

a heavy chain complementarity determining region (HCDR) 1 comprising an amino acid sequence of GYTFTNYW (SEQ ID NO: 1), an HCDR2 comprising an amino acid sequence of VYPGDGDT (SEQ ID NO: 2), an HCDR3 comprising an amino acid sequence of ARGFTAYGMDY (SEQ ID NO: 3); and a light chain complementarity determining region (LCDR) 1 comprising an amino acid sequence of SSVSY (SEQ ID NO: 4), an LCDR2 comprising an amino acid sequence of DTS (SEQ ID NO: 5), and an LCDR3 comprising an amino acid sequence of QQWSSNPLT (SEQ ID NO: 6).

[0005a] In another aspect, the present disclosure provides a use of the humanized anti-ADGRE2 antibody or antigen-binding fragment thereof as described herein in the manufacture of a medicament for treating an ADGRE2 expressing cancer. In another aspect, the present disclosure provides a pharmaceutical composition comprising a humanized anti-ADGRE2 antibody or fragment thereof as described herein and a pharmaceutically acceptable carrier.

[0005b] In some examples, provided herein is a humanized anti-ADGRE2 antibody or antigen-binding fragment thereof comprising:

VSYMHWYQQKSGLSPKRWIYDTSKLASGVDRFSGSGSGTDYFTISRMEPEDFATY
YCQQWSSNPLTFGGGKLEIK (SEQ ID NO: 20);

(c) QVQLQQSGAEVKKPGASVKLSCKASGYTFTNYWMQWVRQAPGQGLEWIGA
VYPGDGDTRYTQKFQGRATLTADTSTSTVYMEVSSLRSEDVAVYYCARGFTAYGM
DYWGQGTTVTVSSGGGGSGGGGSGGGGSEIVLTQSPATMSASPGERVMTMSCASSS
VSYMHWYQQKSGLSPKRWIYDTSKLASGVDRFSGSGSGTDYFTISRMEPEDFATY
YCQQWSSNPLTFGGGKLEIK (SEQ ID NO: 21); or

(d) QVQLQQSGAEVKKPGASVKVSKASGYTFTNYWMQWVRQAPGQGLEWMG
AVYPGDGDTRHTQKFKGRVTMTRDTSTSTVYMEVSSLRSEDVAVYYCARGFTAYG
MDYWGQGTSLTVSSASTGGGGSGGGGSGGGGSEIVLTQSPATLSLSPGERATLSCSA
SSSVSYMHWYQQKPGAPRLLIYDTSKLASGIPDRFSGSGSGTDFTLTISRLEPEDFAV
YYCQQWSSNPLTFGQGTKVEIK (SEQ ID NO: 22).

[0031] In another aspect, provided herein is a nucleic acid sequence encoding an amino acid sequence that is at least 90% identical to any one of SEQ ID Nos: 7-18.

[0032] In another aspect, provided herein is a vector comprising the nucleic acid sequence as described herein.

[0033] In another aspect, the present disclosure provides an isolated cell comprising the vector as described herein. In some examples, provided herein is a cell comprising the vector.

[0034] In some examples, provided herein is a method of treating cancer comprising administering a humanized anti-ADGRE2 antibody or antigen-binding fragment thereof to a subject in need thereof, the humanized anti-ADGRE2 antibody comprising:

(a) an immunoglobulin heavy chain variable (V_H) region comprising an amino acid sequence that is at least about 80% identical to

QVQLQQSGAEVAKPGASVKLSCKASGYTFTNYWMQWIKQAPGQGLEWIGAVYPGD
GDTRHTQKFKGKATLTADKSTSTAYMEVSSLRSEDVAVYYCARGFTAYGMDYWGQ
GTTVTVSS (SEQ ID NO: 7), and

an immunoglobulin light chain variable (V_L) region comprising an amino acid sequence that is at least about 80% identical to

EIVLTQSPATLSLSPGERATLSCSASSSVSYMHWYQQKPGAPRLLIYDTSKLASGIPD
RFSGSGSGTDFTLTISRLEPEDFAVYYCQQWSSNPLTFGQGTKVEIK (SEQ ID NO: 8);

