

CLAIMS

1. A modified immunoglobulin, comprising:
an amyloid-reactive peptide; and
an Ig antibody or functional fragment thereof that binds to human amyloid fibrils, wherein the Ig antibody or functional fragment thereof comprises a heavy chain and a light chain, wherein the peptide and the Ig antibody or functional fragment thereof are joined together at the N-terminal end of the Ig light chain and/or the N- and/or C-terminal end of the Ig heavy chain.
2. The modified immunoglobulin of claim 1, wherein the amyloid-reactive peptide and the Ig antibody or functional fragment thereof are joined together at the N-terminal end of the Ig light chain or the C-terminal end of the Ig heavy chain.
3. The modified immunoglobulin of claim 1 or claim 2, wherein the Ig antibody or functional fragment thereof comprises a light chain variable region (VL) and a heavy chain variable region (VH), wherein:
 - a) the VL comprises a CDR-L1 comprising the amino acid sequence set forth in SEQ ID NO:64, a CDR-L2 comprising the amino acid sequence set forth in SEQ ID NO:21, and a CDR-L3 comprising the amino acid sequence set forth in SEQ ID NO:22, and the VH comprises a CDR-H1 comprising the amino acid sequence set forth in SEQ ID NO:17, a CDR-H2 comprising the amino acid sequence set forth in SEQ ID NO:73, and a CDR-H3 comprising the amino acid sequence set forth in SEQ ID NO:19;
 - b) the VL comprises a CDR-L1 comprising the amino acid sequence set forth in SEQ ID NOs: 64-70, a CDR-L2 comprising the amino acid sequence set forth in SEQ ID NO:21, and a CDR-L3 comprising the amino acid sequence set forth in SEQ ID NO:22, and the VH comprises a CDR-H1 comprising the amino acid sequence set forth in SEQ ID NO:17, a CDR-H2 comprising the amino acid sequence set forth in SEQ ID NO:18, and a CDR-H3 comprising the amino acid sequence set forth in SEQ ID NO:19;
 - c) the VL comprises a CDR-L1 comprising the amino acid sequence set forth in SEQ ID NO: 20; a CDR-L2 comprising the amino acid sequence set forth in SEQ ID NO:21, and a CDR-L3 comprising the amino acid sequence set forth in SEQ ID NO:22, and the VH comprises a CDR-H1 comprising the amino acid sequence set forth in SEQ ID NO:17, a CDR-H2 comprising the amino acid sequence set forth in SEQ ID NO: 71-81; and a CDR-H3 comprising the amino acid sequence set forth in SEQ ID NO:19; or

d) the VL comprises a CDR-L1 comprising the amino acid sequence set forth in SEQ ID NOs: 64-70, a CDR-L2 comprising the amino acid sequence set forth in SEQ ID NO:21, and a CDR-L3 comprising the amino acid sequence set forth in SEQ ID NO:22, and the VH comprises a CDR-H1 comprising the amino acid sequence set forth in SEQ ID NO:17, a CDR-H2 comprising the amino acid sequence set forth in SEQ ID NOs: 71-81; and a CDR-H3 comprising the amino acid sequence set forth in SEQ ID NO:19.

4. The modified immunoglobulin of claim 3, wherein the VL comprises a CDR-L1 comprising the amino acid sequence set forth in SEQ ID NO:64, a CDR-L2 comprising the amino acid sequence set forth in SEQ ID NO:21, and a CDR-L3 comprising the amino acid sequence set forth in SEQ ID NO:22, and the VH comprises a CDR-H1 comprising the amino acid sequence set forth in SEQ ID NO:17, a CDR-H2 comprising the amino acid sequence set forth in SEQ ID NO:73, and a CDR-H3 comprising the amino acid sequence set forth in SEQ ID NO:19.

5. The modified immunoglobulin of claim 4, wherein the VL comprises Leu at position 46 and Phe at position 87, and the VH comprises Leu at position 48, Ser at position 96, Val at position 78, Leu at position 79, Phe at position 80, and Thr at position 94.

6. The modified immunoglobulin of claim 4, wherein the VL comprises an amino acid sequence set forth in SEQ ID NO:36, and the VH comprises an amino acid sequence set forth in SEQ ID NO:55.

