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Mini-Review

An Overview of Human Anti-HIV-1 Neutralizing Antibodies against Diverse Epitopes of HIV-1

Sanjeev Kumar,[$] Swarandeep Singh,[$] and Kalpana Luthra*

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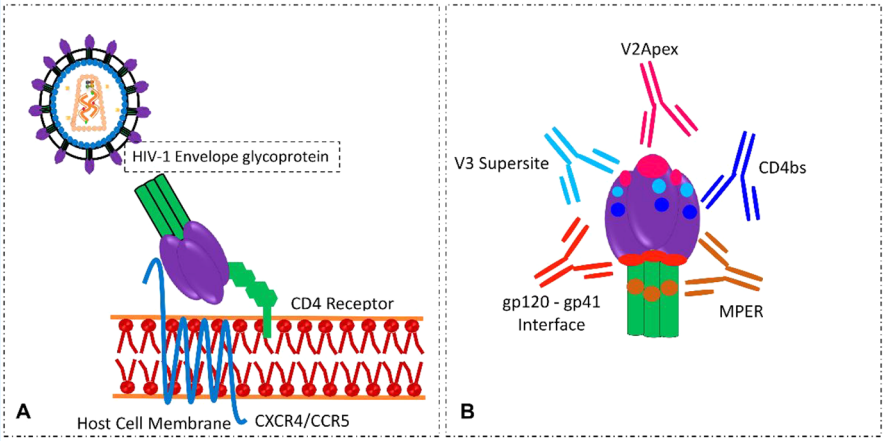

ABSTRACT: In this Review, we have addressed some recent developments in the discovery and applications of anti-human immunodeficiency virus type− 1 (HIV−1) broadly neutralizing antibodies (bnAbs) isolated from infected adults and children. The recent developments in human antibody isolation technologies have led to the discovery of several highly potent anti-HIV-1 bnAbs. Herein, we have discussed the characteristics of recently identified bnAbs directed at distinct epitopes of HIV-1, in addition to the existing antibodies, from adults and children and have shed light on the benefits of multispecific HIV-1 bnAbs and their role in the design of polyvalent vaccines.

INTRODUCTION

Since 1981, the HIV-1 pandemic has continued to infect millions of individuals, without any possible cure. Combined antiretroviral therapy (cART) is currently being given to treat the infected individuals. HIV-1 cART drugs are successful at reducing the viral load, improving quality of life, and delaying disease progression but cannot prevent HIV-1 infection or eliminate latent viruses.[1] To combat the continued surge in HIV-1 infection globally, effective therapeutic neutralizing antibodies (nAbs) and vaccines are of the utmost importance for protection and blocking further infection. About 10−25% of the HIV-1 infected individuals develop broadly neutralizing antibodies (bnAbs), of which less than 1% exhibit elite neutralizing activity (with ability to neutralize at least one virus of each of the four subtypes with 300 ID_{50} titre.[2] During natural infection, however, these bnAbs are often outnumbered by the circulating non−neutralizing antibodies, hence aviremic individuals are rare. In the past decade, several HIV-1 bnAbs have been discovered that are effective against a wide range of strains worldwide.[3−7] Such bnAbs preferentially target key semiconserved epitopes despite the high sequence variation of HIV-1 envelope sequences globally. Using the information on recognition determinants of these bnAbs and applying the Reverse Vaccinology 2.0 approach, researchers are putting intense effort into developing various types of HIV-1 vaccines to elicit potent bnAb responses.[8,9] Moreover, engineered HIV-1 bnAbs, with longer in vivo half-lives, are being explored for their potential usage as therapeutic and prophylactic clinical reagents.[10]

HIV-1 Entry and Envelope Structure. HIV-1 enters the host cells by the binding of its envelope glycoprotein gp160 to its primary receptor CD4, following which the conformational changes induced in the envelope lead to binding with two coreceptors CCR5 and CXCR4.[11] The HIV-1 envelope glycoprotein gp160 is a heavily glycosylated class 1 trimeric

