

LIBRATOR V 1.0 - USER MANUAL

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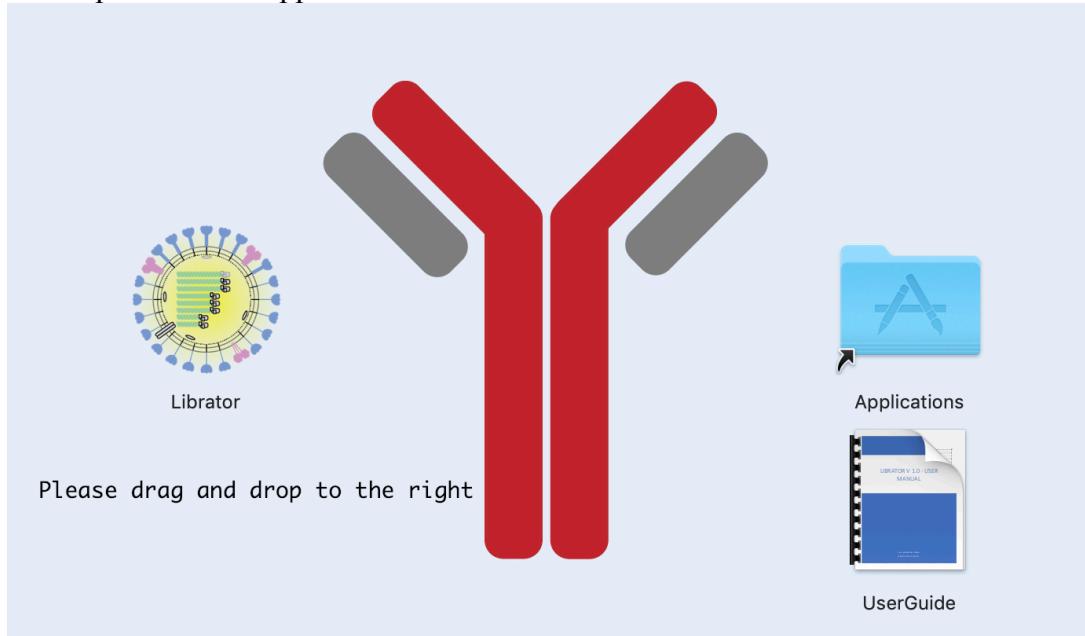
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Installation

Install Librator

Drag and drop Librator to application folder.



Install Dependencies

PyMOL (<https://pymol.org/2/>)

PyMOL for 3D structure visualization of proteins

Download from official website (license required after 30 days free trial)
or install from anaconda

```
conda install -c schrodinger pymol
```

or install from homebrew (recommended)

```
brew install brewsci/bio/pymol
```

or install from MacPorts

```
sudo port install pymol
```

or install from source (not recommended)

```
python setup.py --osx-frameworks install
```

After you installed PyMOL and setup the correct path (see basic function->path setting for details), Librator is ready to work. In case some pre-compile tools don't work on some specific MacOS version, you can download and install those missing packages by following our guidelines:

Clustal Omega

We already included pre-compiled clustal omega with Librator. If our version doesn't work on your computer, please download clustal omega from <http://www.clustal.org/omega/> and then setup the correct path in path setting (see basic function->path setting for details).

Muscle

We already included pre-compiled muscle with Librator. If our version doesn't work on your computer, please download muscle from <https://www.drive5.com/muscle/> and then setup the correct path in path setting (see basic function->path setting for details).

Home brew(<https://brew.sh/>)

The Missing Package Manager for macOS (or Linux). You may need the help form homebrew to install some missing packages. Open a terminal and paste the follow commands, then enter

```
/bin/bash -c "$(curl -fsSL
https://raw.githubusercontent.com/Homebrew/install/master/install.sh)"
```

After you installed home brew, you can install the following packages for better user experience.

GhostScript (<https://www.ghostscript.com/>) + **Pdf2svg**(<https://github.com/dawbarton/pdf2svg>)

This two libraries are required by embedded sequence logo viewer. Users still can view sequence logo on a popup window without this two libraries.

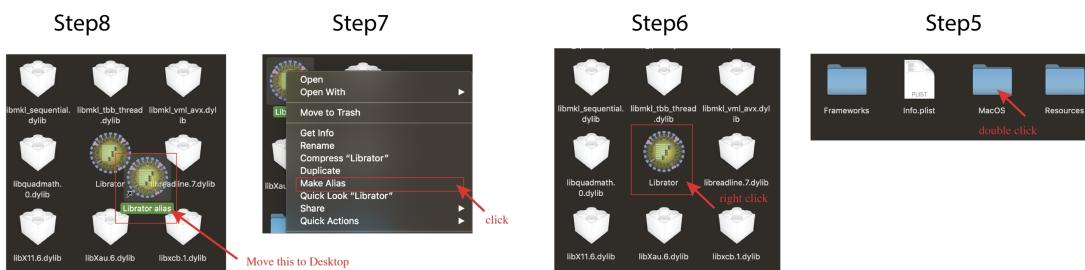
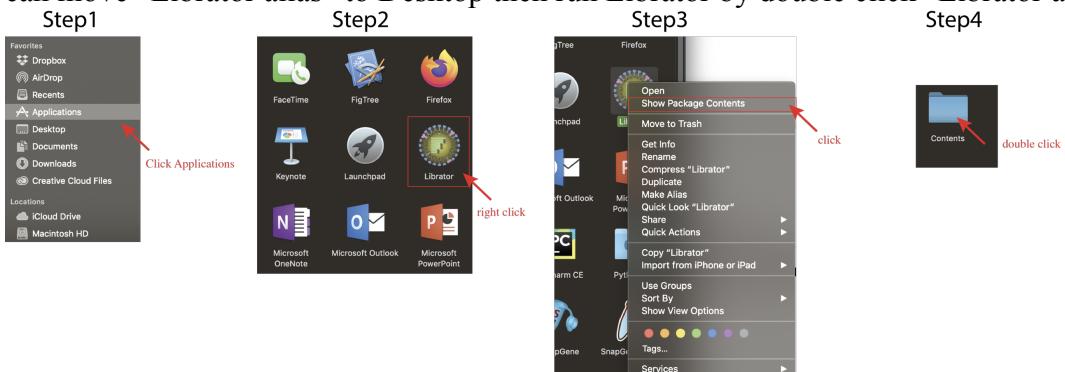
```
brew install ghostscript
brew install pdf2svg
```

Now you're all set!

Run Librator

Users can run Librator directly from Application->Librator (double click).

For better user experience (faster), users can go to Application folder and right click on Librator icon, then click "Show package contents", then go to Contents->MacOS, find file "Librator". Right click "Librator", choose "make alias", then a file called "Librator alias" will be generated. Users can move "Librator alias" to Desktop then run Librator by double click "Librator alias".



User interface

Main Tab (printable)

File Sequences Tools View Edit Sequence Visualization GibsonClone Help Setting

Tool bar

Main Sequence Alignment(RTF) Alignment(HTML) SequenceDB Summary Summary(HTML) FragmentDB

Tab switch

Open recent DB

Recent files: /Users/leil/Documents/Projects/Librator/Librator/DB/Test.lldb

Sequence editing functions

Copy selected sequences into another DB

Generate Sequences: Select report type:

Available sequences: All sequences in the tree can be grouped by Subtype, Role or Form

Group by: SubType

Active sequences: Highlight sequences by Role

Highlight: Unassigned

All sequences in current DB: A/Brasilia/02/2018[EPI_ISL_330190|2018-01-04|H1|1312566
A/Brasilia/59/2007 (H1)
A/California/04/2009 (H1)
A/Denver/1/1957 (H1)
A/Fort Monmouth/1/1947 (H1)
A/New Caledonia/20/1999 (H1)
A/New Caledonia/20/1999 (H1)-A3V(Cocktail)
A/New Caledonia/20/1999 (H1)-D52N(Cocktail)
A/New Caledonia/20/1999 (H1)-E290K(Cocktail)
A/New Caledonia/20/1999 (H1)-K157E(Cocktail)
A/New Caledonia/20/1999 (H1)-L432I(Cocktail)
A/New Caledonia/20/1999 (H1)-N203D(Cocktail)
A/New Caledonia/20/1999 (H1)-N489D(Cocktail)
A/New Caledonia/20/1999 (H1)-R205K(Cocktail)
A/New Caledonia/20/1999 (H1)-R225K(Cocktail)

Active sequences:

Import Sequences Import sequences **Delete Records** Delete records **Clear active sequence panel** **Reset active list**

A/New Caledonia/20/1999 (H1)-E290K(Cocktail)

Current sequence

Users can select one or multiple sequences and change values of those fields

Donor regions:

Role: Generated **Form:** Full HA **Subtype:** H1

100-200 Donor region information

Mutations:

E290K Mutation information information The residue position is count from the first AA of original sequence

DNA Sequence: position = 0 of 1711 total nucleotides

Coding region: Users can adjust coding region for selected sequence. This function is disabled for sequences with mutations

Coding Region: From: 1 To: 5000

Show Reading Frames

Open a popup window show all three Reading Frames

Edit Sequence

Search Motif search function

Amino Acid Sequence: position = 1 of 572 total residues

Nucleotide sequence

Redacted sequence

Design GibsonClones fragments for selected sequences

open PyMOL to visualize selected sequence

3D Visualization

Base Sequence: A/New Caledonia/20/1999 (H1)

Base sequence of current DB. It's unique.

