A longitudinal study on the dynamics of plasma neutralising antibodies and its determinants in HIV-2 infected individuals

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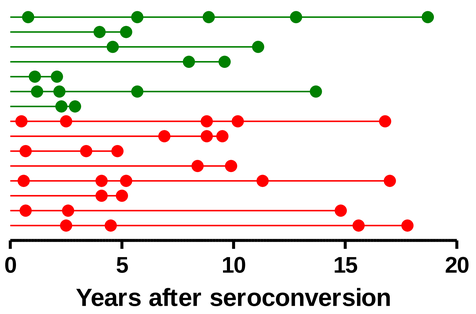
BACKGROUND

- HIV-2 is known to be less virulent and less pathogenic than HIV-1, and the majority of HIV-2-infected individuals remain asymptomatic much longer than HIV-1-infected individuals. Plasma viral load is approximately 1 log unit lower in HIV-2 than HIV-1 when matched for CD4+ T cell count, whereas the proviral load does not appear to differ between the two infections. Therefore, HIV-2 infection represents a model for the studies of immune responses that may control HIV infection and possibly functional cure.
- In HIV-1 infection, broad plasma neutralizing antibody (NAb) response develops in only 15% of individuals after two to four years of infection. Moreover, potency of NAb seems to be low at around 1:300 and fluctuating.
- To study dynamics of NAb response in HIV-2 infected individuals is difficult because of usually unknown seroconversion time and relatively silent disease course.
- Guinea-Bissau is a small country in West Africa with the highest HIV-2 prevalence in the world (2%). In 1990, we initiated an occupational cohort of police officers in Guinea-Bissau. Regular follow-up visits enabled us to investigate evolution of NAb response longitudinally in fifteen HIV-2 infected individuals with known seroconversion time.

QUESTIONS

- When does broad and potent p-NAb response develop in HIV-2 infected individuals?
- What are the modulators of broad and potent p-NAb response

Characteristics of participants



Fourty-six plasma were obtained from 15 individuals from a cohort of police officers in Guinea-Bissau, between 1992-2010.

- None of the participants received HAART.
- Estimated date of infection was defined as the midpoint between the last HIV-2 seronegative and the first seropositive sample.
- Median age (IQR) at seroconversion: 31 years (28-39)
- Median (IQR) follow-up period: 10 year (5-17)
- Median # blood samples: 3 (2-4)
- Median (IQR) CD4+ T cell count/µl: 500 (306-722)
- Median (IQR) CD8+ T cell count/ µl: 475 (351-818)
- Participants were classified into two subgroups based on mean CD4+ T cell count/µl: Immunocompetent (≥500,🟢) versus immunosuppressed (<500 🔴).

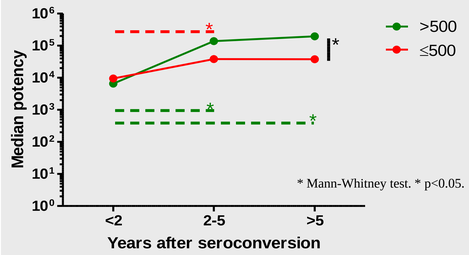
Neutralization assay by plaque reduction

- **Target cell:** GHOST(3)-CCR5 cells
- **Complement-inactivated plasma:** Starting plasma dilution was 1:20.
- Neutralization (%)= (1- $\frac{\text{\#plaques in the presence of virus and plasma}}{\text{\#plaques in the presence of virus}}$)x100

- **HIV-2 panel**

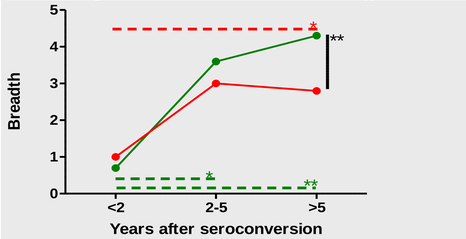
- Cut-off point for neutralization: 30%
- Magnitude of NAb: highest plasma dilution neutralizing corresponding isolate above cut-off point.
- Potency of NAb: summary of magnitude of NAb against corresponding isolates/ # isolates
- Breadth of NAb: # isolates neutralized

Potent NAb response developed during the first two years of infection and was persistent



- During the first two years after seroconversion, seven out of seven participants succeeded to develop potent NAb response at a score of 10000.
- Potent NAb response was persistent during the whole follow-up period in all participants.
- In the immunocompromised group, potency score stabilized at around 30000 in the end of 5 years after seroconersion.
- In the immunocompetent group, potency score showed persistent increase throughout the whole follow-up period and reached approximately to 200000 in the end.
- Potency score was significantly higher at the end of follow-up period in immunocompetent group compared to the immunodeficient group (200000 vs 25000).

