Analysis of HIV-1 Amino Acid Sequences Suggests No Correlation Between Diversity and Progressor Groups

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BIOL 368: Bioinformatics Laboratory
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Outline

- The HIV virus attacks CD4 T cells in the human body and can mutate its genetic structure through changes in amino acids
- Markham et al. 1998 concluded that there was higher diversity among rapid progressor groups
- Amino acid sequences of the first and last visits of four subjects from Markham et al. 1998 were analyzed
- No significant difference was observed between number of mutations observed in rapid versus nonprogressors
- Certain types of mutations may affect the structure of gp120 more dramatically than others
- Future studies consisting of subjects with larger numbers of data points are needed to determine higher rates of diversity among rapid progressors

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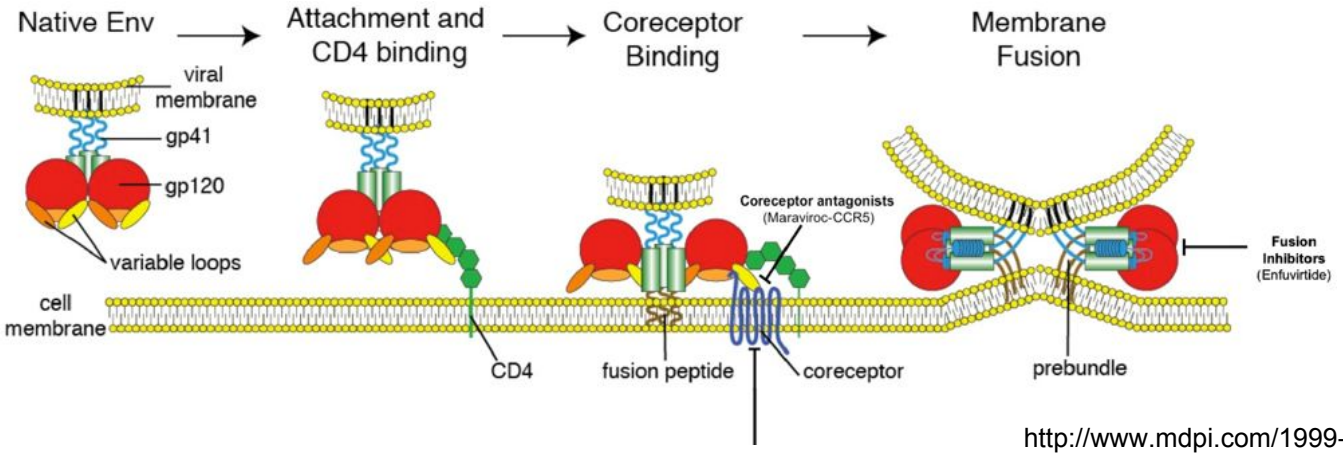
When the HIV virus enters a human host it attacks and infects CD4 T cells.

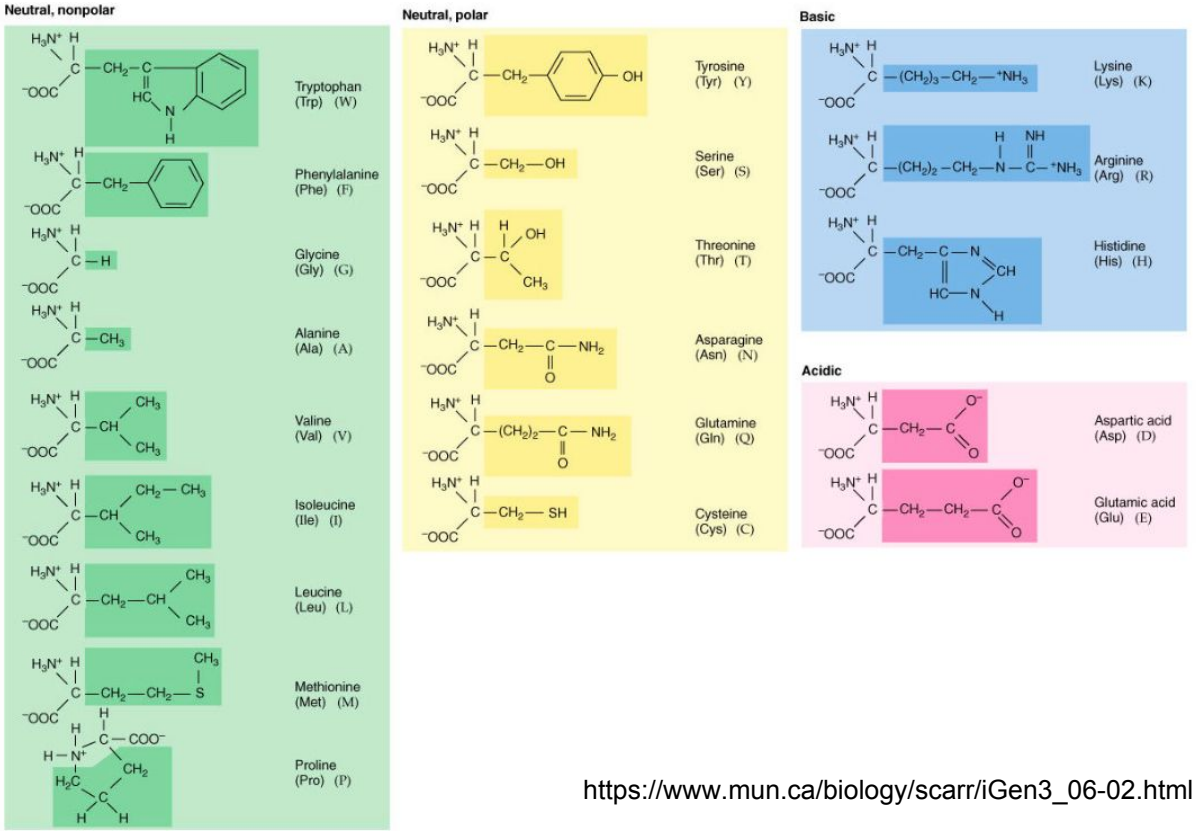
- HIV viruses infect CD4 T cells in human hosts enabling them to proliferate
  - A CD4 T cells' role is to protect the body from infection
  - When a CD4 T cell is infected with an HIV virus, it cannot fight infections
- The V3 loop on the gp120 of a HIV-1 virus helps to infect the host cell by binding to a cytokine receptor