Gibson Clones

Sequence Tab (printable)

File Sequences Tools View Edit Sequence Visualization GibsonClone Help Setting

Main Sequence Alignment(RTF) Alignment(HTML) SequenceDB Summary Summary(HTML) FragmentDB

Sequence Name: A/California/04/2009 (H1) -S75M(HA1)

Subtype: H1

Click feature to show for sequence:

H3 Numbering H1 Numbering Mutations Donor Regions

Sequence Map: Please scroll to refresh the alignment if the content doesn't change

A/California/04/2009 (H1) -S75M(HA1)

```

Position: ....5....10...15...20...25...30...35...40...45...50...55...6
H1-Numbering: -----10...15...20...25...30...35...40...45...
Sequence: MKAILVVLLYTFTANADTLIGIYHANNSTDVTVDLKEKNVTVTHSVNLLEDKHNGLCK

Position: 0...65...70...75...80...85...90...95...100...105...110...115..1
H1-Numbering: 50...55...60...65...70...75...80...85...90...95...100...105..
Sequence: LRGVAPLHLGKCNIAKGWILGNPECEMLSTASSWSYIVETPSSDNGTCYPGDFIDYEELRE

Position: 20..125..130..135..140..145..150..155..160..165..170..175..1
H1-Numbering: 110..115..120..125..130..135..140..145..150..155..160..165..
Sequence: QLSSVSSFEREIFFGKTSSWPNHDNSNKVTAACPHAGAKSFYKNLIVLVKKGNSYPKLSK

Position: 80..185..190..195..200..205..210..215..220..225..230..235..2
H1-Numbering: 170..175..180..185..190..195..200..205..210..215..220..225..
Sequence: SYINDKGKEVLVLWGIIHHPSTSADQSLYQNADTYVFVGSSRYSKKFPEIAIRPKVRDQ

Position: 40..245..250..255..260..265..270..275..280..285..290..295..3
H1-Numbering: 230..235..240..245..250..255..260..265..270..275..280..285..
Sequence: EGRMNYYWTLVEPGDKITFEATGNLVPRYAFAMERNAGSGIIISDTPVHDCNTTCQTPK

Position: 00..305..310..315..320..325..330..335..340..345..350..355..3
H1-Numbering: 290..295..300..305..310..315..320..325-----1..5...10..15
Sequence: GAIANTSPLFQNIHPITIGKCPKYVKSTKLATGLRNIPSIQSRLGAIAGFIEGGWTG

Position: 60..365..370..375..380..385..390..395..400..405..410..415..4
H1-Numbering: ..20..25..30..35..40..45..50..55..60..65..70..75
Sequence: MVDGWWYGHHQNEQGSGYAADLKSTQNAIDEITNKVNVSIEKMNTQFTAVGKEFNHLEKR

Position: 20..425..430..435..440..445..450..455..460..465..470..475..4
H1-Numbering: ..80..85..90..95..100..105..110..115..120..125..130..13
Sequence: IENLNKKVDDGFLDIWTYNAELLVLENERTLDYHDSNVKNLYEKVRSQLKNNAKEING

Position: 80..485..490..495..500..505..510..515..520..525..530..535..5
H1-Numbering: 5..140..145..150..155..160-----
Sequence: CFEFYHKCDNTCMESVKNGTYDYPKYSEEAKLNREEIDGVKLESTRIYQILAIYSTVASS

Position: 40..545..550..555..560....
H1-Numbering: -----
Sequence: LVLVVSLGAISFWMCNSGLQCRICI

Sequence elements: HA1 HA2 stop Transmembrane Trimerization-Avitag-H6 Mutations
H1 Antigenic Sites: Ca1 Ca2 Cb Sa Sb Stalk-MN
Position: Donor Region
```

3D Visualization Base Sequence: A/Wyoming/3/2003 (H3) Gibson Clones

Alignment Tab (HTML)

Sequence DB Tab

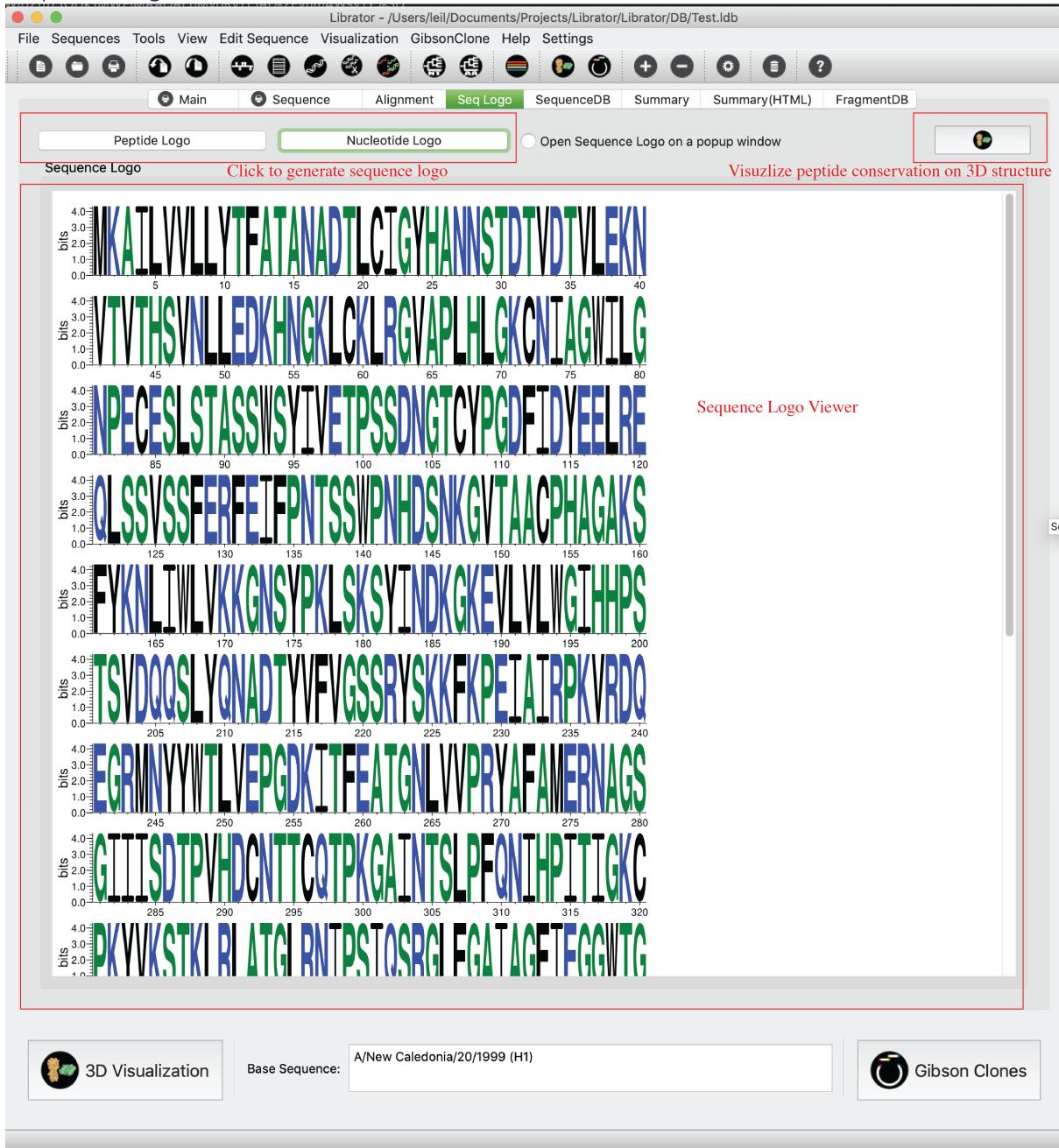
File Sequences Tools View Edit Sequence Visualization GibsonClone Help Setting

Main Sequence Alignment(RTF) Alignment(HTML) SequenceDB Summary Summary(HTML) FragmentDB

Current Sequence Database Double click on the field to edit records when Edit lock is unlocked Click to enable/disable Edit on the table Edit Lock

	SeqName	Sequence	SeqLen	Subtype	Form	VFrom	VTo	Active	Role	Donor	Mutations	ID	Base
4	B/Malaysia/2506/2004	ATGAAGGCA...	1824	B	Other	1	5000	False	Unassigned	none	none	0	
5	B/Florida/4/2006	ATGAAGGCA...	1755	B	Other	1	5000	False	Unassigned	none	none	0	
6	A/yellow shouldered bat/Guatemala/06/2010 ...	AGCAGAA...G	1784	H17	Full HA	1	5000	False	Unassigned	none	none	0	
7	A/turkey/Italy/22015/2002/EPI_ISL_2793 A / ...	AGCAAAAG...	1384	N3	Full NA	1	5000	False	Unassigned	none	none	0	
8	A/turkey/Italy/114845/2002/EPI_ISL_2792 A / ...	AGCAAAAG...	1384	N3	Full NA	1	5000	False	Unassigned	none	none	0	
9	A/turkey/Indiana/16-001403-1/2016 (H7)	TACAAAATG...	1705	H7	Full HA	1	5000	False	Unassigned	none	none	0	
10	A/teal/France/2546/2001/EPI_ISL_28881A / H1N3	GAGATGAAT...	1416	N3	Full NA	1	5000	False	Unassigned	none	none	0	
11	A/swine/Missouri/A01727926/2015 H4	ATGCATATCA...	1713	H4	Full HA	1	5000	False	Unassigned	none	none	0	
12	A/swine/Missouri/4296424/2006 (H2)	TTATTCTGTC...	1780	H2	Full HA	1	5000	False	Unassigned	none	none	0	
13	A/swine/MN/02011/2008 (H1)	ATGAAAGTGA...	1695	H1	Sequence	1	5000	True	Unassigned	none	none	0	
14	A/swine/Jiangsu/40/2011 (H1 avian-swine lineage)	ATGGAAAGCA...	1701	H1	Full HA	1	5000	True	Unassigned	none	none	0	
15	A/swine/Hubei/06/2009 H4	ATGCATATCA...	1738	H4	Full HA	1	5000	False	Unassigned	none	none	0	
16	A/swan/Shimane/227/01/EPI_ISL_5951A / H3N9	AAAGATGAAT...	1426	N9	Full NA	1	5000	False	Unassigned	none	none	0	
17	A/swan/Shimane/190/2001/EPI_ISL_6281A / H6N9	AAAGATGAAT...	1426	N9	Full NA	1	5000	False	Unassigned	none	none	0	
18	A/shoveler/Netherlands/18/1999 (H11)	ATGAAGAAA...	1728	H11	Full HA	1	5000	False	Unassigned	none	none	0	
19	A/shearwater/West Australia/2576/1979 (H15)	AGC AAAAG...	1763	H15	Full HA	1	5000	False	Unassigned	none	none	0	
20	A/hea/North Carolina/39482/1993 (H7)	AGC AAAAG...	1731	H7	Full HA	1	5000	False	Unassigned	none	none	0	
21	A/pintail/Shimane/324/98/EPI_ISL_4981A / H1N9	AAAGATGAAT...	1425	N9	Full NA	1	5000	False	Unassigned	none	none	0	
22	A/mallard/Sweden/81/2002 (H6)	AGC AAAAG...	1742	H6	Full HA	1	5000	False	Unassigned	none	none	0	
23	A/mallard/Netherlands/5/1999 (H2)	AGC AAAAG...	1771	H2	Full HA	1	5000	False	Unassigned	none	none	0	
24	A/mallard/Netherlands/12/2000 (H7)	AGC AAAAG...	1732	H7	Full HA	1	5000	False	Unassigned	none	none	0	
25	A/mallard/Interior Alaska/7MP0167/2007 (H12)	ATGGAAAAAA...	1712	H12	Full HA	1	5000	False	Unassigned	none	none	0	
26	A/mallard/Interior Alaska/10BM01929/2010 (H10)	GGTCACAAT...	1703	H10	Full HA	8	5000	False	Unassigned	none	none	0	
27	A/mallard/Guinea/263/19R2 (H1A)	AGC AAAAG...	1749	H14	Full HA	1	5000	False	Unassigned	none	none	0	

