

WHAT IS CLAIMED IS:

1. An IgM antibody or IgM-like antibody with enhanced serum half-life comprising five bivalent antibody binding units or variants or fragments thereof and a variant J-chain or functional fragment thereof,

wherein each binding unit comprises two IgM heavy chain constant regions or multimerizing fragments or variants thereof, each associated with an antigen-binding domain or subunit thereof,

wherein the variant J-chain or functional fragment thereof comprises one or more single amino acid substitutions, deletions, or insertions relative to a reference J-chain identical to the variant J-chain except for the one or more single amino acid substitutions, deletions, or insertions, and wherein the variant J-chain can affect serum half-life of the IgM antibody or IgM-like antibody; and

wherein the IgM antibody or IgM-like antibody exhibits an increased serum half-life upon administration to a subject animal relative to a reference IgM antibody or IgM-like antibody that is identical except for the one or more single amino acid substitutions, deletions, or insertions in the variant J-chain, and is administered in the same way to the same animal species.

2. The IgM antibody or IgM-like antibody of claim 1, wherein the variant J-chain or functional fragment thereof comprises one, two, three, or four single amino acid substitutions, deletions, or insertions relative to the reference J-chain.

3. The IgM antibody or IgM-like antibody of claim 1 or claim 2, wherein the variant J-chain or functional fragment thereof comprises an amino acid substitution at the amino acid position corresponding to amino acid Y102 of the wild-type human J-chain (SEQ ID NO: 2).

4. The IgM antibody or IgM-like antibody of claim 3, wherein the amino acid corresponding to Y102 of SEQ ID NO: 2 is substituted with alanine (A), serine (S) or arginine (R).

5. The IgM antibody or IgM-like antibody of claim 4, wherein the amino acid corresponding to Y102 of SEQ ID NO: 2 is substituted with alanine (A).

6. The IgM antibody or IgM-like antibody of claim 5, wherein the J-chain is a variant human J-chain and comprises the amino acid sequence SEQ ID NO: 3.

7. The IgM antibody or IgM-like antibody of claim 4, wherein the amino acid corresponding to Y102 of SEQ ID NO: 2 is substituted with serine (S).
8. The IgM antibody or IgM-like antibody of claim 7 wherein the J-chain is a variant human J-chain and comprises the amino acid sequence SEQ ID NO: 4.
9. The IgM antibody or IgM-like antibody of claim 4, wherein the amino acid corresponding to Y102 of SEQ ID NO: 2 is substituted with arginine (R).
10. The IgM antibody or IgM-like antibody of claim 9, wherein the J-chain is a variant human J-chain and comprises the amino acid sequence SEQ ID NO: 5.
11. The IgM antibody or IgM-like antibody of claim 1 or claim 2, wherein the variant J-chain or functional fragment thereof comprises an amino acid substitution at the amino acid position corresponding to amino acid T103 of the wild-type human J-chain (SEQ ID NO: 2).
12. The IgM antibody or IgM-like antibody of claim 11, wherein the amino acid corresponding to T103 of SEQ ID NO: 2 is substituted with alanine (A).
13. The IgM antibody or IgM-like antibody of claim 12, wherein the J-chain is a variant human J-chain and comprises the amino acid sequence SEQ ID NO: 6.
14. The IgM antibody or IgM-like antibody of claim 1 or claim 2, wherein the variant J-chain or functional fragment thereof comprises an amino acid substitution at the amino acid position corresponding to amino acid N49 or amino acid S51 of the human J-chain (SEQ ID NO: 2), wherein S51 is not substituted with threonine (T), or wherein the variant J-chain comprises amino acid substitutions at the amino acid positions corresponding to both amino acids N49 and S51 of the human J-chain (SEQ ID NO: 2).
15. The IgM antibody or IgM-like antibody of claim 14, wherein the position corresponding to N49 of SEQ ID NO: 2 is substituted with alanine (A), glycine (G), threonine (T), serine (S) or aspartic acid (D).
16. The IgM antibody or IgM-like antibody of claim 15, wherein the position corresponding to N49 of SEQ ID NO: 2 is substituted with alanine (A).
17. The IgM antibody or IgM-like antibody of claim 16, wherein the J-chain is a variant human J-chain and comprises the amino acid sequence SEQ ID NO: 7.
18. The IgM antibody or IgM-like antibody of claim 14, wherein the position corresponding to S51 of SEQ ID NO: 2 is substituted with alanine (A) or glycine (G).
19. The IgM antibody or IgM-like antibody of claim 18, wherein the position corresponding to S51 of SEQ ID NO: 2 is substituted with alanine (A).