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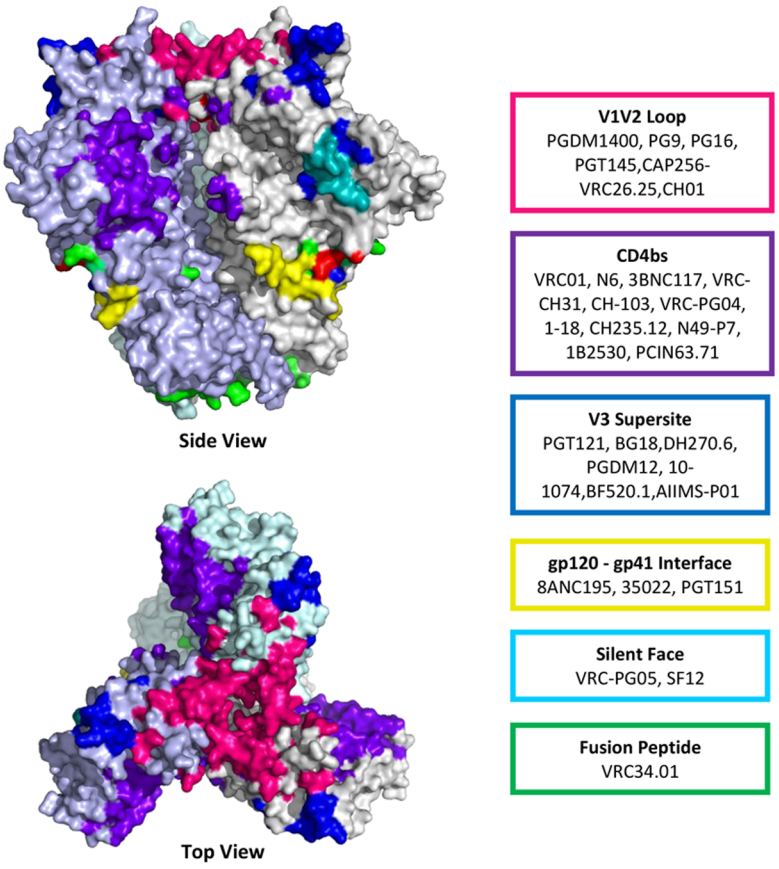

Figure 1. Representation of epitopes of HIV-1 broadly neutralizing antibodies on an envelope glycoprotein trimer. The surface representation of an HIV-1 envelope highlighting major HIV-1 bNAbs epitopes is derived from the crystal structure of BG505 SOSIP.664 (PDB ID. 4ZMJ). The structural figure was generated and highlighted with the program PyMOL (http://www.pymol.org/).

fusion glycoprotein and is a heterodimeric trimer composed of three protomers that are noncovalently associated as gp120 and gp41 monomers. The binding of gp120 to CD4 leads to the opening of the gp120 trimer due to a conformational change that enables the binding of coreceptors. This in turn leads to the insertion of the gp41 fusion peptide into the target cell membrane. Further, the gp41 glycoprotein folds into a hairpin structure, and its two heptad repeats, HR1 and HR2, form a six-helix bundle that brings the host and viral cell membranes into close to complete fusion, resulting in entry of the ribonucleocapsid protein carrying the viral RNA genome into the host cell. HIV-1 gp120 comprises of five variable (V1−V5) and five constant (C1−C5) regions.[11] gp41 domain comprises of the fusion peptide, HR1, HR2, and membrane proximal external region (MPER). Once infected with HIV-1, antibodies are generated against all viral proteins of HIV-1; however, nAbs are developed primarily against the envelope glycoproteins gp120 and gp41. The HIV-1 bnAbs can neutralize the virus by recognizing and blocking the viral entry steps and the gp120 or gp41 regions.

Targets of HIV-1 Neutralizing Antibodies. In 1994, a CD4 binding site (CD4bs) directed antibody b12 was the first HIV-1 bnAb isolated from an asymptomatic HIV-1-infected individual using the phage display technology.[3] Since 2009, with immense efforts from scientists worldwide and technology advancement in the discovery of human monoclonal antibodies, a large panel of second-generation potent HIV-1 bnAbs have been isolated and characterized.[3] These bnAbs primarily target the following seven different conserved epitopes present on the HIV-1 envelope: V2−glycan apex, V3−glycan, CD4bs, gp120−41 interface, MPER, fusion peptide, and silent-face center (Figure 1). The major bnAbs targeting these epitopes are discussed and highlighted in Table 1.

V2−Glycan Apex Epitope-Targeting bnAbs. V2 apex-targeting bnAbs are most potent class of antibodies, with a GMT average of 0.3 μg/mL and coverage of 71% of the global isolates of the HIV-1.[3] The V2−glycan apex epitope includes N160 glycan and a lysine-rich apex region from amino acid position K168 to K171. The key HIV-1 bnAbs that target this V2 apex are PG9, PG16, PGDM1400, CAP256.25, PCT64, and CH01.[3,12] In comparison to bnAbs that target other HIV-1 envelope regions, the V2 apex-directed bnAbs exhibit relatively rare features such as a characteristically long negatively charged CDRH3 region enriched with sulfated Tyr residues and a YYD motif that interacts with the lysine residues of the V2 apex.[3,12] The long CDRH3 loops of V2−glycan bnAbs possess β-hairpin or hammerhead conformations that penetrate between V2−glycans and are typically around 24−36 amino acids longer than those found in V3−glycan bnAbs. A study from sub-Saharan Africa reported that following V3−glycan bnAbs, the V2−apex bnAb precursors were present in 14% of individuals.[13] In our cohort of HIV-1-infected adolescents and infants, we have observed that the earliest bnAb responses are targeted to the V2 region and continue to persist, as observed on longitudinal follow-up of a pair of HIV-1-infected identical twin children.[14,15] V2−apex bnAbs are commonly