(b) an immunoglobulin heavy chain variable (V_H) region comprising an amino acid sequence that is at least about 80% identical to

CLAIMS

1. A humanized anti-ADGRE2 antibody or antigen-binding fragment thereof comprising:
 - a heavy chain complementarity determining region (HCDR) 1 comprising an amino acid sequence of GYTFTNYW (SEQ ID NO: 1), an HCDR2 comprising an amino acid sequence of VYPGDGDT (SEQ ID NO: 2), an HCDR3 comprising an amino acid sequence of ARGFTAYGMDY (SEQ ID NO: 3); and
 - a light chain complementarity determining region (LCDR) 1 comprising an amino acid sequence of SSVSY (SEQ ID NO: 4), an LCDR2 comprising an amino acid sequence of DTS (SEQ ID NO: 5), and an LCDR3 comprising an amino acid sequence of QQWSSNPLT (SEQ ID NO: 6).
2. The humanized anti-ADGRE2 antibody or antigen-binding fragment thereof of claim 1 comprising:
 - (a) an immunoglobulin heavy chain variable (V_H) region comprising an amino acid sequence that is at least 80% identical to
QVQLQQSGAEVAKPGASVKLSCKASGYTFTNYWMQWIKQAPGQGLEWIGA
VYPGDGDT RHTQKFKGKATLTADKSTSTAYMEVSSLRSED TAVYYCARGFT
AYGMDYWGQGTTVTVSS (SEQ ID NO: 7), and
an immunoglobulin light chain variable (V_L) region comprising an amino acid sequence that is at least 80% identical to
EIVLTQSPATMSASPGERV TMSCSASSSVSYMHWYQQKSGQSPKRWIYDTSKLA
SGVPARFSGSGSGTDY TFTISSMEPEDFATYYCQQWSSNPLTFGGG TKLEIK (SEQ ID NO: 14);
 - (b) an immunoglobulin heavy chain variable (V_H) region comprising an amino acid sequence that is at least 80% identical to
QVQLQQSGAEVKKPGASVKLSCKASGYTFTNYWMQWIRQAPGQGLEWIGAVY
PGDGDT RYTQKFQGRATLTADTSISTAYMEVSRLRSDD TAVYYCARGFTAYGM
DYWGQGTTVTVSS (SEQ ID NO: 9), and
an immunoglobulin light chain variable (V_L) region comprising an amino acid sequence that is at least 80% identical to
EIVLTQSPATMSASPGERV TMSCSASSSVSYMHWYQQKSGLSPKRWIYDTSKLA

SGVPDRFSGSGSGTDYFTISRMEPEDFATYYCQQWSSNPLTFGGGTKLEIK (SEQ ID NO: 10);

(c) an immunoglobulin heavy chain variable (V_H) region comprising an amino acid sequence that is at least 80% identical to
QVQLQQSGAEVKKPGASVKLSCKASGYTFTNYWMQWVRQAPGQGLEWIGAVY
PGDGDTRYTQKFQGRATLTADTSTSTVYMEVSSLRSEDNAVYYCARGFTAYGM
DYWGQGTTVTVSS (SEQ ID NO: 11), and

an immunoglobulin light chain variable (V_L) region comprising an amino acid sequence that is at least 80% identical to
EIVLTQSPATMSASPGERVMTSCSASSSVSYMHYQQKSGLSPKRWIYDTSKLA
SGVPDRFSGSGSGTDYFTISRMEPEDFATYYCQQWSSNPLTFGGGTKLEIK (SEQ ID NO: 12); or

(d) an immunoglobulin heavy chain variable (V_H) region comprising an amino acid sequence that is at least 80% identical to
QVQLQQSGAEVKKPGASVKVSKASGYTFTNYWMQWVRQAPGQGLEWMGAV
YPGDGDTRHTQKFQGRVTMTDRDTSTSTVYMEVSSLRSEDNAVYYCARGFTAYG
MDYWQGTLVTVSS (SEQ ID NO: 13), and

an immunoglobulin light chain variable (V_L) region comprising an amino acid sequence that is at least 80% identical to
EIVLTQSPATLSLSPGERATLSCSASSSVSYMHYQQKPGLAPRLIYDTSKLASG
IPDRFSGSGSGTDFTLTISRLEPEDFAVYYCQQWSSNPLTFGQGTKVEIK (SEQ ID NO: 8);