Sequence Logo Tab



Summary Tab



Summary Tab (HTML)



Fragment DB Tab

File Sequences Tools View Edit Sequence Visualization GibsonClone Help Setting

Main Sequence Alignment(RTF) Alignment(HTML) SequenceDB Summary Summary(HTML) FragmentDB

Setup Database information

Local DB (SQLite) Remote DB (MySQL)

Server IP: localhost Port: 3306 DB name: Librator
User Name: root Password: 123456

Connect to Local or remote Fragment DB Connect

DB content Content of fragment DB Edit "in stock" status here

	Name	Segment	Fragment	Subtype	ID	Template	AA seq	NT seq	In stock
1	N9-F3-0005	NA	3	N9	0005	A/duck/Hokkaido/W245/2004 EPI_ISL_645 ...	TCDRNWQGPNRPVIQIDPVAMTHTSQYICSP...	ACATGCAGAGACAATTGGCAGGGCCCAAATA...	Yes
2	N9-F3-0004	NA	3	N9	0004	A/swan/Shimane/190/2001 EPI_ISL_628 A / ...	TCDRNWQGSNRPVIIQIDPVAMTHTSQYICSP...	ACATGCAGAGATAATTGGCAAGGCTCAAATA...	No
3	N9-F3-0003	NA	3	N9	0003	A/swan/Shimane/227/01 EPI_ISL_595 A / ...	TCDRNWQGSNRPVIIQIDSVMTHTSQYICSP...	ACATGCAGAGATAATTGGCAAGGCTCAAATA...	No
4	N9-F3-0002	NA	3	N9	0002	A/duck/Hong Kong/562/1979 EPI_ISL_617 ...	TCDRNWQGSNRPVIIQINPTMMTHTSQYICSP...	ACGTGTAGAGACAATTGGCAAGGCTGAATA...	No
5	N9-F3-0001	NA	3	N9	0001	A/duck/Hong Kong/278/1978 EPI_ISL_619 ...	TCDRNWQGSNRPVIIQIDPTMMTHTSQYICSP...	ACGTGTAGGGACAATTGGCAAGGCTGAATA...	No
6	N9-F2-0006	NA	2	N9	0006	A/pintail/Shimane/324/98 EPI_ISL_498 A / ...	RFYALSQGTTIRGKHSNGTIHDRSQYRALISW...	AGGTTCTATGCTCTCAGCCAAGGGACAACAA...	No
7	N9-F2-0005	NA	2	N9	0005	A/duck/Hokkaido/W245/2004 EPI_ISL_645 ...	RFYALSQGTTIRGKHSNGTIHDRSQYRALISW...	AGGTTCTATGCTCTCAGCCAAGGGACAACAA...	No
8	N9-F2-0004	NA	2	N9	0004	A/swan/Shimane/190/2001 EPI_ISL_628 A / ...	RFYALSQGTTIRGKHSNGTIHDRSQYRALISW...	AGGTTCTATGCTCTCAGCCAAGGGACAACAA...	No
9	N9-F2-0003	NA	2	N9	0003	A/swan/Shimane/227/01 EPI_ISL_595 A / ...	RFYALSQGTTIRGKHSNGTIHDRSQYRALISW...	AGGTTCTATGCTCTCAGTCAAGGGACAACAA...	No
10	N9-F2-0002	NA	2	N9	0002	A/duck/Hong Kong/562/1979 EPI_ISL_617 ...	RFYALSQGTTIRGKHSNGTIHDRSQYRALISW...	AGATTCTATGCTCTCAGCCAAGGGACAACAA...	No
11	N9-F2-0001	NA	2	N9	0001	A/duck/Hong Kong/278/1978 EPI_ISL_619 ...	RFYALSQGTTIRGKHSNGTIHDRSQYRALISW...	AGATTCTATGCTCTCAGCCAAGGGACAACAA...	No
12	N9-F1-0007	NA	1	N9	0007	A/duck/Siberia/700/1996 EPI_ISL_618 A / ...	TCCACTCCCAGGTCCAACTGCACCTCGGTT...	TCCACTCCCAGGTCCAACTGCACCTCGGTT...	No
13	N9-F1-0006	NA	1	N9	0006	A/pintail/Shimane/324/98 EPI_ISL_498 A / ...	TCCACTCCCAGGTCCAACTGCACCTCGGTT...	TCCACTCCCAGGTCCAACTGCACCTCGGTT...	No
14	N9-F1-0005	NA	1	N9	0005	A/duck/Hokkaido/W245/2004 EPI_ISL_645 ...	TCCACTCCCAGGTCCAACTGCACCTCGGTT...	TCCACTCCCAGGTCCAACTGCACCTCGGTT...	No
15	N9-F1-0004	NA	1	N9	0004	A/swan/Shimane/190/2001 EPI_ISL_628 A / ...	TCCACTCCCAGGTCCAACTGCACCTCGGTT...	TCCACTCCCAGGTCCAACTGCACCTCGGTT...	No
16	N9-F1-0003	NA	1	N9	0003	A/swan/Shimane/227/01 EPI_ISL_595 A / ...	TCCACTCCCAGGTCCAACTGCACCTCGGTT...	TCCACTCCCAGGTCCAACTGCACCTCGGTT...	No
17	N9-F1-0002	NA	1	N9	0002	A/duck/Hong Kong/562/1979 EPI_ISL_617 ...	TCCACTCCCAGGTCCAACTGCACCTCGGTT...	TCCACTCCCAGGTCCAACTGCACCTCGGTT...	No
18	N9-F1-0001	NA	1	N9	0001	A/duck/Hong Kong/278/1978 EPI_ISL_619 ...	TCCACTCCCAGGTCCAACTGCACCTCGGTT...	TCCACTCCCAGGTCCAACTGCACCTCGGTT...	No

3D Visualization Base Sequence: A/Wyoming/3/2003 (H3) Gibson Clones

Functions:

Basic functions:

Path setting

Users can set paths for all required tools and databases.

Click *Setting-> Preferences* in menu or click  in *Tool bar*.

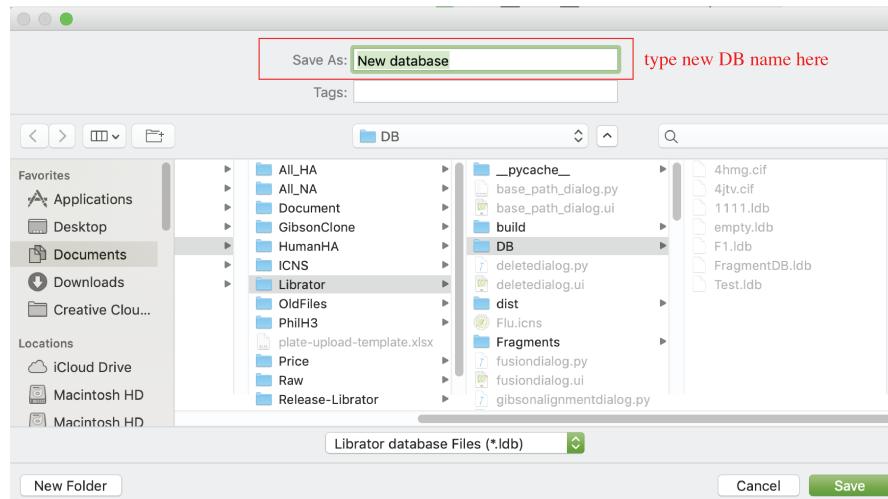


You can type path or click Browse button to choose the correct path.

Create new sequence database

Users can create new sequence database.

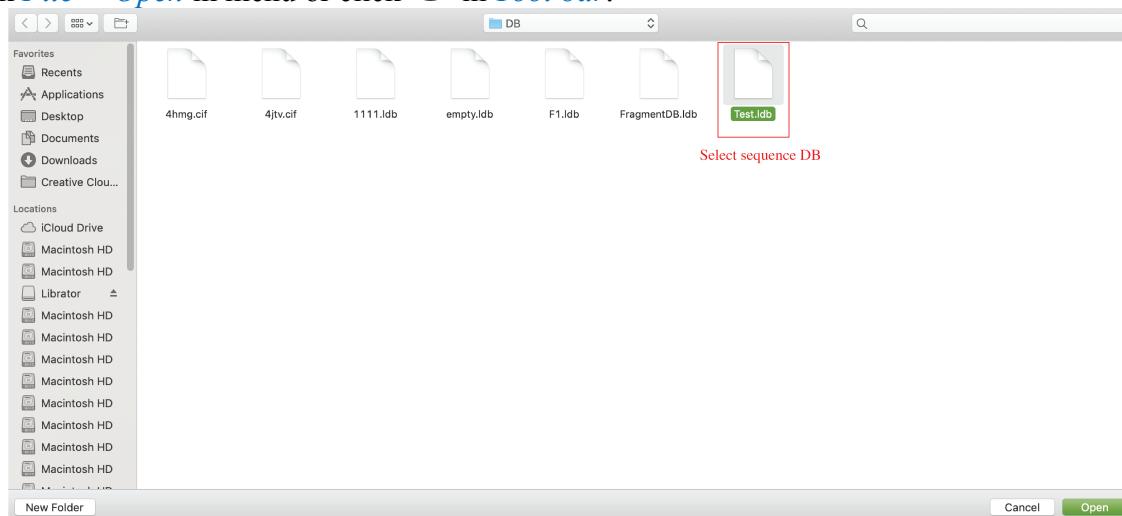
Click *File-> New* in menu or click  in *Tool bar*.



Open existing sequence database

Users can create new sequence database.