NAb response at 1:10000 dilution differentiates immunocompetent from immunocompromised group



- NAbs were able to neutralize only one isolate during the first 2 years after seroconversion.
- In the immunocompromised group, breadth score stabilized at around 3 in the end of 5 years after seroconversion.
- In the immunocompetent group, breadth score showed persistent increase throughout the whole follow-up period and reached approximately 4,5 in the end.
- Breadth score was significantly higher at the end of follow-up period in favor of immunocompetent group (4,5 vs 2,5).

RESULTS

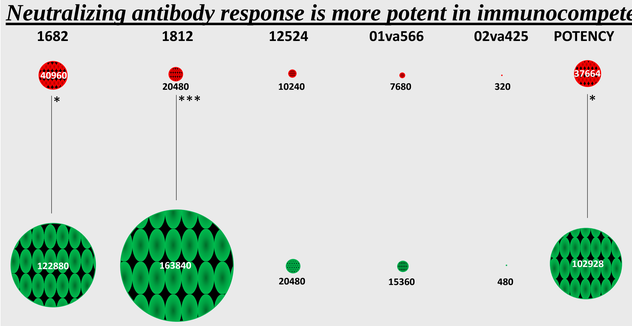
Immunocompetent individuals were infected by HIV-2 at an earlier age

Characteristics	Immunocompetent	Immunodeficient	p value*
Median (IQR) age at seroconversion	28 (27-30)	38 (33-41)	0,014
Gender (% women)	38	0	0,2
Median (IQR) follow up period (yr)	10.4 (3.5-16.0)	10.0 (5.0-17.0)	0,72
Median (IQR) plasma sampling/individual	3 (2.2-4.8)	2 (2-4)	0,22
Median (IQR) CD8+ T cell count/µl	755 (408-1150)	334 (264-394)	0,0027

* Mann-Whitney test.

- Study participants were subgrouped based on mean CD4+ T cell count/µl per individual. Participants with mean CD4+ T cell count ≥500 cells/µl were classified as **immunocompetent** and green color-coded. Participants with mean CD4+ T cell count <500 cells/µl were classified as **immunodeficient** and red color-coded.
- Median follow-up period and number of plasma sampling per individual were similar in both groups.
- The aging of the immune system, also named immusenescence, starts at early 30s and encompasses changes of both humoral and cellular responses in healthy individuals. HIV-2 seroconversion before early 30s may have a role on keeping cellular immunity relatively intact.

Neutralizing antibody response is more potent in immunocompetent group



* Mann-Whitney test. * p<0.05, *** p<0.001.

- Magnitude of NAb response against neutralization sensitive isolates(1682 and 1812) was higher in the immunocompetent group.
- Magnitude of NAb response against resistant isolates (12524, 01va566 and 02va425) did not display significant difference between the groups.
- NAb response was found to be significantly more potent in the immunocomptent group.
- Younger age and relatively intact cellular immunity may play role in more potent humoral immunity in the immunocompetent group.

Modulators of Breadth and Potency of p-NAb Response

- Age at seroconversion correlated negatively with CD4+ and CD8+ T cell count (r=-0.64 and -0.41, respectively, p<0.05)
- There was a positive correlation between breadth and potency (r= 0.75, p<0.05)
- In the immunocpromised group, CD4+ and CD8+ T cell count tended to decrease with years after serconversion (-0.62 and -0.88, respectively, p<0.05). Decreasing number of cellular immunity cells correlated negatively with potency of humoral immunity (-0.55 and -0.78, p<0.05).
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- In the immunocompetent group, both breadth and potency of NAb response tended to increase with years after serconversion (0.53 and 0.51, respectively, p<0.05). Furthermore, potency of NAb response correlated positively with CD4+ T cell count (0.72, p<0.05).

CONCLUSION

- This study represents the most diverse longitudinal seroconversion infection cohort studied to date for HIV-2 neutralization.
- Broad and potent NAb response developed in all participants within the first two years of HIV-2 infection and persisted throughout the whole follow-up period. This strong humoral immunity response may contribute to the control of HIV-2 infection and functional cure.
- Immunosenescence may play a role in the development of humoral immunity against HIV-2. Individuals infected at a younger age seem to keep both humoral and cellular immunity active against HIV-2. However, older individuals with relatively impaired cellular immunity display blunted humoral immunity against HIV-2.
- Our results indicate that CD4+ T cells stimulate potent antibody production in immunocompetent individuals. Interestingly, in immunosuppressed individuals, despite decrease in CD4+ T cell count, potent antibody production continues. We speculate that increasing viremia in the immunodeficient group is responsible for potent antibody production.

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