VARIANT NUCLEIC ACID LIBRARIES FOR CORONAVIRUS

CROSS-REFERENCE

[0001] This application claims the benefit of U.S. Provisional Patent Application No. 63/115,568 filed on November 18, 2020, U.S. Provisional Patent Application No. 63/104,465 filed on October 22, 2020, U.S. Provisional Patent Application No. 63/073,362 filed on September 1, 2020, U.S. Provisional Patent Application No. 63/069,665 filed on August 24, 2020, U.S. Provisional Patent Application No. 63/034,896 filed on June 4, 2020, U.S. Provisional Patent Application No. 63/016,254 filed on April 27, 2020, each of which is incorporated by reference in its entirety.

BACKGROUND

[0002] Coronaviruses like severe acute respiratory coronavirus 2 (SARS-CoV-2) can cause severe respiratory problems. Therapies are needed for treating and preventing viral infection caused by coronaviruses like SARS-CoV-2. Antibodies possess the capability to bind with high specificity and affinity to biological targets. However, the design of therapeutic antibodies is challenging due to balancing of immunological effects with efficacy. Thus, there is a need to develop compositions and methods for the optimization of antibody properties in order to develop effective therapies for treating coronavirus infections.

INCORPORATION BY REFERENCE

[0003] All publications, patents, and patent applications mentioned in this specification are herein incorporated by reference to the same extent as if each individual publication, patent, or patent application was specifically and individually indicated to be incorporated by reference.

~~BRIEF~~ SUMMARY

[0003a] In one aspect, provided herein is an antibody or antibody fragment comprising a variable domain, heavy chain region (VH) and a variable domain, light chain region (VL), wherein VH comprises complementarity determining regions CDRH1, CDRH2, and CDRH3, wherein VL comprises complementarity determining regions CDRL1, CDRL2, and CDRL3, and wherein (a) an amino acid sequence of CDRH1 is as set forth in any one of SEQ ID NOs: 241-255 and 547-575; (b) an amino acid sequence of CDRH2 is as set forth in any one of SEQ ID NOs: 256-270 and 576-604; (c) an amino acid sequence of CDRH3 is as set forth in any one of SEQ ID NOs: 271-285 and 605-633; (d) an amino acid sequence of CDRL1 is as set forth in any one of SEQ ID NOs: 286-300 and 634-662; (e) an amino acid sequence of CDRL2 is as set forth in any one of SEQ ID NOs: 301-315 and 663-691; and (f) an amino acid sequence of CDRL3 is as set forth in any one of SEQ ID NOs: 316-330 and 692-720.

[0003b] In another aspect, provided herein is an antibody or antibody fragment comprising a variable domain, heavy chain region (VH) wherein VH comprises complementarity determining regions CDRH1, CDRH2, and CDRH3, wherein (a) an amino acid sequence of CDRH1 is as set forth in any one of SEQ ID

NOs: 1-50 and 779-919; (b) an amino acid sequence of CDRH2 is as set forth in any one of SEQ ID NOs: 51-100 and 920-1060; and (c) an amino acid sequence of CDRH3 is as set forth in any one of SEQ ID NOs: 101-150 and 1062-1202.

[0003c] In another aspect, provided herein is an antibody or antibody fragment comprising a variable domain, heavy chain region (VH) and a variable domain, light chain region (VL), wherein the VH comprises an amino acid sequence at least about 90% identical to a sequence as set forth in any one of SEQ ID NOs: 721-749, 2104-2118, and 2134-2147, and wherein the VL comprises an amino acid sequence at least about 90% identical to a sequence as set forth in any one of SEQ ID NOs: 750-778, 2119-2133, and 2148-2161.

[0003d] In another aspect, provided herein is an antibody or antibody fragment comprising a variable domain, heavy chain region (VH) comprising an amino acid sequence at least about 90% identical to a sequence as set forth in any one of SEQ ID NOs: 1203-1343 and 2162-2301.

[0003e] In another aspect, provided herein is a nucleic acid composition comprising: a) a first nucleic acid encoding a variable domain, heavy chain region (VH) comprising an amino acid sequence at least about 90% identical to a sequence as set forth in any one of SEQ ID NOs: 721-749, 2104-2118, and 2134-2147; b) a second nucleic acid encoding a variable domain, light chain region (VL) comprising at least about 90% identical to a sequence as set forth in any one of SEQ ID NOs: 750-778, 2119-2133, and 2148-2161; and an excipient.