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Table 1. Characteristics of Select Anti-HIV-1 Broadly Neutralizing Antibodies

S.No.	Antibody	Envelope binding Site	Neutralization Breath	Potency (μg/ml)	Heavy V (IGHV)	Light V (IGKV or IGLV)	CDRH3 Length (a.a.)	CDRL3 Length (a.a.)	VH mutation Frequency (%)	VL mutation Frequency (%)	References
1	PGDM1400	V1/V2 Loop	83%	0.003	IGHV1-8	IGKV2-28	34	9	26.4	11.8	PMC4267403
2	PG9	V1/V2 Loop	84%	0.118	HV3-33*05	λV2-14*01	28	11	12.6	6.3	PMC3335270
3	PG16	V1/V2 Loop	75%	0.02	IGHV3-33*05	IGLV2-14*01	28	11	13.1	12.2	PMC3335270
4	PGT145	V1/V2 Loop	78%	0.29	IGHV1-8*01	IGHLV2-28*01/2D-28*01	31	9	16.2	15.5	PMC3393110
5	CAP256-VRC26.25	V1/V2 Loop	59%	0.001	VH3-30*18	λV1-51*02	38	12	12.2	8.6	PMC4702551
6	CH01	V1/V2 Loop	52%	1.38	VH3–20	VK3–20	24	9	16.7	11.2	PMC3196428
7	PGT121	V3 glycan	66%	0.048	HV4-59*01	λV3-21*01	26	12	19.6	16.5	PMC3393110
8	BG18	V3 glycan	62%	0.032	HV4-4*02	λV3-25*03	23	11	21.5	17.6	PMC7114535
9	DH270.6	V3 glycan	57%	0.151	HV1-2*02	λV2-23*02	20	10	12.8	6.7	PMC4990068
10	BF520.1	V3 glycan	53% *	7.31	VH1–2	VK3–15	18	11	6.6	5.3	PMC4930401
11	AIIMS P01	V3 glycan	67% *	0.26	IGHV4-59	IGLV1-47	19	11	7	5	PMC6364018
12	10-1074	V3 glycan	68%	0.039	HV4-59*01	λV3-21*01	26	12	26.7	30.1	PMC3511153
13	PGDM12	V3 glycan	54%	0.14	VH3–11	VK2–24	19	9	19.1	14.3	PMC4990068
14	PGT128	V3 glycan	68%	0.064	VH4–39	VL2–8	19	10	19.1	7	PMC3393110
15	N6	CD4bs	98%	0.058	HV1-2*02	κV1-33*01	15	5	30.2	22.4	PMC5770152
16	VRC01	CD4bs	91%	0.329	HV1-2*02	κV3-20*01	14	5	31.6	17.2	PMC2965066
17	CH235.12	CD4bs	90%	0.65	HV1-46*01	κV3-15*01	15	8	25	14.8	PMC4826291
18	1-18	CD4bs	97%	0.048	HV1-46*01	kV3-20*01	18	9	26.4	20.2	PMC7042716
19	N49-P7	CD4bs	100%	0.10	HV1-2*02	λV2-11*01	21	5	24.5	14.1	PMC6003858
20	NC-Cow1	CD4bs	72%	0.028	HV1-7*01	–	62	–	–	–	PMC5812458
21	12A12	CD4bs	93%	0.221	VH1–2	VK1–33	13	5	21.9	15.5	PMC3351836
22	3BNC117	CD4bs	89%	0.116	VH1–2	VK1–33	10	5	23.7	14.8	PMC3351836
23	1B2530	CD4bs	72%	3.62	VH1–46	VL1–47	16	11	27.8	15.7	PMC3351836
24	VRC-CH31	CD4bs	84%	0.321	VH1–2	VK1–33	13	5	20.2	15.2	PMC3516815
25	CH103	CD4bs	67%	2.28	VH4–59	VL3–1	13	10	16.9	11.1	PMC3637846
26	VRC-PG04	CD4bs	81%	0.317	VH1–2	VK3–40	14	5	28.6	15.2	PMC3516815
27	VRC-PG20	CD4bs	80%	0.226	VH1–2	VL2–14	13	5	24	14.8	PMC3985390
28	8ANC131	CD4bs	71%	1.78	VH1–46	VK3–20	16	9	25.7	17.2	PMC4683157
29	PCIN63.71I	CD4bs	84%	0.46	VH1–2	VK1–6	13	5	14.6	12.5	PMC6642152
30	PGT151	Interface	72%	0.023	HV3-30*03	κV2D-29*02	28	9	20.8	11.5	PMC4070425
31	8ANC195	Interface	66%	1.115	HV1-3*03	κV1-5*03	22	9	27.2	14.9	PMC3351836
32	35O22	Interface	56%	0.151	HV1-18*03	λV2-14*02	16	10	21.9	22.4	PMC4224615
33	10E8	MPER	98%	0.299	HV3-15*05	λV3-19*01	22	12	21.4	13.4	PMC4854285
34	4E10	MPER	98%	1.765	HV1-69*17	κV3-20*01	20	9	6.9	4.1	PMC7520721
35	DH511.2	MPER	99%	1	HV3-15*05	λV3-19*01	24	12	19.8	14	PMC5905719
36	PGZL1	MPER	84%	6.06	VH1–69	VK3–20	15	9	20.9	11.8	PMC6879610
37	VRC42.1	MPER	96%	4.09	VH1–69	VK3–20	15	9	10.8	5.6	PMC6555550
38	VRC43.1	MPER	63%	1.34	VH4–4	VL7–43	19	9	11.1	8.5	PMC6555550
39	2F5	MPER	58%	2.83	VH2–5	VK1–13	22	9	13.1	11	PMC238102
40	VRC-PG05	Silent Face	53%	3.714	HV3-7*01	κV4-1*01	17	8	9	6	PMC6421865
41	SF12	Silent Face	62%	–	H4-59*01	(K3-20*01	23	6	17	14.6	PMC6591006
42	N123-VRC34.01	Fusion Peptide	73%	0.359	HV1-2*02	kv1-9*01	13	9	15	10	PMC4917739