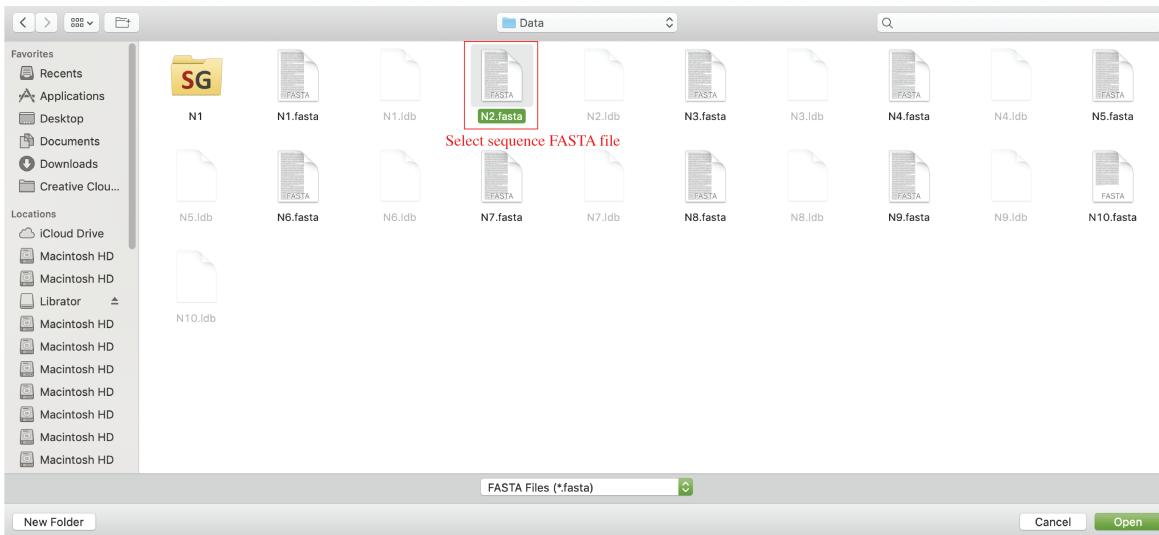
Click *File-> Open* in menu or click  in *Tool bar*.



Import sequences

After load an existing sequence database, users can import sequence into current database. The input files should be in FASTA format. Also please try to make your sequence name concise and easy to recognize. Try not to include space in your sequence name.

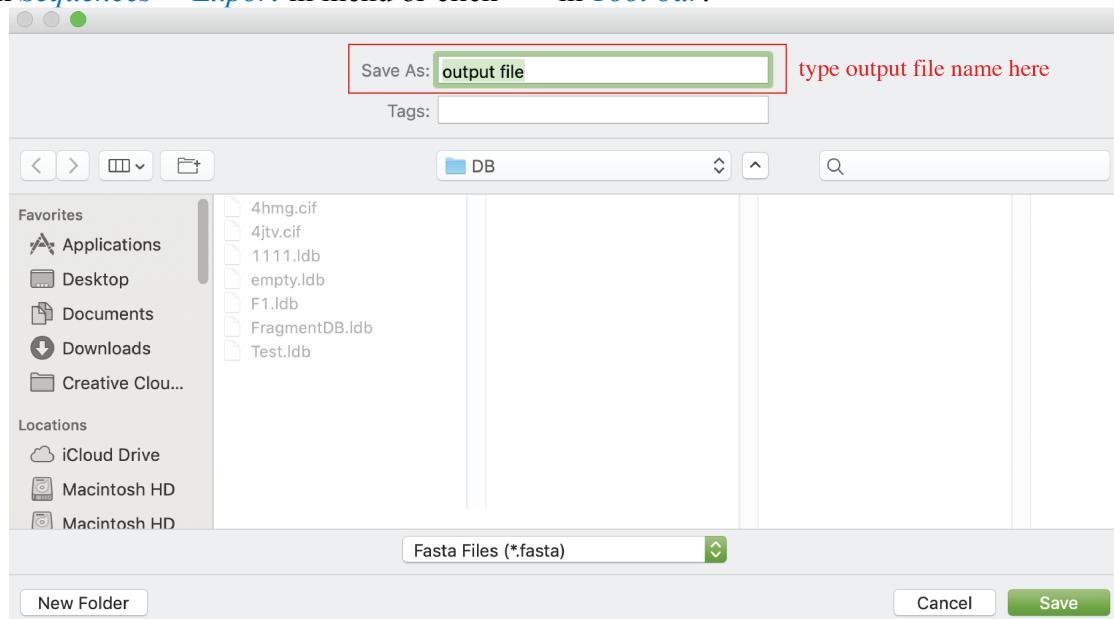
Click *Sequences-> Import* in menu or click  in *Tool bar* or click “import sequences” button in Main tab.



Export sequences

After load an existing sequence database, users can import sequence into current database. Selected sequences will be exported to a FASTA file.

Click *Sequences-> Export* in menu or click  in *Tool bar*.

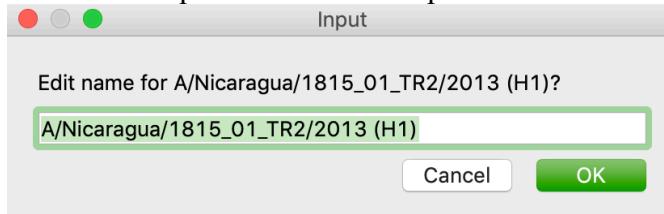


Advanced functions:

Edit information of sequences (on Main tab)

Users can edit information (**sequence name, Role, Form, Subtype, NT sequence, reading frame, donor regions**) of sequences on main tab.

For **Sequence name**, users can click sequence name text input to edit it.

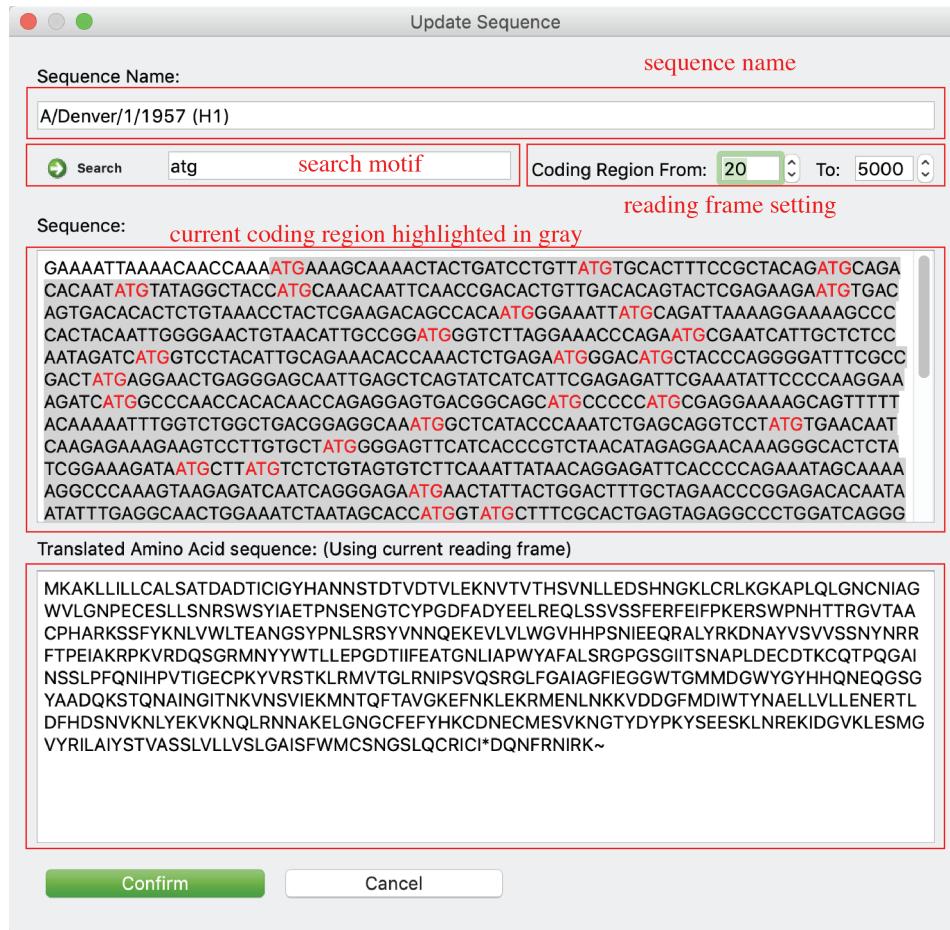


For **Role, Form, and Subtype**, users can edit by the left panel. Users can select multiple sequences in active sequence panel to do bulk update.

The screenshot shows the main application window. On the left, there's a tree view under 'Available sequences' with categories like 'All Sequences', 'Base Sequence', 'B', and 'H1'. Under 'Base Sequence', several sequences are listed with checkboxes. On the right, a list of 'Active sequences' is shown, also with checkboxes. A red box highlights the 'Edit here' section at the bottom left, which contains dropdown menus for 'Role' (Unassigned), 'Form' (Full HA), and 'Subtype' (H1). Below this, there are sections for 'Donor regions' (none) and 'Mutations' (none).

For **reading frame start and end**, users can use the “Coding region” inputs.

For **NT sequences**, users can click “edit sequence” button to edit sequences.



Note: coding region and NT sequence editing is disabled for sequences with mutation information.

For **donor regions** information, users can click “Donor region” input to update information.

HA numbering

Users can access H1/H3 numbering of selected sequence. Two ways:

- 1) Please select your sequence on active sequence panel and then click *Tools-> HA Numbering* in menu or just click “Sequence” tab.
Please see “Sequence” Tab for interface and details.

Sequence Map: Please scroll to refresh the alignment if the content doesn't change

A/Brisbane/59/2007 (H1)

Position:5....10....15....20....25....30....35....40....45....50....55....6
 H1-Numbering: -----10....15....20....25....30....35....40....45....
 Sequence: MKVKLLVLCTFTATYADTICIGYHANNSTDVTVDLKNVTVHSVNLLENSNGKLCL

Position: 0....65....70....75....80....85....90....95....100....105....110....115....1
 H1-Numbering: 50....55....60....65....70....75....80....85....90....95....100....105....
 Sequence: LKGIAPLQLGNCSVAGWILGNPECELLISKEWSYIIVEKPNPENGTCPGHFADYEELRE

Position: 20....125....130....135....140....145....150....155....160....165....170....175....1
 H1-Numbering: 110....115....120....125....130....135....140....145....150....155....160....165....1
 Sequence: QLSSVSSFERFEIFPKESSWPNHTVTGVSASCSSHNGESSFYRNLWLTKNGLYPNLSKS

Position: 80....185....190....195....200....205....210....215....220....225....230....235....2
 H1-Numbering: 70....175....180....185....190....195....200....205....210....215....220....225....2
 Sequence: YANNKEKEVLVLWGVHPPNIGDQKALYHTENAYSVVSSHYSRKFTPEIAKRPKVRDQE

Position: 40....245....250....255....260....265....270....275....280....285....290....295....3
 H1-Numbering: 30....235....240....245....250....255....260....265....270....275....280....285....2
 Sequence: GRINYYWTILLEPGDTIIFEANGNLIAPRYAFALSRGFGSGIINSNAPMDKCDAKCQTPQG

2) Please select your sequence on active sequence panel and then click “Alignment” tab or



Options:

AA NT H1 H3

Display Mode:

Original: Template:

Legend:

H1 highlight region:
H3 highlight region:

Ca1	Ca2	Cb	Sa	Sb	Stalk-MN	Lateral Patch
A	B	C	D	E	Stalk-MN	Lateral Patch

Position AA:	.	.	.	20	.	.	.	25	.	.	.	30	.	.	.	35	.	
H1 numbering	-	.	.	10	.	15	.	20	.	.	.	20	.	.	.	25	.	
H3 numbering	10	.	15	.	20	.	25	.	30	.	.	25	.	.	.	35	.	
Position NT:	50	..	55	..	60	..	65	..	70	..	75	..	80	..	85	..	90	..
A/Brisbane/59/2007 (H1)	A	D	T	I	C	I	G	Y	H	A	N	N	S	T	D	T	V	D
	CAGACACAATATGTATAGGCTACCAT	G	C	T	A	A	C	A	C	G	A	C	G	A	C	T	G	

Identify mutations

Users can identify mutations between any two sequences. If you have a template sequence and a mutated sequence (e.g. escape mutants) and want to quickly know the mutated residues, you can use this function to identify all mutations and annotate mutation information on your mutated sequence.