http://www.mdpi.com/1999-4915/4/2/309/htm

Amino acid mutations can change the structure of the HIV virus.

- HIV viruses actively mutate their genetic structure in the body making it difficult to create drugs to kill the virus
- Amino acids are most often categorized into four sections
  - Nonpolar, Polar, Basic, and Acidic



https://www.mun.ca/biology/scarr/iGen3_06-02.html

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- **Markham et al. 1998 concluded that there was higher diversity among rapid progressor groups based on nucleotide sequences**
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Markham et al. 1998 showed results concluding higher diversity was found among rapid progressors compared to nonprogressors.

- 15 subjects were compared and placed into categories of nonprogressors, moderate progressors, and rapid progressors based on CD4 T cell decline
- Diversity between the clones of each subject was determined by comparing the number of nucleotide sequence mutations
- Rapid progressors were determined to have a higher diversity

Does diversity increase within the amino acid sequences over time more within the rapid progressors compared to nonprogressors?

- **Hypothesis: Since diversity increases over time within the DNA sequences for all progressor types, and more so within the rapid progressors, we would expect to see more diversity within the amino acid sequences over time within the rapid progressors.**
  - **The amino acid sequences in the first and last visits for four subject were analyzed**
  - **Using the multiple sequence alignment and Clustaldist tools within Biology Workbench the first and last sequences were compared**
  - **Amino acid changes were analyzed based on severity**
  - **The locations of the mutations were observed using a structure of gp120 created by Huang et al.**

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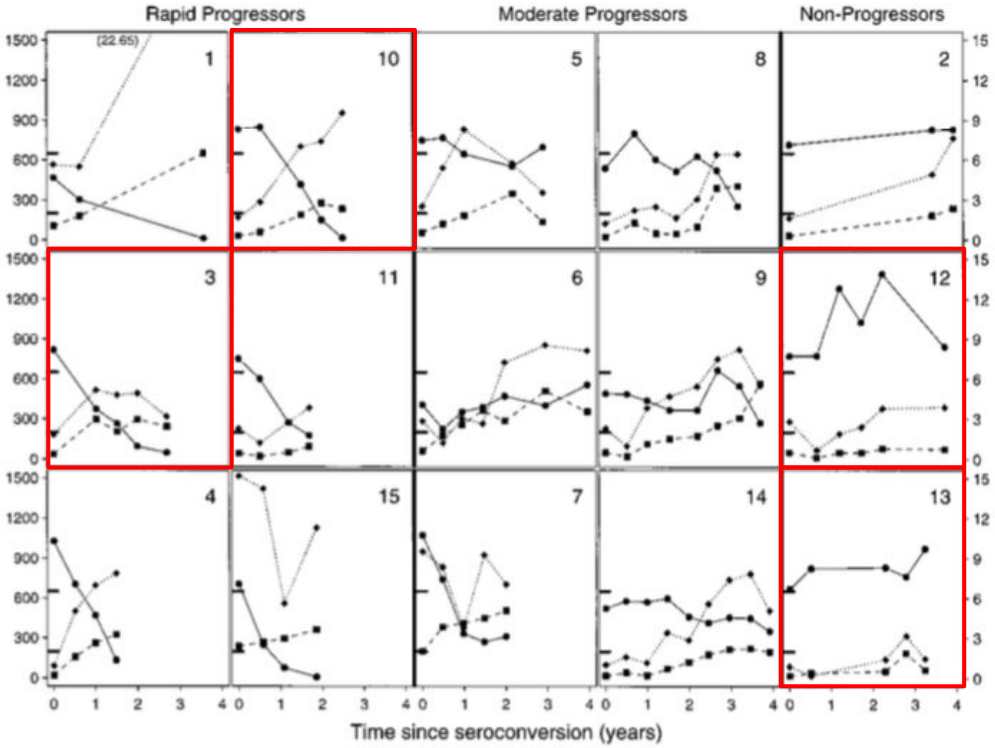
Two subjects from the nonprogressor and rapid progressor groups were chosen comparing each of their first and last visit clones.