encoded by IGHV3−30/33 and IGHV1−8 antibody genes.[3,12] The most potent antibody described so far, CAP256-VRC26.25, is directed at the V2 apex region, with a geometric mean IC_{50} of 1 ng/mL and a breath of 59% against a large

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panel of global isolates of HIV-1.[16] Resolution of the cryo-EM structure of the trimer apex-directed antibody PGT145 along with the envelope trimer and further the affinity of the antibody to the SOSIP.664 envelope trimer in nanomolar concentrations led to the development of PGT145 immunoaffinity chromatography for the purification of stabilized envelope trimers.[17] V2−apex bnAbs were also identified in SHIV-infected macaques, suggesting that their precursor B cells can be steered to elicit such bnAbs.[18]

V3-Glycan-Targeting bnAbs. The most common bnAbs found in HIV-1 infection belong to the V3 supersite.[3,12] The V3−glycan epitope is located between N301 and N332 glycans of the HIV-1 envelope at the base of V3−loop near the GDIR motif (324−327).[19] The V3−glycan-dependent bnAbs are primarily encoded by multiple heavy chain genes (IGHV4−59, IGHV4−39, IGHV4−4, and IGHV1−2). The V3−glycan bnAbs exhibit long CDRH3 regions (18−24 amino acids), often seen with presence of indels in CDRH2 or CDRL1 regions, and high levels of rare somatic hypermutations (SHMs), which are required for HIV-1 neutralization.[3] Like the very long CDRH3 lengths of V2 apex bnAbs, the precursors of V3−glycan bnAbs with long CDRH3 regions are rare. For instance, V3-glycan precursors of the BG18 bnAb B cell lineage have been found to be present at a very low frequency of only 1 in 53 million.[20] V3 supersite-targeting bnAbs show a modest breadth and potency of 71% and 1 μg/mL, respectively.

CD4bs. CD4bs-targeting antibodies typically take longer to develop in natural infection as compared to any other antibody specific to the envelope region and acquire, on an average, the highest level of SHM.[3,12,21] These bnAbs show the highest breath of average 84% among all known bnAbs that target the HIV-1 envelope, with an average GMT of 0.87 μg/mL.[21,22] The CD4bs epitope is a highly conformational and discontinuous epitope. HIV-1 bnAbs that recognize the CD4bs epitope are of two types: CD4 mimetics (recognize CD4 contact residues of the CD4bs epitope by their CDRH2), e.g., VRC01-like bnAbs,[23] and CDRH3 binders (recognize the CD4bs epitope via their CDRH3), e.g., CH103.[24] The characteristic features of VRC01-like bnAbs are frequent IGHV1−2 heavy chain gene usage, common precursors, high levels of somatic hypermutations (∼40%), and the presence of a five amino acid CDRL3 region. VRC01-like bnAbs are most potent and exhibit the highest breadth (90−100%) in comparison to other HIV-1 bnAbs.[23,25] Another CD4bs bnAb N6 potently neutralized 98% of HIV-1 strains, including 16 of 20 that were resistant to VRC01 and others.[3,22] The structural analysis of N6 revealed that it evolved by avoiding steric clashes with glycans, which is a common mechanism of resistance.[22] The CD4 mimetic bnAbs (like the VRC01 and 8ANC31/CH235 class of antibodies) dominantly use the IGHV1−46 heavy chain gene and exhibit the absence of the 5 AA CDRL3 region, 70−97% neutralization breadth, and a high level of SHM (30−40%), while CDRH3 binders like the CH103 class of antibodies primarily use IGHV4−59, with a neutralization breath of around 70%.[24,26] The most unusual bnAbs to the CD4bs, generated by vaccination in cows, have ultralong CDRH3s of up to 70 residues.[27]