Click [Tools->Identify Mutation](#) in menu or just click in [Tool bar](#) to open the function window. Then select one sequence as template, determine the sequence that user want to identify mutation from as target sequence. Then the alignment will show in the HTML viewer and all

mutations will be listed in the bottom text box. Users can save mutation/template information to the target sequence by clicking “Confirm” button.

Template Sequence WT (1761 bp) Template sequence

Target Sequence Mutation (1761 bp) Target sequence

Sequence Alignment

Options:

AA NT H1 H3

H1 highlight region:	Ca1	Ca2	Cb	Sa	Sb	Stalk-MN
H3 highlight region:	A	B	C	D	E	Stalk-MN

Position AA:
H1 numbering
H3 numbering
Position NT:
WT (1761 bp)

Mutation (1761 bp)

Alignment Viewer

Mutations between current template and target sequences:
K142N,A209V,V273L

Mutation information

Confirm **Cancel**

Multiple sequence alignment

Users can align multiple sequences together and check the results in a graphical viewer.

Click [Tools->Multiple Alignment\(HTML\)](#) in menu or just click  in [Tool bar](#) for an alignment viewer in a popup window or click Alignment(HTML) tab for an integrated and alignment viewer. There are two display modes: original sequence mode (panel A) and template mode (panel B). In template mode, users can choose any sequence (including consensus sequence) as template to only highlight sequence differences.

Options: AA NT H1 H3

Display Mode: Original: Template: Alignment display options A

Legend:
H1 highlight region:

Ca1	Ca2	Cb	Sa	Sb	Stalk-MN
A	B	C	D	E	Stalk-MN

H3 highlight region:

Ca1	Ca2	Cb	Sa	Sb	Stalk-MN
A	B	C	D	E	Stalk-MN

Legend

Position AA:
H1 numbering
H3 numbering
Position NT:
A/Brisbane/02/2018IEPI_ISL
A/Brisbane/02/2018IEPI_ISL
A/Brisbane/59/2007 (H1)
A/Brisbane/59/2007 (H1)
A/California/04/2009 (H1)
A/California/04/2009 (H1)-
A/Denver/1/1957 (H1)
A/Fort Monmouth/1/1947
A/Fort Monmouth/1/1947

Position AA:
H1 numbering
H3 numbering
Position NT:
A/Brisbane/02/2018IEPI_ISL
A/Brisbane/02/2018IEPI_ISL
A/Brisbane/59/2007 (H1)
A/Brisbane/59/2007 (H1)
A/California/04/2009 (H1)
A/California/04/2009 (H1)-
A/Denver/1/1957 (H1)
A/Denver/1/1957 (H1)
A/Fort Monmouth/1/1947
A/Fort Monmouth/1/1947

Position AA:
H1 numbering
H3 numbering
Template NT:
Position NT:
A/Brisbane/02/2018IEPI_ISL
A/Brisbane/02/2018IEPI_ISL
A/Brisbane/59/2007 (H1)
A/Brisbane/59/2007 (H1)
A/California/04/2009 (H1)
A/California/04/2009 (H1)-
A/Denver/1/1957 (H1)
A/Denver/1/1957 (H1)
A/Fort Monmouth/1/1947
A/Fort Monmouth/1/1947

Generate phylogenetic tree (ML tree)

Users can generate phylogenetic tree (Maximum Likelihood tree) for selected sequences. Nucleotide and Amino Acid trees are available.

For Nucleotide tree:

Click *Tools-> Generate Maximum Likelihood Tree (nucleotide)* in menu or just click  in *Tool bar*

For Amino acid tree:

Click *Tools-> Generate Maximum Likelihood Tree (Amino Acid)* in menu or just click  in Tool bar.

Alignment Reviewer

Review alignment of your selected sequences:

Sequence: 35 to 35 (1 bases) selected of 581 BP select AA(NT) to see their position

A/Nicaragua/1815_01_TR2/2013_H1
A/California/04/2009_H1
A/California/04/2009_S75M_HA1
A/South_Carolina/1/1918_H1
A/Brisbane/59/2007_H1
A/Solomon_Islands/3/2006_H1
A/PR/8/1934_H1
A/Denver/1/1957_H1
A/Fort_Monmouth/1/1947_H1

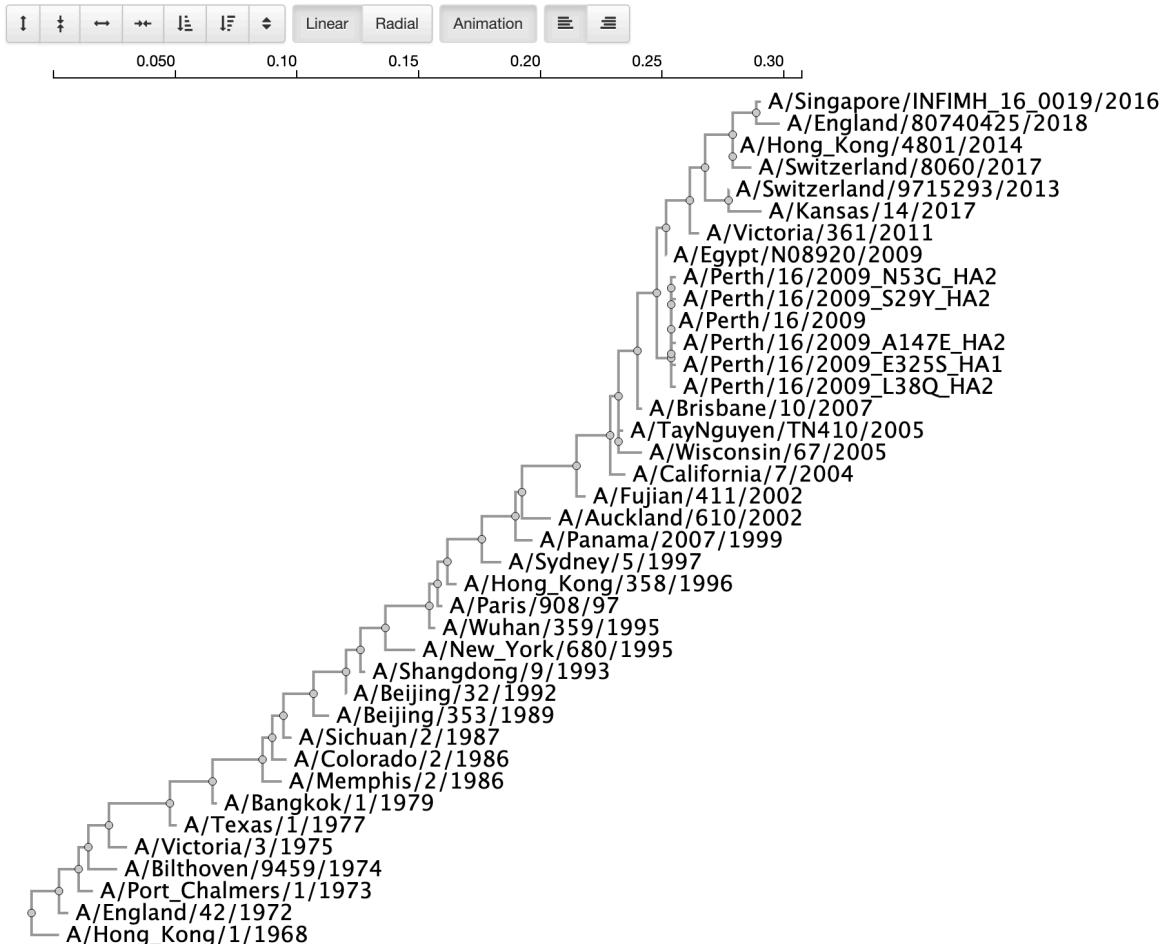
Click sequence name to highlight name and alignment by gray background

Sequence Name Sequence Alignment

Determine region:

Start:(0-5000) 5 End:(0-5000) 570 Sequence region to generate tree Generate Tree Cancel

Users are allowed to review alignments before building the tree. Users can determine a specific region of the alignment to avoid interference from sporadic insertion/deletion (as shown in the figure, selected regions were highlighted by red).

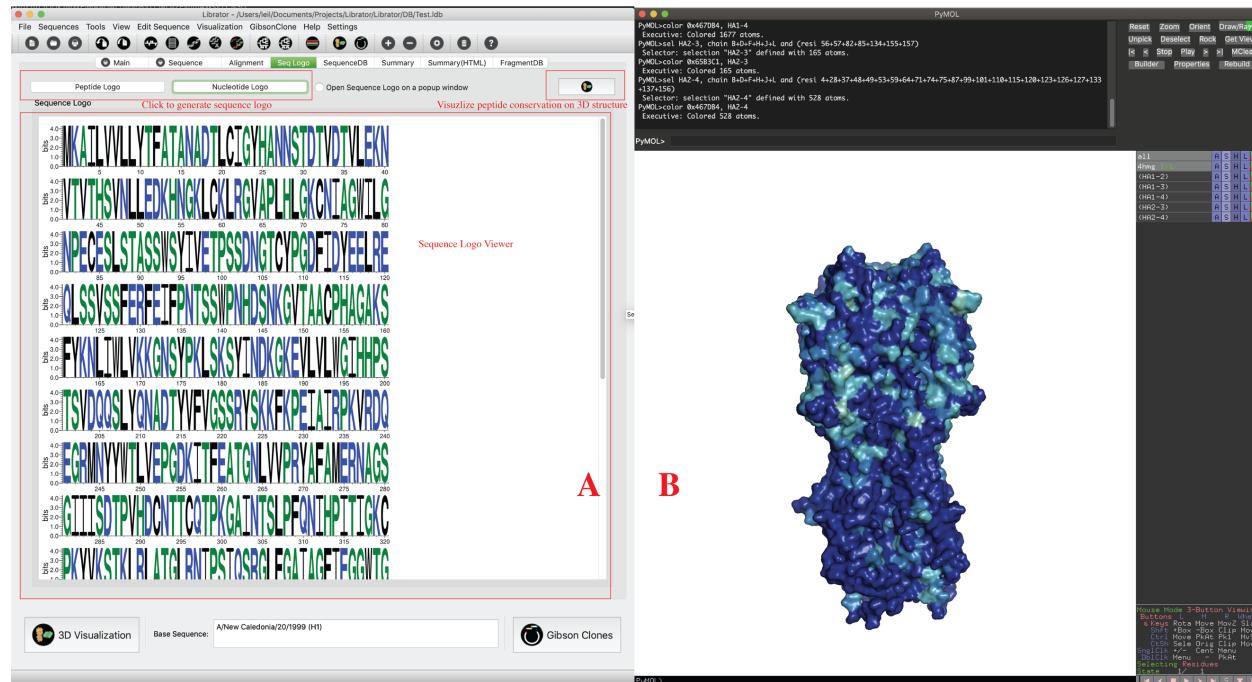


Generate sequence Logo (peptide/nucleotide conservation)

Users can generate sequence logo for multiple selected sequences.