Markham et al. 1998

The overall number of mutations per subject were calculated using multiple sequence alignment.

Group	Subject	Number of Clones	Number of Mutations
Nonprogressors	Subject 13	10	5
Nonprogressors	Subject 12	12	11
Rapid Progressors	Subject 3	10	9
Rapid Progressors	Subject 10	17	24

Sequence alignment of rapid progressors shows large number of mutations from first to last

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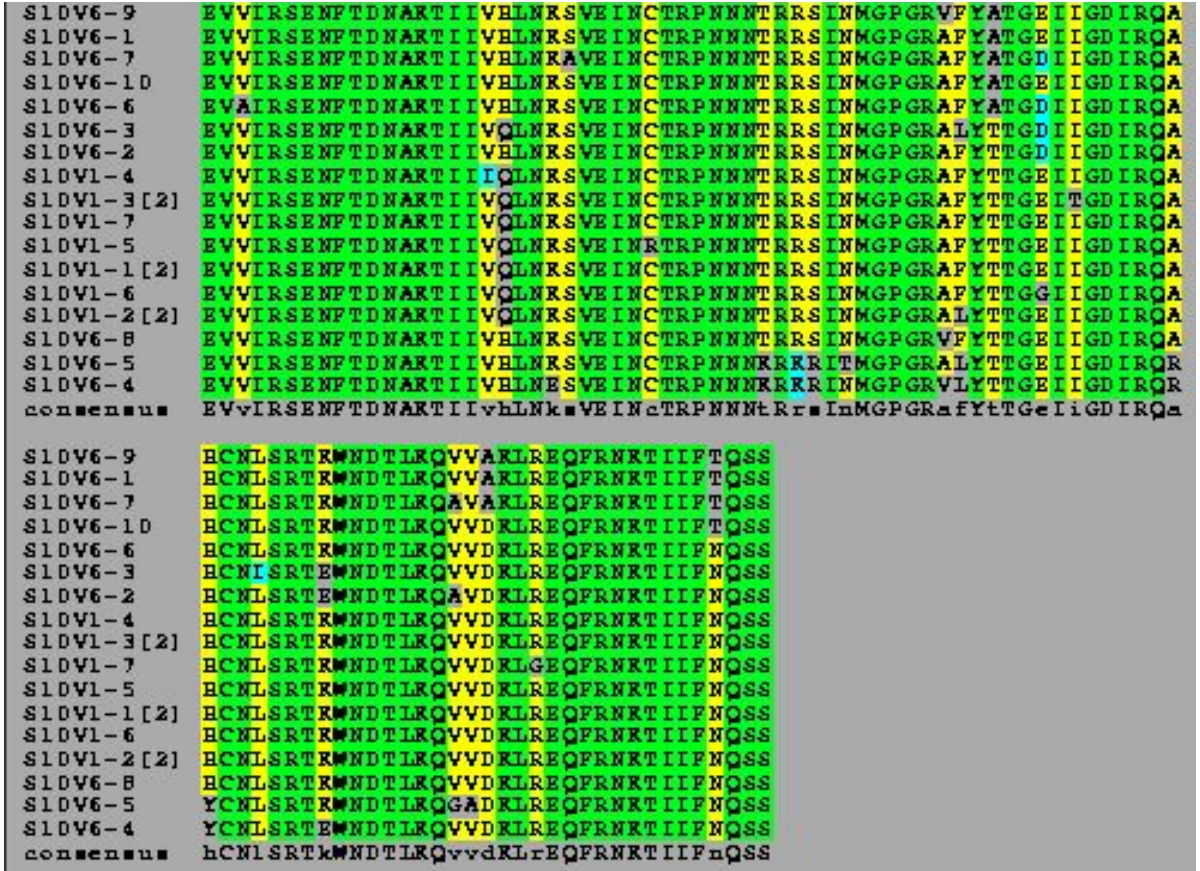
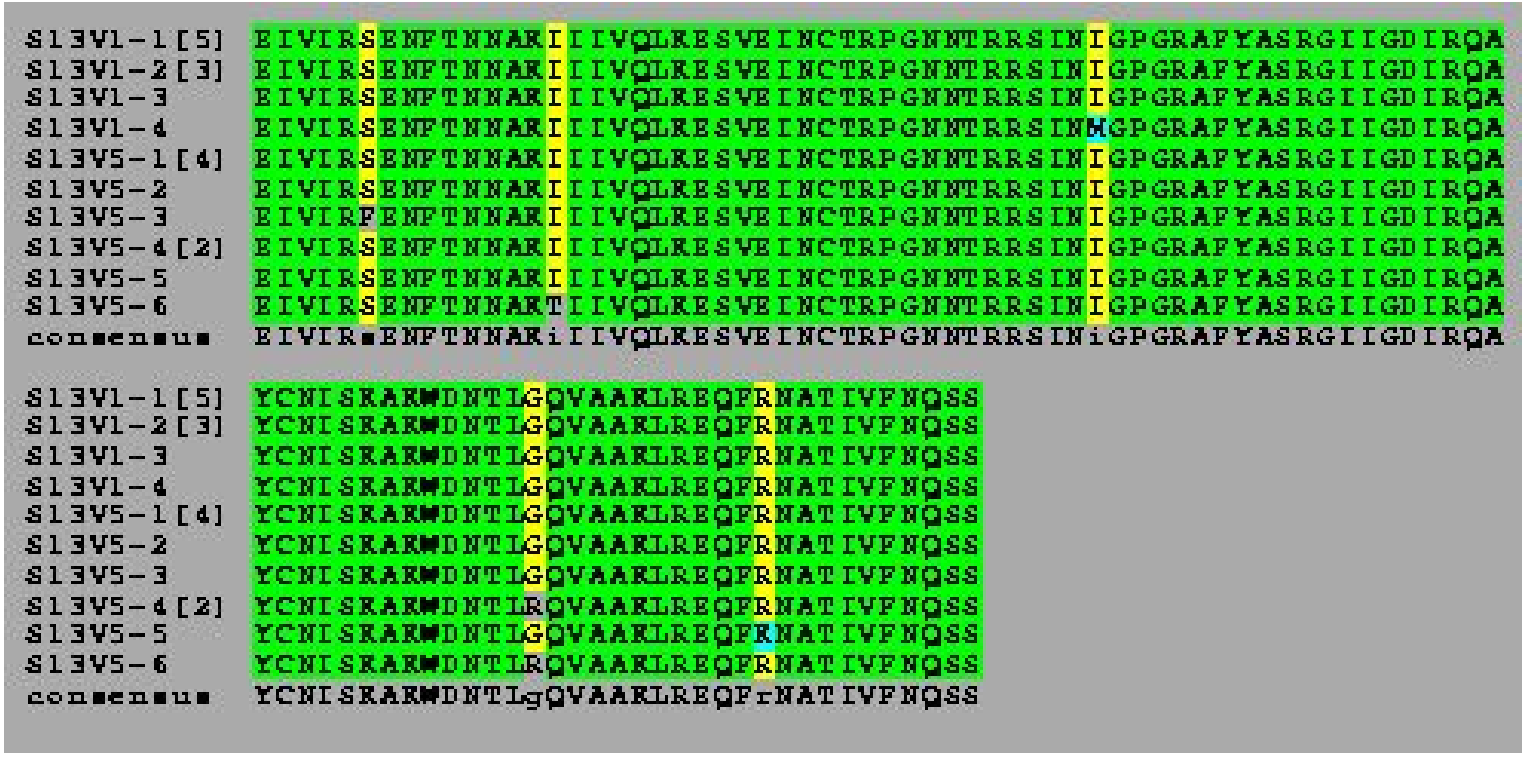
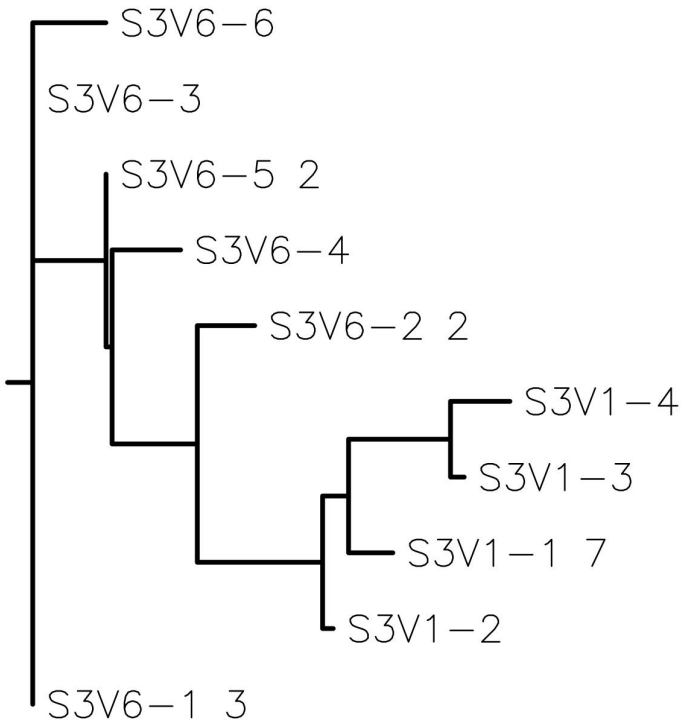


