HIV Database Workshop

www.hiv.lanl.gov

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Theoretical Biology and Biophysics, T-6
Los Alamos National Laboratory

https://tinyurl.com/HIVDB-IEDB-2021

Workshop Topics

Introduction to HIV Sequence Database

HIV in comparison to other viruses

Exploration of HIV evolution, using one Gag epitope as an example.

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Entry page at https://www.hiv.lanl.gov/

HIV DATABASES

The **HIV databases** contain comprehensive data on HIV genetic sequences and immunological epitopes. The website also gives access to a large number of tools that can be used to analyze and visualize these data. This project has been funded in whole or in part with Federal funds from the National Institute of Allergy and Infectious Diseases, National Institutes of Health, Department of Health and Human Services, under Interagency Agreement No. AAI12007-001-00000. Our content is reviewed by an Editorial Board.

SEQUENCE DATABASE ▸ IMMUNOLOGY DATABASE ▸

OTHER VIRUSES ▸

News: Archived News ▸

Variant Visualizer

Variant Visualizer is a new tool that provides new options and output styles for visualizing variants in an alignment of nucleotide or protein sequences. The tool is similar to Highlighter, but the Variant Visualizer has more options for refining the graphical output. The output can be refined interactively without rerunning the tool. *21 July 2021*

Questions or comments? Contact us at seq-info@lanl.gov

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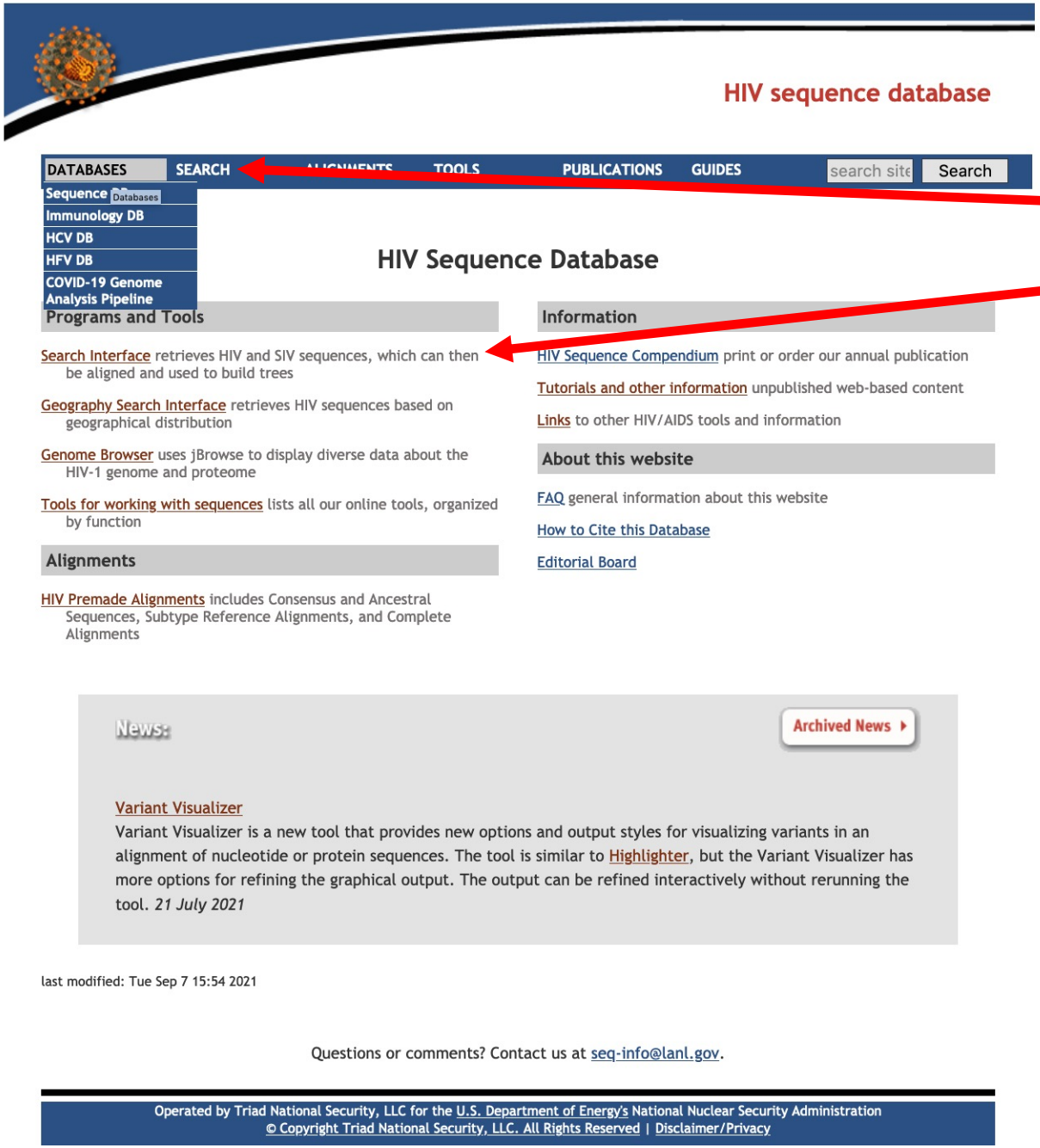

Multiple paths to most tools

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HIV sequence database

DATABASES SEARCH ALIGNMENTS TOOLS PUBLICATIONS GUIDES search site Search

- Tutorials
- CRFs
- HIV-1 Gene Map
- In-depth Annotation
- Neutralizing Antibody Resources & CATNAP
- 3D Structure
- Data Dictionary
- How to Cite this Database
- HIV Database News
- FAQs
- Links

Tutorials and Basic Informatio

Tutorials

HIV Database presentations from conference workshops

FAQs about the HIV Sequence Database

FAQs about the HIV Immunology Database

Sequence quality control explains several common problems with sets of viral sequences

How to make a phylogenetic tree explains how to build a phylogenetic tree

HIV numbering relative to reference strain HXB2

SIV numbering relative to reference strain SIVmm239

Articles

3D views of HIV macromolecular structures provides links to 3D views of HIV proteins

Stalking the AIDS Virus [PDF] article from LANL Research Quarterly (Fall 2003) about HIV Database research on the HIV-immune system interaction as a step toward an AIDS vaccine

Los Alamos research fundamental to first efficacy study for mosaic HIV-1 preventive vaccine 2017 LANL news release about the HIV mosaic vaccine

Reference Inform

Circulating recombinan… etails about all documented CRFs

HIV-1 gene map illustr… s of HIV-1, including HXB2 breakpoints

HXB2 annotated spreadsheet (.xls) provides a fully-annotated sequence of HXB2 with base-by-base detail

HIV and SIV subtype nomenclature gives an overview of HIV and SIV subtype nomenclature, particularly HIV-1 groups and subtypes

Primate immunodeficiency virus nomenclature lists SIV species and nomenclature

How the HIV database classifies sequences explains how recombinants are named and annotated

Common sequence formats for alignments shows examples of common sequence formats for alignments

How to cite this Database explains how to cite this website and the printed HIV compendia

Codes and symbols in sequences decodes the symbols and IUPAC codes that appear in sequences and alignments

Codon table gives the translation of nucleotides into amino acids

Links HIV/AIDS resources and bioinformatics tools on other websites

last modified: Wed Feb 28 14:38 2018

Questions or comments? Contact us at seq-info@lanl.gov.

All kinds of basic information about HIV and about our database

Previous workshop presentations

Yes! We do respond to this e-mail address!

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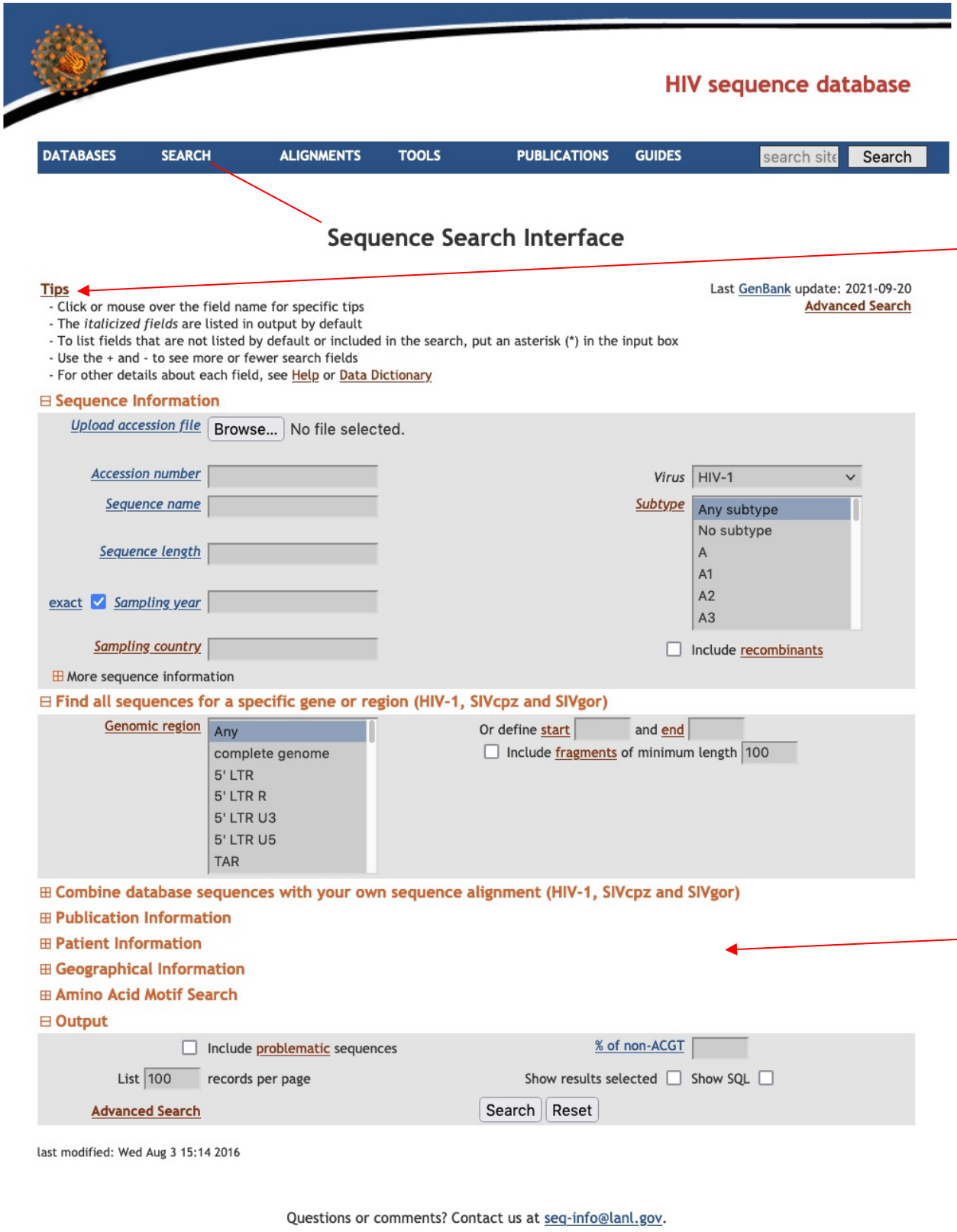