Users should select multiple sequence in main page, then click “Seq logo” tab. On “Seq logo” tab, users can click *Peptide logo* or *Nucleotide logo* to generate sequences logos (Panel A). Users also can check the checkbox called “open Sequence logo on a popup window” to view the sequence logo on a popup window and then can be saved as a EPS file (vectorgraph) for further use.

Users also can click the button on the right top corner to visualize peptide conservations for selected sequences on a 3D structure using PyMOL (panel B).



Mutate sequence

Users can generate new sequences with mutations based on any existing sequence.

Click *Edit Sequence-> Mutation* in menu or just click in *Tool bar*.

As shown below, users can generate mutations on a template sequence using either original position (count from first amino acid, panel A) or H1/H3 numbering (H1 numbering for group1 virus and H3 numbering for group2 virus, panel B). H1/H3 numbering function is only enabled for HA sequences of FLU A.

Users are allowed to input multiple mutations at the same time, and are allowed to decide from two options: 1) generate one sequence with all mutations; 2) generate multiple sequences, each of them with a single mutation.

After the mutated sequence was generated, users can visualize the 3D structure (only enabled for HA now) of generated HA protein (panel C).

A

Please type your mutations below: e.g. R98Y, K141E
Current Sequence: A/California/04/2009 (H1)

Original Position H1/H3 Numbering Count from first Amin Acid

Mutations S86M,L87Q,S88N

HA1 mutations I85Q
HA2 mutations I10M

New SEQ Name A/California/04/2009 (H1) -S86M,L87Q,S88N
 Single Sequence with all mutations Sequences with each single mutation

Generate Sequence Cancel

B

Please type your mutations below: e.g. R98Y, K141E
Current Sequence: A/California/04/2009 (H1)

Original Position H1/H3 Numbering H1/H3 numbering

HA1 mutations I85Q
HA2 mutations I10M

Generate multiple sequences each with a single mutation
New SEQ Name A/California/04/2009 (H1) -I85Q(HA1)I10M(HA2)
 Single Sequence with all mutations Sequences with each single mutation

Generate Sequence Cancel

C

all A S H L D
4jtv 1/1 A S H L D
(ABS-Ca1) A S H L D
(ABS-Ca2) A S H L C
(ABS-Cb) A S H L D
(ABS-Sa) A S H L D
(ABS-Sb) A S H L D
(haimutation) A S H L D

Mouse Mode 3-Button Viewing
Buttons L M R Wheel
& Keys Rota Move MovZ Slab
Shft +Box -Box Clip MovS
Ctrl Move PkAt Pk1 MvSz
CtSh Sele Drig Clip MovZ
SnglClk +/- Cent Menu
DblClk Menu - PkAt
Selecting Residues
State 1/ 1

PyMOL>_

Compare sequences and generate screening mutations

Users can compare sequences and generate consensus sequences (Base biased) or new sequences with screening mutations (Cocktail).

Click *Edit Sequence-> Editing* in menu or just click in *Tool bar*.

Cocktail mode (A):

Compare base sequence to another sequence, identify all mutations between two sequences on donor region of donor sequence (user can setup donor region on this interface), then generate new sequences with those mutations using base sequence as template. Users can choose to only generate sequences with single mutations or all combinations of mutations.

Base biased mode (B):

Generate consensus sequences of base sequence and selected sequences. For each donor sequence, only their donor region will be considered (full sequence will be considered when donor region is none).

The figure shows two panels of the 'Edit Sequences' software. Panel A is active, displaying a 'Donor Sequence List' with various influenza sequences. Panel B is shown as a preview or a second instance of the interface.

Panel A (Active):

- Base Sequence:** A/Brisbane/59/2007 (H1)
- Buttons:** Cocktail, Base Biased
- Donor sequence (single selection):**
 - A/Brisbane/59/2007 (H1) - N52D(Cocktail)
 - A/Brisbane/59/2007 (H1) - Q239R(Cocktail)
 - A/Brisbane/59/2007 (H1) - R162K(Cocktail)
 - A/Brisbane/59/2007 (H1) - V145T(Cocktail)
 - A/California/04/2009 (H1)
 - A/Denver/1/1957 (H1)
 - A/Fort Monmou/1/1947 (H1)
 - A/New Caledonia/20/1999 (H1)
 - A/Nicaragua/1815_01_TR2/2013 (H1)
 - A/PR/8/1934 (H1)
 - A/shoveler/Netherlands/18/1999 (H1)
 - A/Solomon Islands/3/2006 (H1)
 - A/South Carolina/1/1918 (H1)
 - A/swine/Janzen/10/2011/H1 avian-swine lineage
- Donor Region:** From: 100 To: 200
- Sequence Comparision:**
 - Options:** AA, NT, H1, H3 (all checked)
 - H1 highlight region:** Ca1, Ca2, Cb, Sa, Sb, Stalk-MN
 - H3 highlight region:** A, B, C, D, E, Stalk-MN
 - Donor region:** M, ATG
 - Sequence Comparision:** Shows positions 100-115 for AA, 90-105 for NT, and 295-350 for H1/H3. Below is a sequence alignment of the donor region for four strains.
- Buttons at bottom:** Only single mutations, All possible combinations, options
- Buttons at bottom right:** Generate Sequences, Cancel

Panel B (Inactive):

- Base Sequence:** B A/Brisbane/59/2007 (H1)
- Buttons:** Cocktail, Base Biased
- Donor Sequences (multiple selection):**
 - A/Brisbane/59/2007 (H1) - K205R(Cocktail)
 - A/Brisbane/59/2007 (H1) - K290E(Cocktail)
 - A/Brisbane/59/2007 (H1) - K90R(Cocktail)
 - A/Brisbane/59/2007 (H1) - L540R(Cocktail)
 - A/Brisbane/59/2007 (H1) - N52D(Cocktail)
 - A/Brisbane/59/2007 (H1) - Q239R(Cocktail)
 - A/Brisbane/59/2007 (H1) - R162K(Cocktail)
 - A/Brisbane/59/2007 (H1) - V145T(Cocktail)
 - A/California/04/2009 (H1)
 - A/Denver/1/1957 (H1)
 - A/Fort Monmou/1/1947 (H1)
 - A/New Caledonia/20/1999 (H1)
 - A/Nicaragua/1815_01_TR2/2013 (H1)
 - A/PR/8/1934 (H1)
 - A/shoveler/Netherlands/18/1999 (H1)
 - A/Solomon Islands/3/2006 (H1)
 - A/South Carolina/1/1918 (H1)
 - A/swine/Jiangsu/40/2011 (H1 avian-swine lineage)
 - A/swine/MN/02/2011/2008 (H1)
 - A/Texas/36/1991 (H1)
 - A/USSR/1977 (H1)
- Buttons at bottom right:** Generate Sequences, Cancel

Epitope transplant across different subtypes

Users can transplant epitopes/regions from sequences of different subtypes. This function could be helpful for designing chimeric HA protein.

Regions being removed from base sequence and that being inserted from donor sequence are not necessary to have same length.

Users can add multiple replacements on one sequence (click ‘Add’). Those replacements could be from different sequences. Users can review the replacement design in the bottom window.

Users can double click replacement to delete it. Users also can click 'Clear' button to rest current design.

Click *Edit Sequence-> Fusion* in menu or just click  in *Tool bar*. This window has two layouts: high resolution layout and low resolution layout. Liberator will automatically choose the best fit based on current display resolution. Users also can click *Edit Sequence-> Fusion (high resolution)* or *Edit Sequence-> Fusion (low resolution)* to determine the layouts they prefer (As shown below).

Sequence Fusion across subtypes

High Resolution Layout

Select donor sequence:

- A/Brisbane/02/2018/EPI_ISL_330190/20
- A/Brisbane/59/2007 (H3)
- A/California/04/2009 (H1)
- A/California/04/2009 (H1) -10M(HA2)
- A/California/04/2009 (H1) -18Q(HA1)
- A/California/04/2009 (H1) -S75M(HA1)
- A/Denver/1/1957 (H1)
- A/Fort Monmouth/1947 (H1)
- A/Iceland/1815_01/TR2/2013 (H1)
- APR/0/1934 (H1)
- A/Solomon Islands/3/2006 (H1)
- A/South Carolina/1918 (H1)
- A/swine/Jiangsu/40/2011 (H1 avian-swine)
- A/swine/MN/0201/2008 (H1)
- A/Texas/36/1991 (H1)
- A/USSR/1977 (H1)

Active sequence list

Base Sequence: A/Hong Kong/1/1968 (H3)

Base sequence

H3-Numbering: Position: 1...6...11...16...21...26...31...36...41...46...51...56...
H1-Numbering: ...1...5...10...15...20...25...30...35...40...45...50...
Sequence: M K T I I A L S Y I F C L A L G Q D L P G N D N S T A T L C L G H H A V P N G T I V K T I T D D Q I E V T N A T E L V Q

H3-Numbering: Position: 61...66...71...76...81...86...91...96...101...106...111...116...
H1-Numbering: ...10...15...20...25...30...35...40...45...50...
Sequence: S S T G K T C H I R I L L D C T I D A L L G D P H C D V F Q N T D W I L V E R S K A F S N C Y Y D P V

H3-Numbering: Position: 121...126...131...136...141...146...151...156...161...166...171...176...
H1-Numbering: ...105...110...115...120...125...130...135...140...145...150...155...160...
Sequence: Y A S L R S I V A S S G T L E F I T Q N G S S A C K S P G G F S R L N L W T K S G S T P V

H3-Numbering: Position: 181...186...191...196...201...206...211...216...221...226...231...236...
H1-Numbering: ...165...170...175...180...185...190...195...200...205...210...215...220...
Sequence: N V T M P N N D N F U K L Y I N G W H H H P S T N E C T L Y Q A S G R V T V S T R S Q Q T I I P N I G S R P W

H3-Numbering: Position: 241...246...251...256...261...266...271...276...281...286...291...296...
H1-Numbering: ...205...210...215...220...225...230...235...240...245...250...255...260...265...270...275...280...285...
Sequence: G L S R R I S Y W T I V K P G D V L V I N S N G N L I A P R G Y K M R T G K S I M B S D A P I D T C I S E C T P

Position: 301..306..311..316..321..326..331..336..341..346..351..356..