Sequence alignment of nonprogressor 12 shows a slightly lower number of mutations per number of clones than rapid progressors collectively.

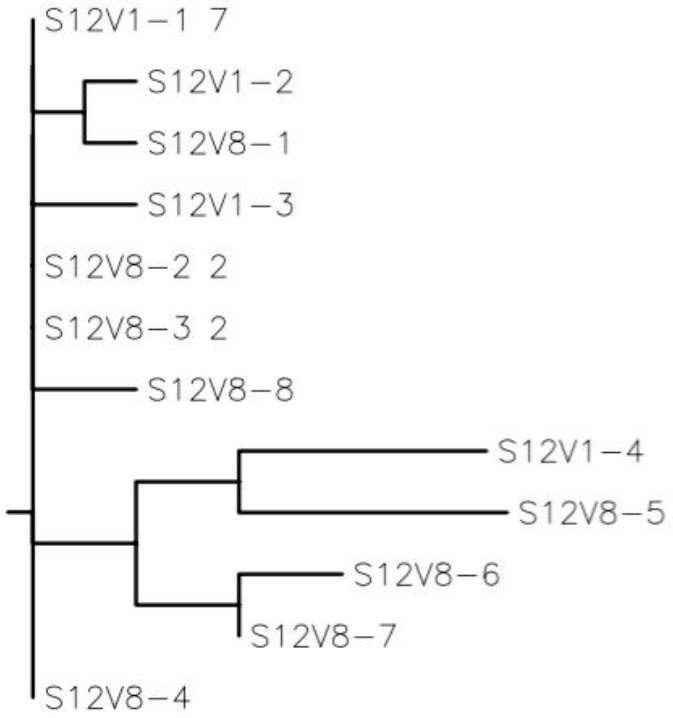
Sequence alignment of nonprogressor 13 shows less mutations than all other subjects observed.



Rooted trees demonstrated no discernable difference between rapid and nonprogressors.



Rapid progressor 3



Nonprogressor 12

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Clustaldist shows higher rate of diversity among rapid progressors by comparing the distances of mutations from the first visit to the last per subject.

Progressor Type	Subject	Distance between first and last visit
Rapid	3	0.010
Rapid	10	0.137
Non	12	0.010
Non	13	0.021

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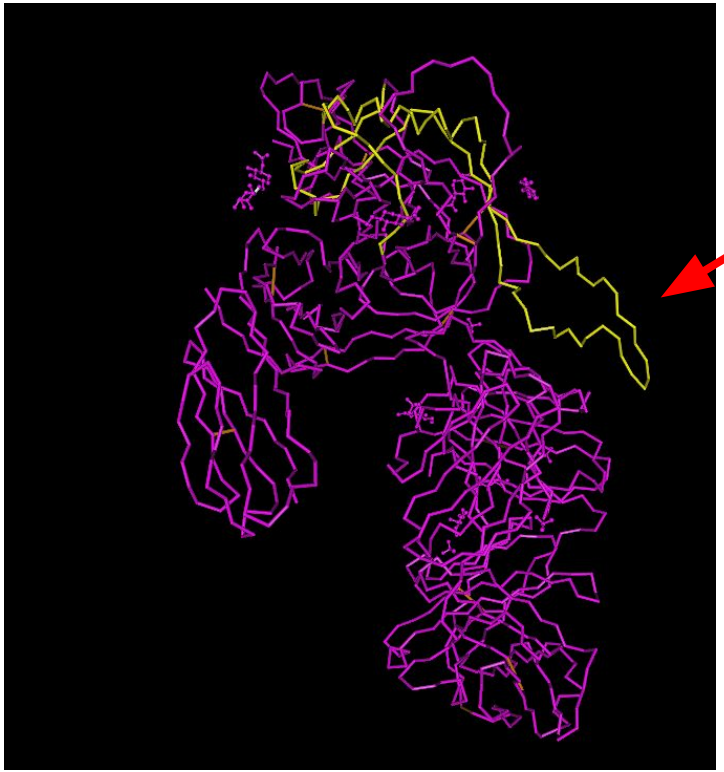
Mutations are identified and located using the structure of gp120 discovered by Huang et al.

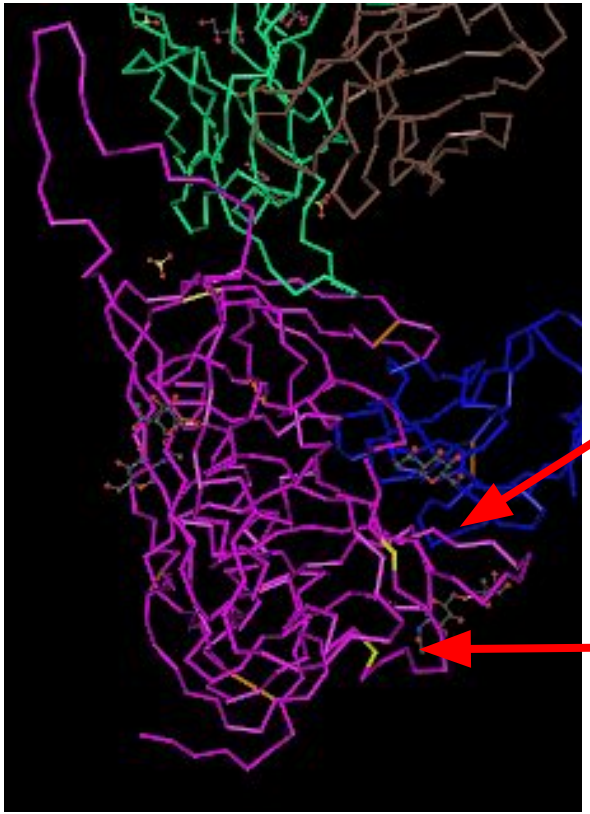


Highlighted in yellow is the V3 region of gp120, the V3 loop can be seen protruding out to the right.

