Contents available at **PubMed**
www.aidsreviews.com

PERMANYER

AIDS Rev. 2019;21:23-27

A Model for Potential B-cell Precursors of Broadly Neutralizing HIV-1 Antibodies Selection and Antibody Affinity Maturation

Radmila Metlas[1] and Tatjana Srdic-Rajic[2]

[1]Center for Multidisciplinary Research, Institute of Nuclear Sciences VINCA; [2]Department of Experimental Oncology, National Cancer Research Center. Belgrade, Serbia

Abstract

The goal of this report was to propose a model, wherein synergy between the B-cell antigen receptor (BCR) and toll-like receptor (TLR) signaling is involved in the selection of the B-cell precursors of HIV-1 broadly neutralizing antibodies (bnAbs) with long heavy chain complementarity determining regions 3, from immature/transitional B cells. The model predicts the involvement of Ab/HIV-1 complexes in a way that Ab from the complex binds both BCRs and HIV-1, while on internalization of HIV-1 TLR ligands such as CpG motifs interacts with TLR9. The result of BCR and TLR9 orchestrated signaling is a formation of somatically mutated memory B cells potential precursors of bnAbs. Generated memory B cells continuously exposed to different Ab/HIV-1 complexes can elicit specific bnAb by stochastic somatic hypermutation rather than in the Darwinian process. This new view of the interaction between Ab/HIV-1 complexes and immune system, leading to affinity maturation of the bnAbs in the absence of nominal HIV-1 antigen and BCR interaction, may have implication for the vaccine designed and passive immunization. **(AIDS Rev. 2019;21:23-27)**

Corresponding author: Radmila Metlas, metlas.r@sbb.rs

Key words

HIV-1 broadly neutralizing antibodies. Precursor of B-cell antigen receptor for broadly neutralizing antibodies. Toll-like receptor. Membrane proximal external region.

Introduction

During the course of natural HIV-1 infection, individuals develop antibodies (Abs) with non-neutralizing and neutralizing capacity unable to clear the infection. In addition, HIV-1 enormous antigenic variability[1] progressively destroys the immune system. However, somatic evolution over several years contributes that Abs achieve the capacity to neutralize a broad range of HIV-1 isolates. These broadly neutralizing antibodies (bnAbs) developed between 10% and 50% of individuals only after 2 or more years of chronic infections[2,3] while approximately 1% develop elite neutralizing Abs[4]. Several key vulnerable regions of the trimeric

Correspondence to:
Radmila Metlas
Institute of Nuclear Sciences VINCA
11000 Belgrade, Serbia
E-mail: metlas.r@sbb.rs

Received in original form: 05-10-2018
Accepted in final form: 24-01-2019
DOI: 10.24875/AIDSRev.19000028

HIV-1 envelope glycoprotein (Env) targeted by bnAbs include CD4-binding site, trimer apex, membrane proximal external region (MPER), gp120-gp41 interface, and high-mannose patch[5]. The MPER a highly conserved region of the HIV-1 Env gp41 subunit situated in the context of diverse envelope states required for HIV-1 fusion event between viral and cellular membranes[6] is among the main targets for vaccine design. Several bnAbs react with linear and partially overlapping epitopes localized in the MPER, the most C-terminal region of the exterior part of gp41[7], only transiently exposed in the native Env spike during membrane fusion[8] when Env gp120-gp41 trimer undergoes substantial conformational changes.

