CLAIMS:

1. A variant Adenosine Deaminase 2 (ADA2) protein or a catalytically active portion thereof, comprising a modification(s) in the sequence of amino acids of an unmodified ADA2 polypeptide or a catalytically active portion thereof, wherein:

i) the variant ADA2 protein, or catalytically active portion thereof, when in dimer form, exhibits reduced heparin binding compared to the corresponding dimer form of the unmodified ADA2 protein of SEQ ID NO:5 or dimer form of the corresponding catalytically activity portion thereof;

ii) modifications that confer reduced heparin binding are selected from among R20A, R20D, R20E, R366A, R366D, R366E, K371A, K371D, K371E, K372A, K372D, K372E, K452E, K11A/R20A, K11A/R20A/K371A, R20A/K371A, K11A/K371A, with reference to amino acid positions set forth in SEQ ID NO:5;

iii) modifications comprise insertions, deletions, and replacements of amino acid residues;

iv) the variant ADA2 protein or catalytically active portion thereof has up to 1, 2, 3, 4, 5, 6, 7, 8, 9, 10 or 10 amino acid modifications in the unmodified ADA2 protein;

v) the unmodified ADA2 protein and catalytically active portion is selected from the group consisting of:

a) a sequence of amino acids selected from the group consisting of SEQ ID NOS: 5 and 380-383;

b) a sequence of amino acids selected from the group consisting of SEQ ID NOS: 5 and 380-383 with all or a part of the PRB domain deleted, wherein the PRB domain consists of residues corresponding to residues 98-156 of SEQ ID NO:5;

c) a sequence of amino acids selected from the group consisting of SEQ ID NOS: 5 and 380-383 with all or a part of the PRB domain deleted and a linker in place of deleted residues, wherein:

the PRB domain consists of residues corresponding to residues98-156 of SEQ ID NO:5; and

the linker is (GGGGS)n, where n = 1 to 5, or is (Gly)n, where n is 2-20;

d) a sequence of amino acids selected from the group consisting of the catalytic domain of the polypeptides of SEQ ID NOS: 5 and 380-383, wherein the catalytic domain consists of residues corresponding to residues 77-473 of the sequence of amino acids set forth in SEQ ID NO:5;

e) a sequence of amino acids selected from the group consisting of the catalytic domain of SEQ ID NOS: 5 and 380-383 with all or a part of the PRB domain deleted, wherein:
the catalytic domain consists of residues corresponding to residues 77-473 of the sequence of amino acids set forth in SEQ ID NO:5; and
the PRB domain consists of residues corresponding to residues 98-156 of SEQ ID NO:5; and

f) a sequence of amino acids selected from the group consisting of the catalytic domain of SEQ ID NOS: 5 and 380-383 with all or a part of the PRB domain deleted, and a linker in place of deleted residues, wherein:
the catalytic domain consists of residues corresponding to residues 77-473 of the sequence of amino acids set forth in SEQ ID NO:5;
the PRB domain consists of residues corresponding to residues 98-156 of SEQ ID NO:5; and
the linker is (GGGGS)n, where n = 1 to 5, or is (Gly)n, where n is 2-20; and

vi) the variant ADA2 protein, when in dimer form, exhibits adenosine deaminase activity to convert adenosine to inosine.

2. The variant ADA2 protein or catalytically active portion thereof of claim 1, further comprising amino acid modifications that confer, to the dimer form, one or more properties selected from among longer serum half-life, altered pH optimum, increased thermal stability, altered receptor binding, and addition of one or more glycosylation sites, compared to the corresponding dimer form of the unmodified ADA2 protein of SEQ ID NO:5 or dimer form of the corresponding catalytically active portion thereof, wherein:
replacements that confer longer serum half-life are selected from among R20E and K371D, with reference to amino acid positions set forth in SEQ ID NO:5;

replacements that confer altered pH optimum are selected from among R20E, K371D, and K371E, with reference to amino acid positions set forth in SEQ ID NO:5;

the replacement that confers increased thermal stability is K371E, with reference to amino acid positions set forth in SEQ ID NO:5;