The types of mutations may affect how the protein function would differ.

Progressor type	Subject	Number of noconsensus mutations	Number of clones	Average number of no consensus mutations per clone
Rapid	3	2	10	0.20
Rapid	10	8	17	0.47
Non	12	5	12	0.42
Non	13	3	10	0.30

Although rapid progressor 3 shows less no consensus mutations than expected, a deletion could alter the entire V3 section.



Valine, a nonpolar amino acid mutates to Alanine, also nonpolar, but then mutates to Glycine, a polar amino acid (nonsynomous mutation)

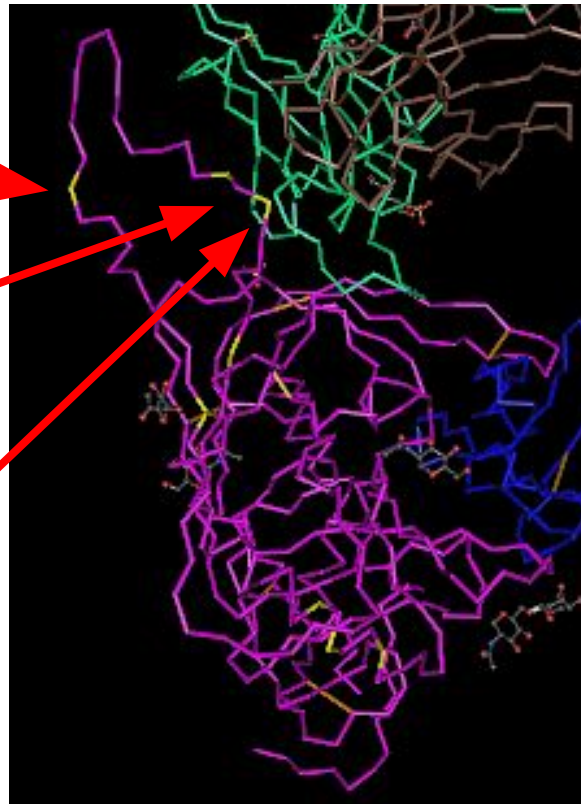
Arginine is deleted in one of the last visit's clones

Rapid progressor 10 shows multiple mutations on V3 loop that may affect binding.

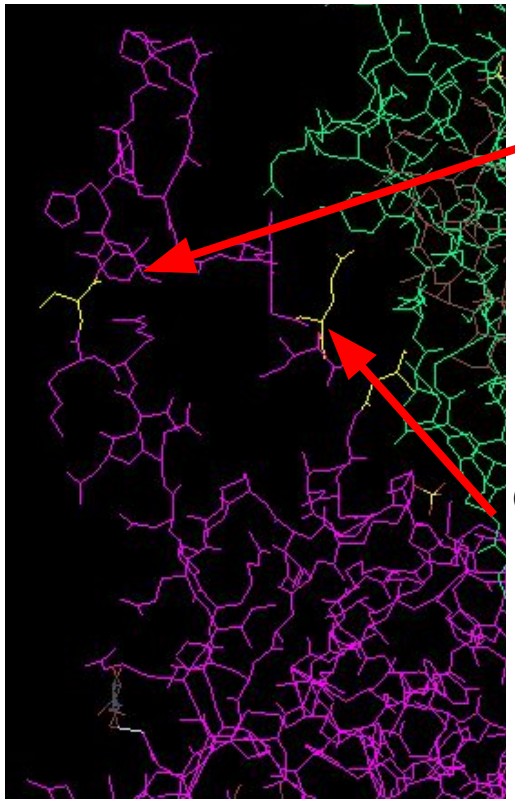
Serine, a polar uncharged amino acid mutates to Arginine, a basic amino acid

Glutamic Acid, an acidic aa mutates to Aspartic Acid, another acidic aa and also mutates to Glycine, a polar uncharged aa

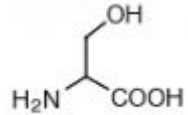
Isoleucine, a nonpolar aa mutates to Theronine a polar uncharged aa (nonsynomous mutation)



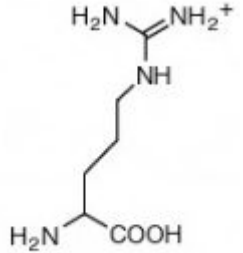
Changes in the structure of each amino acid mutation can affect the severity of the mutation in rapid progressor 10.