Almost all of our pages, tools, etc. have help files explaining the uses and providing help.

Sample data sets to test each tool are also provided.

Each of these sections of the search interface are expandable to provide additional search capabilities.

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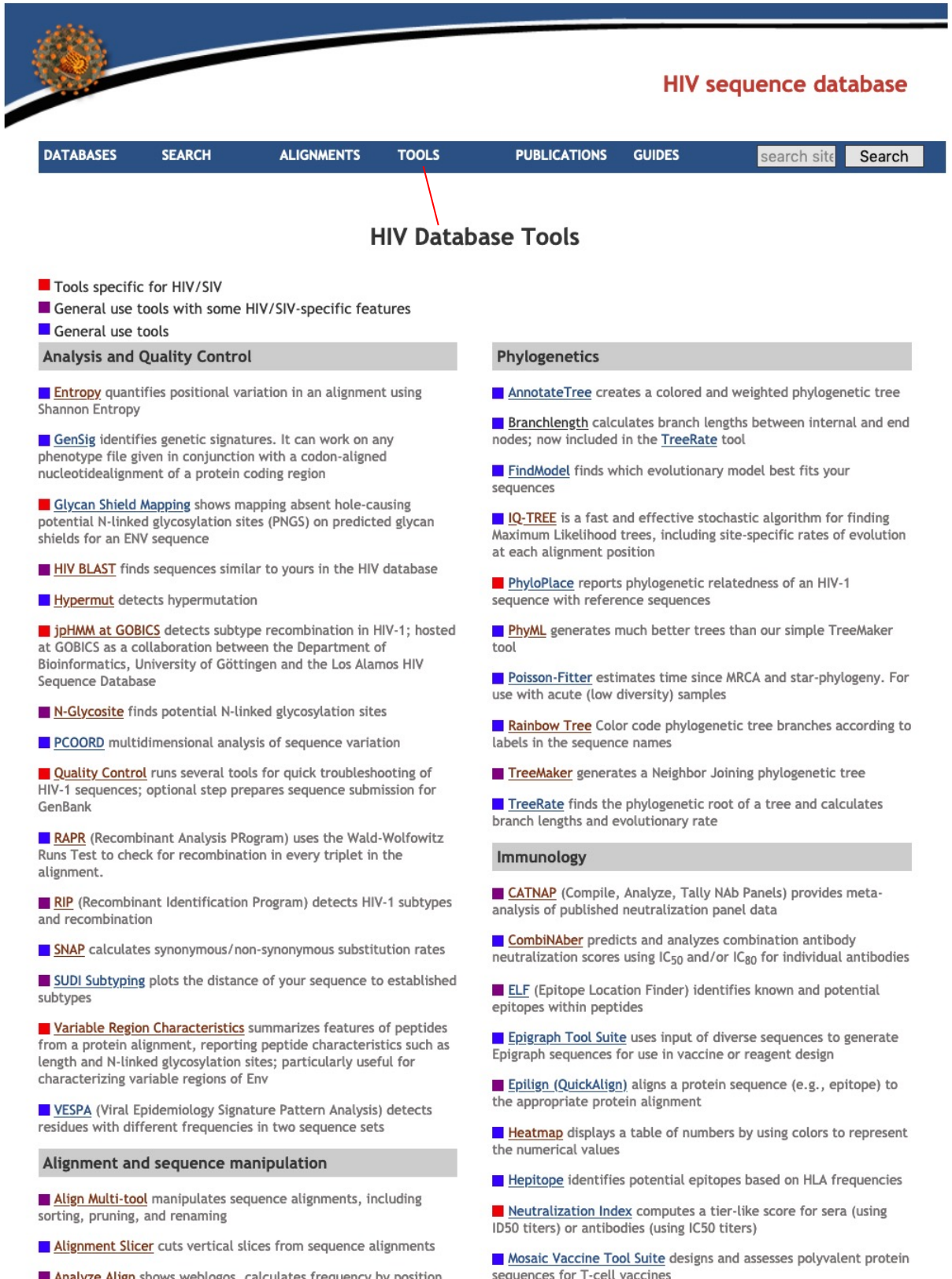

Some of our tools are specific to HIV or lentiviruses, most can be used on DNA or Protein sequences from any organism.

Color-coded square indicates whether a given tool is specific to HIV or not.

Tools are listed alphabetically and organized into general categories.

Write to seq-info@lanl.gov for additional help, or to report problems or suggest new tools or features.

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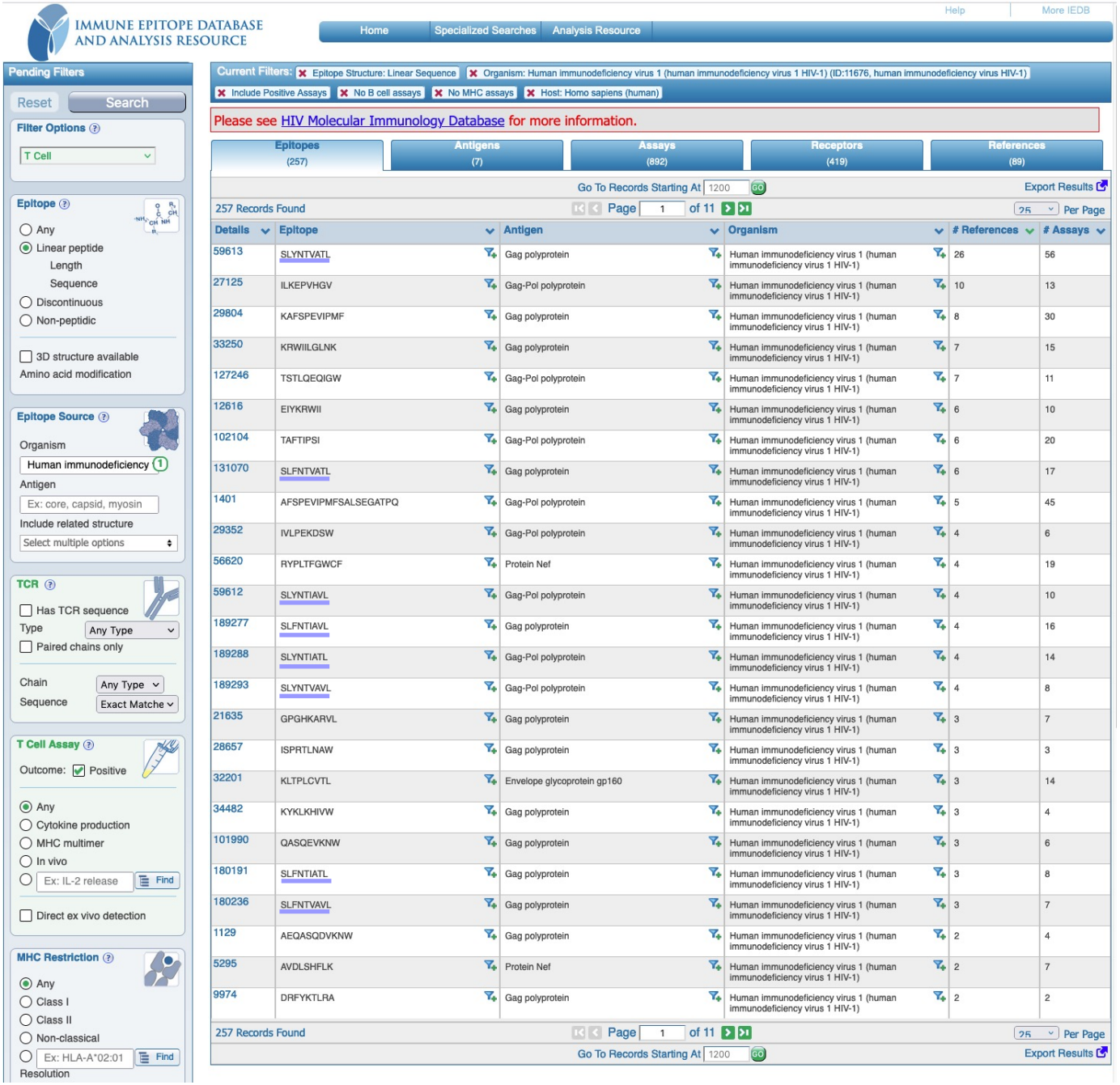
All viruses evolve, but each has unique influences on its epidemiology and evolution

HIV, HCV and some other viruses persist in one host individual long after immune response is mature. Can be passed to recipient individual after significant evolution/selection in donor individual.

