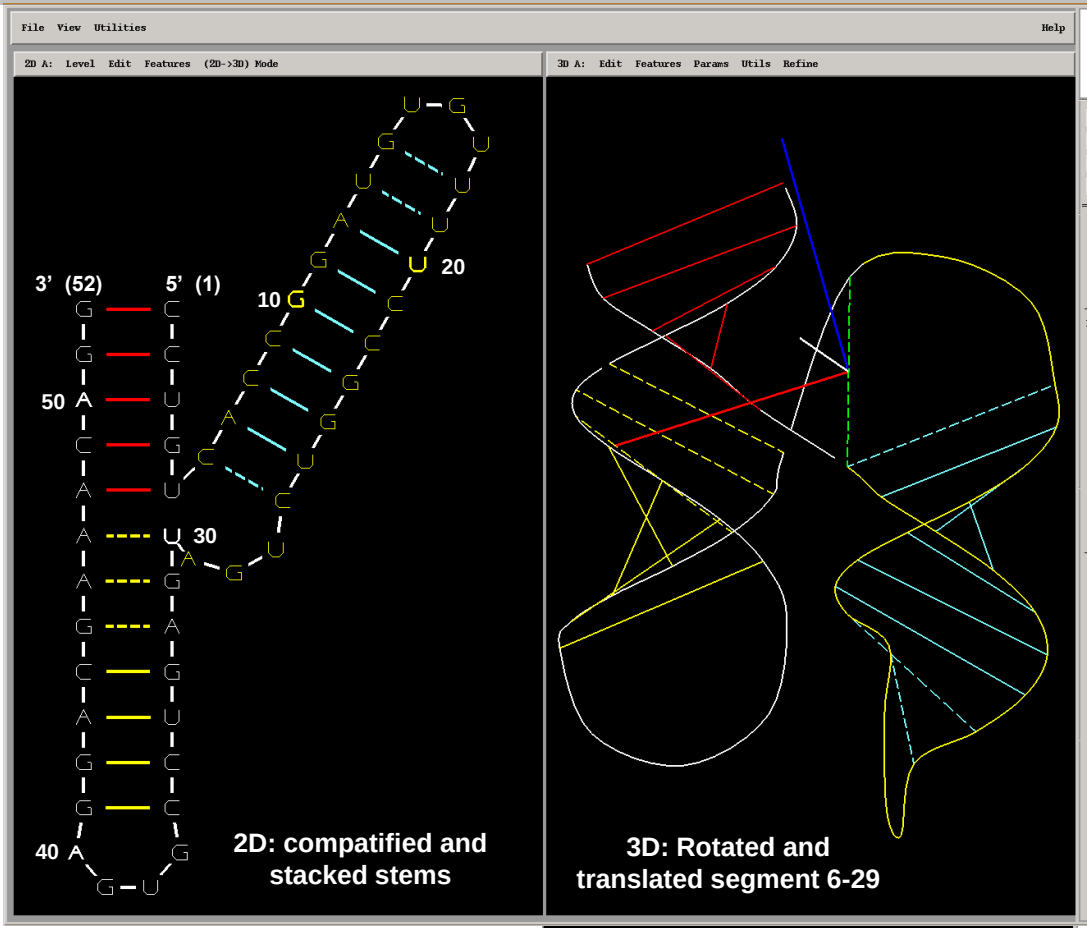


Segment Positioning and Energy Refinement
(Tinker / Amber)

RNA2D3D Modeling: Hammerhead Ribozyme (1)



2D: initial pairing info



Note: BPL files created with different editors may cause problems due to carriage-return & newline combination appended at the end of lines (in MS Word Pad, or othe non-Unix editors). The quickest way to clean them up for use with RNA2D3D is this shell command:

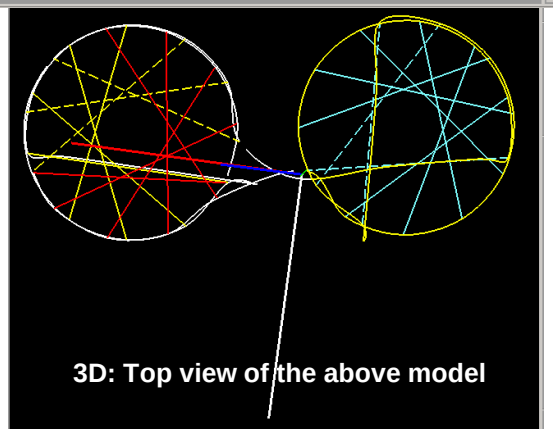
shell-prompt> sed 's|\r||' in-file.bpl > out-file.bpl

Read-in .bpl file: File -> sample BPL -> hammerhead -> Model A
Compactify stems: 2D:Edit -> Compacting -> This Frame
Color stems: 2D:Features -> Stem Coloring (pick stems and select colors)
Stack Stems: 2D:Edit -> Stem Stacking (pick two stems and press Accept)

Manipulate 3D model: 3D:Edit -> Segment (pick positions 6 & 29 in 2D picture)
Segment Position (style=closed; preRef=on):

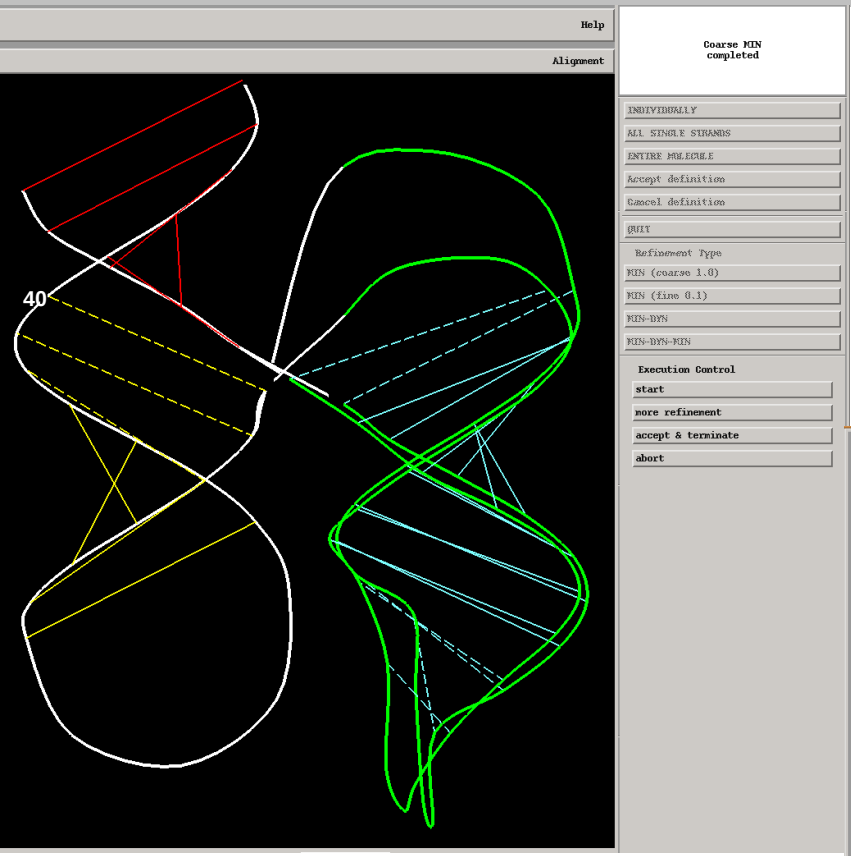
Segment	Axis	Rotation (x,y,z)	Translation (x,y,z)
6-29	X,Y,Z:MP	R=(200,50,75)	T=(-18, -1, 0) - results shown on the right

Note: Specific rotation & translation values will vary with the order and choice of actions



3D: Top view of the above model

RNA2D3D Modeling: Hammerhead Ribozyme (2)

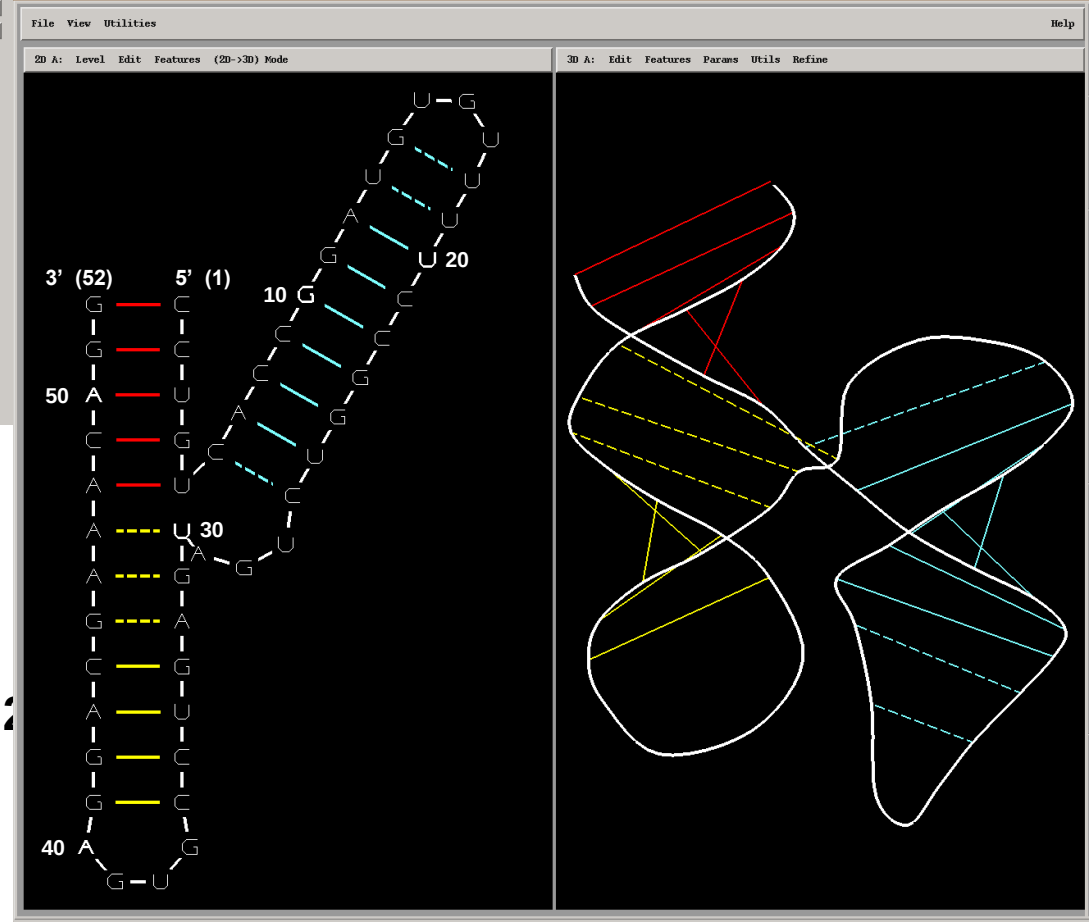


3D:Refine (GC) the 6-29 segment - minimization (coarse 1.0) (left figure)