Donor Sequence: A/Brisbane/59/2007 (H1)

Selected donor sequence

H3-Numbering: Position: 1...6...11...16...21...26...31...36...41...46...51...56...
H1-Numbering: ...1...5...10...15...20...25...30...35...40...45...50...
Sequence: V K L L V L C L T T F A T Y A D T I C G H A N N S T D T V D L E R N V T T H S V N L E N S H N G K C L L R

H3-Numbering: Position: 61...66...71...76...81...86...91...96...101...106...111...116...
H1-Numbering: ...10...15...20...25...30...35...40...45...50...
Sequence: G T A P I Q O N S E V A G N I L G N F C E L L I S E K S W Y I V E K P N E G T C Y P H F H A D Y E E L R Q L

H3-Numbering: Position: 121...126...131...136...141...146...151...156...161...166...171...176...
H1-Numbering: ...0...115...120...125...130...135...140...145...150...155...160...165...
Sequence: S S V S F S P E R F I F K E S N F N H P I C Q A L Y H E T A N V Y V V S S S Y S R K F T E I A K R P K V R V D Q E G R

H3-Numbering: Position: 181...186...186...191...196...201...206...211...216...221...226...231...236...
H1-Numbering: ...170...175...180...185...190...195...200...205...210...215...220...225...
Sequence: N H N K E K V E L P D T I I F E R A N G N L I A P R A F A L S R G F G S G J I N S N A P M D C D A K C Q T P Q G A I

Position: 301..306..311..316..321..326..331..336..341..346..351..356..

Current Base sequence:

A/Hong Kong/1/1968 (H3)

Base sequence name

Base sequence

Replaced region on base sequence: Donor region on donor sequence: Add
60 □ to 70 □ 60 □ to 70 □ Clear

Region on base sequence to be replaced Region on donor sequence to be inserted

Current product

Legend: H1 numbering Cal A Cal B Ca2 C Cb D Sa E Sb F Stalk-MN G Stalk-MN H

Position AA: . . . 5 . . . 10 . . . 15 . . . 20 . . . 25 . . . 30 . . . 35 . . . 40 . . . 45 . . . 50 . . . 55 . . . 60 . . . 65 . . . 70 . . . 75 . . . 80 . . . 85 . . . 90

H1 numbering . . . 5 . . . 10 . . . 15 . . . 20 . . . 25 . . . 30 . . . 35 . . . 40 . . . 45 . . . 50 . . . 55 . . . 60 . . . 65 . . . 70 . . . 75 . . . 80 . . . 85 . . . 90

H3 numbering . . . 5 . . . 10 . . . 15 . . . 20 . . . 25 . . . 30 . . . 35 . . . 40 . . . 45 . . . 50 . . . 55 . . . 60 . . . 65 . . . 70 . . . 75 . . . 80 . . . 85 . . . 90

Original Seq: M K T I I A L S Y I F C L A L G Q D L P G N D N S T A T L C L G H H A V P N G T I V K T I T D D Q I E V T N A T E L V Q

M K T I I A L S Y I F C L A L G Q D L P G N D N S T A T L C L G H H A V P N G T I V K T I T D D Q I E V T N A T E L V Q

FTATYADTICI TVDTVLEKNTV KLCLLKGIAPL

Double click to delete

Insertion information

Confirm Cancel

Sequence Fusion across subtypes

Low Resolution Layout

Select donor sequence:

- A/Brisbane/02/2018/EPI_ISL_330190/201
- A/Brisbane/59/2007 (H1)
- A/California/04/2009 (H1)
- A/Denver/1/1957 (H1)
- A/Fort Monmouth/1947 (H1)
- A/Iceland/1815_01/TR2/2013 (H1)
- A/ICaragua/1815_01(TR2/2013 (H1)
- APR/0/1934 (H1)
- A/Solomon Islands/3/2006 (H1)
- A/South Carolina/1918 (H1)
- A/swine/Jiangsu/40/2011 (H1 avian-swine)
- A/swine/MN/0201/2008 (H1)
- A/Texas/36/1991 (H1)
- A/USSR/1977 (H1)

Active sequence list

Base Sequence: A/Wisconsin/6/2005 (H3)

Base sequence

H3-Numbering: Position: 1...6...11...16...21...26...31...36...41...46...51...56...
H1-Numbering: ...1...5...10...15...20...25...30...35...40...45...
Sequence: M K T I I A L S Y I F C L A L G Q D L P G N D N S T A T L C L G H H A V P N G T I V K T I T D D Q I E V T N A T E L V Q

H3-Numbering: Position: 61...66...71...76...81...86...91...96...101...106...111...116...
H1-Numbering: ...10...15...20...25...30...35...40...45...50...
Sequence: S S T G K T C H I R I L L D C T I D A L L G D P H C D V F Q N T D W I L V E R S K A F S N C Y Y D P V

H3-Numbering: Position: 121...126...131...136...141...146...151...156...161...166...171...176...
H1-Numbering: ...105...110...115...120...125...130...135...140...145...150...155...160...
Sequence: Y A S L R S I V A S S G T L E F I T Q N G S S A C K S P G G F S R L N L W T K S G S T P V

H3-Numbering: Position: 181...186...191...196...201...206...211...216...221...226...231...236...
H1-Numbering: ...165...170...175...180...185...190...195...200...205...210...215...220...225...
Sequence: N V T M P N N D N F U K L Y I N G W H H H P S T N E C T L Y Q A S G R V T V S T R S Q Q T I I P N I G S R P W

H3-Numbering: Position: 241...246...251...256...261...266...271...276...281...286...291...296...
H1-Numbering: ...205...210...215...220...225...230...235...240...245...250...255...260...265...270...275...280...285...
Sequence: G L S R R I S Y W T I V K P G D V L V I N S N G N L I A P R G Y K M R T G K S I M B S D A P I D T C I S E C T P

Position: 301..306..311..316..321..326..331..336..341..346..351..356..

Donor Sequence: A/Nicaragua/1815_01/TR2/2013 (H1)

Selected donor sequence

H3-Numbering: Position: 1...6...11...16...21...26...31...36...41...46...51...56...
H1-Numbering: ...1...5...10...15...20...25...30...35...40...45...
Sequence: R A T K Y M A D V I L V L Y P A T A N A D T C I G H A N N S T D T V D L E R N V T T H S V N L E N S H N G K C L L R

H3-Numbering: Position: 61...66...71...76...81...86...91...96...101...106...111...116...
H1-Numbering: ...10...15...20...25...30...35...40...45...50...
Sequence: S S T G K T C H I R I L L D C T I D A L L G D P H C D V F Q N T D W I L V E R S K A F S N C Y Y D P V

H3-Numbering: Position: 121...126...131...136...141...146...151...156...161...166...171...176...
H1-Numbering: ...105...110...115...120...125...130...135...140...145...150...155...160...
Sequence: Y A S L R S I V A S S G T L E F I T Q N G S S A C K S P G G F S R L N L W T K S G S T P V

H3-Numbering: Position: 181...186...186...191...196...201...206...211...216...221...226...231...236...
H1-Numbering: ...170...175...180...185...190...195...200...205...210...215...220...225...
Sequence: N H N K E K V E L P D T I I F E R A N G N L I A P R A F A L S R G F G S G J I N S N A P M D C D A K C Q T P Q G A I

Position: 301..306..311..316..321..326..331..336..341..346..351..356..

Current Base sequence:

A/Wisconsin/6/2005 (H3)

Base sequence name

Base sequence

Replaced region on base sequence: Donor region on donor sequence: Add
0 □ to 0 □ 0 □ to 0 □ 0 □ to 0 □ Clear

Region on base sequence to be replaced Region on donor sequence to be inserted

Current product

Legend: H1 numbering Cal A Cal B Ca2 C Cb D Sa E Sb F Stalk-MN G Stalk-MN H

Position AA: . . . 5 . . . 10 . . . 15 . . . 20 . . . 25 . . . 30 . . . 35 . . . 40 . . . 45 . . . 50 . . . 55 . . . 60 . . . 65 . . . 70 . . . 75 . . . 80 . . . 85 . . . 90

H1 numbering . . . 5 . . . 10 . . . 15 . . . 20 . . . 25 . . . 30 . . . 35 . . . 40 . . . 45 . . . 50 . . . 55 . . . 60 . . . 65 . . . 70 . . . 75 . . . 80 . . . 85 . . . 90

H3 numbering . . . 5 . . . 10 . . . 15 . . . 20 . . . 25 . . . 30 . . . 35 . . . 40 . . . 45 . . . 50 . . . 55 . . . 60 . . . 65 . . . 70 . . . 75 . . . 80 . . . 85 . . . 90

Original Seq: M K T I I A L S Y I F C L A L G Q D L P G N D N S T A T L C L G H H A V P N G T I V K T I T D D Q I E V T N A T E L V Q