[0003f] In another aspect, provided herein is a nucleic acid composition comprising: a) a first nucleic acid encoding a variable domain, heavy chain region (VH) comprising an amino acid sequence at least about 90% identical to a sequence as set forth in any one of SEQ ID NOs: 1203-1343 and 2162-2301; and b) an excipient.

[0003g] In another aspect, provided herein is a use of the antibody or antibody fragment disclosed herein in the manufacture of a medicament for the treatment of a SARS-CoV-2 infection.

[0003h] In another aspect, provided herein is the antibody or antibody fragment disclosed herein for use in therapy.

[0004] Provided herein are nucleic acid libraries comprising: a plurality of sequences that when translated encode for antibodies or antibody fragments that bind to SARS-CoV-2 or ACE2 protein, wherein each of the sequences comprises a predetermined number of variants within a CDR relative to an input sequence that encodes an antibody, and wherein the library comprises at least 50,000 variant sequences. Further provided herein are nucleic acid libraries, wherein the antibodies or antibody fragments bind to a spike glycoprotein, a membrane protein, an envelope protein, a nucleocapsid protein, or combinations thereof of the SARS-CoV-2. Further provided herein are nucleic acid libraries, wherein the antibodies or antibody fragments bind to a spike glycoprotein. Further provided herein are nucleic acid libraries, wherein the antibodies or antibody fragment bind to a receptor binding domain of the spike glycoprotein. Further provided herein are nucleic acid libraries, wherein the library comprises at least 100,000 variant sequences. Further provided herein are nucleic acid libraries, wherein at least some of the sequences encode for an

antibody light chain. Further provided herein are nucleic acid libraries, wherein at least some of the sequences encode for an antibody heavy chain. Further provided herein are nucleic acid libraries, wherein

CLAIMS

WHAT IS CLAIMED IS:

1. An antibody or antibody fragment comprising a variable domain, heavy chain region (VH) and a variable domain, light chain region (VL), wherein VH comprises complementarity determining regions CDRH1, CDRH2, and CDRH3, wherein VL comprises complementarity determining regions CDRL1, CDRL2, and CDRL3, and wherein (a) an amino acid sequence of CDRH1 is as set forth in any one of SEQ ID NOs: ~~151-165,~~ 241-255~~, 331-357,~~ and 547-575; (b) an amino acid sequence of CDRH2 is as set forth in any one of SEQ ID NOs: ~~166-180,~~ 256-270~~, 358-384,~~ and 576-604; (c) an amino acid sequence of CDRH3 is as set forth in any one of SEQ ID NOs: ~~181-195,~~ 271-285~~, 385-411,~~ and 605-633; (d) an amino acid sequence of CDRL1 is as set forth in any one of SEQ ID NOs: ~~196-210,~~ 286-300~~, 412-438,~~ and 634-662; (e) an amino acid sequence of CDRL2 is as set forth in any one of SEQ ID NOs: ~~211-225,~~ 301-315~~, 439-465,~~ and 663-691; and (f) an amino acid sequence of CDRL3 is as set forth in any one of SEQ ID NOs: ~~226-240,~~ 316-330~~, 466-492,~~ and 692-720.

~~2. The antibody or antibody fragment of claim 1, wherein (a) an amino acid sequence of CDRH1 is as set forth in SEQ ID NO: 155; (b) an amino acid sequence of CDRH2 is as set forth in SEQ ID NO: 170; (c) an amino acid sequence of CDRH3 is as set forth in SEQ ID NO: 185; (d) an amino acid sequence of CDRL1 is as set forth in SEQ ID NO: 200; (e) an amino acid sequence of CDRL2 is as set forth in SEQ ID NO: 215; and (f) an amino acid sequence of CDRL3 is as set forth in SEQ ID NO: 230.~~

~~3. The antibody or antibody fragment of claim 1, wherein (a) an amino acid sequence of CDRH1 is as set forth in SEQ ID NO: 152; (b) an amino acid sequence of CDRH2 is as set forth in SEQ ID NO: 167; (c) an amino acid sequence of CDRH3 is as set forth in SEQ ID NO: 182; (d) an amino acid sequence of CDRL1 is as set forth in SEQ ID NO: 197; (e) an amino acid sequence of CDRL2 is as set forth in SEQ ID NO: 212; and (f) an amino acid sequence of CDRL3 is as set forth in SEQ ID NO: 227.~~