Influenza viruses, coronaviruses, and many other viruses typically infect a naïve host individual and are rapidly passed on to next recipient individual before much host immune selection pressure has taken place.

HIV has been infecting humans for 100 years or more, and has relatively slowly spread around the world. HCV has been infecting humans for more than 20,000 years and likewise has spread quite slowly.

Influenza viruses and coronaviruses can spread around the world in a matter of weeks.



Searched the IEDB for CTL epitopes in HIV-1

Gag SLYNTVATL SLFNTVATL etc are found

How variable is this epitope in HIV-1?

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HIV Sequence Database Genome Browser

View genome or each protein to see features of interest

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HIV sequence database

DATABASES SEARCH ALIGNMENTS TOOLS PUBLICATIONS GUIDES search site Search

HIV Genome Browser

Purpose: Interactive view of the HIV genome and proteome for juxtaposition and exploration of multiple types of data. Help.

Starting Views

NOTE: These are just starting points! Within the genome browser, you can move among any of these views. Please read the quick tips and Help file before you start!

HIV-1 protein-level views:

- Env
- Gag
- Nef
- Pol
- Rev
- Tat
- Vif
- Vpr
- Vpu

HIV-1 proteins, specific examples:

- Env with CTL epitopes + entropy
- Pol with drug resistance sites + entropy

Nucleotide-level views:

- HIV-1 gene map
- SIV Mac239 gene map
- HIV-1 5' LTR

Quick Tips

- Mouseovers! Look for mouseovers to guide you.
- Click and right-click! Features link to loads of information and analysis via click and right-click. If your mouse doesn't have right-click, use Ctrl-click.
- Zoom! There are several ways to zoom in/out. Some features can only be seen when zoomed-in or zoomed-out.
- For details about this interface, see HIV Genome Browser Help.
- Watch the screencast video on the JBrowse website.

References

- Skinner ME, Holmes IH. Setting up the JBrowse genome browser. Curr Protoc Bioinformatics. 2010 Dec;Chapter 9:Unit 9.13. PMID: 21154710

HIV Genome Browser

Select Gag to see Gag protein view

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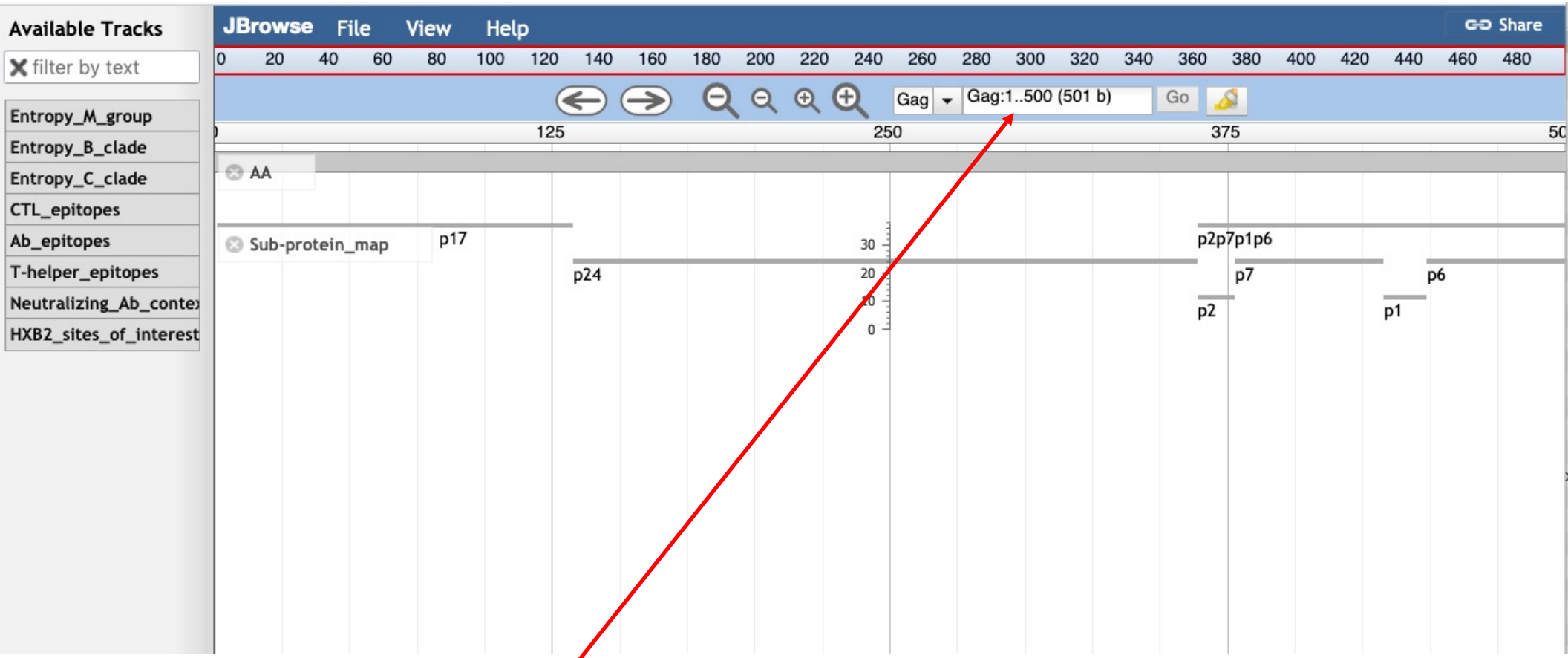

Enter SLYNTVATL to zoom in to that site in the Gag protein.

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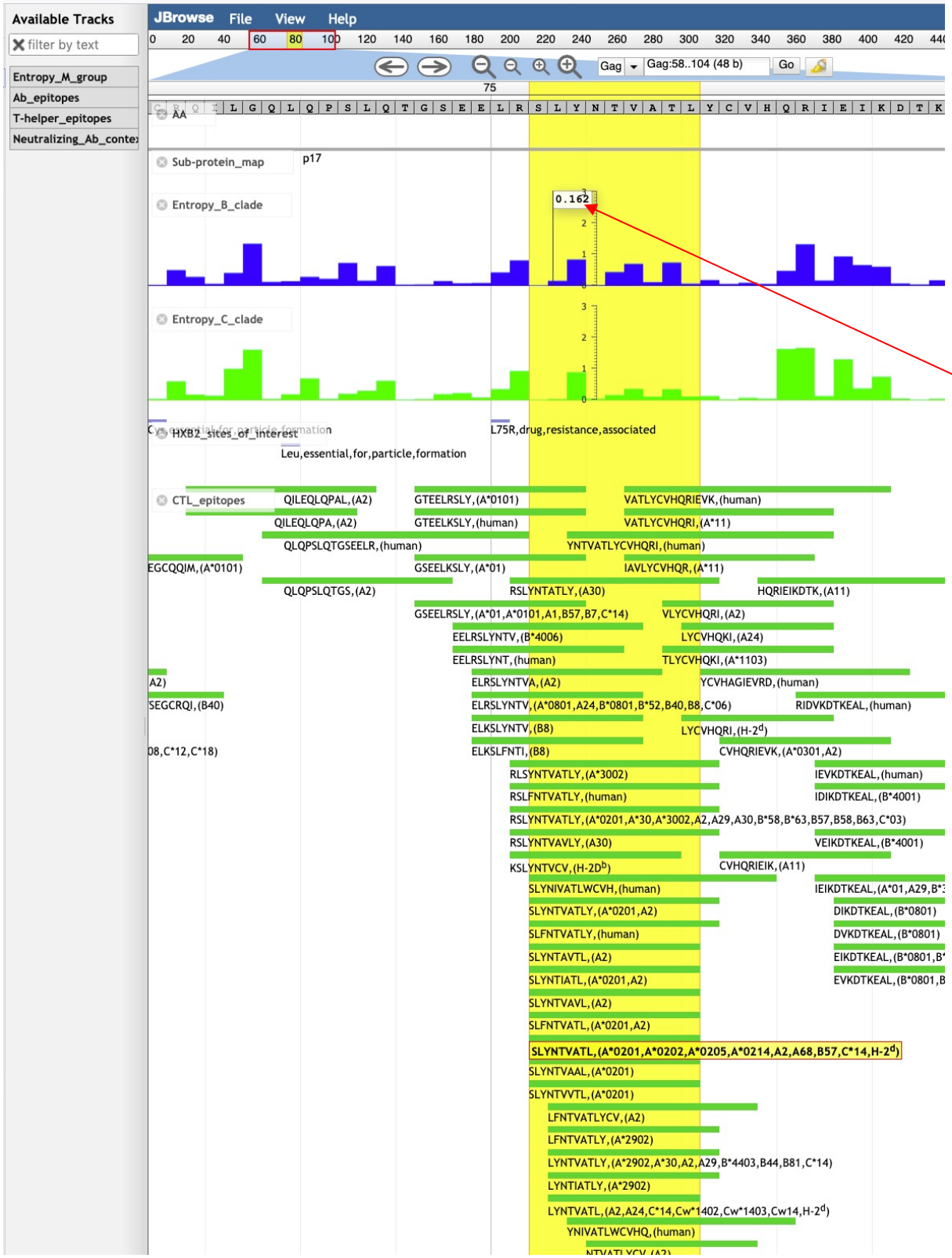