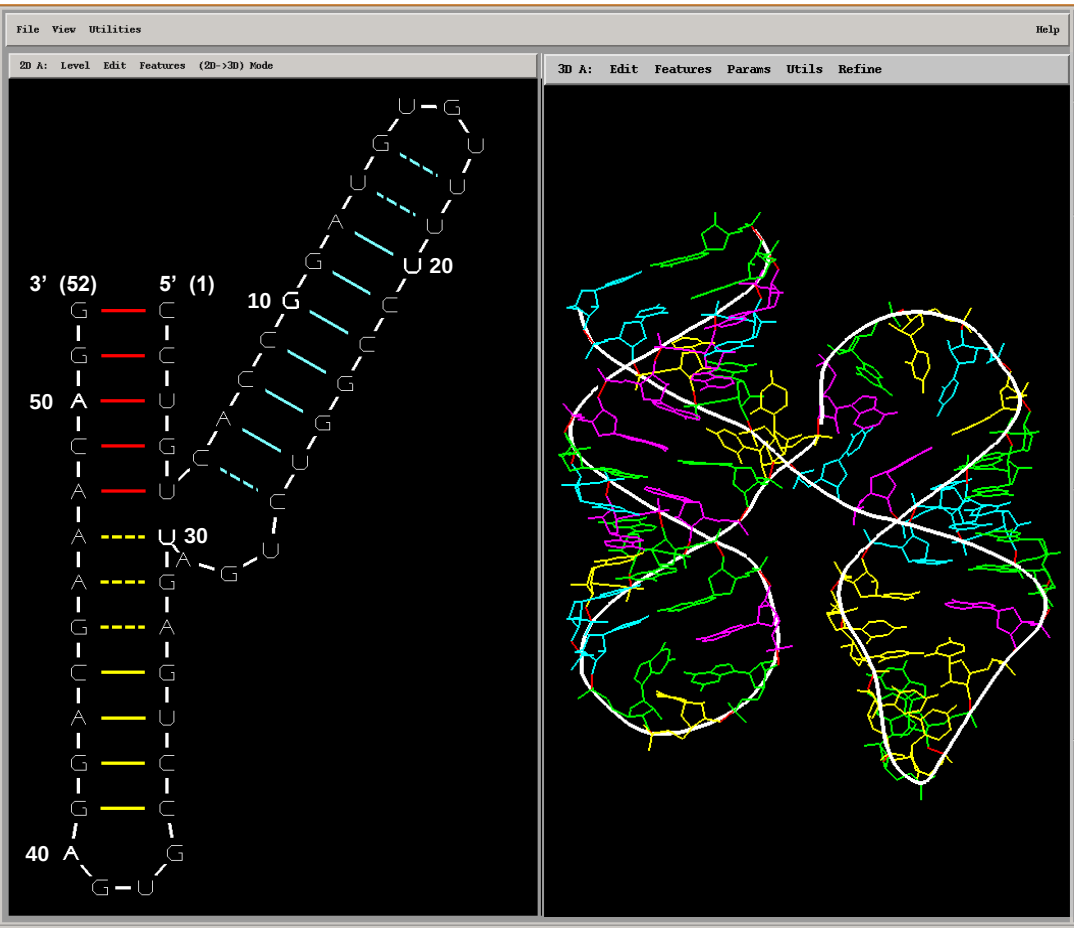
Make sure that the 3WJ single strand (27-29) ends up clearly in front of the other backbone transitioning from red to cyan stem. If it doesn't, go back to previous step and reposition the branches.

3D:Refine (LC) the H-loops and single strands - minimization (coarse 1.0)

3D:Refine (GC) the entire model - minimization (coarse 1.0) (right figure)



RNA2D3D Modeling: Hammerhead Ribozyme (Alt)



Alternative approach selecting the cyan region and the flanking nucleotides from the red and yellow stems.

Manipulate 3D model: 3D:Edit -> Segment (pick positions 5 & 30 in 2D picture)

Note: Specific rotation & translation values will vary with the order and choice of actions

3D:Refine (GC) the 5-30 segment - minimization (coarse 1.0)

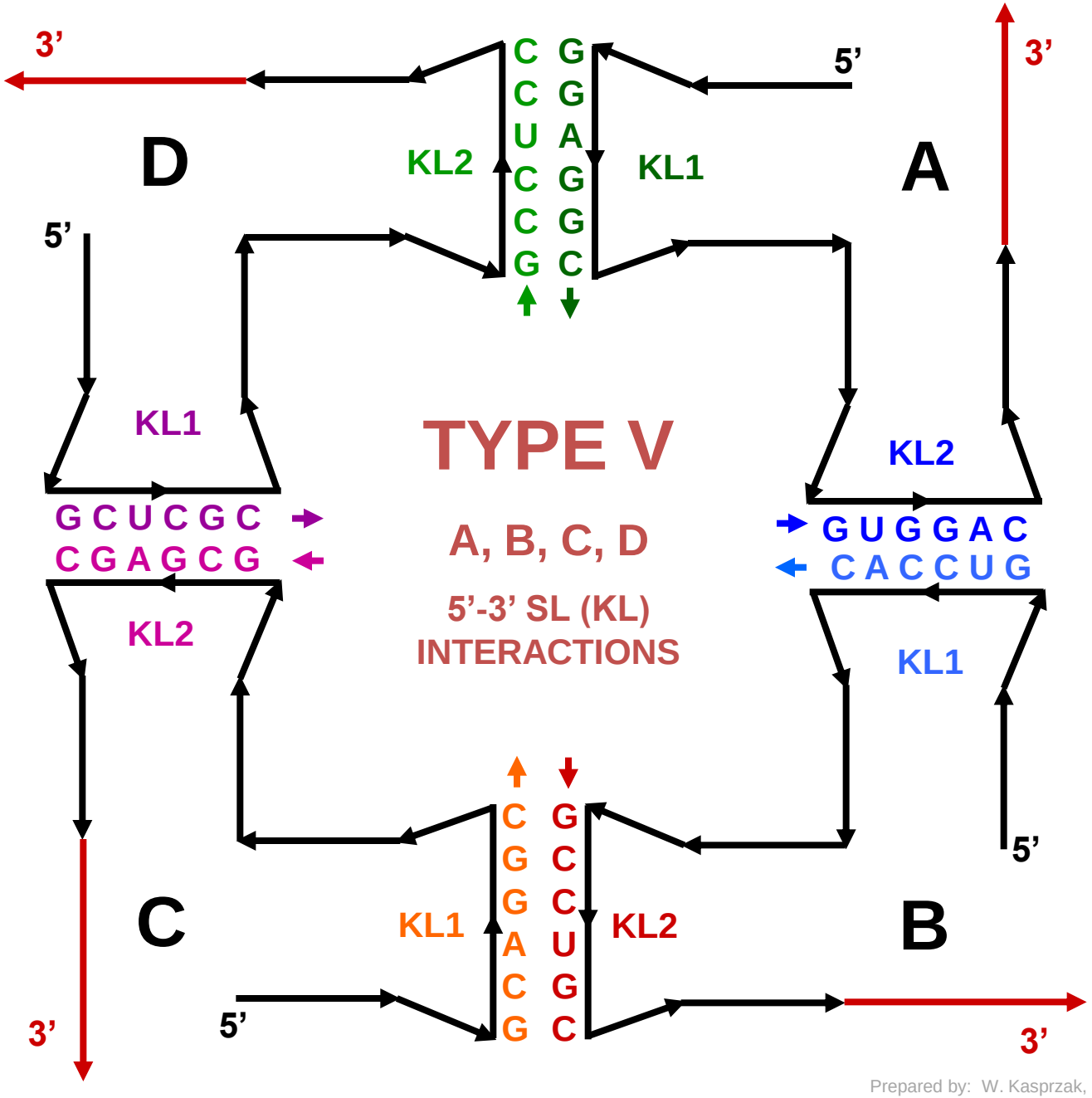
3D:Refine (LC) the H-loops and single strands - minimization (coarse 1.0)

3D:Refine (GC) the entire model - minimization (coarse 1.0)

To see the 3D structure with all nucleotides enable "Nucleotides" in 3D:Params menu and select to "Show All" in the 3D:Utils menu.

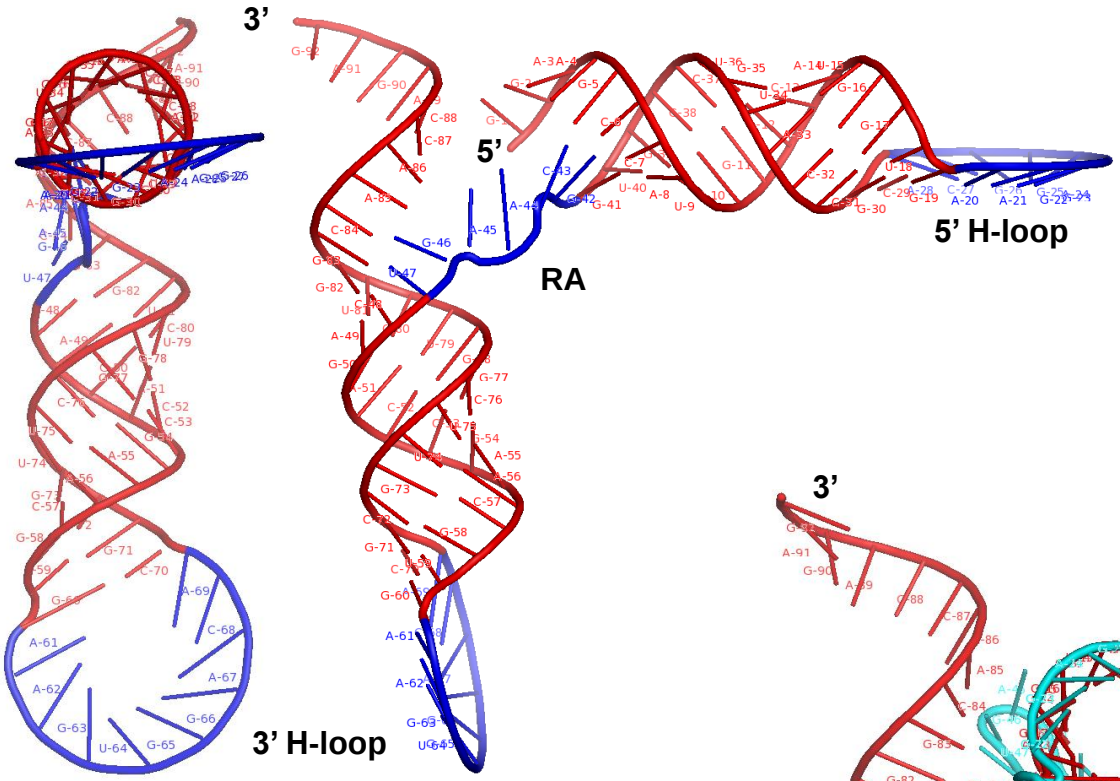
2D & 3D Modeling Example 2: A Tectosquare

Tectosquare of Type V



Prepared by: W. Kasprzak, BSP, SAIC-Frederick, Inc.