Silent-Face Center. This new epitope was identified more recently than the others. It is located on a glycosylated region including N262, N295, and N448 glycans across the CD4bs region. VRC-PG05 and SF12 are the only known HIV-1 bnAbs that target this epitope.[28,29] These bnAbs neutralize HIV-1 by inhibiting the conformational changes required for receptor binding and viral entry. Both SF12 and VRC-PG05 recognize all the three glycans of the silent-face center epitope. The SF12 bnAb is encoded by IGHV4−59*01 and IGKV3−20*01, whereas VRC-PG05 is encoded byIGHV3−7*01 and IGKV4−1*01 antibody heavy and light chain genes, respectively.

Gp120−41 Interface. This conformational epitope is located at the lower region of the gp120 near the gp120−gp41 interface region. Only few HIV-1 bnAbs like PGT151, 8ANC195 and 35022 targeting this epitope have been isolated from infected donors thus far. Few HIV-1 bnAbs (e.g., PGT151) targeting this epitope are trimer specific i.e. these bnAbs preferentially recognize the trimeric conformation of the gp160 over monomeric gp120 protein.[3,30] HIV-1 bnAbs targeting this region neutralize virus by various mechanisms e.g., 3BC176 and 3BC315 can destabilize the trimer whereas 8ANC195, can inhibit the conformational changes needed for membrane fusion. Interface targeting bnAbs demonstrates an average breath of 64% with a mean IC50 of 0.42 μg/mL.

Fusion Peptide. The fusion peptide epitope is present at the N-terminal region of the gp41 region. This epitope is largely conformational and hydrophobic due to its interaction with the host cell membrane during fusion events of HIV-1 entry.[3,31] Due to the proximity to heterogeneous complex glycans on gp41, FP-targeting antibodies are able to neutralize the small subset of viruses. HIV-1 bnAbs that target this epitope neutralize the virus by preventing the fusion of viral and host cell membranes. VRC34 and ACS202 are the key bnAbs that target the fusion peptide region. Their HIV-1 bindin and neutralizing mechanisms are similar to those of the gp120−gp41 interface HIV-1 bnAbs.

Membrane Proximal External Region (MPER) Epitope. MPER bnAbs are typically very broad neutralizers with an average coverage of 85% but have a moderate potency around 2.4 μg/mL.[3,20,32] This hydrophobic, linear epitope is in the gp41 region. HIV-1 bnAbs that target this region exhibit high neutralization breadth (>95%) with low to moderate potency (0.3−5 ug/mL) and a high level of polyreactivity. The 10E8 and DH511 bnAbs are among the most potent ones (∼99% of breadth) that are encoded by IGHV3−15 of the IgG3 isotype, with a hydrophobic 24 amino acid CDRH3 region and high levels of SHM. Others MPER bnAbs, namely, PGZL1, VRC42.1 and 4E10, are encoded by IGHV1−69, which pairs with IGKV3−20 light chains, and exhibit high levels of SHM.

HIV-1 Neutralizing Antibody Responses in Infected Children. The HIV-1 bnAbs isolated from chronically infected adults exhibit signature characteristic features of high somatic hypermutations (SHMs), insertions or deletions (indels), long complementarity-determining region H3 (CDRH3), high potency, and broad viral neutralization breadth.[3] It takes at least 2−3 years of infection in adults for bnAbs to evolve.[2,33,34] HIV-1 infection in children is mostly caused by vertical transmission. The immune system is not fully developed in infants in terms of both innate and acquired immune responses. HIV-1 disease progression is faster and more severe in children as compared to adults. HIV-1-infected infants have been shown to produce de novo anti-HIV-1 neutralizing antibodies at an early age. Further, plasma bnAbs with multiepitope specificities have been shown to evolve in such infants. Plasma mapping revealed that HIV-1 bnAbs can develop early in life and that functional B cells persist in these infants to produce bnAbs irrespective of high viremia and the faster disease progression compared to that in adults.[35] The