HIV Genome Browser has tracks for entropy of each site

SLYNTVATL third amino acid, Y, has high entropy

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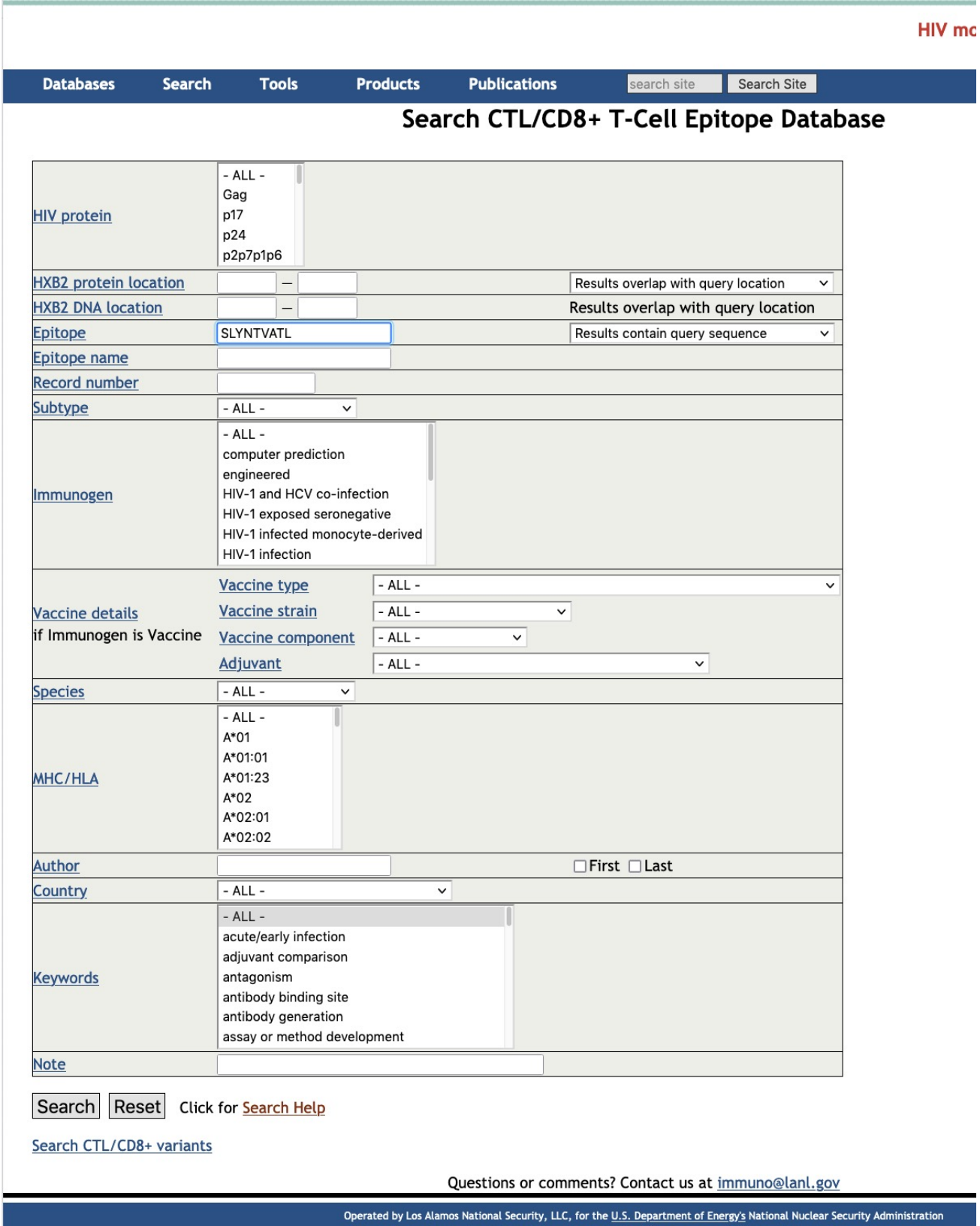

HIV Immunology Database
CTL Search for SLYNTVATL

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HIV molecular immunology database

Databases | Search | Tools | Products | Publications | search site | Search Site

Search CTL/CD8+ T-Cell Epitope Database

Found 301 matching records:

Displaying record number 57

Download this epitope record as JSON.

HXB2 Location	Gag(69-93) p17(69-93) DNA(994..1068)	Gag Epitope Map
Author Location	p17(69-93 BH10)	
Epitope	QTGSEELRSLYNTVATLYCVHQRIE	Epitope Alignment
Species (MHC/HLA)	human(A2)	
Immunogen	HIV-1 infection	
Experimental methods		
Keywords		

Notes

- Gag CTL response studied in three individuals.

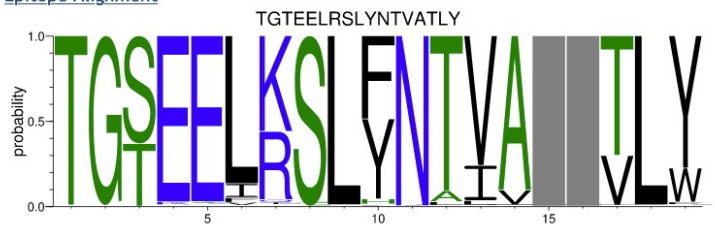
References

Johnson1991 R. P. Johnson, A. Trocha, L. Yang, G. P. Mazzara, D. L. Panicali, T. M. Buchanan, and B. D. Walker. HIV-1 Gag-Specific Cytotoxic T Lymphocytes Recognize Multiple Highly Conserved Epitopes. Fine Specificity of the Gag-Specific Response Defined by Using Unstimulated Peripheral Blood Mononuclear Cells and Cloned Effector Cells. *J. Immunol.*, 147:1512-1521, 1991. This study presented a detailed study of gag-specific CTL from HIV-1 seropositive individuals. Seven p24 and two p17 epitopes were described, that were recognized by class I-restricted CD3+CD8+ CTL. p17 epitopes: KIRLRPGGKKKYKLKHIVWASRELE and QTGSEELRSLYNTVATLYCVHQRIE; p24 epitopes: NPPIPVGEIYKRWIILGLNKIV, VHQAISPRTLNAWVKVVEEKAF, NAWVKVVEEKAFSPEVIPMFSA, SALSEGATPQDLNTMLNTVGGH, GHQAAMQMLKETINEEAAEWDR, and RAEQASQEVK. PubMed ID: 1715361. Show all entries for this paper.

Displaying record number 58923

Download this epitope record as JSON.

HXB2 Location	Gag(70-86) p17(70-86) DNA(997..1047)	Gag Epitope Map
Author Location	Gag(70-)	
Epitope	TGTEELRSLYNTVATLY	Epitope Alignment
Subtype	B, C	
Species (MHC/HLA)	human(C*14)	



301 Records found for SLYNTVATL in CTL epitopes

Each has a pre-done link to an analysis of diversity in sequence database alignment of one sequence per patient.

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HIV sequence database

DATABASES SEARCH ALIGNMENTS TOOLS PUBLICATIONS GUIDES search site Search

AnalyzeAlign

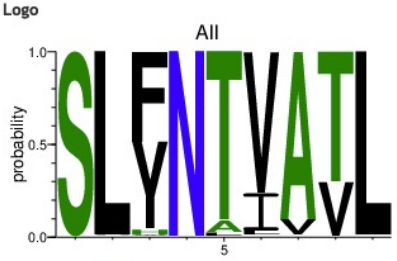
Input options

Alignment: imported from QuickAlign (6144 amino acid sequence(s))
Group the sequences: report results for all sequences as a single group
Logo stack width: 22
Stacks per line: 25
Units: probability
Show x-axis: yes
Show y-axis: yes
Color scheme: hydrophobicity
Draw stacks with largest letters on top: yes
Remove symbols from logo: show all
Mark potential N-linked glycosylation sites: no
Delete gaps and shift alignment to C-terminus: no
Cut-off for calculating frequency by position: 95%
Master sequence for finding variants: SLYNSVATL

Warning: Your sequences had to be padded - please make sure all sequences are of the same length.

Download all results: ZIP

Logo



Download: PNG

Frequency by position

See full raw counts cutoff: 95%

	Percentage and raw count of non-gap	Non-gap/total (percentage)	Gap/total (percentage)
1	S: 99.87% (6136) other: 0.13% (8)	6144/6144 (100.00%)	0/6144 (0.00%)
2	L: 99.09% (6088) other: 0.91% (56)	6144/6144 (100.00%)	0/6144 (0.00%)
3	F: 48.76% (2996) Y: 46.88% (2880) other: 4.36% (268)	6144/6144 (100.00%)	0/6144 (0.00%)
4	N: 99.71% (6126) other: 0.29% (18)	6144/6144 (100.00%)	0/6144 (0.00%)
5	T: 90.89% (5584) A: 6.46% (397) other: 2.65% (163)	6144/6144 (100.00%)	0/6144 (0.00%)
6	V: 76.11% (4676) I: 20.51% (1260) other: 3.39% (208)	6144/6144 (100.00%)	0/6144 (0.00%)
7	A: 90.53% (5562) V: 8.61% (529) other: 0.86% (53)	6144/6144 (100.00%)	0/6144 (0.00%)
8	T: 70.17% (4311) V: 29.30% (1800) other: 0.54% (33)	6144/6144 (100.00%)	0/6144 (0.00%)
9	L: 99.06% (6086) other: 0.94% (58)	6144/6144 (100.00%)	0/6144 (0.00%)

Sequence variants

HIV QuickAlign or AnalyzeAlign tools show that the third site, Y, is often F

Is this due to some clades of virus having Y, others F, or is it F/Y within each clade?

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HIV sequence database

DATABASES SEARCH ALIGNMENTS TOOLS PUBLICATIONS GUIDES search site Search

AnalyzeAlign

Input options

Alignment: imported from QuickAlign (4661 amino acid sequence(s))
Major subtypes only
Group the sequences: separately report results for subsets grouped by user's grouping choice
Logo stack width: 22
Stacks per line: 25
Units: probability
Show x-axis: yes
Show y-axis: yes
Color scheme: hydrophobicity
Draw stacks with largest letters on top: yes
Remove symbols from logo: show all
Mark potential N-linked glycosylation sites: no
Delete gaps and shift alignment to C-terminus: no
Cut-off for calculating frequency by position: 95%
Master sequence for finding variants: SLYNSVATL

Warning: Your sequences had to be padded - please make sure all sequences are of the same length.