20. The IgM antibody or IgM-like antibody of claim 19, wherein the J-chain is a variant human J-chain and comprises the amino acid sequence SEQ ID NO: 8.

21. The IgM antibody or IgM-like antibody of any one of claims 1 to 20, wherein the IgM heavy chain constant regions or multimerizing fragments thereof are variant IgM heavy chain constant regions comprising one or more single amino acid substitutions, deletions, or insertions relative to a reference IgM heavy chain constant region identical to the variant IgM heavy chain constant regions except for the one or more single amino acid substitutions, deletions, or insertions, wherein the variant IgM heavy chain constant regions can affect serum half-life of the IgM antibody or IgM-like antibody, and wherein the IgM antibody or IgM-like antibody exhibits a further increased serum half-life upon administration to a subject animal relative to a reference IgM antibody or IgM-like antibody that is identical except for the one or more single amino acid substitutions, deletions, or insertions in the IgM heavy chain constant regions, and is administered in the same way to the same animal species.

22. The IgM antibody or IgM-like antibody of claim 21, wherein the further increase in serum half-life is additive.

23. An IgM antibody or IgM-like antibody with enhanced serum half-life comprising five or six bivalent antibody binding units or variants or fragments thereof, wherein each binding unit comprises two variant IgM heavy chain constant regions or multimerizing fragments thereof, each associated with an antigen-binding domain or subunit thereof,

wherein the variant IgM heavy chain constant regions or multimerizing fragments thereof each comprise one or more single amino acid substitutions, deletions, or insertions relative to a reference IgM heavy chain constant region identical to the variant IgM heavy chain constant regions except for the one or more single amino acid substitutions, deletions, or insertions, and wherein the variant IgM heavy chain constant regions can affect serum half-life of the IgM antibody or IgM-like antibody; and

wherein the IgM antibody or IgM-like antibody exhibits an increased serum half-life upon administration to a subject animal relative to a reference IgM antibody or IgM-like antibody that is identical except for the one or more single amino acid substitutions, deletions, or insertions in the IgM heavy chain constant regions, and is administered in the same way to the same animal species.

24. The IgM antibody or IgM-like antibody of any one of claims 21 to 23, wherein the variant IgM heavy chain constant regions comprise one, two, three, or four single amino acid substitutions, deletions, or insertions relative to the reference IgM heavy chain constant region.

25. The IgM antibody or IgM-like antibody of any one of claims 21 to 24, wherein the variant IgM heavy chain constant regions comprise an amino acid substitution at the amino acid position corresponding to amino acid R344 of the wild-type human IgM constant region SEQ ID NO: 12.

26. The IgM antibody or IgM-like antibody of claim 25, wherein the amino acid corresponding to R344 of SEQ ID NO: 12 is substituted with alanine (A).

27. The IgM antibody or IgM-like antibody of claim 26, wherein the variant IgM heavy chain constant regions are variant human IgM heavy chain constant region and comprise the amino acid sequence SEQ ID NO: 31.

28. The IgM antibody or IgM-like antibody of any one of claims 21 to 24, wherein the variant IgM heavy chain constant regions comprise an amino acid substitution at the amino acid position corresponding to amino acid E345 of the wild-type human IgM constant region SEQ ID NO: 12.

29. The IgM antibody or IgM-like antibody of claim 28, wherein the amino acid corresponding to E345 of SEQ ID NO: 12 is substituted with alanine (A).

30. The IgM antibody or IgM-like antibody of claim 29, wherein the variant IgM heavy chain constant regions are variant human IgM heavy chain constant region and comprise the amino acid sequence SEQ ID NO: 32.

31. The IgM antibody or IgM-like antibody of any one of claims 21 to 24, wherein the variant IgM heavy chain constant regions comprise an amino acid substitution at the amino acid position corresponding to amino acid S401 of the wild-type human IgM constant region SEQ ID NO: 12.

32. The IgM antibody or IgM-like antibody of claim 31, wherein the amino acid corresponding to S401 of SEQ ID NO: 12 is substituted with alanine (A).

33. The IgM antibody or IgM-like antibody of claim 32, wherein the variant IgM heavy chain constant region are variant human IgM heavy chain constant regions and comprise the amino acid sequence SEQ ID NO: 13.

34. The IgM antibody or IgM-like antibody of any one of claims 21 to 24, wherein the variant IgM heavy chain constant regions comprise an amino acid substitution at the

amino acid position corresponding to amino acid E402 of the wild-type human IgM constant region SEQ ID NO: 12.