The posed goal for the HIV-1 vaccine is to reproducibly elicit potent bnAbs[9]. However, the past efforts to recapitulate eliciting of potent bnAbs by vaccination have failed[10,11]. The unusual traits of bnAbs might be factors that limit their expression including long third heavy chain complementarity determining regions 3 (HCDR3) in the Ab heavy chain[12], extensive somatic hypermutation (SHM) frequencies[13], autoreactivity[14], and large insertions and deletions[15]. In particular, the high evolution rate of HIV-1[16] prevents the antibodies from controlling the infection[17]. The findings that HIV-1 Env and host immunoglobulin (Ig) variable regions (VH) share sequence homology and consequently the existence of complementarity structures between anti-HIV-1 and anti-Ig Abs of non-infected individuals[18-20] might be an additional obstacle in the understanding of HIV-1 bnAbs production. At present, vaccination protocol that can efficiently recapitulate extraordinarily number of mutations that are acquired in HIV-1 bnAbs during affinity maturation process is not available[21]. Furthermore, an important finding is that trimeric HIV-1 Env is generally ineffective in engaging germline B-cell antigen receptors (BCRs) of bnAbs irrespective of their epitope target[22]. The MPER epitope for immunogen design is a challenge as long HCDR3 loops of the MPER-directed Abs corecognize a linear sequence segment in the highly conserved MPER and viral membrane lipids[23,24]. The inability of recently developed immunogens to beside induction of Tier-2 neutralizing activity evoke a bnAb response[25] may suggest that natural infection remains the only system in which we can decipher the parameters that drive the evolution of bnAbs[26]. An additional obstacle for HIV-1 cure research is T regulatory cells (Tregs) which activity in HIV-1 infection may play a central role in shaping the HIV-1 reservoir and compromising the HIV-specific immune responses[27].

Peculiarities concerning MPER-directed HIV-1 bnAbs

In this report, we would like to pay attention on MPER-directed bnAbs and to suggest their developmental pathway:

i. Antibodies that recognize an epitope in the MPER of the HIV-1 gp41 transmembrane glycoprotein in the liposome context, such as 10E8 bnAb appears after years of chronic infection from an unmutated common ancestor with weak MPER affinity[28]
ii. Anti-MPER antibodies are produced in HIV-infected individuals although MPER immunogen is not accessible to B cells in the native Env spike[8] and thus make an obstacle to explain their origin and both design immunogens and immunization protocols
iii. Further, considerable speculation about the origin of MPER-directed antibodies such as 10E8 was raised due to its trait to recognize a gp41 epitope in the membrane context not present in V gene-reverted germline versions, suggesting that this trait is gained only after extensive SHM[29]
iv. Like other strongly neutralizing anti-HIV-1 Mabs, MPER-specific bnAbs use long HCDR3 loops of 20-34 residues compared to the average length of 16 residues of HCDR3 in human B cells[12]
v. It appears that long HCDR3s are generated at the stage of VDJ recombination in immature/transitional B-cell repertoire before the antigen-driven process of antibody affinity maturation, rather than through accumulation of insertions introduced during the SHM process[30].

Alterations in B-cell subpopulations after HIV infection

Numerous immunologic abnormalities may arise shortly after HIV-1 infection while progression of HIV-1 disease is associated with alterations within the B-cell population.

HIV-1 infection is associated with increased terminal differentiation of B cells. These plasmablasts arise early and are maintained at abnormally high levels in viremic individuals. However, the majority of plasmablasts and resting memory B cells are HIV unspecific[31]. Immature/transitional B cells are expanded in the periphery with advancing HIV-1 disease[32]; however, in immunocompromised individuals vaccine can program immature B cells to make antibodies that contribute to protective antiviral immunity[33].

Survival of immature/transitional B cells

Immature/transitional B cells are an important target for negative selection dependent also on the strength of BCR signaling to self-antigens not present in the bone marrow:

i. Due to their immaturity, they can be rescued from cell death induced by BCR stimulation alone, by the second signal[34]
ii. Positive selection of transitional B cells might be promoted by dual BCR/toll-like receptor (TLR) 9 signals in context-dependent manner[35]
iii. In human transitional B-cells, synergism between the BCR and TLR9 increases expression of AID, not Ag selected, that dictates the inclusion of transitional B cells into the memory repertoire with a modified antigen receptor suggesting that TLR9 stimulation might be T independent[36]
iv. In autoimmune disorder, modestly enhanced signaling downstream of the BCR and TLR can promote self-reactive transitional B-cell-positive selection[37]
v. A hallmark of memory B cells formed in response of BCR-antigen interaction is that affinity of specific Abs increases over the course of affinity maturation on reencounter with the antigen, as the result of a Darwinian process that alternates stochastic SHM[38]
vi. For bnAbs, it is found that they are enriched for improbable mutations being a significant barrier for vaccine design and immunization strategy[39]
vii. Studies of B-cell response during chronic HIV infection showed that anti-HIV-gp140 produced by the naive and memory B cells showed heteroligation where one combining site would bind to gp140, whereas the other would bind to any of several other ligands on the viral surface to increase activity[40].

A model for selection of potential B-cell precursors for HIV-1 bnAbs production

Despite enormous efforts, neither detailed pathways promoting positive selection of precursor B cells with the potential to produce bnAbs nor the nature of HIV-1 immunogens capable of generating protective antibodies are solved. The particular problem is how bnAbs might be produced to HIV-1 Env conserved sites that are not accessible on the naive virion such as epitopes in MPER of the HIV-1 gp41 transmembrane glycoprotein. Furthermore, it is not understood how these bnAbs gain the ability to neutralize HIV-1 by recognition of MPER epitope in the context of the membrane. Based on the above-presented data, we proposed a model that at first suggests the selection mode of B-cell potential precursors for bnAbs, as well as their developmental pathway on activation of positively selected B cells.

The model presented is in agreement with the possibility that HIV-specific B cells can develop directly from immature/transitional B cells independently of T-cell help[41]. The model (Fig. 1) predicts that immune complex formed between antibody reactive with HIV-1 genetic variants present in infected individuals play the main role to ensure efficient trafficking and interaction with BCRs of immature/transitional B cell as a potential precursor of HIV-1 bnAbs. Such idiotype-specific antibodies might be isolated even from normal human sera[19]. In another word, polyreactive HIV-1 antibodies by heterogeneous ligand binding or heteroligation are capable to bind HIV-1 ligand with one combining site while other combining site would bind BCR in Id-anti-Id manner. According to our data[18-20], scenario might be that both antibodies combining sites of HIV-1 reactive antibodies bind epitope shared between HIV-1 ligand and BCR.

Interaction of BCR and Ab/HIV immune complex leads to complex internalization and delivery of the virus-associated TLR ligands, such as CpG motifs to autophagosomes, which contain endosome-resident TLRs. Simultaneous engagement of the BCR and TLR9 triggers generation of plasma cells as well as somatically mutated memory B cells from immature/transitional B cells, as potential precursors for bnAbs. Memory B cells that experienced contact with Ab/HIV-1 complexes will on multiple encounters of the Ab/HIV-1 complexes promptly proliferate and initiate the process of SHM. Due to continuous exposure to the HIV-1 genetic variants, immune system is under sustained pressure to respond by producing various anti-HIV Abs. Therefore, different Ab/HIV-1 complexes will be present in circulation able to reactivate memory B cells that in response to BCR and TLR signaling will trigger stochastic SHM. It should be mentioned that these B cells might be under Tregs control if they expose processed BCR peptides as the peptide-MHC complex[42] suggesting that accomplishment of bnAb specificity may need years. Thus, properties of the immune system may control types of Abs produced rather than antigenic epitopes[43] which in HIV-1 case does not inform the rational design of an HIV-1 vaccine.