replacements that confer altered receptor binding are selected from among F119S, F119K, Y224R, Y224N, Y191S, Y191D, F183K, Y191D/Y224R, F109S, F109A, R118D, R118A, Y139T, Y139A, W133S, W133T, P124A, and P124S, with reference to amino acid positions set forth in SEQ ID NO:5; and

amino acid modifications that result in one or more additional glycosylation sites are selected from among --→N1/--→A2/--→S3, R20N/V22S, K371N/D373S, K372N/I374S, T403N/H405S, G404N/P406S, R125N/P126A, S127N/K129S, P126N/E128T, R112N/I114T, I134N/L135C/L136T, I134N/L135S/L136T, R142N/Q144S, E137N/Y139T, and P111N/G113S, with reference to amino acid positions set forth in SEQ ID NO:5.

3. The variant ADA2 protein or catalytically active portion thereof of claim 1 or claim 2, wherein the unmodified ADA2 protein or catalytically active portion thereof has a deletion of all or a portion of the PRB domain.

4. The variant ADA2 protein or catalytically active portion thereof of claim 3, wherein:

the unmodified ADA2 protein or catalytically active portion thereof contains a linker in place of the deleted portion of the PRB domain; and

the linker is (GGGGS)n, where n = 1 to 5, or is (Gly)n, where n is 2-20.

5. The variant ADA2 protein or the catalytically active portion thereof of any of claims 1-4wherein the variant ADA2 protein or the catalytically active portion thereof, when in dimer form, exhibits increased adenosine deaminase activity and reduced heparin binding compared to the dimer form of the unmodified ADA2 protein of SEQ ID NO:5 or the dimer form of the catalytically active portion thereof.

6. The variant ADA2 protein or the catalytically active portion thereof of any of claims 1-5, wherein the variant ADA2 protein or catalytically active portion thereof comprises one or more amino acid replacement(s) selected from the group consisting of R20A, R20D, R20E, R366A, R366D, R366E, K371A, K371D, K371E,

K372A, K372D, K372E and K452E, with reference to amino acid positions set forth in SEQ ID NO:5.

7. The variant ADA2 protein or catalytically active portion thereof of any of claims 1-6, wherein the variant ADA2 protein or catalytically active portion thereof lacks residues N98-N156, C105-E148, C105-T147, or V99-Q144, and optionally, includes a linker in place thereof, wherein the linker is (GGGGS)n, where n = 1 to 5, or is (Gly)n, where n is 2-20.

8. The variant ADA2 protein or catalytically active portion thereof of any of claims 1-5, comprising the sequence of amino acids set forth in any of SEQ ID NOS: 19-24, 27-33, 55-58, 589-594, and 646-657.

9. The variant ADA2 protein or catalytically active portion thereof of any of claims 2-8, wherein, when in dimer form, the variant ADA2 protein or catalytically active portion thereof exhibits one or more properties selected from among longer serum half-life, altered pH optimum, increased thermal stability, altered receptor binding, and addition of one or more glycosylation sites, and has increased adenosine deaminase activity and reduced heparin binding, compared to the dimer form of the unmodified ADA2 protein of SEQ ID NO:5 or dimer form of the catalytically active portion thereof.

10. The variant ADA2 protein or catalytically active portion thereof of any of claims 1-9, wherein the unmodified ADA2 protein is a homodimer, and the monomer form of the unmodified ADA2 protein comprises the sequence of amino acid residues set forth in SEQ ID NO:5.

11. The catalytically active portion of the variant ADA2 protein of any of claims 1-10, wherein the unmodified ADA2 protein is a homodimer of the corresponding catalytically active portions of the polypeptide whose sequence is set forth in of SEQ ID NO:5.

12. The variant ADA2 protein or the catalytically active portion thereof of any of claims 1-11, wherein the unmodified ADA2 protein comprises the sequence of amino acids selected from the group consisting of SEQ ID NOS: 5 and 380-383, or is the catalytically active portion thereof.

13. The variant ADA2 protein or the catalytically active portion thereof of any of claims 1-12, wherein the unmodified ADA2 protein consists of the sequence of

amino acids selected from the group consisting of SEQ ID NOS: 5 and 380-383 or is the catalytically active portion thereof.