~~4. The antibody or antibody fragment of claim 1, wherein (a) an amino acid sequence of CDRH1 is as set forth in SEQ ID NO: 335; (b) an amino acid sequence of CDRH2 is as set forth in SEQ ID NO: 362; (c) an amino acid sequence of CDRH3 is as set forth in SEQ ID NO: 389; (d) an amino acid sequence of CDRL1 is as set forth in SEQ ID NO: 199; (e) an amino acid sequence of CDRL2 is as set forth in SEQ ID NO: 214; and (f) an amino acid sequence of CDRL3 is as set forth in SEQ ID NO: 229.~~

~~5. The antibody or antibody fragment of claim 1, wherein (a) an amino acid sequence of CDRH1 is as set forth in SEQ ID NO: 336; (b) an amino acid sequence of CDRH2 is as set forth in SEQ ID NO: 363; (c) an amino acid sequence of CDRH3 is as set forth in SEQ ID NO: 390; (d) an amino acid sequence of CDRL1 is as set forth in SEQ ID NO: 201; (e) an amino acid sequence of CDRL2 is as set forth in SEQ ID NO: 216; and (f) an amino acid sequence of CDRL3 is as set forth in SEQ ID NO: 231.~~

~~6. The antibody or antibody fragment of claim 1, wherein (a) an amino acid sequence of CDRH1 is as set forth in SEQ ID NO: 158; (b) an amino acid sequence of CDRH2 is as set forth in SEQ ID NO: 173; (c) an amino acid sequence of CDRH3 is as set forth in SEQ ID NO: 188; (d) an amino acid sequence of~~

~~CDRL1 is as set forth in SEQ ID NO: 203; (e) an amino acid sequence of CDRL2 is as set forth in SEQ ID NO: 218; and (f) an amino acid sequence of CDRL3 is as set forth in SEQ ID NO: 233.~~

~~7. The antibody or antibody fragment of claim 1, wherein (a) an amino acid sequence of CDRH1 is as set forth in SEQ ID NO: 551; (b) an amino acid sequence of CDRH2 is as set forth in SEQ ID NO: 580; (c) an amino acid sequence of CDRH3 is as set forth in SEQ ID NO: 609; (d) an amino acid sequence of CDRL1 is as set forth in SEQ ID NO: 290; (e) an amino acid sequence of CDRL2 is as set forth in SEQ ID NO: 305; and (f) an amino acid sequence of CDRL3 is as set forth in SEQ ID NO: 320.~~

~~8. The antibody or antibody fragment of claim 1, wherein (a) an amino acid sequence of CDRH1 is as set forth in SEQ ID NO: 549; (b) an amino acid sequence of CDRH2 is as set forth in SEQ ID NO: 578; (c) an amino acid sequence of CDRH3 is as set forth in SEQ ID NO: 607; (d) an amino acid sequence of CDRL1 is as set forth in SEQ ID NO: 292; (e) an amino acid sequence of CDRL2 is as set forth in SEQ ID NO: 307; and (f) an amino acid sequence of CDRL3 is as set forth in SEQ ID NO: 322.~~

~~9. The antibody or antibody fragment of claim 1, wherein (a) an amino acid sequence of CDRH1 is as set forth in SEQ ID NO: 552; (b) an amino acid sequence of CDRH2 is as set forth in SEQ ID NO: 581; (c) an amino acid sequence of CDRH3 is as set forth in SEQ ID NO: 610; (d) an amino acid sequence of CDRL1 is as set forth in SEQ ID NO: 291; (e) an amino acid sequence of CDRL2 is as set forth in SEQ ID NO: 306; and (f) an amino acid sequence of CDRL3 is as set forth in SEQ ID NO: 321.~~

~~10. The antibody or antibody fragment of claim 1, wherein (a) an amino acid sequence of CDRH1 is as set forth in SEQ ID NO: 554; (b) an amino acid sequence of CDRH2 is as set forth in SEQ ID NO: 583; (c) an amino acid sequence of CDRH3 is as set forth in SEQ ID NO: 612; (d) an amino acid sequence of CDRL1 is as set forth in SEQ ID NO: 288; (e) an amino acid sequence of CDRL2 is as set forth in SEQ ID NO: 303; and (f) an amino acid sequence of CDRL3 is as set forth in SEQ ID NO: 318.~~

~~11.~~2. The antibody or antibody fragment of claim 1, wherein the antibody or antibody fragment binds to ~~a spike glycoprotein~~ACE2.

~~12.~~3. The antibody or antibody fragment of claim 1, wherein the antibody or antibody fragment binds to an extracellular ~~receptor binding~~ domain of ~~the spike glycoprotein~~ACE2.