7. The modified immunoglobulin any one of claims 1-6, wherein the modified immunoglobulin is humanized.

8. An antibody-peptide fusion protein that binds human amyloid fibrils comprising an antibody fused to an amyloid-reactive peptide, wherein the antibody comprises a light chain variable region (VL), and a heavy chain variable region (VH); wherein:

a) the VL comprises a CDR-L1 comprising the amino acid sequence set forth in SEQ ID NO:64, a CDR-L2 comprising the amino acid sequence set forth in SEQ ID NO:21, and a CDR-L3 comprising the amino acid sequence set forth in SEQ ID NO:22, and the VH comprises a CDR-H1 comprising the

amino acid sequence set forth in SEQ ID NO:17, a CDR-H2 comprising the amino acid sequence set forth in SEQ ID NO:73, and a CDR-H3 comprising the amino acid sequence set forth in SEQ ID NO:19;

b) the VL comprises a CDR-L1 comprising the amino acid sequence set forth in SEQ ID NOs: 64-70, a CDR-L2 comprising the amino acid sequence set forth in SEQ ID NO:21, and a CDR-L3 comprising the amino acid sequence set forth in SEQ ID NO:22, and the VH comprises a CDR-H1 comprising the amino acid sequence set forth in SEQ ID NO:17, a CDR-H2 comprising the amino acid sequence set forth in SEQ ID NO:18, and a CDR-H3 comprising the amino acid sequence set forth in SEQ ID NO:19;

c) the VL comprises a CDR-L1 comprising the amino acid sequence set forth in SEQ ID NO: 20; a CDR-L2 comprising the amino acid sequence set forth in SEQ ID NO:21, and a CDR-L3 comprising the amino acid sequence set forth in SEQ ID NO:22, and the VH comprises a CDR-H1 comprising the amino acid sequence set forth in SEQ ID NO:17, a CDR-H2 comprising the amino acid sequence set forth in SEQ ID NOs: 71-81; and a CDR-H3 comprising the amino acid sequence set forth in SEQ ID NO:19; or

d) the VL comprises a CDR-L1 comprising the amino acid sequence set forth in SEQ ID NOs: 64-70, a CDR-L2 comprising the amino acid sequence set forth in SEQ ID NO:21, and a CDR-L3 comprising the amino acid sequence set forth in SEQ ID NO:22, and the VH comprises a CDR-H1 comprising the amino acid sequence set forth in SEQ ID NO:17, a CDR-H2 comprising the amino acid sequence set forth in SEQ ID NOs: 71-81; and a CDR-H3 comprising the amino acid sequence set forth in SEQ ID NO:19.

9. The antibody-peptide fusion protein of claim 8,

wherein the VL comprises one or more amino acid residues selected from the group consisting of:

- a. Tyr at position 36;
- b. Leu at position 37;
- c. Leu at position 46;
- d. Leu at position 85; and
- e. Phe at position 87

and/or wherein the VH comprises one or more amino acid residues selected from the group consisting of:

- a. Val at position 37;

- b. Leu at position 48;
- c. Leu at position 67;
- d. Ser at position 68;
- e. Lys at position 71;
- f. Ser at position 76;
- g. Val at position 78;
- h. Leu at position 79;
- i. Phe at position 80;
- j. Thr at position 89;
- k. Val at position 93; and
- l. Thr at position 94

wherein the amino acid positions are numbered according to the numbering system of Kabat.

10. The antibody-peptide fusion protein of claim 9, wherein the VL comprises an amino acid sequence set forth in the group consisting of SEQ ID NOs: 32-42; and/or the VH comprises an amino acid sequence set forth in the group consisting of SEQ ID NOs: 43-63.

11. The antibody-peptide fusion protein of claim 9, wherein the VL comprises an amino acid sequence set forth in SEQ ID NO:34, and the VH comprises an amino acid sequence set forth in SEQ ID NO:48.

12. The antibody-peptide fusion protein of claim 9, wherein the VL comprises an amino acid sequence set forth in SEQ ID NO:35, and the VH comprises an amino acid sequence set forth in SEQ ID NO:51.

13. The antibody-peptide fusion protein of claim 8, wherein the VL comprises a CDR-L1 comprising the amino acid sequence set forth in SEQ ID NO:64, a CDR-L2 comprising the amino acid sequence set forth in SEQ ID NO:21, and a CDR-L3 comprising the amino acid sequence set forth in SEQ ID NO:22, and the VH comprises a CDR-H1 comprising the amino acid sequence set forth in SEQ

ID NO:17, a CDR-H2 comprising the amino acid sequence set forth in SEQ ID NO:73, and a CDR-H3 comprising the amino acid sequence set forth in SEQ ID NO:19.

