

what is claimed is:

1. An antibody or antigen-binding fragment that specifically binds to DLL3, comprising:

i. a single domain antibody (sdAb) moiety comprising:

(1) a CDR1 comprising the amino acid sequence of SEQ ID NO: 6; a CDR2 comprising the amino acid sequence of SEQ ID NO: 87; and a CDR3 comprising the amino acid sequence of SEQ ID NO: 168;

(2) a CDR1 comprising the amino acid sequence of SEQ ID NO: 21; a CDR2 comprising the amino acid sequence of SEQ ID NO: 102; and a CDR3 comprising the amino acid sequence of SEQ ID NO: 183;

(3) a CDR1 comprising the amino acid sequence of SEQ ID NO: 24; a CDR2 comprising the amino acid sequence of SEQ ID NO: 105; and a CDR3 comprising the amino acid sequence of SEQ ID NO: 186;

(4) a CDR1 comprising the amino acid sequence of SEQ ID NO: 27; a CDR2 comprising the amino acid sequence of SEQ ID NO: 108; and a CDR3 comprising the amino acid sequence of SEQ ID NO: 189;

(5) a CDR1 comprising the amino acid sequence of SEQ ID NO: 34; a CDR 2 comprising the amino acid sequence of SEQ ID NO: 115; and a CDR 3 comprising the amino acid sequence of SEQ ID NO: 196;

(6) a CDR1 comprising the amino acid sequence of SEQ ID NO: 39; a CDR2 comprising the amino acid sequence of SEQ ID NO: 120; and a CDR 3 comprising the amino acid sequence of SEQ ID NO: 201;

(7) a CDR1 comprising the amino acid sequence of SEQ ID NO: 7; a CDR2 comprising the amino acid sequence of SEQ ID NO: 88; and a CDR 3 comprising the amino acid sequence of SEQ ID NO: 169;

(8) a CDR1 comprising the amino acid sequence of SEQ ID NO: 8; a CDR2 comprising the amino acid sequence of SEQ ID NO: 89; and a CDR 3 comprising the amino acid sequence of SEQ ID NO: 170; or

(9) a CDR1 comprising the amino acid sequence of SEQ ID NO: 9; a CDR 2 comprising the amino acid sequence of SEQ ID NO: 90; and a CDR 3 comprising the amino acid sequence of SEQ ID NO: 171; or

ii. an antigen-binding fragment comprising a heavy chain variable region (VH) and a light chain variable region (VL), wherein

- (1) the VH comprises a CDR1 set forth in SEQ ID NO: 498, a CDR2 set forth in SEQ ID NO: 499, and a CDR3 set forth in SEQ ID NO: 500, and the VL comprises a CDR1 set forth in SEQ ID NO: 495, a CDR2 set forth in SEQ ID NO: 496, and a CDR3 set forth in SEQ ID NO: 497; or
- (2) the VH comprises a CDR1 set forth in SEQ ID NO: 504, a CDR2 set forth in SEQ ID NO: 505, and a CDR3 set forth in SEQ ID NO: 506, and the VL comprises a CDR1 set forth in SEQ ID NO: 501, a CDR2 set forth in SEQ ID NO: 502, and a CDR3 set forth in SEQ ID NO: 503.

2. The antibody or antigen-binding fragment of claim 1, wherein

- (1) the sdAb comprises an amino acid sequence having at least about 95% sequence identity to the amino acid sequence of any one of SEQ ID NOs: 274-354; or
- (2) the sdAb is a humanized sdAb that comprises an amino acid sequence having at least about 95% sequence identity to the amino acid sequence of any one of SEQ ID NOs: 355-367.

3. The antibody or antigen-binding fragment of claim 1 or 2, wherein

- (1) the VH comprises an amino acid sequence having at least about 95% sequence identity to the amino acid sequence set forth in SEQ ID NO: 508, and the VL comprises an amino acid sequence having at least about 95% sequence identity to the amino acid sequence set forth in SEQ ID NO: 507; or
- (2) the VH comprises an amino acid sequence having at least about 95% sequence identity to the amino acid sequence set forth in SEQ ID NO: 510, and the VL comprises an amino acid sequence having at least about 95% sequence identity to the amino acid sequence set forth in SEQ ID NO: 509.