~~13.~~4. The antibody or antibody fragment of claim 1, wherein the antibody or antibody fragment comprises a K_D of less than 50 nM.

~~14.~~5. The antibody or antibody fragment of claim 1, wherein the antibody or antibody fragment comprises a K_D of less than 25 nM.

~~15.~~6. The antibody or antibody fragment of claim 1, wherein the antibody or antibody fragment comprises a K_D of less than 10 nM.

~~16. The antibody or antibody fragment of claim 1, wherein the antibody or antibody fragment comprises a K_D of less than 5 nM.~~

~~17.~~7. An antibody or antibody fragment comprising a variable domain, heavy chain region (VH) wherein VH comprises complementarity determining regions CDRH1, CDRH2, and CDRH3, wherein (a) an amino acid sequence of CDRH1 is as set forth in any one of SEQ ID NOs: 1-50~~,~~ and 779-919~~, 1344-1523, and~~

2381-2452; (b) an amino acid sequence of CDRH2 is as set forth in any one of SEQ ID NOs: 51-100, and 920-1061 1060, 1524-1703, and 2453-2524; and (c) an amino acid sequence of CDRH3 is as set forth in any one of SEQ ID NOs: 101-150, and 1062-1202, 1704-1883, and 2525-2596.

18. The antibody or antibody fragment of claim 17, wherein (a) the amino acid sequence of CDRH1 is as set forth in SEQ ID NO: 1414; (b) the amino acid sequence of CDRH2 is as set forth in SEQ ID NO: 1594; and (c) the amino acid sequence of CDRH3 is as set forth in SEQ ID NO: 1774.

19. The antibody or antibody fragment of claim 17, wherein (a) the amino acid sequence of CDRH1 is as set forth in SEQ ID NO: 1447; (b) the amino acid sequence of CDRH2 is as set forth in SEQ ID NO: 1627; and (c) the amino acid sequence of CDRH3 is as set forth in SEQ ID NO: 1807.

20. The antibody or antibody fragment of claim 17, wherein (a) the amino acid sequence of CDRH1 is as set forth in SEQ ID NO: 1474; (b) the amino acid sequence of CDRH2 is as set forth in SEQ ID NO: 1654; and (c) the amino acid sequence of CDRH3 is as set forth in SEQ ID NO: 1834.

21. The antibody or antibody fragment of claim 17, wherein (a) the amino acid sequence of CDRH1 is as set forth in SEQ ID NO: 1344; (b) the amino acid sequence of CDRH2 is as set forth in SEQ ID NO: 1524; and (c) the amino acid sequence of CDRH3 is as set forth in SEQ ID NO: 1704.

22. The antibody or antibody fragment of claim 17, wherein (a) the amino acid sequence of CDRH1 is as set forth in SEQ ID NO: 1363; (b) the amino acid sequence of CDRH2 is as set forth in SEQ ID NO: 1543; and (c) the amino acid sequence of CDRH3 is as set forth in SEQ ID NO: 1723.

23. The antibody or antibody fragment of claim 17, wherein (a) the amino acid sequence of CDRH1 is as set forth in SEQ ID NO: 1487; (b) the amino acid sequence of CDRH2 is as set forth in SEQ ID NO: 1667; and (c) the amino acid sequence of CDRH3 is as set forth in SEQ ID NO: 1847.

24. The antibody or antibody fragment of claim 17, wherein (a) the amino acid sequence of CDRH1 is as set forth in SEQ ID NO: 780; (b) the amino acid sequence of CDRH2 is as set forth in SEQ ID NO: 921; and (c) the amino acid sequence of CDRH3 is as set forth in SEQ ID NO: 1063.

25. The antibody or antibody fragment of claim 17, wherein (a) the amino acid sequence of CDRH1 is as set forth in SEQ ID NO: 782; (b) the amino acid sequence of CDRH2 is as set forth in SEQ ID NO: 923; and (c) the amino acid sequence of CDRH3 is as set forth in SEQ ID NO: 1065.

26. The antibody or antibody fragment of claim 17, wherein (a) the amino acid sequence of CDRH1 is as set forth in SEQ ID NO: 39; (b) the amino acid sequence of CDRH2 is as set forth in SEQ ID NO: 89; and (c) the amino acid sequence of CDRH3 is as set forth in SEQ ID NO: 139.