RNA2D3D: An Idealized Tectosquare Monomer

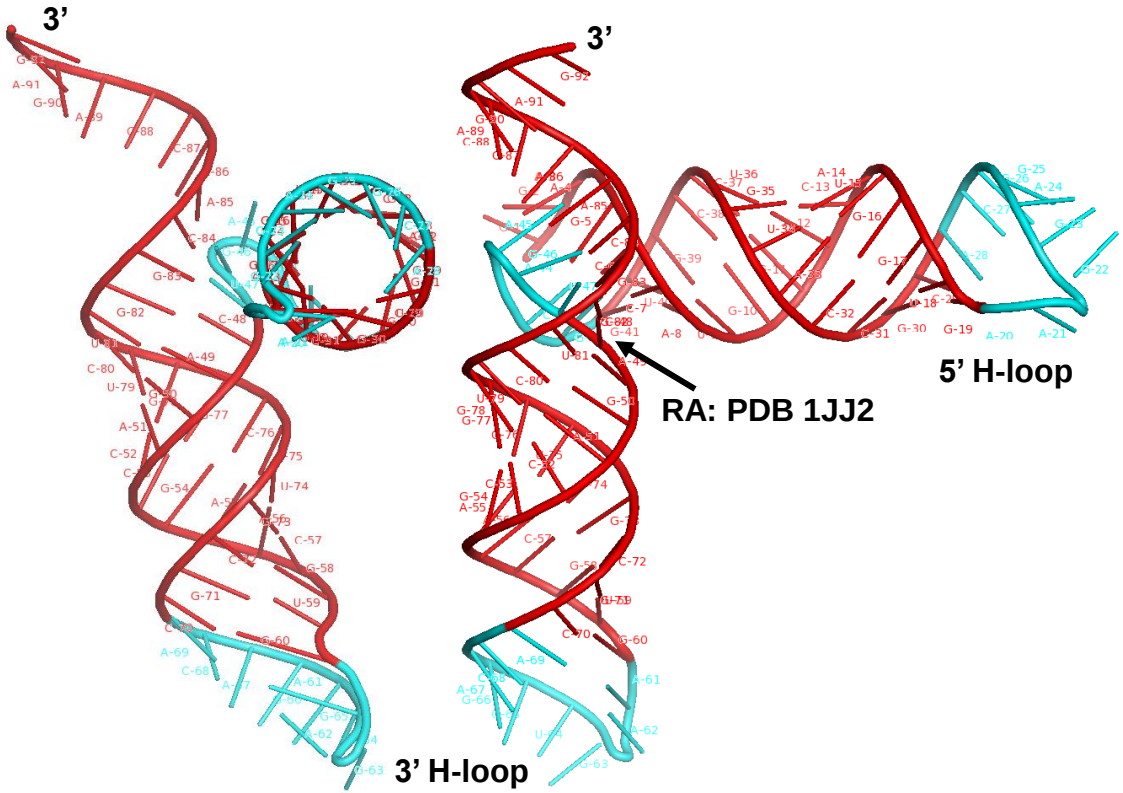


ABOVE: Raw 2D to 3D conversion

- Flat H-loops
- Best-guess RA linker
- Co-planar helix arrangement

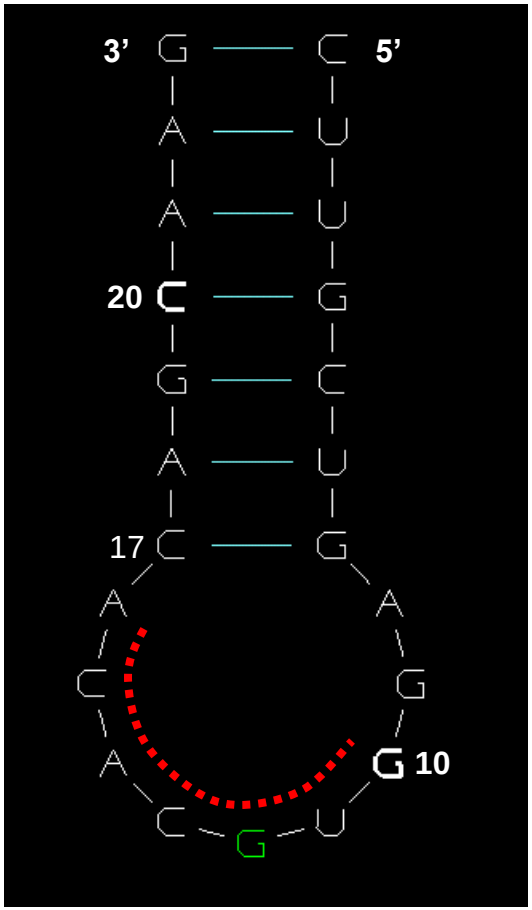
BELOW: Improved 3D structure

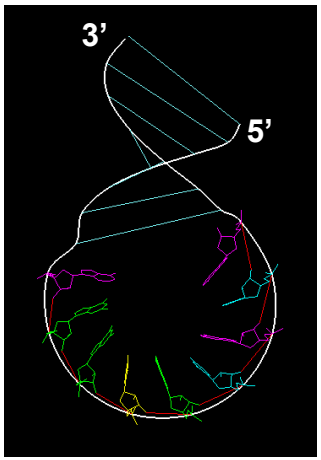
- 3’-end helix extension H-loops
- RA linker from PDB 1JJ2 (50S ribosomal unit of *Haloarcula marismortui*)
- Log cabin helix arrangement

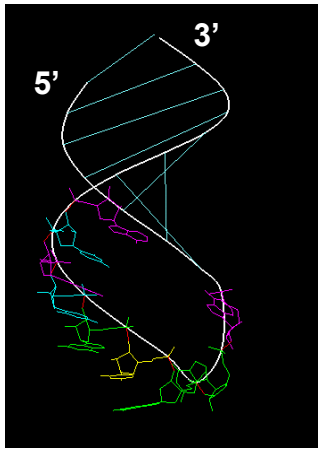


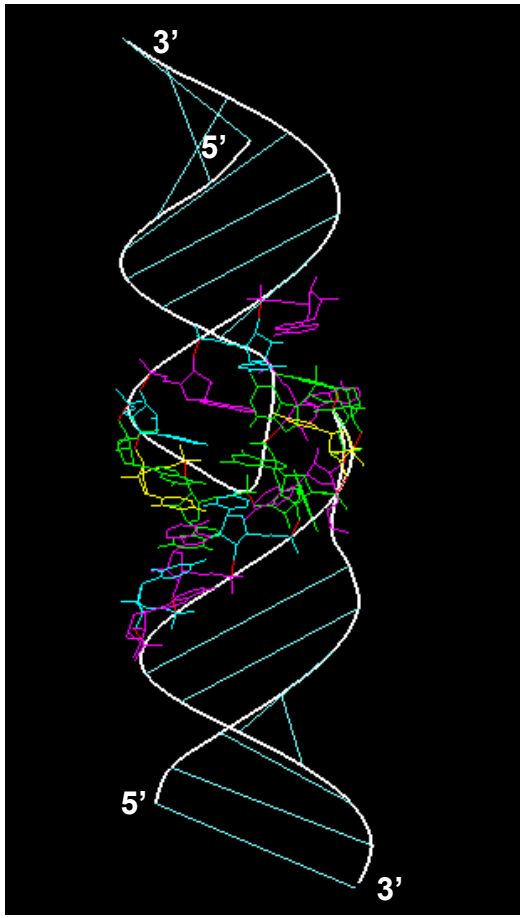
Prepared by: W. Kasprzak, BSP, SAIC-Frederick, Inc.

RNA2D3D: Modeling a Kissing Loop Interaction









From: Martinez *et al.*, JBSD 2008

A. Secondary structure depiction of a stem-loop motif.

B. Initial 3D model with A-form helix and flat H-loop

C. Improved 3D model. H-loop sequence indicated with red dashed line in A is given the A-form helix shape with the help of helix-extending tool: 3D:Edit -> Extend Single Strand Helicity (17..10)

D. The kissing loop complex. Two copies of the stem-loop model connected with the base-alignment tool: View -> 3D models A & B -> Align -> base-pair

Prepared by: W. Kasprzak, BSP, SAIC-Frederick, Inc.

RNA2D3D rev. 5.6+ Procedure to Build a Tectosquare (1)

The Big Picture:

1) Construct monomers and save them as binary 3DModels (and/or ASCII PDB files).
2) Create a Topology file (see TPLfiles) to define interactions between monomers.
3) Manipulate stem length and 3'tail length via "2D Monomer 3D Tectosquare" tools to see global effects (change in 1 monomer is applied symmetrically to all other building blocks/monomers).

Constructing Tectosquare Monomers (4 buidling blocks):

1) Load .bpl file: File -> User bpl/Sample bpl -> slect a file (e.g. LT17-A3.bpl) -> model A
2) Modify hairpin loops by extending the 3' helicity of the supporting stem: Edit -> Extend Single Strand Helicity. For a kissing loop 5'-AAGCGCGCAC-3' click on the 3' nucleotide of the closing basepair (C) and the 5' most G of the self-complementary sequence. Results are immediately visible in the 3D window (View -> 2D A & 3D A mode)
3) Copy Model A to Model B (3D:Utilities)
4) Replace the stem linker single strand (AA) with a PDB right angle motif RA (experimental 3D coordinates) by defining a subset in the full model and its equivalent in the PDB junction structure and replacing the model subset with the PDB subset. (It is a general procedure which allows to replace any subset of the modeled structure with an equivalent fragment from a database).

Prepared by: W. Kasprzak, BSP, SAIC-Frederick, Inc.

RNA2D3D rev. 5.6+ Procedure to Build a Tectosquare (2)

Constructing Monomers (cont'd)

5) Read-in PDB entry into model A: Sample/User PDB -> STANDARD/MSI (BIOSYM)/AMBER format -> select a file (ex: A_TURN.pdb) -> select into model A

6) 2D A: Level -> Subset (pops up subset level control buttons on the right)

   a) Define a subset by picking (button) and click on G8 (5') and U13 (3') bases to highlight the subset (highlighted in green)

   b) Accept definition

   c) Select a subset (after the button click only the subset is displayed in 3D)

   d) Accept Selection

7) Repeat step 6) for model B (where a nanostructure monomer read-in from the BPLfile directory is stored): define by picking (button) and click on G42 (5') and U47 (3') bases to highlight a subset (selection results are highlighted in green).

8) Do the subset substitution from **Model A**: 3D:Utils -> 3DM utils -> Replace a B subset with an A subset. Click on Do substitution and Quit

9) Switch between models A and B via View submenu

10) To view one full tectsquare monomer: View -> 2D B & 3D B; Level -> Molecule

RNA2D3D rev. 5.6+ Procedure to Build a Tectosquare (3)

Constructing Monomers (cont'd):

11) Save the monomer as a 3DM file: Utils -> 3DM utils -> Save as 3DM file -> enter LT17.A3s

    Note that LT17 (Large Tectosquare 17) should be used as a convention (a database name for building tectosquares from multiple monomers, all part of LT17). This file is stored in the user's directory (rna_2d3d_root)./RNA_2D3D/3DModels

Note: The above outlined procedure can also be used to combine PDB-based geometry with the BPL-based preliminary 3D structure. An entire "PDB structure" can be designated as subset A (test carefully if the 2D window shows the whole sequence 1..n or, more likely for the PDB input files 1..n-1), and substituted for the "BPL subset" of identical size (1..n vs. 1..n-1 !).