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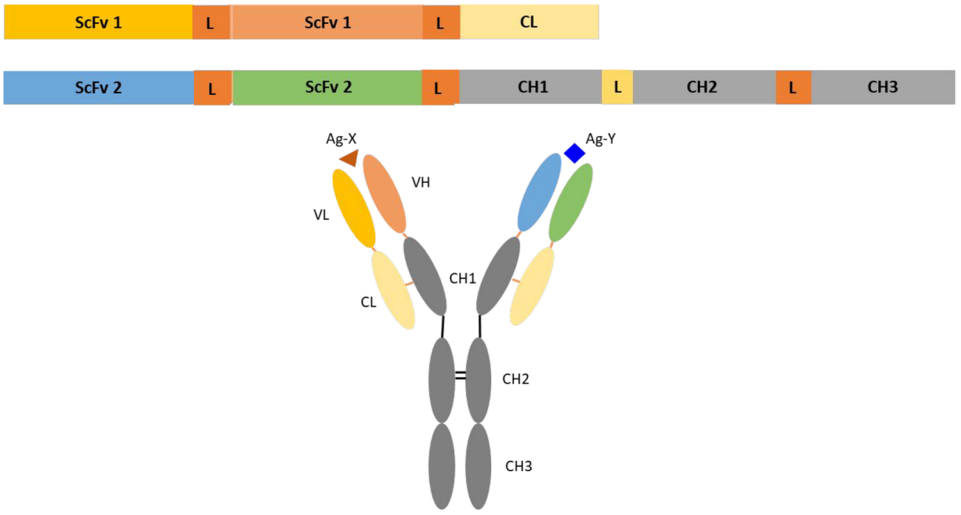

Figure 2. Representation of bispecific antibody design. Bispecific antibodies are the engineered antibodies capable of recognizing the two epitopes of an antigen or two different antigens. The design involves the antigen binding sites of two or more antibodies attached through a flexible linker without altering the constant regions (CH1, CH2, and CH3). ScFv 1 = single-chain variable fragment 1, ScFv 2 = single-chain variable fragment 2, CL = constant light, CH = constant heavy, L = flexible linker, and Ag = antigen.

high viral load, in both infants and adults, plausibly promotes the development of nAb breadth. Our group and others have reported that plasma bnAb responses evolve over time in HIV-1-infected children.[1,6,14,15,35−40] HIV-1 plasma bnAbs, in both adults and children, have been found to target multiple epitopes, including V1V2, V3−glycan N332, CD4bs, and MPER.[6,14,15,35−37] Our plasma mapping studies showed that the earliest bnAb responses in elite and broad neutralizers in the cohort of recruited Indian HIV-1 clade C-infected infants and children are against the V1V2 apex region; moreover, these antibodies continued to persist in a pair of chronically infected antiretroviral naïve monozygotic twin pediatric elite-neutralizers that were longitudinally evaluated.[14,15] In addition to V1V2 responses in chronically infected children, we also reported the presence and evolution of plasma V3−glycan and CD4bs bnAbs, supporting a polyclonal vaccine design. Thus far, only two HIV-1 bnAbs (BF520.1 and AIIMS-P01) have been identified from HIV-1-infected children.[5,41] BF520.1, one of the HIV-1 N332 supersite-dependent nAbs isolated from an infant at 1-year p.i., has shown cross neutralizing activity despite limited SHMs and an absence of indels, unlike the bnAbs isolated from adults, suggesting that infant bnAbs evolved by different pathways than adult bnAbs.[41] Another HIV-1 N332-targeted bnAb AIIMS-P01, discovered by us from a singular pediatric elite neutralizer, showed a 67% HIV-1 neutralization breadth despite low somatic hypermutations and exhibited indels.[5] In future, there is a need to isolate more HIV-1 bnAbs that target multiple epitopes from infected infants and children to understand their defining characteristics and the mechanisms by which they evolve in children, which can guide effective vaccine design.

Multispecific HIV-1 Neutralizing Antibodies. Due to the complexity and high diversity of the HIV-1 envelope, no single bnAb is able to neutralize all the viral strains. A combination of two or more bnAbs as a cocktail can synergistically improve broad coverage against most viruses by lowering the risk of emerging escape mutants. The manufacturing and testing of two or three independent bnAbs are time-consuming, costly, and resource intensive. This has led to the generation of multispecific antibody formats that can incorporate multiple different bnAbs with distinct epitope specificities into one molecule. Several HIV-1 bispecific, trispecific, and multibody bnAbs have been developed by combining the antibody chains of two or three nonoverlapping epitope-targeting bnAbs[10,42,43] (Figure 2). These multispecific HIV-1 bnAbs can effectively neutralize near-pan viral strains and have been shown to be protective in animal models. A study conducted by Mascola et al. demonstrated that the VRC07 and PG9−16 bispecific antibody showed superiority to parental bnAbs in terms of potency and breadth by neutralizing 97% of viruses with an IC_{50} of 0.055 μg/mL.[43] Moshoette et al. reported an engineered bispecific bnAb (bibnAb), an iMab-N6 comprised of the N6 HIV-1 spike-targeting bnAb and ibalizumab (iMab), a host CD4-targeting antibody. The iMab-N6 exhibited a pan-neutralization breadth of 100% coverage with 21 pseudoviruses tested including the global panel.[44] A trispecific bnAb (VRC01/PGDM1400−10E8v4, CODV-Ig) with three independent HIV-1 envelope determinants (CD4bs, MPER, and V1V2 apex) exhibited higher potency and breadth than any previously described single bnAb and conferred complete immunity against several simian-human immunodeficiency viruses (SHIVs) in nonhuman primates as compared to single bnAbs.[42] Recently, Julien et al. successfully demonstrated the design of novel multispecific HIV-1 bnAbs based on a protein nanoparticle format displaying multiple epitopes. These multispecific bnAbs (T-01 MB.v2) showed a remarkable ultrapotent median IC_{50} value of 0.0009 μg/mL and 100% HIV-1 neutralization coverage against a broad HIV-1 pseudovirus panel of 118 isolates,[10] suggesting that multivalent and multispecific HIV-1 bnAbs could be the promising next-generation cost-effective therapeutics against diverse strains of HIV-1.