35. The IgM antibody or IgM-like antibody of claim 34, wherein the amino acid corresponding to E402 of SEQ ID NO: 12 is substituted with alanine (A).

36. The IgM antibody or IgM-like antibody of claim 35, wherein the variant IgM heavy chain constant regions are variant human IgM heavy chain constant regions and comprise the amino acid sequence SEQ ID NO: 14.

37. The IgM antibody or IgM-like antibody of any one of claims 21 to 24, wherein the variant IgM heavy chain constant regions comprise an amino acid substitution at the amino acid position corresponding to amino acid E403 of the wild-type human IgM constant region SEQ ID NO: 12.

38. The IgM antibody or IgM-like antibody of claim 37, wherein the amino acid corresponding to E403 of SEQ ID NO: 12 is substituted with alanine (A).

39. The IgM antibody or IgM-like antibody of claim 38, wherein the variant IgM heavy chain constant region are variant human IgM heavy chain constants region and comprise the amino acid sequence SEQ ID NO: 34.

40. The IgM antibody or IgM-like antibody of any one of claims 1 to 39, wherein the increased serum half-life comprises an increased alpha half-life ($t_{1/2\alpha}$), an increased beta half-life ($t_{1/2\beta}$), or an increased $t_{1/2\alpha}$ and an increased $t_{1/2\beta}$.

41. The IgM antibody or IgM-like antibody of any one of claims 1 to 40, which further exhibits an increased peak plasma concentration (C_{max}), an increased area under the curve (AUC), a modified clearance time, or any combination thereof relative to the reference antibody.

42. The IgM antibody or IgM-like antibody of any one of claims 1 to 41, wherein the IgM heavy chain constant regions or multimerizing fragments or variants thereof each comprise a $C_{\mu 4}$ domain and an IgM tailpiece (tp) domain.

43. The IgM antibody or IgM-like antibody of claim 42, wherein the IgM heavy chain constant regions or multimerizing fragments or variants thereof each further comprise a $C_{\mu 3}$ domain, a $C_{\mu 2}$ domain, a $C_{\mu 1}$ domain, or any combination thereof.

44. The IgM antibody or IgM-like antibody of any one of claims 1 to 43, wherein the antigen-binding domain comprises a single-chain Fv (ScFv) fragment, or a single domain variable region (VHH).

45. The IgM antibody or IgM-like antibody of any one of claims 1 to 43, wherein the antigen-binding domain subunit comprises a heavy chain variable region (VH).

46. The IgM antibody or IgM-like antibody of any one of claims 1 to 45, wherein each binding unit further comprises two light chain constant regions or fragments or variants thereof each associated with an antigen-binding domain or subunit thereof.

47. The IgM antibody or IgM-like antibody of claim 46, wherein the light chain constant regions are kappa or lambda light chain constant regions or fragments or variants thereof, and wherein antigen binding domain comprises an ScFv fragment or the antigen binding domain subunit comprises a light chain variable region (VL).

48. The IgM antibody or IgM-like antibody of any one of claims 1 to 47, wherein the J-chain or functional fragment or variant thereof further comprises one or more heterologous polypeptides directly or indirectly fused to the J-chain or functional fragment or variant thereof.

49. The IgM antibody or IgM-like antibody of claim 48, wherein the one or more heterologous polypeptides is/are fused to the J-chain or fragment thereof via a peptide linker.

50. The IgM antibody or IgM-like antibody of claim 49, wherein the peptide linker comprises at least 5 amino acids, but no more than 25 amino acids.

51. The IgM antibody or IgM-like antibody of claim 50, wherein the peptide linker consists of GGGGS (SEQ ID NO: 25), GGGGSGGGGS (SEQ ID NO: 26), GGGGSGGGGSGGGGS (SEQ ID NO: 27), GGGGSGGGGSGGGGSGGGGS (SEQ ID NO: 28), or GGGGSGGGGSGGGGSGGGGSGGGGS (SEQ ID NO: 29).

52. The IgM antibody or IgM-like antibody of claims 48 to 51, wherein the one or more heterologous polypeptides is/are fused to the N-terminus of the J-chain or fragment or variant thereof, the C-terminus of the J-chain or fragment or variant thereof, or wherein heterologous polypeptides are fused to both the N-terminus and C-terminus of the J-chain or fragment or variant thereof, wherein the heterologous polypeptides can be the same or different.

53. The IgM antibody or IgM-like antibody of claims 48 to 52, wherein at least one heterologous polypeptide comprises a binding domain.