(e) an immunoglobulin heavy chain variable (V_H) region comprising an amino acid sequence that is at least 80% identical to
QVQLQQSGAEVKKPGASVKVSKASGYTFTNYWMQWVRQAPGQGLEWIGAV
YPGDGDTRHTQKFQGRVTMTADKSTSTVYMEVSSLRSEDNAVYYCARGFTAYG
MDYWQGTLVTVSS (SEQ ID NO: 15), and

an immunoglobulin light chain variable (V_L) region comprising an amino acid sequence that is at least 80% identical to

QIVLTQSPATLSLSPGERATLTCSASSSVSYMHWYQQKPGLSPKRWIYDTSKLAS
GVPDRFSGSGSGTDYFTIRRLEPEDFATYYCQQWSSNPLTFGQGKVEIK (SEQ
ID NO: 16);

(f) an immunoglobulin heavy chain variable (V_H) region comprising an amino acid
sequence that is at least 80% identical to

QVQLQQSGAEVKKPGASVKLSCKASGYTFTNYWMQWIRQAPGQGLEWIGAVY
PGDGDTRYTQKFQGRATLTADTSTSTAYMEVSSLRSEDVAVYYCARGFTAYGM
DYWGQGTTVTVSS (SEQ ID NO: 17), and

an immunoglobulin light chain variable (V_L) region comprising an amino acid sequence
that is at least 80% identical to

EIVLTQSPATLSASPGERVMTSCSASSSVSYMHWYQQKPGLAPRRWIYDTSKLAS
GVPDRFSGSGSGTDYFTISRMEPEDFATYYCQQWSSNPLTFGGGKLEIK (SEQ
ID NO: 18); or

(e) an immunoglobulin heavy chain variable (V_H) region comprising an amino acid
sequence that is at least 80% identical to

QVQLVQSGAEVAKPGASVKLSCKASGYTFTNYWMQWIKQAPGQGLEWIGAVY
PGDGDTRHTQKFKGKATLTADKSTSTAYMEVSSLRSEDVAVYYCARGFTAYGM
DYWGQGTTVTVSS (SEQ ID NO: 30), and

an immunoglobulin light chain variable (V_L) region comprising an amino acid
sequence that is at least 80% identical to

EIVLTQSPATMSASPGERVMTSCSASSSVSYMHWYQQKSGQSPKRWIYDTSK
LASGVPARFSGSGSGTDYFTISSMEPEDFATYYCQQWSSNPLTFGGGKLEIK
(SEQ ID NO: 31).

3. The humanized anti-ADGRE2 antibody or antigen-binding fragment thereof of claim 1
comprising:

(a) an immunoglobulin heavy chain variable (V_H) region comprising an amino acid
sequence of

QVQLQQSGAEVAKPGASVKLSCKASGYTFTNYWMQWIKQAPGQGLEWIG
AVYPGDGDTRHTQKFKGKATLTADKSTSTAYMEVSSLRSEDVAVYYCARG
FTAYGMDYWGQGTTVTVSS (SEQ ID NO: 7), and

an immunoglobulin light chain variable (V_L) region comprising an amino acid sequence of

EIVLTQSPATMSASPGERVMTMSCSASSSVSYMHWYQQKSGQSPKRWIYDTSKLA
SGVPARFSGSGSGTDYFTFTISSMEPEDFATYYCQQWSSNPLTFGGGTKLEIK (SEQ
ID NO: 14);

(b) an immunoglobulin heavy chain variable (V_H) region comprising an amino acid sequence of

QVQLQQSGAEVKKPGASVKLSCKASGYTFTNYWMQWIRQAPGQGLEWIGAVY
PGDGDTRYTQKFQGRATLTADTSISTAYMEVSRLRSDDTAVYYCARGFTAYGM
DYWGQGTTVTVSS (SEQ ID NO: 9), and

an immunoglobulin light chain variable (V_L) region comprising an amino acid sequence of