4. A chimeric antigen receptor (CAR) comprising a DLL3 binding domain, wherein the DLL3 binding domain comprises or is derived from the antibody or antigen-binding fragment of any one of claims 1-3.

5. The CAR of claim 4, wherein the CAR comprises, from N-terminus to C-terminus, a signal peptide, the DLL3 binding domain, a hinge domain, a transmembrane domain, and an intracellular signaling domain.

6. The CAR of claim 5, wherein the intracellular signaling domain is derived from CD3 ζ , FcR γ , FcR β , CD3 γ , CD3 δ , CD3 ϵ , CD5, CD22, CD79a, CD79b, or CD66d.

7. The CAR of claim 5 or 6, wherein the intracellular signaling domain further comprises an intracellular co-stimulatory sequence.

8. The CAR of claim 7, wherein the intracellular co-stimulatory sequence is derived from a co-stimulatory molecule selected from the group consisting of CD27, CD28, 4-1BB, OX40, CD40, PD-1, LFA-1, ICOS, CD2, CD7, LIGHT, NKG2C, B7-H3, TNFRSF9, TNFRSF4, TNFRSF8, CD40LG, ITGB2, KLRC2, TNFRSF18, TNFRSF14, HAVCR1, LGALS9, DAP10, DAP12, CD83, ligands of CD83 and combinations thereof.

9. The CAR of any one of claims 4-8, wherein the CAR comprises an amino acid sequence having at least about 95% sequence identity to an amino acid sequence selected from the group consisting of SEQ ID NOs: 476-484, SEQ ID NOs: 485-494, or SEQ ID NOs: 515-516.

10. The CAR of any one of claims 4-9, wherein the DLL3 binding domain comprises two sdAbs linked to each other.

11. The CAR of claim 10, wherein the CAR comprises an amino acid sequence having at least about 95% sequence identity to an amino acid sequence selected from the group consisting of SEQ ID NOs: 518-520.

12. A nucleic acid molecule encoding a CAR of any one of claims 4-11 or the antibody or antigen-binding fragment of any one of claims 1-3.

13. The nucleic acid molecule of claim 12, wherein the nucleic acid molecule comprises a polynucleotide sequence selected from the group consisting of SEQ ID NOs: 368-461, and 511-514.

14. The nucleic acid molecule of claim 12 or claim 13, wherein the nucleic acid molecule further comprises a polynucleotide sequence encoding a PD-1 Dominant Negative Receptor (PD-1 DNR), a PD-1 Chimeric Switch Receptor (PD-1 CSR), or a TGF- β Dominant Negative Receptor (TGF- β DNR).

15. The nucleic acid molecule of claim 14, wherein the PD-1 DNR comprises an amino acid sequence having at least about 95% sequence identity to SEQ ID NO: 523; the PD-1 CSR comprises an amino acid sequence having at least about 95% sequence identity to SEQ ID NO: 524; or the TGF- β DNR comprises an amino acid sequence having at least about 95% sequence identity to SEQ ID NO: 529.

16. The nucleic acid molecule of any one of claims 12-15, wherein the nucleic acid molecule encoding a peptide having at least about 95% sequence identity to SEQ ID NO: 521 or 522, or the nucleic acid molecule encoding a peptide having at least about 95% sequence identity to an amino acid sequence selected from the group consisting of SEQ ID NOs: 525-528.

17. An expression vector comprising the nucleic acid molecule of any one of claims 12-16.

18. An engineered immune cell comprising the nucleic acid molecule of any one of claims 12-16 or an expression vector of claim 17.

19. The engineered immune cell according to claim 18, wherein the engineered immune cell is selected from the group consisting of a cytotoxic T cell, a helper T cell, a natural killer T cell, a $\gamma\delta$ T cell, a NKT cell and a Nature Killer cell.

20. A pharmaceutical composition comprising the engineered immune cell of claim 18 or claim 19, and a physiologically acceptable excipient.

21. Use of an effective amount of the pharmaceutical composition of claim 20 in the manufacture of a medicament for treating a DLL3 associated disorder in a subject, preferably the DLL3 associated disorder is cancer, more preferably the cancer is selected from the group consisting of lung cancer, melanoma, breast cancer, prostate cancer, colon cancer, renal cell carcinoma, ovarian cancer, neuroblastoma, rhabdomyosarcoma, leukemia and lymphoma.