27. The antibody or antibody fragment of claim 17, wherein (a) the amino acid sequence of CDRH1 is as set forth in SEQ ID NO: 832; (b) the amino acid sequence of CDRH2 is as set forth in SEQ ID NO: 973; and (c) the amino acid sequence of CDRH3 is as set forth in SEQ ID NO: 1115.

28. The antibody or antibody fragment of claim 17, wherein (a) the amino acid sequence of CDRH1 is as set forth in SEQ ID NO: 869; (b) the amino acid sequence of CDRH2 is as set forth in SEQ ID NO: 1010; and (c) the amino acid sequence of CDRH3 is as set forth in SEQ ID NO: 1152.

29. The antibody or antibody fragment of claim 17, wherein (a) the amino acid sequence of CDRH1 is as set forth in SEQ ID NO: 889; (b) the amino acid sequence of CDRH2 is as set forth in SEQ ID NO: 1030; and (c) the amino acid sequence of CDRH3 is as set forth in SEQ ID NO: 1172.

30. The antibody or antibody fragment of claim 17, wherein (a) the amino acid sequence of CDRH1 is as set forth in SEQ ID NO: 908; (b) the amino acid sequence of CDRH2 is as set forth in SEQ ID NO: 1049; and (c) the amino acid sequence of CDRH3 is as set forth in SEQ ID NO: 1191.

31.8. An antibody or antibody fragment comprising a variable domain, heavy chain region (VH) and a variable domain, light chain region (VL), wherein the VH comprises an amino acid sequence at least about 90% identical to a sequence as set forth in any one of SEQ ID NOs: 493-519 and 721-749, 2104-2118, and 2134-2147, and wherein the VL comprises an amino acid sequence at least about 90% identical to a sequence as set forth in any one of SEQ ID NOs: 520-546 and 750-778, 2119-2133, and 2148-2161.

32.9. An antibody or antibody fragment comprising a variable domain, heavy chain region (VH) comprising an amino acid sequence at least about 90% identical to a sequence as set forth in any one of SEQ ID NOs: 1203-1343 and 2162-23011884-2063, 2302-2380, and 2597-2668.

33. The antibody or antibody fragment of claim 32, wherein the VH comprises an amino acid sequence at least about 90% identical to SEQ ID NO: 1954.

34. The antibody or antibody fragment of claim 32, wherein the VH comprises an amino acid sequence at least about 90% identical to SEQ ID NO: 1987.

35. The antibody or antibody fragment of claim 32, wherein the VH comprises an amino acid sequence at least about 90% identical to SEQ ID NO: 2014.

36. An antibody, wherein the antibody comprises a sequence comprising at least 90% sequence identity to any one of SEQ ID NOs: 1-2668; and wherein the antibody is a monoclonal antibody, a polyclonal antibody, a bi-specific antibody, a multispecific antibody, a grafted antibody, a human antibody, a humanized antibody, a synthetic antibody, a chimeric antibody, a camelized antibody, a single chain Fvs (scFv), a single chain antibody, a Fab fragment, a F(ab')2 fragment, a Fd fragment, a Fv fragment, a single-domain antibody, an isolated complementarity determining region (CDR), a diabody, a fragment comprised of only a single monomeric variable domain, disulfide linked Fvs (sdFv), an intrabody, an anti-idiotypic (anti-Id) antibody, or ab antigen-binding fragments thereof.

37.10. A nucleic acid composition comprising: a) a first nucleic acid encoding a variable domain, heavy chain region (VH) comprising an amino acid sequence at least about 90% identical to a sequence as set forth in any one of SEQ ID NOs: 493-519 and 721-749, 2104-2118, and 2134-2147; b) a second nucleic acid encoding a variable domain, light chain region (VL) comprising at least about 90% identical to a sequence as set forth in any one of SEQ ID NOs: 520-546 and 750-778, 2119-2133, and 2148-2161; and an excipient.

38.11. A nucleic acid composition comprising: a) a first nucleic acid encoding a variable domain, heavy chain region (VH) comprising an amino acid sequence at least about 90% identical to a sequence as set forth in any one of SEQ ID NOs: 1203-1343 and 2162-23011884-2063, 2302-2380, and 2597-2668; and b) an excipient.

~~39. The nucleic acid composition of claim 38, wherein the VH comprises an amino acid sequence at least about 90% identical to SEQ ID NO: 1954.~~

~~40. The nucleic acid composition of claim 38, wherein the VH comprises an amino acid sequence at least about 90% identical to SEQ ID NO: 1987.~~

~~41. The nucleic acid composition of claim 38, wherein the VH comprises an amino acid sequence at least about 90% identical to SEQ ID NO: 2014.~~

~~42.~~12. ~~A method of treating a SARS-CoV-2 infection, comprising administering the~~Use of the antibody or antibody fragment of any one of claims 1-~~36~~9 in the manufacture of a medicament for the treatment of a SARS-CoV-2 infection.