14. The antibody-peptide fusion protein of claim 13, wherein the VL comprises Leu at position 46 and Phe at position 87, and the VH comprises Leu at position 48, Ser at position 96, Val at position 78, Leu at position 79, Phe at position 80, and Thr at position 94.

15. The antibody-peptide fusion protein of claim 13, wherein the VL comprises an amino acid sequence set forth in SEQ ID NO:36, and the VH comprises an amino acid sequence set forth in SEQ ID NO:55.

16. The antibody-peptide fusion protein of any one of claims 8-15, wherein the amyloid-reactive peptide comprises the amino acid sequence set forth in SEQ ID NOs: 1-14.

17. The antibody-peptide fusion protein of any of claims 8-16, wherein the antibody is a chimeric antibody or humanized antibody.

18. The modified immunoglobulin of any one of claims 1-7 or the antibody-peptide fusion protein of any one of claims 8-17, wherein the amyloid reactive peptide is fused to the N-terminus of the antibody light chain or the C-terminus of the antibody heavy chain.

19. The modified immunoglobulin or the antibody-peptide fusion protein of claim 18, wherein the amyloid reactive peptide is fused to the N-terminus of the antibody light chain by a spacer or the C-terminus of the antibody heavy chain by a spacer.

20. The modified immunoglobulin or the antibody-peptide fusion protein of claim 19, wherein the spacer is a peptide spacer.

21. The modified immunoglobulin or the antibody-peptide fusion protein of claim 20, wherein the spacer comprises the amino acid sequence GGGYS.

22. A humanized antibody that binds to human amyloid fibrils, wherein the humanized antibody comprises a light chain variable region (VL) and a heavy chain variable region (VH), wherein

the VL comprises a CDR-L1 comprising the amino acid sequence set forth in SEQ ID NO:20, a CDR-L2 comprising the amino acid sequence set forth in SEQ ID NO:21, and a CDR-L3 comprising the amino acid sequence set forth in SEQ ID NO:22, and

the VH comprises a CDR-H1 comprising the amino acid sequence set forth in SEQ ID NO:17, a CDR-H2 comprising the amino acid sequence set forth in SEQ ID NO:18, and a CDR-H3 comprising the amino acid sequence set forth in SEQ ID NO:19,

wherein the VL comprises one or more amino acid residues selected from the group consisting of:

- a. Tyr at position 36;
- b. Leu at position 37;
- c. Leu at position 46;
- d. Leu at position 85; and
- e. Phe at position 87

and/or wherein the VH comprises one or more amino acid residues selected from the group consisting of:

- a. Val at position 37;
- b. Leu at position 48;
- c. Leu at position 67;
- d. Ser at position 68;
- e. Lys at position 71;
- f. Ser at position 76;
- g. Val at position 78;
- h. Leu at position 79;
- i. Phe at position 80;
- j. Thr at position 89;
- k. Val at position 93; and
- l. Thr at position 94;

wherein the amino acid positions are numbered according to the numbering system of Kabat.

23. The humanized antibody of claim 22, wherein the VL comprises an amino acid sequence set forth in the group consisting of SEQ ID NOs: 33-42, and/or the VH comprises an amino acid sequence set forth in the group consisting of SEQ ID NOs: 44-63.

24. The humanized antibody of claim 22, wherein the VL comprises an amino acid sequence set forth in SEQ ID NO:34, and the VH comprises an amino acid sequence set forth in SEQ ID NO:48; or wherein the VL comprises an amino acid sequence set forth in SEQ ID NO:35, and the VH comprises an amino acid sequence set forth in SEQ ID NO:51.

25. A humanized antibody that binds to human amyloid fibrils, wherein the humanized antibody comprises a light chain variable region (VL) and a heavy chain variable region (VH), wherein:

a) the VL comprises a CDR-L1 comprising the amino acid sequence set forth in SEQ ID NO:64, a CDR-L2 comprising the amino acid sequence set forth in SEQ ID NO:21, and a CDR-L3 comprising the amino acid sequence set forth in SEQ ID NO:22, and the VH comprises a CDR-H1 comprising the amino acid sequence set forth in SEQ ID NO:17, a CDR-H2 comprising the amino acid sequence set forth in SEQ ID NO:73, and a CDR-H3 comprising the amino acid sequence set forth in SEQ ID NO:19;