RNA2D3D rev. 5.6+ Procedure to Build Tectosquares (4)

Constructing Tectosquares:

1) Once you have created four monomers (stored in the 3DModels directory) to be used as building blocks, create/edit the topology file for a nanostructure to be built out of these blocks. This file is in the directory sample.dir (or RNA_2D3D)/TPLfiles, and it specifies, for example, that the first building block (name listed) interacts with the second, starting at the base positions listed: 1:68-2:22. Interactions are automatically extended to include all contiguous complementary bases. The connections between the last (4th) and the first monomers are left open.
Refer to the sample file LT17 (./sample.dir/TPLfiles/LT17):

File -> Tecto Pairing Lists -> User -> file name -> Display / Apply

```
MONO  1    LT17.A3s    1   1:68-2:22        0:0-0:0
MONO  2    LT17.B1s    1   2:68-3:22        0:0-0:0
MONO  3    LT17.C8s    1   3:68-4:22        0:0-0:0
MONO  4    LT17.D6s    1  *4:68-1:22        0:0-0:0
```

- * denotes connectivity left open

2) Superstructures can be designed in this same way – see the sample file LT17-LT20, in which four tectosqares (LT17 through LT20) are given connectivity information, and individual tectosquares connect with each other via 3' tails (single-stranded areas) forming helices in the final model.

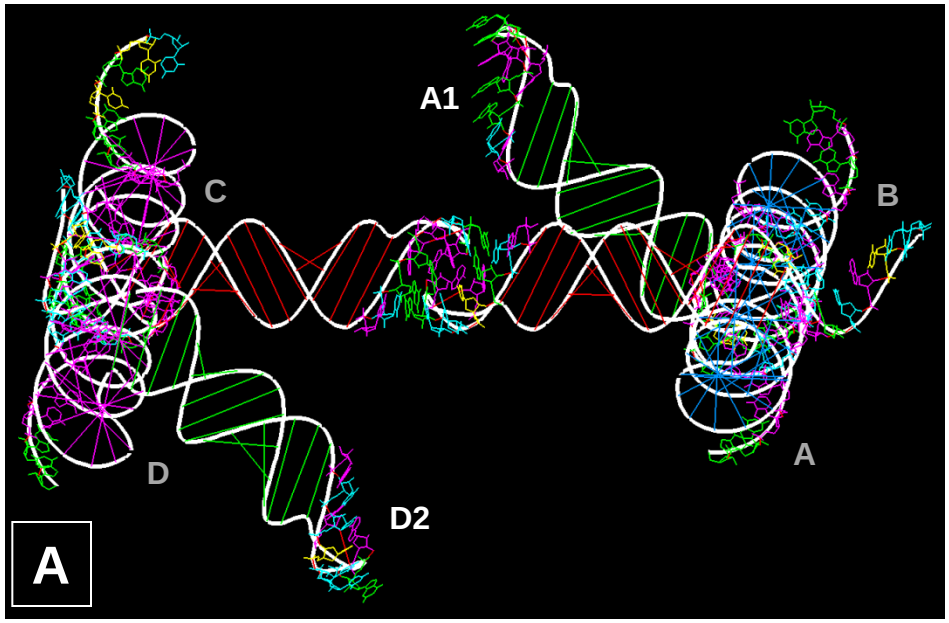
```
MONO  1    LT17.A3s    1   1:68-2:22        1:87-7:92
MONO  2    LT17.B1s    1   2:68-3:22        0:0-0:0
MONO  3    LT17.C8s    1   3:68-4:22        0:0-0:0
MONO  4    LT17.D6s    1  *4:68-1:22        0:0-0:0
MONO  5    LT18.A8ps   2   5:68-6:22        0:0-0:0
MONO  6    LT18.B5ps   2   6:68-7:22        6:92-12:87
MONO  7    LT18.C3ps   2   7:68-8:22        0:0-0:0
MONO  8    LT18.D7ps   2  *8:68-5:22        0:0-0:0
     ...
```

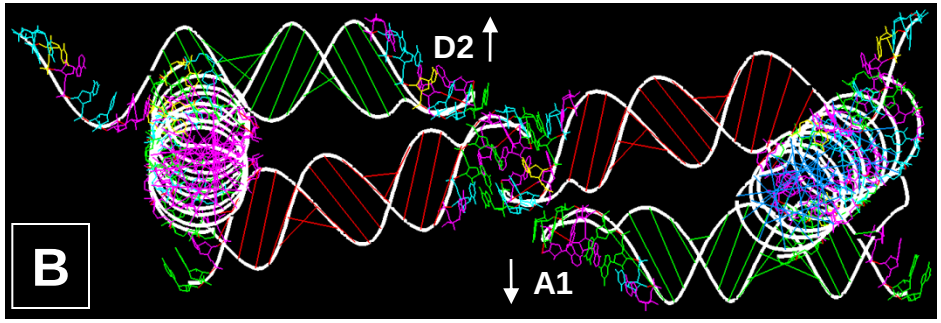
- 3' tail connectivity

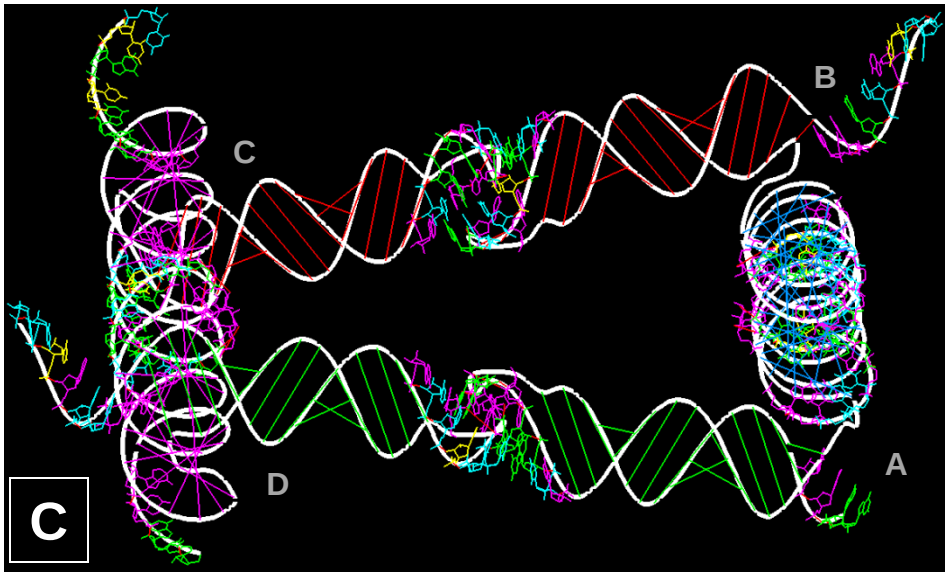
- 3' tail connectivity

Note: the above examples are based on Chworos *et al.*,Science 306, 2004.

RNA2D3D: Type V Tectosquare Closure (1)



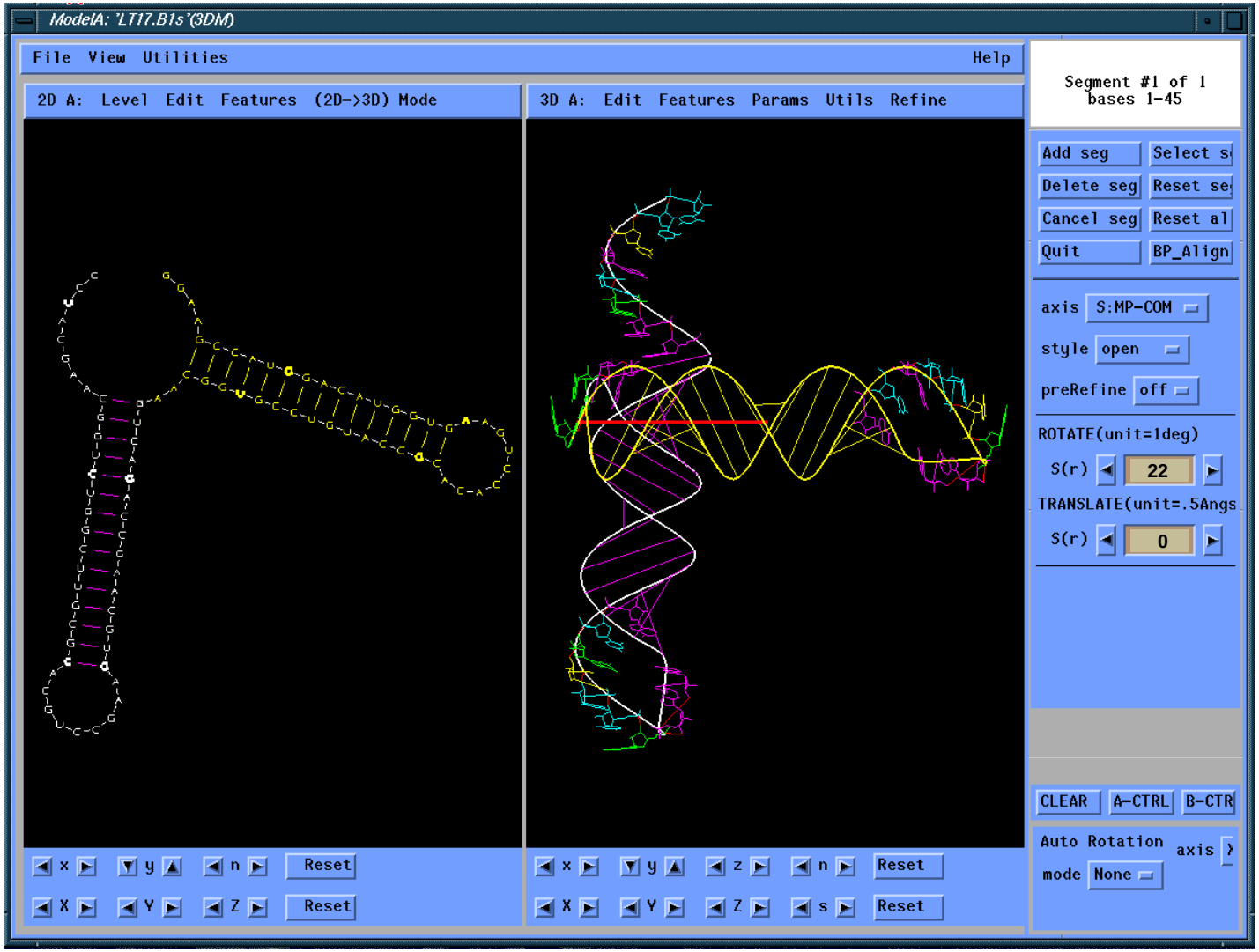




- Tectosqare model: four L-shaped monomers with the improved H-loop shapes and PDB-based RA motifs (1JJ2 - 50S ribosomal unit of *Haloarcula marismortui*). Connectivity (base-sharing) specified in a topology script file.
- **A:** The initial model does not close the tectosquare.
- **B:** Adding 1 bp to every 5' ideal A-form helix of every L-shaped monomer induces rotation of every tectosquare side and causes the A1 and D2 H-loops to move past each other. Thus we know we need an effective rotation of less than 33°.
- **C:** Applied coaxial rotation of each side (via the 5' arm) by **22°** brings the A1 and D2 H-loops into coaxial orientation .

