

CLAIMS

1. An isolated antibody or antigen binding fragment thereof that binds specifically to betacellulin (BTC), wherein the antibody or antigen binding fragment has a dissociation constant (KD) of 5 pM or less.
2. The antibody or antigen binding fragment thereof of claim 1, which
 - a. binds to at least one residue of SEQ ID NO: 157 selected from the group consisting of G34, H35, F36, S37, R38, C39, P40, K41, Q42, Y43, H45, Y46, R51, R53, F54, V56, A57, E58, Q59, T60, P61, A72, R73, E75, and R76;
 - b. binds to R38, C39, P40, K41, Q42, Y43, H45, Y46, F54, Q59, T60, P61, and R73 of SEQ ID NO: 157;
 - c. binds to P40, K41, Q42, Y43, H45, Y46, E58, Q59, T60, P61, A72, R73, E75, and R76 of SEQ ID NO: 157;
 - d. binds to G34, H35, F36, S37, R38, C39, P40, K41, Q42, R51, R53, F54, and V56 of SEQ ID NO: 157; or
 - e. S37, R38, C39, P40, K41, Q42, Y43, H45, Y46, F54, A57, Q59, T60, P61, A72, R73, and E75 of SEQ ID NO: 157.
3. The antibody or antigen binding fragment thereof of claim 1 or 2, which comprises heavy chain variable region complementarity determining region 1 (HCDR1), heavy chain variable region complementarity determining region 2 (HCDR2), and heavy chain variable region complementarity determining region 3 (HCDR3), and light chain variable region complementarity determining region 1 (LCDR1), light chain variable region complementarity determining region 2 (LCDR2), and light chain variable region complementarity determining region 3 (LCDR3), wherein the HCDR1, HCDR2, HCDR3, LCDR1, LCDR2, and LCDR3 are as set forth in:
 - a. SEQ ID NOs: 1, 2, 3, 14, 15, and 16, respectively; SEQ ID NOs: 4, 2, 3, 14, 15, and 16, respectively; SEQ ID NOs: 5, 6, 3, 17, 18, and 19, respectively; SEQ ID NOs: 7, 8, 9, 20, 18, and 16, respectively;
 - b. SEQ ID NOs: 25, 26, 27, 38, 39, and 40, respectively; SEQ ID NOs: 28, 26, 27, 38, 39, and 40, respectively; SEQ ID NOs: 29, 30, 27, 41, 42, and 43, respectively; or SEQ ID NOs: 31, 32, 33, 44, 42, and 40, respectively;

- c. SEQ ID NOs: 25, 49, 50, 58, 59, and 60, respectively; SEQ ID NOs: 28, 49, 50, 58, 59, and 60, respectively; SEQ ID NOs: 29, 51, 50, 61, 62, and 63, respectively; or SEQ ID NOs: 31, 52, 53, 64, 62, and 60, respectively; or
 - d. SEQ ID NOs: 69, 70, 71, 82, 83, and 84, respectively; SEQ ID NOs: 72, 70, 71, 82, 83, and 84, respectively; SEQ ID NOs: 73, 74, 71, 85, 18, and 86, respectively; or SEQ ID NOs: 75, 76, 77, 87, 18, and 84, respectively.
4. The antibody or antigen binding fragment thereof of any one of claims 1 to 3, which comprises a heavy chain variable region (VH) and a light chain variable region (VL) comprising an amino acid sequence with at least 90%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to:
 - a. SEQ ID NOs: 10 and 21, respectively;
 - b. SEQ ID NOs: 34 and 45, respectively;
 - c. SEQ ID NOs: 54 and 65, respectively; or
 - d. SEQ ID NOs: 78 and 88, respectively.
 5. The antibody or antigen binding fragment thereof of any one of claims 1 to 4, which comprises a heavy chain and a light chain with an amino acid sequence as set forth in:
 - a. SEQ ID NOs: 12 and 23, respectively;
 - b. SEQ ID NOs: 36 and 47, respectively;
 - c. SEQ ID NOs: 56 and 67, respectively; or
 - d. SEQ ID NOs: 80 and 90, respectively.
 6. A polynucleotide comprising nucleotide sequences encoding the antibody or antigen binding fragment thereof of any one of claims 1 to 5.
 7. An expression cassette or a vector comprising the polynucleotide of claim 6.
 8. A host cell comprising the polynucleotide of claim 6 or the expression cassette or vector of claim 7.
 9. A pharmaceutical composition comprising an effective amount of the antibody or antigen binding fragment thereof of any one of claims 1 to 8.
 10. ~~A method of treating a subject having an ophthalmic disorder in need thereof, comprising administering to the subject an effective amount of~~ Use of the antibody or antigen binding fragment thereof of any one of claims 1 to 7 or the pharmaceutical composition of claim 9 in the manufacture of a medicament for the treatment of an ophthalmic disorder, wherein an effective amount of the medicament is to be administered to a subject in need thereof and wherein the