EIVLTQSPATMSASPGERVMTMSCSASSSVSYMHWYQQKSGLSPKRWIYDTSKLA
SGVPDRFSGSGSGTDYFTFTISRMEPEDFATYYCQQWSSNPLTFGGGTKLEIK (SEQ
ID NO: 10);

(c) an immunoglobulin heavy chain variable (V_H) region comprising an amino acid sequence of

QVQLQQSGAEVKKPGASVKLSCKASGYTFTNYWMQWVRQAPGQGLEWIGAVY
PGDGDTRYTQKFQGRATLTADTSTSTVYMEVSSLRSEDVAVYYCARGFTAYGM
DYWGQGTTVTVSS (SEQ ID NO: 11), and

an immunoglobulin light chain variable (V_L) region comprising an amino acid sequence of

EIVLTQSPATMSASPGERVMTMSCSASSSVSYMHWYQQKSGLSPKRWIYDTSKLA
SGVPDRFSGSGSGTDYFTFTISRMEPEDFATYYCQQWSSNPLTFGGGTKLEIK (SEQ
ID NO: 12); or

(d) an immunoglobulin heavy chain variable (V_H) region comprising an amino acid sequence of

QVQLQQSGAEVKKPGASVKVSKASGYTFTNYWMQWVRQAPGQGLEWMGAV
YPGDGDTRHTQKFQGRVTMTRDTSTSTVYMEVSSLRSEDVAVYYCARGFTAYG
MDYWGQGTTLVTVSS (SEQ ID NO: 13), and

an immunoglobulin light chain variable (V_L) region comprising an amino acid sequence of

EIVLTQSPATLSLSPGERATLSCSASSSVSYMHWYQQKPGLAPRLLIYDTSKLASG
IPDRFSGSGSGTDFTLTISRLEPEDFAVYYCQQWSSNPLTFGQGTKVEIK (SEQ ID
NO: 8);

(e) an immunoglobulin heavy chain variable (V_H) region comprising an amino acid sequence of

QVQLQQSGAEVKKPGASVKVSCKASGYTFTNYWMQWVRQAPGQGLEWIGAV
YPGDGDTRHTQKFKGRVTMTADKSTSTVYMELSSLRSED TAVYYCARGFTAYG
MDYWGQGTLVTVSS (SEQ ID NO: 15), and

an immunoglobulin light chain variable (V_L) region comprising an amino acid sequence of

QIVLTQSPATLSLSPGERATLTCSASSSVSYMHWYQQKPGLSPKRWIYDTSKLAS
GVPDRFSGSGSGTDYFTIRLEPEDFATYYCQQWSSNPLTFGQGTKVEIK (SEQ
ID NO: 16);

(f) an immunoglobulin heavy chain variable (V_H) region comprising an amino acid sequence of

QVQLQQSGAEVKKPGASVKLSCKASGYTFTNYWMQWIRQAPGQGLEWIGAVY
PGDGDTRYTQKFQGRATLTADTSTSTAYMEVSSLRSED TAVYYCARGFTAYGM
DYWGQGTTVTVSS (SEQ ID NO: 17), and

an immunoglobulin light chain variable (V_L) region comprising an amino acid sequence of

EIVLTQSPATLSASPGERVMTSCSASSSVSYMHWYQQKPGLAPRRWIYDTSKLAS
GVPDRFSGSGSGTDYFTISRMEPEDFATYYCQQWSSNPLTFGGGTKLEIK (SEQ
ID NO: 18); or

(g) an immunoglobulin heavy chain variable (V_H) region comprising an amino acid sequence of

QVQLVQSGAEVAKPGASVKLSCKASGYTFTNYWMQWIKQAPGQGLEWIGAVY
PGDGDTRHTQKFKGKATLTADKSTSTAYMEVSSLRSED TAVYYCARGFTAYGM
DYWGQGTTVTVSS (SEQ ID NO: 30), and

an immunoglobulin light chain variable (V_L) region comprising an amino acid sequence of

EIVLTQSPATMSASPGERVMTMSCSASSSVSYMHWYQQKSGQSPKRWIYDTSK
LASGVPARFSGSGSGTDYFTFTISSMEPEDFATYYCQQWSSNPLTFGGGTKLEIK
(SEQ ID NO: 31).