Prepared by: W. Kasprzak, BSP, SAIC-Frederick, Inc.

RNA2D3D: Type V Tectosquare Closure (2)



Note: Older version of RNA2D3D interface is shown here.

3D:Edit -> Segment Position: Add rotation around the stem axis (select structure segment **1-45**, use **S:MP-COM** axis, style **closed**, preRefine **on,** and the rotation of **22 .**

(Note: Rotate to verify that the displayed red axis is indeed in line with the axis of the stem!)

RNA2D3D: Type V Tectosquare Closure (3)

Constructing Tectosquares:

1) Once all four monomers (stored in the 3DModels directory) have been modified as shown before, create a new topology file for a nanostructure to be built out of these blocks. This file is in the directory RNA_2D3D)/TPLfiles, and it specifies the same interactions as for the generic LT17 tectosquareare.

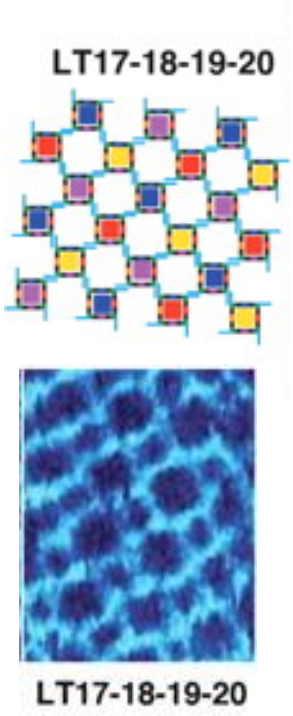
   Refer to the sample file LT17 (./RNA_2D3D/TPLfiles/LT17-SL1p22R1)

   File -> Tecto Pairing Lists -> User -> file name

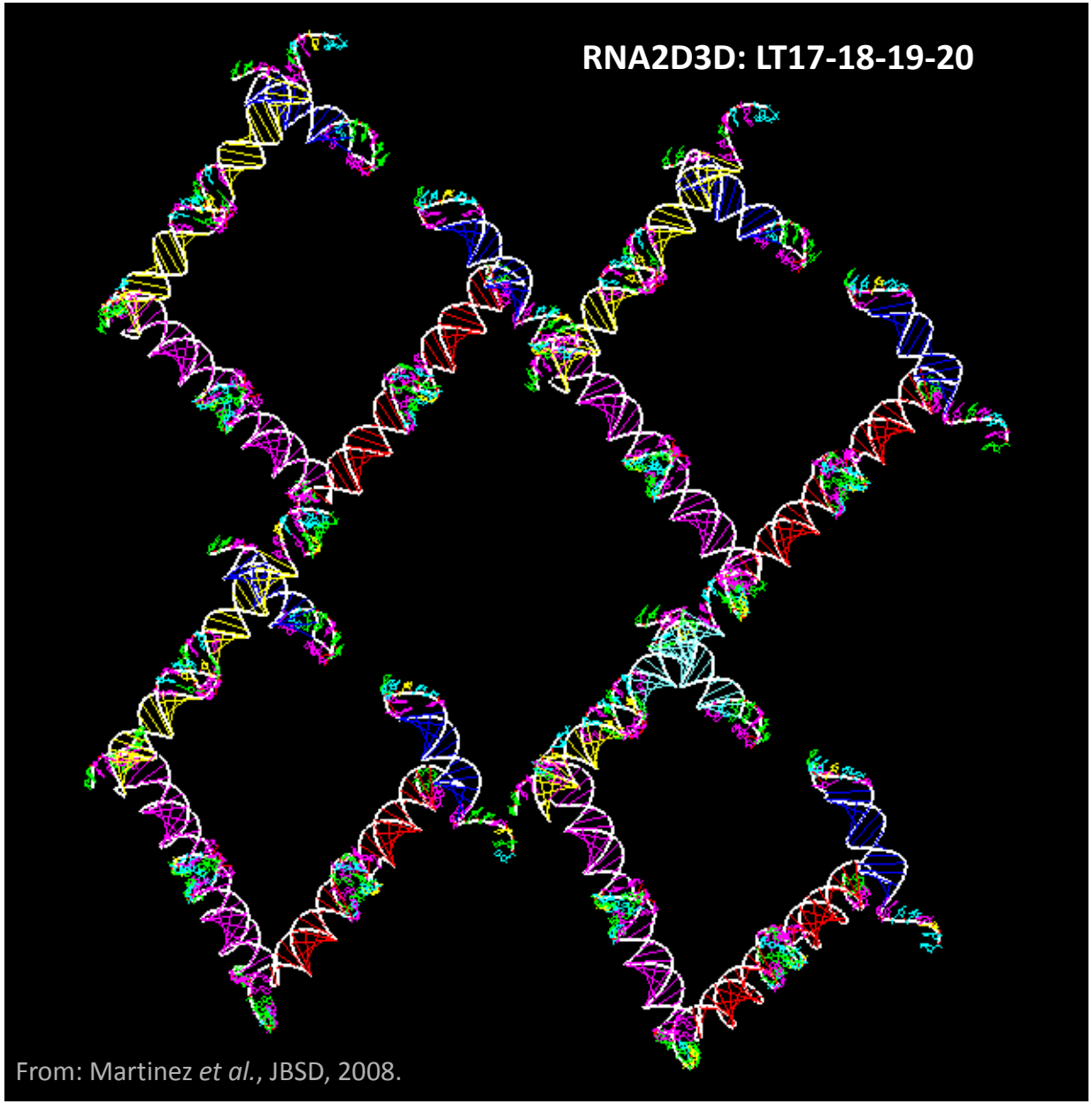
```
MONO  1    LT17.A3s.SL1p22R1    1   1:68-2:22        0:0-0:0
MONO  2    LT17.B1s.SL1p22R1    1   2:68-3:22        0:0-0:0
MONO  3    LT17.C8s.SL1p22R1    1   3:68-4:22        0:0-0:0
MONO  4    LT17.D6s.SL1p22R1    1  *4:68-1:22        0:0-0:0
```

- * open connectivity

RNA2D3D: Type V Tectosquare Mesh



From: Chworos *et al.*, Science 2004.

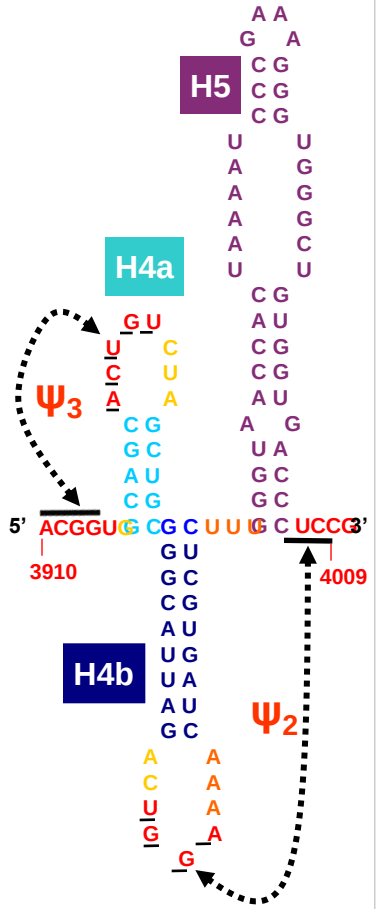


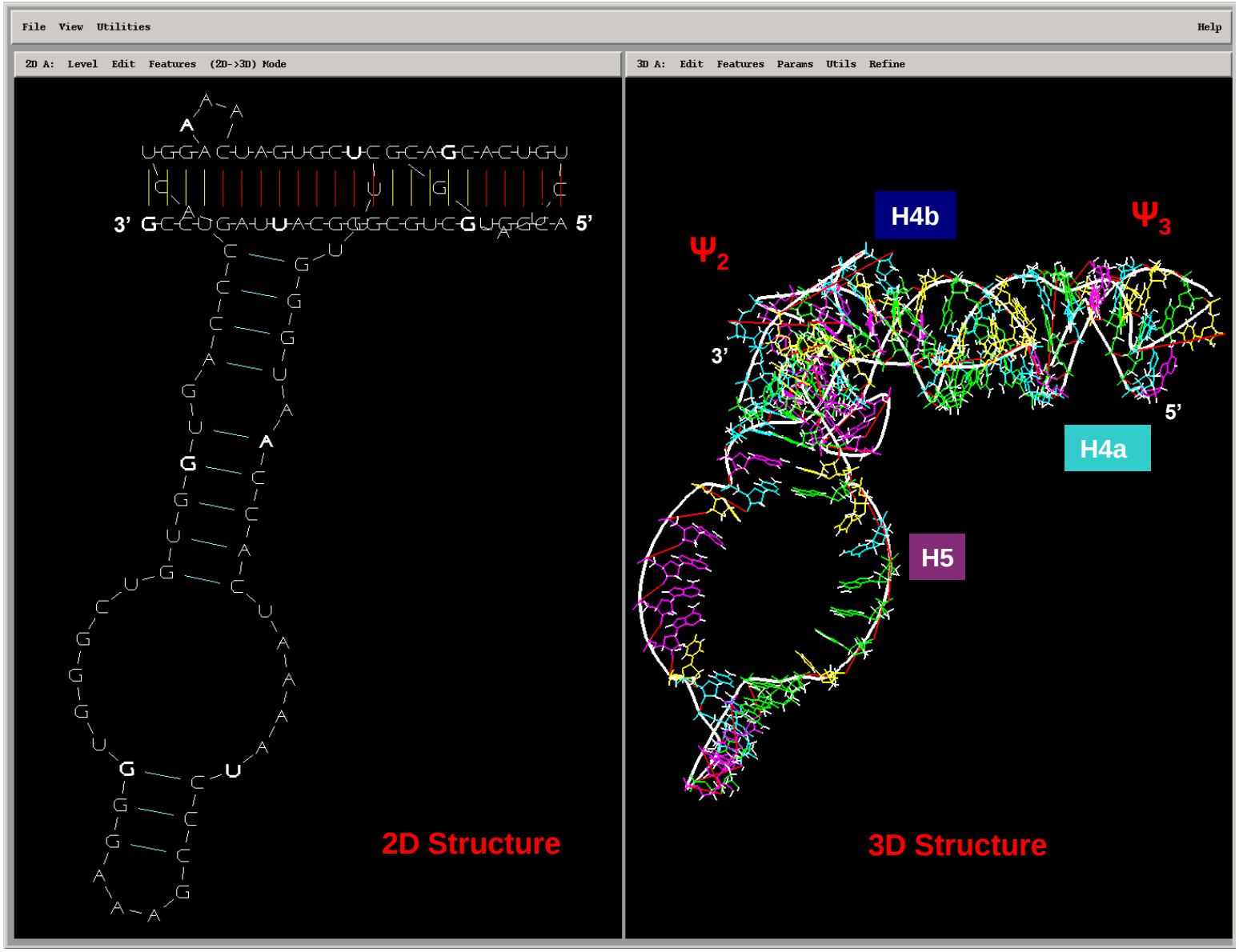
From: Martinez *et al.*, JBSD, 2008.