ophthalmic disorder is selected from the group consisting of diabetic macular edema, age-related macular degeneration, neovascular glaucoma, diabetic retinopathy, macular edema, pathologic myopia, retinal vein occlusions, retinopathy of prematurity, abnormal vascular proliferation associated with phakomatoses, central serous chorioretinopathy, and acute multifocal placoid pigment epitheliopathy.

11. The ~~method~~ use of claim 10, wherein the pharmaceutical composition further comprises an anti-vascular endothelial growth factor (VEGF) antagonist or wherein the method further comprises administering to the subject an anti-VEGF antagonist, wherein the anti-VEGF antagonist is selected from the group consisting of ranibizumab, bevacizumab, aflibercept, brolucizumab, and pegaptanib, or wherein the anti-VEGF antagonist comprises a heavy chain and a light chain as set forth in SEQ ID NOs: 103 and 114, respectively.
12. A multi-specific binding molecule comprising 1) an anti-BTC binding moiety and 2) an anti-VEGF binding moiety.
13. The multi-specific binding molecule of claim 12, wherein the anti-VEGF binding moiety is an anti-VEGF antibody or antigen binding fragment thereof in a format selected from the group consisting of an isolated antibody, a Fab, a Fab', a F(ab')₂, a Fv, and a scFv.
14. The multi-specific binding molecule of claim 13, wherein the anti-BTC binding moiety is an anti-BTC Fab and the anti-VEGF binding moiety is an anti-VEGF Fab, wherein the anti-BTC Fab comprises a heavy chain (HA) and a light chain (LA), and wherein the anti-VEGF Fab comprises a heavy chain (HB) and a light chain (LB).
15. The multi-specific binding molecule of claim 14, wherein the HA and the HB are linked in the format from the N-terminus to the C-terminus: N-HA-linker 1-HB-C, and wherein the LA and the LB are linked in the format from the N-terminus to the C-terminus: N-LA-linker 2-LB-C.
16. The multi-specific binding molecule of any one of claims 12 to 15, wherein the anti-BTC binding moiety comprises HCDR1, HCDR2, HCDR3, LCDR1, LCDR2, and LCDR3 as set forth in:
 - a. SEQ ID NOs: 1, 2, 3, 14, 15, and 16, respectively; SEQ ID NOs: 4, 2, 3, 14, 15, and 16, respectively; SEQ ID NOs: 5, 6, 3, 17, 18, and 19, respectively; SEQ ID NOs: 7, 8, 9, 20, 18, and 16, respectively;