~~43.~~13. The use~~method~~ of claim ~~37~~12, wherein the antibody or antibody fragment is to be administered prior to exposure to SARS-CoV-2.

~~44.~~14. The use~~method~~ of claim ~~43~~13, wherein the antibody or antibody fragment is to be administered at least about 1 week prior to exposure to SARS-CoV-2.

~~45.~~15. The use~~method~~ of claim ~~43~~13, wherein the antibody or antibody fragment is to be administered at least about 1 month prior to exposure to SARS-CoV-2.

~~46.~~16. The use~~method~~ of claim ~~43~~13, wherein the antibody or antibody fragment is to be administered at least about 5 months prior to exposure to SARS-CoV-2.

~~47.~~17. The use~~method~~ of claim ~~37~~12, wherein the antibody or antibody fragment is to be administered after exposure to SARS-CoV-2.

~~48.~~18. The use~~method~~ of claim ~~47~~17, wherein the antibody or antibody fragment is to be administered at most about 24 hours after exposure to SARS-CoV-2.

~~49.~~19. The use~~method~~ of claim ~~47~~17, wherein the antibody or antibody fragment is to be administered at most about 1 week after exposure to SARS-CoV-2.

~~50.~~20. The use~~method~~ of claim ~~47~~17, wherein the antibody or antibody fragment is to be administered at most about 1 month after exposure to SARS-CoV-2.

~~51. A method of treating an individual with a SARS-CoV-2 infection with the antibody or antibody fragment of any one of claims 1-36 comprising:~~

~~(a) obtaining or having obtained a sample from the individual;~~

~~(b) performing or having performed an expression level assay on the sample to determine expression levels of SARS-CoV-2 antibodies; and~~

~~(c) if the sample has an expression level of the SARS-CoV-2 antibodies then administering to the individual the antibody or antibody fragment of any one of claims 1-36, thereby treating the SARS-CoV-2 infection.~~

~~52. A method for optimizing an antibody comprising:~~

~~(a) providing a plurality of polynucleotide sequences encoding for an antibody or antibody fragment, wherein the antibody or antibody fragment is derived from a subject having SARS-CoV-2;~~

(b) generating a nucleic acid library comprising the plurality of sequences that when translated encode for antibodies or antibody fragments that bind SARS-CoV-2 or ACE2 protein, wherein each of the sequences comprises a predetermined number of variants within a CDR relative to an input sequence that encodes an antibody; wherein the library comprises at least 50,000 variant sequences; and

(c) synthesizing the at least 50,000 variant sequences.

53. The method of claim 52, wherein the antibody library comprises at least 100,000 sequences.

54. The method of claim 52, wherein the method further comprises enriching a subset of the variant sequences.

55. The method of claim 52, wherein the method further comprises expressing the antibody or antibody fragments corresponding to the variant sequences.

56. The method of claim 52, wherein the polynucleotide sequence is a murine, human, or chimeric antibody sequence.

57. The method of claim 52, wherein each sequence of the plurality of variant sequences comprises at least one variant in each CDR of a heavy chain or light chain, relative to the input sequence.

58. The method of claim 52, wherein each sequence of the plurality of variant sequences comprises at least two variants in each CDR of a heavy chain or light chain relative to the input sequence.

59. The method of claim 52, wherein at least one sequence when translated encodes for an antibody or antibody fragment having at least 5X higher binding affinity than a binding affinity of the input sequence.

60. The method of claim 52, wherein at least one sequence when translated encodes for an antibody or antibody fragment having at least 25X higher binding affinity than a binding affinity of the input sequence.

61. The method of claim 52, wherein at least one sequence when translated encodes for an antibody or antibody fragment having at least 50X higher binding affinity than a binding affinity of the input sequence.

62. The method of claim 52, wherein each sequence comprises at least one variant in each CDR of a heavy chain or light chain relative to a germline sequence of the input sequence.

63. The method of claim 52, wherein the nucleic acid library has a theoretical diversity of at least 10^{10} sequences.

64. The method of claim 52, wherein the nucleic acid library has a theoretical diversity of at least 10^{12} sequences.

21. The antibody or antibody fragment of any of claims 1-9 for use in therapy.