2D & 3D Modeling Examples: TCV 3’ UTR Translation Enhancer

Prepared by: W. Kasprzak, BSP, SAIC-Frederick, Inc.

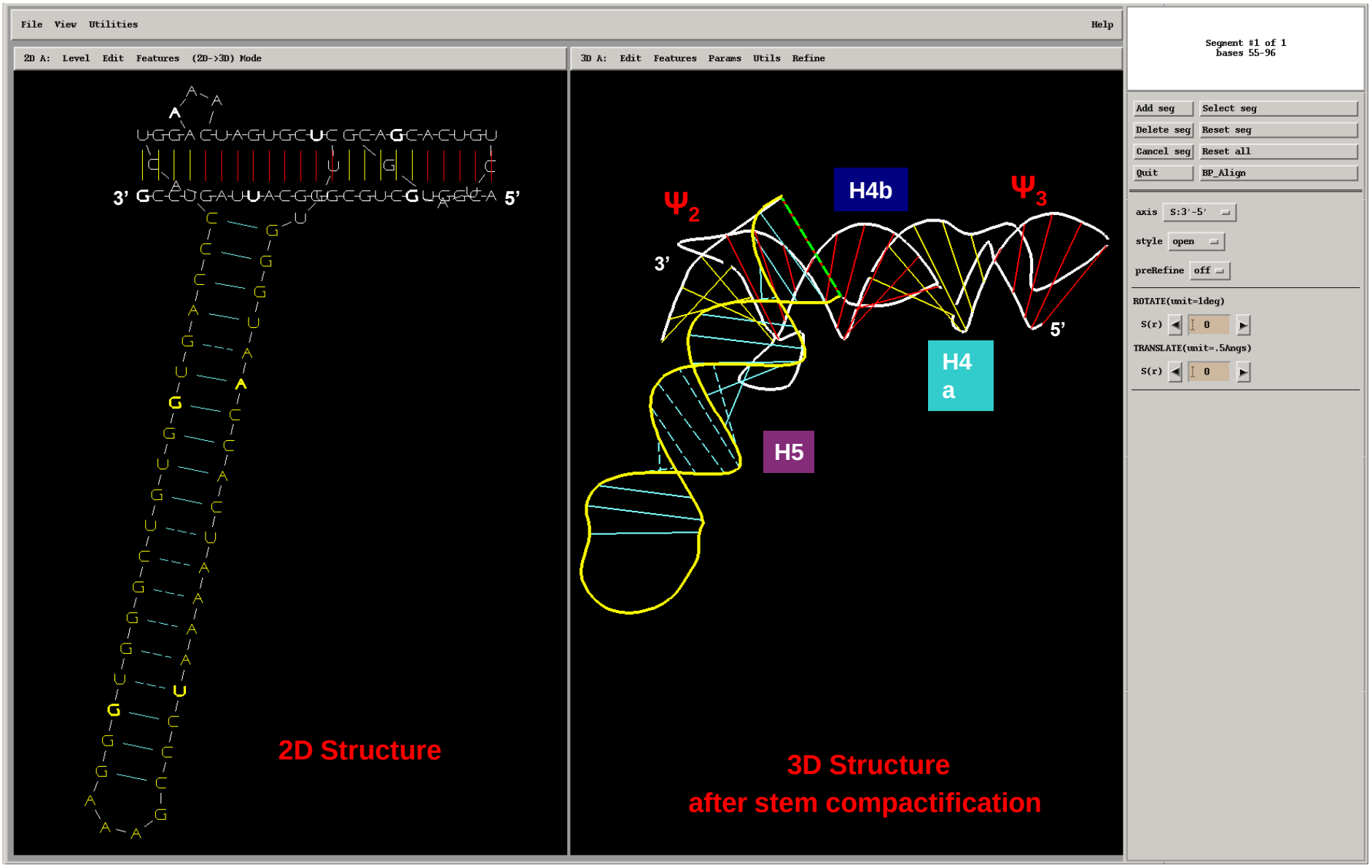
3D Structure Modeling with RNA2D3D (1)





Raw 2D to 3D transformation

3D Structure Modeling with RNA2D3D (3)

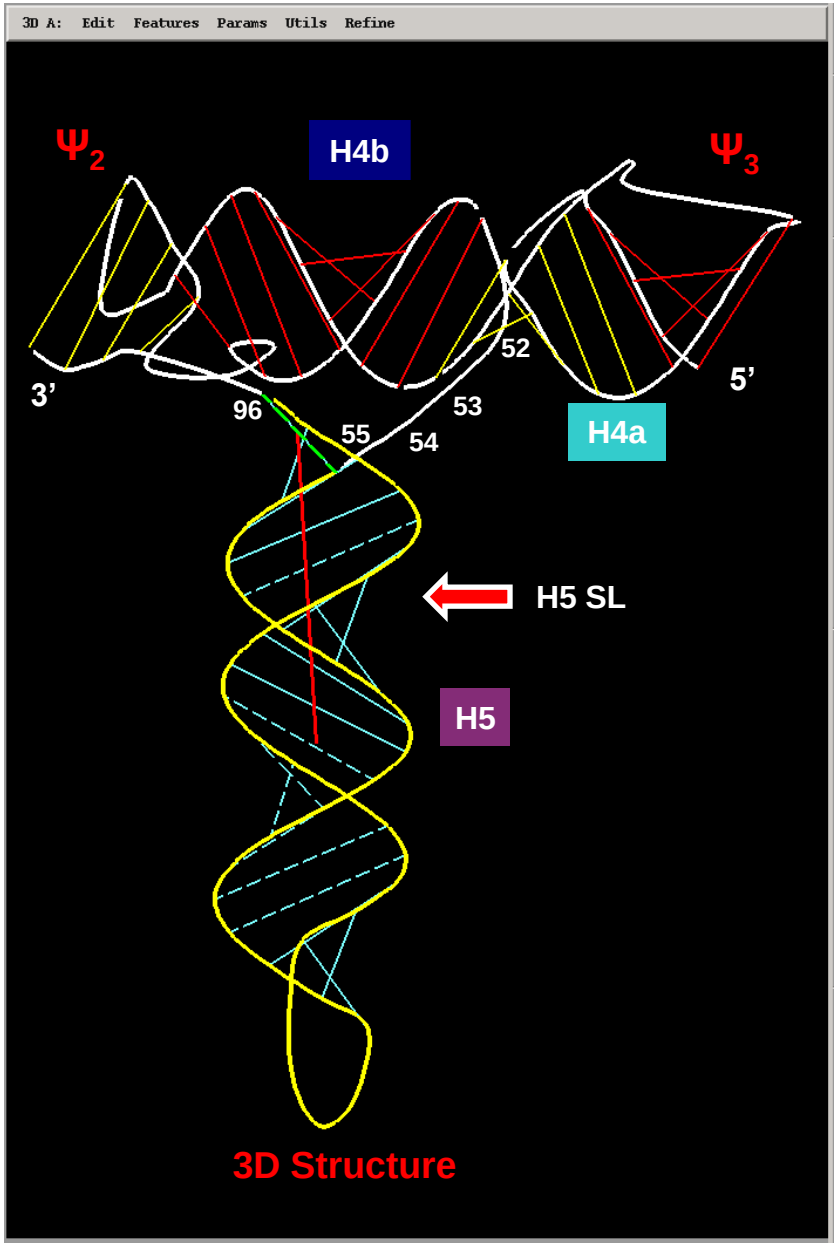


3D:Edit -> Segment Position (style=closed; preRef=on): H5:55-96, S:3'-5', Rotate = -75

3D Structure Modeling with RNA2D3D (5F)

Segment Position (style=closed; preRef=on):

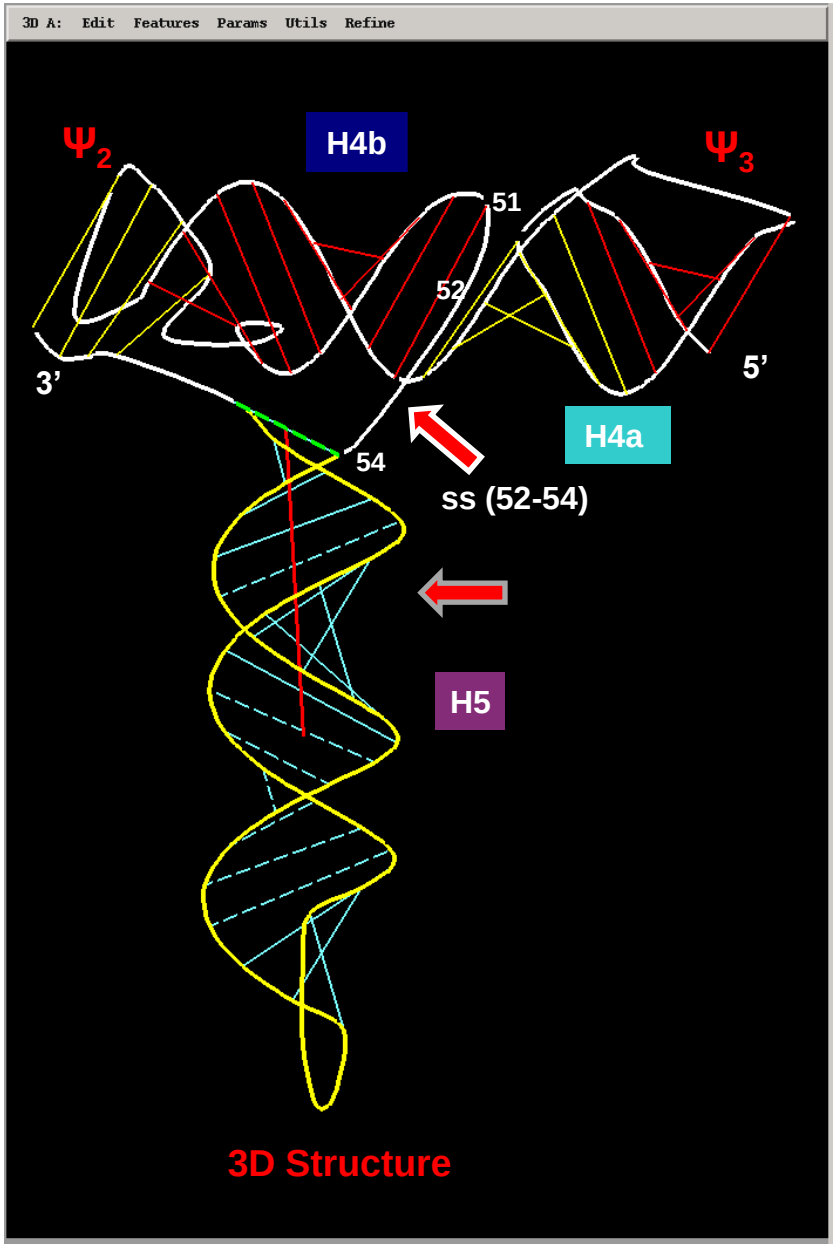
Segment	Axis	Rotation	Translation
H5: 55-96	3'-5'	R= -75 (+285)	
H5+:52-97	MP-COM		T = +5
H5+:53-96	X,Y,Z:3'		T(Y)= -15 (front & down)
H5+:54-96	MP-COM		T =+10 (down)
H5 :55-96	MP-COM		T =+4 (down) (pict. 5F)