Serine (polar uncharged) to Arginine (basic)

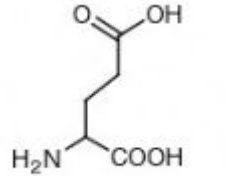


Serine (Ser, S)

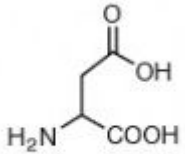


Arginine (Arg, R)

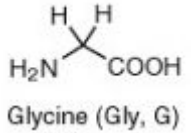
Glutamic Acid (acidic) to Aspartic Acid (acidic) to Glycine (polar)



Glutamic Acid (Glu, E)



Aspartic Acid (Asp, D)



Glycine (Gly, G)

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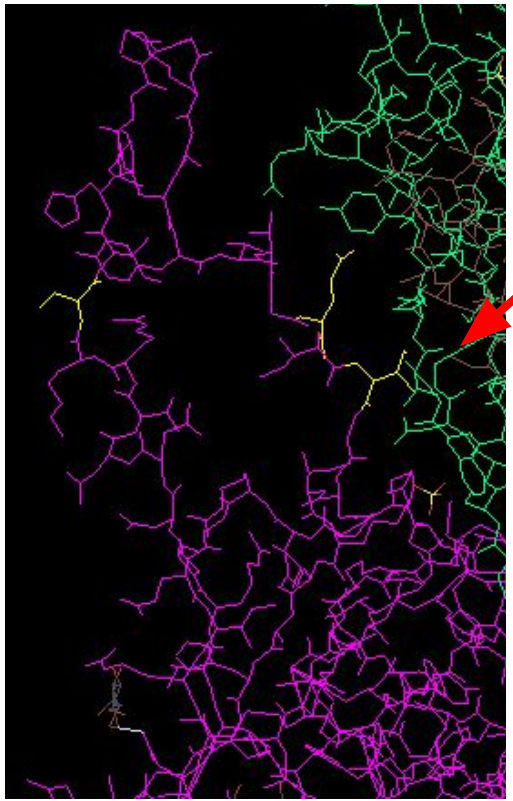
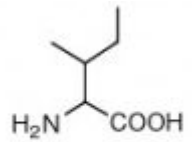
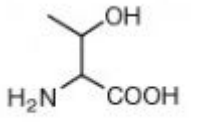


Isoleucine (nonpolar) to Theroine (polar uncharged)



Isoleucine (Ile, I)



Threonine (Thr, T)

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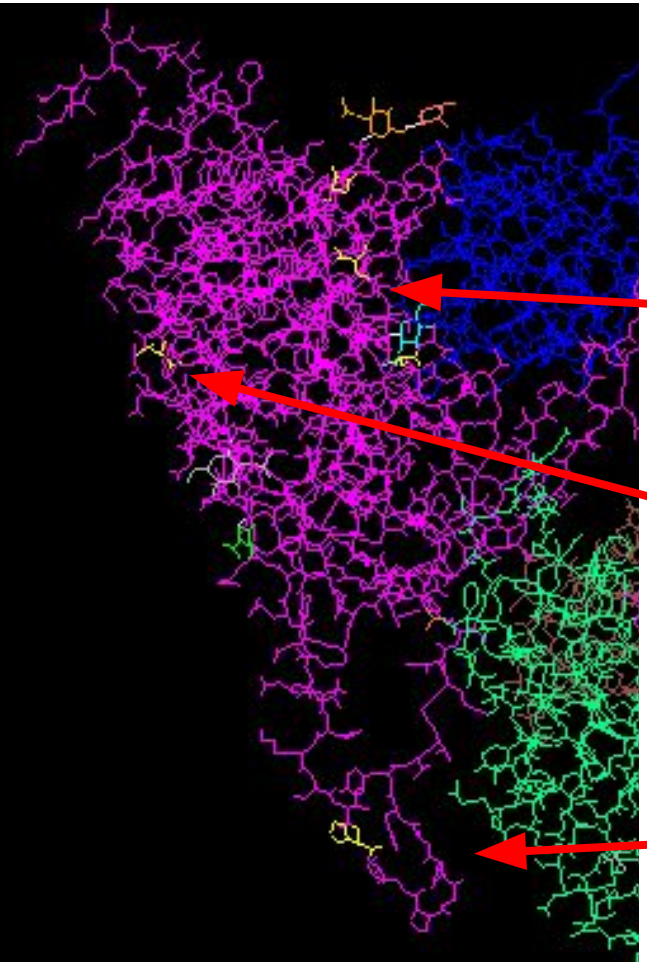
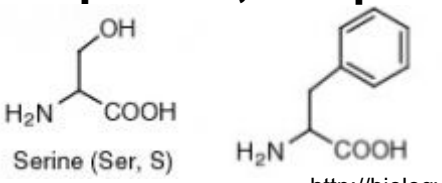


Nonprogressor 12 had a higher number of mutations within the V3 region than nonprogressor 13.

Valine (hydrophobic; nonpolar) to Lysine (positively charged; basic)

Glutamic Acid (acidic; negatively charged) to Theonine (polar)

Serine (polar) to Phenylalanine (hydrophobic; nonpolar)