- b. SEQ ID NOs: 25, 26, 27, 38, 39, and 40, respectively; SEQ ID NOs: 28, 26, 27, 38, 39, and 40, respectively; SEQ ID NOs: 29, 30, 27, 41, 42, and 43, respectively; or SEQ ID NOs: 31, 32, 33, 44, 42, and 40, respectively;
 - c. SEQ ID NOs: 25, 49, 50, 58, 59, and 60, respectively; SEQ ID NOs: 28, 49, 50, 58, 59, and 60, respectively; SEQ ID NOs: 29, 51, 50, 61, 62, and 63, respectively; or SEQ ID NOs: 31, 52, 53, 64, 62, and 60, respectively; or
 - d. SEQ ID NOs: 69, 70, 71, 82, 83, and 84, respectively; SEQ ID NOs: 72, 70, 71, 82, 83, and 84, respectively; SEQ ID NOs: 73, 74, 71, 85, 18, and 86, respectively; or SEQ ID NOs: 75, 76, 77, 87, 18, and 84, respectively.
- 17. The multi-specific binding molecule of any one of claims 12 to 16, wherein the anti-BTC binding moiety comprises a VH and a VL comprising an amino acid sequence with at least 90%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to:
 - a. SEQ ID NOs: 10 and 21, respectively;
 - b. SEQ ID NOs: 34 and 45, respectively;
 - c. SEQ ID NOs: 54 and 65, respectively; or
 - d. SEQ ID NOs: 78 and 88, respectively.
- 18. The multi-specific binding molecule of any one of claims 12 to 17, wherein the anti-BTC binding moiety comprises a heavy chain and a light chain with an amino acid sequence as set forth in:
 - a. SEQ ID NOs: 12 and 23, respectively;
 - b. SEQ ID NOs: 36 and 47, respectively;
 - c. SEQ ID NOs: 56 and 67, respectively; or
 - d. SEQ ID NOs: 80 and 90, respectively.
- 19. The multi-specific binding molecule of any one of claims 12 to 18, wherein the anti-VEGF binding moiety comprises HCDR1, HCDR2, HCDR3, LCDR1, LCDR2, and LCDR3 as set forth in:
 - a. SEQ ID NOs: 92, 93, 94, 105, 106, and 107 respectively;
 - b. SEQ ID NOs: 95, 93, 94, 105, 106, and 107, respectively;
 - c. SEQ ID NOs: 96, 97, 94, 108, 109, and 110, respectively; or
 - d. SEQ ID NOs: 98, 99, 100, 111, 109, and 107, respectively.
- 20. The multi-specific binding molecule of any one of claims 12 to 19, wherein the anti-VEGF binding moiety comprises a VH and VL comprising an amino acid

- sequence with at least 90%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to SEQ ID NOs: 101 and 112, respectively;
21. The multi-specific binding molecule of any one of claims 12 to 20, wherein the anti-VEGF binding moiety comprises a VH and VL encoded by the nucleic acid sequence as set forth in:
 - a. SEQ ID NOs: 102 and 113, respectively;
 - b. SEQ ID NOs: 117 and 123, respectively;
 - c. SEQ ID NOs: 128 and 133, respectively;
 - d. SEQ ID NOs: 138 and 143, respectively; or
 - e. SEQ ID NOs: 148 and 152, respectively.
 22. The multi-specific binding molecule of any one of claims 12 to 21, wherein the anti-VEGF binding moiety comprises a heavy chain and a light chain with the amino acid sequence as set forth in SEQ ID NOs: 103 and 114, respectively, or wherein the heavy chain and light chain are encoded by a nucleic acid sequence with at least 90%, 95%, 96%, 97%, 98%, 99%, or 100% sequence identity to SEQ ID NOs: 104 and 115, respectively.
 23. The multi-specific binding molecule of any one of claims 12 to 22, wherein the anti-BTC binding moiety comprises a variable heavy chain domain (VHA) and a variable light chain domain (VLA) that bind to BTC, and wherein the anti-VEGF binding moiety comprises a variable heavy chain domain (VHB) and a variable light chain domain (VLB) that bind to VEGF, wherein the multi-specific binding molecule is in the format from the N-terminus to C-terminus as: N-VHA-CH1A-linker-VHB-CH1B-C and N-VLA-CKA-linker-VLB-CKB-C, which comprises a heavy chain comprising the VHA, CH1A, linker, VHB, and CH1B, and which comprises a light chain comprising the VLA, CKA, linker, VLB, and CKB, wherein the heavy chain and the light chain is as set forth in
 - a. SEQ ID NOs: 120 and 125, respectively;
 - b. SEQ ID NOs: 130 and 135, respectively;
 - c. SEQ ID NOs: 140 and 145, respectively; or
 - d. SEQ ID NOs: 149 and 154, respectively
 24. A polynucleotide comprising a nucleotide sequences encoding the multi-specific binding molecule of any one of claims 12 to 23.
 25. An expression cassette or a vector comprising the polynucleotide of claim 24.