14. The variant ADA2 protein or the catalytically active portion thereof of any of claims 1-13, wherein the unmodified ADA2 protein comprises the sequence of amino acids set forth in SEQ ID NO: 5 or is the catalytically active portion thereof.

15. The variant ADA2 protein or the catalytically active portion thereof of any of claims 1-14, wherein the variant ADA2 protein or the catalytically active portion thereof is PEGylated.

16. The variant ADA2 protein or the catalytically active portion thereof of any of claims 1-15, wherein the catalytically active portion comprises the ADA domain that corresponds to residues 77-473 of the sequence of amino acids set forth in SEQ ID NO:5.

17. A variant ADA2 multimer, comprising a plurality of variant ADA2 proteins or the catalytically active portion thereof of any of claims 1-16, wherein the variant ADA2 proteins or catalytically active portion thereof are the same or different.

18. A variant ADA2 dimer, comprising the variant ADA2 protein or the catalytically active portion thereof of any of claims 1-16.

19. A conjugate, comprising the variant ADA2 protein or the catalytically active portion thereof of any of claims 1-16, or the multimer of claim 17, or the dimer of claim 18, linked directly or indirectly via a linker to a heterologous moiety.

20. A nucleic acid molecule encoding the variant ADA2 protein or the catalytically active portion thereof of any of claims 1-16, or the variant ADA2 multimer of claim 17, or the variant ADA2 dimer of claim 18.

21. A vector, comprising the nucleic acid molecule of claim 20.

22. An isolated cell or a cell culture, comprising the vector of claim 21.

23. The cell or cell culture of claim 22, wherein the cell is a tumor-infiltrating lymphocyte (TIL), a cytotoxic T lymphocyte (CTL), a natural killer (NK) cell, or a lymphokine-activated killer (LAK) cell.

24. The cell or cell culture of claim 23, wherein the cell is a T cell that encodes and expresses:

a chimeric antigen receptor (CAR) that is specific for a tumor cell antigen; and

the variant ADA2 protein or the catalytically active portion thereof, or the variant ADA2 multimer, or the variant ADA2 dimer.

25. A method of producing a variant ADA2 protein or the catalytically active portion thereof, comprising culturing or expanding the cell of any of claims 22-24, under conditions whereby the variant ADA2 protein or the catalytically active portion thereof is expressed.

26. The method of claim 25, wherein the variant ADA2 protein is a dimer.

27. The method of claim 25 or claim 26, comprising expanding cells to produce expanded cells.

28. Expanded cells produced by the method of claim 27.

29. Use of the expanded cells of claim 28 for treatment of a tumor.

30. A pharmaceutical composition, comprising the variant ADA2 protein or the catalytically active portion thereof, or the dimer or the multimer of the variant ADA2 protein or the catalytically active portion thereof of any of claims 1-18, or the conjugate of claim 19, in a pharmaceutically acceptable vehicle.

31. The pharmaceutical composition of claim 30, wherein the variant ADA2 protein or the catalytically active portion thereof is PEGylated.

32. The pharmaceutical composition of claim 30 or claim 31, wherein the variant ADA2 protein comprises the sequence of amino acid residues set forth in any of SEQ ID NOs: 19-24, 27-33, 55-58, 589-594, and 646-657, or the catalytically active portion thereof.

33. The variant ADA2 protein or the catalytically active portion thereof, or the dimer or the multimer of the variant ADA2 protein or the catalytically active portion thereof of any of claims 1-18, or the conjugate of claim 19, for use for treatment of a tumor, a cancer, a non-cancer hyperproliferative disease, a fibrotic disease, an infectious disease, a vasculopathy, or an immunodeficiency disease.

34. The variant ADA2 protein or the catalytically active portion thereof, or the dimer or the multimer of the variant ADA2 protein or the catalytically active portion thereof, or the conjugate of claim 33, wherein the variant ADA2 protein or the catalytically active portion thereof is PEGylated.

35. The pharmaceutical composition of any of claims 30-32 for use for treatment of a tumor, a cancer, a non-cancer hyperproliferative disease, a fibrotic

disease, an infectious disease, a vasculopathy, or an immunodeficiency disease in a subject.

36. The pharmaceutical composition of claim 35, wherein the variant ADA2 protein or the catalytically active portion thereof is PEGylated.