Role of HIV-1 bnAbs in Vaccine Design. Based on the structural information on the currently available HIV-1 bnAbs, attempts are ongoing to design immunogens that can elicit correlating protection by vaccinations. The neutralizing epitopes defined by HIV-1 bnAbs are being used for vaccine design by applying the Reverse Vaccinology 2.0 strategy.[8,45,46] Current vaccine design approaches seek to trigger rare B cell

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precursors and then steer affinity maturation toward the development of bnAbs in a multistage multicomponent immunization approach.[47] Using structural information on the CD4bs bnAb VRC01, Schief et.al. designed and developed a VRC01 germline-targeting nanoparticle vaccine candidate eOD-GT8 60-mer to prime and elicit VRC01 like bnAbs.[48] Emerging data from the first human clinical trial of the eOD-GT8 vaccine suggests that this vaccine can successfully prime and steer the VRC01 bnAb precursors in humans.[49] Schief et al. also designed N332 bnAb germline-targeting SOSIP trimers to steer PGT121 and BG18 bnAb precursor B cells to elicit such bnAbs upon vaccination.[50,51] The HIV-1 SOSIP trimers developed by Sanders et.al. are excellent antigenic native mimics of the virion-associated HIV-1 envelope, which is inclusive of almost all the known bnAb epitopes, and thus serve as suitable frameworks for vaccine design.[45] Further they have developed VRC01 germline-targeting SOSIP.GT1 trimers to steer VRC01 and other bnAb precursor B cells to elicit VRC01-like bnAbs in the vaccinees.[46] Recently, a SOSIP-based V1V2 germline-targeting vaccine candidate MT145 K was designed by Andrabi et al. that successfully elicited V2 apex antibodies in mice immunizations.[52]

CONCLUSIONS AND FUTURE PERSPECTIVES

Herein we have highlighted the HIV-1 bnAbs identified from adults and children and have shed light on the benefits of multispecific HIV-1 bnAbs and their role in vaccine design. HIV-1 bnAb-based therapeutics are a potential strategy for immediate treatment/prophylaxis or in situations where vaccines are less effective and those involving vaccine hesitancy, unvaccinated infants, children, and immune-compromised individuals. However, the ongoing COVID-19 pandemic has demonstrated that despite a number of mAbs being approved for the treatment of COVID-19, the SARS-CoV-2 virus has shown resistance to most therapeutic mAbs.[53−55] Moreover, treating a disease/infection using monoclonal antibodies (mAbs) is very expensive relative to antiretrovirals and antibacterials. There are limitations with the production of mAbs; protein mAbs require cold-chain storage and transport, in addition to costs of manufacturing and distribution. Few of these hurdles can now be overcome through nonviral synthetic plasmid DNA and mRNA vectors that have been developed to encode optimized mAb genes for in vivo delivery. Developing such therapeutics/prophylactics can eliminate many of the steps involved in bioprocesses, cold-storage, and high production costs.[56,57] In case of HIV-1, to overcome resistance to antiretroviral drugs and develop effective clinical reagents, it is critical to develop a cocktail of bnAbs for HIV-1 therapeutic and prophylactic purposes with increased in vivo half-lifes to be used alone or in combination with ART. The multispecific single-molecule approach displaying two or more HIV-1 bnAbs could be more effective and cost-effective for HIV-1 treatment purposes, especially for low- and middle-income countries (LMICs).