3D Structure Modeling with RNA2D3D (6F)

Segment Position (style=closed; preRef=on):

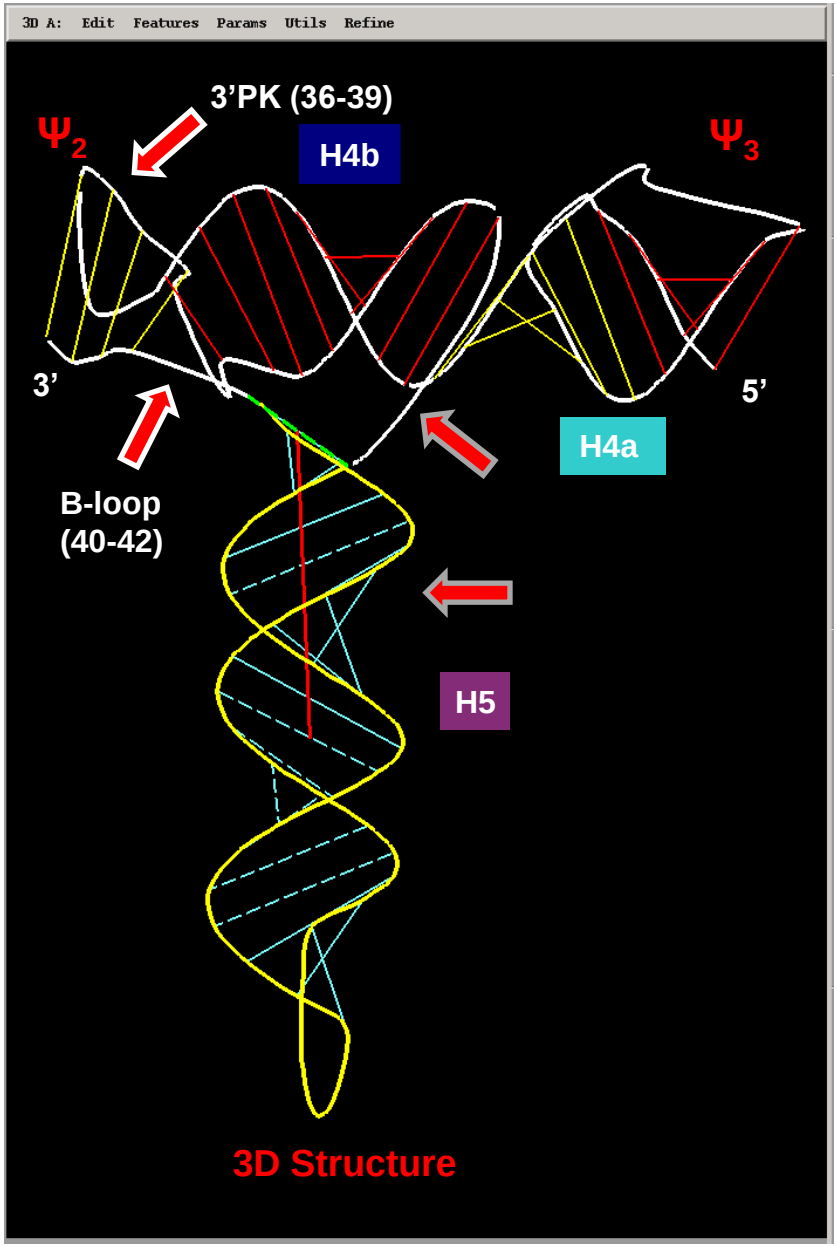
Segment	Axis	Rotation	Translation
H5: 55-96	3'-5'	R= -75	
H5+:52-97	MP-COM		T = +5
H5+:53-96	X,Y,Z:3'		T(Y)= -15 (front & down)
H5+:54-96	MP-COM		T =+10 (down)
H5 :55-96	MP-COM		T =+4 (down) (pict. 5F)
Link:51-54	**3'-5'**	**R= +105**	(move linker to the front)
Link:52-54	**3'-5'**	**R= -85**	(pict. 6F)



3D Structure Modeling with RNA2D3D (7F)

Segment Position (style=closed; preRef=on):

Segment	Axis	Rotation	Translation	
H5: 55-96	3'-5'	R= -75		
H5+:52-97	MP-COM		T = +5	
H5+:53-96	X,Y,Z:3'		T(Y)= -15 (front & down)	
H5+:54-96	MP-COM		T =+10 (down)	
H5 :55-96	MP-COM		T =+4 (down)	(pict. 5F)
Link:51-54	3'-5'	R= +105	(move linker to the front)	
Link:52-54	3'-5'	R= -85		(pict. 6F)
3'PK:36-39	X,Y,Z:3'		T(X)=-10	
3'BI:41-42	3'-5'		T =-10	
3'BI:40-41	3'-5'		T =-5	
3'BI:41-41	X,Y,Z:3'		T(Y)=-5	
3'BI:39-43	MP-COM	R=-30		
3'BI:39-41	MP-COM	R=-40		
3'BI:40-43	MP-COM	R=-10		
3'BI:39-43	3'-5'	R=+35		(pict. 7F)



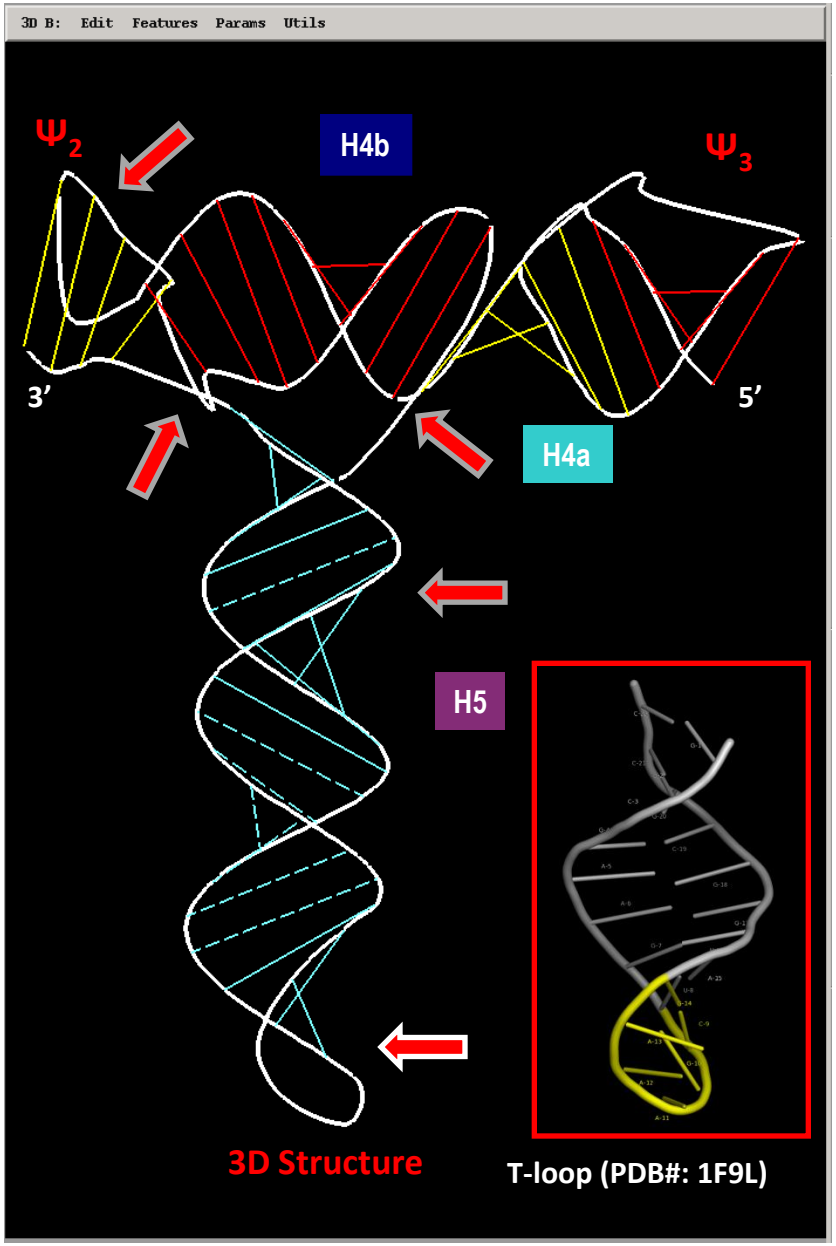
3D Structure Modeling with RNA2D3D (8F)

Segment Position (style=closed; preRef=on):

Segment	Axis	Rotation	Translation	
H5: 55-96	3'-5'	R= -75		
H5+:52-97	MP-COM		T = +5	
H5+:53-96	X,Y,Z:3'		T(Y)= -15 (front & down)	
H5+:54-96	MP-COM		T =+10 (down)	
H5 :55-96	MP-COM		T =+4 (down)	(pict. 5F)
Link:51-54	3'-5'	R= +105	(move linker to the front)	
Link:52-54	3'-5'	R= -85		(pict. 6F)
3'PK:36-39	X,Y,Z:3'		T(X)=-10	
3'BI:41-42	3'-5'		T =-10	
3'BI:40-41	3'-5'		T =-5	
3'BI:41-41	X,Y,Z:3'		T(Y)=-5	
3'BI:39-43	MP-COM	R= -30		
3'BI:39-41	MP-COM	R= -40		
3'BI:40-43	MP-COM	R= -10		
3'BI:39-43	3'-5'	R= +35		(pict. 7F)

Copy Model A -> Model B
read-in GAAA-tloop-1F9L-std into A
Substitute A(9..14) for B (73-78)
Save as 3DM, Copy back A ->B (pict. 8F)

Refinement: MIN ss (1.0); MIN full (1.0);
MIN-DYN-MIN full molecule (pict. 8F-R)