54. The IgM antibody or IgM-like antibody of claim 53, wherein the binding domain of the heterologous polypeptide is an antibody or antigen-binding fragment thereof.

55. The IgM antibody or IgM-like antibody of claim 54, wherein the antigen-binding fragment comprises a Fab fragment, a Fab' fragment, a F(ab')₂ fragment, a Fd

fragment, a Fv fragment, a single-chain Fv (scFv) fragment, a disulfide-linked Fv (sdFv) fragment, or any combination thereof.

56. The IgM antibody or IgM-like antibody of claim 55, wherein the antigen-binding fragment is a scFv fragment.

57. The IgM antibody or IgM-like antibody of any one of claims 53 to 56, wherein at least one heterologous polypeptide can specifically bind to CD3ε.

58. The IgM antibody or IgM-like antibody of claim 57, wherein the J-chain is a variant of the modified J-chain of SEQ ID NO: 9 (V15J)

59. The IgM antibody or IgM-like antibody of claim 58, wherein the modified J-chain comprises the amino acid sequence of SEQ ID NO: 10 (V15J-Y102A).

60. The IgM antibody or IgM-like antibody of claim 58, wherein the modified J-chain comprises the amino acid sequence of SEQ ID NO: 23 (V15J-T103A).

61. The IgM antibody or IgM-like antibody of claim 58, wherein the modified J-chain comprises the amino acid sequence of SEQ ID NO: 24 (V15J-N49A).

62. A composition comprising the IgM antibody or IgM-like antibody of any one of claims 1 to 61, and a pharmaceutically acceptable carrier.

63. A variant J-chain or functional fragment thereof comprising one or more single amino acid substitutions, deletions, or insertions relative to a reference J-chain identical to the variant J-chain except for the one or more single amino acid substitutions, deletions, or insertions, wherein the variant J-chain can affect the serum half-life of an IgM antibody or IgM-like antibody comprising the variant J-chain.

64. The variant J-chain of claim 63, comprising the amino acid sequence SEQ ID NO: 3, SEQ ID NO: 4, SEQ ID NO: 5, SEQ ID NO: 6, SEQ ID NO: 7, SEQ ID NO: 8, SEQ ID NO: 10, SEQ ID NO: 23, SEQ ID NO: 24, or any combination thereof.

65. An isolated polynucleotide comprising a nucleic acid encoding a subunit polypeptide of the IgM or IgM-like antibody of any one of claims 1 to 61, wherein the subunit polypeptide comprises (a) an IgM or IgM-like heavy chain constant region or multimerizing fragment thereof, (b) an antibody light chain, or (c) a J-chain, a modified J-chain, or functional fragment or variant thereof, or (d) any combination thereof.

66. The polynucleotide of claim 65, wherein the subunit polypeptide comprises an IgM or IgM-like heavy chain constant region or multimerizing fragment thereof.

67. The polynucleotide of claim 66, wherein the subunit polypeptide comprises the amino acid sequence SEQ ID NO: 12, SEQ ID NO: 13, SEQ ID NO: 14, SEQ ID NO: 15, SEQ ID NO: 31, SEQ ID NO: 32, or SEQ ID NO: 34.

68. The polynucleotide of claim 65, wherein the subunit polypeptide comprises an antibody light chain.

69. The polynucleotide of claim 65, wherein the subunit polypeptide comprises a J-chain, a modified J-chain, or any functional fragment or variant thereof.

70. The polynucleotide of claim 69, wherein the subunit comprises the amino acid sequence SEQ ID NO: 1, SEQ ID NO: 2, SEQ ID NO: 3, SEQ ID NO: 4, SEQ ID NO: 5, SEQ ID NO: 7, SEQ ID NO: 8, SEQ ID NO: 9, SEQ ID NO: 10, SEQ ID NO: 11, SEQ ID NO: 23, or SEQ ID NO: 24.

71. The polynucleotide of any one of claims 65 to 70, which comprises two, three, or more nucleic acid sequences encoding two, three or more of the subunit polypeptides.