The field of HIV-1 bnAb isolation and vaccine design is continuing to evolve; however, there is still a paucity of information on understanding the HIV-1 bnAb responses at the cellular level. Though studies based on humoral responses have shown that children elicit HIV-1 bnAbs that target multiple epitope specificities, and only two HIV-1 nAbs, both targeting the N332 epitope, have been reported thus far.[5,41] To achieve the goal of a universal and globally effective HIV-1 vaccine suitable for both adults and children, it is necessary to discover more HIV-1 bnAbs that target multiple epitopes from infants/children in addition to the ones from adults to understand their characteristic features. Solving their high-resolution structures can provide useful footprints for HIV-1 vaccine design. A high-throughput antibody discovery technology has been developed by Ward et al.[58] based on cryo-EM based EMPEM and high-resolution structural analysis for novel monoclonal antibody discovery that can further determine detailed structure- and sequence-based information from the pediatric and adult bnAbs of multiple-epitope specificities. Such advanced techniques have benefits over previously used high-throughput phage display technology-based HIV-1 bnAb isolation from random mutant libraries based on recombinant antibodies. However, cryo-EM based techniques, like single B cell sorting technologies, can identify the naturally occurring true heavy an light chain pairing information on a bnAb, which is critical for effective vaccine design based on naturally evolved HIV-1 bnAbs.[20,47] For epitope mapping, the high-resolution based cryo-EM analysis are interesting approaches, but they can have limited resolution. To overcome these limitations, epitope mapping using high-throughput point mutant libraries (shotgun mutagenesis or alanine scanning) can provide near-single-residue resolution to specifically identify recognition/neutralizing determinants of select mAbs.[59,60]

Germline targeting is an interesting approach to developing a vaccine for a very complex HIV-1 virus. The recent human clinical trial of VRC01 germline targeting vaccine candidate eOD-GT8 showed the priming of VRC01 bnAb precursors without any development of VRC01-like bnAbs upon vaccination.[49] Despite years of work, it has still to show proper proof of principle, and its complexity is a serious barrier to real-world implementation. Perhaps the CD4bs-targeted HIV-1 bnAbs have a very high number of SHMs, thereby making them difficult to elicit upon vaccination.[3] It is known from infected adults and children that the early HIV-1 bnAb responses are directed against the V2 apex and V3−glycan region, and it may therefore be effective to coprime various epitope-specific HIV-1 bnAb precursors using their germline-targeting vaccine candidates, adopting a polyvalent strategy.[14,35,41] For example, a cocktail of eOD-GT8 60-mer, BG505.GT1 trimers to steer VRC01 precursors, MD39 for PGT121, and MT145K for V2 apex bnAbs precursors may potentiate the elicitation of HIV-1 bnAbs upon immunization. Our recent studies on HIV-1-infected infants and children have provided key evidence for exploring polyvalent vaccination strategies in the future to prevent/block HIV-1 infection.[14,15]

AUTHOR INFORMATION

Corresponding Author

Kalpana Luthra − *Department of Biochemistry, All India Institute of Medical Sciences, New Delhi, Delhi 110029, India*; orcid.org/0000-0002-7801-2867; Email: kalpanaluthra@gmail.com

Authors

Sanjeev Kumar − *Department of Biochemistry, All India Institute of Medical Sciences, New Delhi, Delhi 110029, India*; orcid.org/0000-0002-8181-3999

Swarandeep Singh − *Department of Biochemistry, All India Institute of Medical Sciences, New Delhi, Delhi 110029, India*

Complete contact information is available at: https://pubs.acs.org/10.1021/acsomega.2c07933

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Author Contributions

Conceptualization: S.K., S.S., and K.L. Writing original draft: S.K. and S.S. Reviewing, editing, and finalization of the manuscript: K.L. All authors contributed to the article and approved the submitted version.

Author Contributions

[$]Co-first authors/equal contribution.

Notes

The authors declare no competing financial interest.

Biographies

Sanjeev Kumar

Dr. Sanjeev Kumar has completed his Ph.D. under the supervision of Prof. Kalpana Luthra, All India Institute of Medical Sciences, New Delhi. His primary research expertise is viral immunology with a focus on the immune response of B cells, neutralizing antibodies, and rational vaccine design. Presently, he is working as a DBT/Wellcome Trust India Alliance Early Career Fellow at Emory Vaccine Center, International Centre for Genetic Engineering and Biotechnology (ICGEB), India, and as a Visiting Fellow at the Emory Vaccine Center, Emory University School of Medicine, Atlanta, GA, USA.

Swarandeep Singh

Swarandeep Singh is registered to All India Institute of Medical Sciences New Delhi under the chief supervision of Dr. Kalpana Luthra as a Ph.D. student. His research is primarily focused on rational vaccine design and anti-HIV-1 monoclonal antibodies generation.

Kalpana Luthra

Dr. Kalpana Luthra is a Professor in the Department of Biochemistry at AIIMS, New Delhi, India. Her lab is actively working in the field of HIV-1 Immunology to understand the coevolution of HIV-1 and antibody responses during natural infection and the generation of recombinant human anti-HIV-1 monoclonal antibodies.

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