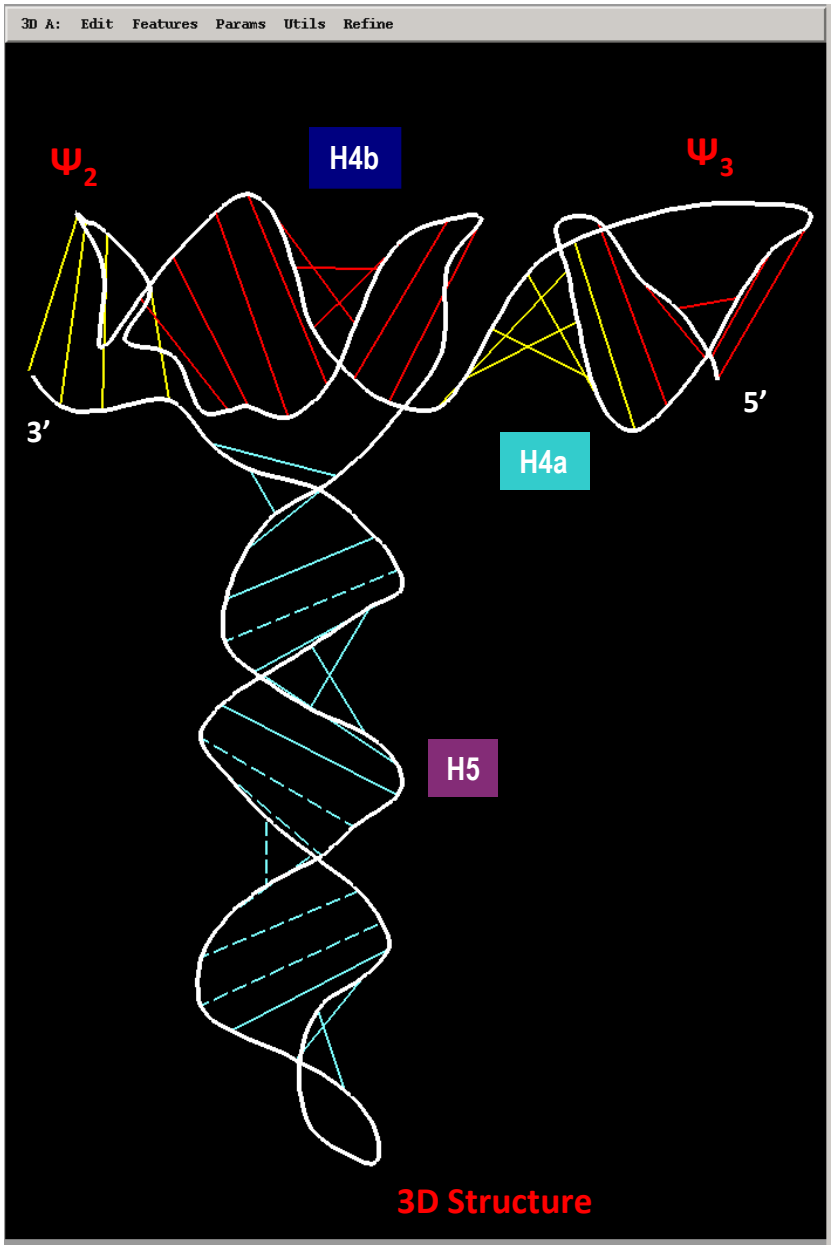
3D Structure Modeling with RNA2D3D (8F-R)

Segment Position (style=closed; preRef=on):

Segment	Axis	Rotation	Translation	
H5: 55-96	3'-5'	R= -75		
H5+:52-97	MP-COM		T = +5	
H5+:53-96	X,Y,Z:3'		T(Y)= -15 (front & down)	
H5+:54-96	MP-COM		T =+10 (down)	
H5 :55-96	MP-COM		T =+4 (down)	(pict. 5F)
Link:51-54	3'-5'	R= +105	(move linker to the front)	
Link:52-54	3'-5'	R= -85		(pict. 6F)
3'PK:36-39	X,Y,Z:3'		T(X)=-10	
3'BI:41-42	3'-5'		T =-10	
3'BI:40-41	3'-5'		T =-5	
3'BI:41-41	X,Y,Z:3'		T(Y)=-5	
3'BI:39-43	MP-COM	R= -30		
3'BI:39-41	MP-COM	R= -40		
3'BI:40-43	MP-COM	R= -10		
3'BI:39-43	3'-5'	R= +35		(pict. 7F)

Copy Model A -> Model B
read-in GAAA-tloop-1F9L-std into A
Substitute A(9..14) for B (73-78)
Save as 3DM, Copy back A ->B (pict. 8F)

Refinement: MIN ss (1.0); MIN full (1.0);
MIN-DYN-MIN full molecule (pict. 8F-R)



3D Structure Modeling with RNA2D3D (9F)

Segment Position (style=closed; preRef=on):

Segment	Axis	Rotation	Translation	
H5: 55-96	3'-5'	R= -75		
H5+:52-97	MP-COM		T = +5	
H5+:53-96	X,Y,Z:3'		T(Y)= -15 (front & down)	
H5+:54-96	MP-COM		T =+10 (down)	
H5 :55-96	MP-COM		T =+4 (down) (	pict. 5F)
Link:51-54	3'-5'	R=+105	(move linker to the front)	
Link:52-54	3'-5'	R=-85		(pict. 6F)
3'PK:36-39	X,Y,Z:3'		T(X)=-10	
3'BI:41-42	3'-5'		T =-10	
3'BI:40-41	3'-5'		T =-5	
3'BI:41-41	X,Y,Z:3'		T(Y)=-5	
3'BI:39-43	MP-COM	R=-30		
3'BI:39-41	MP-COM	R=-40		
3'BI:40-43	MP-COM	R=-10		
3'BI:39-43	3'-5'	R=+35		(pict. 7F)

Substitute T-loop A(9..14) for B (73-78) (pict. 8F)
Refinement: (MIN & MIN-DYN-MIN) (pict. 8F-R)

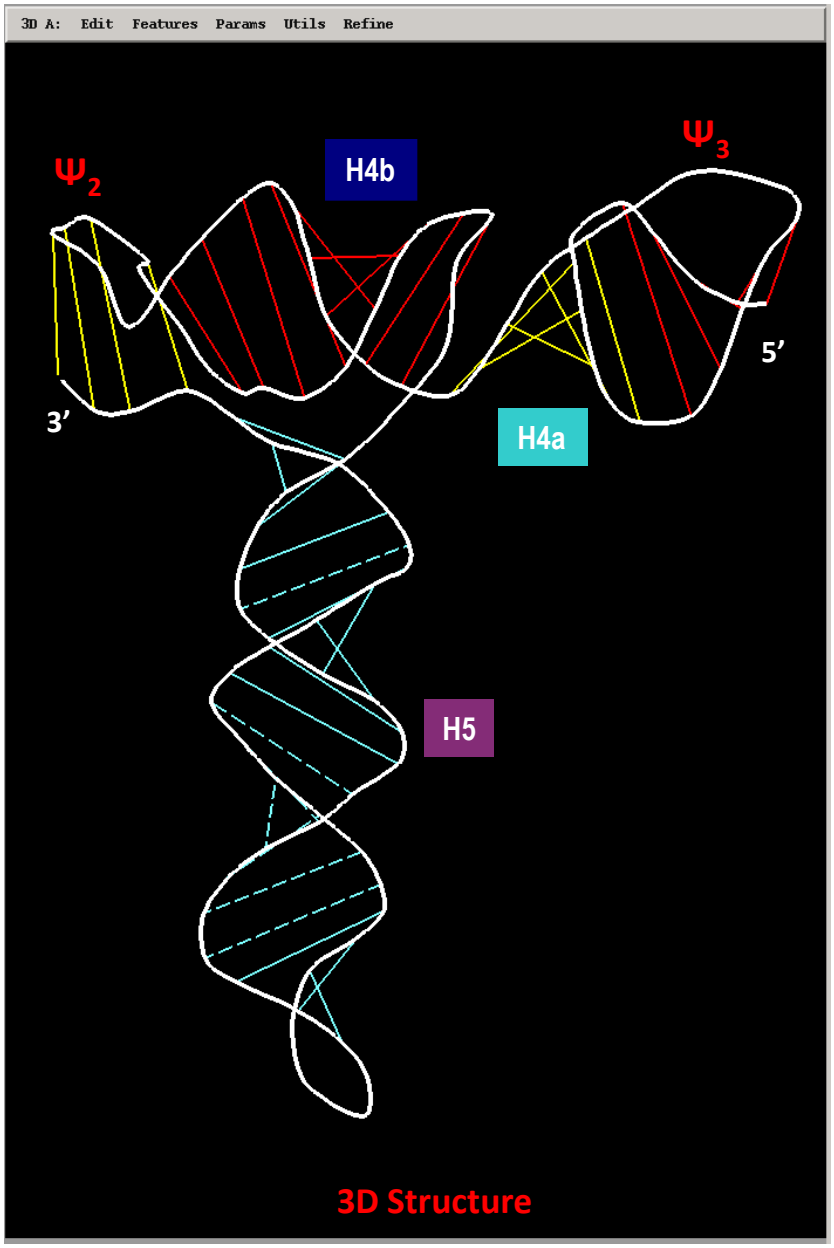
Stem-Group (full stem selection)

3'PK:full	coaxial	R(X)=-15; R(Z)=+20	T(X,Y,Z)=-1,-1,-1
5'PK+H4a	coaxial	R(X)=-10 ;R(Z)=+10	

Segment (as before)

6-6	X,Y,Z:3'	R=+125	T(Y)=-6; T(Z)=-2
1-5	X,Y,Z:5'		T(X)=+5; T(Z)=+10
16-20	3'-5	R=+41	
19-19	X,Y,Z:3'		T(X)=-7
2-2	X,Y,Z:3'		T(X)=-1; T(Y)=+2
1-1	X,Y,Z:3'		T(X)=+2
41-41	X,Y,Z:3'		T(X,Y,Z)=-1,-1,+15

(pict. 9F)



3D Structure Modeling with RNA2D3D (9F-R)

Segment Position (style=closed; preRef=on):

Segment	Axis	Rotation	Translation	
H5: 55-96	3'-5'	R= -75		
H5+:52-97	MP-COM		T = +5	
H5+:53-96	X,Y,Z:3'		T(Y)= -15 (front & down)	
H5+:54-96	MP-COM		T =+10 (down)	
H5 :55-96	MP-COM		T =+4 (down)	(pict. 5F)
Link:51-54	3'-5'	R=+105	(move linker to the front)	
Link:52-54	3'-5'	R=-85		(pict. 6F)
3'PK:36-39	X,Y,Z:3'		T(X)=-10	
3'BI:41-42	3'-5'		T =-10	
3'BI:40-41	3'-5'		T =-5	
3'BI:41-41	X,Y,Z:3'		T(Y)=-5	
3'BI:39-43	MP-COM	R=-30		
3'BI:39-41	MP-COM	R=-40		
3'BI:40-43	MP-COM	R=-10		
3'BI:39-43	3'-5'	R=+35		(pict. 7F)
Substitute T-loop A(9..14) for B (73-78)				(pict. 8F)
Refinement: (MIN & MIN-DYN-MIN)				(pict. 8F-R)

Stem-Group (full stem selection)

3'PK:full	coaxial	R(X)=-15; R(Z)=+20	T(X,Y,Z)=-1,-1,-1
5'PK+H4a	coaxial	R(X)=-10 ;R(Z)=+10	

Segment (as before)

6-6	X,Y,Z:3'	R=+125	T(Y)=-6; T(Z)=-2	
1-5	X,Y,Z:5'		T(X)=+5; T(Z)=+10	
16-20	3'-5	R=+41		
19-19	X,Y,Z:3'		T(X)=-7	
2-2	X,Y,Z:3'		T(X)=-1; T(Y)=+2	
1-1	X,Y,Z:3'		T(X)=+2	
41-41	X,Y,Z:3'		T(X,Y,Z)=-1,-1,+15	(pict. 9F)

REFINE (MIN-DYN-MIN) (pict. 9F-R)