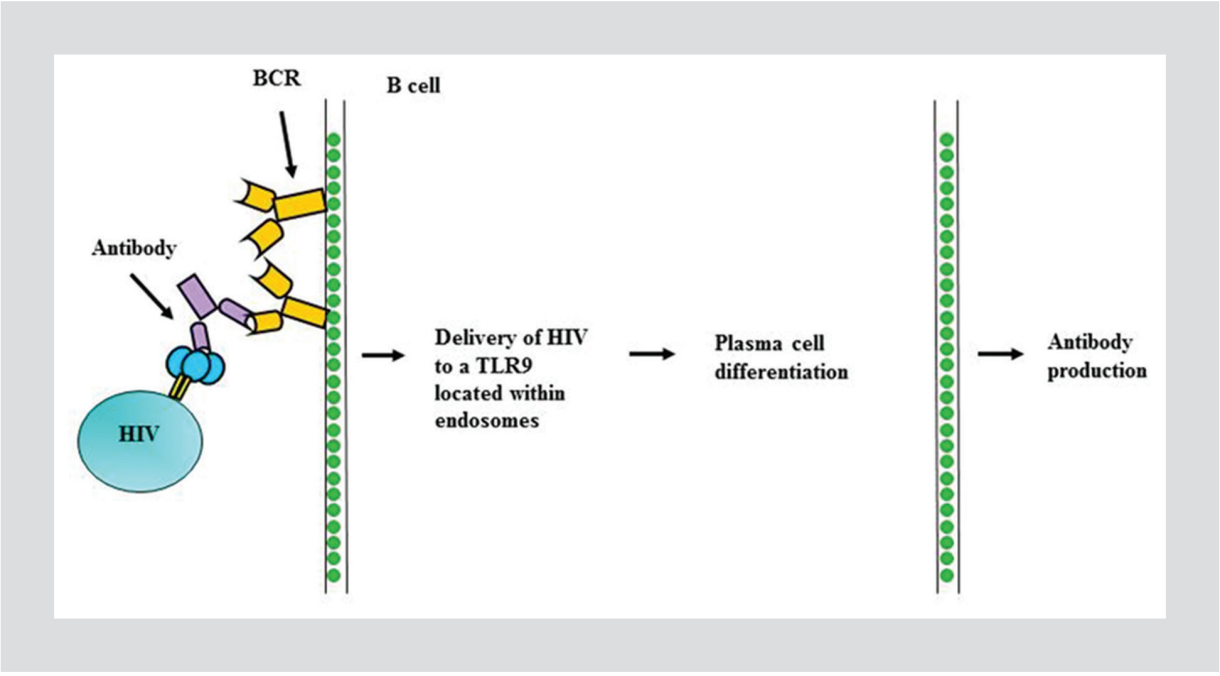

Figure 1. *Model for positive selection and activation signals of transitional B cells. Synergistic signaling events are mediated between B-cell antigen receptor (BCR) and toll-like receptor 9 (TLR9). Antibody from antibody-trimeric HIV envelope protein immune complex binds BCR by the heteroligation process. Interaction between BCR and immune complex triggers endocytosis of the immune complex containing TLR9 ligand and its delivery to endosome-associated TLR9. Activation of TLR9 may govern B-cell selection and differentiation into both plasma and memory B cells.*

In sum, a more complete understanding of how BCR and non-BCR key signals integrate and modify the positive selection process of immature/transitional B cells is likely to provide valuable insights into HIV-1-triggered antibody responses and development of novel therapeutic interventions to elicit bnAbs.

Conflicts of interest

None.

Funding

None.