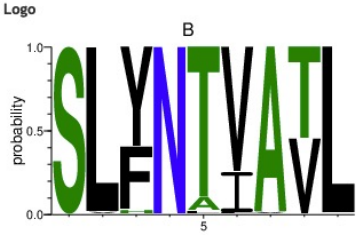
Download all results: ZIP

Groups

B A1 A2 C D F1 F2 G 01_AE 02_AG

Group B Go to top

Logo



Download: PNG

Frequency by position

See full raw counts cutoff: 95%

	Percentage and raw count of non-gap	Non-gap/total (percentage)	Gap/total (percentage)
1	S: 99.84% (1874) other: 0.16% (3)	1877/1877 (100.00%)	0/1877 (0.00%)
2	L: 98.03% (1840) other: 1.97% (37)	1877/1877 (100.00%)	0/1877 (0.00%)
3	Y: 59.51% (1117) F: 37.51% (704) other: 2.98% (56)	1877/1877 (100.00%)	0/1877 (0.00%)
4	N: 99.57% (1869) other: 0.43% (8)	1877/1877 (100.00%)	0/1877 (0.00%)
5	T: 89.50% (1680) A: 8.26% (155) other: 2.24% (42)	1877/1877 (100.00%)	0/1877 (0.00%)
6	V: 74.43% (1397) I: 24.29% (456) other: 1.28% (24)	1877/1877 (100.00%)	0/1877 (0.00%)
7	A: 98.40% (1847) other: 1.60% (30)	1877/1877 (100.00%)	0/1877 (0.00%)
8	T: 54.77% (1028) V: 44.91% (843) other: 0.32% (6)	1877/1877 (100.00%)	0/1877 (0.00%)
9	L: 99.09% (1860) other: 0.91% (17)	1877/1877 (100.00%)	0/1877 (0.00%)

Viewing each subtype separately, we see that the Y vs F at the third site is common in all clades or subtypes of HIV-1

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Exploring that region of the HIV-1 sequence alignment, we see it is a TAC = Y vs TTC = F transversion in the genome.
Nearby, ACC = T vs GTC = V is not co-varying with the Y vs F.

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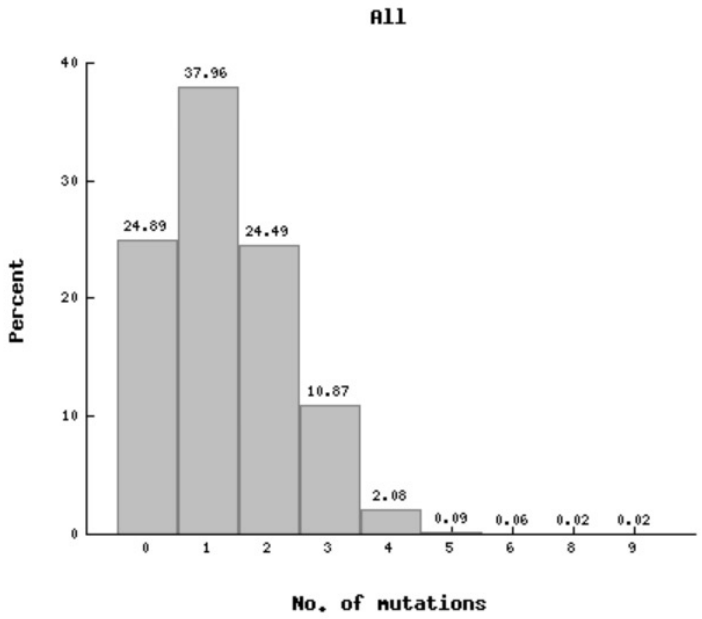
Sequence variants



Variant	Count	Pct.	No. of mutations
SLYNTVATL			
---------	1529	24.89	0
--F------	1383	22.51	1
-------V-	396	6.45	1
--F----V-	389	6.33	2
--F--I---	330	5.37	2
-----I-V-	219	3.56	2
-----I---	181	2.95	1
--F--I-V-	178	2.9	3
--F---V--	108	1.76	2
------V--	107	1.74	1
--F-A--V-	100	1.63	3
--F--IV--	85	1.38	3
--H------	81	1.32	1
----A--V-	56	0.91	2
--F-A----	56	0.91	2

Overall, 24.89% of HIV-1 have SLYNTVATL

22.51% are SLFNTVATL
6.45% are SLYNTVAVL
6.33% are SLFNTVAVL

Years ago, we noted that many sites “toggled” back and forth between two amino acids, and that the top few most common forms of each epitope thus made up the majority.

This was the birth of our ”Mosaic Vaccine Design” which has been proven useful not only for HIV vaccines but also for vaccines against several other viruses.

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The HIV-1 HLA-A2-**SLYNTVATL** is a help-independent CTL epitope.

Kan-Mitchell J, Bisikirska B, Wong-Staal F, Schaubert KL, Bajcz M, Bereta M.

J Immunol. 2004 May 1;172(9):5249-61. doi: 10.4049/jimmunol.172.9.5249.

PMID: 15100263 **Free article.**

The CTL response to the HLA-A*0201-restricted, HIV-1 **p17 Gag**(77-85) epitope (**SLYNTVATL**; SL9) has been extensively studied in patients. ...In contrast to published reports for influenza and melanoma peptides and the HIV **gag** IV9 epitope studied here in p ...

Characterization of cross-reactive CD8+ T-cell recognition of HLA-A2-restricted HIV-**Gag** (**SLYNTVATL**) and HCV-NS5b (ALYDVVSKL) epitopes in individuals infected with **human immunodeficiency** and hepatitis C viruses.

Vali B, Tohn R, Cohen MJ, Sakhdari A, Sheth PM, Yue FY, Wong D, Kovacs C, Kaul R, Ostrowski MA.

J Virol. 2011 Jan;85(1):254-63. doi: 10.1128/JVI.01743-10. Epub 2010 Oct 27.

PMID: 20980521 **Free PMC article.**

The immunologic mechanisms underlying the faster progression of hepatitis C **virus** (HCV) disease in the presence of **human immunodeficiency virus** (HIV) coinfection are not clearly understood. ...Our search for amino acid sequence homology between the HCV ...

Persistant immunodominant anti-**gag SLYNTVATL** responses in HIV-patients with up to 7 years of HAART.

Zidovec Lepej S, Kosor E, Gagro A, Vince A, Remenar A, Poljak M.

Coll Antropol. 2006 Dec;30 Suppl 2:33-8.

PMID: 17508471

Gag SLYNTVATL-specific CD8+ T-cells were detectable in 18 of 26 treated patients (median 5.2 years of HAART) and in 10 of 14 untreated patients. Median percentage of **Gag SLYNTVATL**-specific CD8+ T-cells in treated patients was 0.10 (range 0.00-0.70%). . ...

Intrapatient escape in the A*0201-restricted epitope **SLYNTVATL** drives evolution of **human immunodeficiency virus** type 1 at the population level.

Edwards CT, Pfafferott KJ, Goulder PJ, Phillips RE, Holmes EC.

J Virol. 2005 Jul;79(14):9363-6. doi: 10.1128/JVI.79.14.9363-9366.2005.

PMID: 15994836 **Free PMC article.**

The hypothesis that the intrapatient emergence of cytotoxic T-lymphocyte escape variants contributes to the evolution of **human immunodeficiency virus** type 1 at the population (interpatient) level was tested using the HLA-A*0201-restricted **gag p17** ...

A PubMed search for “SLYNTVATL” hits 55 papers.

HIV Immunology Database at LANL has 301 records for SLYNTVATL and 14 for SLFNTVATL

How do we explore the evolution of this epitope in the HIV Sequences Database?

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HIV Sequence Database
Intrapatient Sets search for Gag-p17 region from early infection.

Does one form of the epitope (SLYNTVATL vs SLFNTVATL) tend to dominate early infections, as the “transmitted/founder” virus?

Apparently not.

In this study only one patient showed heterogeneity of the epitope early in infection, and not at either of the two “toggle sites”.

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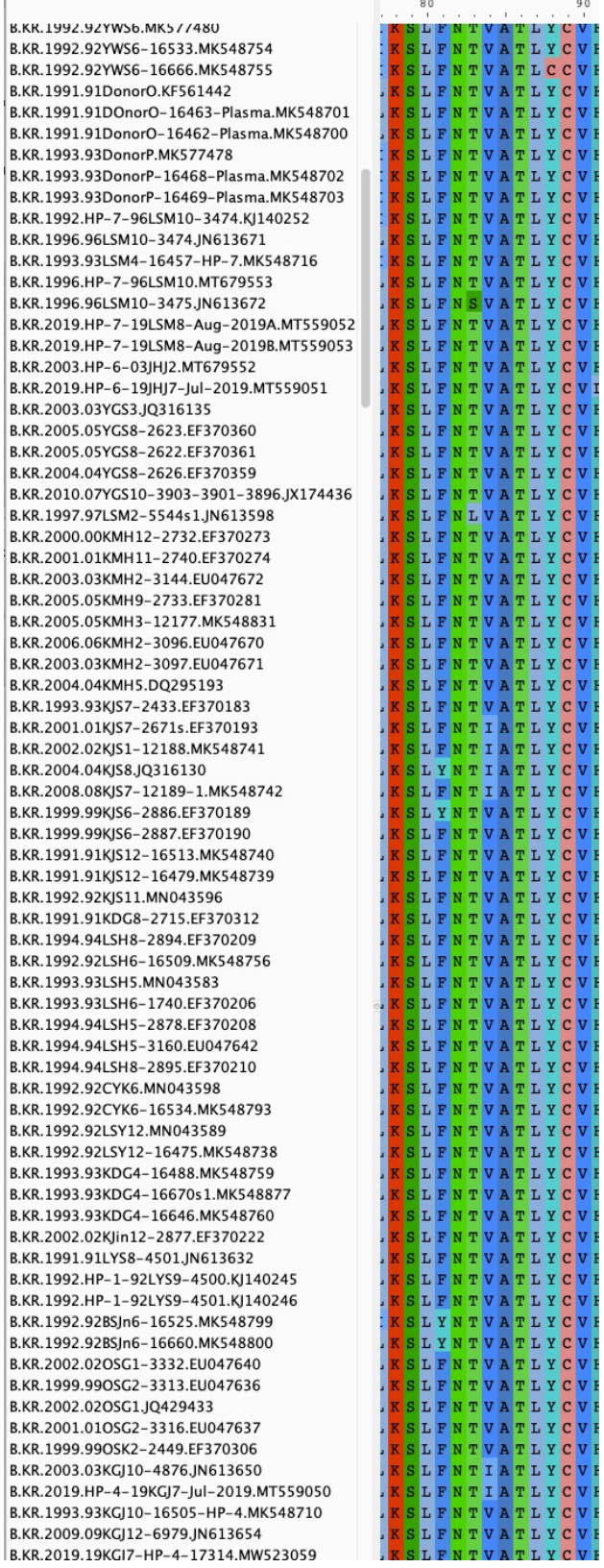

HIV Sequence Database
Korean hemophiliac cohort sampled over many years. All infected from two donors.