b) the VL comprises a CDR-L1 comprising the amino acid sequence set forth in SEQ ID NOs: 64-70, a CDR-L2 comprising the amino acid sequence set forth in SEQ ID NO:21, and a CDR-L3 comprising the amino acid sequence set forth in SEQ ID NO:22, and the VH comprises a CDR-H1 comprising the amino acid sequence set forth in SEQ ID NO:17, a CDR-H2 comprising the amino acid sequence set forth in SEQ ID NO:18, and a CDR-H3 comprising the amino acid sequence set forth in SEQ ID NO:19;

c) the VL comprises a CDR-L1 comprising the amino acid sequence set forth in SEQ ID NO: 20; a CDR-L2 comprising the amino acid sequence set forth in SEQ ID NO:21, and a CDR-L3 comprising the amino acid sequence set forth in SEQ ID NO:22, and the VH comprises a CDR-H1 comprising the amino acid sequence set forth in SEQ ID NO:17, a CDR-H2 comprising the amino acid sequence set forth in SEQ ID NOs: 71-81; and a CDR-H3 comprising the amino acid sequence set forth in SEQ ID NO:19; or

d) the VL comprises a CDR-L1 comprising the amino acid sequence set forth in SEQ ID NOs: 64-70, a CDR-L2 comprising the amino acid sequence set forth in SEQ ID NO:21, and a CDR-L3 comprising the amino acid sequence set forth in SEQ ID NO:22, and the VH comprises a CDR-H1 comprising the amino acid sequence set forth in SEQ ID NO:17, a CDR-H2 comprising the amino acid sequence set forth in SEQ ID NOs: 71-81; and a CDR-H3 comprising the amino acid sequence set forth in SEQ ID NO:19.

26. The humanized antibody of claim 25, wherein VL comprises one or more amino acid residues selected from the group consisting of:

- a. Tyr at position 36;
- b. Leu at position 37;
- c. Leu at position 46;
- d. Leu at position 85; and
- e. Phe at position 87

and/or wherein the VH comprises one or more amino acid residues selected from the group consisting of:

- a. Val at position 37;
- b. Leu at position 48;
- c. Leu at position 67;
- d. Ser at position 68;
- e. Lys at position 71;
- f. Ser at position 76;
- g. Val at position 78;
- h. Leu at position 79;
- i. Phe at position 80;
- j. Thr at position 89;
- k. Val at position 93; and
- l. Thr at position 94;

wherein the amino acid positions are numbered according to the numbering system of Kabat.

27. The humanized antibody of claim 25, wherein the VL comprises a CDR-L1 comprising the amino acid sequence set forth in SEQ ID NO:64, a CDR-L2 comprising the amino acid sequence set forth in SEQ ID NO:21, and a CDR-L3 comprising the amino acid sequence set forth in SEQ ID NO:22, and the VH comprises a CDR-H1 comprising the amino acid sequence set forth in SEQ ID NO:17, a CDR-H2 comprising the amino acid sequence set forth in SEQ ID NO:73, and a CDR-H3 comprising the amino acid sequence set forth in SEQ ID NO:19.

28. The humanized antibody of claim 27, wherein the VL comprises Leu at position 46 and Phe at position 87, and the VH comprises Leu at position 48, Ser at position 96, Val at position 78, Leu at position 79, Phe at position 80, and Thr at position 94.

29. The humanized antibody of claim 27, wherein the VL comprises an amino acid sequence set forth in SEQ ID NO:36, and the VH comprises an amino acid sequence set forth in SEQ ID NO:55.

30. The humanized antibody of any one of claims 22-29, wherein the antibody is a full-length antibody, a Fab fragment, or a scFv.

31. The modified immunoglobulin of any one of claims 1-7 and 18-21, the antibody-peptide fusion protein of any one of claims 8-21, or the humanized antibody of any one of claims 22-30, wherein the antibody comprises an Fc region.

32. The modified immunoglobulin, the antibody-peptide fusion protein, or the humanized antibody of claim 31, wherein the Fc region is of an IgG1, IgG2, IgG3, or IgG4 isotype.

33. The modified immunoglobulin of any one of claims 1-7, 18-21, and 31-32, the antibody-peptide fusion of any one of claims 8-21 and 31-32, or the humanized antibody of any one of claims 22-32, wherein the modified immunoglobulin, the antibody-peptide fusion, or the humanized antibody binds to rV λ 6W1l fibrils, Per125 wtATTR extract, KEN hATTR extract, SHI AL λ liver extract, TAL AL κ liver extract, A β , A β (1-40), IAAP, AL κ 4, Al λ 1, or ATTR fibrils.