References

1. Alizon M, Wain-Hobson S, Montagnier L, Sonigo P. Genetic variability of the AIDS virus: nucleotide sequence analysis of two isolates from African patients. Cell. 1986;46:63-74.
2. Hraber P, Seaman MS, Bailer RT, et al. Prevalence of broadly neutralizing antibody responses during chronic HIV-1 infection. AIDS. 2014;28:163-9.
3. Mascola JR, Haynes BF. HIV-1 neutralizing antibodies: understanding nature's pathways. Immunol Rev. 2013;254:225-44.
4. Simek MD, Rida W, Priddy FH, et al. Human immunodeficiency virus Type 1 elite neutralizers: individuals with broad and potent neutralizing activity identified by using a high-throughput neutralization assay together with an analytical selection algorithm. J Virol. 2009;83:7337-48.
5. McCoy LE, Burton DR. Identification and specificity of broadly neutralizing antibodies against HIV. Immunol Rev. 2017;275:11-20.
6. Montero M, van Houten NE, Wang X, Scott JK. The membrane-proximal external region of the human immunodeficiency virus Type 1 envelope: dominant site of antibody neutralization and target for vaccine design. Microbiol Mol Biol Rev. 2008;72:54-84.
7. Ringel O, Müller K, Koch J, et al. Optimization of the EC26-2A4 epitope in the gp41 membrane proximal external region targeted by neutralizing antibodies from an elite controller. AIDS Res Hum Retroviruses. 2018;34:365-74.
8. Lee JH, Ozorowski G, Ward AB. Cryo-EM structure of a native, fully glycosylated, cleaved HIV-1 envelope trimer. Science. 2016;351:1043-8.
9. Haynes BF, Burton DR. Developing an HIV vaccine. Science. 2017;355:1129-30.
10. Kong L, Sattentau QJ. Antigenicity and immunogenicity in HIV-1 antibody-based vaccine design. J AIDS Clin Res. 2012;S8:3.
11. McCoy LE, Weiss RA. Neutralizing antibodies to HIV-1 induced by immunization. J Exp Med. 2013;210:209-23.
12. Yu L, Guan Y. Immunologic basis for long HCDR3s in broadly neutralizing antibodies against HIV-1. Front Immunol. 2014;5:250.
13. Burton DR, Hangartner L. Broadly neutralizing antibodies to HIV and their role in vaccine design. Annu Rev Immunol. 2016;34:635-59.
14. Kelsoe G, Haynes BF. Host controls of HIV broadly neutralizing antibody development. Immunol Rev. 2017;275:79-88.
15. Kepler TB, Liao HX, Alam SM, et al. Immunoglobulin gene insertions and deletions in the affinity maturation of HIV-1 broadly reactive neutralizing antibodies. Cell Host Microbe. 2014;16:304-13.
16. Simmonds P, Zhang LQ, McOmish F, et al. Discontinuous sequence change of human immunodeficiency virus (HIV) Type 1 env sequences in plasma viral and lymphocyte-associated proviral populations *in vivo*: implications for models of HIV pathogenesis. J Virol. 1991; 65:6266-76.
17. Ackerman M, Alter G. Mapping the journey to an HIV vaccine. N Engl J Med. 2013;369:389-91.
18. Metlas R, Veljkovic V, Paladini RD, Pongor S. Protein and DNA-sequence homologies between the V3-loop of human immunodeficiency virus Type I envelope protein gp120 and immunoglobulin variable regions. Biochem Biophys Res Commun. 1991;179:1056-62.
19. Metlas R, Trajkovic D, Srdic T, Veljkovic V, Colombatti A. Human immunodeficiency virus V3 peptide-reactive antibodies are present in normal HIV-negative sera. AIDS Res Hum Retroviruses. 1999;15:671-7.
20. Metlas R, Trajkovic D, Srdic T, Veljkovic V, Colombatti A. Anti-V3 and anti-igG antibodies of healthy individuals share complementarity structures. J Acquir Immune Defic Syndr. 1999;21:266-70.