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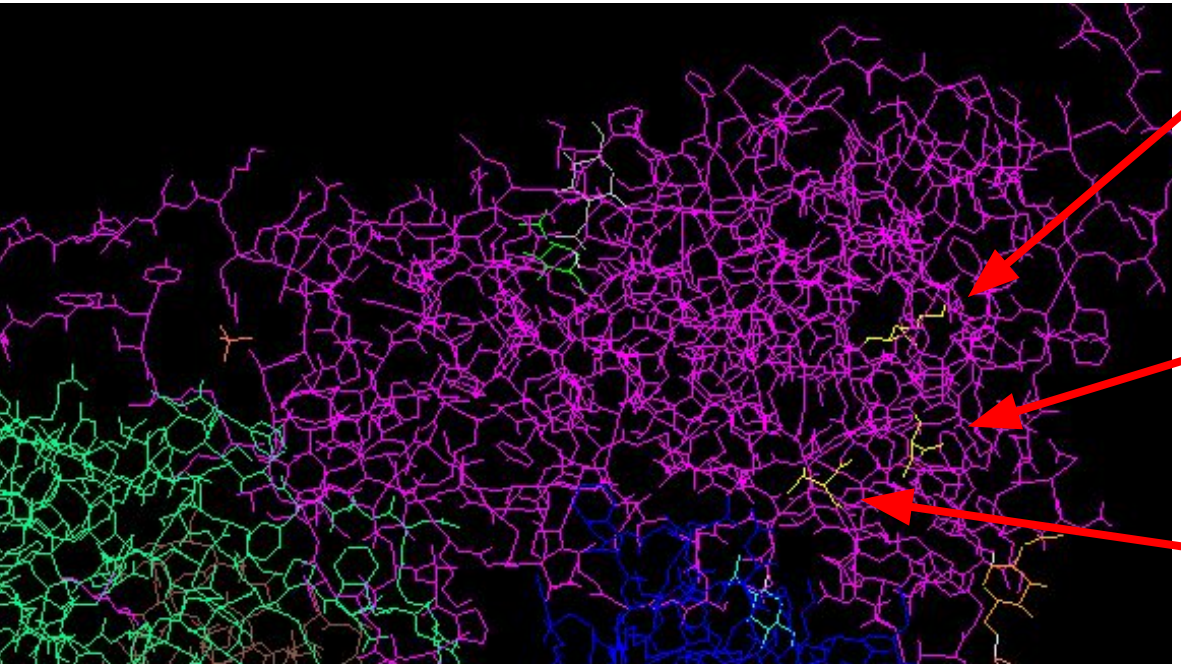
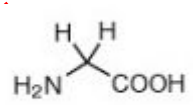
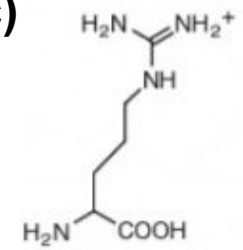
Nonprogressor 13 had fewer mutations which occurred further away from the V3 loop than other subjects.



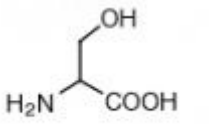
Glycine (hydrophobic) to Arginine (Basic)



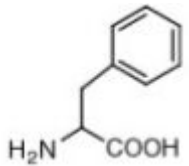
Glycine (Gly, G)



Serine (polar) to Phenylalanine (aromatic; hydrophobic)



Serine (Ser, S)



Isoleucine (hydrophobic) to Methionine (polar)

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Data suggests higher diversity among rapid progressor amino acid sequences than nonprogressors, but a larger sample size is needed to confirm results.

- No mutation patterns were observed within progressor groups
- More mutations occurred within the V3 loop in rapid progressor 10 than the other subjects
- Using more subjects would create a larger sample size
  - Moderate progressor group was overlooked
- More data points per subject could increase statistical evidence
  - Subjects from a more consistent study could be used

Summary

- Mutations which occur within the amino acid sequences of HIV-1 can affect the structure and overall function of the virus
- Markham et al. 1998 concluded that there was higher diversity among rapid progressor groups
- Mutations within amino acid sequences of progressors and nonprogressors from Markham et al. 1998 were analyzed
    - Different types of mutations may affect the structure of gp120 more drastically than others
- No significant difference was observed between number of mutations observed in rapid versus nonprogressors
- To gather more conclusive data, subjects with larger numbers of data points would be needed to determine if there is a higher rate of diversity among the rapid progressor group

Acknowledgements

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References

Huang, C. C., Tang, M., Zhang, M. Y., Majeed, S., Montabana, E., Stanfield, R. L., ... & Wyatt, R. (2005). Structure of a V3-containing HIV-1 gp120 core. *Science*, *310*(5750), 1025-1028.

Kirchherr, J. L., Hamilton, J., Lu, X., Gnanakaran, S., Muldoon, M., Daniels, M., Kasongo, W., Chalwe, V., Mulenga, C., Mwananyanda, L., Musonda, R.M., Yuan, X., Montefiori, D.C., Korber, M.T., Haynes, B.F., & Musonda, R. M. (2011). Identification of amino acid substitutions associated with neutralization phenotype in the human immunodeficiency virus type-1 subtype C gp120. Virology, 409(2), 163-174. DOI: 10.1016/j.virol.2010.09.031

Markham, R.B., Wang, W.C., Weinstein, A.E., Wang, Z., Munoz, A., Templeton, A., Margolick, J., Vlahov, D., Quinn, T., Farzadegan, H., & Yu, X.F. (1998). Patterns of HIV-1 evolution in individuals with differing rates of CD4 T cell decline. Proc Natl Acad Sci U S A. 95, 12568-12573. dos: 10.1073/pnas.95.21.12568