4. The humanized anti-ADGRE2 antibody or antigen-binding fragment thereof of any one of the preceding claims, wherein the anti-ADGRE2 antibody or antigen-binding fragment thereof is selected from the group consisting of an IgA antibody, IgG antibody, IgE antibody, IgM antibody, and bi- or multi- specific antibody..
5. The humanized anti-ADGRE2 antibody or antigen-binding fragment thereof of any one of the preceding claims, wherein the anti-ADGRE2 antibody or fragment thereof is a monoclonal antibody or a single-chain variable fragment (scFv).
6. The humanized anti-ADGRE2 antibody or antigen-binding fragment thereof of any one of the preceding claims, wherein the humanized anti-ADGRE2 antibody or fragment thereof is an antibody comprising an IgG constant region.
7. The humanized anti-ADGRE2 antibody or antigen-binding fragment thereof of any one of the preceding claims, wherein the ADGRE2 antibody or fragment thereof is a single-chain variable fragment (scFv).
8. The humanized anti-ADGRE2 antibody or antigen-binding fragment thereof of claim 7, wherein the scFv comprises a heavy chain variable sequence, a GS-Linker, and a light chain variable sequence.
9. The humanized anti-ADGRE2 antibody or antigen-binding fragment of claim 8, wherein the scFV comprises a sequence having at least 80% identity to:

(a) QVQLQQSGAEVAKPGASVKLSCKASGYTFTNYWMQWIKQAPGQGLEWI
GAVYPPGDGDTRHTQKFKGKATLTADKSTSTAYMEVSSLRSEDNAVYYCARGFT

AYGMDYWGQGTTVTVSSGGGGSGGGGSGGGGSEIVLTQSPATMSASPGERVMT
SCSASSSVSYMHWYQQKSGQSPKRWIYDTSKLASGVPARFSGSGSGTDYTFTISS
MEPEDFATYYCQQWSSNPLTFGGGTKLEIK (SEQ ID NO: 19);

(b) QVQLQQSGAEVKKPGASVKLSCKASGYTFTNYWMQWIRQAPGQGLEWI
GAVYPGDGDTRYTQKFQGRATLTADTSISTAYMEVSRLRSDDTAVYYCARGFT
AYGMDYWGQGTTVTVSSGGGGSGGGGSGGGGSEIVLTQSPATMSASPGERVMT
SCSASSSVSYMHWYQQKSLSPKRWIYDTSKLASGVDPDRFSGSGSGTDYTFTISR
MEPEDFATYYCQQWSSNPLTFGGGTKLEIK (SEQ ID NO: 20);

(c) QVQLQQSGAEVKKPGASVKLSCKASGYTFTNYWMQWVRQAPGQGLEW
IGAVYPGDGDTRYTQKFQGRATLTADTSTSTVYMEVSSLRSEDVAVYYCARGFT
AYGMDYWGQGTTVTVSSGGGGSGGGGSGGGGSEIVLTQSPATMSASPGERVMT
SCSASSSVSYMHWYQQKSLSPKRWIYDTSKLASGVDPDRFSGSGSGTDYTFTISR
MEPEDFATYYCQQWSSNPLTFGGGTKLEIK (SEQ ID NO: 21); or

(d) QVQLQQSGAEVKKPGASVKVSKASGYTFTNYWMQWVRQAPGQGLEW
MGAVYPGDGDTRHTQKFQGRVTMTRDTSTSTVYMEVSSLRSEDVAVYYCARGF
TAYGMDYWGQGLTVTVSSASTGGGGSGGGGSGGGGSEIVLTQSPATLSLSPGER
ATLSCSASSSVSYMHWYQQKPLAPRLLIYDTSKLASGIPDRFSGSGSGTDFTLTI
SRLEPEDFAVYYCQQWSSNPLTFGQGTKVEIK (SEQ ID NO: 22).