21. Ovchinnikov V, Louveau JE, Barton JP, Karplus M, Chakraborty AK. Role of framework mutations and antibody flexibility in the evolution of broadly neutralizing antibodies. Elife. 2018;7:e33038.
22. McGuire AT, Glenn JA, Lippy A, Stamatatos L. Diverse recombinant HIV-1 envs fail to activate B cells expressing the germline B cell receptors of the broadly neutralizing anti-HIV-1 antibodies PG9 and 447-52D. J Virol. 2014;88:2645-57.
23. Irimia A, Sarkar A, Stanfield RL, Wilson IA. Crystallographic identification of lipid as an integral component of the epitope of HIV broadly neutralizing antibody 4E10. Immunity. 2016;44:21-31.
24. Ofek G, Tang M, Sambor A, et al. Structure and mechanistic analysis of the anti-human immunodeficiency virus Type 1 antibody 2F5 in complex with its gp41 epitope. J Virol. 2004;78:10724-37.
25. Pancera M, Changela A, Kwong PD. How HIV-1 entry mechanism and broadly neutralizing antibodies guide structure-based vaccine design. Curr Opin HIV AIDS. 2017;12:229-40.
26. Subbaraman H, Schanz M, Trkola A. Broadly neutralizing antibodies: what is needed to move from a rare event in HIV-1 infection to vaccine efficacy? Retrovirology. 2018;15:52.
27. Kleinman AJ, Sivanandham R, Pandrea I, Chougnet CA, Apetrei C. Regulatory T cells as potential targets for HIV cure research. Front Immunol. 2018;9:734.
28. Apellániz B, Nieva JL. The use of liposomes to shape epitope structure and modulate immunogenic responses of peptide vaccines against HIV MPER. Adv Protein Chem Struct Biol. 2015;99:15-54.
29. Soto C, Ofek G, Joyce MG, et al. Developmental pathway of the MPER-directed HIV-1-neutralizing antibody 10E8. PLoS One. 2016;11:e0157409.
30. Briney BS, Willis JR, Crowe JE Jr. Human peripheral blood antibodies with long HCDR3s are established primarily at original recombination using a limited subset of germline genes. PLoS One. 2012;7:e36750.
31. Buckner CM, Moir S, Ho J, et al. Characterization of plasmablasts in the blood of HIV-infected viremic individuals: evidence for nonspecific immune activation. J Virol. 2013;87:5800-11.
32. Malaspina A, Moir S, Ho J, et al. Appearance of immature/transitional B cells in HIV-infected individuals with advanced disease: correlation with increased IL-7. Proc Natl Acad Sci U S A. 2006;103:2262-7.
33. Giordano D, Draves KE, Young LB, et al. Protection of mice deficient in mature B cells from West Nile virus infection by passive and active immunization. PLoS Pathog. 2017;13:e1006743.
34. Kövesdi D, Pászty K, Enyedi A, et al. Antigen receptor-mediated signaling pathways in transitional immature B cells. Cell Signal. 2004;16:881-9.
35. Rawlings DJ, Metzler G, Wray-Dutra M, Jackson SW. Altered B cell signalling in autoimmunity. Nat Rev Immunol. 2017;17:421-36.
36. Aranburu A, Ceccarelli S, Giorda E, et al. TLR ligation triggers somatic hypermutation in transitional B cells inducing the generation of igM memory B cells. J Immunol. 2010;185:7293-301.
37. Kolhatkar NS, Brahmandam A, Thouvenel CD, et al. Altered BCR and TLR signals promote enhanced positive selection of autoreactive transitional B cells in Wiskott-Aldrich syndrome. J Exp Med. 2015;212:1663-77.
38. Tas JM, Mesin L, Pasqual G, et al. Visualizing antibody affinity maturation in germinal centers. Science. 2016;351:1048-54.
39. Wiehe K, Bradley T, Meyerhoff RR, et al. Functional relevance of improbable antibody mutations for HIV broadly neutralizing antibody development. Cell Host Microbe. 2018;23:759-65000000.
40. Mouquet H, Scheid JF, Zoller MJ, et al. Polyreactivity increases the apparent affinity of anti-HIV antibodies by heteroligation. Nature. 2010;467:591-5.
41. Moir S, Fauci AS. B-cell responses to HIV infection. Immunol Rev. 2017;275:33-48.
42. Metlas R, Srdic-Rajic T, Kohler H. Cooperation of intrathymic T15 idiotype-bearing B and complementary T cells in ontogeny of natural treg cells involved in establishment of T15 clonal dominance. Immunol Lett. 2019;210:52-4.
43. Van Regenmortel MH. Why does the molecular structure of broadly neutralizing monoclonal antibodies isolated from individuals infected with HIV-1 not inform the rational design of an HIV-1 vaccine? AIMS Public Health. 2015;2:183-93.