Does the SLYNTVATL epitope show signs of escape variation?

Apparently not.

In this study the infecting virus had SLFNTVATL and there was little change observed between 1991 donor up to 2019 recent samples.

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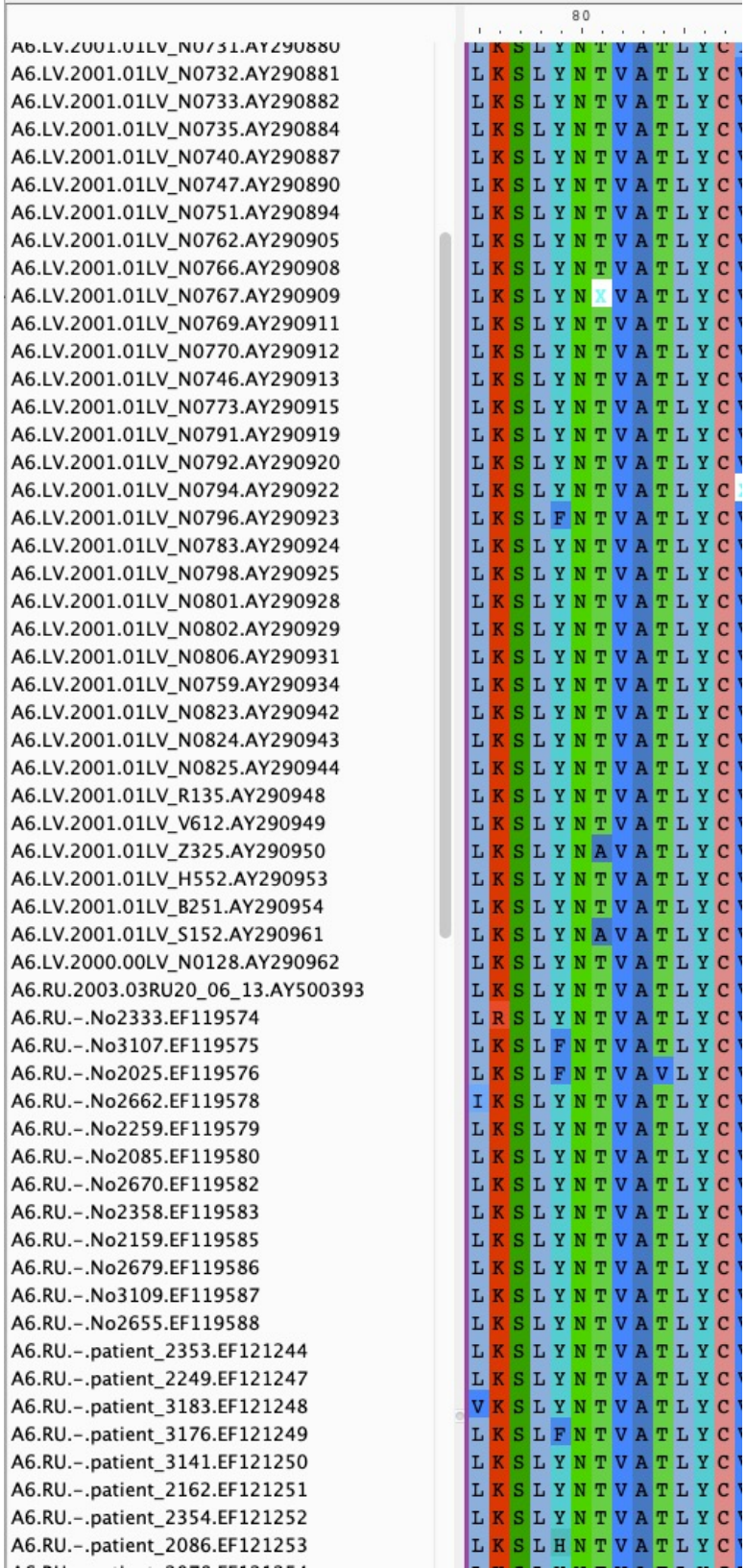

Subsubtype A6 has spread through the former Soviet Union Region, primarily via IVDU, since the mid-1990s.

Not much conversion to SLFNTVATL has taken place.

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Searching Immunology Database for MHC/HLA A*02 and proven diminished response in a Gag epitope, identifies SLFNTIATL as an escape variant.

Displaying record number 54556

HXB2 Location	Gag(77-85)	Gag Epitope Map View variants at this location
Epitope	SLYNTVATL	Epitope Alignment
Variants	SLfNTiATL	diminished response; escape documented in this paper
Epitope Name	SL9	
Species (MHC/HLA)	human(A*02)	

Variant Details

Showing 1 of 4 variants.

Variant ID.	634
Epitope Seq.	SLYNTVATL
Variant Seq.	SLfNTiATL
Mutations	Y/F V/I
Epitope Location	Y3F V6I
HXB2 Location	Y79F V82I
Mutation Type	**DR**: diminished response **E**: escape documented in this paper
Method	CD8 T-cell Elispot - IFNy
Note	Variant detected by week 63. Lower magnitude responses than wt and V/I mutant. Positive selection. Seen with upstream changes E62G/V/A, where the 62A mutation persisted and is suggested to reduce fitness cost of the Y/F escape mutation.

References

Karlsson2007 Annika C. Karlsson, Astrid K. N. Iversen, Joan M. Chapman, Tulio de Oliviera, Gerald Spotts, Andrew J. McMichael, Miles P. Davenport, Frederick M. Hecht, and Douglas F. Nixon. Sequential Broadening of CTL Responses in Early HIV-1 Infection Is Associated with Viral Escape. *PLoS ONE*, 2:e225, 2007. PubMed ID: 17311088. Show all entries for this paper.

Karlsson2007 paper
has link to sequence data
EF396480–EF396891

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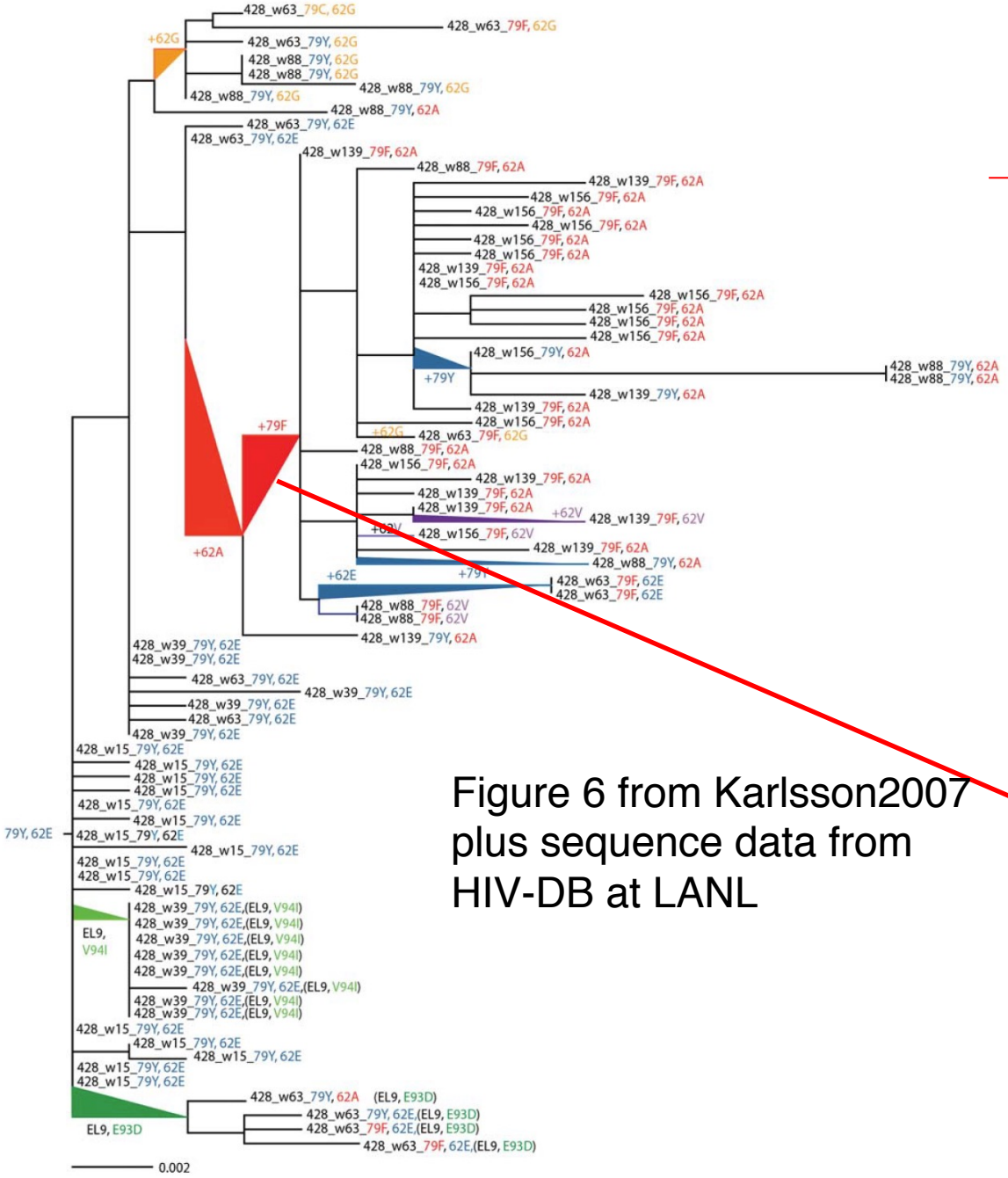
Patient OP428 with SLF/YNTIATL epitope