34. A pharmaceutical composition comprising the modified immunoglobulin of any one of claims 1-7, 18-21, and 31-33, the antibody-peptide fusion protein of any one of claims 8-21 and 31-33, or the humanized antibody of any one of claims 22-33.

35. Nucleic acid(s) encoding the modified immunoglobulin of any one of claims 1-7, 18-21, and 31-33, the antibody-peptide fusion protein of any one of claims 8-21 and 31-33, or the humanized antibody of any one of claims 22-33.

36. A vector comprising the nucleic acid(s) of claim 35.

37. A host cell comprising the vector of claim 36.

38. A method of making a modified immunoglobulin, an antibody-peptide fusion protein, or a humanized antibody comprising culturing the host cell of claim 37 under conditions suitable for expression of the vector encoding the modified immunoglobulin, the antibody-peptide fusion protein, or the humanized antibody and recovering the modified immunoglobulin, the antibody-peptide fusion protein, or the humanized antibody.

39. Use of the modified immunoglobulin of any one of claims 1-7, 18-21, and 31-33, the antibody-peptide fusion protein of any one of claims 8-21 and 31-33, or the humanized antibody of any one of claims 22-33 in the manufacture of a medicament, wherein the medicament is for treating an amyloid related disorder.

40. The use of claim 39, wherein the amyloid related disorder is amyloidosis.

41. The use of claim 39, wherein the amyloid related disorder is selected from the group consisting of AL, AH, A β 2M, ATTR, transthyretin, AA, AApoAI, AApoAII, AGel, ALys, ALEct2, AFib, ACys, ACal, AMed, AIAPP, APro, AIns, APrP, α -synuclein, tau, or A β amyloidosis.

42. The use of any one of claims 39-41, wherein the subject is a human.
43. Use of the modified immunoglobulin of any one of claims 1-7, 18-21, and 31-33, the antibody-peptide fusion protein of any one of claims 8-21 and 31-33, or the humanized antibody of any one of claims 22-33 in the manufacture of a medicament, wherein the medicament is for treating an amyloid-based disease in a subject, comprising:
- determining whether the subject has an amyloid deposit by:
 - detectably labeling the modified immunoglobulin, the antibody-peptide fusion protein, or the humanized antibody of the medicament,
 - administering the medicament to the subject,
 - determining whether a signal associated with the detectable label can be detected from the subject; and,
 - if the signal is detected, administering to the subject an amyloidosis treatment.
44. A method of identifying an amyloid deposit in a subject, comprising detectably labeling the modified immunoglobulin of any one of claims 1-7, 18-21, and 31-33, the antibody-peptide fusion protein of any one of claims 8-21 and 31-33, or the humanized antibody of any one of claims 22-33, administering the modified immunoglobulin, the antibody-peptide fusion protein, or the humanized antibody to the subject, and detecting a signal from the modified immunoglobulin, the antibody-peptide fusion protein, or the humanized antibody.
45. A method of targeting an amyloid deposit for clearance, comprising contacting an amyloid deposit with the modified immunoglobulin of any one of claims 1-7, 18-21, and 31-33 or the antibody-peptide fusion protein of any one of claims 8-21 and 31-33.
46. A method of detecting a ligand, comprising:
- contacting the ligand with the modified immunoglobulin of any one of claims 1-7, 18-21, and 31-33 or antibody-peptide fusion protein of any one of claims 8-21 and 31-33, wherein the modified immunoglobulin or antibody-peptide fusion protein comprises a detectable label, wherein the peptide of the modified immunoglobulin or antibody-peptide fusion protein has binding affinity to the ligand and,
 - determining a signal from the detectable label, thereby detecting the ligand.

47. A method for generating a modified immunoglobulin, comprising:
- providing a first expression vector and a second expression vector,
 - wherein the first expression vector comprises a first nucleic acid sequence encoding an Ig antibody light chain or functional fragment thereof;
 - wherein the second expression vector comprises a second nucleic acid sequence encoding an Ig antibody heavy chain or functional fragment thereof; and
 - wherein the first expression vector and/or the second expression vector comprise a third nucleic acid sequence that encodes a first peptide, the third nucleic acid sequence being located adjacent to the first nucleic acid sequence and/or the second nucleic acid sequence; and
 - inserting the first and second expression vectors into a cell, wherein expression of the first and second expression vectors in the cell results in an immunoglobulin that is joined to the first peptide.