M K T I I A L S Y I F C L A L G Q D L P G N D N S T A T L C L G H H A V P N G T I V K T I T D D Q I E V T N A T E L V Q

TVVLLYTFATAN ANNSTDITVDTV

Double click to delete

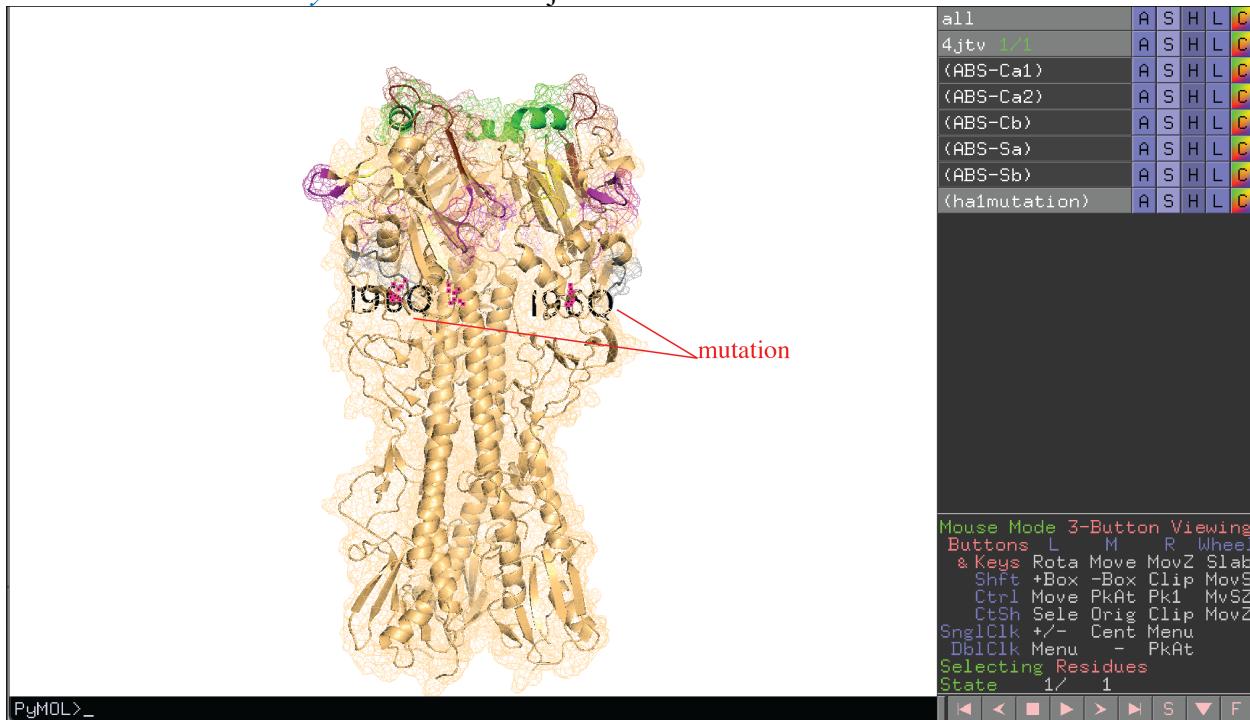
Insertion information

Confirm Cancel

3D visualization via PyMOL

Users can see 3D structure of selected sequence via PyMOL (only for HA for now).

Click *Visualization-> PyMOL* in menu or just click  button.



Generate Standardized Gibson Clone fragments for HA/NA in batches

Users can generate standardized Gibson Clone Fragments for HA/NA sequences in batches. Those fragments are reusable due to they are following the same design.

Click *GibsonClone-> GibsonClone* in *menu* or just click  button on Main tab.

Note: *GibsonClone* only works for sequences within same subtype!

Users can select sequences, determine fragment DB, output path, and joint region plan (panel A), after click “Generate Fragments” button, a dialog will pop up for users to review and confirm (panel B). On panel B, users can review the generated Fragments (both amino acid sequences and nucleotide sequences). Joint region that connect fragment 1 and 4 to the vector not displayed for AA sequences but will be automatically added to NT sequences. ‘-’ in AA alignments indicate incomplete sequences or deletions and will be deleted in NT fragments. Users can click “confirm” button to generate fragments after they confirmed current fragments. After a few second, a notice will be popped up with summarized results (panel C).

Welcome to Gibson Clone Fragment Design page!

Select All

	Name	Subtype
1	A/South Carolina/1/1918 (H1)	H1
2	A/PR/8/1934 (H1)	H1
3	A/Fort Monmouth/1/1947 (H1)	H1
4	A/Denver/1/1957 (H1)	H1
5	A/USSR/1977 (H1)	H1
6	A/Texas/36/1991 (H1)	H1
7	A/Solomon Islands/3/2006 (H1)	H1
8	A/Brisbane/59/2007 (H1)	H1

Sequence candidates

Vector Connector Sequences

Joint region for upstream end (Gibson cloning into the vector):

Joint region for 3' end (instead of transmembrane region): **joint region to vector**

Fragments Database:

Server IP: localhost Port: 3306 DB name: Librator
User Name: root Password: 123456

Gibson clone fragments files output path:

Subtype: H1/Group1 H3/Group2 NA

Joint Region Design: Default User Defined

Fragment 1: 1 to 131 Fragment 2: 123 to 272
Fragment 3: 264 to 411 Fragment 4: 403 to 518

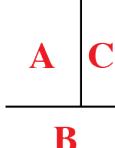
Gibson Clone Fragments Preview

Options: AA NT

Position AA:	Sequence
A/England/80740425/2018	. . . 90 . . . 95 . . . 100 . . . 105 . . . 110 . . . 115 . . . 120 . . . 125 . . . 130 . . . 135 . . . 140 . . . 145 . . . 150 . . . 155 . . . 160 . . . 165 . . .
A/Auckland/610/2002	GDPQCDGFQNKKWDLFVERSRAYSNCYPYDVPDYASLRSVLVASS
A/Memphis/2/1986	GDPHCDGFQNKEWDLFVERSKAFSNCPYDVPDYASLRSVLVASS
A/New York/680/1995	GDPHCDGFQNKEWDLFVERSKAYSNCYPYDVPDYASLRSVLVASS
A/England/80740425/2018	SLRSLVASSGTLERFKNESFNWAGVTQNGKSSACIRGSSSSFFSRLNW
A/Auckland/610/2002	SLRSLVASSGTLERFNNEFSNWTGVAQNGTSSACKRRSDKSFFSRLNW
A/Memphis/2/1986	SLRSLVASSGTLERFINEGFNWTGTVQSGGSYACKRGSVNSFFSRLNW
A/New York/680/1995	SLRSLVASSGTLERFTNENFNWTGVAQDGKSYACKRGSVNSFFSRLNW
A/England/80740425/2018	joint region shared by fragments
A/Auckland/610/2002	
A/Memphis/2/1986	
A/New York/680/1995	

Alignment of Fragments

Selected sequences



Output path

The files were generated under: /Users/eil/Documents/Projects/Librator/Resources/Tmp

Existing fragments used:
H3-F1-0001
H3-F4-0002
H3-F4-0003

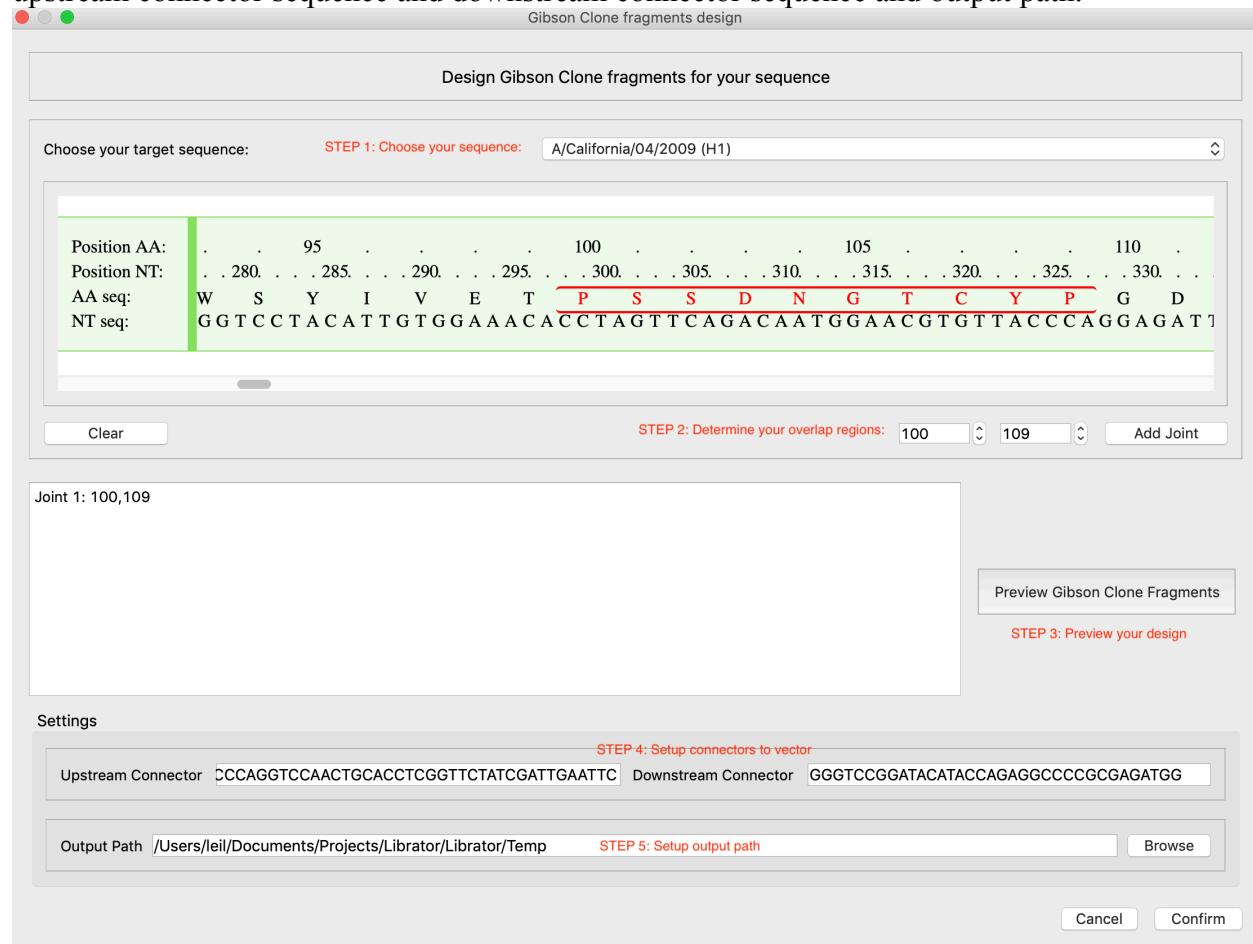
Existing fragments

New fragments generated:
H3-F1-0003
H3-F1-0005
H3-F1-0011
H3-F1-0012
H3-F1-0013
H3-F1-0014
H3-F1-0015
H3-F2-0007
H3-F2-0008
H3-F2-0014
H3-F2-0015
H3-F2-0016
H3-F2-0017
H3-F2-0018
H3-F3-0002
H3-F3-0003
H3-F3-0009
H3-F3-0010
H3-F3-0011
H3-F3-0012
H3-F3-0013
H3-F4-0008
H3-F4-0009
H3-F4-0010

New fragments need to order

Generate Gibson Clone fragments for any sequence

Beside HA/NA sequences, Librator also allows users to split any of their sequences into fragments. Users can use this function for some frontier and novel research, e.g. COVID-19. Users can click *GibsonClone-> GibsonClone for single sequence* in *menu*. Users can view their sequence and add at most 12 joint regions (where to split the sequence) to their sequence. Then users can preview the fragment products under current design. After that, users can setup upstream connector sequence and downstream connector sequence and output path.



Contact

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