10. The humanized anti-ADGRE2 antibody or antigen-binding fragment of claim 8, wherein the scFV comprises a sequence of:

(a) QVQLQQSGAEVAKPGASVKLSCKASGYTFTNYWMQWIKQAPGQGLEWI
GAVYPGDGDTRHTQKFQKATLTADKSTSTAYMEVSSLRSEDVAVYYCARGFT
AYGMDYWGQGTTVTVSSGGGGSGGGGSGGGGSEIVLTQSPATMSASPGERVMT
SCSASSSVSYMHWYQQKSGQSPKRWIYDTSKLASGVPARFSGSGSGTDYTFTISS
MEPEDFATYYCQQWSSNPLTFGGGTKLEIK (SEQ ID NO: 19);

(b) QVQLQQSGAEVKKPGASVKLSCKASGYTFTNYWMQWIRQAPGQGLEWI
GAVYPGDGDTRYTQKFQGRATLTADTSISTAYMEVSRLRSDDTAVYYCARGFT
AYGMDYWGQGTTVTVSSGGGGSGGGGSGGGGSEIVLTQSPATMSASPGERVMT
SCSASSSVSYMHWYQQKSLSPKRWIYDTSKLASGVDPDRFSGSGSGTDYTFTISR
MEPEDFATYYCQQWSSNPLTFGGGTKLEIK (SEQ ID NO: 20);

(c) QVQLQQSGAEVKKPGASVKLSCKASGYTFTNYWMQWVRQAPGQGLEW
IGAVYPGDGDTRYTQKFQGRATLTADTSTSTVYMEVSSLRSEDVAVYYCARGFT
AYGMDYWGQGTTVTVSSGGGGSGGGGSGGGGSEIVLTQSPATMSASPGERVMT
SCSASSSVSYMHWYQQKSGLSPKRWIYDTSKLASGVPDRFSGSGSGTDYTFTISR
MEPEDFATYYCQQWSSNPLTFGGGTKLEIK (SEQ ID NO: 21); or

(d) QVQLQQSGAEVKKPGASVKVSKASGYTFTNYWMQWVRQAPGQGLEW
MGAVYPGDGDTRHTQKFQGRVTMTRDTSTSTVYMEVSSLRSEDVAVYYCARGF
TAYGMDYWGQGTLVTVSSASTGGGGSGGGGSGGGGSEIVLTQSPATLSLSPGER
ATLSCSASSSVSYMHWYQQKPGAPRLLIYDTSKLASGIPDRFSGSGSGTDFTLTI
SRLEPEDFAVYYCQQWSSNPLTFGQGTKVEIK (SEQ ID NO: 22).

11. The humanized anti-ADGRE2 antibody of any one of the preceding claims, wherein the disassociation constant (K_D) is than less than 10^{-8} M K_D , less than 10^{-9} M K_D , less than 10^{-10} M K_D , less than 10^{-11} M K_D , less than 10^{-12} M K_D , or less than 10^{-13} M K_D .
12. The humanized anti-ADGRE2 antibody of any one of the preceding claims, wherein the EC50 is between 1 – 100 nM.
13. The humanized anti-ADGRE2 antibody of claim 12, wherein the EC50 is between 10 – 95 nM.
14. The humanized anti-ADGRE2 antibody of claim 12, wherein the EC50 is between 25 – 75 nM.
15. Use of the humanized anti-ADGRE2 antibody or antigen-binding fragment thereof of any one of the proceeding claims in the manufacture of a medicament for treating an ADGRE2 expressing cancer.
16. The use according to claim 15, wherein the cancer is selected from leukemia, lymphoma, or myeloma.

17. A pharmaceutical composition comprising a humanized anti-ADGRE2 antibody or fragment thereof of any one of claims 1-15 and a pharmaceutically acceptable carrier.
18. A nucleic acid sequence encoding an amino acid sequence at least 90% identical to any one of SEQ ID Nos: 7-18.
19. A vector comprising the nucleic acid sequence of claim 18.
20. An isolated cell comprising the vector of claim 19.
21. The use according to claim 15, comprising that one or more additional agents is to be administered.
22. The use according to claim 21, wherein the one or more additional agents is selected from an antibody, a chemotherapeutic, or radiation therapy.