72. An expression vector comprising the polynucleotide of any one of claims 65 to 71.

73. A host cell comprising the polynucleotide of any of claims 65 to 71 or the expression vector of claim 72.

74. A method of identifying variant J-chains that can increase the serum half-life of pentameric IgM antibodies or pentameric IgM-like antibodies comprising the variant J-chains, the method comprising:

- (a) testing pentameric IgM antibodies or pentameric IgM-like antibodies comprising variant J-chains or fragments thereof for increased serum half-life in a subject animal relative to a reference pentameric IgM antibody or pentameric IgM-like antibody, wherein the variant J-chains or fragments thereof comprise defined amino acid insertions, deletions, or substitutions, and wherein the reference pentameric IgM antibody or pentameric IgM-like antibody comprises a J-chain or fragment thereof identical to the variant J-chains except for the defined amino acid insertions, deletions or substitutions; and
- (b) recovering those variant J-chains or fragments thereof that confer increased serum half-life upon the pentameric IgM antibodies or pentameric IgM-like antibodies relative to reference pentameric IgM antibody or pentameric IgM-like antibody.

75. A method of identifying variant J-chains that can increase the serum half-life of pentameric IgM antibodies or pentameric IgM-like antibodies comprising the variant J-chains, the method comprising:

- (a) testing pentameric IgM antibodies or pentameric IgM-like antibodies comprising variant J-chains or fragments thereof for their level of binding to the Fc alpha-mu receptor (Fc α μ R), the polymeric Ig receptor (pIgR), or both the Fc α μ R and the pIgR, wherein the variant J-chains or fragments thereof comprise defined amino acid insertions, deletions, or substitutions; and
- (b) recovering those variant J-chains or fragments thereof that confer reduced Fc α μ R binding ability, reduced pIgR binding ability, or reduced Fc α μ R and pIgR binding ability upon the pentameric IgM antibodies or pentameric IgM-like antibodies relative to a reference pentameric IgM antibody or pentameric IgM-like antibody comprising a J-chain or fragment thereof identical to the variant J-chains except for the defined amino acid insertions, deletions or substitutions.

76. The method of claim 75, further comprising testing pentameric IgM antibodies or pentameric IgM-like antibodies comprising the recovered variant J-chains or fragments thereof for increased serum half-life in a subject animal relative to a reference pentameric IgM antibody or pentameric IgM-like antibody comprising a J-chain or fragment thereof identical to the recovered variant J-chains or fragments thereof except for the defined amino acid insertions, deletions or substitutions.

77. A method of identifying variant IgM heavy chain constant regions that can increase the serum half-life of an IgM antibody or IgM-like antibody comprising the variant IgM heavy chain constant regions, comprising:

- (a) testing IgM antibodies or IgM-like antibodies comprising variant IgM heavy chain constant regions for increased serum half-life in a subject animal relative to a reference pentameric IgM antibody or pentameric IgM-like antibody, wherein the variant IgM heavy chain constant regions comprise defined amino acid insertions, deletions, or substitutions, and wherein the reference IgM antibody or IgM-like antibody comprises IgM heavy chain constant regions identical to the variant IgM heavy chain constant regions except for the defined amino acid insertions, deletions or substitutions; and

- (b) recovering those IgM antibodies or IgM-like antibodies comprising variant IgM heavy chain constant regions that confer increased serum half-life upon the IgM antibodies or IgM-like antibodies comprising the variant IgM heavy chain constant regions relative to the reference IgM antibody or IgM-like antibody.

78. A method of identifying variant IgM heavy chain constant regions that can increase the serum half-life of an IgM antibody or IgM-like antibody comprising the variant IgM heavy chain constant regions, comprising:

- (a) testing IgM antibodies or IgM-like antibodies comprising variant IgM heavy chain constant regions for their level of binding to the Fc alpha-mu receptor (Fc α μ R), Fc mu receptor (Fc μ R), the polymeric Ig receptor (pIgR), any combination of two of the receptors, or all three of the receptors, wherein the variant IgM heavy chain constant regions comprise defined amino acid insertions, deletions, or substitutions; and
- (b) recovering those IgM antibodies or IgM-like antibodies comprising variant IgM heavy chain constant regions that confer reduced Fc α μ R binding ability, reduced Fc μ R binding ability, reduced pIgR binding ability, reduced ability to bind to any two of the receptors, or reduced ability to bind to all three of the receptors, upon the IgM antibodies or IgM-like antibodies comprising the variant IgM heavy chain constant regions relative to a reference IgM antibody or IgM-like antibody comprising IgM heavy chain constant regions identical to the variant IgM heavy chain constant regions except for the defined amino acid insertions, deletions or substitutions.

79. The method of claim 78, further comprising testing the recovered IgM antibodies or IgM-like antibodies comprising the variant IgM heavy chain constant regions for increased serum half-life in a subject animal relative to a reference IgM antibody or IgM-like antibody comprising IgM heavy chain constant regions identical to the variant IgM heavy chain constant regions except for the defined amino acid insertions, deletions or substitutions.