OP599
OP428

Figure 6 from Karlsson2007 plus sequence data from HIV-DB at LANL

Figure 6. Phylogenetic tree illustrating viral evolution of the HIV-p17 *gag* region (HXB2 coordinates 790–1431) in subject OP428 under selective pressure. Positively selected amino acid sites within the HLA-A2 ((SL9, ^{77}SLYNTIATL^{85} (patient consensus)), HLA-A1 (GY9, ^{71}GSEELRSLY^{79}), and HLA-B8 (EL9, ^{93}EVKDTKEAL^{101}) restricted epitopes. Amino acids corresponding to the consensus B sequence are shown in blue, while the Y79F mutation within SL9 and GY9 is given in red, and the E93D and V94I mutations within EL9 are shown in green on the tree. The variation found at the positively selected amino acid position 62, where a potential compensatory mutation (E62A) seems to occur prior to, and associated with, the Y79F substitution are as follows: variation at position 62: E, blue, G, orange, V purple and A red. Amino acid numbering corresponds to HXB2 Gag. Scale bars signify substitutions/site. w: corresponds to estimated week from infection (e.g. a clone named 428_w15 was obtained from a plasma sample drawn at week 15).
doi:10.1371/journal.pone.0000225.g006

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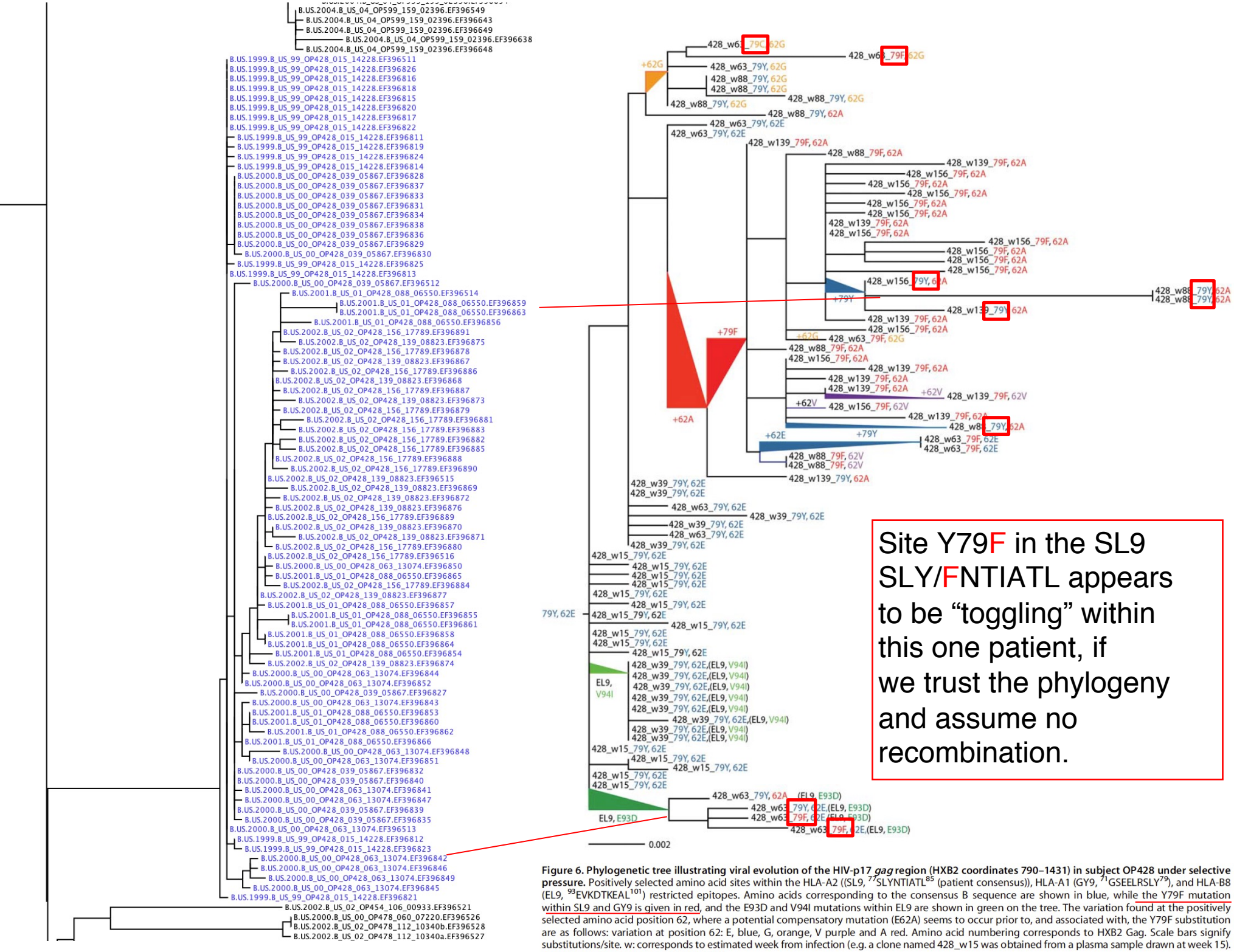
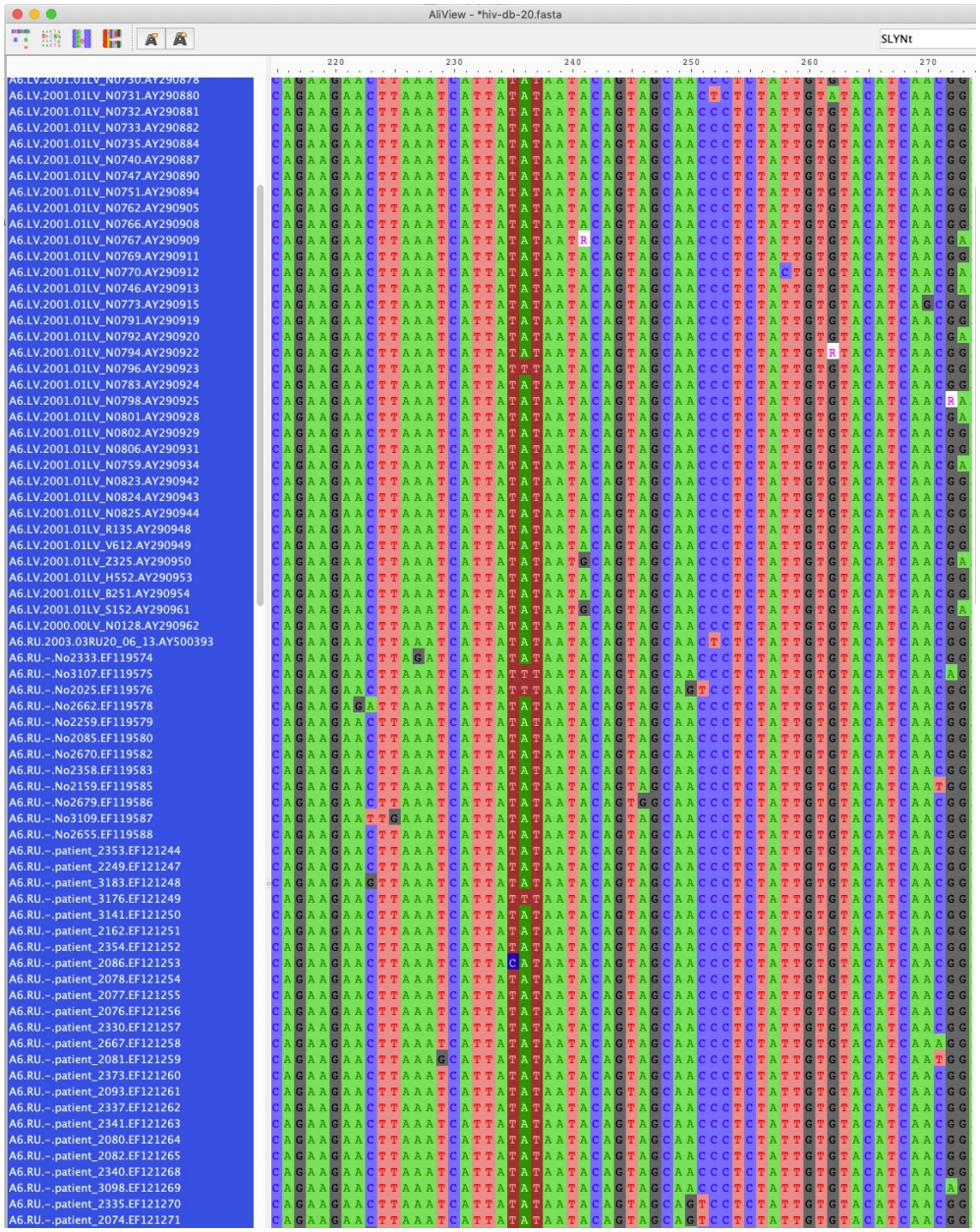


Site Y79F in the SL9 SLY/FNTIATL appears to be "toggling" within this one patient, if we trust the phylogeny and assume no recombination.