26. A pharmaceutical composition comprising an effective amount of the multi-specific binding molecule of any one of claims 12 to 23.
27. ~~A method of treating an ophthalmic disorder in a subject in need thereof, comprising administering to the subject an effective amount of~~ Use of the multi-specific binding molecule of any one of claims 12 to 23 or the pharmaceutical composition of claim 26 in the manufacture of a medicament for the treatment of an ophthalmic disorder, wherein an effective amount of the medicament is to be administered to a subject in need thereof and wherein the ophthalmic disorder is selected from the group consisting of diabetic macular edema, age-related macular degeneration, neovascular glaucoma, diabetic retinopathy, macular edema, pathologic myopia, retinal vein occlusions, retinopathy of prematurity, and abnormal vascular proliferation associated with phakomatoses.
28. A kit comprising the multi-specific binding molecule of any one of claims 12 to 23 or the pharmaceutical composition of claim 26.
29. ~~A method of preventing or treating macular edema, DME, AMD, neovascular AMD, or RVO in a subject in need thereof, comprising administering intravitreally to the subject~~ Use of the multi-specific binding molecule of any one of claims 12 to 23 in the manufacture of a medicament for the prevention or treatment of macular edema, DME, AMD, neovascular AMD, or RVO in a subject in need thereof, wherein the medicament is to be administered to the subject intravitreally at a dose about 0.25 mg/eye, 0.75 mg/eye, 2.5 mg/eye, or 7.5 mg/eye.
30. ~~The method use of claim 29, wherein the dose is~~ 0.25 mg/eye, 0.3 mg/eye, 0.35 mg/eye, 0.4 mg/eye, 0.45 mg/eye, 0.5 mg/eye, 0.55 mg/eye, 0.6 mg/eye, 0.65 mg/eye, 0.7 mg/eye, 0.75 mg/eye, 0.8 mg/eye, 0.85 mg/eye, 0.9 mg/eye, 0.95 mg/eye, 1.0 mg/eye, 1.1 mg/eye, 1.2 mg/eye, 1.3 mg/eye, 1.4 mg/eye, 1.5 mg/eye, 1.6 mg/eye, 1.7 mg/eye, 1.8 mg/eye, 1.9 mg/eye, 2.0 mg/eye, 2.1 mg/eye, 2.2 mg/eye, 2.3 mg/eye, 2.4 mg/eye, 2.5 mg/eye, 2.6 mg/eye, 2.7 mg/eye, 2.8 mg/eye, 2.9 mg/eye, 3.0 mg/eye, 3.1 mg/eye, 3.2 mg/eye, 3.3 mg/eye, 3.4 mg/eye, 3.5 mg/eye, 3.6 mg/eye, 3.7 mg/eye, 3.8 mg/eye, 3.9 mg/eye, 4.0 mg/eye, 4.1 mg/eye, 4.2 mg/eye, 4.3 mg/eye, 4.4 mg/eye, 4.5 mg/eye, 4.6 mg/eye, 4.7 mg/eye, 4.8 mg/eye, 4.9 mg/eye, 5.0 mg/eye, 5.1 mg/eye, 5.2 mg/eye, 5.3 mg/eye, 5.4 mg/eye, 5.5 mg/eye, 5.6 mg/eye, 5.7 mg/eye, 5.8 mg/eye, 5.9 mg/eye, 6.0 mg/eye, 6.1 mg/eye, 6.2 mg/eye, 6.3 mg/eye, 6.4 mg/eye, 6.5 mg/eye, 6.6 mg/eye, 6.7 mg/eye,

6.8 mg/eye, 6.9 mg/eye, 7.0 mg/eye, 7.1 mg/eye, 7.2 mg/eye, 7.3 mg/eye, 7.4 mg/eye, or 7.5 mg/eye.

~~30.~~31. The antibody or antigen binding fragment thereof of any one of claims 1 to 7 or the pharmaceutical composition of claim 9, the multi-specific binding molecule of any one of claims 12 to 23 or the pharmaceutical composition of claim 26 for use in therapy.