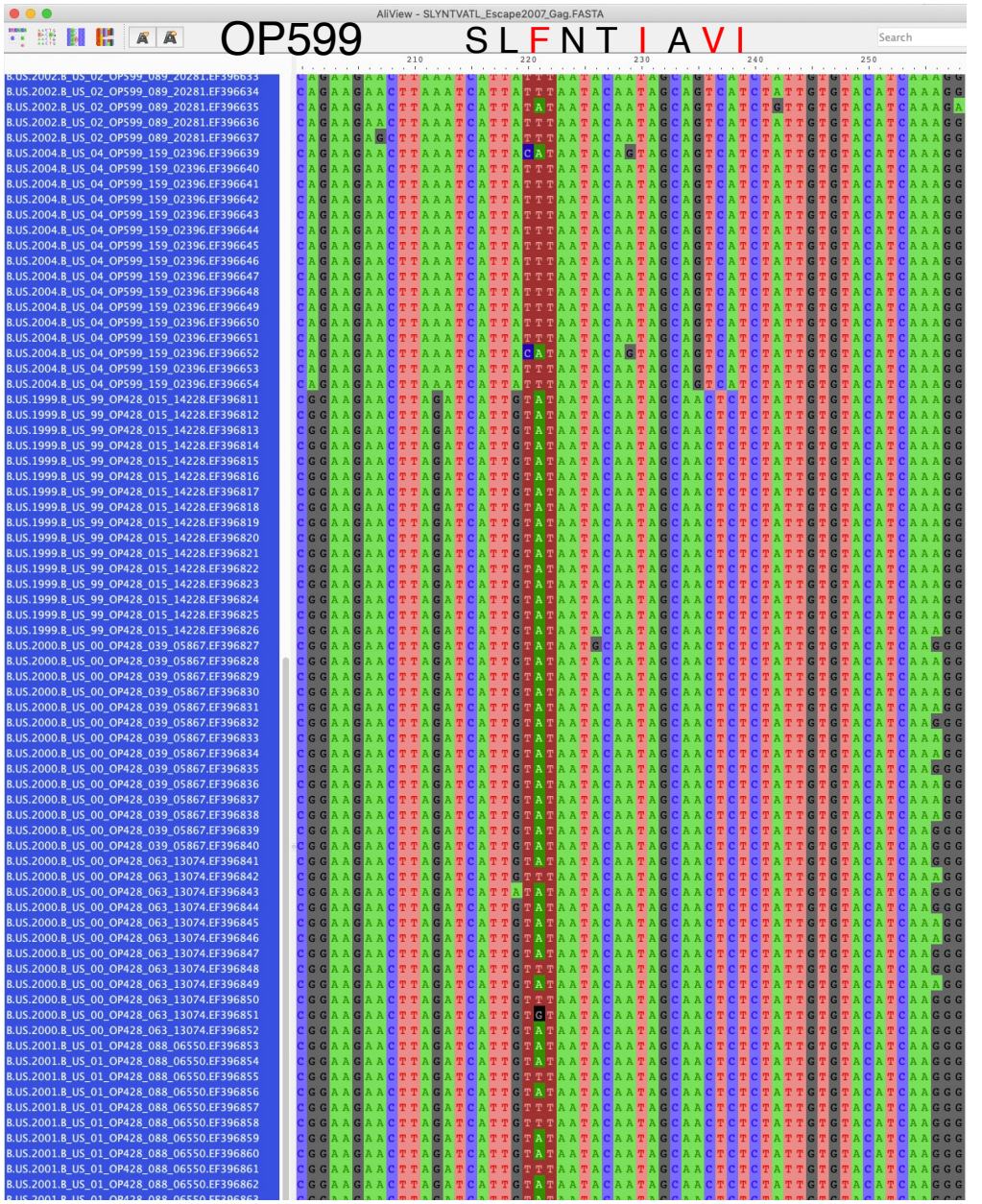
Figure 6. Phylogenetic tree illustrating viral evolution of the HIV-p17 *gag* region (HXB2 coordinates 790–1431) in subject OP428 under selective pressure. Positively selected amino acid sites within the HLA-A2 ((SL9, ^{77}SLYNTIATL^{85} (patient consensus)), HLA-A1 (GY9, ^{71}GSEELRSLY^{79}), and HLA-B8 (EL9, ^{93}EVKDTKEAL^{101}) restricted epitopes. Amino acids corresponding to the consensus B sequence are shown in blue, while the Y79F mutation within SL9 and GY9 is given in red, and the E93D and V94I mutations within EL9 are shown in green on the tree. The variation found at the positively selected amino acid position 62, where a potential compensatory mutation (E62A) seems to occur prior to, and associated with, the Y79F substitution are as follows: variation at position 62: E, blue, G, orange, V purple and A red. Amino acid numbering corresponds to HXB2 Gag. Scale bars signify substitutions/site. w: corresponds to estimated week from infection (e.g. a clone named 428_w15 was obtained from a plasma sample drawn at week 15).
doi:10.1371/journal.pone.0000225.g006

Subsubtype A6 one per patient



S L Y N T V A T L R

Subtype B two patients OP428 is HLA A02



OP599 S L F N T I A V I

OP428 S L Y N T I A T L R
F G

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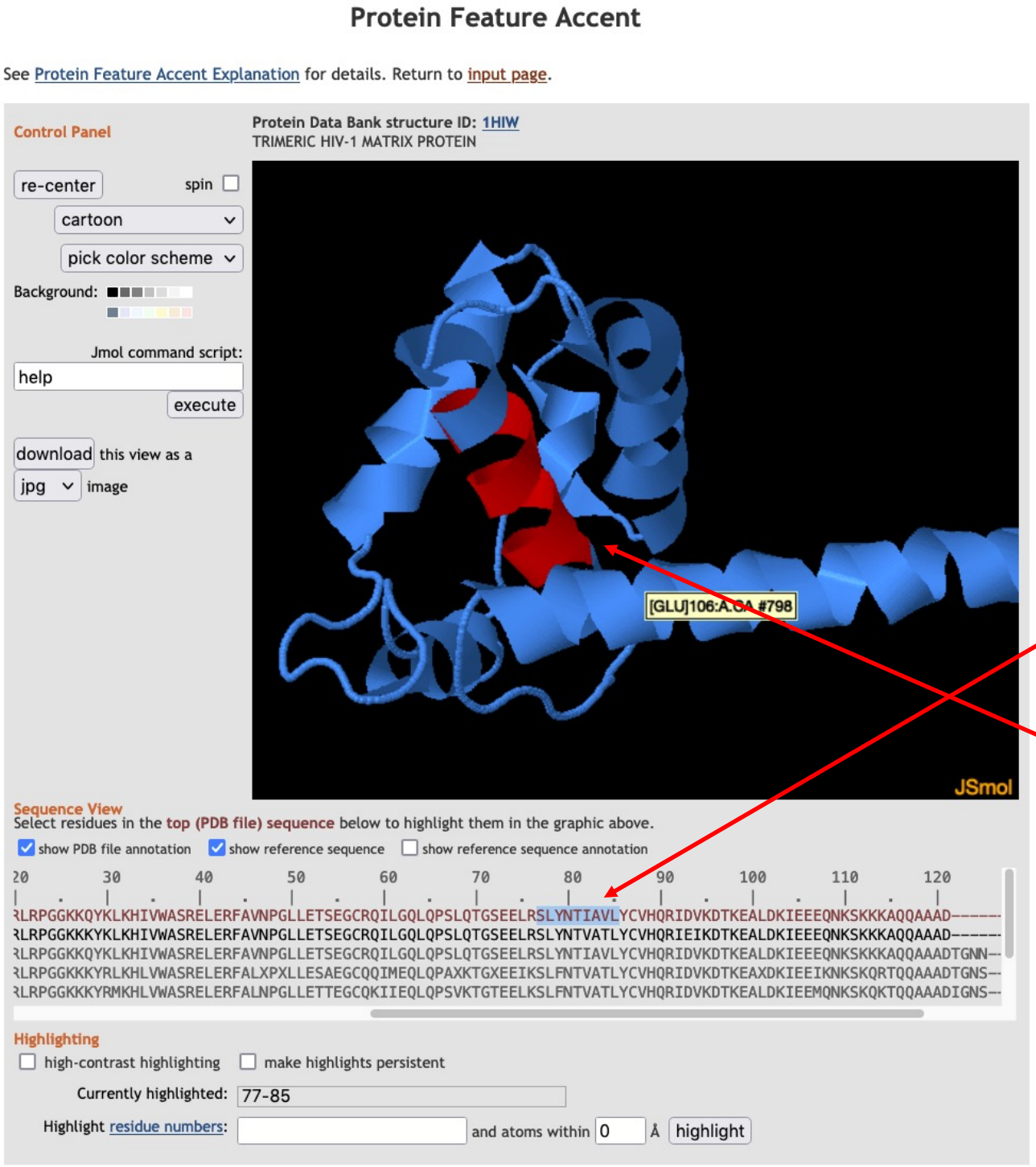

Our Protein Feature Accent tool, loads a PDB 3D structure file and an alignment of protein sequences.

JMol is used to display the protein structure so you can rotate it, change the view from cartoon to spacefill, etc.

Selecting sites or regions in the sequence

highlights that site or region in the structure.

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Gag from all primate lentiviruses.

Looking outside the HIV-1 M group (roughly 100 years of evolution in humans) to SIVs representing at least 100,000 years and more likely over 5 million years of evolution.

Can help determine which amino acids are invariant and thus likely critical for protein function.

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Thank you for attending!

We are happy to help with research questions on the use of our tools and database.

We are thrilled to get ideas for further tool development!

Contact us: seq-info@lanl.gov or immuno@lanl.gov

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Datasets and programs

https://tinyurl.com/HIVDB-IEDB-2021

Aliview

Bioedit

Figtree

slides

datafiles

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